An Epidemiological and Diagnostic Study of The Microfilaria Parasite in Cows in Western Regions of Iraq

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Abstract. The current study was conducted to find out the prevalence of Microfilaria in cows in the western regions of Iraq, by examining 382 cows of both sexes and for ages from 1-11 years during the year 2020. The parasite was diagnosed by examining the skin scraping collected from the infected animals in the laboratory, and the results showed that the cows were infected with the microfilariae parasite by 34%, and the highest infection rate was recorded in Al-Qa'im area 51.6% and Haditha 49.%. Significant differences were observed between males (21.9%) and females (44.6%). The highest incidence rate was recorded in cows aged 3-5 years, at 45.3%, with significant differences from the rest of the ages. Furthermore, the highest incidence rate was observed in Summer seasons, reaching 64.8%. The current study concludes that cattle in the western regions of Iraq are infected with the microfilariae parasite at a high rate, and this necessitates the establishment of an integrated control approach to control parasitic diseases and prevent their spread in farm animals because they cause serious disease effects and heavy economic losses.

1. Introduction
Microfilaria larvae belong to the parasitic nematodes that parasitize many animals such as cattle and buffaloes and also infect humans and cause filariasis [1]. It is an early stage in the life cycle of some nematodes, includes several genera of the Rhabditida order and the Filariid family [2]. Infection occurs by some types of blood-sucking insects, especially black flies, midges, and mosquitoes, which are the intermediate hosts that transmit many species of these parasites [3]. Intermediate hosts, or vectors, feed on the blood of the final host and take with the blood meals the microfilaria [4]. Where they develop into the infectious phase and when blood is absorbed by the insects again the larva moves to the final host to complete its development into adult worms [5]. These larvae resemble worms, and their length ranges from a few millimeters to tens of centimeters [6].

They are found in different places of the host's body such as arteries, subcutaneous tissues, for years and cause local inflammation [7]. It causes many pathological symptoms, the most important of which are severe itching, stiffness of the skin, the appearance of crusts, thickening of the skin, the appearance of many nodules on the surface of the skin, and hair loss [8]. The female filaria lays a huge amount of eggs during her life years, from which the microfilaria larvae hatch and the incubation period is characterized by calmness and lasts between 6 months - 3 years [9]. Filariasis is most common in tropical and subtropical regions in developing countries [10]. Where the prevalence of
infection with this parasite depends on the availability of hot climatic conditions and the availability of intermediate hosts [11].

There are several species of parasitic filarial worms, the most common of which are Wuchereria bancrofti, Onchocerca volvulus, and Brugia malayi [12]. In some types of Onchocercidae, the release of microfilariae by the adult female is cyclic - occurring daily at a specific time of the day or night, this timing increases the chance of getting picked up by a blood-feeding arthropod, which is often most active at certain times of the day [13]. Cows is an important animal and economic wealth in many countries of the world, including Iraq, as it is a major source for the production of meat, milk, and leather [14].

In view of the great economic importance of cows and the lack of studies in the country on microfilariae parasites and their danger to livestock due to the great disease effects and heavy economic losses they cause on different farm animals, this study was conducted to determine the extent of this parasite's spread in cows in the western regions of Iraq to determine the health reality of this wealth Important animal. Further monitoring is warranted to determine the occurrence of this disease, to identify its vectors, and to take appropriate measures to protect the livestock and livestock industry in the country.

2. Materials and Methods

2.1. Collecting and examining a specimens
A field survey was carried out during the year 2020, with 3 visits per week to four rural areas in the western regions of Iraq, which included Included (Al-Qa'im, Haditha, Rutba, and Al- Badgdadi) during which 382 cows' heads were examined for both sexes and for ages 1-11 years. The parasite was diagnosed by examining skin scrapings that were collected from animals infected with skin lesions in the Parasitology Laboratory at the College of Science - University of Anbar.

2.2. Collecting and examining a specimens
Skin scrapings were collected from cows affected by skin lesions after observing the clinical symptoms on the animal, represented by severe itching, the appearance of crusts, thickening, stiffness, and hair loss. A sterile scalpel was used to scrape the skin and placed in glass bottles containing 10 ml of a solution of potassium hydroxide KOH at a concentration of 10% and brought to the laboratory for examination, where it was placed in a water bath of 80 for 20 minutes to get rid of the tissues and fibers as the solution works to dissolve the tissues without affecting the parasite after the specimens were cooled, they were placed in a centrifuge at a speed of 1500 rpm for 5 minutes, after which the clear portion was poured, droplets were taken from the precipitate and placed on a glass slide and examined under a light microscope on a force of 10 x and 40 x magnification to watch cutaneous microfilaria parasite according to a method [15].

2.3. Statistical Analysis
To statistically analyze the data, the chi-square test, was used to determine the lowest significant difference L.S.D at the probability level $P \leq 0.05$ using a set of ready-made statistical programs SAS [16].

3. Results and Discussion
Microfilaria are known to occur among domestic animals almost all over the world, the economically most important and also most abundant filarioid nematodes in cattle, this calls for intensive studies to detect the prevalence of these parasites. The results showed that cows in the western regions of Iraq were infected with larvae microfilaria at a total rate of 34% by examining the scales taken from skin lesions. The cutaneous microfilaria, dyed by the blue methylene stian, appeared in the form of long, cylindrical filamentous larvae that were not sheathed, and their length ranged between 220-260 microns and an average of 240 microns and belonged to the genus *Onchocerca* sp. (Figure 1).
The highest rate of infection was recorded in the Al-Qa'im region, 51.6%, and Haditha, 49%, and the lowest in the Al-Badgdadi region, 11.7%, with significant differences p <0.05 (Table 1). These results are consistent with what Nimisha et al., [17] found, as the incidence rate was 35.4% in northern Kerala, but its higher than that recorded by Al-Lahaibi and Al-Taee [18] in the city of Mosul, where the total infection rate reached 13.2%, and also higher than what Happi et al., [19] found in Nigeria, where the rate of infection with Microfilaria was 5%. As well as higher than what recorded by Chandratre et al., [20]. The reason for the high rate of infection with the microfilaria parasite of the cows in the western regions of Iraq is due to the heavy presence of the intermediate host of the black flies and mosquitoes, where the availability of appropriate climatic conditions, such as high temperatures and appropriate humidity, helps the reproduction and spread of insects.

And also the lack of attention to the cleanliness of the livestock fields and the failure to get rid of the excreta and the gathering of dirt which is a suitable medium for the reproduction and spread of insects, and it was observed that a large proportion of black flies, which are the intermediate host for the transmission of cutaneous microfilaria, were found on the ear areas and around the head of the affected animals in the cows breeding fields. As for the discrepancy between the infection rate between the current study and the studies that preceded it, it is attributed to many reasons, including the difference in the seasons of the study, the methods of breeding in the fields of livestock breeding, the different regions of study, the breed, and age of the animals. (Figure 2) shows the cows infested with microfilaria parasite, where skin thickening, stiffness, hair loss, and crusting are observed.

Table 1. percentage of infection with Microfilaria parasite according to region.

| Region         | Number of cows examined | Number of cows infected | %  |
|----------------|-------------------------|-------------------------|----|
| Al-Qa'im       | 89                      | 46                      | 51.6|
| Haditha        | 104                     | 51                      | 49.0|
| Rutba          | 95                      | 22                      | 23.1|
| Al-Badgdadi    | 94                      | 11                      | 11.7|
| Total          | 382                     | 130                     | 34.0|

Figure 2. Skin lesion of cows infected with Microfilaria parasite.
The results showed that sex had an effect on infection rates, and the statistical analysis showed that there were significant differences P≤0.05 between the infection rates of both female and male buffalo, which amounted to 44.6 and 21.9%, respectively (Table 2). This is in agreement with the Rajendran study [21] in which it was found that the infection rate of females is higher than that of males in India, and the reason for the high rate of infection among females is attributed to the fact that livestock keepers in villages and rural areas keep females for a long time more than they keep males for purposes of reproduction and milk production while Males are bred for fattening purposes and sold early, so the time for females to stay in the fields is more than males, and this increases the chances of females being exposed to disease parasites at a higher rate than males.

The results showed that the highest rate of infection in cows was at older ages more than 3 years, which amounted to 45.3, 42.4% for ages 3-5, and 6-8 years respectively, while the lowest infection rate was 15.2% for ages 2 months - 2 years (Table 3), this is in agreement with the results of Patil et al., [22] and the results of Bordoloi et al., [23] in India. The reason for this is that livestock of large ages is more susceptible to insect stings through the passage of these animals in several seasons, especially in which the spread of insects increases in the fields of livestock breeding which causes a high infection rate. And mentioned [24] that the incidence of Microfilaria increases at older ages. Shahatha [25] reported that younger animals have increased immunity against parasites due to their obtaining of antibodies with colostrum during breastfeeding compared to their mothers, which makes them resistant to parasitic infections and this leads to a lower rate of infection. Kuttler [26] stated that all ages are susceptible to infection with parasites, but that older animals are more susceptible than young animals.

Table 2. Percentage of infection with Microfilaria parasite according to sex.

| sex     | Number of cows examined | Number of cows infected | %   |
|---------|-------------------------|-------------------------|-----|
| Males   | 178                     | 39                      | 21.9|
| Females | 204                     | 91                      | 44.6|
| Total   | 382                     | 130                     | 34.0|

Table 3. Percentage of infection with Microfilaria parasite according to Age.

| Seasons | Number of cows examined | Number of cows infected | %   |
|---------|-------------------------|-------------------------|-----|
| Winter  | 98                      | 16                      | 16.3|
| Spring  | 90                      | 26                      | 28.8|
| Summer  | 94                      | 61                      | 64.8|
| Autumn  | 100                     | 27                      | 27.0|
| Total   | 382                     | 130                     | 34.0|

Table 4. Percentage of infection with Microfilaria according to Seasons of the year.

| Age (years) | Number of cows examined | Number of cows infected | %   |
|-------------|-------------------------|-------------------------|-----|
| 2 months - 2| 92                      | 14                      | 15.2|
| 3 - 5       | 97                      | 44                      | 45.3|
| 6- 8        | 99                      | 42                      | 42.4|
| 9-11        | 94                      | 30                      | 31.9|
| Total       | 382                     | 130                     | 34.0|

The highest infestation rate was in the Summer season, reaching 64.8%, and the lowest was 16.3% in the Winter season (Table 4). This is in agreement with the findings of Phukan et al., [27] and the reason for this is due to the appropriate climatic conditions of temperature and humidity during the
summer months for the reproduction of insects that vector the parasite, such as black flies, midges, and mosquitoes. He indicated de León et al., [28] that the rate of infection with these parasites increases in hot and warm seasons. Shahatha [29] indicated that the multiplication and spread of insects, especially flies and mosquitoes, as well as environmental and climatic conditions favorable for the growth of parasites during the summer season lead to a high rate of infection.

4. Conclusion
This study concludes that cows were infected with Microfilaria parasite the western regions of Iraq with a high percentage. Consequently, the current study provides exploratory indicative data for future monitoring of parasitic diseases of medical importance, identifying risk factors, and reducing economic loss to map strategies for controlling these diseases. Therefore, the veterinary authority must implement a control program to control these diseases by treating them using medicinal plants, improving the standard slaughterhouse system, and eliminating vector hosts.

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