Surveillance and Vector Control of Lymphatic Filariasis in the Republic of Korea

Shin Hyeong Cho, Da Won Ma, Bo Ra Koo, Hee Eun Shin, Wook Kyo Lee, Byong Suk Jeong, Chaeshin Chu, Won Ja Lee, Hyeng Il Cheun

Division of Malaria and Parasitic Diseases, Korea National Institute of Health, Osong, Korea.
Division of Medical Entomology, Korea National Institute of Health, Osong, Korea.
Division of Epidemic Intelligence Service, Korea Centers for Disease Control and Prevention, Osong, Korea.

Abstract

Objectives: Until the early 2000s, lymphatic filariasis would commonly break out in the coastal areas in Korea. Through steady efforts combining investigation and treatment, filariasis was officially declared eradicated in 2008. This study surveyed the density of vector species of filariasis in past endemic areas, and inspected filariasis DNA from collected mosquitoes for protection against the reemergence of filariasis.

Methods: Between May and October 2009, mosquitoes were caught using the black night trap in past endemic coastal areas: Gyeongsangnam-do, Jeollanam-do, and Jeju-do. The collected mosquitoes were identified, and the extracted DNA from the collected vector mosquitoes was tested by polymerase chain reaction for Brugia malayi filariasis.

Results: Ochletotatus togoi, Anophel es (Hyrcanus) group, Brugia malayi filariasis, Ochletotatus togoi, and Culex pipiens were most frequently caught in Jeollanam-do (Geumun Island, Bogil Island, Heuksan Island), Jeju-do (Namone-ri, Wimi-ri), and Gyeongsangnam-do (Maemul Island). DNA of B malayi was not found in Och Togoi and An (Hyrcanus) group as main vectors of filariasis.

Conclusion: Lymphatic filariasis was not found in the vector mosquitoes collected in past endemic areas. However, considering that the proportion of vector species is quite high, there is a potential risk that filariasis could be reemerging through overseas travel or trade. Thus, there is a need to continuously monitor vector mosquitoes of lymphatic filariasis.

KEYWORDS: Anopheles (Hyrcanus) group, Brugia malayi filariasis, Ochletotatus togoi, vector mosquitoes

Received: July 3, 2012
Revised: July 19, 2012
Accepted: July 19, 2012

*Corresponding author.
E-mail: ilcheun7@korea.kr

This is an Open Access article distributed under the terms of the Creative Commons Attribution Non-Commercial License (http://creativecommons.org/licenses/by-nc/3.0) which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

Copyright © 2012 Korea Centers for Disease Control and Prevention. Published by Elsevier Korea LLC. All rights reserved.
1. Introduction

Lymphatic filariasis is a vector-borne nematode infection caused by Wucherea bancrofti, Brugia malayi and Brugia timori, and more than 100 million people are affected worldwide [1,2]. Of this total, 90% are caused by W bancrofti and 10% by B malayi [3]. The first case of elephantiasis in Korea was reported in 1927 [4], and it was called by various names: Soojongdari (leg dropy), Pinaerim (blood down flow), Pijoeng (blood diseases), and Gakmomsal (malaise with arthralgia) [5]. In the 1940s, the infection rate reached as high as 12–26% in the southern parts of the country and Jeju-do [6—8]. The rate remained at a 12% average until the early 1970s (range, between 5.5% and 18.0%) in the more inland areas of Youngju-si (si = district), Gyeongsangbuk-do (do = province). Following the government’s active efforts for treatment, the disease was eliminated in the late 1980s [9—11]. By the mid-1980s, the average rate fell below 1% even in Jeju-do where the infection was most prevalent [12]. Furthermore, no positive cases of filariasis were reported in past endemic areas of Jeollanam-do, Gyeongsan-do, and Jeju-do between 2002 and 2006 [13]. Finally, the World Health Organization verified that filariasis was eliminated in Korea, declaring the country free from lymphatic filariasis [13—15]. However, monitoring of vector mosquitoes should continue to prevent possible reemergence due to increasing overseas travel and climate changes. In this study, we investigated the density and distribution of vector mosquitoes of filariasis, and its DNA from vector mosquitoes was tested.

2. Materials and Methods

2.1. Survey areas

Jeollanam-do [Sa-ri (ri = village), Heuksan Island in Shinan-gun, Geomun-ri, Geumun Island in Yeosu-si (si = district), Baekdo-ri, Bogil Island in Wando-gun], Gyeongsangnam-do (Maejuk-ri, Maemul Island in Tongyoung-si), and Jeju-do (Island) (Wimi-ri, Seoguipo-si) were surveyed (Figure 1).

2.2. Adult mosquito surveillance

Adult mosquitoes were collected twice weekly using the commercial Black Hole® mosquito black light trap (Model: Black Hole, Bio-trap Inc., Seoul, Korea). The traps were placed under the eaves of houses from 19:00 to 06:00 hours the following morning. All mosquitoes were identified to the genus or species level under a dissecting microscope using standard morphological keys [16,17]. An sinensis was assigned to the Anopheles (Hyrcanus) group, because of the difficulty in identifying the species by microscope.

2.3. DNA extraction and PCR condition

The selected Och togoi and An (Hyrcanus) Group were pooled to 1–3 mosquitoes, and total DNA was extracted with the QIAamp DNA kit according to the manufacturer’s protocol (Qiagen, Valencia, CA, USA). Hha1 F 5′-
GGCGATAAATTCATGCAGC-3', R 5'-GGGAAAAAC TTAATTACAAAAGC-3' of two set primers were used [18]. Polymerase chain reaction (PCR) was performed in a 25-μL reactive solution containing 5 μL of DNA template, 20 pmol of each primer, 1.25 U of ExTaq DNA polymerase (Takara Co., Japan), 2 mM MgCl₂, and 250 μM of each dNTP. The following PCR cycle was performed in an iCycler thermal cycler (Bio-Rad Ltd., Hercules, CA, USA): 1/C°C for 5 minutes, 40/C°C for 1 minute, 56°C for 1 minute, 72°C for 1 minute ) ,
1/2 72°C for 10 minutes.

3. Results

3.1. Sa-ri, Heuksan Island, Shinan-gun

A total of 149 mosquitoes were collected in Heuksan Island, consisting of four genera and six species. Among them, Och togoi was the most common (62.4%), followed by Culex pipiens (16.8%), Armigeres subalbatus (13.4%), Culex tritaeniorhynchus (6%), Culex bitaeniorhynchus (0.7%), and An (Hyrcanus) Group (0.7%) (Table 1). However, filariasis DNA was not detected in the vector species of Och togoi and An (Hyicanus) group.

3.2. Baekdo-ri, Bogil Island, Wando-gun

In Bogil Island, 3097 mosquitoes were collected in seven genera and 12 species. Och togoi was the most frequent (84.1%), followed by An (Hycanups) Group (5.3%), Cx tritaeniorhynchus (4.1%), Ar subalbatus (2.5%), Cx pipiens (1.3%), Och dorsalis (1.1%), An sinesis (1.0%), Coh ochracea (0.3%), Och nipponicus (0.1%), Ae vexans (0.03%), and Lutrie vorax (0.03%). The number of collected Och togoi rose sharply between May and June, and remained high until September. Neither Och Togoi nor An (Hyicanus) group had filariasis DNA (Table 2).

3.3. Geomun-ri, Geomun Island, Yeosu-si

A total of 376 mosquitoes were collected in Geomun-ri, Geomun Island, comprising five genera and eight species. Among them, the number of Cx pipiens was highest (45.2%), followed by Och togoi (12%), Arm subalbatus (4.8%), Ae albopirtus (0.8%), Cx inatomi (0.3%), Och koreicus (0.3%), and Lu vorax (0.3%) (Table 3). Filariasis DNA was not found in Och togoi and An (Hyicanus) group.

3.4. Maejuk-ri, Maemul Island, Tongyoung-si

A total of 1213 mosquitoes were collected in Maejuk-ri, Maemul Island, comprising of four genera and seven species. Och togoi was most frequently caught (84.9%), followed by Ar subalbatus (12%), Cx pipiens (2.1%), Cx triraenior (0.5%), Ae albopirtus (0.3%), Och koreicus (0.2%), and Cx bitaenior (0.1%). The number of Och togoi picked up sharply between May and June, and dropped significantly between

Table 1. Seasonal variations of mosquitoes collected in mosquito trap in Sa-ri, Heuksan Island (latitude 34°39’N, longitude 125°25’E), Shinan-gun, Jeollanam-do in 2009

| Month | An (Hyicanus) group | Arm subalbatus | Cx Pipiens | Cx Tritaenior | Och togoi | Mean no./trap |
|-------|---------------------|----------------|------------|--------------|-----------|--------------|
| May   | 1                   | 1              | 3          | 4            | 19        | 31           |
| Jun   | 30                  | 5              | 3          | 10           | 318       | 366          |
| Jul   | 23                  | 16             | 1          | 6            | 39        | 12           | 737          | 835         |
| Aug   | 106                 | 21             | 9          | 16           | 77        | 9            | 2            | 1,005       | 1,245       |
| Sep   | 1                   | 31             | 35         | 13           | 12        | 1            | 527          | 1           | 620         |
| Total | 1 (0.03) 164 (6.3)  | 78 (2.5) 9 (0.3) | 1 (0.03) 41 (1.3) 128 (4.1) 35 (1.1) | 4 (0.1) 2,606 (84.1) 1 (0.03) 3,097 |

Numbers in parenthesis represent data on male mosquitoes.

Table 2. Seasonal variations of mosquitoes collected in mosquito trap in Baekdo-ri, Bogil Island (latitude 34°08’N, longitude 126°32’E), Wando-gun, Jeollanam-do, in 2009

| Month | An (Hyicanus) group | Arm subalbatus | Cx Pipiens | Cx Tritaenior | Och togoi | Mean no./trap |
|-------|---------------------|----------------|------------|--------------|-----------|--------------|
| May   | 4                   | 1              | 3          | 4            | 19        | 31           |
| Jun   | 30                  | 5              | 3          | 10           | 318       | 366          |
| Jul   | 23                  | 16             | 1          | 6            | 39        | 12           | 737          | 835         |
| Aug   | 106                 | 21             | 9          | 16           | 77        | 9            | 2            | 1,005       | 1,245       |
| Sep   | 1                   | 31             | 35         | 13           | 12        | 1            | 527          | 1           | 620         |
| Total | 1 (0.03) 164 (6.3)  | 78 (2.5) 9 (0.3) | 1 (0.03) 41 (1.3) 128 (4.1) 35 (1.1) | 4 (0.1) 2,606 (84.1) 1 (0.03) 3,097 |

Numbers in parenthesis represent data on male mosquitoes.
September and October (Table 4). Filariasis DNA was not found in Och togoi and An (Hyicanus) group.

3.5. Namone-ri, Seoguipo-si

A total of 247 mosquitoes were collected in Namone-ri, Seoguipo-si, Jeju-do, comprising two genera and four species. Cx pipiens was most frequently caught (81.8%), followed by Och togoi (13%), Cx tritaenior (4.9%), and Cx mineficus (0.4%). No filariasis DNA was detected in Och togoi and An (Hyicanus) group (Table 5).

3.6. Wimi-ri, Seoguipo-si

A total of 298 mosquitoes were collected in Wimi-ri, Seoguipo-si, Jeju-do, comprising four genera, five species. Cx pipiens was most frequently caught (77.9%), followed by Och togoi (15.8%), Cx tritaenior (5.4%), Ar subalbatus (0.7%), and Ae albopirtus (0.3%). No filariasis DNA was detected in Och togoi and An (Hyicanus) group (Table 6).

4. Discussion

In the survey, the ratio of Och togoi was highest in Maemul Island, Bogil Island, and Heuksan Island, while Cx pipiens was most frequently caught in Geomun Island, Namone-ri, and Wimi-ri in Jeju-do. This trend remained consistent in the survey between 2002 and 2005 by Cheun et al [19], and Cx pipiens also found out to be the dominant species in Geomun Island and Jeju-do. According to Ree [20], there are nine genera of mosquitoes found in Korea: Anopheles, Culex, Ochletotatus (formerly known as Aedes), Armigeres, Mansonia, Heizmannia, Tripteroides, Culicera, and Tozorhynchites. Among them, Och togoi was recognized as the main vector mosquito of B malayi in Jeju-do, Korea [8,21], whereas An sinensis sensu stricto, associated with rice paddies, is found in inland areas [9,14].

According to Lee [22], two larvae from mosquitoes were found to be infected with B malayi in 464 Aedes togoi collected in Jeju-do; after this, several researchers found that the natural infection rate of Aedes togoi was between 1.4% and 10.7%, and Och togoi was a vector of filariasis [23–25]. However, Ae koreicus and Cu pipens were reported to have poor susceptibility as a vector of filariasis [26]. Furthermore, Kim et al [10] also collected 4351 An sinesis in Youngju-si, Chungcheongbuk-do, inland areas, and found that 14 mosquitoes (0.3%) were infected with B malayi. Therefore, these studies showed that both Och togoi and An sinensis had susceptibility for lymphatic filariasis [10]. In this study, An (Hyicanus) group and Och togoi collected in these past endemic areas, however, were not infected with B malayi, and this fact is proof that these areas are free of lymphatic filariasis.

Next, mosquito larvae are susceptible to salinity and temperature, and their numbers increase after the

### Table 3. Seasonal variations of mosquitoes collected in mosquito trap in Geomun-ri (latitude 34°17’N, longitude 127°23’E), Geomun Island, Yeosu-si, Jeollanam-do in 2009

| Month | Ae alboptirus | Arm sabalbatus | Cx inatomi | Cx pipiens | Cx tritaenior | Och koreicus | Och togoi | Latrie vorax | Mean no./trap |
|-------|---------------|----------------|------------|------------|--------------|--------------|-----------|-------------|--------------|
| May   | 1             | 1              | 1          | 1          | 1            | 1            | 1         | 1           | 1            |
| Jun   | 2             | 2              | 2          | 2          | 2            | 2            | 2         | 2           | 2            |
| Jul   | 3             | 3              | 3          | 3          | 3            | 3            | 3         | 3           | 3            |
| Aug   | 4             | 4              | 4          | 4          | 4            | 4            | 4         | 4           | 4            |
| Sep   | 5             | 5              | 5          | 5          | 5            | 5            | 5         | 5           | 5            |
| Oct   | 6             | 6              | 6          | 6          | 6            | 6            | 6         | 6           | 6            |
| Total | 24 (6.4)      | 1 (0.3)        | 170 (45.2)| 18 (4.8)   | 1 (0.3)      | 158 (42)     | 1 (0.3)   | 376         |              |

Numbers in parenthesis represent data on male mosquitoes.

### Table 4. Seasonal variations of mosquitoes collected in mosquito trap in Maejuk-ri (latitude 34°38’N, longitude 128°34’E), Maemul Island, Tongyoung-si, Gyeongsangnam-do in 2009

| Month | Ae alboptirus | Arm sabalbatus | Cx inatomi | Cx pipiens | Cx tritaenior | Och koreicus | Och togoi | Mean no./trap |
|-------|---------------|----------------|------------|------------|--------------|--------------|-----------|--------------|
| May   | 7             | 7              | 7          | 7          | 7            | 7            | 7         | 7            |
| Jun   | 16            | 16             | 16         | 16         | 16           | 16           | 16        | 16           |
| Jul   | 2             | 2              | 2          | 2          | 2            | 2            | 2         | 2            |
| Aug   | 14            | 14             | 14         | 14         | 14           | 14           | 14        | 14           |
| Sep   | 2             | 2              | 2          | 2          | 2            | 2            | 2         | 2            |
| Oct   | 3             | 3              | 3          | 3          | 3            | 3            | 3         | 3            |
| Total | 4 (0.3)       | 145 (12)       | 1 (0.1)    | 25 (2.1)   | 6 (0.5)      | 2 (0.2)      | 1030 (84.9)| 1,213        |

Numbers in parenthesis represent data on male mosquitoes.
summer monsoon season [19]. As the survey results show, the number began to climb up in June in all survey areas, stayed high July through September, and started to fall in October. According to Nakamura (1988), the number of *Och togoi* larvae begins to increase slowly when winter is over, and peaks in early May. The number falls in summer as the air and water temperature rises with strong sunlight. However, when rainfall increases in June through September, the number increases again due to lower salinity in rock pools [27]. This means that seasonal condition and regional characteristics have a direct influence on the number of mosquito larvae. Compared to a previous study [21], the regional decrease or increase in the number of collected mosquitoes was affected by these conditions. However, this study confirmed that *Och togoi* and *An (Hyrcanus)* group are the dominant species in survey areas.

This is the first study conducted in Korea that investigated filariasis DNA from vector mosquitoes in six remote island areas in Korea, and the negative results indicate that filariasis has been eliminated in the country. In particular, the costal island areas in Jeollanam-do showed a high infection rate until the early 2000s [28], but government, academic, and local clinics have been exerting efforts continuously to bring about its elimination. As the country’s economic growth accelerated from the late 1980s, peoples’ quality of living improved. They had better access to medical treatment, and various mosquito-repelling chemicals and equipment were developed such as mosquito nets. All these factors substantially reduced human contact with mosquitoes [29].

Finally, the survey examined the number of mosquitoes, including vector species for filariasis, in island areas of Korea, and confirmed the negative result in DNA detection. However, filariasis has a latent period of 4–10 years, and there is a possibility of its emergence or reemergence through travel or trade as more people visit areas in the West Pacific, Southeast Asia, and Africa, where filariasis is still active. For this reason, it is important to continue quarantine inspection and monitoring of vector mosquitoes.

### Acknowledgments

We thank our colleagues at the Province and City Bureau of Health Center and the Research Institute of Health and Environment for their devoted support and efforts. This work was supported by a grant from Korea National Institute of Health (NIH-091-4800-4845-300), National Research and Development Program, Ministry of Health and Welfare, the Republic of Korea.

### References

1. Michael E, Bundy DA, Grenfell BT. Re-assessing the global prevalence and distribution of lymphatic filariasis. Parasitology 1996;112(Pt 4):409–28.
2. Ottesen EA. Lymphatic filariasis: treatment, control and elimination. Adv Parasitol 2006;61:395–441.
3. World Health Organization. Lymphatic filariasis. Wkly Epidemiol Rec 2003;78:169–80.
4. Yun IS. Elephantiasis due to filaria in Korea. Chosen Igakai Zasshi 1927;76:326–34.
5. Seo BS. Malayan filariasis in Korea. Korean J Parasitol 1978;16(Suppl.):5–108.
6. Seo BS. Studies on filariasis in Korea: status survey and chemotherapy in Cheju Do. Seoul J Med 1976;17:83–95.
7. Senoo T, Lincicome DR. Malayan filariasis. Incidence and distribution in Southern Korea. U.S. Armed Forces Med J 1951;2(10):1483–9.
8. Lee KT, Kim SH, Kong TH, et al. Malayan filariasis. 2nd report: epidemiological investigations on filariasis due to *Brugia malayi* in the residents of southern Cheju-Do island. J Korean Med Assoc 1964;7:657–64.
9. Kim DC, Lee OY, Lee KW. Epidemiology of malayan filariasis of inland Korea: 1. endemicity of filariasis malayi in Yongju area. Yonsei Rep Trop Med 1977;8:9–22.
10. Kim DC, Lee OY, Lee KW. Epidemiology of malayan filariasis of inland Korea: II. Vector finding and transmission of *Brugia malayi* in Yongju area. Yonsei Rep Trop Med 1977;8:23–32.
11. Lee JS, Kim TS, Lee WJ, et al. Epidemiology of malayan filariasis of inland Korea (III). Report NIH Korea 1992;29:114–22.
12. Lee OY, Lee JS, Son SC, et al. Epidemiological studies on filariasis malayi on Cheju Do and the southern islands. Report NIH Korea 1986;23:407–22.
13. Cheun HI, Lee JS, Cho SH, et al. Elimination of lymphatic filariasis in the Republic of Korea: an epidemiological survey of formerly endemic areas, 2002–2006. Trop Med Int Health 2009;14(4):445–9.
14. Kim DC. Epidemiological studies of filariasis in inland Korea: 4. Vector determination of filariasis malayi in Yongju Area. Abstracts of the 16th Annual Meeting of the Korean Society for Parasitology; 1974.
15. Cheun HI, Cho SH, Lee JS, et al. Successful control of lymphatic filariasis in the Republic of Korea. Korean J Parasitol 2009;47(4):323–35.
16. Tanaka K, Mizusawa K, Saugstad ES. Mosquitoes of Japan and Korea. Contrib Am Entomol Inst 1979;16:148–52.
17. Lee KW. A revision of the illustrated taxonomic keys to genera and species of female mosquitoes of Korea. Department of the Army, 5th Medical Detachment, 168th Medical Battalion, 18th Medical Command; 1998.
18. Mishra K, Kumar Raj D, Dash AP, et al. Combined detection of *Brugia malayi* and *Wuchereria bancrofti* using single PCR. Acta Trop 2005;93(3):233–7.
19. Cheun HI, Cho SH, Lee HI, et al. Seasonal prevalence of mosquitoes, including vectors of brugian filariasis, in southern islands of the Republic of Korea. Korean J Parasitol 2011;49(1):59–64.
20. Ree HI. Medical entomology. Seoul, Korea: Komoon Sa; 1978. p. 1–294.
21. Kim JS, Lee WY, Chun SL. Ecology of filariasis on Cheju Island. Korean J Parasitol 1973;11(1):33–53.
22. Lee WY. A study on *Aedes togoi* as vector of filariasis in Cheju Island. Korean J Parasitol 1969;7(2):153–9.
23. Chun SR. A preliminary survey of mosquitoes of Cheju do related to filariasis, on the species, biology and infection status. Korean J Public Health 1968;5:113–21.
24. Wada Y, Katamine D, Oh MY. Studies on malayan filariasis in Cheju Island, Korea: 2. Vector mosquitoes of malayan filariasis. Jpn J Trop Med Hyg 1973;13(4):197–210.
25. Seo BS. The periodicity of microfilariae of *Brugia malayi* in Cheju Island. Korean J Parasitol 1974;12(2):95–100.
26. Kim DC, Lee OY, Kim TW, et al. Epidemiological studies of human filariasis of inland Korea: endemicity and transmission of human filariasis in Yongju area. Report NIH Korea 1971;8:147–65.
27. Nakamura S, Miyagi I, Toma T. Seasonal appearance of immature populations of *Aedes* (Finlaya) *togo* (Theobald) in Okinawa. Jpn J Sanit Zool 1988;39(2):91–6.
28. Chai JY, Lee SH, Choi SY, et al. A survey of *Brugia malayi* infection on the Heuksan islands, Korea. Korean J Parasitol 2003;41(1):69–73.
29. Korea National Institute of Health, Centers for Disease Control and Prevention. 2007 Elimination of Lymphatic filariasis in Korea (National Documentation for Certification); 2007. p. 1–149.