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Research Article

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Analysis of the relationship between the ratio of man to livestock and malaria incidence in Shandong province, China

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ABSTRACT

Animals play an important role in the host-seeking process of zoophilic and anthropophilic mosquito species. This article discusses existing information and analyzes the relationship between the ratio of man to livestock and malaria incidence, as well as a survey on the number and distribution of people and domestic animals serving as hosts. A correlation analysis was performed to assess the effect to domestic animals. The human blood index of Anopheles sinensis was found to be closely related to the space of livestock enclosures. Specifically, a negative correlation was found between the human blood index and the space of human rooms. Meanwhile, the human blood index was positively correlated with the space of livestock enclosures. A relationship between the ratio of man to livestock and malaria incidence was identified, the regression equation is linear, which is expressed as: y=-377.65+184.97x (correlation coefficient is 0.4997). The relationship between the ratio of man to pig and malaria incidence was more approximately linear, which is expressed as: y=-125.56+54.85x (correlation coefficient is 0.6305). This research has demonstrated the role of livestock in the control of malaria and in the reduction of vector density. Malaria incidence was influenced by the number of livestock and the position of barn in the areas with a predominant An. sinensis population.

Keywords: malaria incidence, *Anopheles sinensis*, the human blood index

INTRODUCTION

Malaria, which is transmitted to humans by the bite of an infected female *Anopheles sinensis*, is a serious disease and causes significant morbidity and mortality. Recently, reports have substantiated that about 3.3 billion people are at risk of malaria, with children under five years of age and pregnant women as the most severely affected (WHO 2011). Outbreaks of malaria had been reported in the 1960s and in the 1970s with the highest prevalence in Shandong province, which is located at the eastern coast of China. *An. sinensis*, the most abundant vector, is common especially in the rice region of the southwestern Shandong. *An. sinensis* is highly zoophilic and anthropophilic. Hence, the number of livestock can affect the human blood index (HBI) of *An. sinensis* and can sequentially influence malaria incidence (Muriu et al. 2008, Pappa et al. 2011). Much speculation has been reported on the prophylactic influence of domestic animals. Over the last century, reports referred to the reduced prevalence of malaria as a result of the active deployment of livestock (Hess & Hayes 1970, Kirnowordoyo & Supalin 1986, Bouma & Rowland 1995, Tirados et al. 2006). Although numerous conclusions had emphasized on the effect of livestock to malaria incidence (Lardeux et al. 2007, Killeen & Smith 2007), the correlations between livestock and malaria incidence have been unconvincing. Most conclusions in previous reports had referred to villages or families as bases. Consequently, the results obtained in a limited area were vulnerable to circumstance. Thus, data on some infectious diseases and on livestock population from 1958 to 1978 in Shandong province were analyzed to explore the relationship between the ratio of man to livestock and malaria incidence. Our findings provided evidence for the formulation of prevention and control strategies against malaria.

EXPERIMENTAL SECTION

The relationships between HBI of An. sinensis and the distribution of people and domestic animals
Field surveys were carried out from 1978 to 1984 in TangKou county, Shandong province, China. People in Tangkou county usually grew rice and raised livestock. Thus, *An. Sinensis*, which has high population density throughout the summer and autumn, is the most common vector to transmit malaria. During the survey and experimentation period, malaria incidence varied between 0.41‰ and 15.72‰. Most families raised pigs and goats, whereas cattle and horse which belonged to commune were all together raised.

Female adult mosquitoes were collected by aspirator every hour from 18:00 to 21:00 and were transported to the laboratory. Traps were spaced at 10–100 m distances from the human dwelling and 30–275 m distances from the barn to evaluate the relationship between the effective distance used in each livestock enclosure and HBI. Blood-meal smears were collected to determine the feeding preferences and to enable HBI estimation. The number of animals and their distance from the collectors were then recorded, and the HBI of *An. sinensis* was calculated.

Appraisal of blood-meal
All identified blood meal samples were tested with anti-human, anti-pig, anti-cattle, anti-horse, and anti-sheep sera using the precipitation method. Antigen blood serum was produced by immunizing white rabbits with animal blood serum. The immunity method is described as follows: White rabbits were injected the day after the antigen blood serum was extracted. The total number of immunizations was nine times. The first injection of 1 ml blood serum from the animals was administered via the ear vein. The second, third, and fourth boosters were then injected subcutaneously at 1 ml each. The remaining boosters were injected in the same way as the first booster. However, the dosages were 1, 2, 3, 4, and 5 ml. Ten days later, the antigen blood serum was extracted.

The filter paper adsorbing blood from the midgut of the mosquitoes was placed into the test tube. Following the specified dosage of blood, 1 to 3 ml normal saline was added to the test tube and left to stand for 8 h. The antigen blood serum (1:2) was diluted through glycerin brine (14%), and then added with the diluent serum (0.1 ml) into another test tube. Subsequently, the sediment serum was blocked with the blood smear extracts for about 0.5 h at 37 °C. Positive reactions were identified with a white neck ring at the antigen-antibody interfaces.

Size of the population and livestock
Malaria incidence information of Shandong province from 1958 to 1978 was supplied by the Shandong Institute of Parasitic Diseases. Data on the size of the human and livestock population were provided by the government department. The data were verified for accuracy through a sampling survey and by checking reports on infectious diseases.

Statistical analysis
The data were plotted using Microsoft Excel (2003) and analyzed using the statistical analysis system (SAS). Pair *t*-tests were used to compare the differences among the human bait and livestock because of the dependence of the data at the same collection site. Other data were examined through regression analysis, and *P* values were calculated.

RESULTS

Relationships between HBI of An. sinensis and the distribution of people and domestic animals
During the course of this investigation, a total of 3932 blood-feeding and 289 human blood-feeding anophelines were collected. Low human landing rates of *An. sinensis* were recorded during the entire study period. The number of *An. sinensis* significantly varied among the human and the livestock subjects (T=4.1, P=0.0018). The data in Table 1 were examined through multiple regression analysis to get an idea of the relationship between HBI of *An. sinensis* and the distribution of people and domestic animals. The formula on HBI (y) and the distances to the human dwelling and to the barn are written as: $y=10.346-0.3875\ x_1+0.1807\ x_2$, where x_1 is the distance to the human dwelling and x_2 is the distance to the barn. The correlation coefficient was 0.7787. Therefore, HBI is higher when closer to the human dwelling.

Table 1 Human blood index and the distribution of people and domestic animals

Distance from the collectors		No. of Blood meals	No. of human bait	Human blood index
house	barn			
30	80	226	40	17.7
10	30	195	33	16.9
100	275	93	9	9.7
5	250	49	40	81.6
50	100	525	27	5.1
50	50	69	1	1.4
100	50	144	2	1.4
37.8	122.8	726	38	5.2
42.4	87.3	512	38	7.4
18.4	124.4	680	36	5.3
21.5	77.3	180	9	5.0
52.9	66.5	533	16	3.0

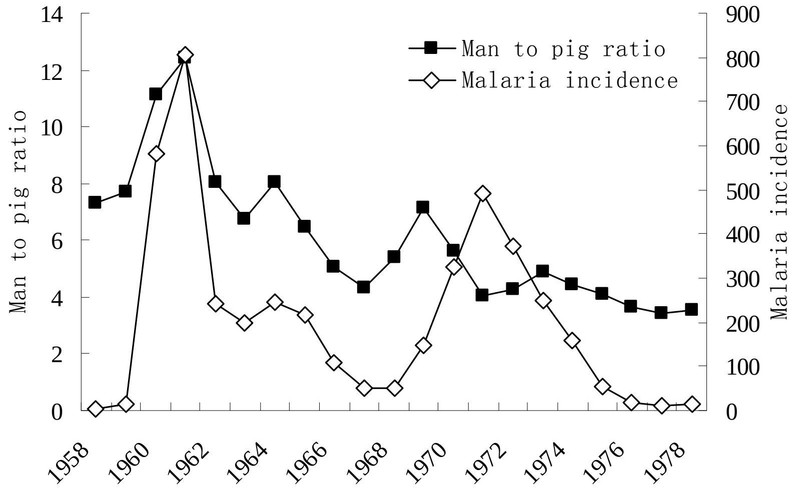

Figure 1 Man to pig ratio and malaria incidence

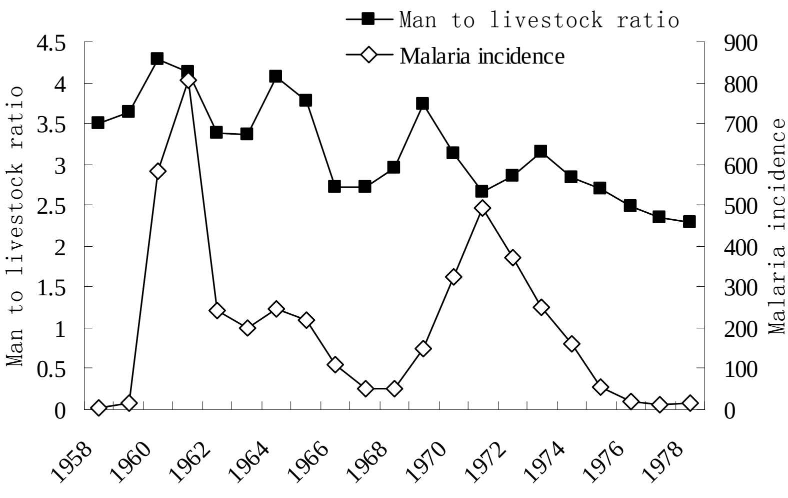

Figure 2 Man to livestock ratio and malaria incidence

Correlations between the ratio of man to livestock and malaria incidence

In this study, the range, probably governed by the varying ratios of man to livestock, was between 2.28 and 4.28. Meanwhile, the ratio of man to pig was between 3.45 and 12.45. Combining the present evidence with the data reported, malaria incidence in Shandong province was between 3.63‰ and 806.17‰ from 1958 to 1978. Figure 1 shows that four peak ratios of man to livestock were observed: 1960, 1964, 1969, and 1973. Meanwhile, the ratio of man to pig exhibited similar trends, as shown in Figure 2. More importantly, peaks of malaria incidence mainly occurred in 1961, 1964, and 1971. A slight difference was observed among the peaks of the man to livestock ratios, the man to pig ratios, and malaria incidence rates. Despite this slight difference, the trend of the correlation between the man to livestock ratio and malaria incidence is clear, that is, with the increase in man to livestock ratio, malaria incidence also increased. Regression analysis was conducted via the regression function in the statistical analysis system. The result showed a significant linear relation between malaria incidence (y) and the man to pig ratio (x_1). The regression equation is therefore linear, which is expressed as: $y=-125.56+54.85x_1$ (correlation coefficient is 0.6305). Meanwhile, a linear relation between malaria incidence (y) and the man to livestock ratio (x_2) was identified. The regression equation is also linear, which is written as: $y=-377.65+184.97x_2$ (correlation coefficient is 0.4997).

DISCUSSION

An. sinensis, the primary malaria vector, is highly zoophilic and exophilic. Hence, they like to feed on blood and express predominantly zoophilic characteristics. *An. sinensis* initially bites livestock when people and animals co-exist in a community. Thus, livestock can possibly be used to reduce the vector's HBI and malaria incidence in the population (Garrett-Jones et al.1980, Kaburi et al.2009). After a seven-year survey in 12 locations, a number of biting mosquitoes were collected. The relationship between HBI and the distance between the collectors and the host were analyzed. The number of animals and their distances from the collectors revealed an osculatory correlation, that is, the closer the collector is from the barn, the smaller the HBI, whereas the closer the collector is from the human dwelling, the bigger the HBI. Correspondingly, a strong correlation between trap spacing and livestock enclosure was observed. Trap spacing between 4 and 7 m apart captured mosquitoes more efficiently than other trap spacing distances (Lee et al.2009). The present study shed further light on the positive impact of livestock in reducing man-vector contact. Consequently, the position of the barn should be determined according to the feeding habit of the vector to reduce its HBI.

This study has demonstrated that a correlation exists between the ratio of man to livestock and malaria incidence for long-term trends. A strong correlation was also evident between the increasing man to pig ratio and the increasing prevalence of malaria. This evidence suggested a direct link among the proximity of livestock, the reduction of human-biting incidences, and the decreased malaria prevalence. Analytical results demonstrated that the number of pigs more efficiently reduced malaria incidence than the other animals in the community. The main reasons are as follows. During the study period, large domestic animals, especially horse and cattle, were raised far from the human dwelling, whereas the pigs were raised in close proximity with their human owners. In addition, the pigs were defenseless against mosquitoes in this study.

Meanwhile, increased prevalence of malaria has been reported due to the diminished number of livestock. Cattle and pigs can actually form barriers between breeding sites and human settlements. Instances where the introduction of livestock has apparently reduced the prevalence of disease have also been reported. Reduction in the incidence rate of malaria has been attributed partly to the increase in the livestock population (Mahande et al.2007). Animals, such as cattle and buffalo, have been used as baits to reduce vector densities (Hewitt & Rowland 1999,Manh et al. 2010). Conversely, a few instances have been cited that the prevalence of malaria was higher in villages where enclosed cattle sheds were closely situated around human dwellings than in villages where open cattle sheds were located far from human dwellings (Hewitt et al. 1994, Huldén & Huldén 2009, Schultz 1989). Unlike the study mentioned above, the present research first analyzed the data in Shandong province from 1958 to 1978. Past studies were conducted in small-scale trials (village and family). In several malaria-endemic countries and territories, the species composition and distribution of *Anopheles* vary in various habitats. Several malaria vectors may even exist in the same location. These differences indicate that malaria vectors necessarily inhabit a region that offers various opportunities for host selection. Livestock plays a different role for malaria transmission on account of the vectors feeding on different blood characteristics. *An. sinensis* was identified as the most common malaria vector in Shandong province. On the other hand, the distribution of livestock in different villages differs from the distribution of people. As a result, the ratio of man to livestock varies in different areas. This heterogeneity affects HBI, which is closely related to malaria incidence. Finally, a shift in agricultural practices toward crop production resulted in a dramatic decrease in livestock numbers in Shandong province, as observed in the 1970s. Meanwhile, the government has a high regard for the elimination of the vector with the use of insecticide-treated nets and for the prevention of malaria through drug prophylaxis or drug treatment. Moreover, long-term social changes had contributed to the obvious reduction of malaria incidence in Shandong Provence.

The prevalence of malaria is affected by several factors. Domestic animals are likely to have a promising prophylactic influence when the vector is zoophilic and anthropophilic. This condition can have an impact on the long-term trend of malaria. Therefore, livestock should be deployed based on the actual environmental conditions to form a barrier between the vector and the human population. Moreover, malaria transmission can be reduced by zooprophylaxis in areas with a predominant *An. sinensis* population.

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