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EVALUATION OF AN OGAWA *MYCOBACTERIUM* CULTURE METHOD MODIFIED FOR HIGHER SENSITIVITY EMPLOYING CONCENTRATED SAMPLES

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Abstract: Two egg-based culture media were evaluated for detection of mycobacteria with Lowenstein-Jensen (L-J) as a gold standard. The conventional culture method was modified to improve laboratory diagnosis of tuberculosis in resource scarce countries by employing an inexpensive but sensitive and specific culture method. Sputum samples were collected from pulmonary tuberculosis suspects who visited the chest clinic at the University Teaching Hospital in Zambia. These samples were processed using three different sample treating procedures (with or without sample concentration) and cultured on L-J and Ogawa media for mycobacteria isolation. A total of 276 sputum samples were collected from 138 pulmonary tuberculosis suspects. When the L-J result was used as a standard, the sensitivity of Ogawa and modified Ogawa was 81.7% and 90.3% respectively. Similarly, the specificities of those methods were 96.7% and 92.3% respectively. In total, 90 samples (32.6%) were smear positive and 108 (39.1%) were culture positive. The positivity of each culture method was as follows: 93 (33.7%) in L-J, 98 (35.5%) in modified Ogawa and 82 (29.7%) in original Ogawa. The contamination rate was 1.1%, 5.1%, and 9.8% for L-J, Ogawa and modified Ogawa respectively. The Ogawa culturing method is economical, simple and quick. Its low sensitivity was overcome by employing the concentration method, the sensitivity significantly improving from 81.7% to 90.3%. Ogawa techniques are ideal in overburdened TB laboratories with poor resources in developing countries.

Key words: Ogawa, culture, tuberculosis, and developing country

INTRODUCTION

Tuberculosis is still one of the main public health problems in most developing countries. Global estimations indicate that approximately 8 million new cases of active tuberculosis are notified every year and that 2 million people die of tuberculosis every year (Davies et al., 1996). Incidences are rising in most parts of the world, especially in developing countries where the epidemic of human immunodeficiency virus (HIV) has had the effect of increasing the number of tuberculosis cases. These countries, apart from facing an increased incidence of tuberculosis, are also overburdened with other HIV and non-HIV related problems including depressed economies from drought, famine and the effects of a worldwide recession (Mwinga, 1994). In Zambia, tuberculosis (TB) is one of the major causes of mortality among adults, accounting for 13% of all adult deaths (ZAMBART, 1999). Incidence of TB countrywide has risen five fold from 100 per 100,000 population in 1980 to approximately 500 per 100,000 population in 1996, making Zambia a country with one of the highest incidences of TB in the world (NASTL, 1996; NTP, 1996; WHO, 2002).

The conventional microscopy for acid-fast bacilli (AFB), which 99% of the country's health services provide, is rapid in obtaining results but it has a relatively low sensitivity. The culture method with egg-based media is a conventional way of isolating mycobacteria, but it is time consuming. Recently, the results of radiometric and nonradiometric BACTEC/MGIT systems demonstrated that they are more efficient in the recovery of mycobacteria (Zanetti et al., 1997). However, they are far too expensive for most developing countries. L-J is the standard method for the culture of mycobacteria in Western countries. Ogawa was developed as an inexpensive alternative but proved to have a relatively low sensitivity as compared to L-J (Ogawa et al., 1960). Thus, the Ogawa method employing concentrated samples (modified Ogawa) may be a better alternative even though it may produce a higher contamination rate. In this study we compared the sensitivity of Ogawa and modified Ogawa cultures with that of L-J to evaluate the usefulness of the modified Ogawa method.

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MATERIALS AND METHODS

Clinical samples

One hundred and thirty eight patients with suspected pulmonary tuberculosis visiting the chest clinic at the University Teaching Hospital in Lusaka, Zambia submitted 2 early morning sputum samples consecutively. Smears were prepared by taking a purulent portion of the sputum and smearing it on labelled slides. The slides were air-dried, heat fixed and stained by the standard auramine-phenol method. These smears were examined by fluorescent microscopy (x200). All procedures were performed by one designated technologist.

Culture methods

Each medium was prepared to standard protocol. (Table1) For the L-J method, the sputum samples were decontaminated by adding 3 volumes of 4% sodium hydroxide (NaOH) to one volume of sputum and incubating at 37 C for 15 minutes. They were then centrifuged at 3,000 g for 15 minutes. After disposing the supernatant by decantation,



Figure 1. The interrelation of culture results among the three methods

Table 1. Composition of L-J and Ogawa medium

| Component | L-J | Ogawa |
|-------------------------------------|--------------|------------|
| Potassium dihydrogen phosphate | 1.2 g | 9.0 g |
| Magnesium sulphate | 0.12 g | - |
| Magnesium citrate | 0.3 g | - |
| Sodium glutamate | - | 3.0 g |
| L-asparagine | 1.8 g | - |
| Glycerol/Sodium pyruvate | 6.0 ml/3.6 g | 18.0 ml/ - |
| Distilled water | 300 ml | 300 ml |
| Malachite green (2%) | 16 ml | 18 ml |
| Egg homogenate | 500 ml | 600 ml |
| Benzyl penicillin (1,000,000 IU/ml) | 1 ml | - |

The above measurements were conducted on 100 tubes of each type of media, each 12 ml capacity tube containing 7 ml of media. The mineral salts were autoclaved at 115 \mathbb{C} for 15 minutes. The egg homogenate and benzyl penicillin (as for L-J) were added asceptically after cooling to about 40 \mathbb{C} . The media was then coagulated at 90 \mathbb{C} for 60 minutes after dispensing. approximately 10 ml of distilled water was added to neutralise the sodium hydroxide. The samples were centrifuged again under the same conditions and the supernatant was decanted. Exactly 0.2 ml of deposit was inoculated onto L-J slope. The inoculated slopes were incubated at 37 € until growth was observed or for up to 8 weeks.

For the Ogawa method, the samples were decontaminated and homogenised by mixing one volume of sputum with 3 volumes of 4% sodium hydroxide and incubated for 15 minutes. Then, 0.2 ml of mixture was inoculated onto 3% Ogawa slopes. The inoculated slopes were incubated at 37 \mathbb{C} until growth was observed or for up to 8 weeks. Modified Ogawa method

The clinical samples were decontaminated and homogenised in the same way as mentioned above. They were centrifuged at 3,000 g for 15 minutes and the supernatant was disposed by decantation. Then 0.2 ml of deposit was inoculated onto 3% Ogawa slopes without neutralisation. The inoculated slopes were incubated at 37 \mathbb{C} until growth was observed or for up to 8 weeks.

Reading of cultures

Cultures were read once a week for any growth. The culture tubes that showed positive growth were removed for identification and report. The contaminated cultures were removed and recorded.

Statistical analysis

The sensitivity, specificity, and positive and negative predictive values were compared among original 3% Ogawa, modified Ogawa and L-J cultures. The values for each method with their 95% confidence intervals (CI) were calculated using EpiInfo version 6.03.

RESULTS

A total of 276 samples from 138 pulmonary tuberculosis suspects were examined. With regard to smear results, 186 (67.4%) were negative but 90 (32.6%) were positive. The breakdown of positives was 14 (5.1%), 12 (4.3%), 3 (1.1%) and 61 (22.1%) for scant, 1 +, 2 + and 3 + (WHO standard) respectively. As for smear negative samples, eight patients showed discrepant culture results between the two consecutive samples. Six of the eight patients had a few colonies of positive cultures on each tube while the other two showed higher positives. No patient showed discrepant culture results in a pair of smear positive samples. Among the 90 smear positives, 80 (88.9%) samples were culture positive, while 28 (15.1%) of 186 smear negative samples were culture positive.

A total of 108 (39.1%) samples were found culture positive overall. In the culture positives, 93 (33.7%), 82 (29.7%) and 98 (35.5%) were detected by the L-J, original

| Table 2. | Comparison of sensitivity of Ogawa with the modified |
|----------|--|
| | Ogawa and Lowenstein-Jensen method on the basis of |
| | sample number |

| | | | L-J method | |
|----------|---------------------------|----------|------------|-------|
| | | Positive | Negative | Total |
| Ogawa | Positive | 76 | 6 | 82 |
| | Negative or contamination | 17 | 177 | 194 |
| | Total | 93 | 183 | 276 |
| Modified | Positive | 84 | 14 | 98 |
| Ogawa | Negative or contamination | 9 | 169 | 178 |
| | Total | 93 | 183 | 276 |

3% Ogawa and modified Ogawa method respectively. (Table2) The isolation sensitivity was not significantly different among these three methods. Seventy-two samples (26.1%) were positive in all three methods. Five samples were culture positive by the L-J method only with contaminations of other methods. One and 9 samples were positive only by 3% Ogawa and modified Ogawa respectively. Four samples were positive by both the L-J and 3% Ogawa method. Twelve samples were positive by the L-J and modified Ogawa method, and another 5 were positive by 3% and modified Ogawa method. The interrelation is shown in Figure1. In total, the discrepant culture results were obtained in 54 of 276 (19.6%) samples tested using these three methods. If the contamination is included into negative culture results, the discrepancy was 8.0% (22/276).

The contamination rates were 1.1% (3 of 276), 5.1% (14 of 276) and 9.8% (27 of 276) in the L-J, 3% Ogawa and modified Ogawa method respectively. There were statistically significant differences between the L-J, and the 3% and modified Ogawa methods (p = 0.0114 and p < 0.0001 respectively). There was no difference between 3% Ogawa and modified Ogawa (p = 0.0514). The major contaminant was fungus. There was no tendency to find contaminated results in the samples treated on a specific day.

DISCUSSION

In this study, we evaluated three culturing methods with two egg-based solid media for the detection of *M. tuberculosis*. Despite the high incidence of tuberculosis (WHO, 2002), the developing countries in Sub-Saharan African region have few diagnostic centres that employ culture techniques in the detection and recovery of mycobacteria from clinical samples. In Zambia, for example, there are only two centres that employ culture techniques in the diagnosis of tuberculosis. In these countries, the lack of culture services in the diagnosis of tuberculosis can be attributed to the scarcity of medical resources.

The L-J method has been the standard technique of re-

covering mycobacteria from clinical samples in African countries. The L-J method requires three steps for sample preparation, i.e. decontamination, concentration, and neutralisation. It takes approximately 30 minutes for completion. On the other hand, the Ogawa method takes approximately half the time of L-J for sample preparation with decontamination only. This means that Ogawa is simple and quick. Additionally, from the aspect of economy, Ogawa is less expensive than L-J because of it requires few ingredients. However, the major disadvantage of Ogawa is its relatively low sensitivity due to the non-concentration of samples. The sensitivity of smear and culture for the detection of mycobacteria is improved by sample concentration (Apers et al., 2003). But, there has been no attempt to introduce the sample concentration step to the Ogawa method. In the present study, the sensitivity of Ogawa was improved by adopting the concentration step in the procedure of sample preparation. The improved sensitivity of Ogawa method was equal to or higher than that of the L-J method. It proved effective to employ the sample concentration procedure as a way of increasing the sensitivity of the Ogawa method, and this modification should be adaptable to the conventional Ogawa method without any complication.

The disadvantage of sample concentration in the Ogawa method as compared to the conventional method is the increased possibility of contamination. The contamination rate was not significantly different between the conventional and modified Ogawa method, although it was higher in the 3% Ogawa and modified Ogawa methods as compared to the L-J method. The higher rate of contamination may be attributable to the shorter exposure time to sodium hydroxide for decontamination because no neutralisation of sodium hydroxide is required in the Ogawa procedure. The non-use of penicillin will also contribute to the higher rate of contamination in the 3% Ogawa and modified Ogawa method as compared to the L-J. Additionally, the contaminated samples were mainly seen in one batch of samples. Some reagents might be contaminated by fungi, although no contamination was seen in the L-J at the same period. Another possible, probably major, reason is that the technologist was not practiced in the Ogawa method because the Ogawa method is not implemented widely in African countries. However, the technologists are familiar with the L-J method, which requires the concentration step for sample preparation. The contamination rate will undoubtedly be reduced by further practice. Therefore, when a laboratory attempts to introduce the modified method, avoiding additional workload and cost, it is advisable to employ the method as indicated in this study. It is also advisable to introduce this method to a new laboratory because of its simplicity.

Several recent reports have highlighted the low positivity of sputum examination in HIV positive patients. Zambian tuberculosis patients also showed low sputum smear positivity in HIV positive patients in past studies (Elliott et al., 1993 and Johnson et al., 1997). The implementation of the culture method will increase the isolation of mycobacteria, without any advanced technologies even in HIV epidemic areas. Then, it is important to choose an economical and sensitive culturing method.

A definitive diagnosis of tuberculosis is still dependent on the isolation of *Mycobacterium tuberculosis* by culturing. Although the culture on solid media is inexpensive, it is time consuming and low sensitive (Somoskovi and Magyar, 1999). Although the liquid medium-based radiometric and non-radiometric systems have reduced the recovery time of mycobacteria and have improved the sensitivity of recovery, the cost of these systems is beyond the reach of most developing countries (Pfyffer et al., 1997 and Pfyffer et al., 1997). As a compromise, a slightly cheaper egg-based culture technique, the modified Ogawa, could be employed in the routine diagnosis of tuberculosis in developing countries where resources are scarce.

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FURTHER EVIDENCE THAT GENOTYPE I AND GENOTYPE II OF CRYPTOSPORIDIUM PARVUM ARE DISTINCT

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Abstract: Three new genes of *Cryptosporidium parvum* were cloned, including a gene encoding methionine aminopeptidase, one encoding chaperonin containing T-complex protein 1 delta (TCP-1 delta) and one with unknown function. DNA sequence analysis indicated that these genes are quite conserved, but there were some base pair differences between genotype I and genotype II isolates. These differences were confirmed by PCR-restriction fragment length polymorphism (PCR-RFLP) analysis of the 3 genes from 41 isolates collected from different hosts and geographical origins. In brief, the band patterns generated by endonuclease Hind III or Hinf I restrictions of the gene of methionine aminopeptidase, Sac I restriction of the gene of chaperonin, or Ava II restriction of the unknown gene could differentiate the isolates of *C. parvum* into genotype I and genotype II. PCR primers based on these genes amplified only *C. parvum* genes. Even a single oocyst was detectable with these PCR primers. Thus the results provided further evidence that genotype I and genotype II are distinct, and our three new primers can be used to detect and characterize *C. parvum* isolates with high sensitivity.

Key Word: Cryptosporidium parvum, methionine aminopeptidase, chaperonin, genotype, PCR-RFLP

INTRODUCTION

An intestinal protozoan parasite, *Cryptosporidium parvum* is the major causative agent of cryptosporidiosis in humans and livestock. Outbreaks of human cryptosporidiosis are often caused by the contamination of water supplies with this parasite throughout the world. Because of the epidemiological importance, it is desirable to develop a sensitive detection technique for this parasite and accurate genetic classification.

To this end, many efforts have been made to characterize isolates of *C. parvum* in the past decade. The difference in virulence, infectivity, pathogenesis and drug sensitivity has indicated that *C. parvum* is not a uniform species or monophyletic (Morgan *et al.*, 1999a; Okhuysen *et al.*, 1999; Pereira *et al.*, 2002). The present working hypothesis is that *C. parvum* is composed of two main genotypes, genotype I and genotype II. Genotype I (human genotype or anthroponotic genotype) is exclusively found in humans, while genotype II (bovine genotype or zoonotic genotype) was first found in cattle and has also been found in a wide range of mammals, including humans. Some other hostadapted-genotypes have been also found among *C. parvum* isolates, such as the pig, dog and mouse genotype (Morgan *et al.*, 1999b; Sulaiman *et al.*, 2000; Xiao *et al.*, 2000a)

With the enormous development of gene technology, gene diversities between genotype I and genotype II have

been reported. The evidence includes sequence and/or PCR-RFLP analysis of several gene loci, such as 18S rRNA (Morgan *et al.*, 1997; Xiao *et al.*, 1999), COWP (Spano *et al.*, 1997; Xiao *et al.*, 2000b), HSP70 (Gobet and Toze, 2001; Sulaiman *et al.*, 2000), TRAP (Spano *et al.*, 1999; Sulaiman *et al.*, 1998), and/Cpgp40/15 (Wu *et al.*, 2003).

In the present study, we cloned 3 new genes of *C. parvum* adapting our previous method to design PCR primers from random amplified polymorphic DNA (RAPD), and further confirmed the distinctness between genotype I and genotype II of *C. parvum* by the sequences and PCR-RFLP analysis of the isolates from different hosts with different geographical origins.

MATERIALS AND METHODS

Parasite isolates

Forty-one isolates of *C. parvum* were used. The details were published in our previous paper (Wu *et al.*, 2003). The host and geographical origins are shown in Table 1.

All fecal samples were preserved in 2% K₂Cr₂O₇ and oocysts were isolated using a sucrose flotation method. The samples were refined, contaminated micro-organisms being reduced to a minimum level using an immuno-magnetic separation kit (Dynabeads anti-*Cryptosporidium*, Dynal A. S., Oslo, Norway).

Oocyst DNA was isolated by the method described

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Table 1. Isolates of C. parvum.

| Code | Host origin | Geographical origin | Genotype |
|-------|-------------|---------------------|----------|
| CGJ1 | Calf | Gifu, Japan | II |
| CGJ2 | Calf | Gifu, Japan | II |
| CGJ3 | Calf | Gifu, Japan | II |
| CGJ4 | Calf | Gifu, Japan | II |
| CGJ5 | Calf | Gifu, Japan | II |
| CGJ6 | Calf | Gifu, Japan | II |
| CKJ1 | Calf | Kobe, Japan | Π |
| CKJ2 | Calf | Kobe, Japan | II |
| CKJ3 | Calf | Kobe, Japan | II |
| CKJ4 | Calf | Kobe, Japan | II |
| CKJ5 | Calf | Kobe, Japan | II |
| CKJ6 | Calf | Kobe, Japan | II |
| CKJ7 | Calf | Kobe, Japan | II |
| CKJ8 | Calf | Kobe, Japan | II |
| CKJ9 | Calf | Kobe, Japan | II |
| CKJ10 | Calf | Kobe, Japan | II |
| CKJ11 | Calf | Kobe, Japan | II |
| CNJ1 | Calf | Nagoya, Japan | II |
| CI1 | Calf | Italy | II |
| CI2 | Calf | Italy | II |
| CI3 | Calf | Italy | II |
| CI4 | Calf | Italy | II |
| CI5 | Calf | Italy | II |
| CI6 | Calf | Italy | II |
| CI7 | Calf | Italy | II |
| CI8 | Calf | Italy | II |
| GI1 | Goat | Italy | II |
| HJ1 | Human | Japan | II |
| HJ2 | Human | Japan | Ι |
| HJ3 | Human | Japan | Ι |
| HI1 | Human | Italy | Ι |
| HI2 | Human | Italy | Ι |
| HI3 | Human | Italy | Ι |
| HI4 | Human | Italy | Ι |
| HI5 | Human | Italy | Ι |
| HN1 | Human | Nepal | Ι |
| HN2 | Human | Nepal | Ι |
| HN3 | Human | Nepal | Ι |
| HN4 | Human | Nepal | Ι |
| HN5 | Human | Nepal | Ι |
| HN6 | Human | Nepal | Ι |

previously (Wu *et al.*, 2000). In brief, oocysts were frozen and thawed repeatedly 5 times, treated at 100 \mbox{C} for 20 min, and then the samples were digested with proteinase K at a final concentration of 200 µg/ml at 55 \mbox{C} for 3 hours. The reaction was stopped by heating at 95 \mbox{C} for 5 min. The treated samples were then directly used as a template for PCR.

Development of PCR primers

PCR primers were produced according to our previously described methods (Nagano *et al.*, 1996). In brief, RAPD was produced from *C. parvum* DNA (Code# CGJ2) by means of arbitrary primed PCR (AP-PCR) using 10 base pair primers. The three target genes thus obtained were RAPD SB281, SB289 and SB012, which were produced by primers tgatgaccgc, gcgtgctcac and cggcccctgt, respectively. The DNA fragments were extracted and sequenced. Based on these sequences, 3 pairs of primers were developed as shown in Table 2.

Detection sensitivity and specificity of primers

The sensitivity of primers was tested by amplifying serially diluted template DNA. The purified oocyst DNA was diluted at 160, 40, 10, 2.5, 0.625, 0.156, 0.039 and 0.001 pg/µl. Tested primers included the primers SB281, SB289 and SB012, as well as 3 other primers to amplify the genes of COWP (GenBank accession No.: AF266273, Xiao *et al.*, 2000b), HSP70 (GenBank accession No.: AF221535, Sulaiman *et al.*, 2000), and TRAP1 (GenBank accession No.: AF017267, Spano *et al.*, 1998), as shown in Table 2. PCR conditions were as follows: one cycle of initial denaturing at 94 \mathfrak{C} for 3 min; 35 cycles at 94 \mathfrak{C} for 30 sec, 51-56 \mathfrak{C} (see details in Table 2) for 30 sec, and 72 \mathfrak{C} for 1 min; and one cycle of final extension at 72 \mathfrak{C} for 10 min.

The specificity of the primers SB281, SB289 and SB012 was tested by detecting various kinds of template DNA, including *C. parvum* and control samples of *C. muris*, human, bovine, *Entamoeba histolytica, Giardia lamblia, Blastocystis hominis, Ascaris lumbricoides, Trichomonas vaginalis, Trichinella spiralis and Escherichia coli.*

DNA sequencing and sequence analysis

The RAPD was produced by primer SB281, SB289 or SB012 from 16 isolates, which included 8 calf isolates (Code# CGJ2, CGJ5, CKJ1, CKJ3, CKJ7, CNJ1, CI2 and CI8) from Japan and Italy, one goat isolate (Code# GI1) from Italy and 7 human isolates (Code# HJ1, HJ2, HJ3, HI1, HI2, HN4 and HN6) from Japan, Italy and Nepal. DNA sequence of the RAPD was determined using a Thermo Sequenase cycle sequencing Kit (USB Corporation, Cleveland, USA) and an automatic sequencer (Model LIC-4200, Aloka Co., Ltd., Tokyo, Japan). The sequence data were analyzed using DNASIS Software (Hitachi Software Engineering, Tokyo, Japan). Homology searching on the nucleotide and protein database was carried out using the BLAST program at NCBI (Bethesda, MD, USA). Pairwise sequence alignment and protein identities were performed using CLUS-TALW 1.8 software (Jeanmougin et al., 1998) and PHYLIP DNADIST software (distributed by Felsenstein, Department of Genetics, University of Washington, Seattle).

PCR-RFLP

PCR and RFLP were performed according to our previously described methods (Wu et al., 1999). PCR prod-

Table 2. Criteria of PCR Primers.

| Primer | Sequence | Annealing Tm | Product size | Target gene |
|--------|---|-----------------|-----------------|---|
| SB281 | 5'-gatagtgttccatttgagagg-3' 5'-ttagatgcaacaaatacaggat-3' | 51.0 | 553 | methionine aminopeptidase |
| SB289 | 5'-cccaattcagttctgtctct-3' 5'-aataatgctcctaacaacgc-3' | 51.0 | 505 | chaperonin containing TCP-1 delta |
| SB012 | 5'-ctccgttcgatgatgcagatg-3' 5'-cggcccctgtagaaataagtca-3' | 51.3 | 434 | unknown |
| COWP | 5'-ggttcctcctatgcctttct 5'-gtgggcattcctttgtgac | 52.7 | 869 | oocyst wall protein |
| HSP70 | 5'-tcagttgccatcagtagagc 5'-tcttcttctcagcctcatca | 52.5 | 841 | heat shock protein |
| TRAP1 | 5'-ggatgggtatcaggtaataagaa 5'-tagcccagttctgactctctg | 52.0 | 1177 | thrombospondin-related adhesive protein |

Table 3. Predicted RFLP pattern of RAPD SB281, SB289 and SB012 PCR products.

| | Size of predicated bands | | | | _ |
|---------|--------------------------|--------------|----------|----------|----------|
| Isolate | SB281 | | SB289 | SB012 | Genotype |
| | Hae III | Hinf I | Sac I | Ava II | |
| CGJ2 | 59, 494 | 75, 478 | 505 | 152, 282 | II |
| CI2 | 59, 494 | 75, 478 | 505 | 152, 282 | II |
| IG1 | 59, 494 | 75, 478 | 505 | 152, 282 | II |
| HJ2 | 59, 204, 290 | 75, 138, 340 | 232, 273 | 434 | Ι |
| HJ3 | 59, 204, 290 | 75, 138, 340 | 232, 273 | 434 | Ι |
| HN6 | 59, 204, 290 | 75, 138, 340 | 232, 273 | 434 | Ι |
| HI2 | 59, 204, 290 | 75, 138, 340 | 232, 273 | 434 | Ι |

ucts were purified by ethanol precipitation, and then digested with an appropriate restriction endonuclease (Hind III, Hinf I, Sac I and Ava II) according to the manufacturer's instructions. The digests were subjected to electrophoresis.

RESULTS

Sequence analysis

Three dense RAPD bands with an appropriate molecular size of 610 (SB281), 600 (SB289) and 550 bp (SB012) were generated from the template DNA of the isolate CGJ2. The RAPD fragments were isolated from the gel and sequenced (GenBank accession No: AY488139, AY488146, AY488153). Based on the 3 sequences, 3 pairs of primers were developed (Table 2). The RAPD from 16 isolates (including 8 calf isolates from Japan and Italy, one goat isolate from Italy and 7 human isolates from Japan, Italy and Nepal) were produced with the 3 developed primers and sequenced (GenBank accession numbers are shown in Figs 1, 2 and 3).

Homology analysis of the RAPD sequence showed that these genes were quite conserved among different isolates, as shown in Figs 1, 2 and 3. The identities of the sequences of SB281 RAPD, SB289 RAPD, and SB012 RAPD were 98.0-100%, 97.6-100%, and 98.2-100% among the 16 isolates, respectively.

The animal isolates shared nearly the same sequence regardless of their origin. The identities of SB281 RAPD, SB289 RAPD, and SB012 RAPD among the 9 animal isolates were 99.3-100%, 99.6-100% and 99.0-100%, respectively. The same was true for human isolates. The identities of RAPD SB281, RAPD SB289, and RAPD SB012 among the 7 human isolates were 98.4-99.6%, 98.0-100% and 99.3-100%, respectively.

Although DNA sequence was conserved, there were point mutations in human origin isolates (except the isolate HJ1) at identical nucleotide sites. The point mutation pattern was different from the animal origin isolates as shown in Figs 1, 2 and 3.

The DNA sequence of the RAPD fragments were deduced into an amino acid sequence and subjected to Gen-Bank for blast searching. The blast results showed that SB281 was a protein gene encoding methionine aminopeptidase. The deduced amino acid sequence shared 52-57% identities with the sequence of methionine aminopeptidase of other organisms, for example, *Drosophila melanogaster*, *Arabidopsis thaliana*, human, mouse and *Caenorhabditis elegans* (Fig 4). The SB289 was a protein gene encoding

| II II I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 1: TTAGATGCAACAAATACAGGATTAAAAGTTGCCGGAATTGATGTTATGTTTTCTGAAATAGGCTCAGCTATAGAAGAAGTCATTAAATCT 1: |
|-----------------------------------|--|--|
| II II I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 91: TATGAATTTGAGTACAAAAGTAAGGTTTATAATATTAAAACCTATTAAAAATCTAAATGGTCATTCAATTCTACCATATCATATCCATGGA 91: |
| II II I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 181: GGGAAATCAGTACCAATTATTGCAACAAATGATGACACAAGAATGGAGGAAAATGAAATATATGCCATCGAAACATTTGCAACCACTGGA 181: |
| II II I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 271: AGAGGCTACGTTACAGAAGGGCTAGATTGTAGCCACTATATGAAATACTATGACAATCCCTTCCTAAACGAAAATTCAACCAGACTTAAT 271: |
| II II I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 361: C. 361: |
| II II I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 451: CATGCACTAGCTCTTAAATCATTGGTAGACTCGGAAATCATTCGGCCGTATCCTCCATTAAACGACATTCCGGGTTCATTCTCCTCTCAA 451: |
| II II I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 541: ATGGAACACTATC 541: |

Figure 1. DNA sequence diversities among *Cryptosporidium parvum* isolates in SB281 (methionine aminopeptidase) gene. The genes from 16 isolates were sequenced and typed into genotype I and genotype II, as indicated in the left margin by I and II. Filled circles in the alignment indicate identical base pairs. The differences between genotype I and genotype II are indicated by shading. GenBank accession numbers of CGJ2, CI2, GI1, HJ2, HJ3, HI2 and HN6 are AY488139, AY488140, AY488141, AY488142, AY488143, AY488144 and AY488145, respectively.

chaperonin containing TCP-1 delta subunit. The deduced amino acid sequence shared 54-61% identities with the sequence of chaperonin containing TCP-1 delta subunit of other organisms, such as *D. melanogaster*, *A. thaliana*, hu-

man, mouse and C. elegans (Fig 5).

Differentiation of C. parvum *isolates by the RFLP method* After PCR with the primer SB281, SB289 or SB012,

| II II I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 1:CCCAATTCAGTTCTGTCTCTCAGCTCCAAAGACAGATATTGGAGAATAATATTGTAGTTAAGGACTATACAGCTATGGATCGATTACT 1: | CAG |
|--|--|--|------|
| II II I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 91: GGAAGAGAGACTTTTGATTGCCAAAATGATTAAACAGATAGCAGCAACAGGCTGCAACGTATTACTTATCCAAAAGAGTATATTGAG 91: | AGA |
| II II I I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 181:GCCGATAAGTACTTTGGCATTAGATTATTGCGCTAAAGCAAAAATTCTTGTTGTCAAAGATATTGAGAGGGATGAAATTGAATTTTT 181: <td>'AAG</td> | 'AAG |
| II II I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 271: TAAAGCTCTGAACTGCTCTCCTATTGCTTCACTTGACCATTTTACTTCAGACAAACTTGGCGCTGCTAACAGAGTTTCTGATGAGGA 271: | .TCT |
| II II I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 361: AGATGGCAATGGCGTATTTGCAGAATTACAGGGATACCAGGAAAAGACATGATGATAATATTTGTAAGGGCATCTAATATGTTGAT 361: | GCT |
| II II I I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 451: AGATGAGACAGAACGTTGCATTCACGATGCATTATGCGTTGTTAGGAGCATTATT 451: | |

Figure 2. DNA sequence diversities among *Cryptosporidium parvum* isolates in SB289 (chaperonin containing TCP-1 delta subunit) gene. The genes from 16 isolates were sequenced and typed into genotype I and genotype II, as indicated in the left margin by I and II. Filled circles in the alignment indicate identical base pairs. The differences between genotype I and genotype II are indicated by shading. GenBank accession numbers of CGJ2, CI2, GI1, HJ2, HJ3, HI2 and HN6 are AY488146, AY488147, AY488148, AY488149, AY488150, AY488151 and AY488152, respectively.

all 41 DNA samples produced the expected size bands of 553 bp, 505 bp and 434 bp, respectively. Thus 123 (3 times 41) RAPDs were analyzed by PCR-RFLP.

Two kinds of band patterns (Panels A and B in Fig 6) were produced by endonuclease Hind III and Hinf I restriction of the RAPD SB281. All of the calf isolates from both Japan and Italy, the isolate from the goat of Italy and one isolate from a Japanese patient (Code# HJ1) showed the same kind of RFLP pattern of genotype II. All of the hu-

man origin isolates, except one Japanese human isolate (Code# HJ1) produced the same kind of RFLP pattern of genotype I, which was different from that of genotype II.

The same results were obtained when the 41 isolates were analyzed by endonuclease Sac I restriction of RAPD SB289 (Panel C in Fig 6) and Ava II restriction of RAPD SB012 (Panel D in Fig 6).

| II II I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 1 : CTCCGTTCGATGATGCAGATGCATTGCAAAGATTTTGTTCTTTTATTCTCCCTATGCTAGATGAAAAAGGTACCCATACAGGTGAGGAAG |
|-----------------------------------|--|---|
| II II I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 91: TTGATCTATCAATAACTGAAAATAATGAATTTATTTATCAACAAATGAAAGTTTCAAAGTTGGTCCATCAGATCAAACACGAGGATGTGA 91: |
| II II I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 181: ACCAAATTTTCAGCATGTACGGAATTCTTTTCGACTTATTTAGTCGTGTAGATAGTTCAAGATTTAAATATACATTCCCAACTCTGGGGT 181: |
| II II I I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 271: ATTGTGCCATAAATCTTATTGAAACAACTTTATCAAAGGAAAAAACAGATAATGAGCCTTCAAAATTATCTGTAAAAAAGATATTTCAGT 271: |
| II II I I I I | CGJ2 CI2 GI1 HJ2 HJ3 HI2 HN6 | 361: TTATTCATAAGATCGCAATAATCCTTGTAACTTGTGCGCCAGAACTAGCTCTTGACTTATTCTACAGGGGCCGT 361: |

Figure 3. DNA sequence diversities among *Cryptosporidium parvum* isolates in SB012 gene. The genes from 16 isolates were sequenced and typed into genotype I and genotype II, as indicated in the left margin by I and II. Filled circles in the alignment indicate identical base pairs. The differences between genotype I and genotype II are indicated by shading. GenBank accession numbers of CGJ2, CI2, GI1, HJ2, HJ3, HI2 and HN6 are AY488153, AY488154, AY488155, AY488156, AY488157, AY488158, and AY488159, respectively.

Detection sensitivity and specificity of primers

The detection sensitivity of primers was tested by means of PCR with serially diluted template DNA, as described in Materials and Methods. The density of bands tended to be faint when a lower concentration of template DNA was used. The primers SB281, SB289 and SB012 gave the highest sensitivity in detecting template DNA of *C. parvum*. The minimum concentration of template DNA necessary for a positive reaction was 0.156 pg with the primer SB281, 0.039 pg with the primer SB289 and 0.156 pg with the primer SB012 (Panels A, B and C in Fig 7), while the control primers of COWP, HSP 70 and PRAP1 showed a minimum detection concentration of 10 pg, 10 pg and 40 pg, respectively (Panels D, E and F in Fig 7).

All 3 kinds of primers produced bands with the expected size from *C. parvum*, while control samples produced negative results, indicating the species specificity of these constructed primers.

DISCUSSION

Epidemiologically, molecular markers of pathogens are useful in determining the importance of responsible reagents, for example, distinguishing between zoonotic and anthroponotic transmissions. Reportedly, both genotype I and genotype II of *C. parvum* are responsible for the sporadic infection and outbreak of human cryptosporidiosis (McLauchlin *et al.*, 1999; Ong *et al.*, 1999; Sulaiman *et al.*, 1998; Xiao *et al.*, 2000a).

In the present study, 3 new target genes were proposed and assessed for genetic analysis. The sequence analysis of these genes from 41 isolates of *C. parvum* from different hosts and geographical origins indicated that there are identical base pair differences between the isolates from human and animal origins. The PCR-RFLP analysis of the 3 genes confirmed that these 3 new genes are useful for the genotyping of *C. parvum*, because all of the 3 genes showed 2 kinds of RFLP patterns which correspond to genotype I and

| C. parvum | LDATNTGLKVAGIDVMFSEIGSAIEEVIKSYEFEYKSKVYNIKPIKNLNGHSILPYHIHGGKSVPIIATN |
|-----------------|--|
| D.melanogaster | KE IRE RLCD A Q ME I. LDG. T. P A. R S R A T VKGG |
| A.thaliana | RE. Y. I. E RLCD. A. Q. M V. ING. VFQV. S. R G. Q. A VKGG |
| Mouse | K I. C RLCDV. E Q ME V. IDG. T. QV R G R A T VKGG |
| Human | K I. C, RLCDV. E Q ME V. IDG. T. QV R |
| C.elegans | RE A. I. E RLCDV. EIV MT. H. V. LDG. S. VV R AQ. R A T VKGG |
| Ŭ | :** :*:: ***** : ::* ::**: *:*,** : :*,*:******* *:**.***** |
| C. parvum | DDTRMEENELVATETFATTGRGYVTEGLDCSHYMKYYDNPFLNENSTRLNSAKTLLGGTNTHEGKLAFCR |
| D. melanogaster | ES. M. D. F. GS. L. HDDM. NF. LFVPL. Q. S. Q. T. KN. T. K |
| A. thaliana | EQ. KM., G. F.,, GS., K., REDLE,, NF. AGHVPL, PR., Q., AT., KN. ST., R |
| Mouse | EA. M. G. V GS. K HDDME NF. VGHVPI. PRT. H. NV. EN. T R |
| Human | EA. M. G. V GS. K HDDME NF. VGHVP I PRT. H NV EN T R |
| C. elegans | EQ. KM |
| | : *:***.* :******.:**:* * . : *****: : ** :* ** *: :*****: |
| C. parvum | RWLDQLGFNKHALALKSLVDSEIIRPYPPLNDIPGSFSSQMEHY |
| D.melanogaster | RA. AT. YQMD. C. KG. VEA C K. CYTA. Y T 56% |
| A.thaliana | . Y RI. ET. YLM N. C. SG. VQ C. VK YV Y T 57% |
| Mouse | R. ES. YLM N. C. LG. VD C K YTA. F T 56% |
| Human | R. ES. YLM N. C. LG. VD C K YTA F T 56% |
| C.elegans | I. R ET. YLM D. C. KG. VD , C. VK. CYTA. F T 52% |
| - | *::*: * *: :***.* * *: **** *: *.: :* ** |

Figure 4. Alignment of the deduced amino acid sequence of methionine aminopeptidase of *C. parvum* and the sequence of *D. melanogaster*, *A. thaliana*, human, mouse and *C. elegans*. Filled circles in the alignment indicate identical base pair and hyphens indicate gaps. The identities of sequence are indicated at the ends of sequences.

| C.parvum | PIQFCLSAPKTDIENNIVVKDYTAMDRLLREERLLIAKMIKQIAATGCNVLLIQKSILREAISTLALDYCAKAKILVVKDIER | DEIEFLS |
|---------------|---|---------|
| A.thaliana | VQI. PQSSQI. KNY. LG. I. K. KD. VTD. S. H. L M. I V. | VT |
| D.melanogaste | er L I MDH. VI. S. A V. K SY. LNIV KKS V D. V. D QHFLD. I. CM V | EDVC |
| mouse | L MD. Q S AQ V AY. LNLV KK D. L. D HFLN. M M V 1 | EDIC |
| human | L MD. Q S. AQ. V AY. LNLV. KK D. L. D HFLN. M. M. I | EDIC |
| C.elegans | LQI.PMQVIITAQA.KQYLLEICK.AD.VNEHFLMMCI | EDY. |
| | *** | **::*** |
| | | |
| C.parvum | KALNCSPIASLDHFTSDKLGAANRVSDEDLDGNGRICRITGIPGKD-MMIIFVRASNMLMLDETERCIHDALCVVRSII | |
| A.thaliana | . T L NIE RAE H. DL. EEAS. GDG-K. LK KDMGRTTSVL G Q. V A SL CLV | 61% |
| D.melanogaste | er . T. H. R | 54% |
| mouse | . TIGTK. V. HI. Q A. M S. ELAEEVS. N. SGKLFK CTSPGKTVT. V G K. VIE. A S I. CLV | 56% |
| human | . TIGTK. V. HI. Q A. M S. ELAEEVN. N. SGKLLK CASPGKTVT. V G K. VIE. A S I. CLV | 56% |
| C.elegans | RI. GCR. V V NA. A Y. DL. EEIPTG. DGKVIKV VQNPGHAVS. LL. G K. V. E. AD. S I. CLV | 54% |
| | : : *:* :::* :: *. *:: :. ::** :. *.** *:::*: *.:******:*.:: | |

Figure 5. Alignment of the deduced amino acid sequence of chaperonin containing TCP-1 delta subunit of *C. parvum* and the sequence of *D. melanogaster*, *A. thaliana*, human, mouse and *C. elegans*. Filled circles in the alignment indicate identical base pairs and hyphens indicate gaps. The identities of sequence are indicated in the ends of sequences.

genotype II. More important is the fact that the 3 primers amplified only *C. parvum* DNA with high sensitivity, making these candidate primers to detect and genotype *C. parvum* in water contamination at the required quality in terms of sensitivity. Using our method, even low numbers of oocysts in sample water may be amplified with any one of the primers. RFLP analysis with a combination of any one of the three genes can identify animal or human genotypes. Such examples are given by primer SB281/Hind III, SB281/ Hinf I, SB289/Sac I or SB012/Ava II.

We have reported that *C. parvum* specific primer SB012 is highly sensitive. The primer SB012 can detect the lowest amount of template DNA necessary for a positive reaction, which was 0.156 pg DNA or even one oocyst, or 50



Figure 6. PCR-RFLP analysis of SB281, SB289 and SB012 genes by the restriction of SB281 with Hind III (panel A) and Hinf I (panel B), SB289 with Sac I (panel C), and SB012 with Ava II (panel D). The isolates from Japanese calf (lanes 1-3), Italy calf (lane 4 and 5), Italy goat (lane 6) and one Japanese human (HJ1 in lane 7) give the same kind of band pattern of genotype. The isolates from the other Japanese human (HJ2 and HJ3) in lanes 8 and 9, Italy human (lane 10 and 11) and Nepal human (lanes 12 and 13) show the same kind of band pattern of genotype I. M is Base-Pair Ladder of molecular weight marker.

oocysts in raw water (Wu *et al.*, 2000). The new primers (SB281 and SB289) had equivalent detection sensitivity to primer SB012. PCR detection indicates that these primers have a sensitivity 50 times higher than that of some other primers developed from COWP, HSP70 and TRAP1 and currently being used for genotyping *Cryptosporidium*.

Thus the analysis of the three RAPD genes provided the same genetic characterization to group the isolates to genotype I and genotype II, further evidence for the distinction between the two genotypes of *C. parvum*. Our new primers may also provide selectable methods for reliable elimination of *C. parvum* from tap water and for genotyping of the contaminated oocysts for further epidemiological survey.

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Figure 7. The sensitivity of constructed primer pair SB281, SB289 and SB012 by detecting template DNA and a comparison with other primers. M is 100 Base-Pair Ladder of molecular weight marker; lanes 1 to 8 are DNA amount of 160, 40, 10, 2.5, 0.625, 0.156, 0.039 and 0.001 pg, respectively. Panels A to F are primer SB281, SB289, SB012, COWP, HSP70 and TRAP1, respectively.

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IN VIVO EVALUATION OF COMBINATION EFFECTS OF CHLOROQUINE WITH CEPHARANTHIN[®] OR MINOCYCLINE HYDROCHLORIDE AGAINST BLOOD-INDUCED CHLOROQUINE-RESISTANT *PLASMODIUM BERGHEI* NK 65 INFECTIONS

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Abstract: The combination effects of chloroquine with Cepharanthin® or minocycline hydrochloride were evaluated against a blood-induced infection with chloroquine-resistant P. berghei NK 65 in ICR mice. The infected mice in an untreated control group showed a progressively increasing parasitemia leading to mouse death. A twoday dosage of 20 mg base/kg of chloroquine alone produced little effect against P. berghei NK 65 infection, and all mice died from day 13 to 15 with an increasing parasitemia. A four-day dosage of 4 mg/kg of Cepharanthin[®] alone produced no antimalarial activity, and all mice died by day 10. A four-day dosage of 50 mg/kg of minocycline hydrochloride alone produced a slight effect, but all mice died by day 18. Furthermore, mice given chloroquine in combination with Cepharanthin[®] died from day 14 to 15. Mice given Cepharanthin[®] plus minocycline hydrochloride also died from day 15 to 17. On the other hand, infected mice treated with chloroquine plus minocycline hydrochloride survived during the experiment. All mice treated with chloroquine alone, minocycline hydrochloride alone, chloroquine plus Cepharanthin® or Cepharanthin® plus minocycline hydrochloride showed low parasitemia levels during drug administration and a few subsequent days, but then malaria parasites re-increased in the bloodstream of the treated mice until death. On the other hand, malaria parasites in the mice given chloroquine plus minocycline hydrochloride decreased on day 6 and then could not be detected by microscopic examination during the observation period. This finding strongly suggests that the combination effects of chloroquine and minocycline hydrochloride are worthy of evaluation in human malaria. The results also clearly demonstrate the necessity and importance of in vivo experiments in estimating the activities of drugs.

Key words: Plasmodium berghei NK 65, Cepharanthin®, minocycline, antimalarial activity, chloroquine-resistance

INTRODUCTION

Malaria is one of the most important tropical diseases in the world. Chloroquine resistance in the human malaria parasite *Plasmodium falciparum* arose first in South America and Southeast Asia (Harinasuta *et al.*, 1962; Young and Moore, 1961). It has now spread to all parts of the world where malaria is endemic and poses a major threat to the elimination of the disease. Although there are several strategies for controlling the disease, chemotherapy is the primary defense against malaria, and thus the worldwide emergence of chloroquine-resistant variants of parasite has stimulated the development of new treatments for malaria (Payne, 1987; Winstanley, 2000). The development of a new drug, however, is an extremely expensive and timeconsuming process, and hence there have been efforts to reevaluate the antimalarial activities of certain drugs already accepted for clinical use in patients with various nonmalarial infectious diseases. White and Olliaro (1996) recently advocated the utility of combination chemotherapy as a rational approach to the containment of drug-resistant malaria. These trials focused on the detection of compounds that display killing activity against multiple drugresistant malaria parasites in vitro (Haruki *et al.*, 2000; Lin *et al.*, 2001). However, the question remains as to which in vitro antimalarial effects can be induced in vivo. Since the discovery of murine malaria parasite *P. berghei* by Vincke and Lips (1948), the model of experimental murine malaria

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has proved to be very convenient both for detection of antimalarial activity (Peters et al., 1975) and investigations into the mode of action with P. falciparum in culture. Furthermore, the mouse infected with P. berghei is generally considered to be a valid model for the primary screening of drugs for eventual use against human malaria (WHO, 1973). In 1971, attempts to confirm the antimalarial activity of Artemisia annua extracts in mice infected with P. berghei led to the isolation of a plant constituent, artemisinin (Klayman, 1985). The use of drug-resistant strains of P. berghei can yield additional information concerning both the mode of action of a compound, and its potential value against drug-resistant strains of human malaria. Thus, we utilized the murine model to investigate the effects of chloroquine - in combination with drugs known to reverse chloroquine resistance in vitro (Haruki et al., 2000; Lin et al., 2001) - against blood-induced chloroquine-resistant P. berghei NK 65 infections in ICR mice.

MATERIALS AND METHODS

Animals and parasites

All animal experiments were performed according to the Guidelines for Animal Experimentation, Hamamatsu University School of Medicine. Outbred male ICR mice, 7 weeks old, purchased from SLC Inc. (Hamamatsu, Japan), were used. Murine malaria parasites, chloroquine-resistant Plasmodium berghei (strain NK 65), were a gift from Professor Y. Wataya (Okayama University, Japan). The blood stage parasites were stored in a - 80 € deep freezer. For experiments, the parasites from frozen stock were injected into two mice. The mouse showing 10-15% of parasitemia was bled under ether anaesthesia to collect the parasitized blood. Experimental mice were given an intraperitoneal injection of the 10⁵ parasitized blood. To determine the effect of the drug, treated mice were monitored for % parasitemia and days of survival relative to control mice up to day 30 post infection. Thin blood smears from the tail vein were prepared and Giemsa-stained throughout the observation period.

In vivo antimalarial activity of chloroquine against chloroquine-resistant P. berghei NK 65 infections

Twenty mice were infected intraperitoneally with 10⁵ parasitized erythrocytes and divided into four per group, as well as an untreated control group, for activity assay of chloroquine diphosphate (Sigma Chemical Co., MO, USA). From day 4 after injection, the mice were given chloroquine orally at 20 mg base/kg body weight (kg) for 2 or 3 days, or 40 mg base/kg for 1 or 2 days, respectively, in the treated groups. An equivalent volume of distilled water was given orally to mice in the untreated, infected group for 3 days.

In vivo antimalarial activity of Cepharanthin[®] and minocycline hydrochloride against chloroquine-resistant P. berghei NK 65 infections

Twenty-eight mice were infected intraperitoneally with 10⁵ parasitized erythrocytes and divided into four per group, including an untreated control group, for activity assay of Cepharanthin[®] (an extract of *Stephania cepharantha*) and minocycline hydrochloride. Cepharanthin[®] and minocycline hydrochloride were purchased from Kaken-Shoyaku Co. (Tokyo, Japan) and Takeda Chemical Industries Ltd. (Osaka, Japan), respectively. From day 4 after injection, the mice were given Cepharanthin[®] at 0.2, 2 or 4 mg/kg intraperitoneally once a day for 4 consecutive days. Mice in the other three groups were given minocycline hydrochloride at 2.5, 25 or 50mg/kg intraperitoneally once a day for 4 consecutive days from day 4 after injection. An equivalent volume of sterile normal saline was given intraperitoneally to mice in the untreated control group.

Combination effects of the compounds against chloroquineresistant P. berghei NK 65 infections

Twenty-eight mice were infected intraperitoneally with 10⁵ parasitized erythrocytes and divided into four per group, including an untreated control group, for activity assay of the compounds in combination with or without chloroquine. From day 4 after injection, the mice in three groups were given chloroquine orally at 20 mg base/kg once a day for 2 days, and furthermore, to detect the activity of Cepharanthin[®] and minocycline hydrochloride in combination with chloroquine, the mice in two out of three of the groups treated with chloroquine received Cepharanthin[®] at 4 mg/kg intraperitoneally once a day for 4 consecutive days and minocycline hydrochloride at 50 mg/kg intraperitoneally once a day for 4 consecutive days. Furthermore, the mice in the other three groups were given Cepharanthin[®] at 4 mg/kg, minocycline hydrochloride at 50 mg/kg, or both drugs simultaneously, intraperitoneally once a day for 4 consecutive days. An equivalent volume of water was given orally to mice in the untreated, infected group for 4 consecutive days.

RESULTS

In vivo antimalarial activity of chloroquine against chloroquine-resistant P. berghei NK 65 infections

As shown in Fig. 1A, mice in the untreated control group died from day 9 to day 11 post infection after a gradual body weight loss. Chloroquine treatment was somewhat effective against the parasites but it did not entirely eradicate them. All mice in the groups given the drug died after a gradual body weight loss as follows: mice given a twoday dosage of 20 mg base/kg died from day 12 to day 14, those given a three-day dosage of 20 mg base/kg died from



Fig. 1. Time-course changes of survival rate (A) and parasitemia in the bloodstream (B) of mice in response to treatment with chloroquine. From day 4 after injection of $10^5 P$. *berghei* NK 65-parasitized erythrocytes, mice were given chloroquine orally at 20 mg base/kg b.wt. for 2 days () or 3 days (), or 40 mg base/kg b.wt. for 1 day () or 2 days () in the treated group. An equivalent volume of distilled water was given orally in the untreated, infected mice () for 3 days.

day 15 to day 16, those given a one-day dosage of 40 mg base/kg died from day 10 to day 13, and those given a twoday dosage of 40 mg base/kg died from day 12 to day 17. Malaria parasites appeared from day 4 post infection in the bloodstream of the control and treated groups (Fig. 1B). Parasitemia levels of mice in the control group gradually increased and all of the mice died by day 11. All of the mice treated with chloroquine showed low parasitemia levels during drug administration and the following few days. Especially, malaria parasites in the mice given a two-day dosage of 40 mg base/kg of chloroquine decreased on day 5 and then could not be detected by microscopic examination for two days. Malaria parasites, however, rebounded in the bloodstream of all treated mice and all eventually died.

In vivo antimalarial activity of Cepharanthin[®] and minocycline hydrochloride against chloroquine-resistant P. berghei NK 65 infections

Mice in the untreated control group died from day 8 to 10 post infection, showing a progressively increasing parasitemia (Figs. 2A and 2B). Four daily doses of 0.2, 2 or 4 mg/kg of Cepharanthin[®] from day 4 after parasite injection produced no antimalarial activity and all of the mice died by day 10, showing a pattern of parasitemia similar to that in the untreated control group (data not shown). A four-day dosage of 2.5 mg/kg of minocycline hydrochloride produced no effect, and all of the mice died by day 10. On the other hand, four daily doses of 25 or 50 mg/kg of minocycline hydrochloride produced a slight effect, but all of the mice died by day 17. Malaria parasites in the mice given a four-day dosage of 2.5 mg/kg of minocycline hydrochloride gradually increased until the mice died. A four-day dosage of 25 or 50 mg/kg of minocycline hydrochloride suppressed



Fig. 2. Time-course changes of survival rate (A) and parasitemia in the bloodstream (B) of mice in response to treatment with minocycline hydrochloride. From day 4 after injection of 10^5 *P. berghei* NK 65-parasitized erythrocytes, mice were given minocycline hydrochloride intraperitoneally at a dose of 2.5 (), 25 () or 50 () mg/kg b.wt. once a day for 4consecutive days. An equivalent volume of sterile normal saline was given intraperitoneally in the untreated, infected group () for 4 consecutive days.

the multiplication of parasites during drug administration and the following few days. Especially, malaria parasites in the mice given 50 mg/kg of minocycline hydrochloride decreased on day 6 but rebounded from day 10 in the bloodstream of all treated mice until death.

Combination effects of the compounds against chloroquineresistant P. berghei NK 65 infections

Mice in the untreated control group died from day 9 to 11 post infection (Fig. 3A). A two-day dosage of 20 mg base/kg of chloroquine alone produced little effect against P. berghei NK 65 infection, and all of the mice died from day 13 to 15. Four daily doses of 4 mg/kg of Cepharanthin[®] alone produced no antimalarial activity, and all of the mice died by day 10. A four-day dosage of 50 mg/kg of minocycline hydrochloride alone produced a slight effect, but all of the mice died by day 18. Mice given Cepharanthin[®] plus minocycline hydrochloride died from day 14 to 17. Mice given chloroquine plus Cepharanthin® died from day 14 to 15. On the other hand, mice given chloroquine in combination with minocycline hydrochloride survived during the experiment. Malaria parasites appeared from day 4 in the bloodstream of the control and treated groups (Fig. 3B). Parasitemia levels of mice in the control and Cepharanthin[®] -treated groups gradually increased and all of the mice died. All mice treated with chloroquine alone, minocycline hydrochloride alone, Cepharanthin[®] plus minocycline hydrochloride and chloroquine plus Cepharanthin[®] showed low parasitemia levels during drug administration and the following few days. However, malaria parasites rebounded in the bloodstream of the treated mice until death. On the other hand, parasites in the mice treated with a combination



Fig. 3. Time-course changes of survival rate (A) and parasitemia in the bloodstream (B) of mice in response to the combined treatment. From day 4 after injection of 10⁵ P. berghei NK 65-parasitized erythrocytes, mice in three groups were given chloroquine orally at a dose of 20 mg base/kg b.wt. once a day for 2 days, and then, except for mice in the chloroquinemedicated control group (), the remaining mice were further given the following drugs: Cepharanthin[®] at a dose of 4 mg/kg b.wt. intraperitoneally once a day for 4 consecutive days () or minocycline hydrochloride at a dose of 50 mg/kg b.wt. intraperitoneally once a day for 4 consecutive days (). Additionally, Cepharanthin[®] alone (), minocycline hydrochloride alone () or Cepharanthin $^{\mathbb{R}}$ plus minocycline hydrochloride () was given to the mice in the three other groups under the same treatment regimens. An equivalent volume of distilled water was given orally to the mice in the untreated, infected group () for 4 consecutive days.

of minocycline hydrochloride and chloroquine decreased on day 6 and then could not be detected by microscopic examination during the observation period.

DISCUSSION

Recently, drugs enhancing the sensitivity of parasites to chloroquine were studied in resistant strains of rodent and human *Plasmodium* (Martin *et al.*, 1987; Bitonti and McCann, 1989; Peters *et al.*, 1990; Kyle *et al.*, 1993). In the present experiment, *P. berghei* NK 65 showed a chloroquine-resistant property, and hence this variant can be a useful model for research on drugs that enhance the sensitivity of parasites to chloroquine (Ishih *et al.*, 2003).

Lin *et al.* (2001) re-evaluated the antimalarial effects of three tetracyclines accepted for clinical use in patients with various non-malarial infectious diseases, and they reported that minocycline was effective against chloroquineresistant *P. falciparum* in vitro. Their electron microscopic examination revealed a number of electron dense vesicles with a single membrane bound in the cytoplasm of minocycline-treated parasites (Lin *et al.*, 2001). The antiparasite action of minocycline in vitro studies may be attributable to the high lipophilicity of this drug, but it is difficult to explain whether the structural changes resulted from the specific action mechanism of tetracyclines. In our P. berghei NK 65 infected ICR model, all mice given minocycline hydrochloride alone showed low parasitemia levels during drug administration and the following few days, indicating the inhibitory effects of minocycline on parasite growth, but they died after re-increase of malaria parasites. In contrast, the combination of chloroquine with minocycline hydrochloride eventually produced a killing activity against P. berghei NK 65 parasites, and consequently, all mice survived during the experiment. Kim et al. (1998) evaluated a combination effect of 5-fluoroorotate and sulfamonomethoxine using this malaria isolate and reported a potent synergistic activity. By definition, chloroquine resistance reversal agents specifically target the chloroquine resistance mechanism and do not enhance the baseline activity of chloroquine against parasites (Martin et al., 1987; Bitonti and McCann, 1989). In the present study, both chloroquine and minocycline hydrochloride showed moderate antimalarial activity against the present parasite isolate when given alone to the infected mice, and thus it remains unclear which drug enhanced the antimalarial activity of the other. This finding, however, strongly suggests that the combination of chloroquine and minocycline hydrochloride extends the clinical utility of chloroquine if the higher dose of minocycline hydrochloride is tolerated. Further studies are necessary to clarify the mechanisms of combination effects of these drugs.

Meanwhile, Haruki et al. (2000) reported that Cepharanthin[®], widely used clinically in Japan, enhanced the activity of chloroquine against the resistant strain of P. falciparum in vitro. The mechanism by which Cepharanthin[®] increases the potency of chloroquine is unclear, but it may be related to enhanced accumulation of chloroquine. In our model, however, Cepharanthin[®] given approximately 20 times in clinical use (Harada et al., 2001) produced neither combination effects with chloroquine nor its intrinsic antimalarial activity, suggesting the difference between in vitro and in vivo assay. The present results indicate that differences in assay systems, such as in vitro and in vivo methods, might lead to different and conflicting results for the antimalarial activities of drugs, and they indicate the necessity for and importance of in vivo experiments in the estimation of the activities of drugs.

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INCIDENCE OF TRAVELERS' DIARRHEA AMONG JAPANESE VISITING THAILAND

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Abstract: A cross-sectional survey of 327 Japanese short-term travelers (\leq 3 weeks) arriving in Bangkok, Thailand was conducted to assess the incidence of travelers' diarrhea (TD) as well as their symptoms and treatment-seeking behaviors. The incidence of the first episode of TD (FTD) was ascertained retrospectively by questionnaire. Reported by 69 travelers, FTD clustered within the first 8 days of arrival in Thailand, and the incidence rate varied from 2% to 8% with the highest incidence on the third day.

Cumulative probability of FTD was 19% for those arriving in Thai directly from Japan, 42 % for those arriving via Southeast Asia, and 25% for those arriving via other regions at Day 7 by the Kaplan-Meier survival analysis. Log rank test revealed a higher FTD risk for travelers arriving via other Southeast Asian countries than for those arriving directly from Japan (P < 0.005). Of all the 69 FTD episodes, 33% had classic TD defined as ≥ 3 unformed stools per 24 hours with at least one accompanying symptom, 49% had moderate TD defined as ≤ 2 unformed stools with at least one additional symptom or more unformed stools without additional symptoms, and 17% had mild TD defined as with ≤ 2 unformed stools without additional symptoms. Cumulative probability of FTD at Day 7 was 12% for classic TD, 25% for classic plus moderate TD and 30% for all the TD. More than 38% of travelers with diarrhea took medicine brought from Japan. Among travelers with classic TD, 35% bought medicine in Thailand, whereas 47-50% of travelers with moderate and mild TD took only rest without any treatment. **Key words:** incidence, diarrhea, travelers, Japanese, Thailand,.

INTRODUCTION

Travelers' diarrhea (TD) is a common health problem among travelers. Annually, over 50 million people from industrialized countries visit developing countries. Ten to 80 percent of travelers suffer from diarrhea (Black, 1990; Cobelens *et al.*, 1998; Steffen, 1986; von Sonnenburg *et al.*, 2000). Although TD is a self-limiting illness that usually resolves spontaneously within a few days, travelers often have to change their itinerary because of it (Hill, 2000; Steffen et al., 1999).

Annually, 3 million Japanese people travel to Southeast Asia (Statistics on Legal Migrants, Ministry of Justice in Japan, 2001). Enteropathogens causing diarrhea were isolated from a high proportion (65.3%) of Japanese travelers who arrived from Southeast Asia and had suffered from TD (Ueda *et al.*, 1996). There is a risk of spread of imported causative agents in homeland countries (Wittlinger *et* *al.*, 1995). Nevertheless, the TD of Japanese travelers has never been studied in Southeast Asia before, although TD was studied among United States troops and US Peace Corps volunteers in Thailand (Beecham *et al.*, 1997; Echeverria *et al.*, 1981; Taylor *et al.*, 1985). Epidemiological data on Japanese TD in Southeast Asia will lead to valuable advice for Japanese travelers regarding prevention and treatment.

We therefore studied the incidence of TD as well as their symptoms and treatment-seeking behaviors for TD by a cross-sectional survey in Japanese travelers on adventurestyle trips (hereafter, backpackers) arriving in Bangkok, Thailand. Here we report on the results in Thailand, a country visited by one million Japanese annually (Statistics on Legal Migrants, Ministry of Justice in Japan, 2001).

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MATERIALS AND METHODS

This study was conducted between January and February 2002 in the Khaosan Road area, Bangkok, Thailand. The subjects were Japanese travelers who were more than 15 years old and stayed in Thailand for 3 weeks or less. Three hundred and eighty four Japanese travelers participated in this study with informed consent. The study was conducted by a self-administered questionnaire. Of the 384 subjects, 57 (15%) who had stayed more than 3 weeks in Thailand before the interview were excluded to reduce recall bias. Thus, 327 Japanese travelers were used for the analysis.

As in previous studies (Steffen *et al.*, 1999; von Sonnenburg et al., 2000), classic TD was defined as the passage of three or more unformed stools per 24 hours with as least one accompanying symptom such as nausea or vomiting, abdominal pain, fever, or blood in stools. Moderate TD was defined as either the passage of one to two unformed stools with at least one additional symptom, or more unformed stools without additional symptoms. Mild TD was defined as the passage of one to two unformed stools without additional symptoms.

Daily incidence rates (IRs) of having the first TD episode (FTD) on Day N were calculated by the following formula:

 $IR(n) = \frac{100 \text{ x Number of travelers who had FTD on Day N}}{\text{Number of travelers who stayed on Day N and who}}$ had never had FTD until Day N-1

The IRs of FTD were presented as simple proportions with 95% confidence interval (95% Cl). Data management was performed with Microsoft Excel (Version 8.0). Cumulative probability of FTD and difference of its risk by preregion was calculated by the Kaplan-Meier procedure and log rank test, respectively. For comparison of symptoms and treatment-seeking behaviors for TD, χ^2 -test was conducted using the Epi Info 6 programme.

The data were also analyzed by region visited prior to arrival in Thailand; i.e., a) directly from Japan, b) via other countries in Southeast Asia, and c) via other countries outside Southeast Asia.

RESULTS

Characteristics of Japanese travelers

The characteristics of the 327 Japanese travelers are shown in Table 1. Men accounted for 66% of the subjects. Mean and median age was 27 and 25 years old, respectively, with 85% of subjects being less than 30 years old. Most travelers were single (91%), and high school graduates

Table 1 Characteristics of 327 Japanese travelers to Bangkok, Thailand

| Characteristic | N (%) | | |
|-------------------------------------|----------------------|--|--|
| Sex, male | 215 (65.7) | | |
| Age* (year) | 26.8 (25/19-66) | | |
| Marital status, single | 298 (91.1) | | |
| Educational level | | | |
| \geq High school | 318 (97.3) | | |
| Occupation | | | |
| Unemployed/Retired | 124 (37.9) | | |
| Student | 104 (31.8) | | |
| Worker | 84 (25.6) | | |
| Housewife | 6 (1.8) | | |
| Others | 9 (2.8) | | |
| Travel with companions | 154 (47.1) | | |
| First visit to Thailand | 116 (35.5) | | |
| Purpose of travel, sightseeing | 300 (91.7) | | |
| Travel style, self-arranged journey | 310 (94.8) | | |
| Pre-regions prior to Thailand | | | |
| Japan | 136 (41.6) | | |
| Southeast Asia | 117 (35.8) | | |
| Other regions | 74 (22.6) | | |
| Duration of stay* (day) | 6.2 (4/1-21) | | |
| Total expenditure per day † (US\$) | 17.9 (9.1/2.3-204.5) | | |
| | | | |

*Mean (median/range). † Mean (median/range), thirteen missing.

(97%). Unemployed /retired persons accounted for 38%, followed by students (32%) and workers (24%). Fifty-three percent of travelers traveled alone, and 35% were first visitors to Thailand. Most travelers visited Thailand for sight-seeing (92%), and arranged their trip by themselves (95%). Forty-two percent arrived directly from Japan while 36% came to Thailand via other countries in Southeast Asia like Vietnam, Cambodia, and Laos. Others came from Nepal, India, Australia, the U.S.A. and so on. Mean and median days of stay in Thailand up to the day of interview were 6.2 and 4.0, respectively. Their mean daily expenditure was 17.9 US dollars with median of 9.1 US dollars.



*Bars present 95% confidence interval for each incidence of FTD Figure 1 Daily incidence of first episode of travelers' diarrhea (FTD) and number of subjects at risk

Number of subjects at FTD risk and incidence of FTD

The number of subjects at risk for FTD was 327 on the first day of stay and decreased to 88 on the seventh day and 28 on the 14th day (Figure 1). Sixty-nine travelers developed diarrhea. Of them, 33% (23 travelers), 49%, (34 travelers) and 17% (12 travelers) had classic TD, moderate TD and mild TD, respectively. Most FTD (91%, 63 travelers) clustered within the first 8 days after arrival to Thailand. The incidence rates during these eight days ranged between 2.7 % and 7.5% with the highest incidence on the third day. Occurrences of FTD were sporadic between Days 9 and 15. No FTD was reported after Day 16.

Cumulative probability of FTD

Survival probability without experiencing FTD up to Day N was calculated by the Kaplan-Meier survival analysis. Then, the cumulative probability of FTD to Day N (%) was calculated by subtracting the above from 1.0. Figure 2 shows the cumulative probabilities of FTD by pre-region (a. directly from Japan, b. via other countries in Southeast Asia, and c. via other countries outside Southeast Asia). The cumulative probability of FTD at Day 7 by pre-regions was 19% for those arriving directly from Japan, 42% for those arriving directly from Southeast Asian countries, and 25% for those arriving from other regions. The cumulative probability of FTD in travelers via Southeast Asia was more than twice as that in those from Japan. The cumulative probability of FTD for those arriving from Japan, Southeast Asian countries and other regions reached 26%, 47%, and 32%, respectively, by Day 14. According to a log-rank statistic test, travelers who arrived in Thailand via other Southeast Asian countries developed FTD significantly more frequently than those arriving directly from Japan (P < 0.005) and showed a marginally higher incidence of TD than those arriving via other regions (P < 0.1).



Figure 2 Cumulative probability of FTD by pre-region among Japanese travelers in Bangkok, Thailand



Figure 3 Cumulative probability of FTD due to classification of FTD among Japanese travelers in Bangkok, Thailand

Figure 3 shows the cumulative probability of classic TD, moderate TD, and mild TD. Cumulative probability of FTD at Day 7 was 12% for classic TD, 25% for classic plus moderate TD, and 29% for all forms of TD including mild cases. Cumulative probabilities reached to 13%, 30%, and 35%, respectively, by Day 14.

Cumulative probability of classic plus moderate TD at Day 7 was the highest among travelers from Nepal (56%), followed by travelers from Laos (42%), Vietnam (39%), and Cambodia (38%), while it was only 12% among travelers who arrived directly from Japan.

Symptoms and treatment-seeking behaviors among 69 travelers with TD

The mean daily number of stools and its standard deviation was 4.4 ± 1.9 for classic TD, 2.8 ± 2.2 for moderate TD and 2.0 ± 1.4 for mild TD (Table 2). Abdominal pain was reported in 91% of classic TD, a rate significantly

| | Table 2 | Symptoms ar | d reactions among | g 69 trave | lers with T |
|--|---------|-------------|-------------------|------------|-------------|
|--|---------|-------------|-------------------|------------|-------------|

| ¥7 · 11 | Classic | Moderate | Mild |
|--|---------------|-------------|---------------|
| Variable | (n=23) | (n=34) | (n=12) |
| Sex: no. male/no. female | 17/6 | 19/15 | 9/3 |
| Mean frequency of unformed | | | |
| stools (\pm SD) of diarrhea | 4.4 ± 1.9 | 2.8 ± 2.2 | 2.0 ± 1.4 |
| Symptoms: % of patients | | | |
| Fever | 13.0 | 8.8 | |
| Nausea/Vomiting | 26.1 | 14.7 | |
| Abdominal pain | 91.3* | 44.1 | |
| Bloody stool | 13.0 | 2.9 | |
| Watery stool | 56.5 | 20.6 | 50.0 |
| Reactions for TD: % of patients | | | |
| Taking medicine carried from homeland | 47.8 | 38.2 | 41.7 |
| Buying medicine in Thailand | 34.8* | 8.8 | 8.3 |
| Taking ORS (oral rehydration salt) | 21.7 | 8.8 | |
| Visiting clinic | 13.0 | 8.8 | |
| Taking only rest without any treatment | 21.7 | 47.1 | 50.0 |

*P < 0.05 compared with Moderate TD.

higher than in cases of moderate TD (44%, P < 0.05). Watery diarrhea was documented in 57% of classic TD, 21% of moderate TD and in 50% of mild TD. Nausea or vomiting was documented in 26% of classic TD and 15% of moderate TD. Fever was reported in 13% of patients with classic TD and in 9% of those with moderate TD. Bloody diarrhea was documented in 13% of patients with classic TD but in only 3% of those with moderate TD.

Table 2 also shows the treatment-seeking behaviors of patients with TD. Regardless of severity, 38% to 48% of patients took medicine that they brought from Japan. Patients with classic TD bought medicines in Thailand significantly more frequently than those with moderate TD (P < 0.05). the proportion of those having taken ORS was also higher in classic TD than in moderate TD. The proportion of those taking only rest without any treatment tended to be less in classic TD (22%) than in moderate (47%) or mild TD (50%). Six travelers who suffered from TD visited clinics. Of them, 2 travelers visited clinics without taking any medicine from Japan. Two travelers visited a clinic after taking medicines brought from Japan, and the remaining two travelers bought medicines in Thailand before they went to the clinics.

DISCUSSION

TD generally resolves within a few days, but 10-20% of cases last for more than 7 days (Cobelens *et al.*, 1998; Mattila, 1994). In the case of long-lasting or continual diarrhea, it is difficult for travelers to distinguish the first episode of TD from subsequent episodes. This study therefore focuses on the first episode of TD after entry to Thailand. Because Japanese travelers stay an average of 5.6 days in Thailand (Immigration Bureau, Police Department in Thailand, 2001), the first episode of TD is more important for Japanese travelers than subsequent episodes.

This study was conducted in the Khaosan Road area, Bangkok, Thailand. This area is a popular venue for young backpackers. Currently, many backpackers visit there from all over the world, because of its friendly atmosphere and low-cost accommodation and food. Discount air tickets and useful trip information are available in the Khaosan Road area. Backpackers are generally considered to prefer an adventurous travel style and to be more exposed to risk of diarrhea (Steffen, 1986). Self-arranged travel is also at higher risk for diarrhea than organized travel (Cobelens *et al.*, 1998). Increasing age is negatively correlated with diarrhea (Steffen, 1986; Cobelens *et al.*, 1998). Steffen *et al.* (1999) found that teenagers and young adults were at a higher risk for TD, probably due to their great appetite and consequent ingestion of a higher dose of pathogens. Since the age of the subjects in the present study clustered undter the age of 30 years, this age group is probably to be at a higher risk of acquiring TD than other older age groups.

FTD clustered within the first 8 days of arrival in Thailand in the present study (Figure 1). As in previous reports (Angust and Steffen, 1997; Steffen, 1986), TD started within the first day after entry to Thailand, and showed the highest proportion of onsets on the third day. About one in three travelers to Thailand (30%) had TD within the first week, 35% of travelers developed TD within 14 days, and 43% within 21 days (Figure 3). This increased proportion of cumulative probability of TD also shows the highest risk of FTD within the first week of stay.

Risk of TD was found to vary by season (Mattila *et al.*, 1992). Since the present study was conducted in the cooler season (January to February) in Thailand, the incidence and cumulative probability of FTD are expected to be higher in the warmer season.

Among the 136 travelers arriving in Thailand directly from Japan, 18 (13.2%) experienced TD. The cumulative probability of FTD was 19% within the first week, 26% within the two weeks, and 35% within three weeks (Figure 2). This shows that TD occurred more frequently during the first week even among those who came directly from Japan. The cumulative probability on Day 14 was similar to the incidence reported by Steffen (1986). The cumulative probability on Day 20 was also similar to the incidence of TD among American troops stationed in Thailand (Beecham *et al.*, 1997). Japanese travelers in Thailand seem to experience TD at a rate similar to that of European travelers (Steffen, 1986). Although most subjects in the present study were young backpackers, this does not seem likely to exert a significant influence on TD occurrence.

Regional variations prior to entry in to Thailand influenced TD occurrence (Figure 2). According to log-rank test, travelers who arrived Thailand via other Southeast Asian countries developed TD at a significantly higher rate than those arriving directly from Japan (P < 0.005). It is suspected that travelers arriving via Southeast Asia were exposed to more TD agents before Thailand than those arriving directly from Japan. The difference in cumulative probability of FTD between travelers arriving via Southeast Asia and from Japan may be due to the dose and type of causative agents ingested in previous regions. The difference was 23 points at Day 7, and 21 points at Day 14. The effect of pre-region on occurrence of TD was stronger during the first week. The evidence of the effect of pre-region indicates that travelers might import pathogens from surrounding countries to Thailand. This may also affect the health of the Thai people.

Pre-regions other than Southeast Asia included high-

risk destinations such as India and Nepal (Hill, 2000; von Sonnenburg *et al.*, 2000). The cumulative probability of FTD via these areas was thought to be high before the analysis. But in reality, it was similar to that from Japan. this may be due to a seasonal variation (Mattila *et al.*, 1992) and to the inclusion of developed countries such as the U.S. A. and Australia in "other pre-region". Since the January to February Period is winter in India and Nepal and the incidence of diarrhea is low (Hoge *et al.*, 1996), the effect of pre-regions might not reflect strongly on the cumulative probability of FTD in the present study.

Cobelens et al. (1998), Hill (2000) and Mattila (1994) reported that 34-80% of patients with TD had abdominal pain, 17-42% had fever, 18-45% had nausea or vomiting, 28 -69% had watery stool, and 0-5% had bloody stools. The present study showed a similar occurrence of these symptoms. Although TD is a self-limiting illness and generally resolves within a few days, the severity of TD influences the activity of travelers: 48% of patients suffering from classic TD took medicine, and 13% visited clinics. These actions of Japanese travelers are similar to the finding of Hill (2000) and Steffen et al. (1999). However, more Japanese travelers with mild TD took medicine than the travelers studied by Steffen et al. (1999; 43% vs. 5%). Japanese travelers tended to take medicine more easily, but 47-50% of travelers with moderate and mild TD took only rest without any medication. Japanese travelers are likely to consider diarrhea as slight or not severe.

A high proportion of Japanese travelers took medicine brought from Japan. However, antibiotic resistances has been found, recently among diarrheal pathogens (Hoge *et al.*, 1998; Matushita *et al.*, 2000) and some antibiotics are not effective for diarrhea. It is important for travelers to know the efficacy of the medicines they carry, and to be familiar with procedures needed to obtain safe and effective medical care.

Finally, three million Japanese travel to South Asia annually, and many of them are at high risk of acquiring TD. Nevertheless, the epidemiological study of Japanese TD has never been conducted in South Asia before. The present study provides basic epidemiological data concerning the incidence of Japanese TD and the reactions to TD. However, many factors such as various pathogens, travel destination, length of stay, food consumption and so on (Ueda *et al.*, 1996; Cobelens *et al.*, 1998; Ryan and Kain, 2000)are related to the occurrence of TD. Therefore, further investigations are needed to provide appropriate and valuable advice to Japanese travelers on how to avoid and treat TD.

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CHILD DEATH AND WOMEN'S OWN EARNINGS ARE ASSOCIATED WITH CONDOM USE IN MADAGASCAR

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Abstract: The prevalence of human immunodeficiency virus (HIV) infection is lower in Madagascar than in continental African countries, but recently it has steadily risen. To identify factors that facilitate or deter condom use among married couples in Madagascar, a cross-sectional study was conducted by means of a questionnaire survey of 977 women attending 10 health facilities for antenatal care and child care. Information on reproductive and socioeconomic factors was obtained, and logistic regression analysis was used to identify factors contributing to condom use. Child death and women's earnings were statistically significant factors for condom use, with an odds ratio of 2.0 (95% confidence interval (CI): 1.2-3.5) for the former and 1.6 (95% CI: 1.1-2.3) for the latter in the logistic regression analysis. The promotion of economic independence among women and the reduction of child mortality deserve greater attention in the planning of HIV prevention policy in Madagascar.

Key words: Madagascar, Condom use, Child health, Women's socioeconomic status.

INTRODUCTION

In the face of a massive epidemic of human immunodeficiency virus (HIV) infection in Africa, Madagascar retains a very low prevalence in comparison with countries on the African continent. However, the estimated prevalence among adults in Madagascar has steadily risen, showing a ten-fold increase in 6 years from 0.03% in 1992 to 0.3% in 1998 (UNAIDS/WHO 2002). This increase seems to have accelerated in recent years based on the seropositivity of 1.0% in 1999 (Ministère de la Santé, Madagascar, 1999). Ravaoarimalala *et al.* (1998) predict that HIV seroprevalence among adults will reach 3% in the low scenario or 15% in the high scenario by 2015.

Heterosexual contact has been the predominant mode of transmission in Africa, making women vulnerable to the infection from the early stage (Piot P 1984, Nzilambi N 1988). More than half of HIV-positive cases in Africa are women, and females aged 15-19 years have a much higher infection rate than their male counterparts (UNAIDS/WHO 2002). These facts reflect both the socioeconomic situation of women and their biologically higher risk of becoming infected: 8 times greater than in men (Padian N 1997).

Women in Madagascar seem to be in a similar situation, and they are under the threat of a surge in the HIV epidemic. Risk factors for its spread also abound in this country: a high prevalence of sexually transmitted infections (Harms G 1994, Behets FM 1996), early sexual debut among youths, increased prostitution related to poverty, increasing tourism and contact with people from countries with high HIV prevalence such as India and the African Continent.

Thus, Madagascar is at a juncture in the effort to prevent and control the HIV epidemic. Condom use is currently one of the major strategies in the prevention and the control of HIV (Population Reports 1999). However, because they lack economic resources of their own and fear of abandonment or violence by their partners (UNAIDS 1997), women at risk seem to have little power in using condoms and otherwise controlling sexual activity. The purpose of this study is to identify factors that facilitate and deter condom use in Madagascar. The information obtained and presented in this paper should be instrumental in the planning and implementation of activities to prevent and control HIV infection in places where the prevalence is still relatively low but timely action is urgently needed.

METHODS

A questionnaire survey was conducted in Mahajanga District I, which has a population of 230,000. Mahajanga District I includes Mahajanga City, a port on Mozambique

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Strait. The directors of the 19 health facilities in the area were approached with a request to cooperate in this study. Facilities that specialized in sexually transmitted infections were excluded. Ten health facilities agreed to cooperate: 2 in semi-urban areas and 8 in the urban area.

Questionnaire sheets were handed to all the currently married women who visited the clinics for antenatal consultation or care of their sick children during a four-week period in February 2000. All the women consented, signed the form of consent to participate in the study, and responded to the questionnaire. Registered prostitutes were excluded. When the number of study subjects reached 100, recruitment at that clinic was terminated. The questionnaire was in French, and those who could read it (about one tenth of the study subjects) were asked to fill in the questionnaire themselves. For women who could not read it, a facility staff member read the questionnaire aloud and obtained their answers. The questionnaire consisted of the following items: age, years of school attendance, number of past pregnancies, live births and stillbirths, deaths and causes of death of children, presence of symptoms suggestive of sexually transmitted infections, personal earnings, and condom use by their partners.

The information obtained was analyzed using PC-SAS (SAS Institute Inc. 1996). Logistic regression analysis was used to assess factors related to condom use.

RESULTS

Replies to the questionnaire were obtained from a total of 977 subjects, and their characteristics and replies are shown in Table 1. The subjects ranged in age from 13 to 57 years, the mean and standard deviation being 28.7 and 7.4 years, respectively. The range of the number of past pregnancies, fetal death and child death were 0-17, 0-9 and 0-9 with mean (standard deviation) 3.8 (2.7), 0.6 (1.0) and 0.4 (0.8), respectively. Three hundred and fifty (35.8%) of the 977 subjects showed STI symptoms, the most common being vaginal discharge (24.0%), genital pain (10.4%), and genital ganglion (1.4%). Regarding condom use, 701 (71.8%) of the 977 responded that condoms were never used, 131 (13.4%) that they were occasionally used, and 38 (3.9%) that they were regularly used by their partners. Ninety-four (9.6%) answered "don't know" with regard to condom use by their partners.

Associations between condom use and the characteristics studied were assessed by the x^2 test as presented in Table 2. Condom use was dichotomized into two categories in the analysis. Answers of "regular" and "occasional" were classified as "use", and answers of "never" were classified as "non-use" with the rationale of dividing study subjects

Table 1. Social and reproductive characteristics of enrolled women

| Characteristics | | Ν | % |
|--|------------|-----|------|
| Age (n=973) | | | |
| - | 13-14 | 3 | 0.3 |
| | 15-19 | 73 | 7.5 |
| | 20-24 | 246 | 25.3 |
| | 25-29 | 232 | 23.8 |
| | 30-34 | 202 | 20.8 |
| | 35-39 | 131 | 13.5 |
| | 40+ | 86 | 8.8 |
| Pregnancy (n=977) | | | |
| | 0 | 29 | 3.0 |
| | 1-2 | 347 | 35.5 |
| | 3-4 | 313 | 32.0 |
| | 5-6 | 150 | 15.4 |
| | 7-8 | 75 | 7.7 |
| | 9+ | 63 | 6.4 |
| Fetal death ^a (n=962) | | | |
| | 0 | 615 | 63.9 |
| | 1 | 223 | 23.2 |
| | 2 | 78 | 8.1 |
| | 3 | 29 | 3.0 |
| | 4+ | 17 | 1.7 |
| Child death ^o (n=977) | | | |
| | 0 | 756 | 77.4 |
| | 1 | 135 | 13.8 |
| | 2 | 51 | 5.2 |
| | 3 | 24 | 2.5 |
| | 4+ | 11 | 1.1 |
| School attendance ^c (n=963) | | 07 | 0.0 |
| | <1 | 87 | 9.0 |
| | 1-5 | 308 | 32.0 |
| | 6-9 | 337 | 35.0 |
| | 10+ | 231 | 24.0 |
| Own earnings (n=967) | V | 169 | 10 5 |
| | Yes | 468 | 48.5 |
| Condom was $(r, 0.77)$ | NO | 498 | 51.5 |
| Condom use $(n=9/7)$ | NI | 701 | 71.0 |
| | Never | /01 | /1.8 |
| | Occasional | 151 | 15.4 |
| | Regular | 38 | 3.9 |
| | Don t know | 94 | 9.6 |
| | No answer | 13 | 1.3 |

a. Number of stillbirths and/or abortion (Number of pregnancy-Number of live births) b. Number of dead children after live birth c.Years of school attended

Table 2. Condom use by characteristics of enrolled women

| | | | Condom use | | |
|-----------------------|------|----------------|------------|------|----------|
| Characteristic | | Respondent (N) | Yes (N) | % | p-value* |
| Age (y) | < 25 | 282 | 51 | 18.1 | 0.522 |
| | 25 | 584 | 117 | 20.0 | |
| Pregnancy | < 3 | 345 | 77 | 22.3 | 0.096 |
| | 3 | 524 | 92 | 17.6 | |
| Fetal Death | 0 | 532 | 100 | 18.8 | 0.534 |
| | 1 | 323 | 67 | 20.7 | |
| Child Death | 0 | 682 | 148 | 21.7 | 0.001 |
| | 1 | 188 | 21 | 11.2 | |
| School Attendance (y) | < 10 | 642 | 111 | 17.3 | 0.010 |
| | 10 | 218 | 56 | 25.7 | |
| Own earnings | Yes | 444 | 105 | 23.6 | 0.001 |
| | No | 416 | 63 | 15.1 | |

* chi-square test

| | Factors | Ν | OR (95% CI) | P-value |
|--------------------------|---------|-----|------------------|---------|
| Pregnancy ^a | | | | |
| | < 3 | 345 | 1.00 | |
| | 3 | 524 | 1.31 (0.85-2.01) | |
| Fetal death ^b | | | | |
| | 1 | 323 | 1.00 | |
| | 0 | 532 | 1.13 (0.77-1.66) | |
| Child death ^c | | | | |
| | 1 | 188 | 1.00 | |
| | 0 | 682 | 2.06 (1.22-3.50) | < 0.01 |
| Mother's age (y) | | | | |
| | 25 | 584 | 1.00 | |
| | < 25 | 282 | 1.39 (0.91-2.12) | |
| School attendance (y | r) | | | |
| | < 10 | 642 | 1.00 | |
| | 10 | 218 | 1.35 (0.92-1.98) | |
| Own earnings | | | | |
| | No | 416 | 1.00 | |
| | Yes | 444 | 1.61 (1.13-2.30) | < 0.05 |

Table 3. Factors associated with condom use by logistic regression analysis

OR= odds ratio adjusted for each variables listed above

CI=confidence interval;

a Number of past pregnancies

b Number of pregnancies - Number of live births

c Number of dead children after live birth

with some intention of condom use from those without any such intention. Factors with numerical values were reclassified as follows for multivariate analysis: age, into under 25 years old and 25 years old or over; number of pregnancies, into less than 3 and 3 or more; number of fetal deaths (abortion or stillbirth), into none and 1 or more; number of dead children, into none and 1 or more; and education, into less than 10 years and 10 years or more. The occurrence of more than 1 child death was significantly associated with non-use of condoms (p=0.001). School attendance for more than 10 years (p=0.01), and having personal earnings (p=0.001) were positively associated with condom use. However, no significant association was found between school attendance for more than 10 years and personal earnings (p=0.15, data not shown).

A multivariate logistic regression analysis was then performed to identify variables that had a significant independent association with condom use. Data from the 158 subjects with missing information for any factor were excluded, and the analysis was conducted on the data from the remaining 819 subjects.

The results of the logistic regression analysis using these variables are shown in Table 3. Child death and personal earnings by the women remained statistically significant factors for condom use. More specifically, women without a child death used condoms 2.0 times more than women with at least one child death, and women with their own earnings making a financial contribution to the household used condoms 1.6 times more than women without their own earnings or contributions. Length of school attendance was not statistically significant in the multivariate logistic regression analysis.

DISCUSSION AND CONCLUSION

Our study showed the proportion of women whose partners used condoms regularly to be very close to the proportion reported in the nationwide study of 1997 (Enquête Démographique et de Santé Madagascar 1998), that is 4.9% among currently married women. The proportion was rather low, and there seemed to be no increase in use over the past few years, even though the prevalence of HIV steadily increased. Condoms seem to be used more often as a method of contraception than as a way to protect against STIs including HIV. The high proportion of women with signs and symptoms of STIs in this study, which was closely consistent with the proportion in a previous study (Harms G 1994), supports this assumption. Indeed, the use of condoms is the third most common contraceptive method among married couples in Madagascar, after injection method (9.8%) and contraceptive pills (8.9%) (Enquête Démographique et de Santé Madagascar 1998).

A history of at least one child death and personal earnings were associated with condom use in this study. The number of live births and years of school attendance were not significant factors for condom use. These results may provide a useful insight for strategies to prevent HIV infection through the use of condoms in Madagascar and other developing countries.

The importance of economic empowerment of women has been relatively underestimated until recently. This study found that the partners of women with their own earnings used condoms more frequently than those of women without their own earnings. The economic independence of women or redress of the power imbalance between partners seems to play a key role in the success of women's attempt to gain their male partners' cooperation and acceptance of condom use. This hypothesis is supported by the report that women economically dependent on men had difficulty in protecting their own reproductive health (Van der Straten et al. 1995). Since education is thought to be a prerequisite for economic empowerment, much attention has been paid to women's education. However, our results suggest that education may not necessarily be contributory or linked directly to condom use. We propose that women's economic independence deserves greater attention in efforts to combat the spread of HIV infection through condom use.

The reduction of childhood mortality seems to be another key issue related to condom use. Women with a history of at least one child death used condoms less frequently than women with no experience of child death. In this study, 221 (22.6%) of the 977 women experienced the death of a child, reflecting the very high mortality among children under 5 years of age in Madagascar: one of the 30 highest among the 191 countries in the world (UNICEF 2000). This high child mortality could dampen the motivation for contraception including condom use in society and particularly among couples who experienced the death of their children. Promotion of women's health certainly plays an important role in the health of children and family (UNICEF 2000, McDermott JM 1996), but our results suggest that the child's health gives women the impetus to improve their own health and therefore that the two are closely related.

The general population does not yet aware of the impending epidemic of HIV infection in Madagascar. Accurate knowledge of HIV infection needs to be disseminated, and education is needed to inform the population that the most efficient method of HIV prevention is, at present, the use of condoms. Dissemination of this information requires that family planning be promoted and widely accepted in Madagascar society, and condom use should be introduced as the first priority. In conjunction with these direct condom use campaigns, the promotion of women's income generation and the reduction of child mortality should be intensified and integrated as closely related programs in planning HIV prevention policy in Madagascar.

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TWO NEW SPECIES OF *SIMULIUM* (*SIMULIUM*) (DIPTERA: SIMULIIDAE) FROM THAILAND

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Abstract: Two new black-fly species of the subgenus *Simulium (Simulium)* are described and illustrated, based on female specimens captured while approaching a human near a mountain summit in Doi Inthanon National Park, Chiang Mai Province, northern Thailand. The first species, *Simulium suchariti* sp. nov., is assigned to the *griseifrons* species-group and is distinguished from most other related species by the cibarium with tubercles near the base of its medial projection; the second, *S. setsukoae* sp. nov., is very similar to *S. rufibasis* of the *tuberosum* species-group but is distinguished from the latter by the shorter length of clustered stout hairs on the seventh abdominal segment and the ovipositor valve with its posteromedial corner widely bare.

Key words: black fly, Simuliidae, Simulium, Thailand, new species, griseifrons species-group, tuberosum speciesgroup

During our recent collections of adult black flies using a human attractant in Doi Inthanon National Park, Chiang Mai, northern Thailand, we obtained females of two undescribed species of the subgenus Simulium Latreille s. str. of the genus Simulium Latreille s. l. The first species seems to belong to the griseifrons species-group (Takaoka and Davies, 1996) in that it possesses five longitudinal vittae on the scutum, simple claws and triangular ovipositor valves; while the second is conspecific to Simulium sp. D, recorded from the same national park by Takaoka and Suzuki (1984). The latter species is very closely related to S. rufibasis Brunetti, 1911 and S. weji Takaoka, 2001, of the tuberosum species-group, by the presence of a pair of clustered hairs on the ventral surface of the seventh abdominal segment of the female (Puri, 1932a; Takaoka, 2001). These two species are new to science and so described below.

The morphological features and terms used herein follow those of Takaoka (2003).

Holotype and paratype specimens will be deposited at the Natural History Museum (BMNH), London, U.K.

Simulium (Simulium) suchariti sp. nov.

DESCRIPTION. **Female.** Body length about 2.4 mm. *Head.* Narrower than width of thorax. Frons slaty black, shiny, widely bare except for several dark stout hairs along each lateral margin; frontal ratio 1.4:1.0:1.2 - 1.3; fronshead ratio 1.0:3.5 - 4.1. Fronto-ocular area (Fig. 1) moder-

ately developed. Clypeus slaty black, white pruinose, shiny, sparsely covered with dark stout hairs except for the medial portion widely bare longitudinally. Labrum about 0.7 times as long as clypeus. Antenna composed of 2+9 segments, all brownish black to black though base of 1st flagellar segment narrowly light brown; 1st flagellar segment 1.7 - 2.0 times as long as 2nd one. Maxillary palp composed of 5 segments, black except 4th and 5th segments greyish black, with proportional lengths of 3rd, 4th and 5th segments 1.0: 1.0:2.0 - 2.1; 3rd segment (Fig. 2) of moderate size; sensory vesicle large, elongate, with rugged surface, about 0.4 times length of 3rd segment, with medium-sized round opening near apex. Maxillary lacinia with 13 - 15 inner and 14 - 16 outer teeth. Mandible with 24 - 28 inner and 12 outer teeth. Cibarium (Fig. 3) with a narrow medial projection directed dorsally, and covered with many minute tubercles on this projection and 10 - 12 somewhat larger ones near its base. Thorax. Scutum black (except anterolateral calli medium brown), shiny, whitish-grey pruinose, with 5 dark longitudinal non-pruinose vittae (i.e., 1 very narrow medial, 2 rather broad submedial, and 2 rather broad lateral vittae), all united on prescutellar area when illuminated dorsally and viewed anterodorsally (this color pattern reversed when viewed from behind); scutum with a pair of white submedial spots near anterior margin when illuminated anteriorly and viewed dorsally; scutum densely covered with yellow recumbent fine hairs interspersed with long upstanding dark hairs on prescutellar area. Scutellum dark brown,

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shiny, whitish-grey pruinose, with long dark and short yellow hairs. Postnotum dark brown to brownish black, shiny, whitish-grey pruinose, without hairs. Pleural membrane bare. Katepisternum longer than deep, and bare. *Legs*. Foreleg: coxa whitish yellow; trochanter dark brown except under surface yellow basally; femur yellow on basal 1/2 and brownish black on apical 1/2; tibia whitish yellow except apical 1/4 brownish black; tarsus entirely black; basitarsus, 2nd and 3rd tarsal segments somewhat dilated; basitarsus about 6.4 times as long as its greatest width. Midleg: coxa brownish black; trochanter dark brown to brownish black except base yellow; femur brownish black except dorsal surface of basal 1/5 and ventral surface of basal 1/4 or 1/3 yellow; tibia white except apical 1/3 brownish black; tarsus brownish black except base of basitarsus dark yellow. Hind leg: coxa dark brown to brownish black; trochanter yellow; femur brownish black except basal 1/4 yellow; tibia white except a little less than apical 1/3 brownish black; tarsus brownish black except basal 1/2 of basitarsus and base of 2nd tarsal segment whitish yellow; basitarsus (Fig. 4) nearly parallel-sided, about 5.6 times as long as wide, about 0.8 and 0.7 times as wide as hind tibia and femur, respectively;



Figures 1-7. Morphological characters of female *Simulium suchariti* sp. nov. 1, fronto-ocular area; 2, 3rd segment of maxillary palp with sensory vesicle; 3, cibarium; 4, basitarsus and 2nd tarsal segment of hind leg (left side and outside view); 5, medial portion of ventral surface of the abdominal segment 7 showing a pair of submedial sternal plates; 6, sternite 8, ovipositor valves, genital fork and spermatheca *in situ* (ventral view); 7, right paraproct and cercus (lateral view). Scale bars. 0.05 mm for all figures.

calcipala distinct, 0.9 times as long as wide and about 0.5 times as wide as the width of apical portion of basitarsus; pedisulcus distinct. All tarsal claws simple, without basal or subbasal tooth. Wing. Length 2.4 - 2.7 mm. Costa with dark spinules and hairs; subcosta haired except apical 1/5 to 2/5 bare; basal section of radial vein bare; R_1 with spinules and hairs; R₂ with hairs only; hair tuft at base of stem vein dark brown; basal cell absent. Abdomen. Basal scale dark brown, with a fringe of dark long hairs on each side; dorsal surface of abdominal segments light to medium brown and somewhat greyish, with many vertical wrinkles except tergites dark brown and smooth; tergite 2 with a pair of silvery shiny dorsolateral patches not connected medially to each other; tergites 3, 4 and 5 small, nearly as long as wide; tergite 6 about 1.5 times as wide as long, tergites 7 - 9 large and wide, tergites 6 - 9 shiny; dorsal surface of segments 2 and 3 almost bare, but that of the remaining segments sparsely covered with dark short hairs. Ventral surface of abdominal segments 2 - 7 ocherous except a pair of submedial sternal plates on segment 7 (Fig. 5) dark brown, and sparsely covered with dark hairs except 2nd and 3rd segments almost bare. Genitalia (Figs. 6 and 7). Sternite 8 well sclerotized, dark brown to brownish black, bare medially, and with 17 - 19 long stout hairs as well as a few short hairs laterally on each side; ovipositor valve triangular in shape, membranous, covered with 12 - 16 stout hairs as well as numerous microsetae; inner border nearly straight, well sclerotized except near apex transparent. Genital fork inverted-Y-shaped, with well-sclerotized stem; arms slender, each with strongly sclerotized apical bulge having a distinct projection directed anteriorly. Paraproct in ventral view with distinct shallow concavity on ventral surface along anteromedial margin; anteromedial surface of paraproct moderately sclerotized, and with a few short sensilla; paraproct much produced ventrally, covered with 14 - 19 short stout hairs on ventral and lateral surfaces. Cercus in lateral view much wider, about 2.7 times as wide as long, covered with many short stout hairs. Spermatheca nearly ovoid, 1.2 times as long as wide, well sclerotized (though duct and large area around the base of duct unsclerotized), with faint reticulate surface pattern; minute internal setae present; accessary ducts subequal in diameter to each other, and somewhat larger than the major one.

TYPE SPECIMENS. Holotype female, collected with a hand net while approaching a human, near the mountain summit of Ang Ka, altitude 2,465 m, Doi Inthanon National Park, Chiang Mai Province, northern Thailand, 21. IX. 2003, by W. Choochote. Paratype 1 female, same data as holotype except date, 19. IX. 2003.

ETYMOLOGY. The species name *suchariti* is in honor of Prof. Supat Sucharit, former head of the Department of Medical Entomology, Faculty of Tropical Medicine, Mahidol University, Bangkok, Thailand, whom the junior author, WC, thanks for his advice and encouragement.

REMARKS. Simulium suchariti sp. nov. is similar in the female to S. kawamurae Matsumura, 1915 from Japan (Bentinck, 1955) in that it has the triangular ovipositor valve with several stout hairs and the cibarium with a medial projection directed anteriorly and covered with tubercles on and near this projection, as well as the scutum with five longitudinal vittae. The female of S. kawamurae differs from that of this new species by the first flagellar segment of antenna about 1.3 times as long as the second one, the sensory vesicle about 0.35 times as long as the third maxillary palpal segment, mid and hind basitarsi whitish on basal 1/2 and on basal 2/5, respectively, and the cercus about 1.6 times as wide as long. This new species is similar to S. digrammicum Edwards, 1928, originally described from female specimens collected in peninsular Malaysia and Thailand (Edwards, 1928; Takaoka and Davies, 1995) and S. griseifrons Brunetti, 1911, from India (Puri, 1932b), but is distinguished by the bare basal portion of the radial vein (cf. fully haired in S. digrammicum and haired or bare in S. griseifrons), a pair of submedial sternal plates (Fig. 5) (cf. one medial sternal plate in the two known species), and the presence of tubercles near the base of the medial projection of the cibarium (Fig. 3). The female of S. suchariti is also similar to that of S. maenoi Takaoka and Choochote, 2002, found in northern Thailand (Takaoka and Choochote, 2002) in that it has the bare basal portion of radial vein, but differs by the presence of many tubercles near the base of the medial projection of the cibarium (Fig. 3).

The male, pupa and larva of this new species are unknown yet.

Simulium (Simulium) setsukoae sp. nov.

Simulium (*Simulium*) sp. D: Takaoka and Suzuki, 1984 (fe-male)

DESCRIPTION. **Female.** Body length 2.2 - 3.1 mm. *Head.* Narrower than width of thorax. Frons slaty black, shiny, with several dark stout hairs along lateral margins; frontal ratio 1.3 - 1.4:1.0:1.1; frons-head ratio 1.0:3.5 - 3.7. Fronto-ocular area (Fig. 8) moderately developed, short and rounded apically. Clypeus slaty black, shiny, with scattered dark stout hairs marginally. Labrum 0.61 - 0.64 times as long as clypeus. Antenna composed of 2+9 segments, dark brown except base of 1st flagellar segment narrowly yellow; ventral surface of 1st flagellar segment dark yellow in some

females; 1st flagellar segment about 1.5 times as long as 2nd one. Maxillary palp brownish black, composed of 5 segments with proportional lengths of 3rd, 4th and 5th segments 1.0:1.0:2.1; 3rd segment (Figs. 9 and 10) of moderate size; sensory vesicle long, 0.39 - 0.49 times length of 3rd segment, with medium-sized opening medially or apically. Maxillary lacinia with 14 - 18 inner and 17 - 19 outer teeth. Mandible with about 34 inner and 13 - 15 outer teeth. Cibarium (Fig. 11) with numerous minute tubercles near the

anterior margin. *Thorax.* Scutum slaty black, shiny, not patterned, moderately covered with copper-colored recumbent hairs (appearing shiny yellow at certain angle of light), interspersed with dark long upstanding hairs on prescutellar area; scutum thinly grey pruinose when illuminated at a certain angle of light. Scutellum dark brown, with dark long hairs. Postnotum dark brown, shiny, without hairs. Pleural membrane bare. Katepisternum longer than deep, and bare. *Legs.* Foreleg: coxa dark yellow or light yellowish-brown;



Figures 8-15. Morphological characters of female *Simulium setsukoae* sp. nov. 8, fronto-ocular area; 9 and 10, 3rd segments of maxillary palp with sensory vesicle; 11, cibarium; 12, basitarsus and 2nd tarsal segment of hind leg (left side and outside view); 13, right half of medial portion of ventral surface of the abdominal segment 7 showing small sternal plate and clustered hairs on it; 14, sternite 8, ovipositor valves, genital fork and spermatheca *in situ* (ventral view); 15, right paraproct and cercus (lateral view). Scale bars. 0.05 mm for all figures.

trochanter and femur dark brown; tibia dark brown with outer median portion largely yellowish white, and extensive white sheen on outer margin when illuminated; basitarsus entirely dark brown, slightly dilated, 6.4 times as long as its greatest width, with short dorsal hair crest; rest tarsal segments dark brown. Midleg: dark brown except basal 1/4 to 2/5 of tibia and a little less than basal 1/2 of basitarsus whitish yellow; tibia with extensive white sheen on posterior surface when illuminated. Hind leg: dark brown with basal 1/3 of tibia, a little more than basal 1/2 of basitarsus and basal 1/2 of 2nd tarsal segment yellowish white; tibia with extensive white sheen on posterior surface when illuminated; basitarsus (Fig. 12) parallel-sided, 6.6 times as long as wide, 0.62 and 0.56 times as wide as the greatest width of hind tibia and femur, respectively; calcipala short, 1.1 times as wide as long, and 0.57 times as wide as basitarsal tip; pedisulcus distinct. All tarsal claws simple, without basal or subbasal tooth. Wing. Length 2.5 - 2.8 mm; costa with spinules and hairs; subcosta haired except apical tip bare; basal section of vein R bare; R1 with spinules and hairs; R2 with hairs only; hair tuft at base of stem vein dark brown; basal cell absent. Abdomen. Basal scale dark brown with fringe of dull long hairs; dorsal surface of abdomen medium to dark brown, with dark hairs; 2nd segment with a pair of silvery iridescent spots dorsolaterally, broadly connected in middle to each other; tergites 3, 4 and 5 small and dull, but tergites 6 - 8 large and shiny. Ventral surface of abdominal segment 7 (Fig. 13) with a pair of small submedial sternal plates, and with a cluster of 11 - 15 stout hairs on each sternal plate (hairs on sternal plates subequal in length to those on the surrounding area of the same segment). Genitalia (Figs. 14 and 15). Sternite 8 well sclerotized, bare medially but with 8 - 11 long stout hairs as well as a few short slender hairs laterally on each side; ovipositor valve triangular in general shape having a small flap -like posteromedial corner with rounded margin, membranous, covered with 5 - 8 short setae as well as numerous microsetae except narrow portion along inner margin and rather wide portion near posteromedial corner bare; inner margins slightly to widely curved, and moderately sclerotized. Genital fork inverted-Y-shaped, with wellsclerotized stem; arms slender, each with strongly sclerotized projection directed anterodorsally. Paraproct much longer than cercus, covered with 26 - 30 short stout hairs on lateral and ventral surfaces; anteromedial surface strongly sclerotized. Cercus rounded posteriorly, about 0.5 times as long as wide, covered with about 20 short stout hairs on outer surface. Spermatheca nearly globular, well sclerotized (though duct and small area around base of duct unsclerotized); no definite reticulate pattern; minute internal setae present; accessary ducts of moderate size, subequal to

that of the main one.

TYPE SPECIMENS. Holotype female, collected with a hand net while approaching a human, near the mountain summit of Ang Ka, altitude 2,465 m, Doi Inthanon National Park, Chiang Mai Province, northern Thailand, 18. IX. 2003, by W. Choochote. Paratypes 3 females, same data as holotype, 1 female, same data as holotype except date, 21. IX. 2003, and 4 females, same data except date, 25. XII. 2001.

OTHER SPECIMENS EXAMINED. 6 females (labelled *Simulium* sp. D) collected with a hand net while biting a human, altitude ca. 2,400 m, Doi Inthanon National Park, 20. II. 1978, by H. Suzuki.

ETYMOLOGY. The species *setsukoae* is named after the wife of Dr. Hiroshi Suzuki, Nagasaki University, who collected female specimens of this new species for the first time in 1978 (Takaoka and Suzuki, 1984).

REMARKS. Simulium setsukoae sp. nov. is very similar to the female of S. rufibasis in many features including the leg color, but is distinguished from the latter by the short size of the clustered hairs on the seventh abdominal segment which are subequal to those on the surrounding area (Fig. 13), as already mentioned by Takaoka and Suzuki (1984). In addition to this difference, this study clarified that the two species differ from each other in the ovipositor valve, i.e., its posteromedial corner widely bare in S. setsukoae (Fig. 14) but moderately setose in S. rufibasis. This new species is also similar to S. weji, found in northern Thailand (Takaoka, 2001), which has also a pair of clustered hairs on the seventh abdominal segment, but the latter species has many different characteristics including the smaller sensory vesicle, the cibarium with a reduced number of tubercles, the hind femur not entirely dark, and the clustered hairs much longer than those of S. rufibasis (Takaoka, 2001; Takaoka and Suzuki, 1984).

The male, pupa and larva of this new species remain unknown.

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President

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| P2 - 47 | Health Problems and Status of Homeless People living in Taito-ku SANYA-Ar | ea |
| | | Abe, A. et al. |
| P2-48 | Health Situation of Homeless People in Shinjuku, Tokyo | Ohwaki, K. et al. |
| P2 - 49 | Current situation of life-style related diseases among Japanese expatriates in | |
| | Kuala Lumpur, Malaysia | Hondo, K. et al. |
| P2-50 | Demand survey of Typhoid vaccine in Japan Overseas Health Administration | Center |
| | | Koga, T. <i>et al.</i> |
| P2-51 | Cystic Hydatidosis - Occurrence of Cases in Japan and its Prevention. | Doi, R. et al. |
| P2-52 | Correlation between the risk recognition by obstetricians about congenital tox | oplasmosis |
| | and their clinical activities | Naoi, K. <i>et al.</i> |
| P2-53 | DNA diagnosis of malaria using microtiter-plate hybridization method | Kim, H. et al. |
| P2-54 | Detection of <i>Plasmodium berghei</i> by ICT P.f./P.v. immunochromatographic | e test (2): |
| | analysis of the detection sensitivity for the panmalarial antigen during the infe | ctious |
| | courses with antimalarial treatment | Arai, M. et al. |
| P2-55 | Recombinant baculovirus virions displaying circumsporozoite protein protect | |
| | agasint Plasmodimu berghei sporozoite infection | Yoshida, S. et al. |
| P2-56 | The target of Atoyaquone in <i>Plasmodium falciparum</i> | Mi-ichi, F. <i>et al.</i> |

P2-56The target of Atovaquone in *Plasmodium falciparum*Mi-ichi, F. *et al.*P2-57Plasmodium berghei NK65 giving rise to recrudescence after chemothrapy.Nakazawa, S.

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| P2-58 | Epidemiological and histological study of Burkitt lymphoma in inland provinces of | Kenya |
|----------------|--|----------------------------|
| | | Toriyama, K. <i>et al.</i> |
| P2-59 | Epidemiological study and Characterization of Japanese encephalitis virus strains is | olated |
| | in Okinawa island, in 2002 and 2003. | Saito, M. et al. |
| P2-60 | A clinical significance of platelet-associated IgG and IgM in dengue virus | Saito, M. et al. |
| P2-61 | Clinical assessment of dengue among children in Metro Manila: 1999-2001 | Honda, S. et al. |
| P2 - 62 | Electron microscopic studies on the multiplication of chikungunya virus in Aedes al | bopictus |
| | | Yamanishi, H. |
| P2-63 | Self-reported blood in urine and water contact in a schistosomiasis endemic area of | coastal Kenya |
| | | Kaneda, E. et al. |
| P2 - 64 | Urinary schistosomiasis in north Zanzibar: An epidemiological study with questionn | aire for |
| | water contact behaviors and self-diagnosis | Ataka, Y. <i>et al.</i> |
| P2-65 | Inhibition of Schistosoma Mansonia Infection by Ultraviolet Cream | Ohwatari, N. <i>et al.</i> |
| P2-66 | Toward the interruption of vectoral transmission of Trypanosoma cruzi - impact of v | vector |
| | control against Chagas disease vector in Guatemala | Nakagawa, J. <i>et al.</i> |
| P2 - 67 | Characterization of procyclic form-specific genes from Trypanosoma brucei bru | lcei |
| | | Ohshima, S. et al. |
| P2-68 | Molecular cloning and characterization of Vacuole Protein Sorting (VPS41) gene fro | om |
| | Trypanosoma brucei brucei | Suzuki, T. <i>et al.</i> |
| P2-69 | Development of an Immunochromatographic Test for the Rapid Detection of Babe | sia equi |
| | Infection in Horses | Igarashi, I. <i>et al.</i> |
| P2-70 | Murine Dendritic Cells Derived from Peritoneal Cavity Macrophages Undergo Rapi | d |
| | Apoptosis in Culture which is Blocked by Toxoplasma Gondii | Makala, L. <i>et al.</i> |
| P2-71 | Possible role of calcium ions and calmodulin in the excystation and metacystic | |
| | development of Entamoeba | Makioka, A. <i>et al.</i> |
| P2-72 | Analysis of farnesyltransferase of Entamoeba histolytica | Kumagai, M. et al. |

President's lecture

K1) ROLE OF EPIDEMIOLOGY IN THE FIELD OF TROPICAL MEDICINE

TAKESUMI YOSHIMURA

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It has been stated that epidemiology should be essential and inevitable in the field of tropical medicine. Recently, two symposia at the scientific meeting targeted an epidemiological approach for tropical medicine. One was held in November 2002 at the 43rd Japanese Tropical Medicine Association scientific meeting (President Prof. Hashiguchi) titled "Toward ideal field work - Epidemiological approaches and ethical issues for field work "The latest symposium titled "Field epidemiology in tropical areas "was held at the 27th Kyushu district's scientific meeting of the Japanese Tropical Medicine Association in February 2003.

In this presentation, I would like to discuss 1) What can be accomplished through epidemiology? 2) What are the limitations of epidemiology? and 3) What should be done in the field of tropical medicine in future? Among many epidemiological studies undertaken by this author, an epidemiological study on onchocerciasis in Guatemala, C.A. was taken as one of the examples. The project requested an epidemiologist to evaluate the effect of vector control on onchocerciasis occurrences. Repeated cross - sectional studies in pilot areas in Guatemala, C.A. on

vector control had continued for 9 years, in order to observe changes of prevalence and incidence of onchocerciasis before and after vector control. For this purpose, census, population registration and annual examinations for onchocerciasis by skin snip biopsy had been carried out. It was found that after vector control by larvicide, prevalence and incidence of onchocerciasis infection declined dramatically. Thus, a well designed epidemiologic study gave us evidence regarding the effect of vector control on onchocerciasis in the project.

In general, epidemiology can 1) evaluate health problems quantitatively, 2) can clarify risk or preventive factors for a health problem and 3) can evaluate the effect of a medical intervention program. But epidemiological approaches are limited because of human nature and ethical or practical reasons. Field epidemiologists should try to do their best to conduct epidemiological surveys, taking into consideration the basic health information, human behavior, local culture etc. Finally I propose that human resources for field epidemiologists should be developed more in our society.

Key note speech (lecture)

K2) REPORT FROM THE FIELD ; SARS CONTAINMENT PROGRAM AT WHO

SHIGERU OMI

The Western Pacific Regional Office, World Health Organization

SARS (Severe Acute Respiratory Syndrome) has received great attention. The SARS outbreak occurred within the jurisdiction of WHO office for the Western Pacific Region, and I directed the countermeasures for four months, beginning in February 2003. Information on SARS, an emergent infectious disease, was limited. It was likely to be a historical catastrophe, with its quick spread and high death rate, and no identified virus. However, all concerned professionals collaborated in planning the countermeasures, based on epidemiological analyses conducted quickly in coordination with WHO. This can be a model for global health risk control.

SARS is also a social phenomenon. Its outbreak took a new form in the current century, accompanied by many people crossing international borders. It crippled the whole health care system through its effects on medical staff. It had a heavy impact on both human beings and economies. Political negotiations, illustrated in the advisory that travel to certain regions that might threaten the nation's basis of existence, were added to the WHO's duties. This indicates that activities to sustain people's health involve work across many sectors.

The lessons are : infectious diseases are a continuous threat ; and disclosure and transparency of information are important in countermeasure activities. It will be a challenge to utilize the revised International Health Regulations based on these lessons. Innumerable health problems, including emergent and recurrent infectious diseases, have universality across national borders. Collaboration is necessary to solve such problems. With this recognition, WHO will continuously endeavor to accomplish the task.

Prize winner's lecture of Japanese Society of Tropical Medicine

K3) MOLECULAR CHARACTERIZATION OF PEROXIREDOXINS FROM HUMAN MALARIA PARASITE *PLASMODIUM FALCIPARUM*

SHINICHIRO KAWAZU

Research Institute, International Medical Center of Japan

The human malaria parasite Plasmodium falciparum is exposed to reactive oxygen species that are generated via its endogenous metabolism (e.g. hemoglobin digestion) and during external attack by the host immune system. In addition, the effects of several anti-malarial drugs (e.g. chloroquine and artemisinin) can burden the parasite with substantial oxidative stress.

Peroxiredoxins (Prxs) are a recently described family of antioxidants that are ubiquitous in living cells for metabolizing hydrogen peroxide and hydroxyl radicals. We have reported the molecular cloning and characterization of 1-Cys and 2-Cys Prxs from *P. falciparum*. The results obtained from a series of experiments are summarized herein, and the physiological roles of these antioxidant proteins in the parasite cell are discussed here.

2-Cys Prx is expressed in the parasite cytoplasm throughout the blood stage. The thioredoxin (Trx) - per-

oxidase activity of 2-Cys Prx revealed in the recombinant protein suggests that Prx is constitutively expressed and, thus, likely plays a housekeeping role in the parasite's intracellular redox control. Disruption of the 2-Cys Prx gene renders the parasite hypersensitive to reactive oxygen and nitrogen species.

In contrast, 1-Cys Prx shows stage-specific expression in blood-stage parasites. The limited expression of 1 - Cys Prx in the trophozoite cytoplasm suggests that 1 -Cys Prx may be involved in the parasite's hemoglobin metabolism, which generates a pro-oxidative heme iron and increases intracellular oxidative stress. Overexpression of 1-Cys Prx makes the parasite less susceptible to chloroquine.

Further studies clarifying the roles of Prxs in *P. falciparum* may lead to use of these proteins as targets for anti-malaria chemotherapies.

Educational lectures

E-1) WEST NILE VIRUS INFECTION

KOUICHI MORITA

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West Nile virus was first isolated from a patient with fever in Uganda in 1937. The virus was subsequently

named after the patient's residence. West Nile virus, an arthropod-borne virus, circulates between birds and mos-

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quitoes. Humans are infected by mosquitoes carrying the virus and show wide range of illness, such as mild fever, meningitis and encephalitis. West Nile virus could be found in Africa, Middle East, West Asia and parts of Europe, but was previously absent from American continent. In 1999, the virus suddenly emerged in New York, USA and caused 62 patients with 7 deaths. Virus transmission was confirmed in four states of the USA in that year. Since then, the areas of the virus transmission have continuously expanded, in the years 2000 and 2001, 12 and 26 states respectively. While, the number of patients did not surge, 18 and 42 cases in the year 2000 and 2001, respectively. However, both the area of virus circulation and number of patients increased explosively in 2002. A total of 43 states reported 3873 patients with 246 deaths. Important characteristics of the West Nile virus in the US include the severe pathogenecity in avian, high neuro -invasiveness in humans, and rapid expansion of the virus affected area. If the West Nile virus epidemic in the US continues, the virus may eventually spread to Japan and

other part of Asia. At present, transmission of West Nile virus from the US or West Asia to the East or Southeast Asia has not been reported. There are frequent birds migrations between Asian countries and Japan. Thus, once West Nile virus invades the Southeast Asia, there is a possibility that the virus quickly transmitted to Japan by these migrating birds. Actually, in the early 1990s, genotype I Japanese encephalitis virus (JEV) was transmitted from Asia to Japan, which is now a dominant JEV strain in Japan. If West Nile virus enters Japan, differential diagnosis from JE is essential, because both viruses have very similar antigenecity and pathogenecity. West Nile virus is transmitted by mosquitoes breeding in urban centers and cause outbreaks in more populated areas than JEV, that is transmitted by Culex tritaeniorhynchus, which breeds in less populated area such as rice fields. Collaboration of experts in clinical, research, public health, and veterinary fields is vital for the response to the possible invasion of West Nile virus and there is an urgent need for vaccine development.

E-2) HEALTH PROBLEMS OF OVERSEAS TRAVELERS

ATSUO HAMADA

Japan Overseas Health Administration Center,

Every year we see an increase in the numbers of Japanese who travel abroad for business or vacation. Naturally, many of these people contract diseases, and we have noticed a great increase in demand for this group's cooperation in addressing health problems internationally. We began our work in the area of occupational medicine, that is, health care for Japanese people sent abroad for business. Since the 1970s, Japanese corporations have increasingly sent people overseas. Illnesses such as diarrhea or malaria were especially problematic, and prevention of these became our highest priority. Since the early 1980s, the health problem of overseas employees became an issue for the Japanese Society of Tropical Medicine. Around 1990, as a country, we realized the need for international cooperation in our approach to health problems of overseas employees. As a result, sponsored medical checkup was offered before and after overseas dispatch. Also, the Japan Overseas Health Administration

Center was established as part of the Labor Welfare Corporation. Meanwhile, the health issues affecting overseas employees have been changing. As always, contagious diseases are still a big problem, but the widespread use of the Internet have made it easier for employees to obtain information necessary for care and prevention. Attention has shifted to new health problems such as lifestyle related diseases and mental health problems. While these health problems are not specific to tropical regions, they do affect overseas residents. Now, in addition to occupational medicine and tropical medicine, we wish to include health issues that are global in scope. In the west, travel medicine is well established, but in Japan, it is still under development. In order for us to advance in the field of travel medicine, we must seek the cooperation and participation of all those who have medical expertise in the field of international health.

E-3) SARS : STRUGGLE AND EXPERIENCE IN CONTROL

HIROSHI OHARA

International Cooperation Bureau, International Medical Center of Japan, Tokyo, Japan

Severe Acute Respiratory Syndrome (SARS), which originated in Guandong Province in China in November 2002, spread to many countries. International collaborations including the dispatch of Japanese Disaster Relief Teams were conducted aiming at putting SARS under control. This presentation summarizes the experiences participated in SARS control as Japanese teams.

Vietnam : A total of 63 nosocomial infection cases with 39 medical staff were reported at the hospital to which the index case was admitted. The Ministry of Health decided to close the above hospital and designated Bach Mai Hospital (BMH) as the sole facility for receiving SARS cases. Since then no case of nosocomial infection has been reported and SARS was contained on April 28.

During March 16 - April 1, the Japanese Disaster Relief Team was dispatched and technical guidance on nosocomial infection control was performed as well as providing protective attires and respirators. In BMH technical cooperation project has been implemented since January 2000 by Japanese experts, which incorporated technical guidance on nosocomial infection control and the medical staff at BMH had reached higher level of skills when SARS broke out.

Guandong Province : After the index case the epi-

demics spread rapidly and more than 300 cases had occurred by the middle of February 2003. A hospital statistics reported that 45 out of 83 cases derived from nosocomial infection. The number of patients decreased in and after April. During April 10 - 15, the Japanese Medical Team was dispatched with the main purpose of protecting Japanese sojourners there.

Beijing City : Since the index case was recognized in March 2002, cases continued to increase. At the initial stage a lot of nosocomial infection occurred with medical staff accounting for 15 - 20% of all cases. Thereafter the Chinese Government made a great effort and succeeded in SARS containment. During May 11 - 16, the Japanese Disaster Relief Team was dispatched and cooperated by providing technical guidance on nosocomial infection control in combination of providing equipment.

Prompt action, adequate nosocomial infection control, accurate information sharing and international cooperation are crucial factors in carrying out effective SARS control. Besides, more importance should be put on training of medical staff to enhance basic techniques and establish control system at ordinary times, not starting after the outbreak of epidemics. Such basis will make it possible to apply stringent nosocomial infection control promptly.

E-4) AMENDMENT OF THE LAW CONCERNING THE PREVENTION OF INFECTIONS AND MEDICAL CARE FOR THE PATIENTS OF INFECTIONS AND THE QUARANTINE LAW

MITSUHIRO USHIO

Tuberculosis and Infectious Diseases Control Division, Health Service Bureau, Organization of the Ministry of Health, Labour and Welfare, Tokyo, Japan.

The basic law to fight against infectious diseases is the Law concerning the Prevention of Infections and Medical Care for the Patients of Infections (hereinafter refer to the law), which was established in 1998 by making fundamental reform of previous Communicable Diseases Prevention Law. After enacting this law, zoonosis such as West Neil Disease or SARS (Severe Acute Respiratory Syndrome) and Bacterial Terrorism are giving a new threat to human-being, especially the case of a foreign medical doctor's visit to Japan who found to have a contact with SARS patient after returning his home country made a panic and gave us an alert for the high possibility of introduction of new diseases.

Initially the law should be amended in 2004 accord-

ing to its supplementary provision, which says the law should be revised at least in five years after enacting, however this process has accelerated to cope with new emerging disease; SARS, epidemic of which is once declared to be ceased by WHO (World Health Organization) but expected to re-emerge in this winter season.

Sub-committee for Infection Prevention of Welfare and Science Council submitted the proposal for the counter measure for infection in new era in 21 August 2003, which become the basis of law amendment.

Followings are major points of amendment;

(1) Strengthen the right and responsibility of the Minister of Health, Labour and Welfare in case of emer-

gency

(2) Inclusion of Action Plan in both the Basic Guidelines established by the Minister and the Prevention Plan made by the each Prefectural Governor

(3) Strengthen the measure concerning import of animals that might transmit the pathogen of infectious diseases

(4) Strengthen the role and right of Quarantine station, especially to prevent the introduction of SARS from foreign countries

(5) Define SARS and Small Pox as the category 1 infections

Symposium 1

S1-1) JAPANESE INTERNATIONAL COOPERATION FOR HEALTH IN THE 21ST CENTURY : ITS POLICY, PRACTICE, RESEARCH AND EDUCATION

MASAAKI SHIMADA¹, MASAMINE JIMBA²

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Since the start of the 21st century, we have experienced many important health matters.

First, we could not control HIV/AIDS within the 20th century and it remains a major public health threat. Then, SARS hit the world this year. Moreover, war and terrorism have emerged once more. Health workers are just paying its debt. Meanwhile, the gap between the rich and the poor is getting bigger and bigger, making it difficult for the poor to receive basic health services.

How can we cope with these issues in the field of international health? What can Japan contribute to solve these problems? Japan had been the top donor country for developing countries in the 1990s, and now it stands at number 2. While quantity is important, the quality of Overseas Development Aid has been emphasized these days in Japan.

To improve quality, the human factor is the key. We also need ideas. Although the total budget tends to decrease, we still have enough. In this symposium, we invite health professionals who may lead the international health of the 21st century and ask them to tell us their ideas.

We selected five professionals from the Ministry of Foreign Affaires, the Japan International Cooperation Agency, two universities, and one non-governmental organization. They are the people who do not just advocate and go away, they are the people who can take responsibility for what they say. Finally, we welcome the audiences' active participation in this symposium. (Jimba)

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S1-2) ROLES AND CHALLENGES OF JAPAN FOR GLOBAL HEALTH

OSAMU KUNII

Research and Programming Division, Economic Cooperation Bureau, Ministry of Foreign Affiars

Japan's ODA had been the top donor for a decade in the 1990s and has long been committed to health improvement in developing countries. However, as the world has been learning many lessons in development assistance for developing countries, Japan needs to catch up with these global trends.

In my opinion, there are five key challenges for Japan's ODA in the health sector, including : internationalization of Japan's ODA, specification and strategizing of assistance strategies, promotion and consolidation of partnerships, activation of civil society, human resource development through specialization and mobility.

To address these challenges, I believe that Japan should keep up with global standards and trends of development assistance. At the same time, the country needs clearly articulate its policy, philosophy and strategies. It might be necessary for Japan to make a health sector assistance strategy, including prioritizing issues, approaches and countries. Real partnerships for better utilization of ODA from planning to evaluation are needed with academia, NGOs, other donors, UN organizations, etc. Engagement of civil society for development assistance should be strengthened, particularly in terms of organizational management and expertise. Human resources and the capacity of Japanese experts in international health should be developed both in quality and quantity.

All of these challenges need strategic planning, followed by intensive, collective discussion among all the stakeholders.

S1-3) HEALTH SECTOR COOPERATION FOR DEVELOPING COUNTRIES BY JAPAN INTERNATIONAL COOPERATION AGENCY (JICA)

AKIRA HASHIZUME

Medical Cooperation Department, Japan International Cooperation Agency (JICA), Tokyo, Japan

Japan started its assistance to developing countries in 1954. When the Japanese government established an organization for technical assistance in 1962, which later became JICA, infant mortality rate of Japan was still 26.4 per 1000 and maternal mortality rate was about 1/1000.In half of the countries at present, IMR and MMR are not as high as this level. If these countries will be more developed and become partners of Japan in helping other developing countries, we can imagine the wonderful future of human kind.

JICA has become an independent administrative agency. As the newly adopted logotype shows, partnership will be a keyword for JICA's international cooperation.

JICA has been so far a bureaucratic organization with top-down decision system. But as it is now an inde-

pendent agency, Ministry of Foreign Affairs and JICA have become partners. In addition, under the leadership of the new president, JICA will put more emphasis on activities in each developing country, i.e. the decentralization of decision process. At the same time, JICA's management system should be such prepared that decentralization would be implemented smoothly. As being an independent administrative agency requires efficiency, more efforts should be put to share, among JICA staff, experiences and failures as institutional memories. This enables us not to repeat the same errors in a similar project.

JICA will adopt theme-wise approaches before next April. In this way, we will be able to share the experiences of similar projects, regardless of region or country the projects exist.

S1-4) HOW CAN NGOS CONTRIBUTE TO INTERNATIONAL HEALTH? _"DIVERSITY" AND "FLEXIBILITY"

TOMOKO FUJISAKI

Health and Development Service (HANDS)

In Japan, interest in non-government organizations (NGOs) has increased in recent years, along with expectations for their work. Often, however, discussions take place without a clear and common understanding of what NGOs are. Also, there is confusion between similar terms, such as NGOs, non-profit organizations (NPOs), private voluntary organizations (PVOs), and community-based organizations (CBOs). In this presentation, the term NGO refers to venues for citizens to pursue public interests.

NGOs differ from each other in their legal status, missions, target populations, and strengths. The diversity among NGOs has value, which should be recognized and appreciated more widely. NGOs are also flexible. Development NGOs begin from problems on the ground among the population of their concern. Unlike government organizations, NGOs are not constrained by predetermined agendas, sectors, or ODA mechanisms. NGOs address problems creatively by mobilizing the resources that are available to them.

In the context of international health, diverse NGOs can make various contributions. Examples include : NGOs' advocacy role in generating support from the international community to produce anti-retro viral drugs for HIV/AIDS, despite strong resistance from the pharmaceutical industry; fast and flexible actions to meet humanitarian needs in emergency situations; and bridging the gap between ODA schemes for emergency and rehabilitation phases.

While NGOs have the potential to contribute to the advancement of people's health globally, we face a number of serious challenges in Japan. To maximize the potential of Japanese health-oriented NGOs, we must overcome three major challenges : accountability, attracting and retaining good human resources, and strengthening organizations.

S1-5) INTERNATIONAL HEALTH COOPERATION IN THE REFORM OF THE JAPANESE UNIVERSITY SYSTEM

NOBUO OHTA

Molecular Parasitology, Nagoya City University Graduate School of Medical Sciences

Many basic/bench researchers in the field if medical sciences are, if not all, not so willing to join international health cooperation activities under the current situation of the Japanese University system. When period of leaving laboratory is long, it frightens their career-up and/or keeping academic position for those researchers, because they are evaluated by indicators such as number of publications, impact factors and so forth. Such survival competition is becoming much more tight in the recent on going reform of the University system in our country. COE 21 or the term contract of university staff in these years pushes them to be more research-oriented. This means that the recent education policy is inhibiting motivation of basic/bench researchers to join international health cooperation activities. University has stock of information, knowledge and technology, and there is no question that those could make contributions for international cooperation. In the Japanese ODA scheme, the basic concept is technical transfer. It is, however, obvious that techniques without scientific background have no big applicability. In this sense, bench researchers at universities should be encouraged to join international cooperation activities. On the other hand, basic researchers become highly motivated when they experience field activities. Field activities could be stimuli for their research in laboratory, and this is surely consistent with the policy of University system in our country. Together with those situations, I would like to propose the future direction for university research staff. The most important point to be considered is incentive for bench researchers. They hate the negative evaluation of their lowered productivity in publication during the period of being out of laboratory. Evaluation system should be, thus, changed, and also ODA scheme, for instance, is also needed to think about their difficult situation of survival in universities. Through more close discussion and mutual understanding between university and the cooperation agencies and/or the field staff, we have to build a new scheme of international health cooperation in the new Century.

S1-6) VIEWS OF AN INSTRUCTOR ON INTERNATIONAL HEALTH AND MEDICAL SERVICE COOPERATION

MIZUKO TOKUNAGA

Department of Nursing, School of health science, Nagasaki University, Nagasaki, Japan

I conducted a questionnaire survey among 1st and 2nd year nursing students to inquire their view toward international cooperation. Of 127 respondents, 91% were interested in international cooperation, and 66% were interested in working in developing countries in the future. They thought it because 40% considered it challenging, 34% wanted to work for the underprivileged, 12% wanted to make use of their skills in developing countries. Their choice of the region (where they wish to work) is Asia (44%), Africa (33.3%) and Latin America (13%). As a prerequisite for taking part in the international cooperation, 24% of the respondents answered the ability of using the second language, 24% knowledge on public health, 23% knowledge on nursing, and 18% knowledge on tropical diseases. The respondents reported that they want more information regarding international cooperation (48%), an opportunity to participate in the training abroad (17%) and learn from those who are involved in international cooperation (15%). The university should provide them with further information and opportunity that help them learn about international cooperation.

Symposium 2

S2-1) WHAT CAN WE DISPATCH FROM LABORATORIES TO THE FIELDS IN THIS GENOMIC ERA?

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²Department of Parasitology and Tropical Public Health, University of Occupational and Environmental Health, Kitakyushu, Japan

The final goal of the basic research on Tropical Medicine will be to elucidate the diseases from which many people in the tropics are suffering and consequently to overcome the diseases by sharing the fruits of the research. Current trends of the basic researches on tropical infectious diseases are on genomes of both infectious agents and human host. Because of the recent establishment of genomic structures of the pathogens such as *Haemophilus influenzae* in 1995 up to *Plasmodium falciparum* in 2002, and indeed, whole genome structure of the human host in 2001, we can say that we are really in the genomic era.

However, on the other hand, environmental factors which support the epidemics or prevalence of the diseases are regarded very important in the 21st century. Researches on ecological environment as well as socio - economic factors are also of great interest for controlling the tropical infectious diseases. Therefore, the questions are always asked on what we can dispatch from laboratory basic science to the field. Here in this workshop, big debate were made among speakers and audience. The topic will be a continuing theme which is to be discussed by members of both Japanese Society of Tropical Medicine and Japan Association for International Health.

S2-2) DEVELOPMENT OF MOLECULAR DIAGNOSTIC METHODS FOR DETECTING FLAVIVIRUS INFECTIONS

ICHIRO KURANE

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Flaviviruses include nearly 70 species of viruses, and approximately 40 are believed to cause illness in humans. Dengue, Japanese encephalitis, West Nile, yellow fever and tick-borne encephalitis viruses are important pathogens among flaviviruses. Laboratory tests are essential for confirmation of infections by these viruses. Serological tests are most widely used and known to be reliable. However, presence of flavivirus-cross-reactive antibodies often confuses the determination of infecting viruses, although the levels of cross-reactivities vary depending on the methods. Further, serological diagnostic methods usually require two blood specimens for confirmation; specimens collected at acute and convalescent stages. Isolation of viruses is the most definite method for determining the causative agent. Infection can be confirmed using only one specimen. However, it takes time and requires an adequate facility. Molecular diagnostic techniques have been developed and widely used in many laboratories.

We have been developing molecular diagnostic techniques for flaviviruses and attempting to transfer the techniques to other laboratories. The molecular techniques can be applied not only to the determination of the infection in humans, but also to the confirmation of the absence of viral contamination in transfused blood, or determination of infected mosquitoes or animals in vector surveillance. Classical reverse transcriptase-polymerase chain reaction (RT-PCR) and TaqMan PCR for dengue viruses and West Nile virus have been developed in our laboratory. In RT-PCR using the primer set covering the E gene ; WNNY514 : Cgg CgC CTT CAT ACA CW (mixed primer) and WNNY904 : gCC TTT gAA CAg ACg CCA TA, West Nile virus was specifically detected. The sensitivity was 0.2-10 plaque forming units (p. f. u.) depending on the strains. In the TaqMan PCR, the sensitivity for West Nile virus, New York strain was 0.1 p. f. u. West Nile virus, Eg101 strain was detected with a low sensitivity. West Nile virus, g226 and FCG strains and other flaviviruses were not detected by this system. Therefore, this system is highly specific and sensitive for some strains of West Nile virus, but not specific for other strains of West Nile virus or other flaviviruses. Thus, further modification is needed to cover all the West Nile virus strains.

Molecular diagnostic methods surely have many advantages over the other diagnostic techniques. However, it is expensive and requires sophisticated equipments. When these techniques are transferred to the laboratories in developing countries, various factors including cost, training and maintenance of the machines should be considered.

S2-3) ROLE OF LABORATORY IN THE FIELD STUDY OF DIARRHEAL DISEASES

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Division of Bacterial Pathogenesis, Graduate School of Medicine, Univ. of the Ryukyus, Okinawa, Japan

Diarrhea is one of the main causes of morbidity and mortality in Tropical countries. Early childhood diarrhea is associated with fitness impairment, cognitive impairment or reduction of immune response. Among the etiologic agents, diarrheagenic Escherichia coli is a major cause of diarrhea. Diarrheagenic E. coli is classified into at least 5 categories and identification of those strains needs to detect the pathogenic properties of the organisms. However, cost and labor make the detection difficult especially in small laboratories of the Tropical countries. In the detection of enteropathogenic E. coli (EPEC), serogrouping of O-antigen is usually carried out, but reliability of O-serogrouping is controversial, because O-antigen does not satisfactorily correlate with the presence of virulence factors. Therefore, identification of diarrheagenic E. coli needs to detect factors that determine the virulence of these organisms. Thus, we developed a new practical reversed passive latex agglutination (RPLA) method to determine EPEC by detecting the virulence factor EspB.

With the popularization of PCR, it has become possible to detect pathogenic genes in bacterial isolates, allowing the rapid diagnosis of diarrheagenic E. coli. We also developed a multiplex PCR assay to detect 6 virulence genes for the categorization of diarrehagenic E. coli in a single reaction tube. Recently, pulsed-field gel electrophoresis is used in a number of large laboratories for epidemiological study of bacterial isolates. However, this method requires special expertise, expensive equipment or reagents. We are attempting to develop an inexpensive and technically accessible epidemiological tool for Vibrio cholerae using antimicrobial susceptibility as epidemiological marker. Our laboratory has cooperated with centers or institutes in Asia countries such as Center for Laboratory or Epidemiology in Laos and National Institute of Health and Epidemiology in Vietnam. In this opportunity, the role of advanced laboratory for the field study of diarrheal diseases introducing our fieldwork in these countries will be discussed.

S2-4) POST GENOME STUDY FOR FIELD APPLICATIONS

SHINJIRO HAMANO

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Post genome study, which develops from deciphered genome information, rings over the extremely wide fields, not only bioscience but also medicine, dentistry, pharmacy, agriculture and engineering and is expected to be a key to solving global problems such as diseases or chronic scarcity of food.

Post genome study also began in our study field along with the open of the genome information of *Homo sapiens* and pathogenic microbes such as *Plasmodium falciparum*. Our final purpose is the prevention, the diagnosis and the treatment of infectious diseases in the field. We are requested to prepare scientific basis for battle against infectious diseases by effectively using genome information of the host and the pathogenic microbes, which, in turn, should help individuals and society suffering from infections. The answer to the question how we can prepare scientific basis is strongly dependent on the background of each scientist. As the theme of this symposium is "From a laboratory to the field" and my special field is "Immunity to the infectious agents", I would like to introduce the study stressing the vaccine development.

Leishmaniasis, which is a disease caused by a protozoan parasite, genus *Leishmania*, is transmitted by sand fly in both old and new world. More than ten million people are suffering from it. The drugs against *Leishmania* often show severe side effect and genus *Leishmania* frequently acquires resistance to the drugs. Therefore, we decided it as a vaccine target disease. The key of the vaccine development lies in the proper understanding of cellular immunity, in which IFN-gamma plays central role. Although IL-12 is the most important cytokine for induction of IFN-gamma production, IL-12 is not always sufficient in some situation.

Recently, novel members of IL-12/IL-12 receptor family, IL-23/IL-23R and IL-27/WSX-1, were identified through the homology search of the genome database. For screening of such candidate molecules, homology search using genome information shows its power. As a result of extensive investigation, we elucidated that IL-27/WSX-1 is required for protection against infection with *Leishmania major* and *Trypanosoma cruzi*. We are further investigating the function of these cytokines for clinical application in the near future and believe that the contribution to the field from a laboratory can be achieved by these approaches just as well in post genome era.

S2-5) FROM THE *PLASMODIUM* GENOME RESOURCE DATABASE TO CLINICAL PRACTICE : APPLICATION OF REAL-TIME QUANTITATIVE PCR FOR DETECTION AND DISCRIMINATION OF GENETICALLY DIFFERENT MALARIA PARASITES

SHINICHIRO KAWAZU

Research Institue, International Medical Center of Japan

Genome sequence data for *Plasmodium falciparum*, the parasite responsible for more than 95% of all malaria deaths, have been available in the on-line database PlasmoDB since the end of 2002. Provision of this database should be of major benefit to us. Efficient use of the data should aid us greatly in the control and eradication of malaria.

We have begun to exploit the genomic information in the database in the diagnosis of malaria. That is, we have applied real-time quantitative PCR with allele - specific probes (TaqMan, registerd trademark, ABI) for detection and discrimination of genetically different malaria parasites. We first used small subunit ribosomal RNA (SSUrRNA) genes of the 4 human-infective malaria parasites (*P. falciparum*, *P. vivax*, *P. malariae* and *P. ovale*) as the targets to be amplified and discriminated in clinical blood samples. We were able to detect and discriminate *P.* *falciparum*, *P. vivax*, and *P. ovale*. The PCR system was sensitive enough to detect *P. falciparum* in 10 micro liter of cultured parasite-infected blood with 0.0001% parasitemia.

We then used the *pfcrt* gene, which has a strong relation to the chloroquine resistance of the parasite, for detection and discrimination of drug-resistant malaria parasites. The gene with the single nucleotide polymorphism that alters the PfCRT protein was amplified from the drug-resistant parasite DNA and was discriminated by the allele-specific probe. We were able to detect and discriminate chloroquine-resistant *P. falciparum* in 10 micro liter of cultured parasite-infected blood with 0.0001% parasitemia. We are planning to apply the PCR system for analysis of genetically mixed malaria parasite infections in clinical blood samples.

Symposium 3

S3-1) CLINICAL APPROACH OF TROPICAL INFECTIOUS DISEASES

KAZUNORI OISHI

Department of Internal Medicine, Institute of Tropical Medicine, Nagasaki University

S3-2) CLINICAL AND MICROBIOLOGICAL CHARACTERISTICS OF COMMUNITY-ACQUIRED BRONCHOPULMONARY INFECTIONS AMONG HIV-INFEVTED PATIENTS IN NORTHERN THAILAND-ANALYSIS OF STREPTOCOCCUS PNEUMONIAE AND RHODOCOCCUS EQUI-

NORICHIKA ASOH¹, HIROSHI WATANABE¹, KIWAO WATANABE¹, KAZUNORI OISHI¹, WEERAYUT KOSITSAKULCHAI², TIPPAYA SANCHAI², SUMPUN KAHINTAPONG², PRASIT THARAVICHITKUL³, THIRA SIRISANTHANA³, TSUYOSHI NAGATAKE¹

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Background : We investigated the pulmonary infections among HIV-infected patients between December 1996 and January 2002 in Thailand. Since *Streptococcus pneumoniae* and *Rhodococcus equi* were common organisms, we conducted the following study.

Purpose : The aim of our study is to examine the antimicrobial susceptibility and serotype distribution of *S.pneumoniae*, and the mutations in the *rpoB* gene associated with rifampicin resistance in *R. equi* isolated from humans in Thailand.

Methods and Results : Ninety-three *S. pneumoniae* strains in Thailand between September 1999 and June 2000 were investigated. Of the 93 isolates, 29 (31.2%) were susceptible, 24(25.8%) showed intermediate resistance, and 40(43.0%) were fully resistant to penicillin G. The predominant serotypes were 6A, 11A, and 19A in susceptible strains, 6B and 23F in intermediately resistant ones, and 6B, 19F, and 23F in fully resistant ones. Molecular typing by pulsed-field gel electrophoresis (PFGE) of multidrug-resistant pneumococci showed four patterns (A, B, C, and D) for 16 isolates of serotype 19F, with pattern B being predominant (12 isolates). Eleven isolates of serotype 23F showed two patterns (F and G), with pattern F being predominant (seven isolates). Moreover, the an-

timicrobial susceptibility of 30 R. equi isolates from 30 humans between 1993 and 2001 was investigated. Of the 30 isolates, 26 were susceptible, one showed a low-level resistance (MIC, 8 microgram/ml) and 3 showed a high level resistance (MIC, more than 64 maicrogram/ml) to rifampicin. PCR amplification and DNA sequencing of the rpoB gene, and molecular typing by PFGE were performed in 8 R. equi isolates from 8 AIDS patients with pneumonia or lung abscess caused by R. equi, including one low- and 3 high-level rifampicin-resistant isolates. Two high-level rifampicin-resistant strains with PFGE pattern A had a Ser531Trp (Escherichia coli numbering) mutation and one high-level rifampicin-resistant strain with PFGE pattern B had a His526Tyr mutation, whereas one low-level rifampicin-resistant strain with PFGE pattern C had a Ser509Pro mutation. Four rifampicin - susceptible strains with PFGE patterns D and E showed the absence of mutation in the rpoB region.

Conclusion : Our results indicates that the resistance of pneumococci to antibiotics is progressing rapidly and that effort should be intensified to prevent any spread of pandemic multidrug-resistant serotype 19F, 6B, and 23F. Also, our data indicates the presence of several types of rifampicin-resistant *R. equi* among AIDS patients in northern Thailand.

S3-3) PREVALENCE AND ULTRASONOGRAPHIC HEPATOBILIARY FINDINGS OF LIVER FLUKE, OPISTHORCHIS VIVERRINI, IN LAO VILLAGES

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Infection with the liver fluke, *Opisthorchis viverrini*, remains a major public health problem in Lao P. D. R. Cholangiocarcinoma caused by the liver fluke is the most common form of liver cancer in Northeast Thailand where an about 60 to 70% of the population are infected with the fluke. The parasite is also common to the low-lands of Lao among people with close ethnic ties to northeast Thais, but the prevalence and incidence of the infection in Lao are still not uncertain. The objective of our report is to investigate the prevalence of chronic infection with *Opisthorchis viverrini* and to identify chlanginocarcinoma within a population-based survey of the infection in Lao.

Stool samples from 670 residents (0-87 y. o) from 3 villages in Khammouane Province were examined for intensity of liver fluke infection. People from varying egg count categories were selected for ultrasound examination

to identify hepatobliary disease. Patients coming to provincial hospital with abdominal symptoms were examined in the same way as the residensts were done. The rate of the parasite was 56% among the residents and reached up to 73% in age group greater than 20 years old. The parasite was found even in 20% of age group below 5 years. Only one subclinical case of cholangiocarcinoma were diagnosed from a total of 158 people based on ultrasonographic evidence. Dilation of intrahepatic bile ducts was observed higher within the moderate and heavy liver fluke-infected group in spite of symptoms with and without symptoms.

This survey suggests prevalence of the liver fluke is higher than previously thought. It also suggested the intensity of the parasite was related with the formation of hepatobiliary lesion, although it could not elucidate the relation with cholangiocarcinoma.

S3-4) IMPORTED INFECTIOUS DISEASES : MALARIA

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In order to clarify the characteristics of imported malaria cases in Japan, we analyzed 218 patients who had visited our hospital for these 11 years (1992-2002). The number of malaria cases in our hospital was from 12 to 29 (mean 20.0) per year, which makes up 17.5 % of total cases in Japan (calculated based upon the number of total patients in Japan ; 114 per year reported by Kimura et al.). Although the number fluctuates year by year, we did not find no obvious increase or decrease in the number. The proportion of Japanese cases was approximately two thirds of total cases, and no increasing tendency was observed. As for malaria classification, P. falciparum made up more than 50 % after 1994 except 1996. This frequency of P. falciparum was similar to that in British

Kingdom and Germany, and higher than total Japanese cases. Africa was the most common areas where patients were infected, and Pacific areas including Papua New Guinea were the second. Drugs used for treatment was analyzed and it was found that chloroquine usage declined year by year. Although, for these 11 years, we experienced 4 severe malaria cases (3 acute renal failure cases and one cerebral infarction), all of them recovered completely. Mortality reported from United Kingdom and Germany was 0.65 % and 3.6 %, respectively, suggesting that we have relatively favorable treatment results although the number of patients was small. Together with information from other hospitals as well as nation-wide surveillance by Kimura et al., the characteristics of im-

S3-5) DENGUE AND DENGUE HEMORRHAGIC FEVER

KUNIKO YOSHIDA

AIDS Clinical Center, International Medical Center of Japan

BACKGROUND: Dengue fever ranks highly among emerging infectious diseases in public health significance. Dengue virus which has four major serotypes is transmitted by the mosquito, *Aedes aegypti*. New infection with a particular serotype causes antibody-dependent enhancement. Dengue fever (DF) causes negligible mortality, but re-infection to the other serotype leads to the risk of developing more severe Dengue haemorrhagic fever (DHF) and Dengue shock syndrome (DSS).

METHODS : We reviewed 8 cases who were consulted to our clinic after returning from tropical and subtropical countries and diagnosed Dengue fever between July 2000 and May 2003.

RESULTS : Among 8 cases,3 were male and 5 were female whose mean age was 25.7 years old. 5 cases from Southeast Asia, 2 from Central America and 1 from Oceania. On avarage, it took 11 days from onset to consultation. To differentiate dengue cases, clinical evaluation and laboratory diagnosis were used. 7 cases (87%) complained high fever at the first examination. The most common presenting features of Dengue were thrombocytopenia (87%) and leucopenia (75%) in laboratory. Clinically, high fever (87%), rash (72%), myalgia (50%), headache (25%), pain in orbit (13%), visual impairment (13%) were inspected. Those who complained visual impairment were pointed as hemorrhage in the eyeground. Hospitalization was needed for 6 cases (75%). The Dengue cases were classified as DF (3 cases, 37.5%), DHF (5 cases, 62.5%), and no DSS. All cases were recovered with conservative treatment and their mean duration with symptoms was 15 days. With serological confirmation, primary antibody response was observed in 6 cases (75%). Dengue virus isolation were done for 5 cases, 4 (80%) were positive. Dengue serotypes were; 2 cases were type1, 1was type 2 and 1 was type4.

CONCLUSIONS : The prevalence of dengue fever appears to be increasing in travelers, which calls special attention for health care professionals. The clinical presenting features provide important guides to establishing the diagnosis.

S3-6) TRAVELER'S DIARRHEA

TAKUYA ADACHI

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Diarrhea is by far the most common health problem encountered with travel to developing countries. 298 cases of traveler's diarrhea and 9 cases of typhoid and paratyphoid fever were summarized as intestinal infection. All patients were affected with either diarrhea or enteric fever during or shortly after their trip overseas and visited the Department of Infectious Diseases at Yokohama Municipal Citizen's Hospital from January 2001 to June 2003. The most common countries of disease acquisition were Indonesia (20%), Thailand (16%), and India (14%). 82% of all cases were acquired in Asian countries. From 133 among 307 patients (43%), 149 strains of causative agents were identified. The leading causes were Shigella (20%), diarrhogenic Escherichia coli (17%), Campylobacter (15%), nontyphoidal Salmonella (11%), and Salmonella Typhi and Paratyphi A (6%). 80% of causative agents were bacteria. Protozoa and parasites were less likely causes, accounting for 19% of infection, including 9% of giardiasis. The most important factor in treating diarrhea is the replacement of lost fluids. Antibiotics are usually not required with acute watery diarrhea, which are usually self-limited with supportive treatment. If blood occurs in the stool, or if there is fever, empiric antibiotic therapy should be considered. A short course of fluoroquinolone can be considered in such cases. There have been increasing cases of enteric fever with decreased susceptibility to fluoroquinolones. Resistance to nalidixic acid on disk diffusion susceptibility testing and clinical fluoroquinolone treatment failure are the key findings of decreased susceptibility strains. Intravenous administration of a third generation cephalosporin in addition to fluoroquinolones is useful to treat such resistant strains. There is a tendency that the longer the traveler stays abroad the greater the risk is to acquire enteric fever or intestinal parasite infection. Attention to food and beverage preparation can decrease the likelihood of developing traveler's diarrhea. Bacteriological stool test is incredible for appropriate diagnosis and treatment.

S3-7) THE INITIAL CLINICAL MANAGEMENT OF COMMUNICABLE DISEASE PATIENTS ENTERING JAPAN

EMIKO IWASAKI

Director of Sendai Quarantine Station, Miyagi, Japan

With continuous developments and wide spread transportation, the situation surrounds us continue to change. These have also caused great changes in communicable diseases. The development has resulted in spreads of sever and highly contagious diseases to every corner of world. In Japan, about 18 million people visit overseas every year. Unfortunately most of traveler lack knowledge of health management and risk associated with travel. Therefore there are some cases that get communicable disease overseas unluckily.

Moreover we have 5 million visitors from foreign countries, and they also have chances to bring communicable diseases into Japan. In Japan, the fact that there are not many medical doctors who can diagnose communicable diseases overseas because of a lack of experiences with these diseases makes situation more complicated.

Under this social background, for preventing public from communicable diseases overseas, we have to provide information to public on prevalence of communicable diseases, vaccination and health consultations before departure and after return using internet, FAX, phone and brochure. And also we have to support health management of public for preventing spread of diseases and instruct them to receive adequate treatment as quickly as possible.

At clinics and hospital, all medical stuffs have to recognize that many of communicable diseases have possibilities that it belong to international communicable diseases coming from overseas. And therefore they should pay attention to take care patients, learn how to protect themselves from communicable diseases and prepare for infection control at medical institutions.

Today, the problem of communicable diseases is not a problem just for one country. It is time to think this problem globally and act globally with international cooperation.

Symposium 4

S4-1) EXECUTIVE SUMMARY OF THE ROUNDUP SESSION OF NINE WORKSHOPS : RECOMMENDATION FOR THE FUTURE PERSPECTIVES ON TROPICAL MEDICINE AND INTERNATIONAL HEALTH

YASUHIDE NAKAMURA

Resrearch Center for Civil Society, Graduate School of Human Sciences, Osaka University

The workshops for nine areas or countries are a specific challenge of the Joint Conference of Japanese Society of Tropical Medicine and Japan Association for International Health. In each workshop, the facilitator made a review of health-related problems and summarized the recommendations in consideration of the relationship between the biomedical approach and the public health approach. The facilitators of the nine workshops presented their summaries and recommendations in the roundup session. In this roundup session, we concentrated on the discussion of the following topics : 1) Communication among researchers, field workers and policy makers is essential. We need to strengthen the mutual understanding among researchers of tropical medicine and international health. 2) Researchers and field workers can

frequently work together, because the targets of tropical medicine and international health are very similar. However, their approaches are very different. 3) Both tropical medicine and international health should work together to encourage the younger generation who are keen to work in developing countries in the future. Continuous efforts are crucial in promoting the relationship between tropical medicine and international health, because their objectives are almost of one voice. We sincerely wish to ensure the quality of life of people in developing and developed countries

Forum

F1) JAPANESE PREDECESSORS CONTRIBUTED INTERNATIONALLY TO PUBLIC HEALTH AND INTERNATIONAL HEALTH—; MESSAGES TO THE NEXT GENERATIONS.

SUSUMU WAKAI

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Five predecessors contributed internationally to the public health and international health presented their outstanding experiences in this field. Dr Shimao tackled against tuberculosis to combat against its insurgency in the early fifties and sixties, while Dr Arita dedicated to eradicate smallpox followed by elimination of polio (WHO declared its eradication in WPRO region in 2001). Dr Irie has been working for tackling micronutrient deficiencies. Dr Kita has been committed herself to emergency relief activities around the globe. Dr Kawahara established an NGO working together with people in Asian countries (AHI or Asian Health Institute). They presented their expertnesses and shred their ideas with next generations.

Workshop 1

W1-1) BANGLADESH

NOBUKATSU ISHIKAWA

Research Institute of Tuberculosis, Japan Anti-Tuberculosis Association

Health indices such as total fertility rate, maternity mortality rate have been improving in the last several decades in Bangladesh. Discussion was made how GO or NGO health projects have been contributing to the health development in the country. Four speakers presented their experiences in health projects as below and discussed the role of the international cooperation in Bangladesh. Several projects and programs show a good success story. However there still exists a weakness in administrative capacity managing the health programs especially in local governments. On the contrary, NGOs activities have been widely and remarkably developed to innovate in or supplement government programs, though the beneficiaries and the long-term sustainability are often limited. Flexible approaches combining them would bring a successful outcome. Human resource development and collaborative research should be the center. Importance of laboratory and scientific technology has been shown in the newly arising environmental problem of arsenic pollution. BRAC experience shows that a success pilot project

W1-2) CURRENT CONDITION OF REPRODUCTIVE HEALTH IN BANGLADESH, EFFORTS GIVEN BY THE GOVERNMENT AND SUPPORTS BY DEVELOPMENT PARTNERS, AND PERSPECTIVES FOR SUPPORT

could lead a larger fund available.

YURIKO EGAMI

Japan International Cooperation Agency Bangladesh Office

Bangladesh has witnessed a great success in decreasing TFR and MMR in the last 30 years, but yet the situation is to be improved, with the annual maternal deaths of 25,000 which counts for 5% of all maternal deaths globally. Under Health and Population Sector Program (1998-2003), the Government developed Maternal Health Strategy in 2001, and development partners give various supports to improve the reproductive health. Japanese Government is supporting to develop the network of EmOC, human resources development, and the grass root level approach in improving the access to reproductive health services. The approach from both service providers and beneficiaries is necessary to promote the safe motherhood. The policy that meets with the needs of clients and community will be developed, and the accountability of service providers should be encouraged. At the same time, it would be necessary to encourage the society that allows the womens autonomous behavior change seeking for better reproductive health. The role of us, the international health workers, would be to support identifying the constant policy based on the clients needs and the scientific findings. The strategy should be developed to identify the methodology and indicators to decrease the maternal mortality should be developed and analyzed. Secondly, the advocacy to secure the necessary human resources and budget should be essential. Thirdly, we should review the several success stories in Bangladesh and various lessons learned by development partners, and find components which can be applied to improving the reproductive health in Bangladesh.

W1-3) INTERNATIONAL COOPERATION FOR A MISSION HOSPITAL IN BANGLADESH BY A JAPANESE NGO

YASUO OYAKE

Japan Overseas Christian Medical Cooperative Service

I worked for a mission hospital in the south-east part of Bangladesh for two years and ten months from September 2000 to May 2003, being sent by a Japanese NGO. The hospital is located in a rural area with 6 Bangladeshi doctors and 120 beds. The job description was to set up the paediatric ward and to provide education about paediatrics to the staff, including doctors and nurses. The three major diseases among paediatric patients in the area were malaria, acute respiratory infection, and diarrhoeal disease. The three major causes of death among paediatric patients were perinatal problems, malaria and acute respiratory infection. I introduced some simple equipment like bed nets, a refrigerator and body weight scales into the paediatric ward. It took much longer for the nursing staff to be able to use them properly than I had expected. If it was the first time that they had seen the equipment, even if it was simple, it was not easy for them to operate it properly.

Seeing so many seriously ill patients in the hospital, I started to go to villages once a week to provide health education, especially for prevention of malaria. In addition, I introduced the Integrated Management of Childhood Illness (IMCI) to the health workers from the department of public health in the hospital. Conclusion :

It is important that Japanese staff work with their counterparts to teach, rather than simply provide financial

assistance. Education is the best way to improve healthcare when resources are very limited.

W1-4) ARSENIC CONTAMINATION IN BANGLADESH AND THE ACTIVITY OF ASIA ARSENIC NETWORK

KAZUYUKI KAWAHARA

Asia Arsenic Network, Miyazaki, Japan

There are about 10 million tube wells in Bangladesh. Twenty five percent of them are contaminated with arsenic of over permissible level (0.05mg/l), and 30 million people drinking the dangerous water face the risk of arsenocosis diseases. Melanosis and keratosis will be the first symptom of chronic arsinic poisoning. In many years, there is high probability of contracting cancer of skin, lung, liver and urinary organs.

Asia Arsenic Network (AAN) is conducting an arsenic mitigation project in Sharsha Upazila, Jessore District, collaborating with JICA. AAN has performed the investigation of thirty two thousand tube wells in Upazila with a field kit. The tube wells of over standard level of arsenic contamination were painted red, the ones under the standard level were painted green, according to the result. Besides this, AAN has installed alternative water supply options such as rain water harvest, modified dug well, pond sand filter and deep tube well.

The function of Local Government in Bangladesh is ineffective, posting only one assistant sub engineer in charge of the water supply in Sharsha Upazila with three hundred thousand population. A lot of local NGOs are concerned with the arsenic mitigation but they do not seem to have enough financial capacity to do by themselves without funds. It is critical to use Local Government in order to carry out the sustainable measures of arsenic mitigation. The most important point of JICA/AAN project is to support the Local Government, and promote the community participation.

W1-5) ROLE OF NGO AND COLLABORATIVE RESEARCH IN DEVELOPING NATIONAL TB PROGRAM : EXPERIENCE OF BRAC

ISLAM AKRAMUL

Bangladesh Rural Advancement Committee

BRAC is one of active NGOs working for health in community development projects in Bangladesh. BRAC small-scale pilot community based TB program in a subdistrict for 200,000 populations was initiated in 1984 with the support from JOCS and RIT in Japan. Health volunteers (SSs) were mobilized as cores of diagnosis and treatment in a community. The model was extended to 11 sub - districts in 1999, and then further extended to 126 sub - districts by 2003 covering 30% of the country population under the special collaboration with the government. A further expansion is underway. Government provides drugs and consumables, technical guidelines, management of referral cases, training of doctors and lab. Technologists, and supervision. The success of BRAC in scaling up TB program is due to : a) BRAC commitment to work as a partner, b) learning from pilot projects, c) supports form development donors, and d) long term support from Research Institute of TB in Japan.

Workshop 2

W2-1) WORKSHOP : RURAL HEALTH : KEYWORD OF HEALTH DEVELOPMENT IN CHINA

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China is achieving remarkable economic growth and being said, "no need of international support any more". However, some specific social problems still can be identified. First is the poverty in rural area, which weakens village health system. Poor villagers can't pay for health cost and village doctors are not enthusiastic for less earning job. Poverty also causes second problem ; it's the floating population. Such jobless people are directing to urban areas in east or developing areas in west to get job. They are often socially unregistered and can't be covered by public health services. Third, health system in China is vertical and not disclosed well. Completeness, adequateness and promptness of health information are sometimes unreliable. In addition, paradoxical social factors such as globalization and nationalism, tradition and modernization, and socialism and market economy are making the situation more complicated.

and more focused, and the strategies should be changed and specified. Based on the activities done, three presenters clarified the expected approach for social health development in China. They are summarized as follows :

(1) Target population : rural and floating population, especially in western poverty provinces,

(2) Capacity building and human exchange under equal partnership rather than one-way donation,

(3) Operational researches to develop realistic measures,

(4) Rural development through self reliance and community participation,

(5) Facilitation of inter-province, inter-institute collaboration in two countries, and,

(6) Academic exchange in specific areas : Infectious diseases such as ARIs, SARS, and HIV/AIDS, and other common health problems in both countries.

Therefore, international resources should be shifted

W2-2) ACTIVITIES OF POLIO CONTROL PROJECT IN CHINA, AND ONGOING WORK

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 ⁴Team leader, Expanded Programme on Immunization Strengthening Project in China

Expanded Programme on Immunization (EPI) was launched in China from the 1980s and gradually extended all over the country. However there were many unvaccinated children, an unintended consequence of the "one child policy", which had led parents, particularly in rural areas, to avoid registering their children. This vulnerable population was attacked by wild polio viruses during 1989's outbreak throughout China. This outbreak resulted in 5,065 polio cases during 1990.

Our polio control project was started in 1990 in Shandong province, located in eastern China, as a pilot project of control of this disease. Activities of this project were gradually expanded to the neighboring provinces, and finally covered a total of 11 provinces with a population of 600 million, by the end of the project. This project has supported the introduction of AFP surveillance system, construction of a polio laboratory network and vaccine supply for NIDs and SNIDs, etc. As a result of the efforts toward polio eradication, a wild polio virus has not been isolated from specimens of AFP patients in China, except in imported cases, since 1996. The government of China declared the eradication of polio in the meeting of WPRO held in Kyoto city in 2000.

Our ongoing project began in 2000 to assure safe injections of vaccines, to strengthen activities of other vaccination programmes, including, measles, HBV, and others, in addition to the consolidation of polio free areas.

W2-3) SEEING THE CHINESE FARMERS' HEALTH PROBLEMS FORM THE INVESTIGATION OF ARSENIC POLLUTION IN INNER MONGOLIA

XIAO JUAN GUO¹, YOSHIHISA FUJINO², KIYOYUMI SIRANE², DONGYUN ZHAO³, TETSUYA KUSUDA⁴, TAKESUMI YOSHIMURA²

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Chronic arsenic contamination in Inner Mongolia was first reported in 1990. It is now known that more than 410,000 persons are chronically exposed to well water highly contaminated with arsenic. We conducted surveys of drinking water and health in Wuyuan County in Inner Mongolia from 1993 to 2003. The results indicated that there were 66 arsenic-affected villages out of 679 villages, and in the affected villages, 90.6% of wells have water with an arsenic concentrations higher than 50µ g/L. The prevalence of arsenical dermatosis was as high as 45.5% in arsenic-affected villages. Other subjective symptoms were also more common, compared with the arsenic-free villages. These multiple systems lesions pose an important public health problem. Local people are af-

fected in life, labor and the marriage of youngsters, since advanced forms of keratosis are painful, and the consequent disfigurement can lead to social isolation. The last two decades have been important in the development of primary health care in China. Recently, however, health insurance has not been widely available in rural areas. This, along with the rapid increase in health costs and insufficient medical facilities have all made it difficult for villagers to visit doctors. Consequently, they suffer from their diseases until they cannot bear them. This trend may lead to declining health and increased mortality. Good medical service and strict public health policy are key in improving the health condition of rural farmers in China. This is an important theme to be resolved in future.

W2-4) A RESEARCH EXCHANGE BETWEEN JAPAN AND CHINA – AN EXPERIENCE ON TUBERCULOSIS CONTROL

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China has the second highest tuberculosis burden in the world. According to the fourth nationwide prevalence survey conducted in 2000, the prevalence of active, bacteriological positive and smear positive pulmonary tuberculosis were 367/100,000, 160/100,000 and 122 / 100,000 respectively. In 1992, the DOTS strategy was initiated in 13 provinces, supported by a loan from the World Bank. In 2001, the DOTS population coverage was

68% and the DOTS Detection Rate was 29% with 95% Treatment Success Rate in the new smear positive cohort in 2000. In 2002, Japan began supporting the expansion of DOTS in China through the Grant Aid project, providing anti-tuberculosis drugs and microscopes to 12 provinces. In addition to those activities, Japan has been supporting human resource development, which includes group training at the Research Institute of Tuberculosis, Japan Anti-Tuberculosis Association supported by JICA, and several trainings for the introduction of the DOTS. Recently, research activities have grown through joint research on the implementation of DOTS, tuberculosis control in urban areas, and so on. Research exchanges have also taken place, such as exchanges of researchers among institutions, and the organization of the Academic Forum of China - Japan - Korea Tuberculosis Research Institutes. Although China has made great progress in tuberculosis control, additional operational research is needed for further achievement in some areas such as the low DOTS Detection Rate, the quality of the microscopy network, the referral system from hospitals to tuberculosis control, etc.

Workshop 3

W3-1) PACIFIC ISLANDS AND HEALTH DEVELOPMENT

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People living in the Pacific Islands have established their own way of life under a variety of geographic and ecological conditions. Christianity and modernization have had a great impact on the transformation of their behavior patterns. From an epidemiological point of view, there is a distinct difference in mortality patterns between Polynesia and Melanesia. In Polynesia, with a history of Christianity and modernization going back more than 200 years, people are suffering from dramatic increases in chronic diseases that are categorized as post - epidemiological transition. In contrast, in Melanesia, which has only recently been modernized, the leading causes of mortality are infectious diseases, and the life expectancy at birth is lower than in Polynesia. Obesity in Tonga and malaria in PNG and the Solomon Islands are representative causes of health loss resulting from post- or preepidemiological transition, respectively. To control obesity, body weight reduction through changes in consumption behavior is virtually the only option. In Melanesia, distribution of insecticide-treated bed-nets and ensuring their regular re-treatment with insecticides are the main tools for malaria prevention. Helping the local people develop the habit of sleeping under bed-nets is a key strategy for malaria control. Changes in behavior patterns, however, are problematic and will require decades to achieve, since the behavior patterns are closely related to culturally characteristic cognitive structures, traditional habits and social beliefs. We thus recommend employing an integrated approach in the life-style management rather than relying on a single existing measure.

W3-2) 1. HEALTH IN OCEANIA : BACKGROUND HISTORY AND ENVIRONMENT

MINATO NAKAZAWA

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Oceania is divided into three sub-regions : Melanesia, Polynesia, and Micronesia. The oldest roots of the Melanesian population left Southeast Asia 40,000-60,000 years ago. Their descendants now mostly live in PNG Highlands and southern coastal regions (except for Australian Aborigines). Other Melanesian people are descendants of the Lapita cultural complex coming from Southeast Asia around 3,000 years ago. These people also populated Samoa and Tonga at about the same time through long-distance canoeing, where they faced a new environment. After 1,000 years, subgroups sailed to other Polynesian islands. Micronesian islands have been populated during 3,500 to 2,000 years ago : some from Melanesia or Polynesia, some from the Philippines, with genetic mixture. Malaria is absent in Polynesia and Micronesia because of the absence of the vector, Anopheles mosquitoes. PNG Highlands are also malaria-free, but other PNG and Solomon Islands are highly malaria - endemic. There are occasional malaria outbreaks triggered by deforestation, which allows breeding of mosquito larvae. People in malaria-endemic area have a high prevalence of G6PD deficiency, because of its resistance against malaria. In PNG and Solomon Islands, malaria and many other infectious diseases played a great role in people's health. People in other parts of Oceania suffer from obesity. Lapita people may have developed "thrifty" genetic features of low-energy expenditure during their long-distance migrations. With the introduction of a fat rich diet including foods like hamburgers, they tend to develop obesity and its associated diseases. It is important to assess health status in relation to the environmental settings and people's lifestyles.

W3-3) MALARIA CONTROL STRATEGY IN MELANESIAN COUNTRIES

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In Papua New Guinea, malaria eradication programme was established in 1950s, continuing until mid -1980s based on residual house spraying with DDT, together with active and passive case detection by microscopic diagnosis. Because cost-effectiveness of the programme was in question, impregnated bed nets (IBNs) have become the principal measure for malaria prevention since 1988. However, IBNs coverage is still low (less than 10% of population at risk). Indoor house spraying is carried out in specific situations, for example epidemics in highland regions. In Solomon Islands, DDT spraying was applied as the principal anti-malarial measure from 1969 to 1980s. In contrast to PNG, IBNs coverage attained 70% within ten years after IBNs programme introduced in 1989. In Vanuatu, DDT spraying was applied in 1973. However, spraying programme was evaluated to have little or no impact on local malaria endemicity. Therefore, this programme was withdrawn in 1982 and focal spraying has been carried out since then to prevent outbreaks. First impregnated bed net programme stared in 1988 and 70% of population at risk became to use IBNs in late 1990s. For uncomplicated malaria treatment,

chloroquine was widely used as a first drug in Melanesian countries. First case on chloroquine-resistant falciparum malaria was identified from late 1970s to early 1980s (1976 in PNG; 1980 in Solomon Islands; 1982 in Vanuatu). Combination therapy of sulfadoxine-pyrimethamine with chloroquine is now implemented as a first-line treatment (1991 in Vanuatu; 2000 in PNG; 2001 in Solomon Islands). The present malaria control activity in Melanesian countries is based on Roll Back Malaria strategy : rapid diagnosis and treatment ; better multi pronged protection using impregnated bed nets and environmental management. Compared to Sub-Saharan Africa, malaria mortality rate is relatively low despite high level of endemicity. Therefore, radical malaria control programme aimed at cessation of malaria death, reduction of incidence, and reduction of malaria parasite transmission may be possible if high level of community involvement is maintained. Some isolated islands in the region can be favorable environment to reduction of malaria transmission; however, it is a difficult task to extend the programme region-wide because of extensive geographical and cultural diversities.

W3-4) RECONSTRUCTION OF HEALTH SYSTEM SINCE THE ETHNIC CONFLICTS IN THE SOLOMON ISLANDS

TAKAO MOCHIDA

Embassy of Japan in Solomon Islands

The resources for the health system have been limited in the Solomon Islands. The ethnic conflicts further devastated the health system that consequently diminished the public health services. The Japan Embassy in the Solomon Islands is currently funding the project for malaria control there. The strategy of the project is mobilizing available human resources nationwide who are familiar with the relevant knowledge and technologies for both diagnosis and treatment of malaria cases, even though their resources are limited. The project also aims at improving the accessibility to the health services in the community, thereby encouraging voluntary health seeking behaviors. As the situation is chaotic, the embassy's assistance should be flexible beyond the existing framework of international cooperation.

W3-5) NUTRITION AND HEALTH PROBLEMS IN WESTERNIZING SOUTH PACIFIC – WITH SPECIAL REFERENCE TO TONGAN OBESITY AND DIABETES

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Investigation research on nutrition and health problems in the South Pacific (especially Polynesia) can be reviewed as follows. Firstly, despite arguments on the best index of obesity, the prevalence of obesity evaluated by BMI (body mass index, Weight/Height²) in Polynesia is generally very high compared with other people in the South Pacific. In this respect, females are more obese than males, while obesity prevalence is higher among city dwellers than isolated islanders. Moreover, Polynesian migrants in U. S. A. or New Zealand also become obese, which turns into a big health problem in those countries. Secondly, Polynesian obesity relates to many health disorders including diabetes mellitus (type2 diabetes, NIDDM), but its association with those death rates is not decisive. Thirdly, this obesity is caused by the combination of many genetic and environmental factors. A lot of obesity related genes (so-called "thrifty genes" suggested by J. V. Neel in 1960s) have been investigated, however, their effects on obesity have not been confirmed yet.

Fourthly, changes of food consumption and subsistence activities accompanied by the changes of their life style with westernization are significant environmental factors affecting obesity. In Polynesia, furthermore, psychocultural factors admitting or rather admiring obesity are especially important promoting factors for obesity. Finally, it has recently been pointed out that body composition of Polynesians are basically different from Caucasians or Mongoloid, i. e., Polynesians show less fat percent and more muscularity for their BMI in comparison with other ethnic groups.

Since late 1990s, we have carried out studies on life style and health condition of the Island people and city dwellers of the Kingdom of Tonga, one of Polynesian countries. This report aims firstly to clarify how the life style of the Tongans has changed by showing some historical data and materials, secondly to show the results of health examinations conducted in Ha'ano Island (rural), Nuku'alofa capital city (urban) and Kolovai village

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(sub-urban). Tongans are genetically homogeneous, and thus any health differences detected between rural and urban residents should be attributed to those in environmental factors (those in life style such as food consumption and subsistence activity) and psycho-cultural factors. This report also examines the relationship between obesity related genes and the prevalence of BMI or DM. Using data set above mentioned, it was tried to make an obesity model for Tongans, which is urgently needed to prevent their obesity by altering their psycho-cultural attitude toward obesity.

Workshop 4

W4-1) HEALTH PROJECTS IN THE CENTRAL AMERICA AND CARIBBEAN REGION

TAMOTSU NAKASA

Buereu of International Cooepration, International Medical Center of Japan

[Introduction] Since 1980's several health projects by JICA have been implemented in the Central America and Caribbean region. Following characteristics in this region should be considered in order to implement health interventions appropriately.

1. The dominant and most available language is Spanish.

2. Political changes occur every four years in most countries.

3. Main products are coffee, banana and woods.

4. This region is a back-yard of USA and strongly influenced by it.

[Characteristic of Project] Three projects were presented.

1. The medical education project by Ohita meical university by Tetsu Yamashiro The cooperation started in the field of digestive organs and was extended to medical education including public health by hole participation of faculty member of Ohita medical university. 2. The regional health project in Nicaragua by Fumie Takagi Integration approach was adopted to involve the every component of the health system including the community and the project has strong relationship and coordination with the national research center and the faculty of medicine.

3. Chagas Disease Control in Guatemala & CA by Yoichi Yamagata Tripartite strategy (joint activities by MOH, PAHO and JICA) in the central level and intersectorial cooperation strategy in the local level were implemented.

[Conclusion] Considering frequent changes of policy makers in Central and Caribbean region, strategies suited to local reality should be implemented through human resource development and capacity building. Also these strategy should be informed and socialized directly, indirectly and continuously to decision makers through the channels of influenced donors and educational institutions.

W4-2) OVERVIEW FOR TWO PROJECTS IMPLEMENTED IN DOMINICAN REPUBLIC –CENTER OF GASTROENTEROLOGY AND CENTER FOR POST-GRADUATE MEDICAL TRAINING-

TETSU YAMASHIRO

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Oita Medical University was initially concerned with the Dominican Republic (DR) at 1990 through a developing aid project, The research and Clinical project for Gastroenterology which had been implemented for 7 years in the country. According to a census conducted by the Department of Epidemiology, Ministry of Health at the late 1980s, acute diarrhea placed the first place in terms of the incidence and the infant mortality rate, and the countermeasure of which was urgently needed. After the Center for Gastroenterology had been established in the Dr. Luis E Aybar hospital by the grant aid from Japanese government, more than 100 university personnel including medical doctors, registered nurses, medical technicians, and epidemiologists had participated in the project as experts in order to reinforce particular fields through transferring techniques or providing instruction to DR counterparts. At the end of the project, however, unsatisfactory level of fundamental knowledge of DR counterparts in the field of the radiology and the epidemiology was pointed out as an impediment for management of the hospital, and their improvement were strongly recommended. Taking the situation into consideration, the Japan-DR cooperative training center for Medical Education (CEMADOJA) was established in the property of Dr. Luis E Avbar hospital supported by the grant aid of Japanese government. A new project, The project for reinforcement of the medical education, aiming principally at reinforcement of 1) comprehensive diagnostic ability using radiological images and of 2) epidemiological skill, for resident doctors as well as medical staff has been implemented since 1999, principally in the CEMADOJA. The center consists of two departments, the department of radiology and the department of epidemiology where didactics, teaching aid, techniques and equipment needed for training of the resident doctors are transferred to DR counterparts who were expected to be professors of the center. The department of epidemiology has a laboratory which is supposed to collect epidemiologically convincing data by which practical epidemiology would be provided to the resident doctors. One of the principle reasons for successful implementation of those comprehensive activities in the project was probably attributed to a substantial support of Oita Medical University to the management of the project. So far, a scientist who belongs to a university has been restricted to be involved in a developing aid project individually; however, the university unity, in turn, is expected to participate in the substantial management of a project on its initiative.

W4-3) THE PROJECT FOR STREGTHENING THE LOCAL SYSTEM OF INTEGRATED HEALTH CARE (SISTEMA LOCAL DE ATENCIÓN INTEGRAL A LA SALUD : SILAIS) OF GRANADA, NICARAGUA

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The province of Granada has a population of 190,000 people (urban 60%, rural 40%). There are 33 public health facilities at the primary level, one at the secondary level and one laboratory center. Most people use the public health services, because they are free of charge. The major health problems are maternal and perinatal mortality and infectious diseases transmitted by mosquitoes. JICA (Japan International Cooperation Agency) is conducting a health project with the Ministry of Health (MOH), SILAIS Granada, for strengthening the local system of integrated health care, focusing on maternal and child health. The purpose of the project is to improve the quality of health services for children under age 5, and women of reproductive age, particularly at community and primary-level health care facilities. We anticipate four outcomes of this project: 1) increased

community participation for the solution of health problems, 2) improved capability of health teams at the primary level, 3) development of a referral and counter - referral system among the community, primary and secondary levels and 4) improved managerial capabilities of health management teams at headquarters and all municipal offices. In Nicaragua, the MOH is conducting many programs and projects with international organizations and donor countiries. At the local level, SILAIS Granada must carry out many tasks required by MOH central authority with limited heath resouces. Supporting the health strategy of MOH, our project coordinates with other programs and projects working with the Health Research and Study Center of Nicaragua and other sectors, like the Ministry of Education.

W4-4) STRATEGY OF JICA PROJECT FOR THE CONTROL OF CHAGAS DISEASE IN CENTRAL AMERICA

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The Chagas Disease Control Project of JICA began in Guatemala in 2000 and was extended to Honduras and El Salvador in 2003. The purpose of this paper is to share the strategies of the Project with those people working in or for JICA and struggling toward the internationalization of JICA.

The strategies followed dialectical thinking in three steps. Transition from traditionally vertical structure of the vector control to a modern horizontal health system was reinforced by the dispatch of Japanese managers, who made temporary "oblique" information channels. The lack of specific knowledge and skills of the Japanese generalists was supplemented by the local body of knowledge and that of Pan American Health Organization (PAHO). The gap of institutional culture between PAHO and JICA was quickly filled by emphasizing the mutual benefit of the joint venture. JICA benefited from PAHO in terms of on-the-job training of the Japanese personnel, as well as of political influence of PAHO to the recipient countries, which otherwise may change their policies. PAHO benefited from JICA's execution of PAHO's policy of Chagas disease control.

In order to play a vital role in the global partnership towards poverty alleviation, JICA needs to reshape her policies, institutions and human resources. This study suggests that JICA can perform in Latin America by following PAHO's policies, utilizing flexible resources such as volunteers, and training them on the job.

Workshop 5

W5-1) TECHNICAL COOPERATION IN HEALTH AND DEVELOPMENT IN VIETNAM

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Occupational Safety and Health Specialist, ILO Subregional Office for East Asia, Thailand

Vietnam has accelerated its self-help efforts in health and development. Rural health care service networks have been strengthened, enlarging immunization programmes and lowering the rates of infectious diseases. Access to basic sanitation facilities such as safe drinking water or hygienic latrines has been widened. Successful technical cooperation programmes have strengthened the local self-help initiative. Vietnam is now increasingly facing new challenges in health and development. The new challenges reflect the changing socio-economic situations of the country such as the widening of the marketing economy, rapid urbanization and industrialization, and the modernization of agriculture. We may be able to point to three major challenges. First, the strategies to tackle against emerging diseases such as HIV/AIDS need to be further strengthened. Changes in the socio-economic conditions and people's behaviour are important contributing factors. The transmission mode of old diseases like tuberculosis is also becoming diverse reflecting such changes. Second, the quality of health care services requires priority attention. People expect warm and human-oriented clinical services. Expanding the health insurance coverage to ensure equal health care services to the people with different economic situations is another challenge. Third, Vietnam has to fight against the new health risks because of industrialization and urbanization. Adverse health effects brought by environmental pollution and hazardous working conditions present typical problems. These three areas of challenges commonly need the strong concerted efforts among scientists, medical care providers, public health practitioners and policy makers. International technical cooperation programmes with Vietnam need the renewed mindset to reach their changing targets.
W5-2) EPIDEMIOLOGY OF MALARIA IN ENDEMIC AREAS OF VIETNAM.

SHUSUKE NAKAZAWA

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To elucidate who provides gametocytes to vectors, malaria survey is conducted in an endemic area in the south Vietnam as a research project in the exchange program titled' Analysis of various factors on emergence and re-emergence of tropical infectious diseases and their control strategy' supported by the Japan Society for the Promotion of Science. People in Phu Trung and Phu Rieng villages can be divided into three groups according to their living places and ethnicity, villagers (Kim tribe), new immigrants (Kim tribe), and Xi Tieng people (Ethnic minority). After obtaining written informed consents, people are interviewed for collecting demographic data and diagnosed by Giemsa stained thick films. At the same time, blood is taken on a filterpaper. DNA is extracted from a dried spot of blood on the filter paper for PCR diagnosis and genotyping of Plasmodium falciparum. RNA is extracted for evaluating gametocyte counts and levels of cytokines. Sera are extracted for measuring specific IgG against erythrocytic forms and gametocytes of Plasmodium falciparum. Plasmodium ovale and Plasmodium malariae are detected more frequently in Xi Tieng people than other two groups. Different gene types of resistance makers are observed among the three groups. Levels of Th1 related cytokines are higher in Xi Tieng people than in other two groups. There is no difference in Th2 related cytokines among the three groups. Levels of serum antibodies against crude antigens of Plasmodium falciparum are increased, as people grow older. Xi Tieng people show the highest levels of the antibodies among the three. To detect asymptomatic carriers of parasites, body temperature is measured when people diagnosed by Giemsa stained blood films. More than a half of slide positive school children in Xi Tieng people have parasites in spite of normal body temperature. These results suggest that parasites are maintained inside the group and that asymptomatic carriers may play an important role in reserving parasites.

W5-3) BACH MAI HOSPITAL ACTIVITIES IN NURSING FIELD

SACHIKO MIYOSHI

Bach Mai Hospital Project for Functional Enhancement

The Bach Mai hospital was established in 1991 and has provided medical care services mostly for the patients coming from the northern part of Vietnam.

JICA's technical cooperation project started in January 2000. The overall goal of the project is to enhance the quality of medical care services and extend its effects to the northern provinces in Vietnam. The project activities include hospital management, nursing management, various clinical activities. The project is also interested in decentralization. JICA experts have come to work in these fields.

After renovating the hospital, the Bach Mai hospital turned to be a general hospital with 1,400 patient-beds with approximately 1,500 staff members. In addition to the clinical activities, the hospital has many other functions, such as training for medical and nursing students,

and providing post-graduate training.

The project is now implementing total care activities. Every year, the total care seminar is organized to report implemented activities and to exchange information with other hospitals. In the nursing field, the training subcommittee was established in December 2000. This subcommittee is carrying out training for the nurses, implementing specialized care activities and promoting nursing research among the head-nurses. Besides, a variety of efforts have been made to improve the patient care by the nurses.

It is necessary to write a report on change in mind and thinking manner of the hospital staff members as well as on professionalism, and on the roles of the nurses for the total care activities.

W5-4) ILO'S TECHNICAL COOPERATION IN OCCUPATIONAL SAFETY AND HEALTH IN VIETNAM

TSUYOSHI KAWAKAMI

Occupational Safety and Health Specialist, ILO Subregional Office for East Asia, Thailand

Occupational safety and health (OSH) is an emerging concern for the socio-economic development of Vietnam. Rapid industrialization and modernization of agriculture have been bringing diverse risks of work-related accidents and diseases to everyday life of local people. Mushrooming small enterprises in Vietnam need acute attention to ensure safe and healthy work environments. Priority areas include safe use of machines and chemicals, elimination of ergonomic hazards, and establishment of productive and human-oriented work systems. Farmers require practical assistance to cope with new safety and health risks such as the use of dangerous machines, agro chemicals or electricity. Construction and mining sites face special hazards and needs strong risk-reduction programmes. The ILO is strengthening its OSH technical cooperation programmes in Vietnam. The scope of the ILO programmes covers both policy and workplace levels.

At the policy level, development of a medium-term national OSH programme with clear targets has been encouraged. ILO Conventions on Safety and Health No. 155 and on Safety and Health in Agriculture No. 184 and other relevant ILO instruments have been referred to. At the grass-root workplace, participatory, action-oriented training programmes directly to workers and employers have been promoted. ILO's WISE (Work Improvement in Small Enterprises) programme is assisting an increasing number of small enterprises in implementing OSH improvements using locally available resources. The ILO technical cooperation needs to be accelerated focusing on people's own initiative and sustainability. Positive inter action between the policy and workplace programmes should be enhanced for the wider coverage of practical OSH protection.

Workshop 6

W6-1) WORKSHOP 6 : ZAMBIA AND MALAWI WHAT IS EFFECTIVE AND WORTHWHILE COOPERATION WITH AFRICA ?

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The aims of this workshop were ; 1) to deepen our understanding of the actual situation of health in Zambia/Malawi and of the cooperation from Japan. 2) To discuss how should we act to cooperate with Africa, which has many difficult issues, and "far" from Japan.

Wakasugi suggested the points of consideration such as, what is effectiveness and being worthwhile for the cooperation with Africa? and how to challenge lack of human resources in Africa? Then we had the presentations of four speakers;

Suzuki presented the achievement of PHC project implemented in urban slums in Zambia, which are IMCI through community-based growth monitoring program, access to safe water /hygiene and the application of GIS to PHC.

Wakasugi presented a trial of laboratory-based program approach "from VCT to Care" for the control of HIV/AIDS and TB in Zambia, which aims to support consistently the process from VCT as an entry point until care and treatment.

Nakano presented the results of needs assessment study for health policy and master plan in Malawi. Six prioritized projects concerning major health issues such as child's nutrition, safe motherhood and drug management were proposed.

Sugishita presented his clinical experience of many

AIDS patients as JOCV in Malawi and proposed an anthropological approach to grasp an emerging faith healing of people facing with the threat of AIDS. Finally we had many comments from speakers and the floor regarding how to challenge lack of human resources and how should we act in Africa.

W6-2) SEVERAL APPROACHES OF THE PRIMARY HEATH CARE PROJECT IN URBAN SLUM OF ZAMBIA

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The situation was found to be worse in Lusaka urban area with its extremely high density of population. Approximately 75% of the population of this area was living under extremely poor health conditions in the urban area that was called "Compound". Lusaka District Primary Health Care Project intended to improve the capabilities of communities, health centers, and particularly of LDHMT. The Project in the past indicated a various positive developments on water and sanitation, growth monitoring program, referral system, and school health service.

With regard to water and sanitation issues, water supply system provided by Japan's Grand Aid Project was effectively followed up by door-to-door health education on usage of safe water as well as on hand washing with soap, and contact tracing/fumigation during the cholera outbreaks. The empowered residents of George Compound succeeded in construction and management of ventilated improved pit (VIP) latrines, drainages and stations for solid waste disposal. As a result, cholera outbreaks have not occurred since then.

WHO and UNICEF develop Integrated Management of Childhood Illness (IMCI) as a strategy to improve childhood survival. This strategy is rationalized as the management of patients at first-level facilities to try to reduce childhood mortality. A well-planned seminar and in-service training was provided by the Project and LDHMT partnership in Zambia. However, those project activities did not carry out effectively.

The project developed new concept and approach for IMCI, what we called GMP^+ . GMP^+ (growth monitoring program plus) was integrated program for child health of under-5 children with growth monitoring, nutritional consultation, immunization, vitamin A supplementation conducted in the community in collaboration with health center staff and community based organizations. Resulting from the provision of GMP^+ in the past, underweight prevalence among unser-5 children declined form 23% in 1999 to 15% in 2000, full immunization overage of under-1 children increased from 15% to 61%, and the measles incidence among under-5 children declined from 5.5/1,000 to 1.8/1,000.

In conclusion, we would like to emphasize that GMP^+ developed in the project may be an useful vehicle to transfer other important messages related in health promotion such as HIV/AIDS, safe motherhood, and family planning. Therefore, we have a future plan to expand GMP^+ program to other areas of Zambia.

Acknowledgements

The project was carried out in collaboration with the Association of Medical Doctors of Asia (Dr. Suganami), JICA experts, and Dr. T. Umenai.

W6-3) A TRIAL OF A CONSISTENT PROGRAM APPROACH "FROM VCT TO CARE" FOR HIV/AIDS CONTROL IN ZAMBIA

NAOMI WAKASUGI

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Situation of HIV/AIDS in Zambia

In Zambia, HIV prevalence in adult is estimated to be 19.95% (UNAIDS2000) ranked fifth in the world and an estimated 1 million people are now living with HIV/AIDS.

JICA project for HIV/TB control

Following Infectious disease control project, which lasted for 11 years, JICA has restarted HIV/AIDS and TB control project from April 2001 with the virology laboratory at University Teaching Hospital as Zambian counterpart. The objectives are; 1) to strengthen and sustain the capability of laboratory systems, and 2) to effectively utilize them for the control of HIV/AIDS and TB.

Program approach "From VCT to Care"

The global situation for HIV/AIDS has recently drastically evolved. The launch of the Global Fund in

2001 and 3 by 5 initiative adopted by WHO targeting to treat 3 million with ARV by 2005. Thus Africa is trying to get out from a mere prevention based on a resigned treatment. Zambia government committed to treat 10,000 people with ARV.

In such situation JICA project is proposing a program approach model, which aims to support consistently the process beginning from VCT as an entry point until care and treatment of infected people. This includes promotion of VCT access, laboratory support for when and how to start ARV therapy, monitoring the effect and the resistance emergence, capacity building of health staffs providing ART, DOTS-like community & home based care, and so on. This should be conducted making full use of JICA schemes such as grass-root technical cooperation and AIDS-JOCV.

W6-4) EXPERIENCE OF HEALTH SECTOR DEVELOPMENT STUDY IN MALAWI

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Between June 1998 and January 2000, "The Master Plan Study on Strengthening Primary Health Care Services (JICA PHC Study)" was executed in the Republic of Malawi. The purpose of the study was to formulate a central region master plan and a national master plan to improve the primary health care (PHC) system, and to transfer essential research, analytical skills, and methodologies to Malawian counterparts. The health investigations covered the referral system, human resources, health finance, policy and management, the health management information system, community participation, logistics systems and health facilities and supplies. For the formulation of the national master plan, three core health issues, including childhood malnutrition, maternal health, and the role of informal drug-sellers' in the PHC system were selected, and finally six prioritized projects were proposed. As a result of the JICA PHC study, the following characteristics of the health situation in Malawi were observed. We found a significant gap between better physical access to health facilities and poor health indicators. Access to good quality health services is impeded by insufficient human resources and poor infrastructure. The health finance and health systems are strongly dependent on donor agencies, and management capacity is low. In developing an aid policy for the health sector in Malawi, we concluded that it is important to strengthen the PHC system, support human resource development for the improvement of health services, and address the problem of infectious diseases such as HIV/AIDS, malaria and tuberculosis along with the basic development approach for the African countries.

W6-5) HIV/AIDS CLINICAL EXPERIENCES AND A PROVISION ON MEDICAL ANTHROPOLOGY IN MALAWI

TOMOHIKO SUGISHITA

JICA Tanzania Morogoro Health Project

In terms of magnitude and scale of the HIV/AIDS pandemic, Sub-Saharan Africa suffers more than any other regions, with 29.4 million people currently thought to be living with this disease. Throughout Africa, the talk one hears about the pandemic evokes many connotations : fear, despair, suspicion, oppression, stigma, inequality and poverty. In these circumstances, while I worked for Zomba Central Hospital as a surgical specialist during 1995-1998 and 2001, I realized that despite their despair, people are vigorously promoting practical knowledge to sustain their substantial mundane lives. Healing practices, both through traditional healers and through a consecutive movement of African Independent Churches (Zionism, Born-again), have gained popularity in recent years. Here, the discursive knowledge around the pandemic was strategically manipulated by signifying practices based on their fundamental cosmology. Actually they make sense of the way of living positively in the era of AIDS and globalization. On encountering the pandemic, healing rituals strive to humanize the symbolic meaning of HIV/AIDS and to mediate connoted sufferings through an innovative transformation between the indigenous context of tradition and the present context of modernity. Moreover, these dynamics illustrate well the flexible interaction between the medical and religious realms. However, these positive and innovative aspects of the response to the pandemic have been neglected in public health academic discourse. To better serve those who suffer from HIV/AIDS, health authorities and researchers must understand the development of indigenous functional autonomy and its background context, which can be best understood by anthropological approach.

Workshop 7

W7-1) SUMMARY OF THE WORKSHOP ON INDONESIA

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Indonesia has shown rapid growth in economic development, while the disparity among populations has been expanded. Economic crisis, conflicts and natural disasters have endangered human security.

The primary concern of the workshop was the role of Japan as the top donor to Indonesia, in terms of; what we have done, and how we can improve our approaches in health sector.

Prof. Koji Kanbara presented his experience in the JICA partnership project on 'malaria control' in Lombok and Sumbawa; Prof. Ikuko Moriguchi presented about human resource development for primary health care taking lessons from her projects in Sulawesi; and Dr. Yoko Watanabe focused on MCH and reproductive health,

introducing the project on MCH handbook.

In the panel discussions many important suggestions were made from both panelists and audience, among which the followings were particularly emphasized. (1) The external assistance should help closing the internal gaps which are often recognized between the initiatives at central level (top-down approach) and those at front-line level (bottom-up approach). (2) JICA's schemes, including JOCV and policy advisor, should be effectively linked or efficiently combined to make approaches objective oriented. (3) pursuit for the goals with perspectives of longer time span is required, (4) Japan's ODA should develop its own capability of assistance in terms of know how, human resources and aid schemes.

W7-2) JICA PARTNERSHIP PROJECT "MALARIA CONTROL IN LOMBOK AND SUMBAWA ISLANDS"

HIROJI KANBARA

Institute of Tropical Medicine, Nagasaki Univ., Nagasaki, Japan

The JICA partnership program is quite unique in the point that a project is selected by JICA from projects proposed by Japanese NGOs including universities and is implemented by the NGO which proposed it, with financial support from JICA. We, the Institute of Tropical Medicine, proposed the project "Malaria control in Lombok and Sumbawa islands" on the basis of our collaborative malaria studies from 1991 to 2000 at Lombok and Sumbawa islands with Tropical Disease Center (TDC), Airlangga University, Indonesia. The project was accepted by JICA and negotiations for an international agreement between Indonesian and Japanese government were executed by JICA. We have begun our activities since October, 2001. First of all we discussed the exact process of the project with Indonesian organs concerned, namely, the provincial health office of Nusa Tenggara Barat (NTB), the district health office of Lombok Barat, the health center of Meninting (the project area), and TDC, and responsible sections of Ministry of Health, and decided parts of the control work and a responsible person of each part. Through activities during these two years, we have noticed the strong and weak points of the JICA partnership project as follows.

1. Strong points

1) The project is dependent on an NGO's idea but not on JICA's request, and carried out by the NGO.

2) The NGO feels more responsibility for the project than for a contracting project.

2. Weak points

1) Executive processes are regulated by both JICA and the belonging organ, especially financial practice, and employment.

Our project contains practical aspects and basic studies, indicating intimate cooperation between members of Society of Tropical Medicine and International Health will be helpful for implementation of this kind of project. Meanwhile, there are many political and social problems in present Indonesia for malaria control activities. One is the problem related to the recent policy of decentralization that made communication between the central government and the local government difficult. The other is the problem related to a variety of social and natural environments that causes different types of malaria endemic. The above problems make malaria control measures complicated and difficult. To solve them we regularly hold the meeting in which representatives from all the organs concerned participate.

W7-3) TECHNICAL COOPERATION IN THE FIELD OF MCH/REPRODUCTIVE HEALTH IN INDONESIA

YOKO WATANABE

Children's Medical Service Section, Medical Service Division, Bureau of Public Health, Tokyo Metropolitan Government

Background Improving MCH/ reproductive health is a high priority in Indonesia, especially with this country's high maternal mortality rate. To help with these efforts, Japan has been assisting in the development of a MCH handbook program, using many resources, since 1993.

Characteristic of Japan's ODA of the MCH handbook program 1) Designed for poor people, so its

target was mainly users of public health center and community maternal post. 2) The purpose of the project was to ensure quality of MCH service through using MCH handbook. Consequently it improves mother's knowledge of MCH. 3) In addition to two technical cooperation projects, there were grants to provide equipment for FP and population activities, dispatch of individual experts, JOCV, and country specified training in Japan (MCH).4) The project activities cover management of the MCH handbook program, development of training materials, and coordination of other donor agencies.

Collaboration between the project and JOCV 1) Since 1999, total 13 JOCVs (midwives, nurse, and nutritionists) have been dispatched to six provinces. 2) Unlike the project activities, JOCVs' activities have been done at the front line of target people using the MCH handbook. 3) The project and JOCVs conducted joint meetings regularly to exchange information and learn from each other.

Outcome of the MCH handbook program1) As of March 2003, 197 among 350 districts are utilized MCH handbook. About 1.4 million copies are printed by JICA, UNICEF, ADB, local governments, and NGOs. 2) Mothers' knowledge on MCH was improved by using the MCH handbook.

W7-4) THE IMPORTANCE OF PHC AND INTERNATIONAL COOPERATION FOR IMPROVEMENT OF PHC WORKERS

IKUKO MORIGUCHI

College of Nursing Art and Science, Hyogo

I have been working in Indonesia since 1984 to improve the training and effectiveness of PHC workers. There have been roughly four phases to this work.

I cooperated in developing community-oriented nursing education, based on PHC and Japanese PHN experience in a nursing training program, as a JICA expert since 1984. New PHC practices were introduced, including community surveys and training of health volunteers. As a result, the students took a greater interest in PHC and community nursing.

Indonesia began deploying community midwives in rural areas for comprehensive PHC service in 1990. To assess the difficulties they encounter in their work, we collaborated in a fact-finding survey from the PHC perspective in South Sulawesi 1992-1996. The survey revealed the need for an assessment of health needs and for supervision of TBA. The midwives also wanted on-the - job training and support from the Health Center.

The JICA Project for Improvement of District Health Services in South Sulawesi began in 1997. We have tried to implement methods to improve the competence of midwives through action research in a pilot district. We provided training for "verbal autopsy". This is done by home visits and case conferences after a maternal death has occurred, allowing the identification of the apparent cause and factors involved. This method has also become popular in other districts.

We started "Nursing in PHC" training for Indonesia nursing leaders in Japan in 2001. In the future, we hope they will take the initiative in PHC and improve the quality of PHC workers.

Workshop 8

W8-1) HEALTH CARE AT THE FOOT OF THE HIMALAYAS

KATSUNORI OSUGA

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The Kingdom of Nepal kept the door closed to the rest of the world until 1950s. When the door was opened, mountain climbers, explorers, and researchers started to visit this country of beauty. Surprised at the health conditions, some individuals and groups offered help to the local communities. Dr. Noboru Iwamura is by far the best known Japanese among those involved in international health. His struggle to help people fight against tuberculosis has continued in JICA projects focusing on TB control in Nepal. Starting with an overview of the Japanese assistance in TB control, this workshop invites speakers involved in various fields of health in Nepal. Dr. Fukai presents dental health based on the dental camp experiences. Although health status of the laborers has become an important issue with the economic progress in recent years, this aspect of health has not been addressed sufficiently in developing countries. Dr. Yoshikawa shares his experience in this field. The gender aspect is discussed by Ms. Murakami, with her experience in a female literacy program in a rural community. Traditional healers cannot be ignored when health-seeking behavior is considered. Dr. Jimba presents his experience in involving traditional healers in health service delivery in Nepal. Attracted by the mountains, peaceful life, and the rich culture of the country, many Japanese visit Nepal. Many are also involved in medical care and health in Nepal. This workshop expects an active discussion, sharing experiences and ideas for the improvement in health status in Nepal.

W8-2) TUBERCULOSIS CONTROL IN NEPAL

KATSUNORI OSUGA

Dept of International Cooperation, Research Institute of Tuberculosis, Japan Anti-Tuberculosis Association, Tokyo, Japan

It may have been only in recent years that tuberculosis began to spread in Nepal. Dr. Iwamura, the first Japanese doctor in Nepal, began his 14 year-struggle against this disease in the 1960s, when the kingdom first opened up to the world. Nepalese doctors began attending the international TB training courses, which have been organized since 1963 at the Research Institute of Tuberculosis, Tokyo. The first bi-lateral collaboration commenced in 1979 as a JICA public health project. Focusing on TB control, this project continued until 1985. To strengthen the National TB Program, the National Tuberculosis Centre was constructed under the grant assistance scheme in 1989. The JICA TB project was implemented from 1989 to 1994, and then extended to 2000, when the new treatment strategy "DOTS" was introduced. The proproject was a success, with an 86.6% treatment success rate among a sputum smear positive pulmonary TB cohort registered from 1996 to 1998. Global attention began to focus on TB control during this time. Many donors and UN agencies began to provide support for TB control in Nepal, in addition to the international NGOs and Japan. The progress of "DOTS" was remarkable, gaining international recognition in recent years. The JICA project, started in 2000, is focused on "lung health" in the community : TB control in urban settings, control of childhood ARI, and community-wide anti-smoking campaigns. Success of TB control in Nepal was due to the commitment by the community as well as the strong leadership of the TB program.

W8-3) IMPLEMENTATION AND EVALUATION OF INTERNATIONAL COOPERATION IN THE FIELD OF ORAL HEALTH

KAKUHIRO FUKAI

Association of Dental Cooperation in Nepal, Kitakyushu, Japan

Nepal, like other developing countries, faces limited resources and personnel for dental care. The levels of dental caries are still generally lower in developing countries than in the majority of developed countries. However, data from many studies support the findings that levels of dental caries are increasing in urban areas of developing countries, where people have easy access to sugar - containing products. The gap between developed and developing countries is increasing with urbanization. Evidence - based preventive measures of oral disease (fluoride application, sugar control, and effective teeth cleaning) have helped to control oral disease in developed countries. The Association of Dental Cooperation in Nepal has performed 16 dental missions since 1989. A total of 446 Japanese volunteers have worked in Nepal, serving 11,761 dental patients and providing 31,117 Nepali people with health education and health care. The activities of ADCN include dental treatment, school dental health, maternal and child health, and the training of oral health workers. Among these projects, dental treatment is the most important to the Nepali people, who have little access to dental care. In the face of limited resources, much greater efforts should be directed towards preventive measures, in line with the concept of primary care and health promotion. We recommended that Nepali dentists take on the role of community-based dentists, who act as leaders in providing primary oral health care and advocate for community participation in oral health. This process would be achieved through a dependent health projects stage, a collaborative stage, and finally a selfreliant stage.

W8-4) DEVELOPMENT OF THE PARTICIPATORY SAFETY AND HEALTH-TRAINING PROGRAMME FOR TRADE UNIONS IN NEPAL

TORU YOSHIKAWA

The Institute for Science of Labour

The paper reports the results of action-oriented occupational safety and health training seminars and discussions their effectiveness in Nepal. Methods : A pilot seminar held in 2000 led to a new training package using the Participation-Oriented Safety Improvements by Trade union Inisiati VE (POSITIVE) methodology for trade union members (Nepal Trade Union Congress : NTUC). Many seminars were organized in different regions jointly by branch offices of the NTUC and the Japan International Labour Foundation (JILAF). Participants were trained to identify and implement low-cost improvements using locally adjusted training tools. Training seminars of 1-4 days were organized to facilitate the safety and health action of workers. Each seminar was aimed at (1) learning from local good examples; (2) applying an action checklist and group dynamics; and (3) increasing trade

union initiative in making practical improvements. *Re-sults*: POSITIVE seminars held in cooperation with different regions in Nepal. They were trained 112 POSI-TIVE trainers. It was found that 21% of trainers trained about 4-day training skills within 18 month, and that 74% trainers trained about 1-day training skills within 30 months had taken voluntary actions of occupational safety and health improvements. It was reported that 748 workers participated to the 1-day seminar during a year (2001 - 2002). *Conclusions*: Effective training of trade union members could be achieved by applying participatory methods focusing on low-cost improvements in a developing country. These experiences may be widely applicable to in promoting occupational safety and health activities in Nepal.

W8-5) HEALTH PROMOTION THROUGH WOMENS ADULT LITERACY PROGRAM

IZUMI MURAKAMI

Japan International Cooperation Agency Bangladesh Office

The School and Community Health Project (SCHP) started form 1992 in Nepal. Its objectives were to strengthen the primary health care activities. Then in second phase, objectives were expanded to improve the

communitys health through health promotion activities and improve health services. Womens Literacy Program was implemented to support above objectives. Implementation process was assessed to know impact of the program. SCHP conducted total 495 different levels of literacy classes with life skill trainings over the 6 years to more than 3500 women. Literacy rate in project area increased from 10 percent to 50 percent. As a result, large numbers of women become able to take action on many of their health and household matters. These women have also built the confidence, developed leadership qualities, managed various income generation programs and improve the kitchen gardens in their homes. Factors which

contributed for this success programs were, supervision which focus on encouragement, needs based delightful activities and integrated community development, health promotion programs such as schools facilities improvement, Child to Child Program, disaster preparedness program etc. Womens literacy can activate community empowerment and it is enforce community development. This can be one of the models for Health Promotion Approaches.

W8-6) SUSTAINABILITY OF TRADITIONAL HEALER TRAINING IN NEPAL

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Traditional healers (THs) are the major healthcare providers in rural Nepal. This paper examines how THs who were trained in western medicine, are handling the first aid (FA) kit in rural Nepal. In 2000, we interviewed 44 THs about how they were using the FA kit, which they received after the initial training in 1996. This study revealed that the FA kit has become an important new tool for the trained THs. We found that approximately 90% of the respondents were using the FA kit even 4 years after the initial training. In addition, nearly 50% of the respondents expect to have more medicine in a bigger FA kit in future. While most THs were enthusiastic about this new tool, many did not like a new practice of collecting cash from their patients. How to keep the FA kit with a sustainable cost-recovery mechanism thus remains a big challenge to these THs. However, as the majority of the trained THs could manage to use their FA kit for four years with minimum support from outside, the FA kit has a potential to become a common tool of more THs in rural Nepal.

Workshop 9

W9-1) INTERNATIONAL COOPERATION IN HEALTH AND TROPICAL MEDICINE IN KENYA AND TANZANIA : SYNOPSIS OF WORKSHOP

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Three medical and health projects in Kenya and Tanzania were reviewed in the workshop. Dr. Matsubayashi reviewed autonomous management of the central research laboratory in his MCH project supported by JICA in Tanzania, emphasizing that not only sustainability of transferred technology but also sustainability of activity management including finance is the key to sustain the laboratory activity after the project. Establishing a good monitoring system was essential to autonomy of the activity. Mr. Kunieda reviewed his NGO's activities in eastern Kenya on health, education, and environmental issues. The CanDo's activities are based on the rural development approach. The project faced difficulties including political turmoil among the community in the beginning. However the project gradually got footholds in the community through dialogues with the community. Dr. Sugiura reviewed another JICA project with Kenya Medical Research Institute (KEMRI). By contrasting the Japanese style international medical and health cooperation against the Western style one, he pointed out the importance of research, research-based cooperation, and the long-term commitment and understanding between the recipient and donor countries. Although the workshop did not deal with all the international health cooperation problems in Kenya and Tanzania, it could clarify the importance of research-based long-term capacity building cooperation in this field, as health of people in Kenya and Tanzania cannot be improved immediately.

W9-2) SELF-SUSTAINABILITY IN PROJECT-TYPE MEDICAL COOPERATION – AN EXAMPLE OF A CLINICAL LABORATORY IN TANZANIA MATERNAL AND CHILD HEALTH SERVICE PROJECT

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PREFACE : A medical facility must be self-sustaining in technique, finances and administrative management if it is to continue after an international cooperation project has ended. Self-sustainability in technique, targeted in Project-Type Technical Cooperation, has been achieved. However, self-sustainability in finance and management has not been achieved satisfactorily, resulting in constraints. We have worked towards these goals in the Pediatric Clinical Laboratory of the Maternal and Child Health Project (1994-2001) in Tanzania, and have achieved them.

METHOD : Cost-sharing was introduced in which test-samples from private clinics and patients over age five have been charged. The administration of the laboratory was reinforced, skilled and empowered. Administrative staff received on the job training (OJT). After the project ended, a supporting committee formed in Japan has been giving necessary suggestions to the administration.

RESULTS :

1) Two-thirds of operating costs (US\$30,000) have been collected by cost-sharing.

2) The administration manages the maintenance of facilities, equipment, reagents, financial control and personnel management.

3) Monthly reports, including sample numbers, types of samples, income, expenses and balances are sent to JICA headquarters and the supporting committee in Japan.

DISCUSSION : The conditions for the self - sustainability are as follows :

1) activities should not be beyond the capability of the donor or recipient,

2) resources should be collected by the organization, and should be revolved without error,

3) monitoring systems should be formed. Although fulfilling 100% of necessary operating costs is difficult, the proportion that the recipient is expected to bring in should be clarified in the project.

W9-3) COMMUNITY DYNAMICS AND INTERNATIONAL HEALTH COOPERATION : A GRASSROOTS PERSPECTIVE

NOBUHIRO KUNIEDA

Community Action Development Organisation, Tokyo, Japan.

Community Action Development Organisation (CanDo) is a Japan-based development NGO working in the areas of education, health, and the environment in Kenya since 1998. Its main target area, Mwingi District is located in a semi-arid area with frequent droughts and disadvantaged in terms of economic and social development while nearly half of the children under 5 years are chronically malnourished and high rates of morbidity and mortality are observed.

Under its community-based health program, CanDo organizes a training course on primary health care for women at reproductive ages, aiming at helping them to gain basic knowledge and skills and to form connections among themselves and intrinsic motivation for promoting sustainable health care activities. Also, a training course for pre-primary school teachers has started to help them integrate child health care aspects into the education centered school activities. CanDo also assists dispensaries in improving the facilities and the management.

Based on the experience through the above - mentioned program, I would like to share my personal views on international health cooperation as follows*:

(1) Constructing community-based health care facilities does not automatically ensure that local people will utilize the facilities for improved health care services. Some local people may try to take advantage of the opportunity for reinforcing their political base and then split the community, which leads to serious dysfunction of community-based health care activities. It is crucial to examine the community's management capacity and its history when designing a project like facility construction;

(2) The "Training of Trainers" approach does not necessarily bring about ripple effects as theoretically predicted. This is particularly the case where the majority of local people are neither equipped with basic health knowledge and skills, intrinsically motivated, nor well connected for promoting health care activities for their own community. In such a case, training of community health workers and/or traditional birth attendants may only encourage them to utilize their knowledge and skills for their personal benefits ; and

(3) Foreign experts have certain roles to play, but only with local experts. Rural communities in developing countries are incomparable with those of developed countries in various aspects including the languages and cultures, infrastructure, utilities, the environment, etc. The roles of local experts, who are likely to be most familiar with such local conditions and available at much lower expenses, could not be overemphasized.

* These views do not necessarily reflect the views of Community Action Development Organisation.

W9-4) ROLE OF INTERNATIONAL HEALTH COOPERATION IN KENYA MEDICAL RESEARCH INSTITUTE

YASUO SUGIURA

International Medical Center of Japan, Tokyo, Japan

How should Japan facilitate international health cooperation in Kenya? Kenya Medical Research Institute (KEMRI) is one of the largest research institutes in Africa. Since the institute was established, JICA and Japanese University have been cooperating with KEMRI in several fields such as studies of parasites, diarrhea, hepatitis, pneumonia, traditional medicine, and HIV/AIDS. Beyond the Japanese research cooperation, the Centers for Disease Control and Prevention in the U.S. and the Welcome Trust in the U.K. have been studying HIV/AIDS and malaria at the KEMRI. Results of their research are published in standard medical journals and sometimes affect health policies. In comparison with these activities, Japanese research at KEMRI is practically invisible. I believe that the Japanese research gets less attention for the following reasons : 1) At the country level, Japan's support for Kenya is based on a concept of international health cooperation. This means the Japanese research at KEMRI is not for Japanese people but for Kenyan people.2) At the research projects level, the Japanese projects stress KEMRI's initiative and sustainability.3) At the personal level, the Japanese experts are always mindful of KEMRI staff as equal counterparts working to improve research activity and develop mutual understanding. The Japanese cooperation at KEMRI focused on the international parasite control and blood screening for HIV and Hepatitis B are going well. These are unique and important activities for Kenya. What kind of further research cooperation can be performed with KEMRI? It is time to consider by

both Japanese and Kenyan sides.

General presentation

P1-1) DEVELOPMENT OF THE CONCEPT OF A POLICY ADVISER DISPATCHED TO HEALTH AUTHORITIES OF A DEVELOPING COUNTRY

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When I began my post as a health policy adviser in Indonesia in 1997, I devised the idea that the tendency of policy advisers to initiate projects with the donor's interests in mind interfares with the self-reliance of the developing countries in which they work. Thus, I developed the concept of a policy adviser providing "a basic idea of primary care", which a group of primary care doctors in National hospitals proposed, because I think that a policy adviser should be like a primary care doctor for a nation. In this light, I propose that : -A policy adviser should not be overly focused on a specific field.-Beyond the obvious manifestations of a problem, a policy adviser should consider underlying factors involving the mind / psychology / society / cultural anthropology. -A policy adviser should consider a profile of independence. -A policy adviser sholld have a variety of skills, and should use an evidence - based approach toward support activities. -If the policy adviser cannot solve a problem, he or she should get support from other experts, the staff of the JICA representative office and / or the Japanese embassy in a timely manner. -A policy adviser should continuously support the country and see the projects through to completion. This concept helped me in supporting the development of a self-reliant health care system in Indonesia. I think that we need to define the skills of policy advisers and make available a training system as well as a support system during the dispatched period.

P1-2) PROCESS AND POWER FOR INTERNATIONAL HEALTH POLICY IN JAPAN : HOW SHOULD WE RESPOND TO THE REALITY OF THE FIELD, RATHER THAN GLOBAL POLITICS, SUCH AS THE UN?

CHUSHI KUROIWA

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Japanese ODA started as war reparations in 1951. Without dispatching the SDF for peace keeping, economic cooperation developed as the main ODA during theCold War. In 2003, however, Japan ranked last among 21 wealthy countries inhelping the poor, according to a survey. In examining the AD syringe case, Iaimed to 1) explore power and process regarding funding in international health policy and 2) demonstrate the undervaluing of opinions of expertsworking in developing countries. In examining this case, I reviewed reports from the JICA project expert andJICA. In addition, I interviewed people involved in the process of policymaking, such as staff members from JICA and MOF.JICA raised concerns about infection risk due to the disposal of ADsyringes, based on expert reports at the WHO/WPRO joint meeting. However, the pressures from WHO/WPRO, UNI-CEF/New York, and MOF were so strong thatJICA HQ urged the Laos office to send the request for AD syringe procurementafter the expert term finished. The original report of "basic research forpolicy making for syringes provision" was changed in numerous places byJICA grant aid division. Once the provision of AD syringes was decided in 1999 by WHO-UNICEF-UNFPA, Japan could not decline the request for the procurement. All Japan, likeexperts, consultants, corporations, NGOs and the government, should beinvolved in the initial stage of the development of global health policy. Otherwise, this Japanese asset would disappear without respect from the world under the current fashionable integrated funding system.

P1-3) INTRODUCTION OF THE OVERSEAS TRAINING AND THE EFFECT IN THE INTERNATIONAL HEALTH EDUCATION A REPORT ON STUDY TOUR IN THAILAND AND CAMBODIA

KAZUE YAJIMA

Gunma paz Gakuen College, Gunmaken, Japan

I have conducted a study tour to Thailand and Cambodia in the past three years. The aim of the tour is learn the health system in developing countries, to learn the role of nursing in international cooperation, to learn through multi-cultural communication and to understand the frame of international cooperation. The tour took participants to the project sites including medical facilities, community and school settings in there two countries. The participants also stayed in the rural Thai community. The participants reported that the was fruitful not only for understanding the value of international cooperation and different culture but also for their reflection of their own life. Many of them also reported that they got familiar with HIV/AIDs problems through the tour. I am sure that the study tour gives an important opportunity of learning particularly for those who wish to work in the field of international cooperation.

P1-4) DEVELOPMENT OF DATABASE ON JAPANESE FACULTIES ACTIVE IN INTERNATIONAL ECUCATIONAL COOPERATION IN MEDICINE

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[Purpose] The International Research Center for Medical Education, at the University of Tokyo was established by the Ministry of Education, Culture, Sports, Science and Technology in 2000 to promote and reinforce international cooperation in medical education. It promotes international cooperation in various fields of medical education, including medicine, dentistry, nursing, public health and nutrition. We conducted a questionnaire investigation of academic instructors and researchers to develop a database of their experiences and plans regarding international medical education. Through this, we hoped to learn more about the human resources available to promote international cooperation in this field.

[Methods] We sent a questionnaire regarding the interest and availability of educators who could contribute to international cooperation to departments of dentistry, nursing, public health and nutrition in public/private universities, totally 540 departments, in December 2001. We received 2,220 replies from 174 departments by March 20th, 2003.

[Results] The response rates were as follows: medical departments 44 out of 80 (55%)replied, with 1,067 people responding; dentistry departments 29 of 29 (100%), 762 people; pharmaceutical departments 30 of 87 (32.2%) 161 people; nursing department 42 of 70 (34.5%)159 people; nutrition and other departments 48 of 274 (17.5%), 85 people. It became clear that despite interest in these projects, there were many obstacles to middle or long-term overseas commitments.

[Conclusion] Medical education is an important field, and Japanese involvement in it shows our commitment to international cooperation. However it is necessary to develop an organizational entity or a network of individual entities to facilitate these interactions.

P1-5) STUDY ON HEALTH PROMOTION - COMPARISON BETWEEN WHO POLICY AND MUNICIPAL POLICY IN JAPAN -

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In 2002, WHO published a document named of Reducing Risks, Promoting Healthy Life, after the introduction of PHC and Health Promotion Policy in the last decades. Japan has achieved high level of health attainment such as longest life expectancy (81years old) and lowest infant mortality rat (4/1000 live birth) in the world. However, it is not clear how the WHO health policy is taken up in Japan. The study was undertaken to examine how WHO health promotion policy was translated into municipal level policy selecting one city in Hokkaido as a study site. In WHO health promotion policy, it should be noticed that WHO mentioned importance of prerequisite for health promotion, since it implies that health promotion policy should be conducted in relation to health needs and level of infrastructure as well as available social and economic resources. The municipal studies had met adequately with the prerequisites. The municipality achieved good health attainment such as lower total mortality and infant mortality rate as well as longer life expectancy compared with those of indicators in Japan. Interesting finding was that the health policy of the municipality was stated only in general terms but not specific terms as WHO health promotion policy, in spite of adequately meeting with prerequisites and high health attainment. Further study is progressing to examine in what way WHO health polices are taken up by national as well as municipal level in Japan.

P1-6) DEVELOPMENT OF GEOGRAPHICAL INFORMATION SYSTEMN IN THE POPOLATION-BASED COHORT, CHIANG RAI, THAILAND

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Purpose : Chiang Rai Province is located in the northern part of Thailand, with 16 districts and 123 subdistricts. In this province, all tuberculosis cases, HIV/AIDS cases and death certificates are registered through a computerized information system. We estimated the geographic prevalence tuberculosis cases at the sub - district level. Methods : By utilizing the tuberculosis database, we calculated the average number and prevalence rate of cases recorded in 1998-2000 and 2000-2002. All the data were put onto the map of Chiang Rai district using GIS software. Results : 1) Regional distribution of patients : we found a higher number of patients around the sub-districts with the most human traffic. 2) Prevalence and distribution of health facilities : we found divergence between the areas with a high prevalence of disease and the locations of health facilities, such as district hospitals or district offices. Discussion : The recognition of the divergence between the need for health care and the facilities that provide it help to identify target areas for tuberculosis treatment and prevention. Conclusion : By using geographic information systems, we can find more effective ways for tuberculosis control intervention.

P1-7) INTEGRATION OF COMPLIMENTARY MEDICINE INTO NEW HEALTH POLICY IN THAILAND - PRELIMINARY FIELD RESEARCH IN KHON KAEN DISTRICT : PART 1-

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The Thai government established a health program for villages in the 1960's, which has yielded satisfactory outcomes. However, other health issues remain, such as rapidly rising health costs, and the great disparity in health services between Bangkok and more remote areas. Consequently, the Thai government decided to reform the health policy not only to reduce the cost, but also to promote traditional philosophy. The northeastern region has received much less health service than the rest of the nation. However, people in this area traditionally use complimentary and alternative medicines, and their daily diets include many kinds of herbs. Thus, people in this area have retained cultural and spiritual treatments. The objective of the study was to analyze the design and structure of alternative care in the service setting. We selected Khon Kaen district as our research area. Participatory observation and in-depth interviews were conducted with people in this area. We found that the village administration has developed a network of villagers, health centers and hospitals. Key persons, who were selected from among the villagers, took the initiative of conducting an exercise program, which integrates traditional Thai exercise into modern community medicine. Women in the village run a center of Thai traditional massage in collaboration with a Non-government organization, which provides them with better job opportunities and a chance to work while looking after their children. Future research will be expected to assess the practitioner's perspective on conditions, equality development, and to assess the client's opinion toward the level of care.

P1-8) ESTABLISHMENT OF HEALTH INFORMATION NETWORK BY WIRELESS RADIO IN OUDOMXAY, LAO PDR

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Background : Telecommunications in Laos is underdeveloped. According to the 2000 statistics, there are only about 50,000 telephones in the country. Sharing information precisely and promptly is a major problem in Lao PDR. Purpose : Establishment of a health information network by wireless radio between a provincial health office (PHO) and six district health offices (DHO) in Oudmxay province. Methods : Establishing the network involved : (1) organizing a network committee in the PHO, (2) introducing a logbook to record the radio communications, (3) creating a format for daily reports, (4) refresher training for PHO and DHO staff, and (5) visiting DHOs to check that the radios have been set up. Results : Daily communication by the radio between PHO and the six DHOs began in February 2003. The report included the number of inpatients and outpatients in the district hospitals and epidemiological data for infectious diseases. Through daily communication, the PHO could recognize the condition of the radios and responded quickly in case of problems. Discussion : Introducing the network system was a major advance, allowing the PHO to receive daily reports from the DHOs. Although over one hundred radios are in use in this country, there is no regulation of their usage, except for a priority time for each province. Therefore, the experience with the network in Oudomxay could be repeated in the other provinces. Conclusions : We established a health information network by wireless radio between Oudomxay PHO and

P1-9) A STUDY OF QUALITY ASSURANCE FOR LOCAL HEALTH FACILITIES AND HOW JAPAN CAN SUPPORT HEALTH IN THE PHILIPPINES

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Background :

Sentrong Sigla (SS), a program to assure the quality of local health facilities, was launched by the Department of Health in the Philippines in 1999. The SS focuses primarily on Rural Health Units (RHU). As of 2002, 1,125 (47%) of RHU, 251 (2%) of Barangay Health Station (BHS), and 84 (13%) of provincial hospitals were SS certified.

When SS was begun in 1999, the Japanese government assisted in the construction of 83 health facilities in Central Luzon (Region III). Despite these efforts, only 9 RHU (50 %), 16 BHS (27%), and 1 hospital (20%) were SS certified in 2003. Therefore, we investigated why these facilities failed to get SS certification.

Method :

All 83 health facilities supported by the Japanese government were investigated with a questionnaire con-

ducted in March 2003.

Results :

We found that all of the buildings were being properly used, and the equipment needed to fulfill the SS was available. However, equipment use was variable. For example, we found that among the BHS staff, only 47.5% used hemometers and 45 % used forceps frequently, while 35.6 % did not use hemometers and 20 % did not use forceps at all. It is notable that most of the facilities where the equipment was not used were not SS certified.

Conclusion :

Providing equipment and buildings is not enough to assure quality health care. Developing skilled human resources in each facility may require further assistance to the Philippines. This highlights the importance of matching assistance with each country's health policy.

P1-10) MEDICAL INFORMATION ON OBSTETRICS IN DEVELOPING COUNTRIES

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Medical records are necessary for diagnosis and treatment of individual patients, and for appropriate provision of health care by facilitating sharing of the information among health personnel. They can also be utilised as a tool for evidence-based decision-making and for establishment, monitoring and evaluation of health policy. However, it is frequently observed in developing countries that medical records are not properly recorded or stored. The objective of this study was to observe how medical records are kept and managed in developing countries in order to develop strategies to assist in improvement of this process. We investigated storage conditions and contents of obstetric medical records in Afghanistan, Bolivia, Cambodia, and Madagascar from December 2000 to August 2003. An identification number is utilised at 10 out of 11 facilities investigated. However, the numbers are often duplicated, due to lack of an appropriate file management system. All facilities have storage of medical records, however the conditions were different ; e.g. central storage, stored at each ward, or by each doctor. This may be due to lack of legal regulations for the medical records. Observations during labour are not always recorded, and only a few facilities record postpartum observations. Appropriate recording and storage of medical information is essential for improvement of quality of health care and function of a health facility. The medical record is often the only source of health information in some developing countries. Therefore, we suggest that assistance in the area of medical record management is essential for developing countries.

P1-11) MODE OF SOIL CONTAMINATION WITH ASCARIS AND TRICHURIS EGGS IN A VILLAGE IN THE SUBURB OF HANOI, VIETNAM

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Many studies on soil contamination with eggs of soil - transmitted helminth (STH) infection have been done to obtain better understanding of the infection. But there are few studies on how the soil is contaminated with parasite eggs. To establish a more effective strategy to control STH infections we have launched a project in a village located in the suburb of Hanoi, Vietnam. We found prevalence of Ascaris lumbricoide, Trichuris trichuira and hookworm infection was 37.7 %, 65.2 % and 8.9%, respectively. As soil contaminated with Ascaris and Trichuris eggs is a potential source of infection, we had examined 186 samples of soil in various sites of the study area in both rainy and dry seasons. The examination showed that more than 83 % of soil in rice fields and vegetable fields and around households in the village was contaminated with Ascaris sp and Trichuris sp eggs including those from animals, which were kept by the villagers. We herein analyzed the cause for the soil contamination in these sites by questionnaire survey. The survey revealed as follows ; in most of rice and vegetable fields, the contamination occurred by disposal of human or animal (mainly pig) feces as fertilizer but eggs were

also detected in some fields where any feces was not dispersed at all. Fecal disposal to the field did not significantly relate to the level of egg contamination. In the 33 households surveyed, 20 had one-pit latrine, 9 had water sealed latrine with septic tank and 4 had no latrine. Number of eggs detected from soil around houses did not depend on the type of latrine. Among households with one pit latrine, very interestingly, a significant higher number of eggs were found in households they did not use night soil than in those they used. The difference was found in rainy season. Our another survey showed that about 25 % of secondary school children defecate in the place other than latrine and 6-33 % of 180 dust samples collected from various sites was contaminated with the eggs. From these results, the soil contamination may occur by not only direct fecal disposal to the environment but also by another factors such as wind, rain (flood) or transportation by human and animals (pigs, chicken and dogs).In conclusion, the results of this study emphasize the importance of collective efforts of personal and household hygiene such as defecation habits and fecal disposal, and proper sewage system.

P1-12) TRIAL OF NOSOCOMIAL INFECTION CONTROL IN BACH MAI HOSPITAL PROJECT IN VIETNAM AND APPLICATION TO SARS CONTROL

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The frequency of nosocomial infection in developing countries is considered high, and in fact proper measures are insufficient in most hospitals there. In the Bach Mai Hospital Project for Functional Enhancement, which has been implemented at Bach Mai Hospital (BMH) in Hanoi, Vietnam, one of the programs incorporates technical guidance on nosocomial infection control. When Severe Acute Respiratory Syndrome (SARS) spread in Vietnam, BMH contributed greatly to putting SARS under control, implementing rapid and appropriate measures. This presentation deals with SARS control at the hospital level and reviews technical cooperation in the project.

Establishment of an appropriate system for nosocomial infection control, development of manuals and teaching materials, staff training and evaluation, and a fact-finding study on 1,679 in-patients were the main components of technical cooperation. Setting up the basis of nosocomial infection control along with upgrading basic knowledge and skills were undertaken through these activities. The nosocomial infection control system at BMH, including surveillance and reporting, has been functioning well. Results of tests conducted before and after training courses showed a 29.3 score elevation with strong satisfaction among trainees. In the fact-finding survey, 100 cases were considered to be nosocomial infection cases and in 49 cases the causative agents were identified. BMH provided medical care for 35 SARS patients but reported no nosocomial infection cases by April 28, 2003, the date on which Vietnam declared SARS to be under control there.

A swift reaction, effective nosocomial infection control, proper advice by the Ministry of Health and other elements are considered important factors in the success of SARS containment at BMH. In addition, the fruits of technical cooperation conducted under the project are regarded, to no small extent, as having formed the basis for this.

Nosocomial infection control is a crucial factor in achieving high-quality medical care as well as SARS control at the hospital level.

P1-13) SELF-ASSESSMENT OF THE EFFECTS OF EPQI AT PUSKESMAS IN NORTH JAKARTA

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Introduction

The Jakarta Health Office has introduced GKM (Quality Control circle/team), for the first time, to health center services in North Jakarta. A questionnaire survey was conducted after eighteen months had passed, with the aim of providing baseline data for monitoring perceptions that would change with time as the project progressed.

Method

Forty-eight GKM leaders in North Jakarta responded to the questionnaire, as well as 13 facilitators in Sarawak state, Malaysia, where similar activities had been run for more than a decade.

Results

1. When asked to identify "recognized changes" answers included; "SOPs were standardized" (94%) and "attention to patient satisfaction was enhanced" (90%). "Improvement of problem-solving capacity" was raised by 44% in Jakarta, and by 100% in Sarawak.

2. In Sarawak, when asked to name "incentives for GKM" respondents referred to factors related to achieve-

ment, such as "improvement of capacity" (69%), although teams in Jakarta referred to external factors like "encouragement of supervisors" (58%).

3. With regard to "difficulties encountered", both Jakarta and Sarawak named "unawareness of the importance of the activities" (98%, 77% respectively) and "busy" (98%, 70%). The teams in Jakarta chose "lack of visible changes "and "boredom", but none referred to these problems in Sarawak.

Summary

The survey suggested that the sense of success could serve, by itself, as a good incentive. The North Jakarta project should develop a critical mass of qualified instructors and facilitators to lead teams to success in terms of tangible achievement in quality improvement.

P1-14) EAR HEALTH CARE PROJECT IN INDONESIA(REPORT : 14) SUPPORT FOR THE DEAF IN INDOSIA AND NEIGHBORING COUNTRIES

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Hearing loss is invisible and underestimated. Hearing International (HI), non-governmental organization was established in 1992. It aims to facilitate worldwide prevention and treatment of deafness, which largely results from otitis media. The organization works in partnership with the International Federation of Otolaryngological Society and NGOs working in the field of deafness. A Japanese chapter of HI (HIJ) was founded at the same time, and has been operating in Southeast Asia with cooperation and financial support from the Ministries of Post and Telecommunication, the Ministry of Foreign Affairs, JICA, Rotary Clubs and NGOs. The "Project for Ear Health Care in Indonesia" began in 1995 at the University Hospital of Indonesia in Jakarta. Seven pairs of Japanese otologists have been dispatched to the project sites for three months at a time. Also, two ear care specialists stayed in Jakarta for a year and contributed to the training of local ear health care personnel. In 1999, the tempral bone dissection room for the ear surgery training was completed. Since this time, the training in the treatment of otitis media has been regularly provided at this facility. HI has been extending its project sites from Jakarta to another major cities in Indonesia since 1998. Moreover, it is expected to expand the programs to neighboring countries, using a newly built ear care training center near Jakarta Airport, with support from the JICA. The development of good-quality low-cost hearing aids and publication of books to raise public awareness of hearing problem are proceeding as well.

P1-15) HEALTH STATUS AND SUBJECTIVE ECONOMIC SATISFACTION IN WEST PAPUA

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We have performed a medical assessment of indigenous people living in Senggo, a rural village in West Papua/Irian Jaya, Indonesia, with a population of 2000 people. Economic conditions are generally poor in this village. We used questionnaires to assess subjective quality of life, performed physical examinations, and measured total cholesterol and haemoglobin concentrations. Subjective quality of life was rated on a 100 mm visual analogue scale (worst on the left, best on the right). A total of 227 indigenous people (115 men, 112 women, mean age 36.4 years [SD 13.5]) participated in the study, and 153 agreed to undergo blood tests. The mean concentration of total cholesterol was 147.3 mg/dL [42.9] and of haemoglobin was10.6 g/dL [1.8]. The lower concentrations of total cholesterol and haemoglobin could be explained by the widespread and chronic infections of malaria, tuberculosis, and parasites, in addition to general poor nutrition. There was a significant linear association between total cholesterol and haemoglobin concentrations, and, more surprisingly, both correlated significantly with subjective economical satisfaction. These correlations were stronger among men than women. Total cholesterol and haemoglobin, therefore, can be used as indicators of objective nutritional and chronic infectious state in such regions. Although economic conditions are recognised as being associated with health and nutritional state, asymptomatic medical disturbances such as low levels of cholesterol or haemoglobin were closely associated with subjective economical satisfaction in West Papua.

Refference

Wada T, Matsubayashi K, Okumiya K et al. Health status and subjective economic satisfaction in West Papua. Lancet 2002 ; 360 : 951.

P1-16) DIVERSITY OF HEMOGLOBIN AND TOTAL CHOLESTEROL LEVELS IN DEVELOPED AND DEVELOPING REGIONS

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Objectives

Developing regions are plagued with malnutrition, anemia, and chronic infections such as malaria and tuberculosis, in addition to overall poor economic conditions. However, there are few comparative studies of nutrition and hemoglobin (Hb) levels amoong healthy subjects in developed and developing regions. In this study, we performed screening blood tests of total cholesterol (T-cho) and Hb in West Papua, Indonesia, Bago Mountainous Region, Myanmar, and Japan.

Methods

The subjects of this study, all of whom consented to blood tests, were residents of Senggo, West Papua / Indonesia (population about 2000; N=225, M : F=109-116, mean age 39.5 ± 11.6 y. o) and of Sain yay village, Bago Mountainous Region, Myanmar (population about 250; N=184, M : F=86 : 97, mean age 38.1 ± 15.7 y. o.). Studies were conducted in 2/2002 and 11/2002, respec-

tively. They were compared with subjects who work in a Japanese emergency hospital (N=271, M : F=71 : 200, mean age 36.5 ± 10.9 y.o).

Result

Mean (S.D) T-cho levels were 186.8 (36.5) mg/dl in Japan, 147.3(42.9) mg/dl in West Papua and 112.6 (34.2) mg/dl in Myanmar. Mean (S.D) Hb levels were 13.3 (1.7) g/dl in Japan, 10.6 (1.8) g/dl in West Papua and 9.1 (1.9) g/dl in Myanmar.

Discussion

In developed regions, a high T-cho level is a risk factor for arteriosclerosis and cardiovascular disease. However in poor regions, where infectious disease is the primary concern, mean T-cho level is very low, presumably as a result of malnutrition. Further discussion is needed to determine whether international standards for T-cho or Hb levels are appropriate for developing regions.

P1-17) THE HEALTH CARE CONSULTATION BEHAVIOR OF VILLAGERS IN MYANMAR.

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Objectives : The purpose of this study is to clarify the role of rural health centers or rural health sub-centers (HC) and to identify obstacles faced by villagers who consult health institutes in Myanmar. Methods : We conducted interviews with 77 villagers using a questionnaire in Nyanug U and Meiktila townships, Myanmar in 2002. Results : 60 villagers (80%) answered that they go to HC when they feel sick, when they get pregnant, when they need immunization, or to help health workers. 55 villagers (82.1%) go to HC on foot, and most said it takes less than 30 minutes. However, 6 villagers (9%) said it takes more than 2 hours from their house to the HC. 39 villagers (60%) feel that the consultation fee of HC is appropriate, and all the villagers were satisfied with the health workers. The reasons they do not go to HC are distance, lack of necessity or use of another health institute. When they feel sick, 33 villagers (44.6%) go to HC, 17 villagers (23%) take medicine on their own. When they decide to go to HC because they feel sick, distance and consultation fees are not major concerns. However, when they need to go hospitals in town, the consultation fee is too expensive for them. Conclusion : Though villagers go to health institutions when necessary, economic and transportation considerations often prevent villagers from seeking medical treatment, especially in a hospital. A variety of funding systems, such as an emergency fund, and standing medical equipment for emergencies are needed.

P1-18) AN INVESTIGATION OF HEALTH SEEKING BEHAVIOR ON PEOPLE LIVING IN A VILLAGE IN LAO PDR

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[Purpose] To investigate health-seeking behavior of people living in a village in Lao PDR

[Methods] Cross-sectional interviews were carried out in the village, (population 446 in 74 households). Interviews were completed in 67 out of 74 households.

[Results] We found a pattern of sequential health seeking behavior in the village. At first, most people chose traditional medicines when they became sick. Secondary, many people selected modern medicines from a village health volunteer or a private pharmacy. Thirdly, about half of the people who had not recovered from their illness went to the hospital. In this study we divided 67 households into two groups, those that were using some preventive behaviors against illness, and those who were not. Interestingly, the two groups showed different health seeking behaviors. The group that used preventive behaviors (group A) followed the sequential pattern but the other group (group B) did not. Further, we found perceptions about the cause of diseases were different in the two groups. The people in group A tended to believe that the cause was related to non-hygienic behaviors. On the other hand, the people in group B tended to believe that the illness was inevitable.

[Conclusions] The sequential pattern, 1) traditional medicines, 2) modern medicines, and 3) hospital, was recognized as a health seeking behavior in the village.

The two groups that did/did not use preventive behaviors showed different patterns of health seeking behavior and different perceptions of the cause of diseases.

P1-19) A CASE OF A GIRL WITH DERMATITIS IMPROVED THROUGH SKIN-CARE AT A VILLAGE IN LAO PDR

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Case : 2 years old, Laotian

Main Complaint: Yellowish-green scabs on head, dermatitis on limbs.

History: I visited the home of a girl with serious dermatitis at a village in Oudomxay Province, northern Laos, in early November 2002. I noticed an unpleasant odor from her wounds and body. Her skin condition developed ten days after birth, and has persisted.

Observation: Her consciousness was normal, facial skin was red and swollen, scalp had discharge and scabs. Height was 65cm, weight was 6800g, limbs were thin, and her lower abdomen was swollen. Limb mobility was normal, but she was unable to walk or speak any words.

Process and Assessments: I thought she had infectious dermatitis. I instructed her mother to wash her skin using soap, then to spread oil on affected areas, and to not put on her hat. Five days later, most scalp scabs had cleared, but the odor persisted. On the 20th day, facial swelling persisted, and there were reddish dermatitis-like circles on her back and limbs. I instructed her mother to apply antibiotic ointment to her face after washing, and steroid ointment to the reddened areas. On the 22nd day, she began taking antibiotics (AMPC) for 1 week. On the 26th day her head and face skin were improved. She continued synthetic vitamin liquid for 4 months. I advised about diet, encouraging her family to give the girl food whenever she wanted it.

Conclusion : I realized clearly that skin-care (by soap and oil) is important for dermatitis in developing countries.

P1-20) ISOLATION OF FOOD-BORNE PATHOGENS FROM THE MARKET OF THE LAO P.D.R.

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Introduction

The aim of this study was to clarify the actual situation on the bacterial contamination at some food markets in Vientiane, capital of Lao PDR.

Methods

(1) Study site : Vientiane city, capital of Lao PDR.

(2) Study period : The study was performed during 199-2002

(3) Equipment for collecting samples : "Fuki-Fuki test kit" (EIKEN Kagaku, Tokyo) and "Seed-swab No.1"

(EIKEN Kagaku, Tokyo) were used. The surfaces of food samples were wiped with this equipment.

(4) Food samples : Objective foods were various kinds of domestic cattle and fowl meets (beef, water buffalo, pork, chicken, duck etc), domestic fowl eggs, fishery products (fresh water fish, marine fish and shells), frogs, snails, some vegetables (bean sprouts etc.) and cooking material water vessel, cutting board etc, sold at the markets.

(5) Culture and identification of the bacteria : Col-

lected sample was applied to both DHL and TCBS media by direct plating and by growth culture using Rapaport broth, alkaline peptone broth and saline-polymixine broth. The identification was made by *invic* test and commercially available kits.

Results and discussion

1. Contamination by halophilic vibrios in almost of all the meats samples was demonstrated. *Vibrio parahaemolyticus* and *Vibrio fluvialis* were mainly confirmed in every year.

2. *Salmonella* species were detected very low frequency in both marine and fresh water fishes in 2000. However, there were no detected cases on *Salmonella* spices in 1999, 2001 and 2002.

3. Halophilic *Aeromonas* strains were recovered from fowl meats, fresh water fishes, cut board in 2001, and from pork and crabs in 2002.

P1-21) LIFE STYLE AND BRONCHIAL ASTHMA AMONG CHILDREN IN RURAL BANGLADESH : ANTI-ASCARIS IGE IS AN INCREASING RISK FACTOR FOR ASTHMA

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Background : Bronchial asthma is a major cause of morbidity in rural Bangladesh, which is counter to worldwide trends. To identify risk factors for asthma that could explain this unexpected situation, community-based cross-sectional and case-control studies were done. Methods : The sample population was 1705 5-year -old children in rural Bangladesh. Among the 1587 respondents, all asthma cases and randomly selected controls were asked to participate in the study. The cases were defined based on responses to a questionnaire from the International Study of Asthma and Allergies in Childhood (ISAAC). Tests for serum total and specific IgE levels to dust-mite, cockroach, Ascaris and Alternaria, stool tests for helminth infection and tests for indoor housedust mite concentration, were done on 219 cases and 183 controls. Information on history of pneumonia before age 5 years

and other risk factors was also obtained. Findings : The prevalence of wheezing during the last 12 months was 16.2%. The risk for wheeze independently increased in relation with anti-ascaris IgE levels (odds ratio per quartile 1.28 [1.06-1.56], p=0.011), anti-DP IgE levels (odds ratio per quartile 1.24 [1.03-1.50], p=0.021), and total IgE (odds ratio per quartile 1.28 [1.06-1.55], p=0.010). Other independent risk factors were history of pneumonia before age 5 years (odds ratio of history of pneumonia at the age of 2 was 7.27 [2.74-14.18], p=0.000), family history of asthma (odds ratio 2.80 [1.15-6.82], p=0.023). Interpretation : High prevalence of Ascaris infection, with a high titer of anti-ascaris IgE might contribute to the increase of asthma symptoms among children in rural Bangladesh.

P1-22) RISK FACTORS INFLUENCING FOR ACQUIRING JAPNESE ENCEPHALITIS IN WESTERN TERAI, NEPAL - A case contro study-

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Using a case-control study design, we attempted to measure risk factors influencing Japanese Encephalitis (JE) in Nepal. The study population consisted of 56 cases and 112 controls with a matched case and control ratio, 1 : 2. JE confirmed cases of aged 1 through 15 years came from JE hyper-endemic districts (Kailai, Bardiya, Banke and Dang). They had been admitted in Bheri Zone Hospital in August - September, 2000 and were recruited as the cases. The control of each case was selected randomly matching the same age, sex and place of residents of the respective cases. The study revealed that when the parents' education level was lower, the JE risk got higher. Children of illiterate parents had 4.09 and 2.53 times higher risk of JE. Fathers' occupation having domestic animals in household was associated with a high JE risk (OR=2.78). History of previous JE cases in family was highly associated with JE risk (OR=11.96). Sleeping under a bednet or a blanket and wearing long sleeved clothes was found highly protective for acquiring JE (OR=0.44, 0.33 and 0.06, respectively). Working / playing in the rice field, staying outside during twilight were highly associated with JE risk (OR=5.15 and 17.47 respectively). Fishing activities in summer monsoon and using open field for passing stool were associated with JE risk (OR=4.16 and 4.92). Poor quality of houses including mud / cow dung plaster walls, floors, brick soling floor, poor cleanliness and dampness were associates increasing the risk of JE (OR=0.14 and 0.10), respectively.

P1-23) NEED FOR HEALTH PROMOTION BASED ON EVIDENCE IN CROSS SECTIONAL SCHOOL HEALTH DATA

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With the purpose of evaluating the health condition of children in public primary schools in the province of Cordoba in Argentina, cross sectional data were collected in the School Health Program of the Ministry of Health of the Cordoba Province in 2001. To conduct an analysis, a total of 25,701 pupils of the first and the seventh grades from one hundred and fifty primary schools were studied. The data were consisted of variables in three groups : i.e., nutrition, vaccination and disease groups. Prevalence rates were calculated for the variables and descriptive statistics showed the distribution of the health conditions. Pearson correlation coefficients were also calculated. Factor analysis was carried out and six factors were extracted out of original 27 variables. Regression analyses were executed to reveal the risk factors of the diseases. According to the statistical analysis it was clarified that there was reciprocal influence between lower nutritional profile and scoliosis. The absence of vaccination of BCG worked as a risk factor both to lower weight and to scoliosis.

We concluded that it is necessary to modify the primary preventive conducts in the school health program by focusing activities in two preventive areas : firstly, health promotion giving comprehensive health education and life-skills training, reinforcing protective factors while reducing risk behaviors and secondly, community participation, involving all school and community members in making decision and carrying out interventions to promote learning healthy lifestyles and community health promotion projects.

P1-24) IMPLEMENTATION AND EVALUATION OF SCHOOL DENTAL HEALTH IN THE FIELD OF INTERNATIONAL COOPERATION

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The Association of Dental Cooperation in Nepal began its school oral health activities in 1992. Specific activities have included the introduction of oral health education into annual curriculum, fluoride application and training for teachers. In this study, we evaluated oral health activities in 17 schools of two villages near Katmandu.

Of the 17 schools, 13 replied to our inquiries regarding oral health activities in the school. The teeth brushing instruction were practiced in 8 schools, and 6 schools carried out the other dental health education more than twice a year. Dental checkup by teachers performed in 7 schools. The fluoride mouth rinsing program has been continued in 14 schools. One school discontinued the program because the teacher in charge left the school. In schools where we started the fluoride application program in 1994, the DMFT index among students aged from 11 to 13 years was 0.8, but it had decreased to 0.6 by 2002. The DMFT of all other schools in the same villages was 1.3 in this year. Longer the program has been conducted, better the dental caries prevalence rate appeared.

Since we started training oral health workers in 1994, at least one worker has been trained in each school today. They continue to play an important role in oral health activities at their school.

Judging from the above, oral health promotion in school is very effective measure. To ensure long-term success of this program, it will be necessary to establish a system of training by Nepalese teachers.

P1-25) THE STATUS QUO AND PROBLEM OF SCHOOL HEALTH IN SRI LANKA-THOROUGH BASELINE SURVEY FOR AMDA PEACE BUILDING PROJECT

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Background :

AMDA Peace Building Project has started since last February as a two-year project in Sri Lanka. As a part of this project, school health programme has been conducted. The baseline survey was done in Hambantoa, Deep South district in last March.

Objectives of survey :

- To find out how to organize the program

- To understand the school health system, overall school situation, and major health problems

Methodology :

- Four divisions of Hambantota district are selected for target areas.

- Conducted school sanitation surveys and interviews with related organizations

- Gathering information from governments and UNICEF

Findings :

- More than 60% of schools do not have basic school facilities, such as toilets, urinals, water supply, or electricity facilities. Dental caries, malnutrition, and chronic poverty are problems. Insufficient first aid supply and poor maintenance of toilet and urinal facilities were observed in most of schools.

- Teachers have little recognition what major health problems are among their students. Health Club, which is recognized as a key component of school health, does not exist in most of schools.

- Principal and Public Health Inspectors (hereinafter referred to as PHIs) is the direct responsible for school health. It seems, however, neglected issue and has little understanding. PHIs actually visit a school only a few times a year.

Recommendation for the programme :

Enhancement of knowledge and awareness toward student's health among PHIs and principal are needed. PHI and principal should take a leadership in conducting basic health education.

P1-26) SCHOOL HEALTH PROJECT – TROUGH AMDA HEALTH NEWSPAPERS

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Background :

As a part of AMDA Peace Building Project, School Health Programme has been started in Hambantota district, the south part of Sri Lanka since March 2003.

Overall Goal :

- To improve students' health condition for their future healthy life

Specific Goals :

- To provide students with health information and habits through seminars using AMDA Health Newspaper (hereinafter referred to as AHN)

- To strengthen Public Health Inspectors (hereinafter referred to as PHIs) and teachers' responsibility for their students' health

Methodology:

- To publish AHN

- To conduct related seminar including practice for the students and their family members using AHN at each target school

- To hold a regular meeting with PHIs at four divisions

- To monitor the students' health knowledge by conducting questionnaire and short quiz

Expected outcome

- PHIs and principals have more responsibility for school health.

- Health awareness among PHIs, students, their family members, and teachers are promoted.

- Students' general health condition is improved. Students' health habits are acquired.

- Students learn how to use sanitation facilities properly.

P1-27) COMMUNICABLE DISEASE CONTROL IN DEVELOPING COUNTRIES, FOCUSING ON CHILDREN UNDER 5 YEARS OLD

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Primary Health Care (PHC) has greatly contributed

to improving the level of children's medical care in de-

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veloping countries. In an effort to improve areas where PHC has not prevailed, the Integrated Management of Childhood Illness (IMCI) programme has been introduced in some countries, and good outcomes have been reported. However, many children still die of infectious diseases, such as acute respiratory infection, infectious diarrhea and measles. To get an overview of infectious diseases among children, we asked medical personnel who participated in JICA training courses on preventive medicine to provide information through a questionnaire and follow-up interview. We asked about measures against infectious diarrhoea, vaccine- preventable diseases, acute respiratory infection, malaria, tuberculosis and HIV, as well as the effects of IMCI. By analyzing the data, we considered what kinds of control programmes are suitable for effective and long-term operation. Questionnaires were returned by 15 JICA participants in 11 countries. The results indicated that the programmes carried out until now, including Expanded Programme on Immunization (EPI), the use of oral rehydration therapy (ORT) and Global Polio Eradication Programme, are successful. However, many types of infectious diseases among children are not addressed by these programmes. Therefore, new programmes, such as IMCI, are welcomed. The improved availability of safe water, and maintenance of public health infrastructure are indispensable for infectious disease control. Moreover, it is essential to improve community participation, education and motivation.

P1-28) THE MOTHER AND CHILD HEALTH ACTIVITISE AT DAPAKEL VILLAGE IN NEPAL

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Purpose A basic survey conducted between August and December 2001 in Dhapakhel Village, Nepal. Province in Nepal found that about 10% of 56 children under the age of one were suffering from malnutrition, with a score on the Kaup index of 13 or less. As part of this study, we instituted a bodyweight measurement-training program aimed at improving the nutritional expertise of health volunteers.Results1. Discussions regarding health issues We explained the results of the survey to local leaders (5) and health care volunteers (7) in August 2002. Local leaders informed us of the need for mother-child health care activities, but did not have any specific suggestions. We proposed a new program for the health care volunteers.2. Training/Implementation For two days in December 2002, we conducted bodyweight measurement training for health care volunteers. After instruction, the health care volunteers measured the bodyweights of 84 infants in various regions. 3. Impressions of the health care volunteers after the program Health care volunteers from a neighboring village also wanted to learn how to perform measurements.

P1-29) CURRENT SITUATION OF ANTENATAL CARE IN PHNOM PENH CITY, KINGDOM OF CAMBODIA

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Prolonged civil war and genocide left it with shortage of health facilities and personnel in Cambodia. The estimated maternal mortality ratio in 1995 was 437 per 100,000 live births. Antenatal care (ANC) is a key component of the Safe Motherhood Programme (SMP). The objective of this study was to investigate the current situation of ANC in the capital, Phnom Penh City to establish a strategy to support SMP in Cambodia. Women who gave birth between October 9, and November 7, 2003 were interviewed at all the public and private health facilities in Phnom Penh City. 1204 responses were received from Phnom Penh residents, representing 84% of the estimated number of deliveries in the city. Of these, 89% were from public health facilities. The proportion of low birth weight (LBW) babies was significantly higher in the group who received ANC two times or less (10% vs. 6%, p=0.02). There was no difference in the prevalence of complications (haemorrhage, hypertensive disorders, convulsion, and fever) either group. Living in a rural area, multipara, having dropped out of primary school, house-hold income less than US\$ 200 per month, and delivery at public health facility were frequently observed in the group that had attending ANC 2 times or less. This suggests that these socio-economic factors are related to the number of ANC visits and the proportion of LBW. Social backgrounds of women should be considered in establishing implementation strategies for SMP. In addition, further investigation for the ANC role on SMP should be carried out.

P1-30) MIDWIFERY EDUCATION IN CAMBODIA

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To review the historical background of midwifery education in Cambodia, we interviewed the people involved in midwifery education, Cambodian Ministry of Health, and National Center for Maternal and Child Health, along with the literature review. In Cambodia, midwifery education started in 1949. In the post Pol Pot regime, the midwifery education was reformed in 1979 to produce midwives quickly with two different courses : one for training a primary midwife (8 year basic education plus 1 year technical education) from 1979 to 1988, and another for training asecondary midwife (11 year basic education plus 3 year technical education) from 1980 to 1993. In November 2002, midwifery education is aone - year training for registered nurses with three-year nursing education. JICA and American NGO started to provide post-graduate training in 1997. There are three different 4-month post-graduate midwifery-training programs for secondary nurses. In 2003, the Ministry of Health stared to plan their own post-graduate midwifery training. In1975 there were 1380 midwives but it reduced to be 431 by the end of the Pol Pot regime. Today there are 1500 primary midwives and 1800 secondary midwives nationwide but less in rural areas. Quality of the midwives aswell as midwifery education is relatively low. Besides, there are noboard exams for midwives that raise further concerns.

P1-31) SITUATION ANALYSIS AND ITS POTENTIAL CAPACITY BUILDING ON HEALTH PROVIDERS FOR REPRODUCTIVE HEALTH, NARSINGDI DISTRICT, BANGLADESH

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Japan International Cooperation Agency Bangladesh Office

The situation Analysis was conducted form July to

Sep, 2003 at 4 upazilas in Narsingdi District. The general

objective of the situation analysis study was to describe the gap between ideal and actual situation of health services and monitoring, supervisory system of service providers for Reproductive Health from community up to district Level. Selected results are, Supervision and Monitoring, the promise of supervision was not achieved. Mostly supervisors tend to focus on administrative issues, such as inspection facilities, supply logistics, use of resources, review of records and directions from higher to lower levels. Training, the most of the staff interviewed has had their basic training 10 years before. Similarly most of them had their refresher training 2 years before. However, in most of the study sites, staffs have been trained for different type of short time training. Care Seeking Behavior, Place for delivery, almost all the deliveries were conducted at home, 52 out of 60. This tendency of delivery at home finds support from a recent study in Bangladesh reported that 94 percent of the deliveries in rural Bangladesh were conducted at home, husbands house. As a conclusion, improving quality of care through supervision is also another aspect but current supervision produces little evidence towards that. Continuous training or follow up is necessary for keeping up providers motivation. Delivery with skilled birth attendant should be promoted.

P1-32) IN SEARCH OF BETTER REPRODUCTIVE HEALTH PROVISION IN BANGLADESH, HRDRH INTENDS TO FIND OUT THE NECESSARY ASSISTANCE WITHIN THE DISTRICT TO GET UTMOST IMPACT OF THE MIDWIFERY TRAINING AND POSSIBLE SOLUTION. ITS PROCESS AND THE POSSIBLE SUGGESTIONS WILL BE PRESENTED.

YURIKO EGAMI, ICHIKO YMMASHITA, RIKA MASUYAMA, KAY SUZUKI, IZUMI MURAKAMI

Japan International Cooperation Agency Bangladesh Office

Due to the efforts of the Government and the Development Partners, TFR has decreased from 7.7 to 2.8 and MMR from 3000 to 300 in the last 30 years in Bangladesh. However, much work remains in a country with 1.3 million unwanted pregnancies annually, 87% of deliveries without skilled attendance, and only 5% of mothers with complications during pregnancies and deliveries having access to medical care. The Human Resources Development in Reproductive Health (HRDRH) Project aims to improve the quality of training for providers of RH, so that they are well-oriented and skilled after needs-based training. The clinical and training skills (especially clinical training) at the Maternal and Child Training Institute (MCHTI) have been improved, and field health providers are trained in midwifery, EmOC and ICMI. During the rest of the project, we would like to develop effective support to enable our trainees to maximize the training results at their sites, and we wish to develop a mechanism so that lessons learned at the site are reflected to stake holders and concerned authorities for better training and heath provision. The approach from both service providers and beneficiaries is necessary to promote safe motherhood and to decrease maternal mortality. A policy that meets with the needs of clients and the community will be developed and implemented throughout the country, and the accountability of service providers should be encouraged. At the same time, it is necessary to encourage the society to allow the women with greater autonomy seeking better RH.

P1-33) THE 6-MONTHS MIDWIFERY TRAINING FOR FWVS ARE BEING CONDUCTED IN BANGLADESH IN ORDER TO DEVELOP THEIR MIDWIFERY AND HEALTH EDUCATION SKILLS. THE EVALUATION AND THE IMPACT OF THE TRAINING WILL BE PRESENTED. THE SELECTION OF TRAINEES IS ESSENTIAL.

RIKA MASUYAMA, YURIKO EGAMI, ICHIKO YAMASHITA, KAY SUZUKI, IZUMI MURAKAMI

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In Bangladesh, MMR is still high as 360 (per 100,000 live births). Deliveries with skilled attendance remains 13 percent and most of deliveries are conducted by TBAs. Since 2001, the Government conducted training for Family Welfare Visitors (FWVs) on Midwifery skills. Human Resources Development in Reproductive Health (HRDRH) Project by JICA has improved training skills at Maternal and Child Health Training Institute (MCHTI) where we reviewed the progress of the technical skills through pre- and post-test and follow-up visits three months after completion of the training. Overall, all trainees who received the training improved the scores of

tests after the training. We conducted the follow-up visits to 10 trainees three months after the training, and have found out the increase of their services, 7 trainees provided more ANC services (167 percent from before the training) and 5 trainees more delivery services at home (178 percent), the main reasons of which were regarded as the increase of self-confidence and the increase of reliance by the community clients after the training, especially when trainees had ever conducted deliveries at their community. The selection criteria of the trainees should be examined for better impact of the training.

P1-34) PHILIPPINES INTEGRATED PROJECT ON REPRODUCTIVE HEALTH/NUTRITION

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From 1999 to 2002, JOICFP executed the Integrated Project on RH/Nutrition with Batangas Province and its 5 selected municipalities, financially supported by Ajinomoto. The project aimed to help improve the quality of people's life in Batangas through RH and improved nutrition. The short-term objectives were : (1) to have increased the proportion of women and individuals using RH services by 20%; and (2) to have reduced by 30% the percentage of malnourished children, relative to baseline data, by the end of the project period. The main strategies undertaken were developing the capacity for self-reliance, integration of RH and nutrition, and human resource development.

An external evaluation was conducted in March 2002 to assess the project performance through document

review, interviews with people concerned, and field observations.

Project Performance

1. CPR has risen in the project sites and more women, men, children, adolescents and elderly individuals are using health services, particularly RH services.

2. Nutrition levels have been improved, with 32.8% out of 131 malnourished children in the supplementary feeding program having been restored to a normal nutritional state after only two years.

Lessons Learned

1. Properly oriented LGU officials can be strong partners in ensuring the institutionalization of certain activities and maintenance of the initial project gains.

2. Integrating nutrition into RH could bring about a

broader perspective toward health in the life cycle, and involve more sectors.

3. Capacity building of project personnel is the key

to sustaining project gains and activities, even after project completion.

P1-35) ECTOPIC PREGNANCY IN YAOUNDE, CAMEROON : PROBLEMS IN THE MATERNAL HEALTH CARE SYSTEM

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The present study was conducted in order to assess the incidence of ectopic pregnancy (EP) in Yaounde, the capital of Cameroon. In 2002, all women admitted for an EP to hospitals in Yaounde were systematically enrolled. Socio-demographic information on the women and their reproductive histories were collected during a face - to face interview. Medical and obstetrical data (clinical findings at hospital entry, medical history, type of surgery and final vital status) were collected from gynecological and surgical files and admission registers. Finally 320 cases were analyzed, giving an estimated EP incidence rate of 0.8%. Laparotomy was performed in 98% of cases and hemoperitoneum observed in 93%. Three maternal deaths were recorded, corresponding to a mortality rate of 1%. A high percentage of women (83.9%) had first consulted in a local health facility before the hospital admission and most of them had undergone an ultrasound scanning in each facility. This repetitive consultations and medical check-ups led to a substantial delay to the treatments in the hospital. The estimated EP incidence rate in Yaounde was lower than that currently observed in industrialized countries. In developing countries, more effective use of ultrasonography in local medical facilities should be encouraged in order to shorten the delay between the first system and treatments reducing morbidity and mortality due to EP.

P1-36) POPULATION BASED STUDY ON CHILDBIRTH WITH COLLABORATION FROM TRADITIONAL BIRTH ATTENDANTS IN SAINT LOUIS city, SENEGAL

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There is always a risk of recruitment bias when we conduct hospital-based epidemiological research on childbirth, especially in developing countries. The objective of this study was to conduct a population-based study on childbirth in collaboration with 76 traditional birth attendants called "matron" in Saint Louis City, Senegal. We recorded all childbirths that occurred in February 2003 in the Saint Louis area. Medical, obstetrical and sociodemographical information was obtained through medical records and a structured questionnaire. During this period, 106

we recorded 529 child births : 247 in the hospital, 74 in the clinic, and 208 at home with birth attendants. Twenty - eight home births were referred to the hospital, mainly due to excessive hemorrhaging or multiple pregnancies. The factors influencing the place where women chose to give birth were number of living children and the domicile address of the women. Women tended to choose the hospital for their first child, and those who lived nearby showed the same tendency. Eighty-three percent of the birth attendants palpated the fetal presentation, and 88.1% of them practiced an endovaginal examination by using a glove or a fingerstall. A large proportion of the population of Saint Louis gives birth at home. Since the birth attendants have certain medical knowledge and practices, as do hospital midwives, it is logical to make use of the birth attendants, enhancing their skills, rather than promoting hospital child birth in Saint Louis.

P1-37) FEMALE GENITAL MUTILATION (FGM) IN BURKINA FASO – CASE REPORT FROM DELIVERY WARD -

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Background : The Female Genital Mutilation (FGM) has been commonly conducted in Kompienga, Burkina Faso. The form of the FGM is the excision of the clitoris with prepuce of clitoris. We investigated the influence of the FGM scars on the childbirths among the women in Kompienga.

Methods : We have observed four women delivered babies at Kompienga Health Center, Kompienga city, Kompienga State, southeast of Burkina Faso since January 2003. We observed their FGM scares, progresses of labors, vulvae after childbirths and lacerations on the FGM scars.

Results : All cases had the FGM scars. All cases were multipara and transvaginal deliveries. Three of the

four cases had the lacerations on the FGM scares on their labors. Two of four cases had the perineal lacerations without episiotomy on the labors.

Discussion : The FGM scares were more likely to cause the lacerations on childbirths. The lacerations on the FGM scars were formed because of the limited elasticity of the FGM scares. The lacerations on the FGM gave severe pain for the women after childbirths. The pain appears not only during urination but also after urination as they wash vulvae with water. The women suffer from the pain three to five days after the childbirths. The FGM causes the pain and risk of infection for the women after childbirths. As a result, the FGM badly affects the safety and comfort of the women after childbirths.

P1-38) FACTORS CONTRIBUTING TO MATERNAL DEATHS IN LUSAKA DISTRICT, ZAMBIA BY VERBAL AUTOPSY APPROACH

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This paper described factors associated with maternal deaths in Lusaka. Data are drawn from the University Teaching Hospital (UTH) 2001 records and from the household heads of the deceased women (verbal autopsy; VA). There were 122 maternal deaths recorded during the study period and 50 (41%) cases were visited, 18 (36%) deaths were due to direct obstetric causes and 27 (54%) died from indirect causes. The common causes were malaria 18%, tuberculosis 14%, hemorrhage 10%, and abortion 10%. Maternal death causes as perceived by the relatives were multi-factorial. These included traditional beliefs, hospital negligence, inadequate medical supplies

and staff, delayed referral, the existence of HIV / AIDS and poor health workers attitudes to work. The findings

also suggest that traditional beliefs are still associated with maternal death.

P1-39) COST OF ANTI RETROVIRUS THERAPY FOR HIV/AIDS PATIENTS IN KHON KAEN, THAILAND

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Purpose : Thailand originally had two health insurances schemes ; Civil Servant Medical Benefit Scheme (CSMBS) and Social Security (SS) for the company workers. In 2001, a new health insurance scheme was introduced for overall insurance coverage, with the examination fee at public hospitals fixed at 30Bahts (30BS) (1). This study examined the health care costs for anti - retroviral drugs (ARV), which is anticipated to be the government's burden. Methods : Study site : Khon Kaen, northeast Thailand. Duration : April 1st to September 30th, 2002. Participants : citizens who visited an infectious disease hospital in Khon Kaen for HIV/AIDS or related symptoms. Research items : age, gender, living area, type of medical insurance, type and fee of clinical test, CD4 and the amount of virus, diagnosis, amount and cost of prescriptions, other fees, and co-payment. Results and

Discussion : Among 476 persons (260 men, 216 women), non-ARV recipients paid 623B in total and 528B (84.8%) for prescriptions. ARV recipients paid 7,562B in total, 7,382B (97.6%) for prescriptions of which 7,290B (96.4%) was spent on ARV-related-prescriptions. CSMBS had the highest average total fee per visit among non -ARV recipients (CSMBS; 1,163B, SS; 486B, 30BS; 665B, co-payment; 495B), and in ARV recipients (CSMBS; 10,269B, SS; 7,600B, 30BS; 8,244B, copayment; 4,012B). The average costs of ARV prescription per visit were similar (CSMBS; 9,783B, SS; 7,518B, 30BS; 8,094B, co-payment; 3,478B). Numerous 30BS patients were non-ARV recipients (CSMBS ; 10.5%, SS ; 50.0%, 30BS; 75.0%, co-payment; 77.8%). The results showed that medical costs differ among types of insurance. (1) 1 Baht? 2.6 Yen.

P1-40) HERBAL MEDICINE USE AND QUALITY OF LIFE AMONG PEOPLE LIVING WITH HIV/AIDS IN NORTHEASTERN THAILAND

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Background : People living with HIV/AIDS (PHA) use herbal medicine as alternative therapies in Thailand, particularly when curative options are limited.

Methods : Participants were 132 HIV-positive Thai adults who attended the PHA's self-help group meetings beginning in June 2002. Health-related quality of life

(QOL) scores were measured by a self-administered questionnaire from the Medical Outcomes Study-HIV Health Survey. Dimensions of physical function (PF) and mental health (MH) in QOL were assessed in this study. Additional data were collected on herbal medicine use, socio-demographic, psychosocial and HIV-related characteristics. QOL scores were compared between herbal medicine users and non-users, and were statistically evaluated by Mann-Whitney U-test, and then stratified by all the characteristics.

Results : The herbal medicine users had significantly better MH scores than the non-users (p=0.03). When stratified, herbal medicine users with the following characteristics had significantly better MH scores than the non-users : female (p=0.04), widowed (p=0.03), having no income (p=0.03), reporting any HIV-related symptom (p=0.02), having no instrumental support (p1<0.01) or receiving subsidies (p<0.01).

Conclusion : Herbal medicine use was associated with better MH especially among socially vulnerable PHA. This study suggests the importance of herbal medicine use in the strategy of care for PHA to improve the MH aspect of their QOL.

P1-41) TRENDS IN HIV/AIDS CASE NOTIFICATION IN CHIANGRAI, THAILAND

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The purpose of this study was to examine the correlation between HIV/AIDS prevalence and sex-ratio using the annual AIDS/Symptomatic HIV Case data in Thailand. The sex-ratio used in this study is the ratio of male prevalence to female prevalence. We plotted the annual data at the national and provincial levels, respectively. In addition, we investigated sex - ratio trends among various age groups in a single province, Chiangrai. At the national level, we found a negative relationship between prevalence and sex-ratio, meaning that as the prevalence of HIV/AIDS increased, the sex - ratio decreased, as relatively more women became infected. In 1993, the prevalence was 4.4 per 100,000 and the sex-ratio was 7.3. By 2001, the prevalence had reached 41.0 and the sex-ratio was 2.0 in 2001. The data at the provincial level showed the same pattern. The prevalence was 6.0 and the sex - ratio was 9.5 in 1991, and the prevalence was 186.3 and the sex - ratio was 1.4 in 1999. In examining HIV / AIDS infection among various age groups, we found that the sex - ratio generally declined between 1994 and 1999. However among ages 0-4 and 5-9 years, the sex - ratios remained stable at about 1.0. The results of this ecological study suggest a negative relationship between HIV/AIDS prevalence and sex - ratio at several levels. We will follow these data continuously and investigate the meaning of this inverse relationship.

P1-42) SIMULATIONS ON THE PREVALENCE OF HIV/AIDS AMONG FEMALE PROSTITUTES IN THAILAND USING A MATHEMATICAL MODEL

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There were rapid increases in the number of HIV epidemics and AIDS cases in Thailand in 1990s. In par-

ticular, the risk of HIV infection was very high among female prostitutes who played a large part in the trans-

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mission of HIV infection. The prevalence of HIV was higher in northern Thailand than any other province, due to sprightly sexual activities, acts of prostitution. In response to the rapid increase of the HIV/AIDS prevalence, the 100% condom program was undertaken by the Ministry of Public Health in Thailand to promote condom use in commercial sex. By the way of prevention against HIV/AIDS transmission, the condom usage rate rose from 10-20% in early 1989 to 80-90% in late 1990 in northern Thailand, which produced a decrease in HIV prevalence. We formulated a mathematical model for the prevalence of HIV in female prostitutes infected via sexual intercourse with male guests with due regard to young females entering and withdrawing from prostitution. The simulations were carried out during 10 years, from 1989 to 1999 for HIV prevalence of female prostitutes in Chiang Rai, northern Thailand, and Bangkok, the capital of Thailand, based on the AIDS surveillance and the sexually behavioral reports, which showed that the HIV prevalence of the above two regions reached a peak, 49.7% and 17.8%, in 1991 and 1992 and that both the peak and the overall prevalence rate were higher in Chiang Rai than in Bangkok. We applied to our model for above two situations with a comparative view. In due consideration of female sex workers' over-reporting their condom usage rate, the simulations were carried out in the assumption that HIV blocking rate would be the reported condom usage rate multiplied by 0.7-1.0 (in the steps of 0.1). The simulations of our model for Chiang Rai and Bangkok showed that if the condom usage were not recommended, the transit of HIV prevalence in female prostitutes was much higher than the real one, while on the assumption that the condom usage in commercial sex was attained at the rate of 80-90%, the real transit ran within the range of the simulation output, and that HIV prevalence would decrease in the future. It may be given as the result of the simulations that the 100% condom program is helpful in preventing the female prostitutes from HIV infection.

P1-43) THE PROGRESS OF HIV/AIDS CARE SYSTEM IN NORTHERN THAILAND

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²JICA Model development for comprehensive HIV/AIDS prevention and care

The development of the HIV/AIDS care system in northern Thailand Yasuda Tadashi1)2), Miyamoto Hideki1)2), Mori Chiyoko2)1)International Medical Center of Japan, 2)Former expert of JICA AIDS project in Thailand Thailand is well known as a country which successfully developed a comprehensive care and support system in response to the rapid spread of HIV/AIDS. We analyzed this process to learn strategies that other countries could use to develop this kind of system quickly. A literature review of the process revealed that it was developed gradually, one component after another, such as basic medical, social, community and advanced medical components. Advanced medical care was one of the last components. Cross sectional analysis in 2001 showed a relatively insufficient medical care component compared to social support. Introduction of HAART in 2001, however, had a significant impact on the health system and improved the medical care service. The increasing role of PHA in the development of comprehensive care and preparation for the introduction of HAART is also noteworthy. Although it took Thailand fifteen years to develop its comprehensive HIV/AIDS care, late-coming countries should be able to shorten this process and catch up quickly. In the case of Thailand, HAART had a significant impact on this process and other countries could potentially utilize HAART as a first step in developing a comprehensive care system.
P1-44) COMPETENCY-BASED EDUCATION FOR PUBLIC HEALTH

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Objective

In order to obtain practical information applicable to competency-based education and training for workforce development, we examined utilization and limitations of core competencies for public health professionals.

Materials and Methods

We assessed the level of proficiency and the training needs in current assignments, among JICA participants of a seminar held at NIPH. A questionnaire, which was a modified form of the Core Competency Model developed by the Council on Linkages, was the primary tool that we used. We also employed the Likert technique.

Result and Discussions

The core competencies represent a set of skills, knowledge and attitudes necessary for the practice of public health. The core competencies were divided into eight domains. Results from the level of proficiency in each competency, which was assessed by five degree scales, showed scores of more than four points in the Domains of Communications, Community Dimensions of Practice and Basic Public Health Science Skills. As for the training needs, most of the competencies under the Domains of Policy Development/Program Planning and Financial Planning and Management Skills were over four points. This reflects the participants' qualifications, educational background, job position and job category in certain public health practice settings.

Conclusions

It was suggested that the core competencies help guide curriculum and content development of public health education and training programs for workforce development in public health practice.

P1-45) COMPETENCY BASED-EDUCATION ON MPH

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Background

Broadly defined, competencies are actions that are observable in the execution of one's work. The purpose of this article is to review public health programs already using competency-based curricula and the need to expand resources that are applied to the tasks of public health.

Objective

To examine utilization of self-assessment, evaluation tools and alumni surveys in Master of Public Health programs currently using competency-based curricula in the United States of America.

Method

Competency sets for eighteen schools of Public Health currently using competency-based curricula were reviewed using core competency sets in ten essential public health services, developed by the Council on Linkages. Data collection was through websites of the schools using the competency sets for MPH students.

Results

All use MPH curricula based on the core competencies, along with their functional competencies. The learning approach, competency sets and community-based teaching are universal but only eight (44%) of the curricula had self-assessment, evaluation tools and alumni surveys.

Conclusion

Basic training of health workers in the field of public health was comprehensive. However since less than 50% of curricula included self-assessment and evaluation tools for students, these elements should be introduced into more training programs. Periodic examination of competencies for field health workers through alumni surveys will ultimately bridge deficiencies between academia and the field in public health. We suggest that the use of competency-based curricula will improve and standardize the current training programs. A more com-

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other countries may be necessary.

P1-46) STUDY ON HUMAN RESOURCE TRAINING FOR HEALTH CARE PROVIDERS IN DEVELOPING COUNTRY

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To investigate training needs for conducting human resource development, we interviewed 11 experts involved in health-related projects in eight different developing countries including Indonesia, Honduras and Vietnam in the past five years and 5 Vietnamese counterparts of the on-going development projects in Vietnam. Of the 11 experts, all have ever conducted the training for their staff development mainly for nurses, medical assistants, midwives and health workers. They suggested it important to understand training needs, to share the problems regarding the health system at the lower level with the counterparts, to make a plan including monitoring and evaluation in advance. Besides, they also suggested it important to develop the skills of conceiving the project following the evaluation as well as of collecting and analyzing the data for the evaluation. For the training to be more effective and practical, the mangers themselves should be convinced to value it. Meanwhile, the counterparts suggested the lack of human resource for the training as a problem that then results in heavy workload. Foreign experts need to reconsider the content and extent of the training given to their counterparts.

P1-47) PRESENT SITUATION AND PERSPECTIVE OF THE IN-SERVICE TRAINING FOR NURSES AND MIDWIVES IN SENEGAL

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Introduction The PADRHS (Project for the Development of Human Resources in Health in Senegal), the first JICA project in French-speaking Africa, started in November 2001. Among the objectives of this project is the establishment of an adequate in-service training system for nurses and midwives. Objective The objective is to collect and analyze information about in-service training to help develop an education policy in Senegal. Method The first step was to collect material from the ministry of health, international organizations, and NGOs. The second step was the distribution of questionnaires to personnel in each medical region. The third step was to develop a needs assessment. Results Material collected during the first step revealed that there were many vertical health programs, but no coordination between them. The results of questionnaires distributed during the second step showed a policy of decentralization in the field of in-service training. In preparing the needs assessment during the third step, we found that the needs of nurses and midwives are to be in charge of the computer, the management of emergencies, the retraining and the IMCI. Discussion The role of the nurse in Senegal is to deliver medical treatment. However, it is important to coordinate their training, so that they can continue to learn while on the job. To do this we recommend the creation of a committee of coordination that links the ministry of health, international organizations, and NGOs. Conclusion The present situation of the in-service training for nurses and midwives in Senegal was clear.

P1-48) THE SITUATION OF COMMUNITY HEALTH WORKER IN THE REPUBLIC OF SENEGAL

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The Health Field Human Resource Development Project of Republic of Senegal (PADRHS) supported by JICA has started since November 2001 for the period of 5 years. In order to train the sufficient number of Community Health Workers (ASC), surveys were conducted by the Division of PHC, Directorate of Health, MSHP, to analyze the historical context, training method and geographical distribution of ASC. Community Health Workers (ASC : Agent de Sante Communautaire or matrone) have been trained since the late 1970's in accordance with the Alma Ata declaration of the Primary Health Care. They can roughly be divided into 3 categories, those who work as assistants to regular medical staff in health centers and posts, those who work as a head of a health hut, and those who are in charge of health promotion activities in the village. They are trained in health centers and health posts by the medical staffs for 3 to 6 months and are supported by different donors and NGOs if available. The number of ASC amounts up to 77% of total health staff in community health structure and estimated to be about 7,000.The unexpectedly high number of Community Health Workers has demonstrated the importance of ASC in the Senegalese health system and revealed the necessity of national standardization and support for their training and working conditions, as well as the provision of an adequate career path to satisfy the need for health personnel in Senegal.

P1-49) WOMEN'S HEALTH ISSUES IN CAMBODIA DURING POST-CONFLICT DEVELOPMENT PROCESS (1) – AN ANALYTICAL REVIEW OF OFFICIAL PUBLICATIONS

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In this study we analyzed the factors affecting women's health in Cambodia during the conflict and post - conflict periods, based on the available official publications.

During the Pol-Pot Regime between 1975 and 1978, most of the skilled personnel were lost. In 1980s, the number of health personnel and facilities grew, however, they often failed to meet minimum standards of quality. Thus, the under-five mortality rate has not declined since 1986.

After the peace process began in 1991, overseas assistance increased dramatically. It contributed to the rehabilitation and development of the health services. The new Cambodian government, with technical assistance of donor agencies, designed basic health policies to improve the quality of health services.

The maternal mortality ratio in Cambodia is still high compared to neighboring countries. One of the reasons for this is poor access to health services for women in rural areas. The ratio of deliveries assisted by skilled attendants in rural areas is about half that of cities and about one third that of Phnom Penh.

Information about family planning has spread rapidly since 1995. However, contraceptive use did not increase as well, suggesting that poor access to these services or the existence of socio-cultural obstacles.

These findings indicated that access to health services in rural areas is still limited, despite the overall improvement of health services in Cambodia during the past two decades. It is necessary to focus on strengthening health systems in rural areas, to improve women's health, and particularly to decrease maternal mortality.

P1-50) WOMEN'S HEALTH ISSUES IN CAMBODIA DURING POST-CONFLICT DEVELOPMENT PROCESS (2) -- A STUDY ON THE PERIOD OF POST-CONFLICT BASED ON INTERVIEWS OF CAMBODIAN WOMEN

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Women's Health Issues in Cambodia during the Post - conflict Development Period-A study on the post - conflict period based on interviews with Cambodian women -Etsuko Kita1, Hideo Uno2, Shiori Ui2, Tomoko Murata2, Atsuko Aoyamam21. The Japanese Red Cross Kyushu International College of Nursing2. Department of International Health, Nagoya University School of Medicine This presents a preliminary study on "post-conflict" process, based on an analysis of interviews with Cambodian women. All expressed physical or mental complaints within general malaise. Even among those who had bitter experiences during the past conflict, who currently lived in stability, seemed to be able to manage the influence of their experiences. However, women with handicapped husbands, or whose husbands or other family members lacked secure jobs, had greater difficulty coping with the

memory of the past war. These findings indicate that a stable life is extremely important to the rehabilitation process. Although no clear definition of "post-conflict" period is established, it is a dynamic and unstable phase of a political process between the active conflict and stable developmental stage. It is characterized by a) lack of security, b) no reliable authority or counterpart for cooperation, c) shortage of expert professionals or skilled workers, d) lack of, or malfunction of, social infrastructure, and e) moral decay of community. Although donors currently rush to provide assistance to "post-conflict" countries as Afghanistan or Kosovo, it is necessary to develop appropriate intervention methods for "post-conflict" periods when emergency humanitarian assistance or sustainable development assistance alone is not sufficient or appropriate.

P1-51) JAPAN MEDICAL TEAM FOR DISASTER RELIEF AND HEALTH SERVICE IN NORTHERN ALGERIA EARTHQUAKE

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Japanese Medical Team supported by JICA were dispatched May 25 to June 6 after the Earthquake in northern Algeria May 22 2003.Total 2266 people were killed, more than 10000 injured, 80000 suffered or homeless. Japanese Medical team was composed of 4 doctors with international health background, 7 emergency nurses, 114

2 paramedics, one pharmacist and 7 health logistics workers. Field clinic with 5 tents were opened in the afternoon on May 27. Also the first night shift of Japanese clinic were provided soon after the biggest aftershock occurred that day. Most of total 1628 patients were severely shocked and vanished, female dominant with various symptoms as abdominal, lumbar and back pain. Due to deteriorations of life style and lack of sufficient safe water, skin, eye, ear diseases were increased day by day. Health service and facilities of the local government were completely destroyed there. Instead of them, various primary health care and service were planned. One Japanese expert of water and toilet made a emergency lavatory plan and specifications using local materials written in Arabic. Public health leaflets in Arabic enhancing personal hygiene, nutrition, vaccination and early signs of infections were also distributed widely. Medical resources coming from remote area in Algeria were filled gradually but no health coordinators and workers there. In the period of no health service provided by local government in suffered country, Intervention of international health are considered to be effective for many evacuated people as well as emergency medical care services hereafter.

P1-52) AFGHANISTAN NATIONAL HEALTH RESOURCE ASSESSMENT AND ITS ROLE FOR HEALTH SECTOR RECONSTRUCTION PLANNING

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Health and Development Service (HANDS)

OBJECTIVE : To evaluate the feasibility and usefulness of a national assessment of existing health resources in Afghanistan at the transitional stage between emergency to reconstruction phases.

METHOD : All known health facilities in 32 provinces in Afghanistan were visited by data collectors. Three types of questionnaires were used to obtain basic information about : (a) health facilities (location, infrastructure, services, equipment, staff and qualifications, service volume) ; (b) community-based health providers ; (c) retail pharmacies. A database with assessment information was established and shared with the Afghan Ministry of Health (MOH), donor agencies, and local and international NGOs as a basis for rational planning of health sector reconstruction. A series of provincial level planning workshops were conducted in collaboration with MOH. **RESULTS :** The assessment confirmed, among other issues, that :

Great inequality exist in the distribution of more than 1,050 health facilities and services throughout the country;

One third of all districts failed to reach the national goal to establish one health facility per 30,000 people;

40% of basic health facilities do not have any female health workers; and

Most health facilities are sustained by multiple sources of support and NGOs are playing important role.

CONCLUSION : A national assessment of available health resources such as this can be a powerful tool, if it can be conducted at an early stage of post-conflict nation building. These findings established a common ground for identifying priority needs and areas, and thus encouraged rational allocation of resources provided by various players for health sector rehabilitation.

P1-53) TO INTRODUCE MATERNAL AND CHILD HEALTH HANDBOOK IN KANDAHAR, AFGHANISTAN

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The population In Kandahar is about 700,000, where most of people are pashtune tribe.

We have opened Karez clinic since Aug.2002 to cure of patients of poverty layer and who live in refugee's camp.

Because high rate of maternal death rate & Infant mortality, it seems most appropriate to introduce Maternal

and Child health Handbook to draw women's attention into care of them selves.

Obstacles to be resolved are total shortage of midwife & OB doctor & health worker, lack of birth registration system, and very low rate of literacy.

Pictorial Handbook would be helpful at this moment.

P1-54) A REPORT ON THE ACTIVITIES OF THE KAREZ'S MEDICAL AND EDUCATIONAL SERVICES IN AFGHANISTAN

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After the 23 years' of conflict and war in Afghanistan the infant mortality rate increased to 170/1000.

The Karez association was established in April 2002 to assist the remote area of Kandahar, Afghanistan through medical and educational services. The mobile medical team consisted of 10 people, working at the remote villages and refugee camps, served 19,360 people in the past year. The most common diseases were diarrhea and dysentery, (25%) and gastric diseases, (20%). Acute respiratory infections including TB were the next common category of diseases. Foe educational support, we opened five school classes and provided education for 209 students in the same area. In addition, we collected two thousand medical textbooks from Japan, and then shipped them to Afghanistan and distributed them to the medical students in the medical colleges of Kabul and Kandahar, in order to improve their medical knowledge.

A Maternal and Child Health Handbook is planned to be introduced in the field, to help protect the mothers and children from the nutritional and infectious disorders

P1-55) CONGENITAL MALFORMATION AND CEREBRAL PALSY AMONGST AFGHAN CHILDREN

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The refugee situation has affected all Afghans, especially mothers and children. Due to the use of all kinds of artilleries and increasing displaced people during the war, congenital malformations among children has been increasing in Afganistan.

Many Afghan pregnant women suffered from intol-

erable war stress and lack of high quality food. Thus they have been subjected to infection, and have experienced a high risk delivery with congenital anomalies.

In this study, we investigated a number of parents and children affected by congenital malformation at JIFF Medical Center for Afghan refugees in Peshawar and Kabul. This study is based on interviews with the parents of malformed children at JIFF Medical Center in Peshawar. Most children were living in the areas where bombing attacked.

As a result, we found about 7% of all live births were suffered from congenital malformation. Major abnormalities included CNS abnormality, spina bifida, encephalocele, microcephaly, congenital heart disease, alimentary system abnormality, cleft lip and palate, ectopic anus, skeletal deformity, limb defect, vertebral defect, achondroplasia, chromosomal abnormality, mongolism, urogenital abnormality, extrophy vesicle. Minor abnormalities included, limb abnormality, Talipes, Congenital dislocation of hip, and Polydactyly.

This study revealed the type of abnormalities among children with congenital malformations in Afganistan. Probably, such congenital malformation of Afghan infants will be ascribed to many factors occurred by the war. At present, however, it is so difficult for mothers to consult with well-arranged medical clinics in Afghanistan, although it is very important to prevent them from infectious diseases.

P1-56) PARASITIC INFECTION : A CONCERN FOR MIGRANT WORKERS FROM LATIN AMERICA

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In recent years, most South Americans who travel to Japan for work have been the descendents of Japanese migrants to Brazil, Peru and Bolivia. Thease Japanese have long suffered from a variety of parasitic diseases. Most of the current Japanese-American in the South America come from the Amazon area and Parana state in Brazil, and from Santa Cruz in Bolivia. Thease area have sevral endemic parasitic disease, including malaria, Chagas' diseaae, Leishmaniasis, filariasis and cysticercosis. Many of thease parasitic infections have a long latent period from the time of infection to the time when the patient becaomes symptomatic. This creates a difficult situation for medical workers in Japan. Diagnosis can be difficult. Chagas' disease, for example, is often asymptomatic, and is not easily detected. Some patients suffering from Chagas' disease develop fatal myocarditis or megacolon, which can be easily misdiagnosed in Japan, where this condition is rare. Beyond the medical workers, these patients themselves often do not know that are at risk for these diseases. We must be prepared to detect and treat these disease to maintain the public health in Japan.

P1-57) THE KNOWLEDGE, ATTITUDE AND PRACVTICE ABOUT SEVERE ACUTE RESPIRATORY SYNDROME (SARS) AMONG THE FOREIGN COUNTRY STUDENTS OF JAPANESE SCHOOL

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Foreign students of Japanese schools are one of the groups in high risk for severe acute espiratory syndrome (SARS). This study investigated knowledge, attitudes, and behaviors of foreign students in Japanese schools for SARS and tried to provide basic data for effective preventive measures. From June to July 2003, we conducted a self-administered questionnaire survey in all students of three Japanese schools in Tokyo. 96.3% of all students completed the questionnaire. Results : The mean age of

the participants was 22.7, with 51.4% males and 48.7% females. Among them, 76% came from China mainland, 12.7% HONG KONG and Taiwan, 10.6% Korea. The males had seeking information about SARS more frequently than the females. However, there was no difference between different areas. As to knowledge of SARS, the answer's correct rate of the females was slightly higher than that of the males. That of students from Korea, Hong Kong, and Taiwan was obviously higher than that

of China. Nearly 50% of the Chinese student could not answer the questions correctly. For the attitudes toward SARS, the percentage of the females who had answered "SARS is terrible "and" will not travel to epidemic area of SARS" was obviously higher than that of the males. As for the preventive methods, "Hand washing" was chosen by 80% of all subjects.

P1-58) CONSTRUCTION OF A COMPUTER-AIDED SYSTEM TO PROVIDE MEDICAL INFORMATION SERVICES TO FOREIGN RESIDENTS IN JAPAN

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[Introduction] The number of the foreign residents in Japan reached approximately 1.78 million in 2001. However, Japanese society has not changed, and specific concerns about the presence of foreign residents persists. Hence, current circumstances in Japan cannot be considered friendly for foreign residents. Therefore, some organizations have begun to provide counseling services to meet the needs of this population. For the aforementioned reasons, we developed two systems. One is to help the survey the problems faced by foreign residents when they seek medical services. The other is to provide necessary medical information to those who need it. [System Outline] 1. The first system is a computer-aided consulting service system. This system was developed as a Web based database, which has data searching, browsing, adding, and editing functions, using the Web browser. This system enables contact staff and supportive organizations to exchange information with each other. 2. The second is a multilingual medical information service system on the Web. To identify the location of the controlled information, this system has a message-sending function. Using this function, an administrator sends e-mail to registered readers whenever the Web page is updated. At present, the information is provided in the Web page of Kansai International Network for Solidarity (KINS). URL : http : //homepage3.nifty.com/kins/index.htm

P1-59) REPORT OF THE NUTRITION CONSULTATION IN MEDICAL MISSION FOR MIGRANTS

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SHARE and Minatomachi Medical Center have held medical missions for migrants around Tokyo with local government collaboration in these 8 years. We launched the nutrition consultation from 2001 onward. Here we report 17-nutrition consultation missions between 2001 and 2002.Nutrition consultation was requested by doctors who found any abnormalities such as obesity, urine sugar positive, and hypertension at medical consultation. Then we asked the clients about eating habits, and explained how to choose and cook foods for health, and handed illustrated pamphlets about diet. They are totally 100 persons, 58 men 42 women. Their nationalities are 48 Philippines, 9 Myanmar, 8 South Korea, and 35 persons of other 12 nations. Their main health problems are obesity, diabetes, and hypertension. Minor problems are anemia, emaciation, allergy, constipation, gastric ulcer, overdrinking, and unbalanced diet of child etc. We think nutrition education is very important to prevent sickness and to promote health. The main obstacles of nutrition consultation I feel are barrier of language and the difference of lifestyle and diet habits from Japanese. Hence food models and volunteer translators who are sent by nonnative communities are much helpful to explain details about diet control. We should learn more about their lifestyle and diet habits with nonnative community collaboration. We wish to extend the network of nonnative translators and volunteer dieticians.

P1-60) OBESITY AMONG FILIPINO IN JAPAN –SURVEY FROM FREE MEDICAL CHECK-UP BY NGO-

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We investigated obesity among 374 Filipino (199 men) aged 20 years or above living in Japan, using the records of free medical check-up offered by the SHARE, NGO, compiled from 1996 to 2002. According to the WHO criteria for obesity (BMI of 25 or above), 45.3% of men and 22.5% of women both aged from 30 to 39 were obese. The obese tended to be men, older and had a

higher level of blood pressure. There were no differences in the time in Japan and the proportion of health insurance holders between these two groups. Since the prevalence of obesity is high in Filipino population in Japan, their lifestyle and dietary pattern should be taken into account when the health program is provided.

P1-61) THE MAINTENANCE BEHAVIOR OF LONG LASTING INSECTICIDE-TREATED BED NETS (LLITBNS) PREVIOUSLY INTRODUCED INTO BOURAPAR DISTRICT, KHAMMOUANE PROVINCE LAO PDR

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Background As a part of JICA grant aid in 1999 - 2000, 4,676 LLITBNs (Olyset net Sumitomo Chemical. Co., Japan) were distributed in Bourapar district. Whilst the net coverage reached 80%, the provincial malaria infection rate remained high (13.3% in 2001), and further increases are possible.

Objective

1. To investigate the actual net conditions, and identify possible factors affecting maintenance behavior

2. To examine the impact of appropriate maintenance on malaria incidence during the last year. **Methods** This study was conducted in February -March 2003. In the selected 10 villages of the district, 240 households were visited, where the questionnaire interviews were undertaken, together with observation of net condition.

Results The proportions of households who followed the appropriate washing frequency (twice a year), and who followed the appropriate treatment for regenerating the insecticide were 33.8% and 73.3%, respectively. About half the households (53.3%) had holes and splits in the nets, of which only 32.8% of the households had repaired them. Factors affecting the maintenance behavior

were income, ethnicity, education level, literacy, and willingness to pay for Olyset Net. There were 180 malaria cases (11.2%) reported during the last year. Appropriate washing frequency had a statistically significant association with malaria incidence, showing a preventive effect.

Discussion It is essential to enhance the appropriate ITBN maintenance behavior, to sustain the effect of intervention. Our finding that malaria education had no impact on the maintenance behavior indicates that, future malaria education programs should be designed to be more widely accepted by community members.

P1-62) MALARIA ENDEMIC AND ITS CONTROL AT LOMBOK ISLAND, INDONESIA

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Since 2001, we have been conducting the malaria control project at the Meninting Health Center covering area in West Lombok. So far, the following epidemiological characteristics have been made clear.

1. Malaria endemics were found at coastal areas and hilly forested areas.

2. Malaria at coastal areas is transmitted mainly by *Anopheles sundaicus* which breeds in lagoons. Huge lagoons are formed in the dry season when exits of rivers to the sea are closed by sand carried by waves and flow of sea water.

3. Malaria at hilly forested areas is transmitted exclusively by *Anopheles balabacensis* which breed in small water pools along small streams or small springs.

4. Malaria endemicity has been lowered at coastal areas mainly by larvicide treatment on lagoons.

Based on these data, initially we have applied the modified active case detection to all the malaria endemic areas as a control measure to reduce the number of human sources. Case detection teams were organized in the health center, and visited malaria endemic subvillages regularly, found malaria patients by means of clinical signs and ICT diagnostic kits and treated them immediately. The result showed the case detection was effective in hilly forested areas at meso-endemic degree but not in coastal areas at hypo-endemic degree. Exceptionally at one subvillage in hilly forested areas, no effect was seen. This may be caused by differences in human behaviors, environmental conditions and drug resistance of parasites between this subvillage and others. Trials of indoor residual spray of insecticide (ISI) and of impregnated mosquito-nets (IMN) were made at some subvillages. The result revealed IMN was quite effective at a sampled subvillage in coastal areas and moderately at a sampled subvillage in hilly areas, while ISI had no effect at two sampled subvillages in hilly areas. On the basis of these results, we decided to continue the case detection activity in all the areas and apply IMN to all the areas. In hilly forested areas other measures including educational programs are under investigation.

P1-63) INTEGRATED COOPERATIVE RESEARCH FOR MALARIA CONTROL PROJECT – MALARIA EPIDEMIOLOGY IN EAST SEPIK PROVINCE, PAPUA NEW GUINEA

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The Integrated Cooperative Research for Malaria Control Project has been implemented by University of Papua New Guinea and Tokyo Women's Medical University, funded by JICA. The objective is to study the interaction of diversity among malaria parasites, vectors, and humans in different ecological zones and among different language groups in East Sepik Province. We conducted four malariometric surveys from September 2001 to August 2003 and examined 10,836 individuals in ten communities. The overall prevalence of malaria parasites was 24%. Positive cases included Plasmodium falciparum (68%), P. vivax (27%) and P. malariae (5%). No P. ovale was microscopically observed. Malaria prevalence was age-dependent, and was highest in the 1-9 year age group. Significant geographical variations were seen. In the wet season, malaria prevalence in foothill and plain areas (28 - 44%) was generally higher than in island and costal areas (13-22%). Seasonal variation was remarkable in a foothill community due to fluctuating P. falciparum prevalence. In island and costal areas, Anopheles farauti was predominant followed by A. Koliemseis. In the plains, A. farauti, A. Kokiensis and A. punctulatus were found. Human biting rate in the foothill communities was lower and predominant species could not be identified. The following eco-epidemiological malaria zones in East Sepik were differentiated: (1) island and costal zones with moderate endemicity, transmitted by A. farauti and A. koliensis, (2) foothill zones with seasonal epidemics, (3) plain zones with high endemicity, transmitted by A. farauti, A. punctulatus and A. koliensis. This diversity may have major implications on disease patterns and malaria control strategies.

P1-64) GLUCOSE-6-PHOPHATE DEHYDROGENESE VARIANTS IN FLORES ISLAND, REPUBIC OF INDONESIA

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We conducted a survey for malaria diagnosis and treatment in four primary schools in Flores Island, one of the Indonesian islands with an area of 17,000 Km² and a population of 1.8 million. Of these examined, 24.4% were diagnosed as having malaria (90/363), and administered medicine immediately. Glucose-6-phophate dehydrogenese (G6PD) test was performed at the same time, and sixteen persons (4.4%) were diagnosed as G6PD deficient. Eleven persons consented to analysis of the G6PD ge-

nome. We analyzed these subjects and found 1 case of G6PD Vanua Lava (T383C), 5 cases of G6PD Coimbra (C592T), 1 case of G6PD Viangchan (G871A), 1 case of G6PD Chatham (G1003A) and 3 cases of G6PD Kaiping (G1388A). These were unexpected findings because five different G6PD variants were found in such a small population. This suggests that people of Flores Island are derived from various ancestries.

P1-65) MALARIA TRANSMISSION-BLOCAKDE OF GENETICALLY-ENGINEERED MOSQUITOES EXPRESSING THE IMMUNOTOXIN GENE

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The release of mosquitoes that are genetically manipulated to destroy the malaria parasite is being considered as a possible method for malaria control. We have previously constructed a gene encoding of a single-chain antibody fragment (scFv) specific for Pbs21 molecule expressed on the ookinete surface. The scFv, designated 13.1 scFv, bound to the surface of P. berghei ookinete, and blocked oocyst development in the mosquito midgut by at least 93%, as assessed by oocyst counts in mosquitoes. To examine whether expression of 13.1 scFv in the mosquito midguts could lead to transmission-blockade of malaria in the mosquito stage, we generated two kinds of transgenic Anopheles stephensi mosquitoe lines that either intracellularly or extracellularly express the 13.1 scFv-Shiva gene under the control of the gut-specific and blood-inducible An. gambiae carboxypeptidase (AgCP) promoter using Minos germ line transformation. The embryos were microinjected by using glass needles with a mixture of Minos-13.1 scFV-Shiva transfer plasmid and Minos transposase plasmid. The Minos transposase mediates integration into the genome of An. stephensi cells. Hatched larvae were analysed on an inverted microscape at a wavelength of 490 nm to detect EGFP expression. Real-time PCR analysis showed that expression of 13.1 scFv-Shiva mRNA is specifically induced (8-fold) in the guts of transgenic mosquitoes, with peak expression at 3 h after a blood meal. By 48 h after a blood meal, mRNA abundance returns to a level close to that present before a blood meal. To examine inhibition of oocyst formation in the transgenic lines, wild-type and transgenic mosquitoes were allowed to feed on the same P. berghei-infected mouse. Oocyst formation in one of transgenic lines were significantly reduced (50%) compared with that in wild type.

P1-66) LEISHMANIASIS RECIDIVA CUTIS DUE TO *LEISHMANIA (VIANNIA) PANAMENSIS* IN SUBTROPICAL ECUADOR : ISOENZYMATIC CHARACTERIZATION

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Leishmaniasis recidivans (LR) is also known as relapsing, lupoid or tuberculoid chronic form of cutaneous leishmaniasis in the Old World (WHO, 1984). As synonymous term, leishmaniasis recidiva cutis (LRC) is used in the New World cutaneous leishmaniasis (Paes-Oliveira, 1977). Leishmaniasis is an endemic and wide distributed parasitic disease in Ecuador. The disease is present in most of its recognized tegumentary forms : localized cutaneous, diffuse cutaneous, disseminated cutaneous and mucocutaneous form. In the Ecuadorian tropical and subtropical areas the predominant species are L. (V.) panamensis and L. (V.) guyanensis (Armijos et al., 1997). The mucocutaneous form or espundia is present in the Amazon region and is caused by L. (V.) braziliensis (Calvopina et al., 2001). The present study was performed at north-west of Ecuador. Six patients had lesions that healed clinically 2 to 4 years ago, but presented later with two or more reddish papulas expanding at the periphery

and leaning towards the scarring resolution in the central area. Parasites were isolated by syringe aspiration and cultured in USMARU and RPMI medium. The tissue sections taken by biopsy showed a granulomatous dermal infiltrate. The anamnestic data, together with the clinical and histopathologic findings, support the diagnosis of LRC in Ecuador. Cellulose acetate electrophoresis analysis of the isoenzyme profiles was performed; 5 isolates of these 6 patients were identified as L. (V.) panamensis. There are some discussions about if the recurrent lesions are relapsing or reinfections (Saravia et al., 1990). In our patients there is unlikely to be occurred reinfection because the lesions recidives at the border of an old scar of previous ulcer suspected to be CL. Further, in this population the proportion of incidence rates are lower; hence the proportion of incident cases due to relapses rather than new infections is likely to be relatively high. The persistence of living parasites around or in the "cured leishmaniasis" had been demonstrated in several studies, suggesting that reactivation is the likely mechanism for these patients. Finally, we believe that LRC is not uncommon in the New World leishmaniasis, but is unreported.

P1-67) CASES OF LEISHMANIASIS IN ECUADOR (2002)

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We report our field study on leishmaniasis in Ecuador (October 15 - October 24, 2002). Six out of 238 primary school-children (5 - 14 years old) and 8 out of 14 inhabitants who wished skin examination (1 - 42 years old) exhibited cutaneous leishmaniasis. In addition, 42 school-children (17.6%) exhibited scars which likely results from old skin leishmanisis. Eight patients received skin biopsy for histological confirmation. Protozoans were cultured from six of them. All cultured protozoan belonged to subgenus *Viannia*. In two cases, DNA sequences of the cytochrome b gene of cultured protozoan matched to that of *L*. (*L*.) *braziliensis*. Prevalence of leishmaniasis appeared to be an important issue in Ecuador.

P1-68) IDENTIFICATION OF SPECIFIC ANTIGEN COMPONENTS OF *LEISHMANIA* DONOVANI BY TWO DIMENSIONAL GEL ELECTROPHORESIS

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Visceral leishmaniasis (VL) is one of the severe parasitic diseases. Though demonstration of causative parasite in the aspirates of spleen, bone marrow and lymph nodes is the most accurate way of VL diagnosis, these methods are not suitable in the field due to lack of sensitivity and difficulties in sample collection. Therefore, the diagnosis is greatly depending on serological tests. We previously reported a urine-based ELISA for the diagnosis of VL and found that the Leishmania donovani crude antigen used in that study showed some cross-reactions with non-VL urine samples. Visceral leishmaniasis (VL) is one of the severe parasitic diseases. Though demonstration of causative parasite in the aspirates of spleen, bone marrow and lymph nodes is the most accurate way of VL diagnosis, these methods are not suitable in the field due to lack of sensitivity and difficulties in sample collection.

Therefore, the diagnosis is greatly depending on serological tests. We previously reported a urine-based ELISA for the diagnosis of VL and found that the Leishmania donovani crude antigen used in that study showed some cross-reactions with non-VL urine samples. In this study, we tried to identify the specific antigen components of L. donovani using 2-D gel electrophoresis to decrease the cross-reactions. We extracted the soluble antigens of L. donovani with Ready Prep TM, Sequential Extraction Kit, BIO-RAD. The antigens were then separated with 2-D SDS-PAGE and transferred on to PVDF membrane. After applying urine or serum samples of VL patients and healthy control, specific spots for VL patients were determined. Among the spots detected, 4 spots with a molecular weight of about 69-kD, 65-kD, 55-kD, and 50-kD were specifically detected by all VL patient's samples.

Determination of amino acid sequences with MALDI-TOF Mass Spectrometry and molecular cloning of the identified proteins is underway.

P1-69) CATHEPSIN L IS CRUCIAL FOR TH1-TYPE IMMUNE RESPONSE DURING LEISHMANIA MAJOR INFECTION

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Prior to the activation of CD4⁺ T cells, exogenous proteins are digested by endo/lysosomal enzymes in antigen-presenting cells (APC) to produce antigenic peptides that are presented on MHC class II molecules. Studies described here inspect the functional significance of cathepsin L for antigen processing and Th1/Th2 differentiation in experimental leishmaniasis. We first demonstrated that cathepsin L is one of candidates for endo / lysosomal enzymes in processing of soluble *Leishmania* antigen (SLA) by using CLIK148, a specific inhibitor of cathepsin L. Treatment of BALB/c or DBA/2 mice with CLIK148 exacerbated the disease by enhancing SLA-specific Th2-type response such as IL-4 production. CLIK148 did not exert any direct influence on either of *L. major* promastigotes themselves or the course of *L. major* infection in SCID mice. Taken together, these findings suggest that treatment of host mice with CLIK148 affects the processing of SLA in APC, resulting in the potentiation of Th2-type immune responses and thus leading to exacerbation of the disease. Furthermore, endo/lysosomal cathepsin L was found to be functionally distinct from previously described cathepsins B and D.

P1-70) PRODUCTION OF MONOCLONAL ANTIBODY THAT RECOGNIZES THE PROMASTIGOTE AND INTRACELLULAR AMASTIGOTE STAGES OF LEISHMANAI MAJOR

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Leishmania major (L. major) is a primary causative agent for cutaneous leishmaniasis. mAbs specific to the promastigotes and amastigotes stages, of Leishmania are needed for accurate, rapid, early clinical diagnosis, and efficacy of drug therapy. In this study, to recognize intracellular amastigote and promastigote stages of L. major, two monoclonal antibodies (mAbs) were developed by immunizing female BALB/c mice with 1x109 L. major parasites by i.d injection. The sensitized mice spleen cells were fused with a mouse myeloma cell line, SP2/0-Ag14, using polyethylene glycol 1500, three days after a final booster immunization of sonicated L. major promastigote via i.p injection. Hybridomas producing mAb reactive to L. major parasites, were screened by indirect immunofluorescence (IF) staining. Hybridomas showing positive results were termed as W-2 and W-10. The hybridomas (5x 105) producing mAb against L. major were i.p injected to SCID mice. After 8-14 days mice were sacrificed and peritoneal fluid was collected and processed for purification. Newly generated mAb efficacy was analyzed by indirect IF staining, Western blotting, Confocal laser micrscopy, Immunohistochemistry, and flow cytometry. The purified mAbs, recognized a conformational epitope present at 62-kDa component, as verified by Western blotting. Indirect IF disclosed that the antigen

recognized by both mAbs distributed homogeneously on

the parasite surface. Intracellular (J-447 and THP-1)

amastigote identification, by using both mABs, was con-

firmed by confocal laser microscope and flow cytometry. Immunohistchemistry performed on L. major infected BALB/c mice (frozen section) and J-447 cell line macrophages, showed positive results for intracellular amastigotes. Both mAbs showed negative reaction to L. (L.) tropica, L (L.) aethopica, L. (L.) donovani, L. (L.) infantum, L. (L.) chagasi, L. (L.) mexicana, L. (L.) amazonensis, L. (L.) granhami, L. (V.) braziliensis, L. (V.) panamensis and L.(V.) guyanensis. We conclude that both mAbs are expected to be useful in clinical, epidemiological studies and also for the efficacy of new drug therapies against L. major.

P1-71) CREATION OF HYBRID TETRAPLOID BY CROSS-BREEDING BETWEEN PARTHENOGENETIC *FASCIOLA* SP. TRIPLOID AND *FASCIOLA HEPATICA*

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In the common liver flukes two species, Fasciola *hepatica* and *F. gigantica*, which are diploids (2n=2x=20)and reproductively bisexual are known. Parthenogenetic flukes Fasciola sp. occurring in Japan, Korea and southeast Asia include three forms, diploid (2n=2x=20), triploid (2n=3x=30) and mixoploid (consisting of diploid and triploid cells, 2x/3x). To elucidate the origin of the parthenogenetic flukes, we conducted cross-breeding between triploid Fasciola sp. and F. hepatica to raise hybrid flukes. Four rats were inoculated with metacercariae of F. hepatica and triploid Fasciola sp. and mixed infection was established in two rats. The flukes taken out from the host rats were F. hepatica which had spermatheca filled with sperms and Fasciola sp. which had spermatheca without sperms or with few sperms, if any. Intrusion of sperm of F. hepatica into primary oocytes of Fasciola sp. was confirmed. Metacercariae raised from the eggs collected from the flukes of Fasciola sp. were inoculated into 21 rats, with 6 to 20 metacercariae to each rat, and twenty adult flukes were obtained. Observation of chromosomes in spermatogonia of the flukes disclosed that

the flukes were tetraploids showing 2n=4x=40 proving that the flukes were hybrids raised from fertilization between triploid egg (n=30) of Fasciola sp. and haploid sperm (n=10) from F. hepatica. In the tetraploid hybrid most of the primary spermatocytes showed so complicated configurations of chromosome pairing that precise analysis was not successful. However, in a few primary spermatocytes, 10 trivalents and 10 univalents were confirmed. The fact implies that the hybrids are allotetraploids consisting of three sets (genomes) of chromosomes derived from the parthenogenetic triploid Fasciola sp. and a set (genome) from diploid F. hepatica. Spermatogenesis and spermiogenesis were quite abnormal in the tetraploid hybrids. It is to be noticed that 30 univalents in the meiocytes of triploid Fasciola sp. showed pairing in trivalents in the meiocytes of the allotetraploid hybrids in which genomes of Fasciola sp. and F. hepatica were coexisting in the cells. In our previous observation hybrid flukes (2n=3x=30) between diploid Fasciola sp. (2n=2x=20) and F. hepatica (2n=2x=20)showed 10 bivalents and 10 univalents in the meiocytes (Terasaki *et al.*, 2003). Therefore, it is possible to infer that diploid and triploid forms of parthenogenetic *Fas*-

ciola sp. have common genomes which are remotely related to that of *F. hepatica*.

P1-72) PRESENT SERIOUS SITUATION OF TAENIASIS AND CYSTICERCOSIS IN INDONESIA CONFIRMED BY APPLICATION OF IMMUNOLOGICAL AND MOLECULAR DIAGNOSTIC TECHNOLOGIES

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Taeniasis and cysticercosis in Indonesia is briefly reviewed. WHO has declared that taeniasis/cysticercosis of Taenia solium is one of the most serious but eradicable parasitic zoonoses worldwide 2003. In Indonesia, north Sumatra, human taeniasis caused by Taenia asiatica (= Taenia saginata asiatica) is still endemic through traditional consumption of minced pork and viscera. Bali has well known to be highly endemic of cysticercosis of T. solium but recently it is rather not easy to detect human cysticercosis and impossible to detect pigs infected with T. solium. This is due to the drastic change in pig farming from roaming to indoor and block of scavenging of human feces. However, in contrast, taeniasis of Taenia saginata has become highly endemic, since local people want to enjoy beef as well as pork now. It is unclear the reason why T. saginata prevalence is so high in Bali. In Irian

Jaya, taeniasis and cysticercosis due to Taenia solium is serious and may be the worst in the world : 47.8% of adult local people, 70.4 & of pigs, 10.9% of dogs show specific antibody responses to Taenia solium glycoproteins. Local people of 15.9% in 1999 in villages and 8.6% in 2002 showed copro-antien positive responses for taeniasis and expelled T. solium adult worms. Subcutaneous cysts from local people, cysts removed from pigs and dogs and adult worms and eggs expelled from taeniasis carriers have been confirmed to be T. solium though mitochondrial DNA analysis and morphological confirmation of metacestodes developed in NOD/Shi-scid mice injected with in vitro hatched oncospheres. It is stressed that modern technologies of immunology and molecular biology are essential for crucial evidence of infection.

P1-73) APPLICATION OF MULTIPLEX PCR TO MOLECULAR EPIDEMIOLOGY AND CONTROL OF TAENIASIS/CYSTICERCOSIS

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Multiplex PCR was established for differential diagnosis of taeniasis/cysticercosis including their causative agents. Cytochrome *c* oxidase subunit 1 gene (*cox 1*) was amplified as a target gene. For the molecular identification of taeniid cestodes, mitochondrial DNAs were prepared from a total of 57 parasite materials and multiplex PCR was performed. Taeniid species- and *Taenia solium* genotype-specific forward primers and a reverse primer common to taeniid species were used. Multiplex PCR yielded evident products unique for *Taenia saginata*, *T. saginata asiatica*, and Asian and American/African genotypes of *T. solium*. For the detection of taeniasis patients or tapeworm carriers using multiplex PCR, copro -DNA samples were extracted from a total 28 fecal samples with *T. saginata* (n=5), *T. solium* from Indonesia (n=9) and *T. solium* from Guatemala (n=14). Fecal samples from individuals not infected with any parasites were examined as negative control. By the multiplex PCR using copro-DNA, diagnostic products were detected from all carriers with *T. saginata*, whereas diagnostic products were amplified from 5 and 7 *T. solium* carriers from Indonesia and Guatemala, respectively. It was worth that multiplex PCR product was detected form a worm carrier who expelled only immature proglottids, indicating that it is possible to detect tapeworm carriers before they release mature gravid proglottid as an infectious source. This leads to effective control of taeniasis.

P1-74) CHANGES OF MATERNAL ANTIGEN SPECIFIC IGG4 IN URINE SAMPLES FROM JINFANTS IN BANCROFTIAN FILARIASIS ENDEMIC AREA

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Background

We developed an ELISA with urine samples for diagnosis of lymphatic filariasis and have been using the method for epidemiological surveys. Urine can be collected safely and easily even from babies. Antigen specific antibody levels in young generation give information on a recent figure of transmission of the disease in the area. In a filariasis endemic area in Sri Lanka, we found that antigen specific IgG4 antibody levels become positive at 3 years old or older. At the same time, we found antibody positives in infants and the antibodies were suspected to be from their mothers. In this study, we selected antibody levels of their babies to observe (1) the antibody levels in babies born from the positive mothers and (2) changes of the antibody levels.

Subjects & Methods Urine samples from 14 pairs of mothers and their babies were collected monthly up to 6 months and 12 months after birth in a filariasis endemic

area in Matara, southern part of Sri Lanka. Urine collection bags were used to collect urine from babies. Anti-*Brugia pahangi* IgG4 levels in the urine samples were measured by ELISA.

Results & Discussion

In urine samples of 12 out of 14 babies born from mothers with anti-B. pahangi IgG4 positives, collected 1 week after birth, the IgG4 levels were almost the same with their mothers' levels. The IgG4 levels were maintained 3 to 6 months after birth, then declined and disappeared by 1 year. In other two cases, no anti-B. pahangi IgG4 was detected throughout the period studied. The IgG4 levels of babies given artificial milk earlier showed earlier decline.

These results suggest that (1) anti-*B. pahangi* IgG4 in babies come from their mothers through placenta and (2) the levels were maintained by supply of the antibodies through breast milk.

P1-75) PHYLOGENETIC RELATIONSHIP AMONG SEVEN SPECIES OF FROM A VIEWPOINT OF A CO1 REGION OF MITOCHONDRIAL DNA

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In order to clarify molecular phylogenetic relationships among Japanese species in the genus *Cercopithifilaria*, base sequences in the CO1 gene region of mitochondrial DNA were determined from seven Japanese species (Japanese serow-derived 5 species, *C. shohoi*, *C. multicauda*, *C. minuta*, *C. tumidicervicata* and *C. bulboidea* ; Japanese sika-derived 2 species, *C. longa* and *C. crassa*), and then, tree analysis was carried out by computer on phylogenetic relationships among these 7 species using the base sequences. First of all, no base substitutions were observed between *C. bulboidea* and *C. longa*, suggesting that recent host switching of a lineage of *C. bulboidea* from the serow family to the cervid family gave rise to *C. longa*. Secondly, in all computer analyses a morphologically ancestral type, *C. bulboidea*, was branched last, suggesting this species is of a derived type. *C. multicauda* was branched first in the computer analyses. It seems therefore that *C. multicauda* is the oldest of the seven species.

P2-1) FEATURES AND CHALLENGES OF THE GHANA POVERTY REDUCTION STRATEGY (GPRS) HEALTH COMPONENT

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[Background]

In September 1999, the World Bank and the IMF launched the HIPC (Highly Indebted Poverty Country) Initiative, which allows debt relief for the eligible countries and gives supplementary IDA loans to facilitate the poverty reduction process. To qualitfy, the HIPCs should prepare a Poverty Reduction Strategic Paper (PRSP) and implement a three-year comprehensive poverty reduction strategy. Health is a popular component of PRSP because it is closely related to poverty. In April 2002, Ghana joined the Initiative after developing the PRSP called 'the Ghana Poverty Reduction Strategy; GPRS'.

[Purpose]

To describe the features and challenges of the GPRS health components in comparison to the Health Sector Wide approach (SWAp) which has been implemented since the mid 90's.

[The features of the GPRS Health Component]

The GPRS health component consists of four sub chapters; HIV/AIDS, Population Management, Health, and Safe Water and Environmental Hygiene, in Chapter VII 'Human Development' along with other sectors. Due to a budgetary delay and shortfall, the presidentially prioritised programmes; HIV/AIDS control, Establishment of the Model Health Centres and Introduction of the Health Insurance and Exemption were listed in the annex.

[Challenges]

1. Since the contractors outside of the Ministry of Health developed the GPRS, it is not consistent with the Health SWAp. These efforts should be coordinated to avoid duplication of funding and human resources.

2. In addition to the HIV/AIDS programme, the

GPRS should also describe the malaria programme based on the high morbidity and mortality of this disease and tremendous socio-economical impact.

P2-2) SITUATION ANALYSIS OF HEALTH MANAGEMENT INFORMATION SYSTEM IN MOROGORO PROVINCE, TANZANIA

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General Objectives

The Health Management Information System (HMIS) in Morogoro Health Project by JICA was analyzed.

Methods and Materials

A participatory workshop was conducted to identify problems concerning HMIS. We interviewed responsible personnel and examined recording/reporting formats at various health institutions.

Findings and Discussions

1. HMIS of Council Health Management Teams (CHMTs) was not functioning well due to unreliable data, inadequate training, inadequate coordination among CHMT members, and logistics. Poor supervision led to difficulty in preparing Comprehensive Council Health Plan (CCHP).

2. Several new indicators such as occupational health-related indicators, population growth rate, death due to accidents, community health fund participation rate, etc. were proposed for CCHP, in addition to the essential indicators.

3. It is necessary to develop peer review-type horizontal supervisory mechanisms, as well as to strengthen current vertical supervision among health centers and dispensaries.

P2-3) THE IMPACT OF THE HEALTH SECTOR REFORM IN YEMEN

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Background : The government of Yemen began a structural adjustment facility in1998 with support from the IMF. Health sector reform is a part of this strategy. In October 2002, the Poverty Reduction Strategy Paper (PRSP) was release, and was well received by the World Bank and IMF, as well as by major bilateral donors. This paper will broadly review the possible effects of the PRSP. Present situation : The minister of public health revealed that more than half of the government health staff lives below the poverty line. This is considered the one of main reasons for the deterioration of the quality of health services. Moreover there is almost no government or private health insurance system available for the public. Accordingly, patients are asked to pay out-of-pocket for exami-

nations and treatments. This situation causes serious economic hardship for families. Future plan : According to the PRSP, the Yemen government will increase the health expenditure from 1.5% to 2.2% by the year of 2005. Privatization also will be introduced in certain medical fields. Interpretation : Health sector reform will increase the budget for opening new basic health units and employing the new health staff. The introduction of the private sector into public health care may affect the quality of health services. A follow up study should be carried out to evaluate the degree to which health sector reform supported by international organizations and Yemen government has benefited the people of Yemen.

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P2-4) IMPACT OF REALLOCATION OF HEALTH STAFF AT THE PRIMARY LEVEL ON USAGE OF THE PRIMARY HEALTH SERVICES IN NANDAIME, NICARAGUA

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Reallocation of health staff at the primary level was conducted on December 2002 in Nandaime, with technical support of CIES, a JICA health project. Nandaime has a population of 40,000 people (70% rural). In rural areas, 4 health posts were staffed permanently. Prior to the reallocation, one full-time assistant nurse and one general physician a few days per week were allocated to the health posts. In 2003, one assistant nurse, one professional nurse and one general physician were allocated as permanent staff at health posts. By comparing the use of health services over three months from January to March of 2002 and 2003 using statistics of Nandaime municipal health office, the impact of reallocation of health staff was evaluated. The total number of health consultations of the 4 health posts was increased from 2974 in 2002 to 4191 in 2003. With regards to the health services, 0 year child health check 266 to 280, 1-4 year child health check 317 to 713, prenatal care 138 to 204, post-partum health check 16 to 36, family planning 552 to 708, all were increased. Consultations with doctors increased 1658 to 2194, and with nurses 1316 to 1997. On the other hand, consultations at the health center were not decreased. In developing countries, limited health resources should be used effectively. By conducting research for health reorganization and reallocation of health staff at the primary level, we could increase the usage of health services of people in rural areas without negative impact.

P2-5) THE SURVEY OF THE RATIONAL USE OF DRUGS IN THE REPUBLIC OF HODURAS

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[Background and object] In Orancho district in Honduras, the district health office has a medical storehouse. From the storehouse, medicines and medical equipment are sent to local health facilities. This distribution used to be performed only once or twice a year, due to the lack of a transportation system. However, the distribution cycle has been shortened to once in three months after improving the stock management. As a proper management of medicines is a key issue in the district, we have investigated the rational use of medicines from April to May, 2003.[Method]In total we collected 156 cases from the San Francisco hospital and ten health centers. In addition to basic information, such as a patient name, age, sex, and diagnosis, we recorded the conversation between the doctor and the patient. We then evaluated the use of medicines by studying the doctor's explanation about the contents of prescription and medicine, directions for use of the medication, etc. [Result] Of 156 cases, 122 (78.2%) were prescribed medicines, and 26 (21.3%) prescriptions were not suitable for the rational use of drugs. The major problems included, medicines not being prescribed according to evidence-based medicine, and excessive use of antibiotics, iron, and folic acid. [Consideration] In Honduras, it is very difficult to assure the rational use of drugs

due to the lack reliable clinical data. Despite such difficulties, the Honduras Ministry of Health needs to develop basic manuals for the rational use of drugs, and to provide more training to the health workers.

P2-6) PARTICIPATORY EVALUATION IN HONDURAS

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Introduction : Participatory evaluation is a form of self-evaluation conducted by internal staff members as well as by a wide range of external personnel. Although this method has been embraced by international donor agencies since the 1980's, it is rarely used in Japan. Our report is about a participatory evaluation conducted during the 3rd year of the Project. Method : The total of 184 people participated in the midterm evaluation from the Project, the Ministry of Health of Honduras, JICA, the evaluation mission from Japan, nine international donor agencies, and the Health Project of JICA Nicaragua. The midterm evaluation was a 5-day workshop of 3 sessions ; 1) introductory workshop to collect questions from the Project participants including Japanese experts and the

evaluation mission from Japan. The five items of the evaluation of DAC and two additional items, strategy and tactics, were adopted for evaluation, and 3) final workshop to answer the questions asked at the introductory workshop, and to conclude the evaluation with the external evaluators. Conclusion : The objectives of participatory evaluation are 1) to improve managerial competence, 2) to encourage the local ownership of the project, 3) to provide effective feedback, and 4) to improve accountability. We could achieve all these objectives except improvement of managerial competence. It was concluded that participatory evaluation is a more effective method than conventional evaluation, especially for midterm evaluation, in which prompt and effective feedback to a project is required.

P2-7) EVALUATION OF ORAL HEALTH PROMOTION IN NEPAL : HAS OHI-S (DENTAL PLAQUE/STONE) DECREASED IN THE PAST TEN YEARS?

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Purpose we began conducting dental surveys and providing dental treatment in Nepal in 1989. Our services include dental treatment, nutritional advice, and oral health care education. With the progression from treatment to oral health care education, our strategy has shifted toward encouraging self-reliance among the local inhabitants. Now the majority of local people are aware of the importance of oral health and can maintain their own oral health. Therefore, we evaluated our work over the past 10 years by measuring changes in the oral hygine index (OHI-S). Method We compared the oral hygiene index (OHI-S) of 2002-2003 with that of 1992-1993 for people over the age of 10 years. OHI-S evaluates dental plaque and calculus attached to the tooth surface. Result Debris index (DI) and Calculus index (CI) increased with increasing age. However, the OHI-S for 2003 was lower than for 1993. DiscussionReduction of the bacterial count from the oral cavity is very important in preventing caries and periodontal diseases. At present, an effective method to eliminate dental plaque and calculus is using a tooth brush and a plaque-removing device. In Nepal ten years ago, few people brushed their teeth. We have seen an improvement in oral hygine (OHI-S) and a decrease in oral disease in the area where we have carried out our work. We will continue to educate the local inhabitants about oral health care and encourage their self-reliance. We hope that this work becomes a model project and that it can be used throughout Nepal.

P2-8) THE EVALUATION OF DENTAL TREATMENT IN THECHO VILLAGE, NEPAL

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We, members of the Association Dental Cooperation in Nepal, have made 16 visits to Nepal to carry out dental missions. We have treated an especially large number of patients in Thecho village. Recently, we have been shifting our activities from medical care towards health care, but, as always, many villagers require dental treatment. Therefore, we will shape our so that that we can bring medical care and health care together, and still provide dental services as needed. To best accommodate the needs of the villagers, we wanted to analyze the chief complaints and treatments needed by dental patients in Thecho village. We analyzed dental treatment data for Thecho villagers from the 3rd mission to 16th mission. We classified the chief complaints into seven groups : tooth pain, request for extraction, request for filling, request for scaling, painful or bleeding gums, suspected caries, and others. We classified treatment into five groups : extraction, filling, scaling, endodontics and others. We calculated the percentage that each category represented for each of the missions and statistically analyzed the results. Despite our efforts, circumstances and habits have been changing in Thecho village, and a few problems have appeared, such as increased sugar intake. However, the decrease in patients complaining of tooth pain is the fruit of our long labor. Nonetheless, dental problems persist, reflected in the many extractions we performed. To solve these problems, we plan to incorporate a greater emphasis on preventive treatments for younger people into the project in the future.

P2-9) INVESTIGATION OF FOOD AND LIVING STYLE IN LOCAL AREA FOR DENTAL HEALTH DEVELOPMENT OF NEPAI

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For dental health development in local area of Nepal, investigations oftarget villages were carried out. The investigations consisted of food style, infrastructure of village, personal income and other life conditions relatedto dental health. Results are as follow. Population of Chapagaun village was about 10,000 and Sunacoci was about6,000. Income was 6,000 to 8,000 Rp (1Rp=3Yen) /month /family for Chapagaonvillagers and 5,000 to 6,000 132

Rp /month /family for Sunacoci villagers. Condition of tap water, toilet and kitchen were not so bad. Especially, thekitchen style was changing in Chapagaon. Several families had refrigerators, electric rice cookers and LPG cooking equipment. The food life style was changing from 2 meals a day to 3 meals a day in both villages. Also, they are starting to eat bread and cracker instead of tradifional food like a rice or corn. In these villages, bakery stores and meat shops were observed. Jelly shops, Candy shops and sugar shops were observed at many places in villages. 36 of these kinds of shops were in Chapagaon, and 45 shop in Sunacoci. This investigation showed that these two villages had little differences, but mostly the two village were leaving the traditional life style for a city life style. From these investigation, we have conformed that dental health supports in these two villages, are needed.

P2-10) INTERNATIONAL DENTAL HEALTH EDUCATION BY FUKUWARAI GAM

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[Introduction]

Dental health is critically important, particularly in developing countries, where basic dental education is needed. In cooperation with a dental nurse in Cambodia, we present a broad view of dental education and strategies for improving dental health in primary schools.

[Method]

- 1) Dental education for the nurse and teacher.
- 2) Making a game and textbook for children.
- 3) Drawing a dental health picture for the family. [Material and Contents]

The FUKUWARAI game is shown in the figure. In this game, children can remove eyes, tongue and teeth. The children complete the face, observing its features and oral structures, and become curious. We ask them questions "Why do you need any teeth for the body?" After the explanation, we show pictures of caries, gingivitis, and tooth brushing, distribute a mirror, dental brush and cloths, and then practice tooth brushing.

[Conclusion]

1) The resident nurse has used this dental health activity in another primary school and village.

2) After the dental health activity, 100% of 50 children who were surveyed answered, "we had a lot of fun"

3) The oral hygiene of the teacher and school children in the primary school has been improving.

[Discussion]

Education is an important worldwide problem. The use of pictures and games make this an inexpensive and convenient approach to education, and one that be easily continued in a developing country. The visual approach provides knowledge of dental health naturally, and helps improve oral hygiene.

P2-11) THE PROBLEMS ON THE PERIPHERAL NERVOUS SYSTEM OF THE LOCAL RESIDENTS DRINKING ARSENIC-CONTAMINATED WATER IN INNER MONGOLIA, CHINA

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Arsenic (As) is a naturally occurring element with no smell and notaste. Most arsenic compounds dissolve in water. Arsenic is known as a hazard to human health. According to the WHO Guidelines for Drinking-Water Quality (1993), the guideline value of arsenic is 0.01 mg per liter. Recently in some regions in the world, its adverse health effects have appeared among people drinking arsenic-contaminated water for long time. In some areas Inner Mongolia, China, well-water for drinking has been found contaminated by arsenic exceeding WHO guideline value.

In September 2002, we visited to investigate local residents in a village in Wuyuan prefecture, Inner Mongolia, where well-water for drinking is contaminated by arsenic exceeding WHO guideline value. The subjects are 84 persons (45 men and 39 women) of age between 20 and 59, with informed consent. Arsenic concentration ([As]) of their drinking water is quantitatively analyzed. Nerve conduction velocity examination was conducted to measure distal latency of motor nerve (DL), motor nerve conduction velocity (MCV), and sensory nerve conduction velocity (SCV). Linear regression analysis was conducted, using [As] as independent variable, and DL, MCV and SCV as dependent variables.

Correlation between [As] and DL is, y = 0.003x + 3.682 (R² = 0.032).

Correlation between [As] and MCV is, y = 0.009x + 55.347 ($R^2 = 0.005$).

Correlation between [As] and SCV is, y = -0.005x + 54.979 ($R^2 = 0.001$).

So far, little has been known about the effect of lower-level, long-term exposure to arsenic on nervous system. Further investigation is needed.

P2-12) EVALUATION OF A SIMULATION OF WATER-DRAWING BURDEN AFTER THE SELECTIVE SEALING OF ARSENIC-CONTAMINATED WELLS

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Arsenic contamination of drinking water wells in Inner Mongolia, China has emerged as a problem since the 1990s. The sealing of heavily contaminated wells is the most immediate countermeasure to prevent arsenic poisoning. It was observed that within a village, nearly all wells had levels of arsenic well above the safe standard set by the Chinese Government and the WHO, and that the levels of contamination varied widely from place to place. The goal of this study was to determine how much extra work would be involved in drawing water if the most heavily contaminated wells were sealed one by one, in order of contamination. The burden of drawing water (1 km/day) was defined as the product of the quantity of water used per day (l/day) and distance between a well and a water jar (km). Assuming that if the closest well was sealed that villagers would use the next nearest well, the total burden was repeatedly calculated for each additional sealed well. If the allowable burden was 20 to 40 l km/day, our simulation showed that we could immediately reduce arsenic poisoning without dramatically increasing the work of drawing water by sealing 12 wells or by setting maximal acceptable levels of arsenic at 178ppb. However, it should be noted that villagers would continue to use wells with arsenic levels above the safe standard. The approach used in this study provides evidence that the sealing of the most heavily contaminated wells could be an effective new approach for controlling arsenic tox-

P2-13) HOW DID DIFFERENCE OF IODIZED SALT USAGE PATTERNS AFFECT TO GOITER RATES?

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Purpose : The purpose of this study was to compare occasional uses of iodized salt in the reduction of goiter rates by a longitudinal study in Mongolia. To do this, we provided iodized salt at different intervals to households that had never used it, and then monitored the goiter rate of children in these households over the course of one year.

Methods : In two provinces in Mongolia, selected households of 297 children (age : 7 to 11 years old) were randomly assigned to one of the three groups. Each group received 2 kg of iodized salt either monthly, bimonthly, or quarterly, from November of 2000 until April of 2002. At the pre and post study examination, the size of each child's thyroid gland was measured using ultrasonography and the goiter rates were recorded.

Results : The initial goiter rate was 56.6 percent. We found goiter rates of less than 5 percent in each group in 2002.

Conclusion : The goiter rates of each of the above three groups was normalized to less than 5 percent by the addition of iodized salt to their diets. This indicates there was no longer iodine deficiency among any of the groups. This suggests that households that use a certain amount of iodized salt, even though they did not use it regularly, should be considered as effective iodized salt users in Mongolia.

P2-14) IODINE PROPHYLAXIS AROUND THE SEMIPALATINSK NUCLEAR TESTING SITE, REPUBLIC OF KAZAKHSTAN

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Objective : The aim of this study was to clarify the iodine deficiency status in the Semipalatinsk region, which was contaminated by radioactive fallout from nuclear testing during the Soviet era.

Design : As the part of the Japan-Kazakhstan joint project of adult cancer screening around the Semipalatinsk Nuclear Testing Site (SNTS), spot urine specimens were collected at random in each village from May to October 2002. Separately, children aged 5-15 years from around the SNTS were chosen at random and their spot urine specimens were collected as well.

Subjects : A total of 2609 adults aged over 40 years from 16 settlements in 3 regions and one city, and 298 children aged 5-15 years from 2 regions and one city.

Results : Median urinary concentrations of adults and children in all regions ranged from 116.0-381.7 and 127.7-183.0microg/l, respectively. The highest prevalence of values less than 50microg/l (14.1%) did not exceed 20%. Distributions within each group, adults and children, showed almost the same pattern, except for one region. More than 50% of samples were above 100microg/l in iodine concentration in every group.

Conclusions : In agreement with our previous studies, the urinary iodine concentration data showed no clear evidence of iodine deficiency around the SNTS. Kazakhstan is geographically and nutritionally at moderate risk of iodine deficiency disorders without fortification of iodine replacement by iodized salt. The socio-medical prophylaxis against iodine deficiency has been successfully maintained in East Kazakhstan.

P2-15) DESCRIPTIVE EPIDEMIOLOGY OF GASTRIC CANCER IN CENTRAL AMERICA

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Background : There is little etiological information about stomach cancer in Guatemala. Therefore, we studied the epidemiology of stomach cancer in this country.

Method : We analyzed available epidemiological data, such as the official publication by the Ministry of Health of Guatemala from 1985 to 1989 and IARC Scienentific publications.

Results : The number of total cancer death is consistently increasing from 1960s. With improvement in sanitary conditions leading to fewer deaths from infectious disease, cancer is expected to become a more common cause of mortality. For both sexes, stomach cancer is the third most frequent cancer. For male, the mortality rate for stomach cancer is 8.3 per 100,000, while the female corresponding rates were 9.5 in 1989. One surprising finding is that the male : female ratio was consistently below 1 from 1985 to 1989. To confirm this findings we specifically analyze various cases form urban areas, which are likely to be more accurate due to availability of medical services, and found the tendency was generally the same.

Conclusion : The mortality rate for stomach cancer is higher among females than males. At the present time, it is difficult to identify the causes of this discrepancy, because of difficulties in confirming this type of information in this country. Further follow up studies are needed to better understand the etiology of stomach cancer in Guatemala.

P2-16) RESULTS OF THE PROJECT ON STRENGTHENING OF HEALTHCARE IN THE SOUTHERN REGION IN JAMAICA-FOR THE THIRD COUNTRY TRAINING

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Our project, Strengthening of Health Care in the Southern Region of Jamaica, ended in May of 2003. One

of the major accomplishments of this initiative was the establishment of health examination systems for lifestyle diseases, which are prevalent in this country. It is the first regional disease prevention program in Jamaica. We also established a diabetes class, targeting obese residents who are at greater risk for this disease. The course was run by our C/P themselves. This program triggered the formation of community health programs throughout the southern region of Jamaica. Our efforts also addressed health concerns throughout the Caribbean. The Ministry of Health and the Southern Regional Health Authority has been holding classes for health promotion for Caribbean countries, supported by JICA (third-countries-trainee). Our C/P is also preparing a training course for the public health sector of Jamaica's other regional health authorities before training a third country. This would allow them to spread their health promotion activities throughout the country. We believe that a third country trainee is able to be effective in motivating our C/P to continue their efforts after our project is completed.

P2-17) HOW CULTURAL FACTORS INFLUENCE ILLNESS : COMBINING QUALITATIVE AND QUANTITATIVE APPROACHES TO TUBERCULOSIS DIAGNOSIS AND TREATMENT IN YEMEN

JUNKO DATE

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Background : San'a, the capital of Yemen, has experienced accelerated socioeconomic development since the union of North and South Yemen in 1990. However, the people of San'a and the surrounding area maintain their traditional customs. The relationship between socio cultural factors and tuberculosis has been previously studied in several countries, but not in Yemen. Methods : The research was conducted from August 2000 to March 2003. Qualitative data were collected using standard anthropological methods. Quantitative data were collected for diagnostic delay analysis and treatment outcome, and was statistically analyzed by SPSS. Relationships between socio-cultural factors and tuberculosis were interpreted for pulmonary tuberculosis cases. Findings : Illiterate people have a longer diagnostic delay (median 12 weeks) than literate people (median 8 weeks). Illiterate patients are more likely to assume that they have a common illness, such as 'cough' or 'chest', rather than the unknown illness 'tuberculosis'. More female patients (69.7%) than males (46.3%) complete treatment. Many female patients have the full support of their male family members throughout their treatment. Interpretation : The tendency to assume that one has a 'common illness', without consideration of symptoms and diseases, causes a longer diagnostic delay among illiterate patients than among literate patients. The social role of men in Arab tribal society influences the different treatment outcomes between male and female patients. These findings are clear from the results of both the qualitative and quantitative approaches. It is interesting that different socio - cultural factors affect tuberculosis diagnosis and its treatment in the same population in this study.

P2-18) HEALTH PERCEPTIONS OF DIABETICS AND THEIR RELIANCE ON THERAPY AT DR. SARDJITO HOSPITAL IN YOGYAKARTA, CENTRAL JAVA, INDONESIA

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Aims : The incidence of diabetes has been increasing, not only in developed, but also in developing countries due to a rapid change in lifestyle. Information about how patients perceive and understand their own cases of diabetes is important in the management of this and other lifestyle-related diseases because our perceptions about health vary. Health perceptions may influence therapeutic compliance and self-care management. The purpose of this study was to examine the relationship between the health perceptions of diabetics and their reliance on treatment in Indonesia. Subjects and methods : Type 2 diabetics (n=157) were recruited at the outpatient unit of the Dr. Sardjito Hospial in Yogyakarta, Indonesia. They were requested to answer the following questions : 1) What do you think are the causes of your disease? and 2) What therapies influence your disease? Results : Using the chi - square test, we found that the diabetics who re-

lied on drug therapy believed that super-natural factors (destiny) and internal factors (character or predisposition) were responsible for their ill health. On the other hand, the diabetics who relied on non-drug therapy (diet or exercise) believed external factors (lifestyle-habit) were the cause of their illness Conclusion : For the management of lifestyle-related diseases such as diabetes, information about the health perceptions of patients is useful.

P2-19) DIABETIC PATIENTS' HEALTH LOCUS OF CONTROL IN INDONESIA

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Introduction. The diabetic population is rapidly increasing, both in developed and developing countries. We researched the patients' Health Locus of Control(HLC), for considering comprehensive approaches. **Methods.** From July 25th - November 25th, 2002, type2 diabetics (50-70 years of age) were recruited at the outpatient department of Dr. Sardjito hospital, Yokyakarta, Indonesia. They were requested to reply to questionnaires about HLC. High - HLC - score means that the people think that their health depends on their own behaviors and efforts, so that they have good adherence to self-care. We analyzed the patients' HLC with a Microsoft Excel. **Result.**

The total HLC scores in Indonesia were not significantly different from those of Japan. In Indonesia, men scored significantly higher than women. This is likely to be related to the Islamic culture of Indonesia. In Islamic society, men have greater power and higher positions, giving them the final say in their families or the societies. Consequently, they are accustomed to taking responsibility and making their own decisions. It can connect to their strong internal factors. In Indonesia, HLC depends on sex. We can use HLC and these results in considering effective disease management approaches.

P2-20) THE MEANING OF THE MEDICAL ANTHROPOLOGICAL POINT OF VIEW IN INTERNATIONAL HEALTH AND MEDICAL COOPERATION -THE CASE OF MALARIA IN THE SOLOMON ISLANDS

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Objective : This research shows the Solomon Islander's view of malaria and their concept of health, and considers the meaning of the anthropological perspective with respect to international health and medical cooperation in the future. Method : Semi-structured interviews were carried out based on results of a questionnaire administered to 102 residents in 3 different areas of the Solomon Islands. We asked about the respondents view of traditional medicine and its use, their basic knowledge of malaria, and their values with regards to health. Results and discussion : 1.Traditional and/or modern medicine The respondents seem to choose traditional and/or modern medicine depending on the type of disease they are suffering from, among other considerations. 2. Residents and malaria The respondents understood that malaria is carried by mosquitoes, but the mechanism by which it occurs was not widely known. 3. A sense of values and view of health Religious expressions were seen in answers to the questions regarding their knowledge of malaria. In addition, the residents have great desire to maintain their health, and they often found value in health. Conclusion : Medical anthropology is a tool to understand the human being. It facilitates the understanding of society, the environment and how they interact. Therefore, it allows us to recognize our shared values with people who live in target countries. We think that the medical anthropological perspective in international health and medical cooperation is a valuable approach to learn more about the communities in which we work.

P2-21) ONCHOCERCIASIS AS METAPHOR : A MEDICAL ANTHROPOLOGICAL APPROACH TOWARD ILLNESS CONCEPT OF JAPANESE-MEXICANS IN CHIAPAS, MEXICO

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ABSTRACT

This research was conducted to clarify the concept and metaphor of Onchocerciasis (river blindness) among the Japanese-Mexicans in Chiapas. Due to ecological habitat segregation, health workers reported no Onchocerciasis cases amongst the Japanese-Mexican community. Therefore, the narratives of 'Onchocerciasis' given by Japanese-Mexicans were most likely based on inaccurate rumors, due to their limited experience and knowledge of the illness.

Through careful interview data analysis, the following points were derived; Firstly, the Japanese-Mexicans' concept of Onchocerciasis was that it was an illness of the 'others', who were outside of their community, particularly 'poor Mexicans and Guatemalans'. Secondly, the metaphor of Onchocerciasis reflects images of poverty and misery, and was used to stigmatize or scapegoat those whom they wished to target.

P2-22) THE CHARACTERISTIC BALANCE OF CULTURE, NORM AND HE BALANCE OF MENTAL HEALTH IN THE SUBTROPICAL ZONE

HITOMI SAEKI

Human Care Associaton

The life in the subtropical zone is different from ours in the industrialized countries. It seems to be important to know what we can learn from their values on human lives.

To explore the people's perception on the value of life and death in the subtropical zone, I did fieldwork in Okinawa (Japan), Calcutta (India), Dhaka (Bangladesh), and in Pompey (Micronesia).

The data collection was done by the participant observation and interview.

As a result, I found the people in the subtropical zone appreciate the indigenous culture.

The people in Pompey are proud of their roots and put high value on human bonds.

However, in Calcutta and Dhaka, where moderni-

zation has affected the urban life, the residents show much more favor on the modern technology.

The people in the subtropical zone also regard death as a God's will and naturally accept it. They also revere spiritual aspects of life. All the new-born lives are blessed.

They believe in the Great Power of God(s). They seem to be mentally healthy as they have deep senses of forgiveness in human life.

In conclusion, we should learn such values of the people in the subtropical zone to make our lives more balanced in the modern society.

P2-23) STUDY ON SEXUAL CONSCIOUSNESS AND SEXUAL BEHAVIOR OF ADOLESCENTS IN JAPAN -PROBLEM IDENTIFICATION AND A STEP TOWARD CHALLENGE-

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In Japan, sexual behavior among the younger generation has been changing, with a shift in the age at first sexual intercourse to the early teen years. The abortion rate, as well as the incidence of sexually transmitted infections (STI) among of teenagers (15-19 years olds), are increasing. A KAP (Knowledge, attitudes, practices) study on sexual consciousness and behavior conducted in one prefecture in 2002, which surveyed 3,056 high school students, showed similar results to the national study reported above.

The abortion rate among the younger generation is rising in Japan, while the contraception use rate remains low. This is in contrast to other countries, such as the Netherlands, where the abortion rate is decreasing while the use of contraception is high. One important difference between these two countries is that in the Netherlands, the promotion of sex education in the home, school and community has been well developed, while it has not yet been adequately developed in Japan.

In our study, we investigated how midwives, nurses, and nursing students consider the above-mentioned situation. As a first step, a discussion was conducted among the target population. Our preliminary results show that the younger generation is struggling to find their own way that is acceptable to them.

P2-24) DOCUMENTAION ON BEST PRACTICES OF ENGENDERING COMMUNITY SUPPORT FOR ADOLESCENT SEXUAL & REPRODUCTIVE HEALTH PROGRAMS IN ASIA

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Objectives

The recent rapid increase in unwanted pregnancies and HIV/AIDS/STIs amongst adolescents affects one sixth of the world's population. JOICFP believes the lack of information and services for adolescent sexual reproductive health (ASRH) is due to adults not recognizing these needs, and that gaining adult understanding and support is essential for implementing ASRH programs. JOICFP has made a documentary on the best practices for engendering community support for ASRH programs, supported by the United Nations Population Fund (UN-FPA) and International Planned Parenthood Federation (IPPF).

Procedure

Considering religious, socio-cultural and geographical representation, six cases out of 20 were documented : Cambodia : Sewing a Healthy Future, India : A Life Blooms, Indonesia : Hanging Out, Islamic Republic of Iran : A Way of Life, Japan : Adults Who Care, and Mongolia : Dual Protection. They were chosen, based on their own reports, from surveys sent to twelve Asian

countries.

Strategies common among the six cases

1. Establishing a multi-sectoral network including GO, NGO, health and education sectors, mass media, research institutions, police and community groups

2. Simultaneously working on community leaders, guardians, schools and business managers

3. Targeting the inclusion of religious leaders

4. Extending policy and legislative advocacy

Lessons Learned

Program implementers need to

1. Apply both formal and informal approaches to maintain links with other organizations.

2. Cultivate contacts with gatekeepers, with personal contacts concerned.

3. Advocate for favorable laws and policies.

Findings

In responding to the needs of youth, activities that help gain the adults' understanding and support are of utmost importance.

P2-25) THE DIFFERENCE IN PERINATAL MEDICINE BETWEEN PUBLIC HOSPITALS AND PUSKESMAS IN INDONESIA

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In this study, we aimed to identify the roles of 'puskesmas' and public hospitals in maternal and child health care in Indonesia. We carried out this study in two phases.

First, from May to December in 2002, we collected

data through literature review and interview with Dr. Danawati. Next, in March 2003, we visited two public hospitals and one puskesmas and then compared their perinatal care activities.

As a result, we found the following differences be-

tween two institutions.

1. In puskesmas, where poorly equipped with medical instruments, midwives and nurses conduct normal deliveries and call medical doctors only when delivery-related emergencies take place.

2. In the public hospitals, where many medical equipments are available, medical doctors take care of pregnant women and their babies through regular check -

ups as well as conducting delivery services.

When emergency care is needed for a pregnant women or a baby, puskesmas staff members are expected to refer them to a public hospital. However, while we were visiting Indonesia, we found such a referral system was not properly functioning. In order to reduce the high mortality rates of pregnant women and infants, the referral system should be much more strengthened.

P2-26) PRENATAL CARE VISITS AND INFANT'S WEIGHT AT BIRTH AMONG ETHNIC MINORITY MOTHERS IN MOUNTAINOUS BAC KAN PROVINCE, VIETNAM

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Prenatal care is considered a mean to reduce the risks of low birthweight (LBW). The aim of this study was to investigate association between frequency of prenatal care visit as well as timing of first prenatal care visit and infant's weight at birth among ethnic minority mothers in mountainous Backan province of Vietnam. Study infants were recruited over a one-year period beginning August 2001 to December 2002 in this cross-sectional study. The questionnaire about their frequency and timing of prenatal care visits was administered to 64 ethnic minority women with a singleton birth. Of these, thirty-two normal birthweight (NBW) infants were selected as controls. Each LBW infant was individually matched to the next an eligible control by sex and mother's age. The mean number of prenatal care visits was 3.4 ± 1 for mothers of NBW infants and 2.8 ± 0.9 for LBW infants (p=0.02). Mothers of NBW infants initiated their prenatal care visit at 13.1 ± 5.7 weeks before their infants were born, earlier than mothers of LBW infants group at 14 ± 5.7 weeks. Frequencies of prenatal care visit were probably positive associated with infant's weight at birth. Although initial prenatal care visit varied by 1 week between two groups, the difference was not found to be significant.

P2-27) PREVALENCE AND ASSOCIATED FACTORS OF REPRODUCTIVE TRACT INFECTIONS AMONG PREGNANT WOMEN IN NGHE AN PROVINCE, VIETNAM

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This study investigated the prevalence and associated factors of seven reproductive tract infections (RTIs) among pregnant women in Nghe An Province. The pilot study took place at two sites. Twenty-nine of 30 antenatal care attendants at the MCH/FP center, and 17 of 24 pregnant women registered at the commune health center and 9 non-registered pregnant women in one commune participated. The participation rate in the pilot study showed the feasibility of the survey. In the main study, 373 (86%) of 435 registered pregnant women and 183 non-registered women in 10 communes joined the survey. The overall RTI prevalence was 36%, with a wide regional variation. The most prevalent infection was *Candida* (17%) followed by hepatitis B (10%). Sexually transmitted diseases were rare; 7 cases of *Trichomonas* and no gonorrhea or syphilis were found. The prevalence of bacterial vaginosis

and group B *Streptococcus* was 7% and 4%, respectively. Three factors decreased the risk of infection : use of tap water for genital washing, condom use, and higher financial status. Compared with laboratory diagnosis, women's self-reported abnormal discharge had a sensitivity of 93% and specificity of 11%, while physicians' finding of vaginitis had a sensitivity of 54% and a specificity of

55%. The main study suggests first, that RTI prevention should consider regional differences and financial status. Secondly, health education is needed regarding sanitation, condom use and basic knowledge about RTI. Thirdly, RTI diagnosis should be based on laboratory tests or at least speculum examination rather than self-reported symptoms alone.

P2-28) AN ANALYSIS OF TRENDS IN THE ANNUAL NUMBER OF ARTICLES ON THE PATENT PROBLEMS OF HIV DRUGS AND THEIR BACKGROUND

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Recently, much attention have been paid to the patent problems of HIV drugs, the situation of which is rapidly changing with price reductions in patent drugs, renouncement of the patent by major pharmaceutical companies and production of generic drugs by developing nations. In the present study, we analyzed the trends in the annual number of articles on the patent problems of HIV drugs and investigated their background. The annual number of articles on the patent problem of HIV drugs was surveyed from 1998 to 2002 with Asahi newspaper database. For an investigation on the background of changes in their trends, search engines and other web sites were referred. The results show that the annual number soared to 9 in 2000 with a 200% yearly increase after recording 4 and 3, in 1998 and 1999, respectively, In 2001, it surged to 20 with a 100% annual growth. Hence, some factors of importance are naturally assumed in the background of a rapid rise in the number from 1999 to 2000. We speculate that the demonstration against WTO meeting, or anti-globalization protest, in Seattle in 1999 is

one of the most important factors. After that, multinational companies, including major pharmaceutical manufacturers, found it necessary to consider a contribution to environmental protection and local society as well as to raise their profits. In May the next year, 5 major multinational pharmaceutical companies announced price reductions in their patent drugs for HIV. On the patent problems of HIV drugs, various conflicts of interests are observed, such as between 'developed and developing nations' or 'profits and ethics'. In WTO, the problems are still under controversy among developing nations with or without generic drug production and developed nations with or without the patent. On the other hand, some major drug companies, under pressure not only from NPO but also from international investors with a new but gradually prevailing investment criteria, or SRI (Social Responsible Investment), have stated their own business strategies with the social contribution. Earnings of them are sound currently.

P2-29) HIV PANDEMIC AND INVENTION OF NOBLE TRADITION

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JICA Tanzania Morogoro Health Project

An existential world has been articulated and manipulated by particular agents and institutions by the way of which favorable knowledge has been elaborately produced and reproduced. However, despite of an aggregation of the discursive knowledge of HIV/AIDS pandemic, a traditional healer in Kilombero district, Tanzania, strives to mediate social sufferings into a new mode of knowledge of "tradition" through his healing rituals and herbal practices. Here, the deterministic knowledge of HIV/AIDS in a sense of biomedical reductionism has been invented and transformed into their practical and substantial knowledge of "ukimwi" by articulating meaningful live and society in the reality. The encounter with the external knowledge of HIV/AIDS has been dealt by a revitalization of traditional icons and a reconstitution of the unity between body and mind, which was distorted by the pandemic. Generally this innovative aspect of knowledge transformation represents the strategic knowledge system, where people can manipulate an intruded truth

into practical and mindful knowledge in a sense of individual satisfaction and communal fruition. Indeed, knowledge transforms the society and, at once, the society transforms the knowledge. In the mundane reality of Tanzania, the knowledge of the pandemic magnifies the discursive gap between the centre and the peripheral, where the power and the control of the knowledge are unevenly distributed. The autonomous knowledge innovation through enterprising efforts by the traditional healer demonstrates well a process of signification and appropriation of the substantial knowledge in the era of pandemic and marginalization.

P2-30) A PILOT OF VCT AND THE PREVENTION OF MOTHER-TO-CHILD TRANSMISSION OF HIV IN A PRIMARY HEALTH CENTER AT BAMAKO, MALI

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Objective :

Our study aims were ; 1) to assess the feasibility of VCT (Voluntary Counseling & Testing) and prevention of mother-to-child transmission of HIV (PMTCT) at a community - based primary health center. 2) To explore the factors associated with HIV infection among pregnant women.

Method :

A woman doctor and 2 midwives at a communitybased primary health center at Bamako proposed VCT and a questionnaire to pregnant women after 36 gestational weeks from May to November 2001. HIV testing, ARV (Nevirapine) for PMTCT and formula milk were provided free of charge when accepted.

Result :

Out of 362 pregnant women, 343 (94%) accepted HIV testing, and 20(5.8%) were found HIV seropositive, much higher than official statistics (2.7%). All 20 women

and their newborn took a single dose of NVP at home or in the clinic. Twelve out of 20 chose not to breastfeed. One mother and 3 newborns died during the study period. High education level and the age of first sex were positively associated with positive HIV (p=0.05, p=0.04). Condom use, polygamy and female genital mutilation were not significantly associated. Having one's own earning and the number of abortion were positively associated with condom use (p<0.05, p<0.01).

Discussion:

It seems feasible to introduce VCT into antenatal care at urban community-based primary health centers, and to perform PMTCT using NVP. The advantage of this approach was high acceptance of VCT. The disadvantage was insufficient supports for post-test counseling to sero negatives, safe formula feeding and follow-up of mother and child.

P2-31) THE SITUATION OF VCT SERVICES IN AFRICA AND FUTURE COOPERATION STRATEGIES THROUGH JAPANESE ODA

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In Africa, the situation of HIV/AIDS is getting worse. To improve it, a voluntary counseling and testing (VCT) service has been recommended.

In this study, we aimed to analyze the situation of VCT service in Africa. We visited six African countries, Uganda, Ghana, South Africa, Zambia, Kenia, and Tanzania where VCT-related data were known to be available. We collected qualitative data through key informant interviews, secondary literature survey, and observation in these countries.

As a result, we found that several different NGOs were implementing VCT services in all the countries. Some VCT services were also expanding. To improve the

quality of VCT services, we identified the following needs. First, VCT service sites should be identified and expanded, and then HIV test kits should be constantly given to the service providers. Second, each government should issue a guideline on VCT, carry out advocacy activity for VCT service promotion, strengthen management capacity for VCT services, and finally improve monitoring and evaluation system.

To meet such needs, Japan should combine the past HIV/AIDS-related activities with VCT services and then make a more comprehensive approach in these African countries.

P2-32) MAOIST INSURGENTS AND STI EPIDEMIC IN NEPAL : IMPLICATION OF THE SERO-SURVEILLANCE

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Introduction : After the Maoist-attacks in November 2001, the government of Nepal mobilized its security forces throughout the country. One of the biggest army camps in Far Western Region is located in Doti district and coincidently this is the field where the authors conducted an epidemiological survey of HIV and sexually transmitted infections (STIs).

Methods: In April 2001, blood from 149 men (15 to 45 years old) was tested for HIV and Syphilis in Doti. Based on the results, a simulation model (inc. mapping) to project the prevalence has been created.

Results : Of the 149 subjects (All males, mean age=27 years, SD= \pm 7.7) 11 (7%) tested positive for HIV infection and 38 (26%) for syphilis. Over two years, we predict that out of 2000 soldiers, 32 new HIV cases will

occur. If Voluntary Testing and Counselling are not available, after 10 years, HIV cases would number more than ten thousands.

Discussion : Several studies indicated promiscuous behaviour among soldiers and STI transmissions. The authors believe that the reported low prevalence of HIV/STIs in Kathmandu may give impressions to urban residents that rural area is much safer in terms of transmission of these diseases. Consequently, deployed soldiers may be less likely to use condoms in their sexual encounters in Doti. Considering the high STI prevalence in Doti, and soldiers as mediators of HIV/STIs transition, we are concerned that an outbreak of HIV infection will occur soon unless appropriate measures are taken.

P2-33) CARETAKER'S KNOWLEDGE ON ANTIRETROVIRAL DRUGS PRESCRIBED FOR CHILDREN UNDER 14 YEARS

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To improve the effectiveness of antiretroviral (ARV) treatment for HIV/AIDS, the patients' adherence to the drugs is critical. However, improving such adherence is not easy. Though several studies were done among adults, few studies were conducted for the children's adherence. In this study, we aimed to investigate the caretakers' knowledge about food limitation while the children were taking ARVs.

We conducted a cross-sectional study at Sao Paulo State Center of Reference and Training for HIV/AIDS from December 2002 to March 2003. The participants were the caretakers of the HIV positive children under 14 years old. The trained interviewers interviewed 47 caretakers using the structured questionnaire. We also used the medical records of the children, which included the type of treatment and CD4 cell count.

We found that 37 out of 47 caretakers were expected to know about the food limitation ; of 37, 60 % had little knowledge about it. Still, most of them believed that they knew about it. The caretakers indeed knew about the drug dose as well as when to take them, but not so much about food limitation. In addition, the caretakers, who were also under the ARV treatment, understood the children's treatment methods much better than theirs

The results revealed dissociation between the caretakers' perception about the correct use of the ARVs and their practices. The health care providers should be more aware of what the caretakers should know and practice for ARV treatment. Then they should develop innovative methods to improve the caretakers' knowledge.

P2-34) ANALYSIS OF THE TIME TREND OF HIV/AIDS PREVALENCE AND SEX RATIO

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The purpose of this study was to examine the usefulness of the sex ratio of the number of HIV/AIDS cases as a substitute for the sex ratio of case prevalence. Both ratios are calculated annually at the national, zone, and provincial levels. In addition, both sex ratios are calculated for each 5-year age group within the Chiangrai population. Results of the time series data analysis from 1993 to 2001 at the national level showed similarities between both sex ratios. The case number sex ratio (male : female) was 7.3 and, and the case prevalence sex ratio was 7.3 in 1993. These values fell to 1.9 and 2.0, respectively in 2001. Data at the zone level also showed similarities between the sex ratios. The ratios of the 12 zones ranged from 5-17 in 1993, and then fell to 2-3 in 2001. The results of the time series data from 1989 to 1999 in Chiangrai showed similar results. The sex ratios were 1.0 and 1.0 in 1989, and went up to 9.5 and 9.4 in 1991, and then fell to 1.4 and 1.4 in 1999. The age group data from 1994 to 1999 followed the same trend. The results showed similarities between both sex ratios at each level and in each age group in Chiangrai. The sex ratio of the number of cases can thus be used as a substitute for
the sex ratio of case prevalence.

P2-35) THE 10 YEARS FIELD EXPERIENCES OF INTERNATIONAL COLLABORATIVE RESEARCH ON TB AND HIV/AIDS IN CHIANG RAI, THAILAND

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The TB/HIV Research Project under RIT was established in 1995 based on a long history of cooperation since 1963, after Thai doctors returned from the first international TB training at RIT. The Project is working collaboratively with various institutions in Thailand. The mission of the Project is to conduct research and related activities such as human resource development and technical cooperation on Tuberculosis and HIV/AIDS. Our goal is to provide a scientific basis for the control strategies of these diseases, not only in both countries but also internationally. There were six major research areas : 1) Population-based longitudinal study of TB patients and their contacts ; 2) Research and prevention of nosocomial Mycobacterium tuberculosis transmission ; 3) The socio cultural dimensions of TB in HIV/AIDS Epidemic Areas ; 4) TB screening and prevention in a cohort of people living with HIV registered at day care centers; 5) Cohort study of drug users in Mae Chan Hospital; and 6) Research and development of blood bank system. The results were fed back to the control program by various means including 30 publications by 2002. In addition, the Project coordinated Thai health personnel to be trained at RIT. The "TB/HIV Research Fellow" program was set up to collaborate with Thai universities to support their graduate students in the field research. The Project organized the annual provincial TB meeting, Knowing TBessential knowledge for the general public in HIV epidemic area" which was published. A project member is a core member of the TB/HIV working group of WHO.

P2-36) COMPLIANCE AMONG HIV POSITIVE PATIENTS AT A RURAL HOSPITAL IN THAILAND

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This study aimed at describing the use of medical services among HIV/AIDS patients and examining the factors associated with continuity in receiving medical services. Subjects were the adult HIV/AIDS patients who lived in Khon Kaen and visited the outpatient department of Northeast Hospital (Srindhorn General Hospital)from 1st April to 30th September 2002. Dates of visits, sex, age, healthinsurance, HIV stage, and use of antiretroviral

drugs (ARV) werecollected from the medical charts. Among those who visited the hospital from April to July, the patients who continued to visit the hospital from August to September were classified as the continuing group and those who did not do so were classified as the discontinued group. From April to July, 340 HIV/AIDS patients visited the hospital. There were 145 patients in the continuing group and 195 patients in the discontinued group. Logistic regression analysis indicated that HIV stage (OR 1.9, symptomatic vs asymptomatic) and use of ARV (OR 2.0, use vs. no use) were significantly associated with the continuing group. It is possible that some of the patients discontinued their visits simply because they changed hospitals, were hospitalized, or died. Further study is necessary to follow-up with the patients who discontinued

visiting the hospital in order to find out the reasons whythey did so, and to investigate whether they were able to receive appropriate medical services. This research was supported by a Grant for Health Cooperation Research (13-C3) from the Ministry of Health, Labor and Welfare of Japan.

P2-37) MOLECULAR EPIDEMIOLOGICAL INVESTIGATION OF HIV/AIDS EPIDEMIC IN YUNNAN PROVINCE OF CHINA : IMPLICATION FOR THE GENESIS OF THE EXPANDING EPIDEMIC IN CHINA

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Objectives : To investigate the molecular epidemiology of HIV in Yunnan Province, an epicenter of the expanding AIDS epidemic in China, where the initial HIV-1 outbreak among injecting drug users (IDUs) occurred in 1989, and to analyze the genesis and the interrelationship of the epidemic with that in other areas in China.

Design : A molecular epidemiological investigation was conducted among IDUs in three Prefectures in Yunnan Province, including Wenshan (east) and Honghe (southeast), and Dehong (west) to elucidate the mechanism of the expanding AIDS epidemic in China.

Methods : Thirty nine specimens were collected from consenting IDUs in 2000-2001. The nucleotide sequences of 2.6-kb gag-RT and 340-bp env (C2/V3) regions were determined. Phylogenetic tree and recombination breakpoint analyses were performed to elucidate the detailed recombinant structure of prevalent strains.

Results : The circulating recombinant form (CRF), CRF08_BC, predominated in east Yunnan near Guangxi Province (89% in Wenshan and 81% in Honghe), while it was not detected in Dehong (0 of 14) in the west. In contrast, 71% (10 of 14) of the Dehong isolates were unique recombinant forms (URFs), mostly between subtypes B' (Thailand variant of subtype B) and C, with distinct profiles of recombination breakpoints. The subtype B' accounts for the remaining 29% (4 of 14) of Dehong isolates. The detailed recombination breakpoint analyses revealed that two Honghe isolates (2 of 16) were new class of recombinants between CRF07 BC and CRF08 BC.

Conclusion : New recombinant strains are arising continually in western part of Yunnan Province near Myanmar border. Some appeared to be the second generation recombinants comprised of previously established CRFs in China. The uneven distribution of subtypes, CRFs and URFs suggests the presence of independent transmission networks and clusters among IDUs in Yunnan. The findings in the present study would provide new insights into the genesis of expanding AIDS epidemic in China.

P2-38) EPIDEMIOLOGICAL ANALYSIS OF THE SUBTYPE OF HIV-1 IN CHINA

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Objective : Our research are try to reveal the epi-

demic modes of HIV in China, and acquire the general

information of the subtypes of HIV-1 prevalence including location, transmission pattern in China. Methods : This study was based on existing sets of HIV-1 nucleic acid sequences of China. All the sequence data are publicly available from the HIV sequence database at the Los Alamos National Laboratory We also reviewed the medical literature and published documents from the Ministry of health P.R. China and from the sentinel surveillance site. In this study, phylogenetic analyses were carried out using the neighbor-joining algorithm, based on a genetic distance matrix calculated using the Kimura two parameter method. Results : The cumulative reported of people with HIV/AIDS reached 40, 560, and HIV transmission is focused primarily among IDUs . Subtype B is the most common strain in China. Subtype B/C is now responsible for most infections among IDUs. B/C is most common in Kunming, and AE is most common in guangxi. The most likely explanation for the finding is the different epidemiologic patterns

P2-39) FIVE S FOR QUALITY MANAGEMENT ACTIVITY IN SRI LANKA

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Introduction

Health indicators of Sri Lanka are better than any other developing countries because of its higher standard of education and primary health care system. The alumni of the trainees who were trained in Japanese industry introduced the quality control activities. Five "S", Seiri, Seiton, Seiketsu, Seisou and Shituke, are well-known and utilized in the public sectors as well as industries in Sri Lanka society.

Cases

1) Electric factory : The factory was run by Sri Lanka owner and is producing antenna, cable, semiconductors for the Japanese or US market. The number of the employees was about 500. Five S and "Kaizen" are shown in walls of the production line. The environment of the factory was managed nicely.

2) Castle Street Maternity hospital : This hospital is

a governmental hospital and awarded by the Asian Productivity Organization (APO) on the good quality management in the public sector. Management of hospital assets and waste, prevention of nascomial infection, improve quality of medical care were extensively done on the concept of 5S and "Kaizen".

Assessment and Conclusion

Quality control activity in Sri Lanka is much common than in Japan although it is initiated in Japan. Participatory approach in the community health care will be applicable in Sri Lanka society. TQM (Total Quality Management) in Japanese medical care was introduced from the management of Japanese industry to prevent the medical incidents or control nasocomial infection. Their effort teach Japanese medical personnel to enhance the quality improve management in medical systems.

P2-40) DEVELOPMENT AND EVALUATION OF A TRAINING PROGRAM FOR ORAL HEALTH WORKERS IN NEPAL

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Since 1994, we have conducted the training for oral health workers in Nepal. In 2001, the graduates and counterpart members established the Committee of Oral Health Workers (COHW) and we extended the training programs to different areas for a year. In 2002, we assessed their knowledge about oral health before and after the training. A total of 39 participants in basic and advanced courses were evaluated. Moreover the advanced course at on-going and new project sites was compared. Although participants in the basic course had less knowledge about oral health, they were aware of the importance of oral health education. After the training, their knowledge had improved. Compared to the participants in the basic course, the participants in the advanced course had more knowledge and understanding. Among the participants in the advanced course, those from the new project site had less knowledge compared to those from the on going project site. In sum, it is necessary to continue the training in order to maintain a certain level of knowledge and skills among oral health workers. We therefore expect that the COHW can play an important role in continuing and expanding the project.

P2-41) HEALTH EDUCATION IN TEN MINITUES WITH TRIGER TALE OF TRAINER'S EXPERIENCE - TRAINING OF TRAINER IN EAST TIMOR

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Background : An obstacle to health education is the fact that most people are busy and are unable to concentrate on listening to long explanations. Therefore, simple and short health education should be developed to help people understand the learning facilitators' message easily, and help them make positive changes in their health behavior.

Activities : Within the SHARE (a Japanese NGO)'s Health Project in the East Timor, a Trial of Trainers' Training has been implemented using;

1. a micro-teaching method, which starts with (1) a trigger phrase, trigger tale or trigger drama as an introduction to get people's attention, (2) a clear objective, (3) a main message, (4) an assessment to check people's understanding and (5) a summary. This whole process takes only 10-15 minutes. 2. active listening and silent listening skills to encourage people to talk about their understanding of the material that was presented.

3. facilitation skills rather than mere teaching skills to make a comfortable learning environment. Even within 10 minutes, the personal experiences of the learning facilitators attract people to the health message, a first step in taking action to improve their own health.

Result : At the beginning, 'health educators' tended to talk for longer than 30 minutes. After learning microteaching procedures and practicing several times, all trainees learned the skill of the 10 minute attractive talk. Although this kind of talk takes more time to prepare, it is more effective than a longer talk. This method can be utilized in other places as well.

P2-42) HOW COULD LOCAL STAFF BE ASSISTED TO GRASP THE HEALTH PROBLEMS OF RESIDENT PEOPLE AND BE EMPOWERED TO ADDRESS THEM? -FROM A COMMUNITY SURVEY ON HEALTH, ERMERA DISTRICT, EAST TIMOR-

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SHARE : Services for the Health in Asian & African Regions

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Introduction The Health Education Promotion Project in East Timor has been carried out by SHARE since April 2002. A health professional dispatched in 2002 conducted a community health survey, and the process were shared with local staff. This session will explore how the PRA approach helped local staff become further motivated and gain confidence in their work. Objectives of the Survey1. To reveal health status, knowledge, and perception of prevention in the community.2. To use the data to devise training programs for health education promotors.Method1. Area: 15 villages selected from 5 sub-districts.2. Sample : n=473 (Both sexes, between 13 and 50 years.)3. Method : Home visit, direct interviews.4. Questionnaire: The survey consisted of the following four categories : Health Status, Prevention, Health Education, and Life Condition. The results were categorized

into Knowledge, Attitude, and Perception, and analyzed according to Age, Sex, and Life Condition. Major Findings Most respondents did not know how to prevent diseases, and did not think that they could prevent diseases themselves. They believed that prevention required doctors, nurses, and medicine. Seventy-seven percent believed that curative methods are better than prevention. Conclusion In addition to attaining our objectives, local staff gained greater awareness of peoples' health problems and of their views of prevention, allowing them to provide better health education. They gained confidence by participating in the planning, execution, analysis, and presentation of the survey. Their progress was displayed at training of trainers (TOT) of health staff in Ermera held in August 2003.

P2-43) TRIAL CASE OF THE DEVELOPMENT OF MEDICAL IN-SERVICE TRAINING SYSTEM IN GHANA

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In-service training (IST) is defined as "a training for health workers currently in service that enables them to gain updated professional knowledge, skills and morals necessary for providing local populations with better quality health services". IST is one of the most effective means of updating the training of health workers and maintaining the quality of health services. However, for various reasons, ISTs in Ghana have not produced the expected results, despite tremendous investment. In order to improve this situation, JICA and the Ministry of Health/Ghana launched the development of the IST system, which consists of four system components : 1) IST official classification ; 2) IST information system ; 3) IST logbook ; and 4) IST monitoring system. At the end of the project, these have been well developed, and contributing to the improvement of IST status in the health sector. It should be emphasized that this is the first case of the development of an IST system in Africa. Thus, some other African countries are interested in this system. Observation trips have been made by other countries' government officials, and one-day workshops have been held in Senegal, Kenya and Tanzania. Other ministries in Ghana which regularly conduct ISTs, e.g. the Ministry of Education, also expressed interest in the introduction of this system. Our Project focuses on the development of essential systems in the health sector in Ghana, and its impact is expanding to various areas. This system development approach could be an appropriate model of JICA's future cooperation with other developing countries.

P2-44) DELAY AND RELATED FACTORS OF FOREIGN PATIENTS WHO VISITED AN EMERGENCY ROOM

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Objectives : To describe characteristics of foreign patients who visited an emergency room, and explore their delay in seeking medical help. Methods : The study was conducted at a hospital in a local city in Mie. From March to September 2002, 9497 people visited the ER, and of these, 307 were of foreign nationality. Information on diagnosis and delay were collected from medical and nursing records. Results : Of the 307, 149(48.5%) were male and 158(51.5%) were female, most were South Americans. Age distribution was bimodal, with one peak at under 5 years old and another at 20-30 years old. Reasons for visiting the ER included : injury (27.0%), respiratory diseases (26.4%), and digestive diseases (13.4%). Males were more likely to suffer from injuries (72%), with traffic and industrial accidents most common. Under - five children with upper respiratory infections and

asthma were dominant in respiratory diseases (43%). Digenstive diseases, including gastritis and enteritis, were more common in females (56%). The delays before seeking treatment ranged from : less than 24 hrs(44.0%), 24 -48 hrs(20.2%), 48-72 hrs(8.1%) and more than 72 hrs(17.6%). Of the 54 who delayed more than 72 hrs, most had respiratory diseases. Among the actions taken during the "wait-and-see" period, self-medication was most common. Some delayed more than one month with serious diseases such as pneumonia, pyelonephritis, and heart problems. **Conclusion** : One in six foreign emergency patients delayed more than 72 hrs before visiting the ER, including some with serious health problems. This indicates a need for better medical service access for foreign patients.

P2-45) STUDY ON MATERNAL AND CHILD HEALTH SERVICE UTILIZATION AMONG LATIN AMERICAN WOMEN LIVING IN JAPAN WITH FOCUS ON PREGNANCY AND CHILDBIRTH

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We investigated maternal and child health service utilization among Latin American women living in Japan who have ever become pregnant and given a birth in Japan, using a Spanish and Portuguese version of a selfadministered questionnaire. There were 43 respondents. We asked them the frequency of taking a maternal health checkup, whether they wish to give a birth again at the same hospital in the future, and the attitude of health personnel toward the respondents throughout their contacts for pregnancy and childbirth. The attitude of doctors and nurses at the health checkup was not associated with the frequency of taking health checkup. However, their wish to give a birth again at the same hospital in the future was related to the attitude of nurses at the health checkup, the attitude of doctors at delivery, attitude of nurse, and the attitude of doctor at childbed. Employment, language proficiency, time in Japan and delivery costs were not associated with both the frequency of taking health checkup and their wish of delivery at the same hospital. The attitude of health personnel does not hinder their health service utilization but it is unknown whether they understand their explanation. Yet, the language is less likely to be a matter for the satisfaction of Latin American women with health services but the attitude of the health personnel toward them.

P2-46) ORAL HEALTH CHARACTERISTICS OF NEWCOMERS IN JAPAN

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We studied differences in oral health among 1007 newcomers who were examined at sixteen health check ups for immigrants in Tokyo, Chiba, and Saitama, between 1998 and 2002. Filipino's oral health conditions were the worst, with an average of 2.4 decayed teeth, 7.4 missing teeth, and 2.8 filled teeth per person, even though they brush their teeth an average of 2.7 times a day, and 35% use mouth rinse and 15% use dental floss. Few had crown prostheses and 37.4% had attached dentures. In the Philippines, it is common to extract teeth and make dentures to treat caries that affect the dental pulp. In Japan, however, extraction is the last resort, while a root canal followed by a metal crown or bridge prosthesis is preferred. This difference may cause some Filipinos to distrust Japanese dentistry. Migrants from Myanmar had few decayed teeth, just 1.3 per person, although 60.0% of them had never had dental treatment, oral health education or tooth brushing instruction. The top reason for not seeking dental care is unwillingness to pay for expensive treatments. The other main reasons given were as follows : difficulty in taking time from work to visit a clinic, difficulty in obtaining information about dental clinics willing to treat foreign patients, and lack of understanding of the Japanese health insurance system. It is important to provide oral health education that addresses the characteristic dental conditions of people from each foreign country, and to give immigrants information that will help them gain access to dental clinics.

P2-47) HEALTH PROBLEMS AND STATUS OF HOMELESS PEOPLE LIVING IN TAITO-KU SANYA-AREA

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Abstract Due to the economic recession in Japan, an increasing number of people have lost their jobs, incomes and homes. This situation forces some of them to live on the streets. Most of the homeless people are excluded from the social security system, such as the national health insurance plans. Many are at risk for health problems. We carried out this study to learn more about the health situation of the homeless people in SANYA-Area in Tokyo, and to explore possible interventions to solve their health problems. We carried out interviews with 110 homeless men, (average age : 65.2 years old). We found that 85.5% of the interviewees had some health complaints. The most common complaint was untreated hy-

pertension, followed by joint/back pain. The major causes of these health problems were the unstable food supply, insufficient rest, and fatigue due to carrying their heavy belongings. To solve these problems, it is necessary to find ways to stabilize the lives of homeless people by helping them to find employment, or by giving them access to public livelihood assistance. It is also vital to have well coordinated health programs. These programs should include health information, which would enable homeless people to understand the severity of their health conditions, and help them plan strategies to overcome their health problems.

P2-48) HEALTH SITUATION OF HOMELESS PEOPLE IN SHINJUKU, TOKYO

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There are over 25,000 homeless people living in Japan, and their health is in a critical state. Sinjuku - renrakukai is an organization that supports the health of homeless people in Shinjuku, Tokyo. This organization has been providing monthly health consultations since 1996.This study was conducted by analyzing data from health consultations performed between January and December 2002. During this time, 369 homeless people, including 12 women, sought health consultation. For this report, we classified them into 3 age groups : less than 50 y.o. (110 homeless people), fifties (160), or 60 or over (99). Ninety-three homeless people had utilized hospital care after their consultations. The age distribution of those who were hospitalized was 20 in the less than 50 group, 44 in the fifties group, and 29 in the 60 or over group. They suffered from a variety of diseases, including hypertension, tuberculosis, phlegm on, and gastric ulcers. The characteristic diseases in each age group were as follows. Hypertension was seen in a large fraction (31%) of the 60 or over group. Gastric ulcers were seen in 11.3% of the fifties group. In each case, the incidences of these two diseases were remarkably high, relative to the other two age groups. Tuberculosis was also common, occurring in 9.1% of the fifties group and 20.7% of those 60 or over. Many also suffered from previously diagnosed illnesses, including gastric ulcers, tuberculosis, hypertension, diabetes. In the fifties group, gastric ulcers, diabetes were common, relative to the other two age groups.

P2-49) CURRENT SITUATION OF LIFE-STYLE RELATED DISEASES AMONG JAPANESE EXPATRIATES IN KUALA LUMPUR, MALAYSIA

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Purpose : Japanese people living abroad long-term may contract diseases due to changes in their lifestyle. We have conducted research into the lifestyle-related diseases of senior Japanese people (more than or equal 40years) living in Malaysia, Kuala Lumpur.

Method : We conducted medical examinations locally via interview, body measurement, blood pressure measurement, urine and blood sugar examination. We also took blood samples and examined in Japan for liver function (GOT, GPT, γ GTP), lipid (Cholesterol, Triglycelide) and uric acid with "Postal Medical Examination", which is produced by Aichi Medical Foundation of Diagnostic.

Result : Our subjects were 63 Japanese people, 44 of whom were workers from local corporations, or unem-

ployed. From the result, alcohol consumption was around 70%, which is somewhat high. Those who exercised regularly comprised 46%, higher than domestic figures. Twenty-two of the subjects (34.9%) visited hospitals for treatment of lifestyle-related diseases. The results also showed an obesity rate of 27.4%, high compared to domestic result. The percentages of abnormal result in blood pressure (22.2%), lipid (37.0%), and blood sugar (11.7%) were also high compared to domestic figures.

Conclusion : Although many of the subjects visited hospitals for their lifestyle-related diseases, the ratios of their abnormal results in the examinations of the diseases were high. It is indicated that efforts at management of lifestyle-related diseases are still not sufficient in the group. Henceforth, we wish to focus our examinations on

P2-50) DEMAND SURVEY OF TYPHOID VACCINE IN JAPAN OVERSEAS HEALTH ADMINISTRATION CENTER

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Introduction

Vaccines for typhoid fever are readily available overseas, but are not permitted in Japan. Presently, people are importing vaccines themselves, and being vaccinated. Since the year 2000, the Japan Overseas Health Administration Center has been vaccinating those who wish to be vaccinated, using Typhim Vi vaccine.

Objective

Investigate the demand for typhoid fever vaccine, and its side effects.

Procedure

During the six month period from September 1, 2002 to March 31, 2003, we targeted outpatients who requested, through their doctors, typhoid fever vaccine from the Japan Overseas Health Administration Center. We calculated the straight-line distance from the Center to the homes of the people receiving typhoid fever vaccine. We randomly selected the same number of patients from a group receiving tetanus toxide, similarly calculated the distance, and compared the numbers. During the same time period, we also asked people to respond to a questionnaire regarding post-vaccination side effects.

Results

From September 1, 2002 to March 31, out of 448 outpatients who requested vaccination, 40 were vaccinated with the typhoid fever vaccine. Those who were vaccinated with tetanus toxide numbered 222. The average straight-line distance (Center to home) of those who requested typhoid fever vaccine to this center was 82.8km, while the average distance of the 40 people randomly sampled from the tetanus toxide group was 20.0km, a significant difference. (p<0.05)

Regarding side effects, we sent questionnaires to 15 people who received typhoid fever vaccine, and we received 11 responses. Out of these, 7 people complained of local pain, but there were no other symptoms such as fever and/or swelling.

Conclusion

Despite doubts regarding the necessity of typhoid fever vaccine, there is demand for vaccines in Japan. We have seen that people will travel great distances to receive this type of medical treatment. As for side effects, the main complaint is local pain. However, if the patient feels that vaccination is necessary, some pain is tolerable. It is our conclusion that the benefits of this vaccine far outweigh the drawbacks.

P2-51) CYSTIC HYDATIDOSIS - OCCURRENCE OF CASES IN JAPAN AND ITS PREVENTION

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Cystic hydatidosis (CH) in Japan was reviewed on

the basis of case reports and other references since 1881

to 2003. A total of 76 cases were identified as CH patients which 47 of them were reported prior to 1945 and 29 thereafter. Those cases were divided into 2 stages before and after the Slaughterhouse Act (SA; 1906). The endemic foci of CH was suggested before SA, especially in southwestern Japan. Development of cattle breeding and unsanitary small slaughterhouses were thought to be the cause of endemism. Japan hastened cattle breeding on behalf of dietary protein source and also various military accouterments for soldiers in the past. China had been a transit port to Japan from Europe and also a big endemic area of CH. Japan had sent a huge number of soldiers and citizens to China in Japanese-Sino War, Japanese-Russo

War and etc. A number of cattle, pet dogs and people have also came to Japan from China. After the enforcement of SA in 1906, the number of CH decreased dramatically, and only 2 cases were reported for 30 years between 1920 and 1950. These situations of CH suggest that there had been the endemic foci in southwestern Japan, though there is no direct evidence because no one had found E. granulosus from dogs in Japan. Recently almost all of CH patients are supposed to be infected in overseas endemic countries. These facts suggest strongly that the sanitary control of slaughterhouses is essential for preventing CH in endemic areas.

P2-52) CORRELATION BETWEEN THE RISK RECOGNITION BY OBSTETRICIANS ABOUT CONGENITAL TOXOPLASMOSIS AND THEIR CLINICAL ACTIVITIES

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Among four congenital infections, or toxoplasmosis, *rubella*, HCV and *Parvo* B19, attitudes for explanation and serum antibody tests to pregnant women, risk recognition for each disease, such as possibility of primary infection in pregnancy and number of cases experienced by gynecologists were compared.

No statistically significant difference was observed in both number of cases per clinician and percentage of clinicians who have experienced any cases among the four congenital infections. In risk recognition, *rubella* enjoyed the highest rank in four out of six factors, while toxoplasmosis was estimated lowest in four out of six. Toxoplasmosis also recorded lower risk estimates than *Parvo* B19 in five out of six. The result indicates that gynecologists in the area hold relatively lower risk recognition on toxoplasmosis.

In clinical activities, gynecologists showed as positive attitudes for toxoplasmosis as for *rubella*. On the other hand, their attitudes for toxoplasmosisi were more positive than for *Parvo* B19. The result indicates that gynecologists in the area hold relatively positive attitudes for explanation and serum antibody tests on toxoplasmosis in pregnancy.

As we previously reported, gynecologists in Obihiro area, Hokkaido, Japan, held relatively negative attitudes for clinical activities on congenital toxoplasmosis, probably backed by their lower risk estimates on the disease. Although some epidemiological evidence is in contradiction to their recognition, their risk estimates and clinical activities are in consistency. The present study indicates that gynecologists in Edogawa area have some discrepancy between their risk estimates and clinical activities, in other words, what they think and what they do. It is speculated that this discrepancy may reflect information gap between these two areas as well as difference in recognition about congenital infection by pregnant women. In order to establish EBM (Evidenced Based Medicine) for congenital infections, more comprehensive epidemiological studies are required.

P2-53) DNA DIAGNOSIS OF MALARIA USING MICROTITER-PLATE HYBRIDIZATION METHOD

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The conventional malaria diagnostic method, microscopic examination of Giemsa-stained blood films, is labor-intensive and requires highly trained microscopists for accurate identification especially when the number of parasites is very low. We have developed a simple diagnostic method "microtiter plate-hybridization" (MPH) for the purpose of detecting human malaria parasites by those without experience in identifying 4 species of human malaria parasites, or detecting low-grade parasitemia. In this method, the target DNA sequence of the 18S ribosomal RNA gene is amplified by polymerase chain reaction and hybridized with the species-specific probes immobilized on a microtiter well. The PCR products bound on a well are visualized by the biotin-streptavidin system and the following chromogenic reaction. Our MPH method could detect as few as 13 parasites in 10 microliters of whole

blood with high sensitivity and specificity including P. ovale-variant which we found during the course of this study. Here we report the usefulness of the MPH method with clinical blood samples obtained from patients with imported malaria in several hospitals of Japan between 1991 and 2003. We obtained blood samples by finger puncture from 344 asymptomatic donors. Among the 344 samples, 126 (36.6%) were P. falciparum-positive, 87 (25.3%) were P. vivax-positive, 16 (4.7%) were P. ovale positive, 6 (1.7%) were P. ovale-variant, 3 (0.9%) were P. malariae-positive and 106 (30.8%) cases were malaria negative. The results of our DNA diagnostic method showed good correlation with those microscopic test. This result sugget that Our MPH method is good tool for diagnosis of imported malaria in Japan and epidemiologic study of malaria infection in endemic areas.

P2-54) DETECTION OF *PLASMODIUM BERGHEI* BY ICT P.F./P.V. IMMUNOCHROMATOGRAPHIC TEST (2) : ANALYSIS OF THE DETECTION SENSITIVITY FOR THE PANMALARIAL ANTIGEN DURING THE INFECTIOUS COURSES WITH ANTIMALARIAL TREATMENT

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We evaluated the ICT Malaria P.f./P.v. immunochromatographic test for the detection of the panmalarial antigen (PMA) using a rodent malaria model. Mice were infected with *Plasmodium berghei* by mosquito bite, and blood was examined by microscopy and the ICT test. Artemether treatment was started when the parasite density reached over 70,000/microL. The ICT PMA band appeared when the parasite density reached more than 2,000/microL, but it continued to be 'positive' after the parasitemia became negative in response to the drug treatment. When all the test results were divided into increasing phase (IP) and declining phase (DP), the sensitivity in DP was significantly higher than that in IP, suggesting that the reactivity of the ICT PMA is significantly influenced by persistent and accumulated PMA after drug treatment and longer duration of infection in DP. Recognizing that the patient population in a clinical situation would be a mix of individuals in IP and DP, it should be emphasized that the individual history of recent fever, duration of illness and drug treatment must be considered carefully for the interpretation of the ICT results.

P2-55) RECOMBINANT BACULOVIRUS VIRIONS DISPLAYING CIRCUMSPOROZOITE PROTEIN PROTECT AGASINT PLASMODIMU BERGHEI SPOROZOITE INFECTION

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Recently, the baculovirus Autographa californica nuclear polyhedrosis virus (AcNPV) has been shown to be capable of displaying a foreign protein on the virion surface. This display system is achieved by the use of the major surface glycoprotein of AcNPV, gp64, to display foreign proteins on the virion surface by in-frame fusion between the gp64 signal sequence and mature foreign protein domain. For example, the external domain of HIV envelope protein gp120 was displayed on the virion surface and possessed binding activity for its ligand, CD4, indicating that baculovirus displaying specific ligand binding proteins may serve as an effective tool for targeted gene therapy to specific cells. The baculovirus display system has several potential advantages as a vaccine vehicle. Use of purified virions as an immunogen alleviates the need for the addition of an extraneous adjuvant to the vaccine formulation due to the intrinsic immunostimulatory effect often associated with viral immunogens. Additionally, presentation of an antigen in its proper conformation on the virion surface makes it readily accessible for interactions with cellular components of the immune system. The absence of the preexisting antibodies against baculovirus in animals can benefit to induce primary antibody responses to foreign proteins expressed in this manner prior to the production of neutralizing antibodies against the viral virions. In fact, pathogen antigens such as the HIV envelope protein gp41, foot - and mouth disease virus capsid protein and Theileria parva p67 antigen have been expressed using this system, and some of them induced high titers of antigen-specific antibodies. To our knowledge, however, the efficacy of the baculovirus display system as a protective vaccine delivery system has not been previously reported. To evaluate the baculovirus display system as a vaccine vehicle, we have generated a recombinant baculovirus (AcNPV-CSPsurf) that displays the rodent malaria Plasmodium berghei circumsporozoite protein (PbCSP) on the virion surface as a fusion protein with the major baculovirus envelope glycoprotein gp64. The PbCSP-gp64 fusion protein was incorporated and oligomerized on the virion surface and led to a 12-fold increase in the binding activity of AcNPV-CSPsurf virions to HepG2 cells. Immunization with adjuvant-free AcNPV-CSPsurf virions induced high levels of antibodies and gamma interferon secreting cells against PbCSP, and protected 60% of mice against sporozoite challenge. These data demonstrate that AcNPV-CSPsurf displays sporozoite-like PbCSP on the virion surface and possesses dual potentials as a malaria vaccine candidate and a liver-directed gene delivery vehicle.

P2-56) THE TARGET OF ATOVAQUONE IN PLASMODIUM FALCIPARUM

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Atovaquone is effective against chloroquine - resistant strains and is already being used for therapy in epidemic regions in Africa and Thailand. However, its target is still ambiguous. The effect of the drug on mitochondrial respiration and the structural similarity of naphtoquinone to atovaquone and ubiquinol suggest that the ubiquinol - cytochrome c reductase (bc_1 complex, also referred to as complex III) is the drug target. Mutation studies involving cytochrome b in the resistant strain of *Plasmodium* and *P. carinii* also suggest that bc_1 complex is a target of atovaquone. Furthermore, atovaquone is thought to inhibit dihydroorotate dehydrogenase (DHOD) in *Plasmodium* mitochondria, which catalyzes the conversion of dihydroorotate to orotate and transfers a reducing equivalent to ubiquinone in the respiratory chain.

The reason that the target of atovaquone remains unclear is that no active *Plasmodium* mitochondria have been available for biochemical study. In the previous study, we established the method of preparing mitochondria from *P. falciparum* with high activity by the N₂ cavitation method. In this study, we established reproducible assay system of bc1 complex, and directly showed that atovaquone inhibit *Plasmodium* mitochondrial bc_1 complex selectively rather than DHOD.

Specific inhibition of *Plasmodium* cytochrome bc_1 complex by atovaquone was observed. IC₅₀ for *P. falciparum* cytochrome bc_1 complex was 0.052 nM, while that for rat liver mitochondria was 32.3 nM. DHOD was fully active even in the presence of 100 microM atovaquone. These results clearly indicate that the target of atovaquone is *Plasmodium* cytochrome bc_1 complex.

P2-57) PLASMODIUM BERGHEI NK65 GIVING RISE TO RECRUDESCENCE AFTER CHEMOTHRAPY.

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Recrudescence of Plasmodium berghei NK65 infection was studied to elucidate the factors related to recrudescence. Treatment with a high concentration of chloroquine did not prevent recrudescence, although the reappearance of recrudescent parasites exposed to a higher concentration of chloroquine a few days delayed than that exposed to a lower concentration of chloroquine. A longer duration of treatment prevented recrudescence more efficiently than a shorter duration of treatment when the same concentration of chloroquine was administered. Infection with a larger number of parasites caused recrudescence more frequently. Recrudescent parasites were as sensitive to chloroquine as those were before recrudescence. The results demonstrated that drug sensitive parasites recrudesced in a mouse model of malaria after drug treatment. To examine whether phagocytosed parasites cause recrudescence, phagocytes were decreased in number or were hindered from phagocytosis. Mice splenectomized and administered carbon particle were infected with parasites and treated with chloroquine. The number of mice showing recrudescence were significantly larger in a mice group that were splenectomized and administered carbon than in one that were given sham operation and PBS. The results demonstrated that phagocytes contributed to suppress recrudescence. These results did not suggest that merozoite stage of parasites might escape the effect of chloroquine by hiding in phagocytes, but suggested that latent parasites like dormant ring-form stages of parasites caused recrudescence.

P2-58) EPIDEMIOLOGICAL AND HISTOLOGICAL STUDY OF BURKITT LYMPHOMA IN INLAND PROVINCES OF KENYA

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Burkitt lymphoma (BL) is one of high-grade B-cell

lymphomas and classified to endemic, sporadic and im-

munodeficiency-associated types. Almost always endemic BL is related to Epstein-Barr virus (EBV) infection and occurred geographically in tropical Africa, Papua New Guinea and low altitude areas of South America, where are holoendemic areas of falciparum malaria (FM). The results of our epidemiological study on African endemic BL in inland provinces of Kenya showed that a higher prevalence rate of BL is found among the Luo ethnic group, who are living around Lake Victoria. The lake basin area of Kenya is a holoendemic area of FM and our findings support to hypothesize that frequent infections of FM in childhood has an indirect condition to develop endemic BL in combination with EBV infection. BL is histologically characterized by "starry sky" appearance, which is manifested by the scattering of macrophages that appear within lacunae surrounded by proliferating lymphoma cells. The virus in most EBV-infected lymphocytes remains in latent cycle, whereas some viruses enter the lytic cycle to produce EBV virions. Although these two infection stages have been demonstrated in established cell lines derived from BLs, the relationship between the infection stages of EBV in the BL tissues and the development of their "starry sky" appearance has not been dclarified. We studied the relation between EBV infection stages and the formation of "starry-sky" apperance in African endemic BL, using EBV-encoded small RNAs (EBERs) as a marker of latent infection of EBV, and BamHI H Left Frame 1 (BHLF1) and BamHI Z EBV (ZEBRA) as lytic cycle markers of EBV. In all 44 cases, EBER was positive in most of the lymphoma cells, and BHLF1 and ZEBRA were positive in the lymphoma cells within and around the lacunae in "starry sky" appearances among 73% of all cases, respectively. The mean numbers of lacunae per unit area in cases positive for both or one of lytic cycle markers (BHLF1 and/or ZEBRA) were significantly higher than that of negative cases. These findings suggest that the transition to the lytic cycle of EBV in the lymphoma cells is the histomorphogenetic factor influencing the formation of "starry sky" appearance in

P2-59) EPIDEMIOLOGICAL STUDY AND CHARACTERIZATION OF JAPANESE ENCEPHALITIS VIRUS STRAINS ISOLATED IN OKINAWA ISLAND, IN 2002 AND 2003

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Japanese encephalitis (JE) had been one of the major public health problems before 1970's in Japan, including Okinawa island. No case has been reported officially in Okinawa island since 1974, but transmission of JE virus in nature is demonstrated by the high prevalence of JE antibody in swine sera, and by the virus isolation from swine sera and mosquitoes. Polymorphism of JE virus is recognized serologically and genetically. Five genotypes of JE virus are reported at present. Genotype 1 was distributed in northern Thailand and Cambodia, and genotype 3 was widely distributed in Asia including Japan, Korea, China, India, and Nepal. Over 150 strains of JE virus were isolated in Okinawa island before 1992 and all of the studied strains belonged to genotype 3. Recently genotype 1 strains have newly been emerged in Japan, Korea, Vietnam, and Australia. Okinawa island is located between the East China Sea and Pacific Ocean, 2000 km south of mainland, Japan. We carried out epidemiological study of JE virus to clarify recent change of its distribution in Asia.

African endemic BL.

One and four strains of JE virus were isolated in 2002 and 2003, respectively, from swine sera in Okinawa island. Interestingly, genetic analysis based on 240 nucleotides of core/premembrane gene region showed all of the five isolates belonged to genotype 1. To investigate the pathogenesis of newly emerged Okinawan isolates, we compared the neurovirulence and neuroinvasiveness of Oki431S strain (genotype 1 isolate in 2002) with those of Na 54 strain (genotype 3 Okinawan isolate from swine sera in 1985). Neurovirulence and neuroinvasiveness were indicated as LD50 values by inoculation intracerebrally and intraperitoneally, respectively, to ICR mice. The pathogenesis using both indications showed that of Oki431S in mice was apparently weaker than that of Na 54. To clarify antigenic change of JE virus in Okinawa island, neutralization titers against Nakayama (prototype), Oki431S, and Na 54 strains were surveyed in swine sera in 1985-1988 and 2002. The swine sera in 1985-1988

tended to neutralize all strains, while most of those in 2002 neutralized Oki431S strongly but did not neutralize Na 54.

These results indicate that genetic and antigenic dis-

tribution of JE virus has changed in Okinawa island during this decade, and suggest that exogenous JE virus has arrived naturally or artificially, and spread to Okinawa island.

P2-60) A CLINICAL SIGNIFICANCE OF PLATELET-ASSOCIATED IgG AND IgM IN DENGUE VIRUS

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Although the public health impact of dengue is increasing rapidly, the mechanism of thrombocytopenia in this disease remains unknown. We already reported about the significant inverse correlation between platelet - associated IgG and platelet count of the patients in the acute phase of secondary dengue infection. We also demonstrated that anti-dengue virus antibody is concerned in PAIgG of these patients. But the correlation between PAIgG and disease severity is unclear. In this study, we investigated the correlation between PAIgG, PAIgM and platelet count, PAIgG, PAIgM and type of disease severity on dengue patients.

We examined PAIgG or PAIgM level by competition ELISA method, using the platelet from 67 dengue patients, admitted in San Lazaro Hospital in Metro Manila from September to November, 2002. Then classified their diagnosis followed WHO criteria in 1997. Hemaggulutinin Inhibition test was performed to determine whether their infection was primary or secondary. The mean age of the enrolled 67 patients was 19.8. Thirteen of them were diagnosed as DF, and 54 were DHF. Sixty patients had secondary infection. Based on the serum sample taken from 67 patients during the acute phase of infection, the platelet count had an inverse correlation not only with PAIgG, but also with PAIgM value. The value was significantly higher than those of the convalescent phase. The value of PAIgG and PAIgM for DHF patients tended to be higher than those for DF patients. These finding suggest that, PAIgG and PAIgM may play a role in the mechanism of the thrombocytopenia on dengue virus infection. More case study are needed to really definite the clinical significance of PAIgG, PAIgM on primary and secondary infection as well as with disease severity.

Collaborators : San Lazaro Hospital ; AM Robles, MTP Alera, EM Dimaano. St. Luke's Med Center ; DJM Cruz, RR Matias, FF Natividad.

P2-61) CLINICAL ASSESSMENT OF DENGUE AMONG CHILDREN IN METRO MANILA : 1999-2001

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Dengue is mosquito-borne febrile illness. Recently its breakout is seemed around Metro Manila in Philippine and which is the most important medical and public health problems. Dengue illness has two types, dengue fever (DF) has relatively light symptoms and dengue hemorrhage fever (DHF) has possible more severe ones. We examined the difference of clinical feature and laboratory data between DF and DHF among pediatric patients with dengue. We enrolled 504 patients with clinically suspected of having a dengue virus infection at the pediatric ward of St. Lukes Medical Center between Jan 1999 and Dec 2001. Of these, 356 pediatric patients were diagnosed of having a dengue virus infection by IgM capture ELISA or RT-PCR using multiple plasma samples and the grading severity of DHF were diagnosed by WHO definition (1997). Precise clinical information including the severity of disease and disseminated intravascular coagulopathy (DIC) were evaluated using the Japanese Ministry of Health and Welfare, 1988. In our study period, outbreak was in 2001, and the peak point of year is among Sep- Aug and the average of age is 10.5 y.o. Of these enrolled 356 pediatric patients, 113 (31.8%) were diagnosed as DHF, and DHF grade I is 1.9%, II is

30.6%, III is 0.6%, and IV is 0.3%. The mortality rate was low (0.3%). Clinical symptoms such as abdominal pain and epistaxis were more frequently seen in DHF than in DF at the time of admission. Peripheral platelet counts were constantly lower in DHF than in DF. Incidence rate of DIC was low but significantly higher in DHF than in DF. Our data could be helpful for predicting DHF in clinical practice and understanding of its pathophysiology. Collaborators : St.Lukes Med Center ; Cinco MTD, Carlos C, Cruz DJM, Matias RR, Natividad FF.

P2-62) ELECTRON MICROSCOPIC STUDIES ON THE MULTIPLICATION OF CHIKUNGUNYA VIRUS IN AEDES ALBOPICTUS

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Virus replication in the midgut, salivary gland and brain after experimental infection was observed with electron microscope for analyzing vector competence. The multiplication of the chikungunya virus was observed in Aedes albopictus infected with the virus. In the midgut, the virus increased by the method of the budding in the basal labyrinth. In the salivary gland, the virion was observed in various regions. The basal labyrinth is distributed like the labyrinth in the salivary gland cell. One or several virions were found in the basal labyrinth inside of the cell. Large number of enveloped virus particles was observed in the area surrounding plasma membrane and basal lamina. In this region, the multiplication image by the budding was observed only the small number. The position which seemed to be the multiplication factory near the region was observed. The virion which was completed in the vacuole of the cytoplasm was observed . The virus multiplication process in the salivary gland was estimated from these observations.

P2-63) SELF-REPORTED BLOOD IN URINE AND WATER CONTACT IN A SCHISTOSOMIASIS ENDEMIC AREA OF COASTAL KENYA

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The relation of self-reported blood in urine with water contact activities was studied in a schistosomiasis endemic area of coastal Kenya. The study focused on how it varied in two different rivers and among types of water contact activities.

We visited the homes of 1,248 villagers, 569 males and 679 females in 2002 to collect data on self-reported blood in urine and other urinary schistosomiasis-related subjective symptoms. We also inquired if they ever came into contact with Kadingo river [K] and/or Manolo river [M] in the 14 days previous to the interview, and for what purposed (fishing, bathing, washing, recreation or crossing). The results were as follows :

1. The prevalence of self-reported blood in urine was an average of 14% (Male : 15%, Female : 13%) ; the highest of which was 33% (Male : 38%, Female : 28%) at

ages ranging from 10-19 years old.

2. About 30% out of the 1, 248 subjects (Male : 39%, Female : 24%) had contacts with K river or M river and/or both ; 181 for K river, 186 for M river, and 12 for both rivers. The remaining 868 of the subjects had no contact with either river. The prevalence of self-reported blood in urine was higher among those who had contact with river(s) (25%) than those who had not (9%, $x^2=51.704$, p<0.001). The prevalence of self-reported blood in urine of those who had contact with K river (33%) was higher than those who had contact with M river (16%, $x^2=12.587$, p<0.001).

3. Contact with the rivers in general were : 28% for bathing and washing, 20% crossing, 14% recreation, and 8% for fishing. For the 11-19 years old group, the figures were 39%, 36%, 25%, and 20%, respectively.

4. Crossing and recreation percentages in K river were significantly high, while bathing, washing and fishing were significantly high for M river.

5. With a multiple logistic regression analysis, the odds ratio of self-reported blood in urine was 3.2 (95% CI : 1.8-5.9) for contact with K river to M river, 2.1 (1.2 - 3.7) for contact for recreation to no contact for recreation, and 1.9 (1.0-3.5) for fishing to no fishing.

Conclusions : Self-reported blood in urine in this study area differed between rivers and also differed among type of water contact activities. Recreational activities in K river would be most risky for the schistosome infection. Warnings accompanied with health education regarding specific activities in specific rivers are more comprehensive and effective than ambiguous prohibitions.

P2-64) URINARY SCHISTOSOMIASIS IN NORTH ZANZIBAR : AN EPIDEMIOLOGICAL STUDY WITH QUESTIONNAIRE FOR WATER CONTACT BEHAVIORS AND SELF-DIAGNOSIS

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The estimation of exposure to the cercarial infested water is indispensable for eco-epidemiological study in schistosomiasis endemic area. We have been conducting researches to evaluate the accuracy of interview data, compared to direct observation method, on water contact behaviors, and concluded that the 24-hour recall method, with certain adjustment, was a practical means to accurately quantify water exposures in Lower Moshi area, Tanzania, where Schistosoma mansoni has been endemic. However, long-term survey using these methods need time and cost, especially for a large sample. The present study was conducted for evaluating the reliability of rapid questionnaire on water contact behaviors and self - diagnosis in Bandamaji, a urinary schistosomiasis endemic area in north Zanzibar (Unguja), Tanzania. The number of subject was 648 (65% of whole population), and 50.3% (males : 62.6%, females : 39.4%) of the subjects was infected with S. haematobium. The egg-positive rates gradually decreased with increasing age groups, with its peak at 70.7% (males : 83.3%, females : 58.2) for the 6 -18 age group. The mean number of eggs was 17.4 eggs/10ml (geometrical mean; males: 21.6, females:

12.9); the intensity was highest among 6-18 age group with 23.5 eggs/10ml (males : 33.5, females : 14.3). These results imply that the prevalence of schistosomiasis was high, more that 50% in total, while the intensity of infection was moderate. The subjects who used river and pond water, i.e. risk water for infection, for bathing and laundry purposes showed significantly higher rate of morbidity than those who do not use ($p \le 0.001$). On the contrary, there was no significant relationship between schistosomiasis infection and well-water use for the same purposes (p=0.980). Bivariate analyses demonstrated that self-diagnosis was a practical factor for screening (odds ratio : 13.5, p0.001), as is the case with age and sex. Logistic regression analysis also clarified that above - mentioned factors were significant and had potential to be applied as useful factors in rapid questionnaire for schistosomiasis infection. Although question on self-diagnosis have been considered as one of the useful factors for the rapid and inexpensive screening method, mainly due to this disease's evident symptom, blood in urine, the accuracy of data by rapid questionnaire may be improved by adding simple questions for the water contact behaviors.

P2-65) INHIBITION OF SCHISTOSOMA MANSONIA INFECTION BY ULTRAVIOLET CREAM

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We experimented on two types whether ultraviolet (UV) cream influences for cercaria and its' growth or not.

On the first experiments, it investigated the influence of UV cream (Anessa, SHISEIDOU CO., Japan) for growth of cercaria by in-vivo experiments. Experimental animals were used 55 mice of C57-Black in this study. Abdominal skin of mouse was cut hair at 4 cm² and UV cream put on the skin area. Mice were parted in control group without UV cream and in three group by dose of UV cream at 1, 2, 4 mg/cm². It applied each mice 150 cercaria through the skin painted UV cream by ring method under anesthesia. We evaluated the inhibitory effect of UV cream for growth of cercaria by recovery rate of adult worm at 7 weeks after cercaria infection.

As results in the first experiment, invasion rate of cercaria were not recognized the statistical significant difference compared among four groups. However, inhibitory recovery rate of adult worm in UV cream groups were 53.6% of 1 mg/cm² UV cream, 58.9% of 2 mg/cm² UV cream and 78.9% of 4 mg/cm² UV cream as comparison with control group with statistical significants.

On the second experiments, it investigated the direct influence of UV cream for cercaria by in-vitro experiments. UV cream, which six dose were 32 mg to 1 mg, painted on each hole bottom of 96 hole multi-plate, and put cercaria with dechloridation water of 0.1 ml in each hole for half and one hour. After that, cercaria were dyed with methylene blue stain. The activity of cercaria was evaluated by color concentration in cercaria. As results in this experiments, there are positive correlations during damage in cercaria and dose of UV cream or exposure time. There was a statistical different between over 16 mg/cm2 of UV cream and control.

In this study, UV cream has a depression effect to growth of cercaria of Schistosoma Mansonia in final host at usual dose. And UV cream in high dose has a casualties action for cercaria with. These results suggest that UV cream inhibits aggravation of the state of Schistosomiasis. We experiment now to specify the material with inhibitory effect for Schistosoma Mansonia in UV cream.

P2-66) TOWARD THE INTERRUPTION OF VECTORAL TRANSMISSION OF TRYPANOSOMA CRUZI - IMPACT OF VECTOR CONTROL AGAINST CHAGAS DISEASE VECTOR IN GUATEMALA

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This study determined the effects of the vector control operation against *R. prolixus* and *T. dimidiata* in the departments of Jutiapa and Zacapa. First of all, an entomological survey was conducted to identify infested villages. Residual spraying of pyrethroid insecticides was performed in the infested villages by *R. prolixus* and *T. dimidiata*. A post-spraying survey evaluated the impact of spraying by comparing entomological indices before and after the spraying.

The majority of *R. prolixus* was captured in Zacapa, and 15.8% of villages were infested with low bug-density level. *T. dimidiata* was widely distributed, and more houses were infested in Jutiapa. Residual spraying was performed, and three types of pyrethroid insecticides (beta-cyfluthrin, deltamethrin and lambda-cyhalothrin) were sprayed. A post-spraying vector survey demonstrated that the insecticide spraying was effective. The percentage of infested houses by *R. prolixus* fell by 97.7%. The percentage of infested houses by *T. dimidiata* fell by 75.3% in Jutiapa, and by 90.6% in Zacapa. Reinfestation were observed mainly inside the houses, probably indicating that some indoor bugs survived the spraying round. Elimination of *R. prolixus* is attainable by spraying 100% of the infested villages. To reach the goal of elimination, residual spraying of 100% of the infested houses needs to be assured. In order to control the *T. dimidiata* population, the efficacy of the insecticide spraying needs to be improved, spraying techniques need to be reviewed, and insecticides need to be re-applied at regular intervals. An effective vector-surveillance system with community participation is also essential.

P2-67) CHARACTERIZATION OF PROCYCLIC FORM-SPECIFIC GENES FROM TRYPANOSOMA BRUCEI BRUCEI

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Trypanpsoma brucei brucei protozoa spreads between the mammalhost and the tsetse fly vector in its lifecycle. In order to obtain information for control of trymanosomiasis, we analyzed geneexpression profiles of bloodstream and procyclic (insect) forms of *T.b.brucei* using the method of fluorescence differential display method. About 12,000 cDNAs were surveyed,and one cDNA was found to be markedly up-regulated in procyclic forms. A full length cDNA was obtained by 5'RACE method and sequenced. The gene was 3304bp long and had an open reading frame of 813 amino acids. A sequence homology search with Gene Bank database revealed that this gene has 38% identity with *Trypanosoma cruzi* (*T. cruzi*) Cyclin 5 (TzCYC5) gene. Suggesting this gene (named TbCYC 5) that also plays an important role in the cell cycle control similar to TzCYC 5.RT-PCR analysis demonstrated that the gene was not expressed in long slender bloodstream form, while it was expressed in short stumpy (pre-adapted) form, which can be changed to procyclic form when the blood is sucked by tsetse fly. Therefore, it seems likely that this gene has an important function in lifecycle change of *T. b. brucei* from bloodstream to procyclic forms. The growth rate of the procyclic form cells was not changed by RNAi-mediated TbCYC 5 gene knocked down (silenced) analysis.

Electron Microscopical analysis of the TbCYC 5 - knocked down cells and identification of interacting molecules by yeast two hybrid system are now under way.

P2-68) MOLECULAR CLONING AND CHARACTERIZATION OF VACUOLE PROTEIN SORTING (VPS41) GENE FROM *TRYPANOSOMA BRUCEI BRUCEI*

TAKASHI SUZUKI¹, SHAOHONG LU², YOSHISADA YABU¹, NARUSHI IIZUKA³, SHIGERU OHSHIMA¹, MARIKO HATO¹, NOBUO OHTA¹

¹Dept. Mol. Parasitol., Nagoya City Univ. Grad. Sch. Med. Sci., Aichi, Japan., ²Inst. Parasitic Diseases, Zhejiang Acad. Med. Sci., Zhejiang, China, ³Dept. Microbiol. and infect., Nagoya City Univ. Grad. Sch. Med. Sci., Aichi, Japan. African trypanosomes are causative protozoan agent for sleeping sickness in human and nagana disease in cattle. Incubation of bloodstream forms of *Trypanosoma brucei* with an iron chelator, deferoxamine leads to growth inhibition, suggesting iron-uptake system is a potential target for chemotherapy. But the iron-uptake mechanism of *T. brucei* cells has yet to be investigated in detail and remains almost completely unexplored at the molecular level.

In order to clarify the iron-uptake mechanism of *T. brucei* cells, *T. brucei* genome database (TIGR) were searched for genes involved in iron-uptake, and a potential coding sequence of homologue of *Saccaromyces cerevisiae VPS41* (*ScVPS41*) was found, which has been reported to be important for iron-uptake. By 5' RACE and 3' RACE, full - length *Trypanosoma brucei VPS41* (*TbVPS41*) cDNA (gene) was cloned and sequenced. The full - length *TbVPS41* was 3580 bp long, with an open

reading frame of 1092 amino acids and two domains : WD40 and clatrin heavy chain repeat (CHCR). TbVPS41 gene showed moderate similarity (about 40% positives) with yeast, human and tomato homologues, and was expressed equally in RNA-level between bloodstream and procyclic (insect) forms. RNAi-mediated TbVPS41 gene knock down analysis demonstrated the interference of the VPS41 gene expression causes lower growth rate of Trvpanosoma cells in iron-deficient medium and fragmented lysosome-like phenotype. Heterologous expression of TbVPS41 gene in $\Delta vps41$ yeast cells demonstrated that *TbVPS41* gene could suppress the growth of $\Delta vps41$ yeast cells in iron-deficient medium, while it could not suppress fragmented-vacuole phenotype of $\Delta vps41$. These results indicate that TbVPS41 has evolutionally conserved iron uptake function, while its function to maintain membranous organelle seems to differ between T. brucei and S. cerevisiae.

P2-69) DEVELOPMENT OF AN IMMUNOCHROMATOGRAPHIC TEST FOR THE RAPID DETECTION OF *BABESIA EQUI* INFECTION IN HORSES

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To develop a rapid, simple, and reliable immunoassay for the diagnosis of *Babesia equi* infection, a highly purified recombinant truncated *B. equi* merozoite antigen - 2 (rEMA-2t) was conjugated with gold colloid as a conjugated antigen and immobilized on a nitrocellulose membrane as a capture antigen. An immunochromatographic test (ICT) was sequentially developed and evaluated using sera from 11 and 8 horses infected experimentally with *B. equi* and *B. caballi*, respectively, and 20 normal horses.

The specific antibodies were detected in all serum samples from *B. equi*-infected horses but in none of those from *B. caballi*-infected and normal horses. In sequential sera from 2 *B. equi*-infected horses, the antibodies were detectable as early as 4 days post-infection, which might be due to the fact that this system is capable of detecting various classes of antibodies, such as IgM, IgA, and IgG. In the detection of specific antibodies in field sera from 61 horses, the result of the ICT was compared with those of Western blot analysis and the enzyme-linked immunosorbent assay. The agreement of the three tests was 95.1% (58/61). The ICT might be more sensitive than the others.

The results suggest that the ICT is rapid, simple, accurate, and relatively inexpensive. All these characteristics would make it a suitable diagnostic tool in both laboratory and field conditions.

P2-70) MURINE DENDRITIC CELLS DERIVED FROM PERITONEAL CAVITY MACROPHAGES UNDERGO RAPID APOPTOSIS IN CULTURE WHICH IS BLOCKED BY TOXOPLASMA GONDII

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P2-71) POSSIBLE ROLE OF CALCIUM IONS AND CALMODULIN IN THE EXCYSTATION AND METACYSTIC DEVELOPMENT OF *ENTAMOEBA*

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The effect of calcium ions (Ca^{2+}) and calmodulin (CaM) on the excystation and metacystic development of Entamoeba invadens was examined by transfer of cysts to a growth medium containing calcium antagonists and CaM inhibitors. Excystation, which was assessed by counting the number of metacystic amoebae, was inhibited by the cacium chelators EGTA and EDTA, with EDTA being more potent than EGTA. The inhibitory effect of higher concentrations of these chelators on excystation was associated with reduced viability of cysts. Metacystic development, when determined by the number of nuclei in an amoeba, was delayed by EGTA, because the percentage of four-nucleate amoebae was higher than in controls at day 3 of incubation. EDTA made metacystic development unusual by producing a large number of metacystic amoebae with more than ten nuclei. The inhibition of excystation by these chelators was partially abrogated by their removal. A putative antagonist of intracellular calcium flux, TMB-8, also inhibited the excystation and metacystic development, but had little effect on cyst viability. The slow Na⁺-Ca²⁺ channel blocker bepridil but not verapamil inhibited the excystation and metacystic development, associating with reduced cyst viability at higher concentrations. The inhibitory effect of bepridil on excystation was abrogated by removal of the drug. The CaM inhibitor trifluoperazine (TFP) but not W-7 inhibited the excystation and metacystic development. The inhibitory effect of TFP on excystation was also abrogated by removal of the drug. These results indicate that extracellular calcium ions, amoebic intracellular calcium flux, calcium channels, and a CaM-dependent process contribute to the excystation and metacystic development of E. invadens.

P2-72) ANALYSIS OF FARNESYLTRANSFERASE OF ENTAMOEBA HISTOLYTICA

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Ras proteins function as a molecular switch of signal transduction in cell proliferation and cell differentiation. Ras proteins require a post-translational lipid modification called protein farnesylation in order to become membrane-associated and functional. Farnesyltransferase (FT) catalyzes the protein farnesylation. Since constitutively active mutations induce carcinogenesis, FT has been attracted attention as a target for cancer chemotherapy. We have already cloned the genes of alfa and beta subunit of FT from *Entamoeba histolytica* and expressed the proteins in *Escherichia coli*, showed its enzyme activity, and identified one of its intrinsic substrates (*Eh*Ras-CVVA) belonging to Ras superfamily proteins. Phylogenetical analysis revealed that *Eh*FT was independent of FTs from other species. Among the Ras superfamily proteins of *E. histolytica*, *Eh*Ras-CVVA located philogenetically between Ras/Rap and Rho/Rac families. *Eh*FT was highly (about 1,000fold) resistant to both farnesyl analogue and peptide mimetic inhibitors for human FT. These results suggest that *Eh*FT is remarkably different from human FT in binding to substrates and inhibitors, which highlight this enzyme as a novel target for chemotherapy against amebiasis.