The Progression of Taiwan Ferret Badger Rabies from July 2013 to December 2016

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Abstract

On July 17, 2013, rabies was confirmed in ferret badgers (Melogale moschata) in Taiwan, which is the sole reservoir host. The epidemic happens solely in Taiwan Island. This research is to show the progression of ferret badger rabies at Taiwan, using the samples examined from July 2013 to December 2016. Rabies data are periodically released by the Bureau of Animal and Plant Health Inspection and Quarantine (BAPHIQ) in the http://www.baphiq.gov.tw/ governmental website index.php. The authors downloaded necessary data to investigate the epidemiology with kind permission of BAPHIQ. From July 2013 to December 2016, a total of 644 samples of ferret badgers were taken from 7 cities/ counties of all the 16 cities/counties in Taiwan Island. None (0%, 0/644) were confirmed as rabies-positive. Of the 789 samples of ferret badgers taken from the other 9 cities/counties, 553 (70.1%, 553/789) were confirmed as rabies-positive. Until December 2016, a total of 77 townships were confirmed as rabies-enzootic. A total of 76 out of the 77 enzootic townships were mountainous. Only 1 out of the 77 townships was urbanized. Chi-square test indicates that there is an extremely significant difference in the incidence of ferret badger rabies between townships with mountainous landscapes and urbanized townships (p<0.001). Ever since the outbreak of ferret badger rabies in July 2013, the epidemic has sustainably been constrained by the original geographical barriers.

In 2014, 2015, and 2016, the progression of ferret badger rabies in total added 18 enzootic townships.

Keywords: Taiwan; Ferret badger; Rabies; Epidemiology

Introduction

Rabies Virus (RABV) is the prototype virus of the genus *Lyssavirus* in the family Rhabdoviridae and is the causative agent of classic rabies in human and all mammals. Once the infection is established and has reached the brain, RABV will cause a fatal encephalomyelitis. Rabies infection in terrestrial animals is maintained in two epidemiological cycles, one urban and one sylvatic. In the urban rabies cycle, dogs are the main reservoir host. In the sylvatic rabies cycles, reservoirs comprise foxes, skunks, mongooses, raccoons, coyotes, raccoon dogs etc. [1]. On July 17, 2013, RABV was detected in Taiwan ferret badgers in Taiwan Island [2]. Ferret badger is the reservoir host for the rabies virus, and the epidemic is only prevalent in Taiwan Island [2,3].

The ferret badger is omnivorous wild-life mustelid, feeding on invertebrates, vertebrates, fruits, etc. Most important food sources are earthworms and insects [4]. Reports of mean body mass for adults were 0.90 kg \pm 0.14 kg [5], or 0.8 kg \sim 1.6 kg [6]. The animal has poor visual perception, small teeth and weak biting force, which is considered as a weak competitor and not found in urbanized areas accordingly [7]. Ferret badgers exhibit strong nocturnal activity patterns, mainly known to occur at elevation below 2000 m, being seen throughout southern China, Taiwan, Hainan, the northern portions of Vietnam, Laos, Thailand, and Burma, Bangladesh, and North Eastern India [6,7].

Tsai et al. indicated that the rabies is badger-associated and confined to the above mentioned 9 cities/counties in Taiwan sylvatic environments [8]. The phylogeny of nucleoprotein and glycoprotein genes of ferret badger associated RABV revealed the RABV to be clustered in two distinct groups, TWI and TWII, consistent with the geographic segregation into the East and West areas separated by the Central Mountain Range. Chang et al. also indicated that there were the Main (West) and East epidemic areas separated by the Central Mountain Ranges [3]. Chiou et al. indicated that RABV could be cryptically circulating in the environment [9]. An understanding of the underlying mechanism might shed light on the complex interaction between RABV and its host. Lin et al. indicated that the origin of the epidemic could be in the Eastern Taiwan, and then the Formosan ferret badger rabies moved across the Central Mountain Range to western regions and separated into two branches [10]. A retrospective study of Formosan ferret badgers rabies indicated that the ferret badger population in Taiwan had been affected by rabies prior to 2010 [11].

Objectives of this research are to identify enzootic area and progression characteristics of ferret badger rabies and to study the association between the landscapes of townships and the incidence of ferret badger rabies.

Materials and Methods

Rabies surveillance

To determine the spread of the disease and the enzootic areas within Taiwan Island, all suspected animals were submitted and tested. Suspected animals, mainly ferret badgers, included road-kills, those exhibiting neurological signs, and those found dead. Animals submitted for testing were collected by veterinary officers of local authorities in Taiwan Island, with assistance from local law enforcement officers, animal inspectors, and the general public. The national Animal Health Research Institute (AHRI) performed diagnosis. The most widely used test for rabies diagnosis was Fluorescent Antibody Test (FAT), which is the gold standard recommended by both WHO and OIE. However, in the case of human exposure or samples from suspicious animals with human contact history, samples were always conducted by FAT and PCR simultaneously. For FAT test, 2 sample tests per brain were conducted concurrently. In cases of inconclusive results from FAT, samples were further inoculated into MNA (Murine Neuroblastoma) cell line to detect the replication of the virus. From 2016, AHRI has joined the inter-laboratory test for rabies diagnosis and serology proficiency test that organized by the Anses-Nancy laboratory for rabies and wildlife, an OIE reference laboratory. The tests AHRI succeeds are FAT, RT-PCR, real-time RT-PCR (for rabies diagnosis), FAVN and RFFIT (for serological test). Future reference to a "positive" or "infected" ferret badger indicates a rabies-confirmed test result.

Landscapes of townships and rabies prevalence

To determine whether the landscape condition of a township is associated with the incidence of ferret badger rabies, the chi-squared test for two-way contingency table was used to assess whether mountainous townships are associated with the incidence of ferret badger rabies, using positive or negative cases of ferret badger rabies from July 2013 to December 2016 in Taiwan Island. Information of the landscape of townships were retrieved on governmental official websites as well as Taiwan Google 3D map (https://www.google.com/maps/@24.3981937,120.777506,21451m/data=!3m1!1e3? hl=zh-TW). The townships with landscape having greater or equal to 200 m are categorized as mountainous townships.

Statistical analysis

The chi-squared test for two-way contingency table was used to determine whether the landscape of townships was associated with the incidence of ferret badger rabies. Calculation was carried out using Graph Pad Software at https://www.graphpad.com/quickcalcs/contingency1.cfm.

Results

Rabies surveillance

Table 1 indicates that all of the 644 ferret badgers sampled from 7 cities or counties were tested negative. They were considered as rabies free area. **Figure 1** identified the locations of the 7 cities or counties, which are Taipei City, New Taipei City combined Keelung City, Taoyuan City, Hsinchu County combined Hsinchu City, Miaoli County, Changhua County and Yilan County. Keelung City and Hsinchu City are too small to mark in **Figure 1**.

Table 2 shows the diagnosis of rabies in 789 ferret badgers sampled in the other 9 cities or counties in Taiwan Island. The number of confirmed rabies cases in the 9 cities or counties is 553. Three counties of them are located east of the Central Mountain Range. They are categorized as "East Epidemic Area", from north to south including: Hualien County (37 cases), Taitung County (210 cases), and Pingtung County (21 cases). The other 6 of the 9 enzootic cities or counties are located west of the Central Mountain Range. They are categorized as "West Epidemic Area", including: Taichung City (67 cases), Nantou County (113 cases), Yunlin County (34 cases), Chiayi County with Chiayi City (26 cases), Tainan City (26 cases), Kaohsiung City (19 cases), as shown in **Figure 1**. The Chiayi City is too small to mark in **Figure 1**.

Figure 2 illustrates the progression of the ferret badger rabies front in geographical units of township from July 2013 to December 2016, which shows 59 enzootic townships in 2013; 3 more rabid enzootic townships were confirmed in 2014; 10 more townships were confirmed in 2016. In 2014, the 3 confirmed new enzootic townships were Mudan in Pingtung County, Fuli in Hualien County and Changbin in Taitung County. In 2015, the 10 confirmed new enzootic townships were Chichi and

2017

Vol.1 No.1:2

Nantou City in Nantou County; Meishan in Chiayi County; Baihe and Yujing in Tainan City; Taimali in Taitung County; Henchuen, Taiwu, Fangliau and Chunrih in Pingtung County. In 2016, the 5 confirmed new enzootic townships were Fengyuan in Taichung City, Douliou in Yulin County, Dalin in Chiayi County, Fengbin in Hualien County, and Dawu in Taitung County.

Table 3 compares the progression of the ferret badger rabies front in geographic units of city, county, as well as township from July 2013 to December 2016. The number of enzootic area of ferret badger rabies does not change annually when counted by the units of city or county. However, the number of enzootic townships increased annually when counted by the unit of township. In **Table 3**, three rabid enzootic townships were confirmed and added to the count for enzootic townships in 2014; 10 townships were added in 2015 and 5 townships were added in 2016. **Figure 2** shows the annual demographic expansion in geopolitical unit of township from the outbreak of the epidemic in July 2013, where the colour of expanded townships is different annually.

Landscapes of townships and rabies prevalence

Table 4 indicates that there are a total of 215 townships in East and West Epidemic areas. The number of enzootic townships with mountainous landscapes is 76. The number of enzootic townships with urbanized and in level areas is 1. Among the other 138 (215 minus 77) townships where no cases of ferret badgers rabies were found, the number of mountainous townships is 23. The number of townships with urbanized and in level areas are 115.

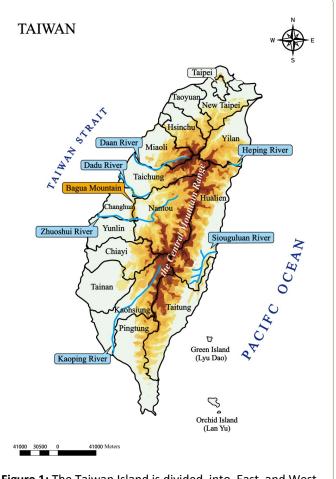


Figure 1: The Taiwan Island is divided into East and West Epidemic Areas of ferret badger rabies by the Central Mountain Range.

 Table 1: Non-enzooticareas of ferret badger rabies: Taiwan, July 2013 ~December 2016. Note: Data are expressed as "number tested".

No.	Counties/Cities	2013	2014	2015	2016	Subtotal
1	Taipei City	31	6	6	7	50
2	New Taipei City	173	10	15	12	210
3	Taoyuan City	10	2	1	3	16
4	Hsinchu County	82	11	18	22	133
5	Miaoli County	137	15	20	16	188
6	Changhua County	9	0	2	0	11
7	Yilan County	22	5	3	6	36
	Total	464	49	65	66	644

 Table 2: Prevalence of ferret badger rabies: Taiwan, July 2013~December 2016. Note: Data are expressed as "number positive/

 number tested". ND: Not done.

No.	Counties/Cities	2013	2014	2015	2016	Subtotal
1	Hualian County	3/20	15/22	16/29	3/6	37/77
2	Taitung County	118/124	79/80	9/11	4/4	210/219
3	Pingtung County	9/13	5/12	7/11	0/3	21/39
4	Taichung City	32/55	17/28	11/17	7/9	67/109
5	Nantou County	54/86	15/27	27/44	17/32	113/189
6	Yunlin County	16/22	10/16	5/8	3/4	34/50
7	Chiayi County	10/45		0/44	5/44	00/07
7	w/Chiayi City	13/15	N.D.	8/11	5/11	26/37
8	Tainan City	17/33	5/8	3/3	1/2	26/46
9	Kaohsiung City	14/15	1/1	2/5	2/2	19/23
	Total	276/383	147/194	88/139	42/72	553/789

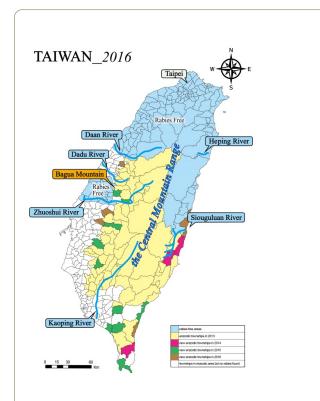


Figure 2: Demographic epidemic expansion of ferret badger rabies in geographic unit of township, July 2013 ~ December 2016.

Statistical analysis

The critical value of chi-square (X²) distribution is 10.827 when α =0.001, df=1. Chi-square test of the Epidemic area at **Table 5** shows that: X²=133.87>10.827, which indicate that there is an extremely significant difference in the incidence of ferret badger rabies between townships with mountainous landscape and urbanized townships (p<0.001).

Discussion

Tables 1 and 2 indicate that ever since the outbreak of ferret badger rabies in July 2013, the epidemic has sustainably been constrained by the following geographical barriers, as illustrated in Figure 1: 1) The Central Mountain Range separates the East Epidemic Area and the West Epidemic Area. Tsai et al. also indicated that the phylogeny of nucleoprotein and glycoprotein genes of ferret badger associated RABV revealed the RABV to be clustered in two distinct groups, TWI and TWII, consistent with the geographic segregation into the East and West Areas separated by the Central Mountain Range [7]; 2) The Daan River, which originates from the west of Central Mountain Range, blocks the epidemic spreading from the southern side to the North; 3) The Kaoping River, which originates from the southwestern tip of Central Mountain Range towards the Taiwan Strait, blocks the East and West Epidemic Areas on both sides; 4) The Siouguluan River, which originates from the eastern side of Central Mountain Range towards to the Pacific Ocean, blocks the epidemic spreading to the North. However, in 2014, wildlife officers of Forestry Bureau found two rabid ferret badgers in the tributaries of Siouguluan River (unpublished data). Currently it is controversial whether the two rabid ferret badgers have broken through Siouguluan River because it is difficult to identify the side of the north or south among the complex tributaries; 5) The Changhua County, while is located in the south of Daan River, is considered protected by the following boundaries: The Dadu River is located north; The Bagua Plateau is located east; The Zhuoshui River is located south. The Taiwan Strait is in the west. Since the outbreak of ferret badger rabies in July 2013, the 7 cities or counties, though still exhibiting ferret badgers, do not have any positive ferret badgers since; therefore it is identified as rabies free area. Rabies epidemics among wildlife reservoir host populations within defined regions frequently follow a distinct course. Following an initial epidemic of rabies, which is typically the largest of a possible series of epidemics that may emerge over

time as wildlife rabies enter into a new region to infect previously naive population, a series of successively smaller epidemics may occur at increasing frequency; over time, the periodic epidemic structure of rabies epidemics may become indistinguishable against a background level of sporadic disease [12]. According to the current epidemiologic data, the Taiwan ferret badger virus is still sequestered to the mountainous regions (**Figure 2**).

 Table 3: Incidence of Taiwan ferret badger rabies, Taiwan, July 2013 ~ December 2016.

Items	2013	2014	2015	2016	Total
No. positive ferret badgers	276	147	88	42	553
No. total enzootic cities/counties	9	9	9	9	9
No. enzootic townships	59	62	72	77	77
No. progressed new enzootic townships	-	3	10	5	18

Table 4: The status of ferret badger rabies in East and West Epidemic areas, Taiwan, July 2013 ~ December 2016. Townships of Green Island and Orchid Island in Taitung County were not counted in this research.

No.	Cities and Counties	No. enzootic townships		No. townships no cases found	Total	
		With mountainous landscape	Urbanized and located in level areas	With mountainous landscape	Urbanized and located in level areas	
1	Hualien	4	0	9	0	13
2	Taitung	13	0	1	0	14
3	Pingtung	8	0	4	21	33
4	Taichung	8	0	2	19	29
5	Nantou	12	0	0	1	13
6	Yunlin	2	0	1	17	20
7	Chiayi	7	0	1	10	18
8	Tainan	10	1	2	24	37
9	Kaohsiung	12	0	3	23	38
	Total	76	1	23	115	215

Table 5: Contingency table of Chi-square analysis of ferret badger rabies in Epidemic area Taiwan, July 2013 ~ December 2016. Note: X^2 =133.87>10.827; p<0.001 (the critical value of X^2 =10.827 when α =0.001, df=1).

Landscapes	Enzootic townships	No case found townships	Total
Townships with mountainous landscape	76	23	99
Townships with urbanized and in low-lying area	1	115	116
Total	77	138	215

Studies of ethology and habitat of ferret badgers in China and Taiwan indicate that it is unlikely to see a ferret badger in urbanized areas, because leaves in city parks are cleaned routinely, which causes the soil not moist enough for the growth of earthworms and the like [4]. In addition, the ferret badger is always captured or killed by people or dogs wherever found. Therefore, the urbanization condition in human community is inadequate for ferret badgers' survival. Tung et al. estimated that the populations of household dog and stray dog in Taiwan were 1,565,156 dogs and 86,244 dogs respectively [13]. Studies of the human-animal bond indicate that relying on human resources, dogs mainly live with people, establishing a longstanding and strong bondage with humans to protect their territory [14]. Additionally, when considering the factors of the body mass, poor visual perception, small teeth and weak biting force, the ferret badger is not able to win the competition against dogs in urbanized cities [7]. **Tables 4** and **5** also showed this fact. Additionally, mountainous townships usually provide abundant habitat needs that elevate population density of ferret badgers. Density of ferret badgers above the critical threshold value corresponds to a high probability of rabies occurrence [12].

Conclusion

There are a total of 215 townships in above mentioned 9 enzootic cities or counties. Among the total of 77 enzootic townships, 76 of them are mountainous, only 1 township is urbanized and located in level areas. Chi-square analysis also indicates that townships with mountainous areas are extreme associated with the incidence of ferret badger rabies (p<0.001). Consequently, the difficulty of ferret badgers to survive in urbanized areas seems to be an important factor that affects the epidemic expansions.

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