

# *Anopheles arabiensis*: abundance and insecticide resistance in an irrigated area of eastern Sudan

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توافر الأنوفيلة العربية ومقاومتها للمبيدات في منطقة مروية في شرق السودان  
يوسف الصافي حمدان، مصطفى دكين، الأمين الرياح، إسحاق آدم

**الخلاصة:** درس بواهر الأنوفيلة العربية ومدى تأثرها بالمبيدات في منطقة حلفا الجديدة، شرق السودان، خلال الفترة من آذار/مارس 1999 وحزيران/يونيو 2000. ومن بين الإناث من الأنوفيلات التي تم جمعها والتي بلغ عددها 4854 كان 4847 منها (99.9%) من الأنوفيلات العربية، فيما كان 7 منها (0.1%) من الأنوفيلات الفرعونية. إن الإناث من الأنوفيلات العربية تواصل سراها (تكاثرها) طيلة العام مع فصلين للنسب الكثيف، أسدما خلال الفصل المطير (حيث وجدت 158.4 أنثى/اليوم/الغرفة مع 84.7 من اليرقات في كل عشر غمسات) والثاني خلال فصل الري (حيث وجد 136.8 من الإناث/اليوم/الغرفة مع 44.8 من اليرقات في كل عشر غمسات). وكان متوسط نشاط العض 28.8 عضه/شخص/ليلة، وتوزع العضات على مدار الليل، وبشكل خاص خارج المنزل. وقد كان معدل تأثر الأنوفيلات العربية بالدد.د.ت 97.8% وبالملاثيون 96.3% وبالفينيتريون 100%. إن الأنوفيلة العربية هي الناقل الوحيد للملاريا في المنطقة، وهي ذات نشاط مستمر كامل العام ولا يقتصر على فصل دون آخر.

**ABSTRACT** The abundance of *Anopheles arabiensis* and its susceptibility to insecticides was studied in New Halfa, eastern Sudan, from March 1999 to June 2000. Of 4854 females anophelines collected, 4847 (99.9%) were *An. arabiensis* and 7 (0.1%) *An. pharoensis*. Female *An. arabiensis* were breeding throughout the year, with 2 peak densities, during the rainy (158.4 females/room/day and 84.7 larvae/10 dips) and irrigated seasons (136.8 females/room/day and 44.8 larvae/10 dips). The mean biting activity was 28.8 bites/person/night, found throughout the night, mainly outdoors. Susceptibility of *An. arabiensis* to insecticides dichlorodiphenyl-trichloroethane (DDT), malathion and fenitrothion was 97.8%, 96.3% and 100% respectively. *An. arabiensis* is the sole malaria vector in the area and is perennial rather than seasonal.

## *Anopheles arabiensis* : abondance et résistance aux insecticides dans une zone irriguée du Soudan oriental

**RESUME** L'abondance d'*Anopheles arabiensis* et sa sensibilité aux insecticides ont fait l'objet d'une étude à New Halfa (Soudan oriental) de mars 1999 à juin 2000. Sur 4854 anophèles femelles récoltées, 4847 (99,9 %) appartenaient à l'espèce *An. arabiensis* et 7 (0,1 %) à *An. pharoensis*. Les femelles *An. arabiensis* se reproduisaient toute l'année, avec 2 pics de densité, pendant la saison des pluies (158,4 femelles/pièce/jour et 84,7 larves/10 prélèvements) et la saison d'irrigation (136,8 femelles/pièce/jour et 44,8 larves/10 prélèvements). La densité agressive moyenne était de 28,8 piqûres/personne/nuit, constatée pendant toute la nuit, principalement à l'extérieur. La sensibilité d'*An. arabiensis* aux insecticides dichloro diphenyl-trichloroéthane (DDT), malathion et fénitrothion était de 97,8 %, 96,3 % et 100 % respectivement. *An. arabiensis* est le seul vecteur du paludisme dans la région et la transmission y est pérenne plutôt que saisonnière.

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## Introduction

Malaria is a major health problem in the tropical countries, especially sub-Saharan Africa, where about 90% of the clinical cases occur. There are nearly 500 million clinical cases of malaria worldwide each year and 1.1 to 2.7 million people die annually [1]. The *Anopheles gambiae* complex is the main malaria vector [2]. In Sudan, malaria constitutes around 40% of all infectious diseases and *Plasmodium falciparum* is the predominant species, which is responsible for over 90% of the infections [3]. The spread of drug-resistant *P. falciparum* strains worldwide has hampered the control of malaria, and vector control remains the best approach for protecting the community against malaria [4].

*An. arabiensis* is the major malaria vector reported from all parts of the country, co-existing with *An. gambiae sensu stricto* (*s.s.*) and *An. funestus* in southern Sudan [5-8]. Vector control by chlorinated hydrocarbon insecticides was started in Sudan in 1948, when dichloro-diphenyl-trichloroethane (DDT) sprayed with oil was used as a residual indoor insecticide. Thereafter, organized control by these chemicals was initiated in 1950/51 in the Gezira scheme (central Sudan) but by the early 1970s, resistance to DDT was reported. Thereafter, the organophosphate insecticides malathion and fenitrothion replaced DDT [9,10].

Understanding the behaviour of the malaria vectors and their abundance is essential for malaria control operations. Furthermore, understanding the resting, biting and breeding habits of the vectors and their susceptibility to insecticides is extremely important for planning, implementing and monitoring vector control measures. The objective of this study was to establish this data for the New Halfa area of Sudan.

## Methods

### Study area

The study was carried out at 2 localities (Dibaira camp and Heielmasakine) in the north and south of New Halfa town, an area which is surrounded by freehold farms. The New Halfa area is located in the semi-arid belt of the Sudan approximately 500 km east of Khartoum in the middle of an agricultural scheme. During the study period, the total rainfall was 431.6 mm and the average temperature was 30 °C. *P. falciparum* is the predominant malaria parasite species, and has been shown to be 75% and 9.6% resistant to chloroquine and quinine respectively [11].

### Mosquito collection

Hand capture and pyrethrum spray methods were used to make monthly collections of indoor resting mosquitoes during the period March 1999 to June 2000. Collection by hand capture was carried out from 06:00 to 08:00 hours. Two collectors spent about 10-15 minutes in each hut (total 10 huts) using a torch, aspirators and paper cups to search for resting anopheline mosquitoes. Then 0.3% pyrethrum in kerosene was sprayed and knocked-down mosquitoes were collected from 08:00 to 10:00 hours. Females collected were classified as according to their blood meal stages unfed, fed, half-gravid/gravid.

The monthly night biting collection was carried out in fixed collection sites during the main transmission season by 2 people (1 indoors and 1 outdoor) sitting from 18:00 to 06:00 hours. Females collected were kept in separate paper cups until identification and dissection for parity determination. The ovaries were dissected to determine the parity rate using the method of Detinova [12]. Blood spots from freshly fed females of *An. arabiensis* caught by

pyrethrum spray catches were harvested on filter paper, the source of each blood determined (human biting index) using the Ouchterlony radial diffusion technique [8].

The surveys of immature stages were carried out from 13:00 to 15:30 hours using a standard dipper. Monthly samples of 100 dips were taken at 10 positive breeding sites selected randomly to cover the whole town. Adult and larval mosquitoes were identified on a morphological basis using the standard keys [13,14].

### Susceptibility testing

The susceptibility of *An. arabiensis* to DDT, malathion and fenitrothion was examined in 3 locations (Wad el Naeem, Heiomasakine and Umgargoor). According to World Health Organization (WHO) recommendations [15], fed females were exposed for 1 hour to 4% DDT, 5% malathion and 1% fenitrothion to determine their susceptibility to these insecticides.

### Statistical analysis

Data was entered into a computer database and SPSS software was used for statistical analysis. The difference between variables was evaluated using the chi-squared test. Student *t*-test was used to evaluate the difference in the density of the vector in the 2

localities and the human biting rate between the outdoor and the indoor sites. One-way analysis of variance was used to compare the human biting rates at different times of the night. A *P* value of less than 0.05 was considered significant.

### Results

Out of 4854 females collected, 4847 (99.9%) were *An. arabiensis* and 7 (0.1%) were *An. pharoensis*. Most of the *An. arabiensis* specimens were collected by pyrethrum spray (4164, 85.9%) rather than hand capture (683, 14.1%).

The mean vector density in the Heiomasakine area (pyrethrum collection) was 29.3 females/room/day, while it was 23.1 females/room/day in Dibaira camp (Table 1). The highest density was recorded in September 1999 by the end of the rainy season (158.4 females/room/day) in Heiomasakine while the lowest density in this area was recorded in April 1999 (0.2 females/room/day). The peak density of adults was reflected by the peak of immature stages in the same month (84.7 larvae/10 dips). The lowest density of adult females was recorded in March 2000 (0.4 females/room/day) in Dibaira camp. The

Table 1 Total number of adult *Anopheles arabiensis* specimens collected during the study period by hand capture and pyrethrum spray and mean density per room per day in the 2 localities

Locality	Pyrethrum spray		Hand capture		Total	
	Total collected No.	Mean $\pm$ SE density No./room/day	Total collected No.	Mean $\pm$ SE density No./room/day	Total collected No.	Mean $\pm$ SE density No./room/day
Dibaira camp	1821	23.1 $\pm$ 8.8	311	3.7 $\pm$ 1.4	2132	26.9 $\pm$ 10.1
Heiomasakine	2343	29.3 $\pm$ 13.0	372	4.6 $\pm$ 1.5	2715	33.9 $\pm$ 14.5
Total	4164	26.0 $\pm$ 7.9	683	4.2 $\pm$ 1.0	4847	30.3 $\pm$ 8.9

SE = standard error.

lowest density of immature stages was recorded in that month (0.54 larvae/10 dips). A minor peak of female density was recorded during the period of irrigation (March 1999), 136.8 females/room/day in Dibaira camp, coupled with a minor peak of immature stages (44.8 larvae/10 dips) in the same month (Figures 1 and 2).

Of 2132 females collected from indoors resting sites in Dibaira camp, 14.1%, 42.5% and 43.3 % were unfed, fed and half-gravid/gravid respectively. While from Heicmasakine, 14.4%, 36.5% and 49.1% were unfed, fed and half-gravid/gravid. The ratio of fed to gravid/half gravid females in the 2 localities considered together was 1:1.2 in favour of gravid/half-gravid. This indicates that this species is more endophilic than exophilic. A total of 4.1 females/room/day were captured on thatched

walls compared with 1.6 females/room/day on mud walls.

Of the *An. arabiensis* females captured while trying to bite human baits, 76.2% were caught outdoors and 23.8% indoors. The average human biting rate during the rainy season was significantly higher for outdoor biting activity (28.8 bites/person/night) than indoors (9.0 bites/person/night) ( $P < 0.001$ ). The indoor biting activity started initially with a moderate value at 18:00–02:00 hours (0.88 bites/person/hour), followed by a peak of biting activity at 20:00–22:00 (1.13 bites/person/hour) and then dropped to lower activity at 24:00–02:00 (0.25 bites/person/hour). A minor peak was observed at 02:00–06:00 (0.75 bites/person/hour). In contrast, the outdoor biting activity started with a peak value at 18.00–20.00 hours (5.0 bites/per-

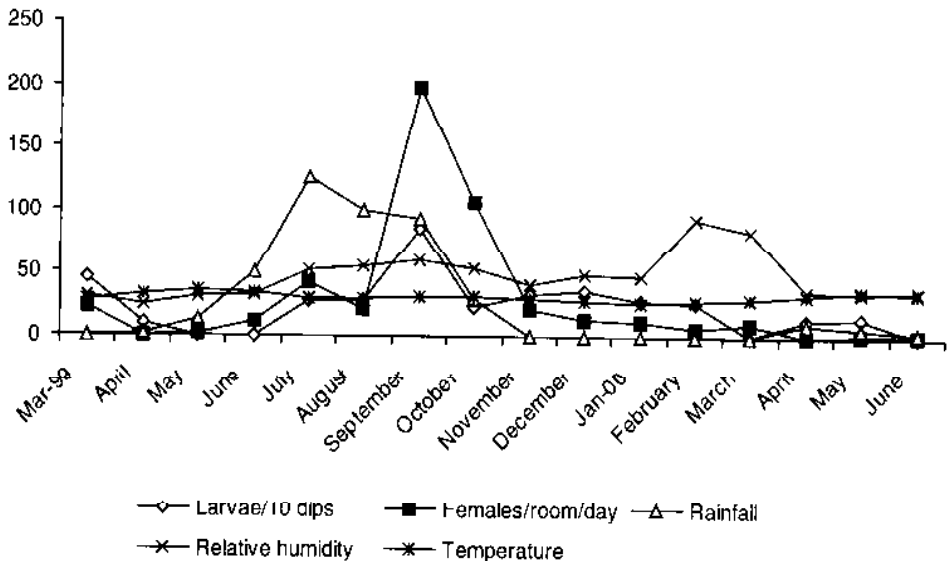


Figure 1 Abundance of both adult females and larvae of *Anopheles arabiensis* in relation to rainfall (mm), relative humidity (%) and temperature (°C) in Dibaira camp

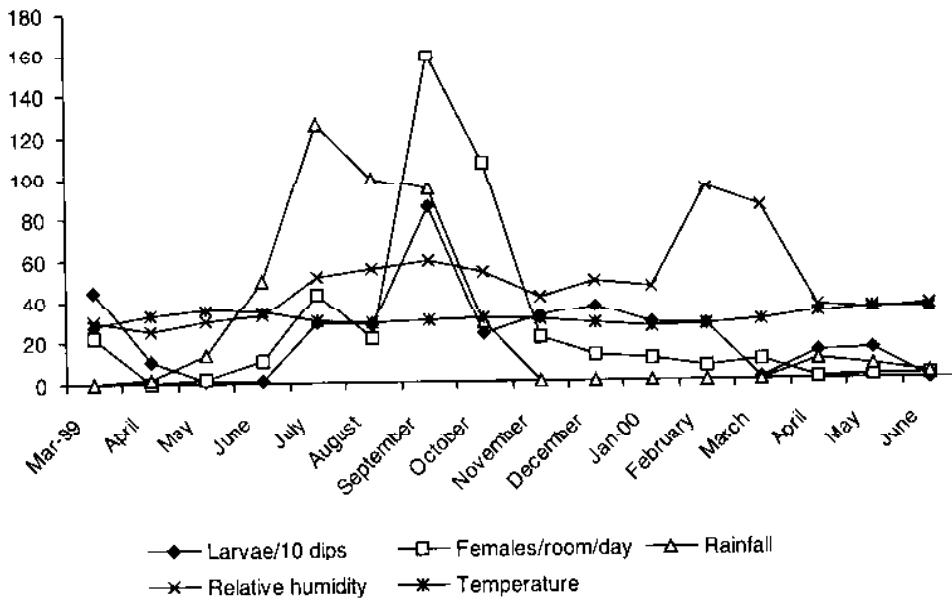


Figure 2 Abundance of both adult females and larvae of *Anopheles arabiensis* in relation to rainfall (mm), relative humidity (%) and temperature (°C) in Heielmasakine

son/hour) decreased at 20:00–22:00 to 3.25 bites/person/hour and dropped to lower values (0.25 bites/person/hour) between 02:00–06:00. The difference was not statistically significant ( $P > 0.05$ ).

The monthly parity rates of *An. arabiensis* during the rainy season were 21.4%, 6.7%, 42.5% and 36.4% in July, August, September and October respectively, with a mean of 32.2% during this period. The human biting index of the 2 localities were 76.4% and 81.1% in Dibaira camp and Heielmasakine respectively, with a mean of 78.7% of the total mosquitoes examined.

Out of a total of 3849 larvae collected, 21.1%, 29.2%, 24.6% and 15.7% were first, second, third and fourth instar larvae respectively. Most (82.5%) of these larvae were collected from the shallow sunlit

pools resulting from broken water pipes. Other breeding sites were rain pools (11.6%), leakage from irrigation canals (3.1%) and water excavations for human and animal use (2.9%). The mean total number of larvae collected during the study period was 24.3 larvae/10 dips.

The susceptibility of *An. arabiensis* to the 3 insecticides was 97.8%, 96.3% and 100.0% for 4% DDT, 5% malathion and 1% fenitrothion respectively (Table 2).

## Discussion

This entomological study was conducted in an agricultural area in eastern Sudan, characterized by a high level of chloroquine-resistant falciparum malaria [11]. *An.*

Table 2 Susceptibility to insecticides of adult *A. arabiensis* collected from 3 localities

Locality	DDT 4%			Malathion 5%			Fenitrothion 1%		
	Total tested	Suscep	Resist	Total tested	Suscep	Resist	Total tested	Suscep	Resist
	No.	%	%	No.	%	%	No.	%	%
Wad el Naeem	75	97.3	2.7	75	98.7	1.3	75	100.0	0
Heielmasakine	60	98.3	1.7	40	100.0	0	40	100.0	0
Umgargoor	100	100.0	0	100	93.0	7.0	100	100.0	0
Total	235	97.8	2.2	215	96.3	3.7	215	100.0	0

*Suscep* = susceptible.

*Resist* = resistant.

*arabiensis* was the main vector (99.9%) found in the area; only 0.1% were *An. pharoensis* and no other species were detected. This agrees with a previous study from the nearby area (Gedaref) in the eastern Sudan where *An. arabiensis* was the main vector, besides 2 other species, *An. pharoensis* and *An. funestus* [8].

*An. arabiensis* existed throughout the year with 2 peaks, a major one at the end of the rainy season (September) and the other during the irrigated season of the scheme. The second peak is most likely due to presence of breeding sites that were formed from the puddles of irrigation canals around the area. Thus *An. arabiensis* has become perennial instead of seasonal because of irrigation and agricultural practices. Similar findings were reported from an irrigated area in the Gezira scheme in central Sudan [16], but unlike our study, a single peak of vector density was observed at the end of the short rainy season that dropped gradually to disappear in the long, hot dry season [8]. The most important factor leading to high breeding density during the rainy season was the optimal temperature (28.9–30.1 °C) and high relative humidity, as it is known that high temperature and low humidity adversely affect adult and immature stages [17]. This trend

of mosquito abundance differs from other tropical areas that are characterized by longer rainy seasons and more humid conditions, e.g. southern Sudan and many other African countries. In these regions, anopheline mosquitoes are known to be prevalent throughout the year [18].

Our study showed that *An. arabiensis* clearly preferred thatched to mud walls as a resting surface. A similar finding was observed from central Ethiopia [19].

In this area *An. arabiensis* is strongly exophagic. This finding is supported by the previous study in Gedaref state and central Ethiopia [8,19]. Three factors seem to determine the biting cycle of the *An. arabiensis*: rhythmic activity of the mosquitoes, the microclimate and human habits [20]. Winds and temperature are the climatic factors affecting the peaks of the biting cycle [18]. In the Zwai area of central Ethiopia the peak of outdoor biting occurred from 22:00 to 24:00 hours, whereas the peak of indoor biting took place early in the evening from 18:00 to 20:00 [19]. However, the vector was aggressive throughout the night and exhibited 2 peaks in the indoor biting activity at 20:00 to 22:00 and later at 02:00 to 06:00, with the peak outdoor biting activity at 18:00 to 22:00.

The low parity rate of female mosquitoes in this area is likely to be related to the application of the insecticide K-Othrine (a water-based formulation of deltamethrin) during the period of the study in August 1999, as shown by the low parity rate that month (6.7%). Mixed feeding by mosquitoes is common, but with *An. arabiensis* this may be determined by the host available [21]. In this area, the presence of domestic animals is most likely to be very rare in human dwellings. Therefore, the human biting index recorded (78.7%) was relatively high, similar to that in Gedaref state [8].

Although there is seldom a large difference in susceptibility between the sexes, female mosquitoes (preferably blood-fed) should be used exclusively in field tests. This is because they survive better and show lower control mortality [15]. The susceptibility to DDT, malathion and fenit-

rothion was found to be 97.8%, 96.3% and 100.0% respectively, indicating that the mosquito population in this area had a low level of resistance to insecticides. Similar findings were reported in central Ethiopia [19]. The comeback susceptibility of *An. arabiensis* to DDT may be due to stoppage of chlorinated hydrocarbon insecticides for more than 20 years. Nevertheless, in the Gezira irrigated scheme the vector developed resistance to several insecticides in the 1970s, which was influenced by the overuse of crop-spraying with insecticides [10].

It is important to note that the establishment of the New Halfa agricultural scheme in the area has resulted in a serious abundance of *An. arabiensis* throughout the year. More studies are needed in order to assess the role of this species in malaria transmission in different seasons.

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