

# Prevalence of canine dirofilariasis in Taiwan

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## Abstract

Between 1993 and 1997, 837 stray dogs from North Taiwan were necropsied and examined for dog heartworm infections. A thick smear from 20 ml of peripheral blood from each dog was also prepared and examined for microfilariae (mf). The overall prevalence of adult worms in the dog population was 57%. The prevalence of mf in 1228 house dogs from different parts of Taiwan was also determined from 20 ml of peripheral blood in the same way. The overall prevalence of mf was 25%, with a value of 30% in the main island of Taiwan, this being 15 times higher than that in the offshore islands (2%). In Taiwan, the prevalence ranged from 4% in Hualien County, East Taiwan, to 41% in Nantou County, Central Taiwan. The mf prevalences on offshore islands were 1% on Liuchiu and 2% on Lanyu. The mf density per 20 ml blood in 82 house dogs was found to be 23 mf per dog, with a range of 3–97 mf per dog. A total of 477 stray dogs were found to be infected with adult worms of *Dirofilaria immitis*. The mean number of 7 worms per dog was obtained, with a range of 1–55 worms per dog. These results indicate that the prevalence of canine dirofilariasis has increased in Taiwan over the past 10 years. Moreover, the prevalence may be related to the wind speed, temperature, relative humidity, and altitude in the different areas surveyed.

## Introduction

The dog heartworm *Dirofilaria immitis* has a worldwide distribution. Although it is a common parasite of the dog, the timber wolf, red fox, grey fox, coyote, domestic and wild cats, bear, beaver, sea lion, harbour seal, muskrat, otter, nutria and man have also been reported to harbour this nematode (Beaver *et al.*, 1984). Therefore, it is an enzootic parasite. Sixty cases of human infection with the dog heartworm have been reported in the USA (Cifferi, 1982), one in Brazil (Leonardi *et al.*, 1977) and ten or more in Japan (Yamane *et al.*, 1977; Kumada *et al.*, 1980). In two cases worms removed from the abdominal cavity were identified as *D. immitis* (Yoshimura *et al.*, 1980a,b). A case of human pulmonary dirofilariasis has also been reported in Taiwan (Chiang *et al.*, 1992).

Canine dirofilariasis in Taiwan was first reported by Miyamoto & Uchida (1935). One imported dog from Mainland China and another from Japan were found to be

infected. However, among 75 indigenous dogs, negative results were obtained. Hsieh & Chuang (1956) examined 75 dogs from Central Taiwan and revealed the infection in one imported and three indigenous dogs. Huang *et al.* (1957) reported 4.5% of 290 stray dogs in North Taiwan being infected with this parasite. These findings indicate an increasing trend in the prevalence of infection.

In the past 20 years, a large number of dogs have been imported from the endemic areas of dirofilariasis, including the United States and Japan. Chen *et al.* (1982) reported a prevalence of 5.2% among 440 imported dogs examined by the nuclepore membrane technique in Taipei County. Yang & Hsieh (1987) reported 16.8% of 155 dogs were infected in Central Taiwan. In 1988, we reported a value 24.8% amongst 306 stray dogs in Taipei City (Wu *et al.*, 1988a). Four years later, we obtained a value of 42% amongst 265 stray dogs in the same city (Wu *et al.*, 1992). Since then a number of reports concerning this infection have been published (Ku *et al.*, 1992; Tin & Wang, 1994; Liang *et al.*, 1996; Wang, 1997; Fang *et al.*, 1999; Su, 1999; Fan *et al.*, 2000). However, these studies were limited to stray dogs or house dogs from a city area or a remote aboriginal district. In the present study, we

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examined not only stray dogs in three cities but also house dogs in ten counties of Taiwan. The results of this long-term study (1993–1997) are likely to reflect the current status of dog heartworm infection in Taiwan.

## Materials and methods

### *Stray dogs*

From 1993 to 1997, 837 stray dogs from Taipei City (1993–1997), Miaoli City (1993–1996), and Taoyuan City (1993) were examined by necropsy. After injection with an overdose of pentobarbital, the heart and lungs of each dog were removed and examined for adult *D. immitis*. The length of individual worms was also measured. Male worms equal to or longer than 13 cm and female worms equal to or longer than 19 cm were considered mature. In order to determine the presence of microfilariae, a thick blood smear was prepared from the heart blood. Each smear was made from 20 ml of blood and spread to a diameter of 1.5 cm. The smears were air dried overnight, stained with 7% Giemsa stain solution, and examined microscopically.

### *House dogs*

In addition to stray dogs, 1228 house dogs from Chiai County (1993–1994), Hsinchu City (1993–1994), Nantou County (1993–1996), Taitung County (1994), Hualien County (1994), Pingtung County (1994), Ilan County (1995), Yunlin County (1995), Tainan County (1995) and Kaohsiung County (1995) were examined for dirofilariasis by peripheral blood examination. A volume of 20 ml of peripheral blood was drawn from each dog and a thick blood smear was spread to a diameter of 1.5 cm. The smears were air dried overnight, stained with 7% Giemsa stain solution, and examined microscopically.

## Results

### *Prevalence in stray dogs*

Of the 837 stray dogs necropsied, 477 were found to be infected with *D. immitis*. The overall prevalence was 57.0%, with 56.3% of male and 57.9% of female dogs being infected. In Taipei City, the prevalence was 67%, with 66% of male and 67% of female dogs being infected. In Miaoli City, the prevalence was 42%, with 36% of male and 47%

of female dogs being infected. In Taoyuan City, the prevalence was 40%, with 36% of male and 44% of female dogs being infected. A total of 3437 worms were recovered from infected dogs. The overall worm recovery was 7.2 worms per dog. The values in Taipei, Miaoli and Taoyuan were 8.1, 5.2 and 3.8 worms per dog respectively. The overall mf rate was 23.1%, with 24.5% of male and 21.1% of female dogs being infected. The values in Taipei, Miaoli and Taoyuan were 26.3%, 17.8% and 17.1% respectively (table 1).

### *Prevalence in house dogs*

Of 1228 house dogs examined, 326 were found to be infected. The overall mf rate was 26.5%, with 23.4% of male and 27.3% of female dogs being infected. On the main island of Taiwan, the prevalence was 27.9%. The highest rate of infection was found in Nantou County (40.7%) followed by Hsinchu City (36.4%) and Tainan County (35.2%). The prevalences in dogs from Kaohsiung, Yunlin, Taitung, Ilan, Pingtung and Chiai Counties were 25.0%, 18.9%, 18.0%, 12.4%, 8.3% and 5.3% respectively. Hualien County (4.0%) had the lowest rate. The prevalence on offshore islands was 1.6%: Lanyu 1.8% and Liuchiu 1.4% (table 2).

### *Microfilarial density in house dogs*

The mf density per 20 ml peripheral blood was determined in 82 house dogs collected from Nantou, Pingtung and Tainan counties. A total of 1855 mf were detected. Each dog was infected with 22.6 mf on average: 18.5 mf per male dog and 27.9 mf per dog (table 3).

### *Adult worms in stray dogs*

Of the 477 stray dogs examined, a total of 3437 adult worms were collected. Each dog was infected with 7.2 worms on average: 7.3 worms per male dog and 7.1 worms per female dog (table 4). The length of adult worms of *D. immitis* in stray dogs varies from 6 to 35 cm among females and 6 to 21 cm among males. However, most female worms are 26–30 cm in length and males 16–20 cm (table 5).

Table 1. Prevalence of dirofilariasis in stray dogs from three cities in North Taiwan from 1993 to 1997 by examination of microfilariae and adult worms.

Year	Endemic area	Sex of dogs	No. of dogs examined	Microfilariae		Adult worm		Worm recovery	
				No. infected	Prevalence (%)	No. infected	Prevalence (%)	Total no.	No./infected dog
1993–1997	Taipei City	M	318	90	28.3	211	66.4	1678	8.0
		F	202	47	23.3	135	66.8	1113	8.2
1993–1996	Miaoli City	M	124	22	17.7	45	36.3	234	5.2
		F	123	22	17.9	58	47.2	306	5.3
1993	Taoyuan City	M	36	5	13.9	13	36.1	62	4.8
		F	34	7	20.6	15	44.1	44	2.9

Table 2. Prevalence of microfilariae of canine heartworm in stray and house dogs in Taiwan from 1993 to 1996.

Year	Endemic area		Sex of dogs	No. examined	No. infected	Prevalence (%)
	County	District				
1993–1994	Hsinchu	Hsinchu City	M	10	5	50.0
			F	12	3	25.0
1993–1996	Nantou	Mingchien, Hsini, Jenai	M	226	79	35.0
			F	317	142	44.8
1993–1994	Chiai	Alishan	M	26	1	3.8
			F	12	1	8.3
1994	Hualien	Siulin	M	21	1	4.8
			F	29	1	3.4
1994	Pingtung	Manchow, Nuipu	M	103	11	10.7
			F	54	2	3.7
1994	Taitung	Haituan, Chengkung	M	42	9	21.4
			F	19	2	10.5
1995	Ilan	Tatung, Nanao	M	53	7	13.2
			F	60	7	11.7
1995	Yunlin	Kukung, Tsouling	M	24	6	25.0
			F	13	1	7.7
1995	Tainan	Hsinhua City	M	62	27	43.5
			F	46	11	23.9
1995	Kaohsiung	Liukuei	M	24	7	29.2
			F	8	1	12.5
Offshore islands						
1993	Taitung	Lanyu	M	25	0	0
			F	31	1	3.2
1995	Pingtung	Liuchiu	M	38	0	0
			F	33	1	3.0

#### Relation of microfilariae and adult worms in stray dogs

Among the 477 stray dogs examined, 57.8% were positive for mf and harboured mature male and female worms. Only 10.2% were positive for mf in 49 dogs with only mature male worms and 11.8% in 51 dogs with only mature female worms. No mf were found in 15 dogs with mature male and immature female worms, 27 dogs with immature male and immature female worms or 24 dogs with immature worms. However, there were 5 dogs with mf but without worms (table 6).

### Discussion

In 1988, we reported that 24.8% of 306 stray dogs from Taipei City were infected with adult worms of *D. immitis*

and 9.2% showed microfilaraemia (Wu *et al.*, 1988a). In 1992, the prevalence increased to 42% of 265 stray dogs from the same city (Wu *et al.*, 1992). Between 1976 and 1992, 10.2% of 2593 dogs examined in the Veterinary Teaching Hospital of National Chung Hsing University in Taichung City were infected with dog heartworm (Ku *et al.*, 1992). From September 1993 to November 1994, 28.8% of 300 stray dogs from the same city were found to be infected with *D. immitis* (Tin & Wang, 1994).

Liang *et al.* (1996) reported that 361 pet dogs were admitted to National Taiwan University Veterinary Hospital from July 1994 to May 1995 for microfilaraemia and adult heartworm antigen. Amongst these dogs, 5.5% were found to have microfilaraemia by microscopic examination and 17.7% showed positive serological results. In the infected dogs, 25% were positive for both

Table 3. Microfilarial density per 20 ml peripheral blood of *Dirofilaria immitis* in house dogs from Nantou, Pingtung and Tainan counties, Taiwan.

Mf density per 20 cmm blood	Male dog			Female dog		
	No. of dogs examined	Microfilariae		No. of dogs examined	Microfilariae	
		Total no.	No. per dog		Total no.	No. per dog
1–10	27	95	3.5	16	55	3.4
11–30	8	146	18.3	8	161	20.1
31–50	5	196	39.2	3	117	39.0
51–80	5	324	64.8	7	486	69.4
81–100	1	89	89.0	2	186	93.0
Total	46	850	18.5	36	1005	27.9

Table 4. The occurrence of adult worms of *Dirofilaria immitis* in stray dogs from three cities in North Taiwan (1993–1997).

No. of adult worms per dog	Male dog			Female dog		
	No. of dogs examined	Total no. of worms	No. of worms per dog	No. of dogs examined	Total no. of worms	No. of worms per dog
1–5	159	428	2.69	126	320	2.5
6–10	60	464	7.73	38	296	7.8
11–15	16	204	12.75	23	297	12.9
16–20	15	260	17.33	10	181	18.1
21–25	8	180	22.5	3	71	23.7
26–30	3	85	28.33	2	54	27.0
31–35	2	68	34.0	2	64	32.0
35–40	1	37	37	1	38	38.0
41–45	1	44	44	1	45	45.0
46–50	3	149	49.66	2	97	48.5
Over 50	1	55	55	0		
Total	269	1974	7.3	208	1459	7.1

parasitological and serological examinations, 70% were only positive for serological examination, and 5% positive for parasitological examination but negative for serological examination. The majority of dogs under 1 year of age was found to be negative for dog heartworm using both methods. However, adult dogs (>1 year old) had a positive rate of 19.7%. Although no significant difference was found in the positive rate by dog sex (male dogs 19.5% and female dogs 17.2%), large breed dogs (German Shepherd dog 61.5%, Rottweiler 30.8%) had higher prevalences than the small breed dogs (Pomeranian 11.4%, Maltese 0%, Miniature Poodle 0%). Wang (1997) reported that 60.6% of 180 stray dogs from North Taiwan were found to have canine filarial infections by postmortem examination. The prevalence of *D. immitis* was 55% and *Dipetalonema reconditum* 12.2%. Each positive dog with *Dirofilaria immitis* was found to harbour 1–102 worms. *Dipetalonema reconditum* was the first record in Taiwan but few morphological characteristics were described.

Su (1999) examined 94 stray and 54 house dogs in Middle-Southern Taiwan by microscopic examination for microfilariae and ELISA for adult worms and found that 36.2% of strays and 20.4% of house dogs were infected. However, the mf rate and the prevalence of *D. immitis* were 35.1% and 16.7%, respectively. No significant differences were found in the positive rates for adult worms nor the mf rates relative to dog sex. In the same year, Fang *et al.* (1999) examined 87 stray dogs in Kaohsiung by postmortem examination for

adult worms of *D. immitis*, blood examination for microfilariae, and ELISA for adult antigens. The prevalence was found to be 44.8% and there was no significant difference relative to dog sex (males 42.9% and females 47.4%). Each infected dog was found to harbour 4.9 adult worms. Fan *et al.* (2000) examined 20 stray dogs from the Dawu and Danzen Districts of Taitung County of East Taiwan by ELISA and 10% were found to be positive.

In the present study, a high prevalence of 57% among 837 stray dogs in Taipei City, Taoyuan City, and Miaoli City in North Taiwan was recorded by necropsy. The prevalence in Taipei City was found to be 67%, which was higher than that reported by Wu *et al.* (1988a, 1992), i.e. 24.8% of 306 stray dogs in 1988 and 42% of 265 stray dogs in 1992. These findings indicate an increasing trend in the prevalence of canine dirofilariasis in the past 10 years. Moreover, the prevalence in 1228 house dogs in 10 counties were also determined by peripheral blood examination for microfilariae. Although the overall mf rate was 25%, the rate ranged from 4% in Hualien County, East Taiwan, to 41% in Nantou County, Central Taiwan.

Amongst 477 stray dogs examined in Northern Taiwan, 10% were found with only mature male worms positive with mf and 12% with only female worms positive with mf. Male or female worms in these dogs may have died. Those with both mature male and female worms but some without mf may have been caused by occult infections (Wong, 1974).

Table 5. The length of adult worms of *Dirofilaria immitis* in stray dogs.

	Range in length (cm)					
	6–10	11–15	16–20	21–25	26–30	31–35
No. of females	11	67	181	383	578	102
No. of males	34	309	892	10*	0	0

\* The maximum length of males is 21 cm.

Table 6. Examination of microfilariae (mf) of *Dirofilaria immitis* among 477 stray dogs from three cities in North Taiwan (1993–1997).

Worm development	No. of dogs examined	Mf positive		Mf negative	
		No. of dogs infected	Prevalence (%)	No. of dogs infected	Prevalence (%)
Mature males only	49	5	10.2	44	89.8
Mature females only	51	6	11.8	45	88.2
Mature males and mature females	306	177	57.8	129	42.2
No immature and mature worms	5	5	100		
Immature females and mature males	15			15	100
Immature males and mature females	27			27	100
Immature males and immature females	24			24	100

Length of mature male worms  $\geq 13$  cm. Length of mature female worms  $\geq 19$  cm.

Dogs which were mf positive but without immature and mature worms may be infected with *Dirofilaria* sp. occupying the subcutaneous tissues (Beaver *et al.*, 1984). As the dogs may have received prophylactic injections or treatment, both male and female may be present but without mf.

Mosquitoes are widespread in Taiwan and the climate is favourable for the development of larvae of *D. immitis* in their mosquito vectors (Lien, 1978). In the present study, we found *Culex quinquefasciatus* and *Aedes albopictus* naturally harbouring the third stage larvae of *D. immitis*. In our previous experimental study, we found that *A. albopictus*, *A. togoi*, *C. alis*, *C. quinquefasciatus*, and *C. tritaeniorhynchus* were susceptible to *D. immitis* in the laboratory (Wu *et al.*, 1988b). Except for *A. togoi*, these potential vectors are common mosquitoes near human habitations and may transmit the infection. Moreover, natural infections of this parasite have also been found in *C. quinquefasciatus* and *A. albopictus* in North Taiwan (Wu *et al.*, 1997). However, the distribution of these mosquitoes may vary in various parts of Taiwan because of differences in wind speed and high altitude. According to the Central Weather Bureau, the wind speed in Taipei City ranged from  $72.2 \text{ m s}^{-1}$  in June to  $116.2 \text{ m s}^{-1}$  in December whereas corresponding figures for the Hsincheng District of Hualien County were  $8.7 \text{ m s}^{-1}$  in August to  $28.7 \text{ m s}^{-1}$  in January and June. In general, low prevalences of dirofilariasis were found in areas with high altitude or high wind speed. Moreover, temperature and relative humidity also vary significantly in different parts of Taiwan. These environmental factors may significantly influence the density of mosquito populations and hence the rate of dog heartworm infection. Moreover, high prevalences in remote areas may imply that dog heartworm infection is transmitted indigenously before the import of dogs from endemic areas. However, this suggestion needs further investigation.

#### Acknowledgements

The authors wish to thank the National Science Council, ROC, for support of a research grant (No. NSC85-2331-B010-034), the Department of Physiology, Dentistry, and Experimental Surgery in National Yang-ming University for the donation of stray dogs, and

Taoyuan City Environmental Protection Bureau, Miaoli City Environmental Protection Bureau, Hsini District Health Station, Nantou County and Haituan District Health Station, Taitung County, for their kind help and co-operation during the field survey of *Dirofilaria immitis* in Taiwan between 1993 and 1997.

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(Accepted 12 September 2002)  
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