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Letter to the Editor

Dear Sir

Proactive plan to help contain pandemic influenza in Asia

Received 28, June, 2006

Ferguson and colleagues' model of influenza transmission[1] indicates that elimination of a nascent influenza pandemic might be feasible using a combination of geographically-targeted antiviral prophylaxis[2] (particularly neuraminidase inhibitors), together with social distancing measures, and that the speed of distribution of antivirals will be crucial.

Participants at the Japan-WHO Joint meeting on Early Response to Potential Influenza Pandemic, held in Tokyo in January[3], reached the same conclusion. In addition, there was consensus that early containment measures would be essential to help slow the spread of pandemic influenza, helping to buy time to develop an effective vaccine against any newly-evolved influenza strains.

In Asia, where more than 80% of human Avian flu cases have so far been reported, the level of preparedness with respect to antivirals to contain a potential influenza pandemic is generally less than adequate. Stocks of drugs for containment and treatment purposes in Indonesia total less than 20,000 courses, Cambodia and Lao PDR less than 200, and Myanmar almost nil, whereas stockpiles in Thailand and Singapore are approximately 100,000 and 700,000 respectively. Moreover, it is likely that there will be significant logistical delay in antivirals currently stockpiled by WHO in Europe and North America for global containment purposes being mobilised and distributed in Asia.

To help contain any regional Avian flu pandemic threat, Japan has proactively decided to provide 500,000 courses of anti-flu drugs to Association of South East Asian Nations (ASEAN) countries with the intention of storing them in the international airport in Singapore, where they will be available for rapid distribution. Should an outbreak occur, following guidance from WHO, the Government of Japan, the affected country, and the ASEAN secretariat will coordinate delivery of antivirals to the international airport nearest to the location of the outbreak, ideally within 12 hours. This should effectively fill the gap until antiviral drugs stockpiled by WHO became available. A Plan of Action is currently being devised to ensure rapid deployment of the stockpile when the need arises. Indicative of the nation's intention to fully tackle Avian Flu, according to a recent World Bank report, Japan remains one of only three nations to fully commit funds pledged at the Avian Flu funding meeting in Beijing in January, disbursing $158 million thus far to a variety of countries and organizations at the regional and global levels. The European Union's recent failure to implement a plan to stockpile Tamiflu indicates that Europe's commitment to Avian Flu pandemic preparedness may be on the wane.

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CRYOPRESERVATION OF NEWBORN AND MUSCLE-STAGE LARVAE OF *TRICHINELLA NATIVA*

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Accepted 17, May, 2006

Abstract: We examined the basic conditions for cryopreservation of newborn and muscle-stage larvae of *Trichinella nativa*. Two types of cryoprotectants were examined: dimethyl sulfoxide and glycerol. For cryopreservation of newborn larvae, dimethyl sulfoxide had a cryoprotective effect, and a maximum survival of 96% was obtained under conditions of 10% dimethyl sulfoxide and 30 min equilibration. Muscle-stage larvae, on the other hand, demonstrated a relatively high survival rate (approximately 80%) with 25% glycerol and three hours equilibration regardless of freezing rate. The infectivity of cryopreserved newborn and muscle-stage larvae was examined using different infection methods: direct injection into the abdominal cavity in the former, and either oral infection or direct injection into the duodenum in the later. Cryopreserved newborn larvae showed infectivity to all the subjected mice. By contrast, muscle-stage larvae did not show any infection when infected orally, but one mouse was positive when the larvae were injected directly into the duodenum.
Keywords: cryopreservation; *Trichinella nativa*; cryoprotectant; freezing

INTRODUCTION

Trichinella spp. is a nematode parasite, the larvae of which are known to encyst in the muscles of mammals, including humans. There are many species of *Trichinella*, all of which can cause the food-borne illness trichinellosis or trichinosis. This illness occurs when raw or improperly cooked meat containing the larvae of the parasite is consumed by mammals, including humans. Clinical manifestations of infection vary from asymptomatic to moderate gastrointestinal distress to cardiac and neurological complications and possibly even death [18]. Consequently, many laboratories are currently investigating the biological and biochemical characteristics of a large number of *Trichinella* spp. isolates using laboratory animals. However, conventional methods for the propagation and preservation of parasites *in vivo* and *in vitro* have a number of limitations, including the need for labor, bacterial/fungal contamination, and changes in the original biological and metabolic characteristics [7, 15, 21]. All of these disadvantages are considerably reduced by cryopreservation, which to date has been applied to several species of parasites.

Some protozoan parasites such as *Trypanosoma* spp. and *Plasmodium* spp. have been successfully cryopreserved with high survival rates [5, 15]. These single cells respond in a relatively uniform way to physical and chemical stresses during the freezing procedures and can be frozen easily without cryoprotectants or precise control of freezing rate [5, 15, 22]. On the contrary, in multicellular systems like helminths, the response to cryopreservation protocols is not uniform, thus making successful cryopreservation difficult to achieve. Although many studies on the cryopreservation of helminthic parasites have been conducted [7, 10, 14], only a few species, mainly nematodes, have been successfully cryopreserved, i.e., microfilariae of *Dirofilaria corynodes*, *Brugia malayi*, and *Wuchereria bancrofti* [14]; first-stage larvae of *Haemonchus contortus*, *Trichostrongylus colubriformis*, and *Ostertagia circumcincta* [7]; second-stage larvae of *Toxocara canis* [20]; and various staged larvae of *Trichinella* spp. [11, 19].

Pozio *et al*. (1988) [19] carried out cryopreservation of newborn larvae (NBL) and muscle-stage larvae (MSL) of different *Trichinella* spp.. Although approximately 80% of the NBL were found to be motile after thawing, only 8% of *T. spiralis*, 6% of *T. nativa*, and 1% of *T. pseudospiralis* were infective to mice. On the contrary, none of the MSL were infective to mice despite motility rates of 80 ± 7%. Similarly, Genan and James (1988) [11] reported cryopreservation of *Trichinella* spp. using liquid nitrogen. They pre-treated the MSL with 10% dog's bile at 37℃ for one hour and then adopted a freezing rate of 5,100℃ min^{-1}. Of the MSL frozen, 80% were motile, and of these, 1.1 ± 0.4% grew to adults when inoculated in mice. Pozio *et al*. (1988) [19] reported that cryopreservation of adult worms

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always failed and that motile NBL were frequently observed beside broken female worms damaged during freezing.

Although numerous authors have studied the cryopreservation of parasites, studies related to *Trichinella* spp. are limited [11, 19]. Therefore, the fundamental conditions needed for cryopreservation remain relatively unknown. In the present study, we examine the basic conditions (the type and concentration of cryoprotectant, equilibration time, and freezing rate) required for cryopreservation of *Trichinella nativa*.

MATERIALS AND METHODS

Preparation of parasites

We used *T. nativa* parasites kindly supplied by the Department of Parasitology, Gifu University School of Medicine, Japan. The parasites were maintained using ICR mice, and the NBL and MSL were subjected to cryopreservation. To obtain NBL, the ICR mice were infected orally with 300–400 MSL. On the sixth day post infection, each mouse was sacrificed and then the entire small intestine was removed and slit longitudinally to recover the adult worms. The recovered adult worms were washed twice in Medium 199 (Gibco, Tokyo, Japan) containing horse serum 10% (v/v) (100 UI/ml penicillin; 100 mg/ml streptomycin; 2 mg/ml liquid fungizone). They were then placed in a 10-ml petri dish with the same volume of culture medium and incubated at 37 ℃ in a CO_2 incubator. After 48 hrs, NBL were recoverable from the culture medium. The culture medium containing NBL was washed in sterile warm phosphate-buffered saline (PBS) then centrifuged at 450 × *g* for 10 min at room temperature.

To obtain MSL, the muscles of infected mice were subjected to artificial digestion for one hour using digestive solution (1,000 ml distilled water; 10 ml HCl; 1 g pepsin). MSL were recovered and washed twice in PBS then treated with 0.25% cetyltrimethylammonium bromide (CTAB) (Gibco, Tokyo, Japan) or 10% dog's bile in culture medium at 37 ℃ for one hour. They were then cultured in culture medium as above for NBL.

Cryoprotectants

Two cryoprotectants were tested, namely, dimethyl sulfoxide (Me_2SO) (Wako, Osaka, Japan) and glycerol (Wako). The cryoprotectant was added to a 1.2-ml plastic cryogenic vial (Wako) and adjusted to a final concentration of 5–30% as appropriate for the different subcultures. The entire mixture was then incubated at 37 ℃ for 0.25–3 hrs (equilibration).

Freezing and thawing

The freezing procedure used in this experiment was based on Miyake *et al*. (2004) [16]. Briefly, a cryogenic vial containing the *T. nativa* suspension was dipped in 99.5% alcohol. The freezing rate was then controlled by adding small pieces of dry ice to the alcohol. Freezing rates of the NBL and MSL to −40 ℃ were 0.3, 1, and 3 ℃ min^{-1}, followed by 3 ℃ min^{-1} to −70 ℃. The vials were then plunged into liquid nitrogen. The precise freezing rate at this point is unknown. The seeding method was that reported by Miyake *et al*. (2004) [16]. Briefly, ice crystals (about 2 mm in diameter) obtained by freezing physiological saline were seeded in suspension at −4 ℃.

The frozen suspension was thawed at a rate of 70 ℃ min^{-1}, which was obtained by keeping the suspension in a water bath at 37 ℃ for 1 min. The thawed larvae were then diluted in PBS by 1:10.

Viability assessment

After thawing, the suspension was observed under a light microscope to check the viability of the NBL and MSL. If thawed NBL showed the same active motility as unfrozen NBL, the parasites were judged viable. In the case of MSL, the cryopreserved larvae were classified into two groups: normal and abnormal. Normal MSL showed active motility without any damage to intestinal organs, while abnormal MSL showed obvious damage to these organs despite their motility.

The infectivity of NBL and MSL were confirmed after cryopreservation. NBL (3,000) were injected into the abdominal cavity of mice, and then on the 30th day post infection, the mice were sacrificed and examined for the presence of MSL. MSL (2,000), on the other hand, were given to ICR mice *per os* or injected into the intestinal duodenum of laparotomized mice. On the 30th day post infection, the mice were sacrificed and examined for the presence of MSL.

RESULTS

Cryopreservation of NBL

Table 1 shows the effects of cryoprotectant type and concentration as well as equilibration time on the survival rate of NBL after thawing. Of the two kinds of cryoprotectant examined, Me_2SO had a higher protective effect than glycerol. When the NBL suspension was kept at 37 ℃ for 30 min (equilibration) prior to freezing, the survival rate of NBL after thawing was higher than that with other equilibration times. The concentration of Me_2SO was also critical for survival with the maximum survival of 96% obtained under conditions of 10% Me_2SO and 30 min equilibration.

Cryopreservation of MSL

Table 2 shows the effects of cryoprotectant type and concentration as well as freezing rate on the survival of MSL after thawing. Glycerol had a higher protective effect

Table 1. Effects of type, concentration, and equilibration time of cryoprotectants on survival rates of NBL

Type of cryoprotectants	Equilibration time (min)	Concentration of cryoprotectants (%)				
		5	10	15	20	25
Me_2SO	15	78±7	69±15	66±7	48±20	25±12
	30	84±15	96±3	93±3	80±11	39±14
	45	47±2	64±11	61±5	49±8	20±1
	60	53±22	54±24	39±6	32±13	12±7
Glycerol	15	15±8	37±7	48±6	8±4	10±6
	30	40±10	55±17	34±12	10±7	23±6
	45	36±14	39±18	58±4	26±8	15±6
	60	52±16	69±18	47±2	35±3	19±3

Cryoprotectants were added at 37℃. After equilibration, the samples were cooled to −40℃ at 1℃ min^{-1} and to −70℃ at 3℃ min^{-1}. Each experiment was performed three times.

Table 2. Effects of cryoprotectant and freezing rates on survival rates of MSL

Type of cryoprotectants	Concentration of cryoprotectants (%)	Freezing rate (℃/min)	No. of MSL examined	Survival	
				No.	%
Glycerol*	10	0.3	147	66	45
	15		165	81	49
	20		189	123	65
	25		180	147	82
	30		186	147	79
Glycerol*	10	1	330	180	55
	15		327	225	69
	20		339	258	76
	25		354	303	86
	30		339	258	76
Glycerol*	10	3	156	75	48
	15		171	111	65
	20		153	123	80
	25		177	159	90
	30		153	123	80
Me_2SO†	5	1	273	21	8
	10		261	39	15
	15		300	39	13
	20		255	38	15

*Equilibration time = three hours.
†Equilibration was not performed.

than Me_2SO; that is, a higher survival rate was obtained with glycerol at a concentration of 25% regardless of the freezing rate used. Freezing rates of 0.3, 1, and 3℃ min^{-1} resulted in maximum survival rates of 82, 86, and 90%, respectively, without significant differences ($P > 0.05$, students t-test).

Figure 1 shows the effects of pre-treatment on the survival rate of MSL after thawing. In this experiment, the following conditions were used: 25% of glycerol as a cryoprotectant, three hours equilibration at 37℃, and a freezing rate of 1℃ min^{-1} between 37 and −40℃. Relatively high survival rates, approximately 80%, were obtained from both the untreated and two pre-treated groups, without significant differences ($P > 0.05$). However, when the appearance of normal larvae was considered, rates differed among the three groups: the bile-treated group showed the highest percentage of normal larvae (63%), while the CTAB-treated and untreated groups showed 51 and 44%, respectively.

Infectivity of NBL and MSL after thawing

Of the five mice infected with cryopreserved NBL, all were positive for MSL on the 30th day post infection. On the other hand, mice orally infected with cryopreserved MSL showed no infectivity regardless of pre-treatment of the larvae. One infected mouse was obtained when MSL were pre-treated with dog's bile and infected directly into the mouse duodenum after thawing (Table 3).

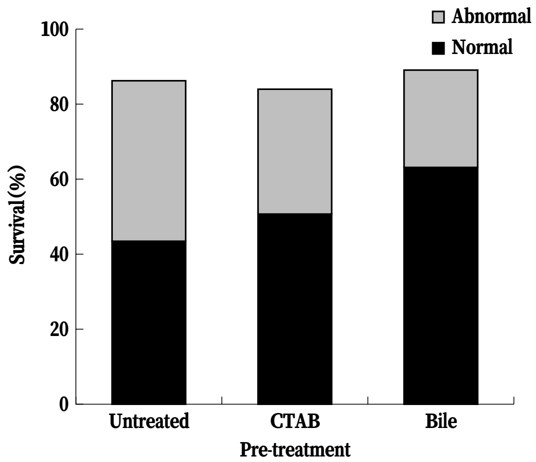

Figure 1. Effect of pre-treatment on survival rates of MSL.

DISCUSSION

Since the first successful cryopreservation of *Plasmodium* spp. in 1939, a number of parasite species have been successfully cryopreserved, including protozoa such as *Toxoplasma gondii* [1], *Entamoeba histolytica* [6], and *Trypanosoma cruzi* [5], as well as helminths such as trichostrongylidae [7], microfilariae of *Dirofilaria corynodes* and *Brugia malayi* [14], and *Toxocara canis* [20]. Some authors have also reported cryopreservation of *Trichinella* spp., although none have obtained satisfactory results [2-4, 11, 19]. Infective MSL of *T. spiralis* have been recovered from polar bear muscle frozen at −15℃ for 12 months [4], wolf muscle frozen at −10℃ for 18 months [3], and arctic fox frozen at −15℃ for 14 months [2]. However, the preservation periods of these experiments were limited to several months. In addition, when the parasites were preserved using laboratory animals such as mice and/or rats, the survival period decreased. Cryopreservation of *Trichinella* spp. at an ultra low temperature has been reported by Gegan and James (1988) [11] and Pozio *et al*. (1988) [19]. Pozio *et al*. (1988) [19] obtained infective NBL of both *T. spiralis* and *T. nativa* when the larvae were cryopreserved using 10% Me_2SO, but MSL cryopreserved using the same method did not show any infectivity to mice. Similarly, Gegan and James (1988) [11] reported that 80% of *T. spiralis* MSL used in their cryopreservation study survived after thawing, but infectivity of these larvae was extremely low. In our study, on the other hand, not only NBL but also MSL of *T. nativa* showed infectivity to mice after thawing, although the survival rate of MSL was low. Our experiment therefore suggests that the infection method is also an important factor in confirming the infectivity of the larvae after thawing. Although we have cryopreserved muscle stage larvae of *T. spiralis*, the survival rate after freezing was always lower than that of *T. nativa* (data not shown). Therefore, this paper represents the results obtained using *T. nativa*.

As reported by Pritchard *et al*. (1985) [17], CTAB-treated larvae lose an excretory/secretory antigen that coats the surface of their body, thus causing an decrease in the susceptibility of these larvae to digestive juice. It seems that a similar phenomenon arises in larvae treated with bile. In the present experiment, we assumed that larvae introduced directly to the duodenum of the recipient mice would not be affected by digestive juices.

In our study, Me_2SO and glycerol were used as cryoprotectants and their effect on the survival of NBL and MSL was investigated. Me_2SO has been used as a cryoprotectant for the cryopreservation of many species of parasites [11, 14-16, 19, 20]. In the present study, the highest survival rate of NBL was obtained with 10% Me_2SO. This result is similar to that of Pozio *et al*. (1988) [19] who reported that Me_2SO at a concentration of 10% yielded 80% motile NBL after thawing. On the other hand, Mutetwa and James (1984) [15] reported that glycerol had similar cryoprotective effects as Me_2SO. With both Me_2SO and glycerol, the protective effect on frozen cells requires intracellular permeation, and the difference between Me_2SO and glycerol is the speed at which they permeate the cell: glycerol permeates more slowly than Me_2SO. Therefore, equilibration is thought to be necessary when glycerol is used as a cryoprotectant [15]. In our study, the duration of equilibration for glycerol was three hours.

Table 3. Infectivity of cryopreserved MSL to mice

Infection method	Pre-treatment	No. of MSL infected	No. of mouse used	No. of mouse showing positive
Per os	Untreated	3,000	5	0
	CTAB	3,000	2	0
	Bile	3,000	2	0
Injection*	Untreated	2,000	1	0
	CTAB	2,000	3	0
	Bile	2,000	2	1

*Injected in the intestinal duodenum in laparotomized mouse.

For MSL, in contrast to NBL, the most effective cryoprotectant was glycerol. Few reports have mentioned that the cytotoxicity of glycerol is less than that of Me_2SO [11, 16]. MSL have a more complicated cellular system than NBL and therefore require a longer equilibration time for cryoprotectant penetration. This might explain why glycerol was more effective with MSL in the present study. Moreover, the cytotoxic effect of glycerol was shown, for the first time, to remain low even when exposed for a longer period of time.

We also examined the effects of freezing rate on MSL survival rates after cryopreservation. Ham and James (1983) [8] reported that the highest survival rate (95%) was obtained with a freezing rate of 5 ℃ min^{-1} compared to 1, 2, and 10 ℃ min^{-1}. In our study, the maximum survival rate of 90% was obtained at a freezing rate of 3 ℃ min^{-1}, followed by 86% at 1 ℃ min^{-1} and 82% at 0.3 ℃ min^{-1}. There were no significant differences ($P > 0.05$) between survival rates despite the different rates used, but the reason for this remains unknown. Therefore, further experiments to determine the optimum freezing rate are needed.

Permeability of cryoprotectants is often a critical factor in the successful cryopreservation of a number of helminth species. *Schistosome schistosomula* cannot be cryopreserved until the cercarial glycocalyx is lost [9], and for several nematode species, excystment of third-stage larvae is essential for survival during cryopreservation [12, 13]. In the case of MSL, facilitating the penetration of cryoprotectants is also an important factor in obtaining high survival rates [11]. Pritchard *et al*. (1985) [17] reported that the use of 0.25% CTAB at 37 ℃ for one hour to remove excretory/secretory antigens can increase surface permeability. In addition, Gegan and James (1988) [11] reported that larvae pre-treated with 10% dog's bile at 37 ℃ for one hour can increase surface permeability. In our study, the effects of pre-treatment with CTAB and bile were examined. When MSL were pre-treated with CTAB and bile, the percentage of normal larvae was higher than with untreated larvae after thawing (Figure 1). In addition, infective MSL were also obtained in bile pre-treatment group (Table 3). From the results obtained, increasing cryoprotectant permeability to prevent damage during cryopreservation of MSL also seems to be an important factor [11].

In summary, the basic conditions of cryopreservation (the type and concentration of cryoprotectant, equilibration time, and freezing rate) of NBL and MSL of *T. nativa* were examined. The cryopreservation of both larvae was possible in liquid nitrogen; however, the infectivity of the MSL remained low even though that of the NBL was high. Further studies to improve the infectivity of MSL are therefore required.

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THE ROLES OF WESTERN BIOMEDICINE AND FOLK MEDICINE IN RURAL SOLOMON ISLANDS: A QUANTITATIVE ANALYSIS OF VILLAGERS' RESPONSE TO ILLNESS

Takuro Furusawa
Accepted 26, May, 2006

Abstract: This study aimed to explore factors determining treatment-seeking behaviors of villagers in a rural Roviana society of the Solomon Islands. Participants (n = 116) were interviewed every evening for 42 days about the occurrence of illnesses and how they had been treated. The study period was divided into two: 22 days during which a nurse was stationed in the village and 20 days when the nurse was absent. As the results show, nurse's presence had a negative effect on traditional folk medicine use (OR = 0.39, [95% CI: 0.21–0.72]). Fever or headache was treated more preferably with biomedicine (3.82 [1.81–8.07] or 6.75 [2.75–16.55], respectively), whereas *putuputu*, an illness with an indigenous etiology, was treated with the latter (34.7 [3.13–384.41]). In addition, biomedicine was preferably used (7.72 [2.65–22.44]) for the treatment of severe illnesses. While folk medicine was used in 40% of all ill person-days, it has likely been partly displaced by effective Western biomedicine. Still, some folk medicine functioned as a culturally indispensable element in treating indigenous illnesses.
Keywords: Folk medicine, Biomedicine, Medical pluralism, International Health, Roviana, Solomon Islands

INTRODUCTION

Indigenous medical systems, hereafter "folk medicine," have been developed in many societies with markedly different characteristics [1, 2]. Folk medicine has undergone changes since coming under the influence of Europeans who introduced Western medicine and discouraged traditional folk medicine [3, 4, 5, 6, 7, 8, 9, 10]. Previous studies in Oceania have reported the existence of medical pluralism, which is defined as coexistence of different medical systems in a single society [11]. For instance, in Melanesian societies, traditional folk medicine is chosen to cure indigenous illnesses or those thought to have supernatural causes, while Western biomedicine is used to treat introduced illnesses and those with non-supernatural causes [4, 9, 12, 13]. On the other hand, the quantitative analysis on this coexistence is still very limited.

Treatment-seeking behaviors might be determined not only by the illness type but also by individual features such as sex and age, availability of medical infrastructure and ecological resources, modernity, and severity of illnesses [4, 14, 15, 16, 17, 18, 19, 20]. Quantitative analyses taking several different factors into account will be able to detect the determining factors and, therefore, make progress in the debate on how Western biomedicine and folk medicine coexist in a single society.

The purpose of this study was: (1) to explore the role of different types of treatment based on daily records of responses to illnesses; and (2) to clarify which and how different factors affect treatment-seeking behaviors of the villagers in a single rural society of Solomon Islands, an area experiencing the rapid Westernization over the last few decades. Special attention was paid to the effect of the type and severity of illnesses, availability of a biomedical professional, patient's sex and age, and socioeconomic status. Multivariate analyses were applied for detection of determinant factors.

PARTICIPANTS AND METHODS

The permission for research was obtained from the Solomon Islands Government (Minister for Education and Training and Director of Immigration Office). At the community level, the chief and elders also consented to the study. The study was performed according to international, national, and institutional rules and was approved by the Ethics Committee of Graduate School of Medicine, University of Tokyo. All of the data presented in this paper were collected after gaining informed consent from the head and other members of participant households.

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Study site

Several traditional practices including herbal treatment and other types of folk medicine are still in widespread use in Solomon Islands [3, 5, 21, 22, 23]. Christian missions initially introduced Western biomedicine and other services, and the government later took over the supply and support of these services [21, 24]. However, their availability is still limited in rural communities. In particular, there is a lack of Western biomedical professionals (e.g., medical doctors and registered nurses) [25]. Registered nurses are not stationed throughout the year in village aidposts, which are the most accessible facilities for the residents of rural areas.

The fieldwork on which this paper is based was conducted from January to October 2001 in Olive village, an inner-Roviana-Lagoon village located in the southern part of New Georgia Island, Western Province. Olive village belongs to Saikile Customary Land, in which four villages and several small hamlets are under the Saikile Paramount Chief today [26, 27]. The inhabitants were 189 males and 158 females belonging to 49 households in February 2001. All villagers were engaged in horticulture of tuberous crops and fishing.

The first Christian mission arrived in Roviana in 1902, and most of the people had been converted by the mid 1920s. Western biomedicine came slowly to inner-Roviana villages including Olive. Establishment of a provincial aidpost equipped with a variety of biomedical drugs opened the way for most villagers to be treated with biomedicine by the 1980s. The nurse, however, did not always stay in the village. For instance, a registered nurse left in November 2000 and another arrived in July 2001. At the time of the study, a small fee (approximately 1 US$) was required to consult the nurse, although garden crops could be substituted as payment. There was a mission hospital with modern facilities in Munda (about 30 km west of Olive) and a better-equipped national hospital in Gizo, the provincial capital, about 80 km west of the village. Although medical treatment at the two hospitals in the province was free or inexpensive, the villagers seldom visited them because of the difficulty of transportation.

Recording responses to illnesses

For the interview survey concerning treatment of illnesses, 14 households (65 males and 51 females, accounting for 33.4% of the total population) were sampled. The participants included a proportionate number of villagers from each age group and sex. Each member of the participant household (or the parent for small children) was interviewed by the author every evening for 42 days about the occurrence of illness experienced over the past 24-hour period and how it had been treated. It is noted that the daily interview was advantageous to record all cases however minor (e.g., small wounds) and to avoid dropouts. On occasions where the participant was absent, the interview was conducted the following evening or later.

The names of illnesses reported by the participants were initially recorded in the Roviana language. The author later identified the symptom(s) related to each illness, referring to discussions with a group of healers (*tie sinalaṉa* in Roviana language) comprised of six males and three females. It should be noted that single symptoms (e.g., fever, cut/wound, and cough) or pain/ache in a specific part of body are called a ‘simple illness,’ while an illness with two or more symptoms included in a name (e.g., malaria) is referred to as a ‘complex illness’ in this study (see Table 1).

In this paper, both treatment (*sinalaṉa* in Roviana language) and no treatment for an illness are considered a ‘response.’ The number of person-days during the 42-day period was used as the basic unit of analysis. No nurse was stationed in the village for the first 20 days of the study period, while a nurse was present at the aidpost for the last 22 days. Therefore, the whole study period was divided into two periods.

Responses to an illness could be broadly divided into three categories: (1) Western biomedicine, (2) folk medicine, and (3) no treatment. The Western biomedicine was divided into two subcategories. The first referred to curing behaviors with modern drugs (*meresena vaka* in Roviana language), which had been purchased at stores in town or obtained at the hospital or aidpost in previous cases of illness and kept in the patient’s house (hereafter called ‘modern drug use’). The second was treatment following consultation with the nurse at the aidpost (*vetu moho*) and using modern drugs (consulting a nurse); some treatment was carried out for every consultation. An important characteristic of consulting a nurse is that modern-drug prescriptions based on Western biomedical standards and injections such as penicillin are available, while biomedical check-ups for malaria and other illnesses, i.e. blood test, were not available at the aidpost. Treatments using folk medicine were also divided into two subcategories. One was the use of herbal medicine (herbal medicine use) and the other was massage (*mono*) including acupressure. It should be noted that the study village also has a tradition of treatment using shells and other faunal resources, but these practices were not observed during the study period.

Interviews on individual and household-level characteristics

Each participant or a parent was asked for his/her age and sex. Male and female household heads (e.g., male head and his spouse and widowed female head and her first son)

Table 1. List of English illness names used in this study and the corresponding Roviana names (asterisk indicates illnesses recognized as fatal or severe)[a]

English name	Roviana names
Simple illness	
Chest ache	*Sigiti raqaraqa*
Cough	*Kohu*
Cut/wound	*Bakora*[b], *poraka**
Diarrhea	*Malohoro tia*[c], *huru**
Eye (inflammation)	*Mata kare*
Fever	*Diana (manini)*[d], *nunuale**, *rarabuana**
Headache	*Sigiti batu*
Muscle ache	*Sigiti tini*[e], *sigiti mudi*, *sigiti lima*, *sigiti avara*
Rhinitis	*Puna*
Skin disorder	*Moqo*[f]*, *tubu*, *tini hikare*
Sore throat	*Sigiti rua*
Stomach ache	*Sigiti tia*
Toothache	*Sigiti livo*
Vomiting	*Lua*[g]*, *iqolo*, *bekaha*
Complex illness	
Illness with imported name	
Malaria	*Malaria hokara*[h], *malaria tia*
Pneumonia	*Niumonia*
Illness with folk etiology	
N. A.	*Putuputu (kolo)*, *tuku ibibu*, *lagu hite*, *popome*, *mateana tia (tia hikare)*

[a] Classifications of illness and severity were based on the group discussion of healers (*tie sinalana*).
[b] *Bakora* means cut/wound generally, while *poraka* means lacerated wounds recognized by the villagers as severer than the former.
[c] *Malohoro tia* is generally referred to as diarrhea, while *huru* means watery, probably bacterial, diarrhea.
[d] Both *diana* (literally 'cold') and *manini* ('hot') refer to the same condition of fever, while *nunuale* refers to shaking chills and *rarabuana* to 'paralysis' recognized by villagers as the result of high fever.
[e] Aches in whole body (*tini*), arm (*lima*), back (*mudi*), and shoulder (*avara*) were classified as muscle ache.
[f] *Tubu* and *tini hikare* refer to pimples and chronic dermatitis, respectively, and *moqo* to inflamed or ulcerated ones.
[g] *Lua* refers to vomiting, *iqolo* to nausea and *bekaha* to nausea with abdominal complaints.
[h] *Malaria hokara* literally means 'genuine malaria,' while *malaria tia* means 'abdominal malaria.'

were asked about their place of birth, age, education, experience of living in towns, and experience of waged job. To ensure accuracy, both household heads confirmed the information. Dates of births were verified by referring to the written record, or using local history of well-known events such as Independence Day and the date logging operations in this area commenced.

Statistical analyses

Logistic regression analyses were performed so as to determine which factors influenced the villagers' responses to illnesses. Factors included in the models were (A) type of illness, (B) severity: (b1) the number of days counted from the onset of illness and (b2) recognized severity (the name of illness which is recognized as severe = 1, others = 0), (C) availability of nurse (during the period of nurse's presence = 1, during his absence = 0), (D) individual characteristics: (d1) age and (d2) sex, (E) household-level socioeconomic characteristics: (e1) type of house (Western-style permanent house = 1, others = 0), (e2) age, (e3) education history (educated in secondary or higher school = 1, others = 0), (e4) place of birth (Saikile = 1, others = 0), (e5) experience of living in town (yes = 1, no = 0), and (e6) experience of waged labor (yes = 1, no = 0) of household head.

These procedures were performed using the SAS for Windows version 9.1 (SAS Institute Inc., Cary, NC). A statistically significant association was assumed at $p<0.05$.

RESULTS

Occurrence of illness

Fifty-eight males (89.2%) and 39 females (76.5%) reported at least one episode of illness during the 42-day period. The total number of person-days in which responses were observed (including "no treatment") was 450 out of 4872 person-days (116 persons times 42 days). Out of the 450, two illnesses were recorded for 131 person-days, and three illnesses were recorded for 21 person-days. No case involved four or more illnesses simultaneously. Most illness episodes ended rapidly; 66.1% of cases occurred in only one day and 89.6% lasted for three days or less. None of the participants visited a hospital in town.

Table 1 shows the illness names in Roviana and the corresponding English names. The latter are sorted by alphabetical order. In total, 33 types of illness were observed, although it should be noted that none were diagnosed with a biomedical check-up. Out of these, 15 illnesses corresponded to single symptoms (i.e., three types of fever, rhinitis, cough or sneeze, three types of vomiting, two types of diarrhea, three types of skin disorders, and two types of cut or wound) and seven to aches (*sigiti* in the Roviana language) in specific parts of the body (e.g., head, stomach, chest, throat, and tooth). Aches in the arm, shoulder, back, and whole body were classified as muscle ache. Roviana names such as *nunuale* or *rarabuana*, *huru*, *lua*, *moqo*, and *poraka* refer to fatal or severe conditions of fever (*diana*), diarrhea (*maloholo tia*), vomiting (*iqolo/bekaha*), skin disorders (*tini hikare*/*tubu*), and cuts (*bakora*), respectively.

Roviana names, such as *malaria* and *niumonia*, the words for malaria and pneumonia borrowed from English or Pidgin (the lingua franca of the Solomon Islands), were used. *Malaria* consists of two different types: *malaria hokara* (literally, "genuine malaria") and *malaria tia* ("abdominal malaria"). The former is associated with fever and shaking with chills, whereas the latter does not show a fever symptom but has complicated abdominal disorders diagnosed by palpation. Both illnesses were believed by the villagers to be caused by malaria *nokinoki* ("germs"). *Niumonia* showed fever and respiratory disorders (e.g., cough, chest pain, and sore throat).

It was difficult to determine the English equivalents of five illnesses, i.e., *putuputu*, *tuku ibibu*, *lagu hite*, *popome*, and *mateana tia*, because of their complicated natures. *Putuputu* (also called as *kolo*) showed combined symptoms of abnormal heartbeat, abnormal ventilation, sleep disorder, fatigue, epigastric pain, and nausea. This illness was believed to be caused by problems with blood flow: several respondents said that it was caused by hypertension. *Tuku ibibu* means literally "the pit of the stomach is closed" and features not only pain in the pit of the stomach but also abnormal respiration and anorexia. *Popome* was regarded as an illness in which the lung was rotten or damaged by an ulcer. Both *lagu hite* and *mateana tia* (also called *tia hikare*) were referred to as disorders of the intestinal organs, though the former and latter were usually associated with the appendix and liver, respectively. These five illnesses were categorized as illnesses with indigenous names and etiologies in further analyses of this study.

Patterns of treatment

Figure 1 shows participants' responses to illnesses by absence (207 person-days) and presence (243 person-days) of the nurse. Throughout the two periods, the most frequent response was modern drug use (45.9% and 38.7% in the absence and presence of nurse, respectively), followed by herbal medicine use (29.0% and 27.8%). Consulting the nurse accounted for 17.8% of person-days during the presence of the nurse. Massage was used in 13.3% and 9.9% of treatment during the absence and presence of nurse, respectively (accompanied by other types of treatment in about a half of cases). No treatment was observed in approximately 18% during both periods. The total frequency of Western biomedicine use (45.9%) and folk medicine use (40.1%) was nearly equal during the period of the nurse's absence, but the former (51.9%) increased and the latter (32.9%) decreased during the period of the nurse's presence.

Table 2 shows all illnesses that occurred during the study period. Cases combining two or more illnesses simultaneously were counted for all and those undergoing two types of treatment were also counted for both. Illnesses were sorted by the order of the number of person-days of modern drug use. Three significant observations were made

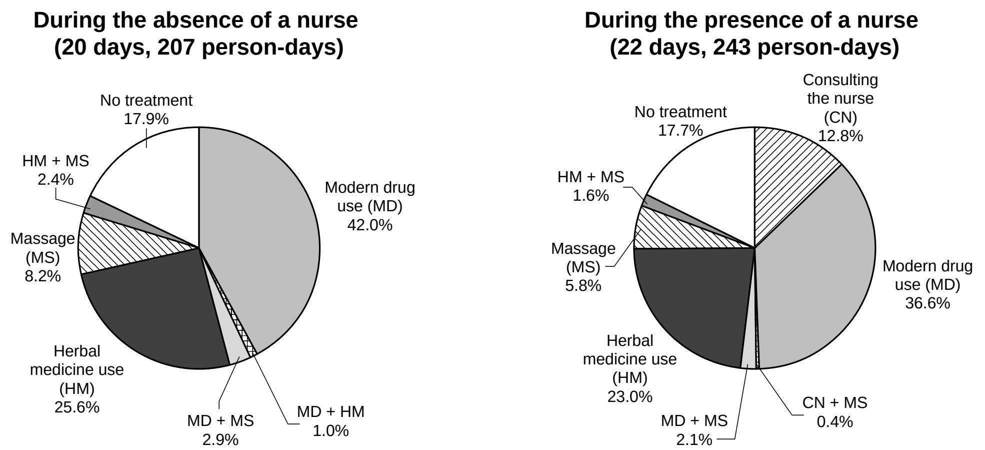

Figure 1. Responses by type of treatment during the period of a nurse's presence in the village (left) and that of the nurse's absence (right)

Table 2. Responses to illnesses occurring during the study period[a, b]

	Western biomedicine			Folk medicine			No treatment	Total
	Modern drug use	Consulting a nurse	*Total*	Herbal medicine use	Massage	*Total*		
Simple illness								
Fever	67	8	*75*	1	9	*10*	8	93
Headache	48	9	*57*	0	5	*5*	8	70
Muscle ache	27	6	*33*	7	7	*14*	16	63
Rhinitis	24	2	*26*	0	2	*2*	4	32
Cough	23	4	*27*	6	2	*8*	3	38
Stomach ache	20	1	*21*	11	5	*16*	4	41
Vomiting	12	0	*12*	5	4	*9*	2	23
Chest ache	11	2	*13*	2	3	*5*	0	18
Toothache	5	0	*5*	0	0	*0*	1	6
Cut/wound	5	3	*8*	20	2	*22*	19	49
Sore throat	4	2	*6*	0	5	*5*	3	14
Diarrhea	4	1	*5*	5	0	*5*	1	11
Skin	3	4	*7*	57	14	*71*	9	87
Eye	1	1	*2*	0	0	*0*	1	3
Complex illness								
Illness with imported name								
Malaria tia	15	0	*15*	1	1	*2*	1	18
Malaria hokara	5	2	*7*	0	0	*0*	0	7
Niumonia	11	0	*11*	0	2	*2*	1	14
Illness with folk etiology								
Lagu hite	4	0	*4*	1	1	*2*	0	6
Putuputu	3	0	*3*	9	7	*16*	0	19
Tuku ibibu	2	3	*5*	0	0	*0*	0	5
Popome	1	0	*1*	2	0	*2*	0	3
Mateana tia	0	0	*0*	4	1	*5*	0	5
Total	295	48	*343*	131	70	*201*	81	625

[a]Cases where two or more illnesses occurred simultaneously were counted for all and those induced more than one responses were also counted for both.
[b]Illnesses were sorted by the order of the number of person-days for modern drug use.

regarding the association between illness and response. First, Western biomedicine, especially modern drug use, was preferred in cases of headache, fever, cough, and rhnitis whereas folk medicine was preferred for skin disorders and wounds. Abdominal pain and diarrhea were treated either with Western biomedicine or folk medicine. Second, illnesses with names and etiologies introduced from abroad (*malaria hokara*, *niumonia*, and *malaria tia*) were treated mainly with Western biomedicine. *Malaria hokara* was treated only with modern drug use. Although not shown in the table, all treatments using herbal medicine or massage as responses to *niumonia* and *malaria tia* were accompanied simultaneously by the use of Western biomedicine. Third, among the illnesses with indigenous names and etoiologies, *putuputu*, *popome*, and *mateana tia* were mainly treated by folk medicine and *lagu hite* and *tuku ibibu* by Western biomedicine.

Factors determining an individual's response

Western biomedicine was used in 48.7% and 50.0% of treatments for males and females, respectively, and folk medicine in 36.9% and 34.9%, respectively. 'No treatment' was observed in 17.8% and 18.4% of treatments for males and females, respectively. Overall, there was no association between responses and sex.

Figure 2 shows the responses broken down by age groups (top) and by different days from the onset of illness (bottom). Western biomedicine was used for the treatment of infants, children, and younger adults accounting for approximately 50% of person-days, while the most frequent use was observed in the 30-60 year-old group (64.7%) and the lowest among elders (8.3%). Folk medicine was used most frequently for the treatment of elders (87.5%), followed by that of infants (45.2%), but less in other age groups (range: 23.4 to 29.3%). The proportion of no treatment was low in the age groups of infants and elders who are generally vulnerable to disease.

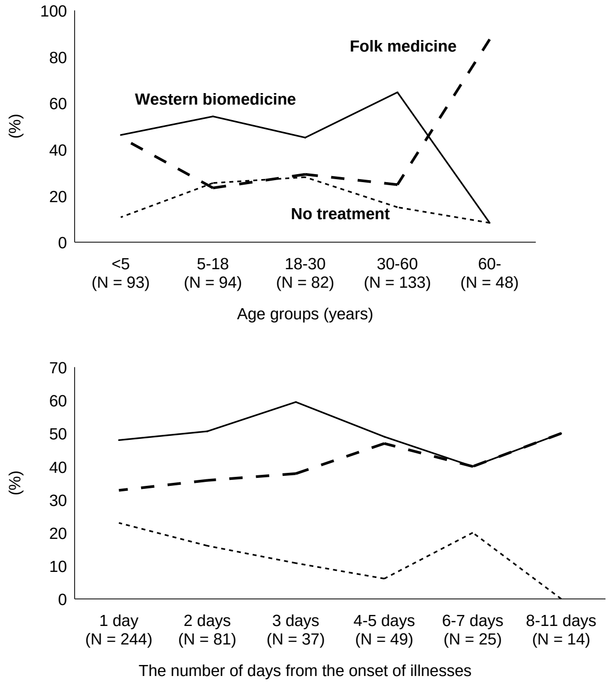

Figure 2. Frequency of response choices in different age groups (top) and different days from the onset of illness (bottom). The number of person-days is shown in parenthesis.

The frequency of Western biomedicine use reached a peak (59.5%) on the third day from the onset of illness, while that of folk medicine use peaked (46.9%) on the fourth day. The frequency of both treatments decreased at the sixth-to-seventh day and then increased again at the eighth-to-eleventh day, and folk medicine was used most frequently (50.0%) in this class of days. The proportion of no treatment was highest on the first day (23.0%) and then constantly decreased, although a second peak (20.0%) appeared at the sixth-to-seventh day.

Logistic regression analysis was used to explore the determining factors for responses as shown in Table 3 (only those factors with significant effects were shown). First, participants aged 60 years or more used Western biomedicine less frequently (OR (odds ratio) = 0.14 [95% CI (confidence interval) = 0.03, 0.76]) than other participants. Secondly, with regard to socioeconomic status, only the type of house (living in Western-style permanent house) had a positive effect on biomedicine use. Socioeconomic status did not have an effect on folk medicine. Third, the effect of the nurse's presence was negative for folk medicine use (OR = 0.39 [0.21, 0.72]). Fourth, Western biomedicine was used more frequently on the third day of illness than other days. Fifth, Western biomedicine was used more frequently (OR = 7.72 [2.65, 22.44]) and folk medicine less frequently (OR = 0.16 [0.06, 0.48]) for the treatment of severe illness.

Sixth, regarding illnesses, patients with fever or headache were treated more frequently with Western biomedicine (OR = 3.82 [1.81, 8.07] or 6.75 [2.75, 16.55], respectively) and less frequently with folk medicine. Cuts/wounds or skin disorders were less likely to be treated with Western biomedicine (OR = 0.23 [0.07, 0.78] and 0.13 [0.05, 0.52], respectively) but were not related with the use of folk medicine. *Putuputu*, the illness with a folk etiology, was a positive factor for folk medicine treatment (OR = 34.7 [3.13, 384.41]).

Discussion

The Role of folk medicine

In the community examined in the present study, folk medicine was used in approximately 40% of all ill person-days. There have been few quantitative studies on the practice of traditional folk medicine in Melanesia and other indigenous societies. A previous study based on fortnightly interviews in Huli, a region of the Papua New Guinea Highlands where Christianity and European culture had arrived in the 1950s, showed that folk medicine was used rarely, accounting for only 4% of all ill person-days in the 1970s [4]. Even taking into account the fact that the interview period was different from this study, it is judged that folk medicine has played a greater role in Roviana society than in Papua New Guinea Highlands.

The results of logistic regression analysis revealed the factors involved in the use of folk medicine. Firstly, the fact that none of the socioeconomic variables was related to the use of folk medicine was important because it indicates the familiarity of folk medicine as an indigenous traditional practice among villagers. It should be noted, however, that the preferred use among elders might reflect a strong cultural affinity continuing mainly in this age group while younger generations are more likely to use Western biomedicine than folk medicine.

The analyses also revealed that folk medicine was preferred during the absence of the nurse and in the treatment of less severe cases. These results suggest that folk medicine serves as an alternative to Western biomedicine and that it is less important than Western biomedicine. This assumption is supported by the other result that both the nurse and the villagers themselves turned to Western biomedicine instead of folk medicine in cases of severe illness.

One of the most important findings, on the other hand, was that folk medicine was used for the treatment of *putuputu*, an illness with an indigenous name and etiology. Several informants believed that Western biomedicine had no effective way to deal with this illness whereas their own medical system did. This belief is thought to have deter-

Table 3. Factors for choosing Western biomedicine or folk medicine, determined by logistic regression analysis (odds ratio and 95% confidence limit in parentheses)[a]

	Western biomedicine (450 person-days)		Folk medicine (450 person-days)	
Individual characteristics				
Age group (years)				
60- (yes = 1; no = 0)	0.14	(0.03, 0.76)	6.27	(1.30, 30.13)
Household socioeconomic status				
House (permanent = 1; leaf = 0)	3.70	(1.11, 12.33)		
Aidpost (open = 1; closed = 0)			0.39	(0.21, 0.72)
Severity				
Number of day				
3rd day (yes = 1; no = 0)	2.91	(1.02, 8.31)		
Class (severe = 1; else = 0)	7.72	(2.65, 22.44)	0.16	(0.06, 0.48)
Illness				
Simple illness				
Fever (yes = 1; no = 0)	3.82	(1.81, 8.07)	0.23	(0.08, 0.61)
Headache (yes = 1; no = 0)	6.75	(2.75, 16.55)	0.26	(0.08, 0.84)
Cut/wound (yes = 1; no = 0)	0.23	(0.07, 0.78)		
Puna (yes = 1; no = 0)			0.16	(0.03, 0.92)
Skin (yes = 1; no = 0)	0.13	(0.03, 0.52)		
Complex illness				
Putuputu (yes = 1; no = 0)			34.70	(3.13, 384.41)
Wald statistics	116.55		102.29	
P	<0.0001		<0.0001	

[a] Variables with significant effects were shown; other variables included in the models were:
(a) Individual characteristics:
Sex (male = 1), 5-18-year-old, 18-30-year-old, and 30-60-year-old age groups,
(b) Household socioeconomic status:
Birthplace of household head (Saikile = 1; others = 0), age of household head, household head's education higher than primary school (yes = 1; no = 0), household head's experience of living in town (yes = 1; no = 0), household head's experience of waged job (yes = 1; no = 0)
(c) Characteristics of illness:
All illnesses shown in Tables 1 and 2 (yes = 1; no = 0).

mined their treatment-seeking behavior and suggests that the villagers continue to need folk medicine in addition to newly introduced biomedicine. Overall, most of the folk medical practices will likely be displaced by effective Western-biomedical alternatives, but others, although limited in number, persist as culturally indispensable treatments for indigenous illnesses.

The Role of Western biomedicine

In general, Western biomedicine was used preferably for the treatment of headache, fever, and introduced illnesses (i.e., malaria and pneumonia), although the effect of the introduced illnesses disappeared when other factors were adjusted. According to the respondents, medical personnel and schoolteachers discouraged the use of folk medicine for treatment of malaria and pneumonia because these illnesses can be fatal and instead recommended the use of Western biomedicine as a powerful measure. In fact, the villagers believed that almost all deaths in the village were caused by these illnesses even though few of them had had a biomedical check. Related illnesses such as fever and headache were therefore treated preferably with biomedicine. "Abdominal malaria (*malaria tia*)" was treated exclusively with biomedicine because of the villagers' recognition that it is caused by a kind of "germ" which they described as being transmitted by flies. This indicated some confusion regarding the cause of the illness. The Malaria Control Programme has been the most intensively promoted health program in Solomon Islands, but it seems to have caused confusion regarding the cause of malaria. Indeed, misunderstandings as to malarial etiologies have also been reported in medical-sociological research in Malaita Province [29].

In the logistic models, housing type and severity of illness were also factors for the use of biomedicine. It is reasonable that Western biomedicine was preferably used in the households with a permanent Western-style house since both correlate with modernity. The fact that biomedicine was used more frequently in severer cases and in the later days of illness suggested that the villagers in the study community recognize that biomedicine wields stronger curing power than folk medicine.

Coexistence of folk medicine and Western biomedicine

Ethnographic studies have pointed out the existence of medical pluralism, which is defined as the coexistence of different medical systems, in Melanesian societies [4, 9, 30, 31]. The authors argue that traditional folk medicine and Western biomedicine are complementary rather than competitive. The patterns of the community in the present study where Western biomedicine is used for the treatment of illnesses originating from other areas and folk medicine for *putuputu*, were similar to that of previous reports [4, 9, 12, 13]. It is thus judged that Roviana society, like other societies [4, 30], is receptive to innovations that are readily available and effective, while maintaining useful indigenous technologies.

This study, on the other hand, reveals the complicated nature of treatment choice as follows. First, the fact that *tuku ibibu*, an illness with an indigenous name and etiology, was treated exclusively with Western biomedicine rather than folk medicine (Table 2) indicated that medical systems and illnesses should be recognized, not as dichotomous, but as dynamic or plural. Second, the fact that the villagers' behavior was strongly affected by the availability of a nurse indicated that the role of both Western biomedical and folk medical systems is dependent on availability of a biomedical professional even within a single society. In conclusion, this quantitative analysis supported the existence of medical pluralism in a rural society of the Solomon Islands, but suggested that this plurality is dynamic and subject to change depending on the availability of a biomedical infrastructure.

Implications for International Health

The results of this study, which showed how villagers accepted introduced biomedicine and often misunderstood it while continuing to use their own folk medicine, are useful from the viewpoint of international health. For instance, the improvement of the biomedical infrastructure by the continuous presence of a nurse, the appropriate supply of drugs, and the provision of regular medical check-ups for malaria is an essential prerequisite for health promotion. In the process, however, the appropriate dosage and use of drugs should be monitored because villagers often misunderstand Western biomedical notions and because the improper use of drug is potentially fatal [18, 31].

At the same time, from the viewpoint of encouraging primary health care and conserving indigenous knowledge, the effectiveness of folk medicine should also be positively re-evaluated [2, 32]. Although traditional folk medicine has undergone changes due to governmental health education and policies, the Olive villagers still conserve knowledge of their own medicine and practice it at a high frequency. Since the biological effectiveness of some herbal plants found in the Solomon Islands has already been recognized [33, 34], the appropriate use of folk medicine can help to reduce the cost of medical care. Moreover, when traditional knowledge is disappearing quickly in the world, societies still using folk medicine should be encouraged from the viewpoint of the conservation of indigenous environmental knowledge [35].

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