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# The host preferences of *Chrysops silacea* and *C. dimidiata* (Diptera: Tabanidae) in an endemic area of *Loa loa* in the Congo

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The analysis of 404 blood meals from *Chrysops silacea* and *C. dimidiata* which had been collected in the forests of the Chaillu mountains (People's Republic of the Congo) has demonstrated for the first time that both species also feed on non-human hosts. For both species the feeding patterns are fairly similar. However, man remains the main host in 89–90% of the cases. *C. silacea* and *C. dimidiata* took 6 and 4% respectively of their blood meals from hippopotamuses, 2 and 0% from rodents, 2 and 4% from wild ruminants, and 0·8 and 0·7% from monitor lizards. Whenever a differentiation has been made between the different members of the primates, only meals from humans but not from chimpanzees or baboons have been detected. The results may provide an explanation for the distinct barrier between the simian and human loiasis infections. They also give evidence for the ability of both vectors to travel over prolonged distances.

On the occasion of a conference on loiasis in 1950, Buxton had pointed out that, 'knowledge of the biology of African tabanids stands at present about where our knowledge of *Glossina* stood in 1905–1910' (Gordon *et al.*, 1950). In particular, there was a lack of knowledge regarding host preferences of the vectors. In those days no tests for identification of blood meals had been performed, and Gordon *et al.*, stressed that 'such tests are certainly important and will have to be done in the future'. But even though this strong statement had been made, no information regarding the origin of blood meals from African *Chrysops* has been published since then.

It is well known that there are two different types of *Loa*: the simian type with a nocturnal periodicity of microfilariae and the human type with a diurnal periodicity (Duke, 1972), but it has been demonstrated that the human type may develop in monkeys under experimental conditions (Duke and Wijers, 1958). This behaviour posed the question about an animal reservoir for this human filariasis (Fain, 1978).

This study presents results from blood meal analyses performed on 404 samples of *Chrysops dimidiata* and *C. silacea*, vectors of loiasis from an endemic area in the Congo.

## MATERIALS AND METHODS

### Study Area (see Fig.)

The investigation concentrated on five old villages and a pygmy camp in the forests of the Chaillu mountains (Lékoumou region). It covered a period from February 1986 to February

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1987. Three villages (Missama, Mapati and Loyo) are situated in the Sibiti district (Sibiti: 03°40′S; 13°20′E); the two others, Mouala and Moetche, as well as the Pygmy camp at Moutalango, are located in the Komono district (Komono: 03°15′S; 13°15′E). The altitude varies between 400 and 600 m. The population consists of Bantus (Bateke) and Pygmys, who live either on the outskirts of the Bantu villages or in their camps in the forest. The population density is two to 10 inhabitants $km^{-2}$.

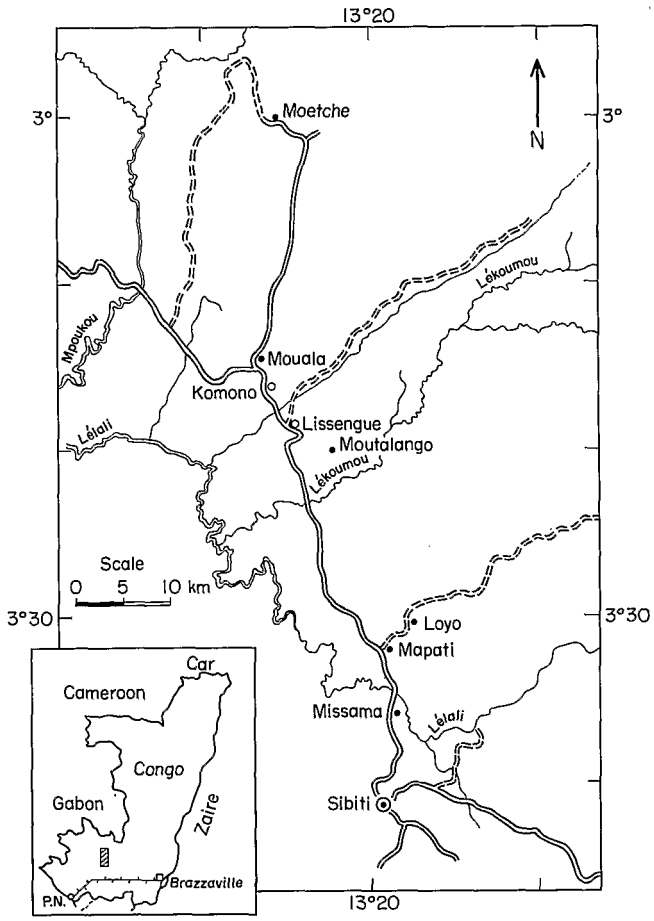


**Fig.** Area of investigation.

The vegetation is essentially of equatorial type (dense and humid forest) with some small scattered savanna areas, which are sometimes created by the inhabitants of nearby villages. The primary forest is in constant retreat because of the extensive agricultural practice of burning.

A variety of wild animals can be found: monkeys, especially gorillas and chimpanzees, are abundant in that area. Antelopes, buffalos and also some elephants and hippopotamuses are mentioned by hunters. The villagers frequently keep sheep and goats, but only rarely pigs.

The climate is of the southern Congo type, with an annual rainfall of 1400 to 1600 mm and constantly high air humidity, even during the dry season between May and August.

TABLE 1

*Host preferences of* Chrysops silacea *and* Chrysops dimidiata. *Prevalence of various sources of blood meals and standard deviation*

| | *Percentage of blood meals found in* | |
|---|---|---|
| *Host* | C. silacea | C. dimidiata |
| Man | 88·9±3·8 | 90·4±4·8 |
| Hippopotamus | 6·1±2·9 | 4·1±3·2 |
| Rodent | 1·9±1·7 | 0·0 |
| Wild pig | 0·0 | 0·7±1·3 |
| Wild ruminant | 2·3±1·8 | 4·1±3·2 |
| Monitor lizard | 0·8±1·1 | 0·7±1·3 |

TABLE 2

*Prevalence of sources of blood meals at different points of capture for* Chrysops silacea

| | *No. of flies which had fed on* | | | | |
|---|---|---|---|---|---|
| *Village* | *Man* | *Hippopotamus* | *Rodent* | *Wild ruminant* | *Monitor lizard* |
| Moutalango | 12 | 1 | 0 | 0 | 0 |
| Mouala | 31 | 1 | 0 | 1 | 0 |
| Missama | 148 | 5 | 5 | 4 | 2 |
| Mapati | 16 | 0 | 0 | 0 | 0 |
| Loyo | 6 | 0 | 0 | 0 | 0 |
| Moetche | 20 | 9 | 0 | 1 | 0 |

TABLE 3

*Prevalence of sources of blood meals at different points of capture for* Chrysops dimidiata

| | *No. of flies which had fed on* | | | | |
|---|---|---|---|---|---|
| *Village* | *Man* | *Hippopotamus* | *Wild pig* | *Wild ruminant* | *Monitor lizard* |
| Moutalango | 37 | 0 | 0 | 0 | 0 |
| Mouala | 11 | 0 | 0 | 1 | 0 |
| Missama | 53 | 0 | 0 | 2 | 1 |
| Mapati | 7 | 0 | 0 | 0 | 0 |
| Loyo | 5 | 0 | 0 | 0 | 0 |
| Moetche | 19 | 6 | 1 | 3 | 0 |

**Sampling Methods**

*Chrysops* were caught with hand-nets by field collectors over a period of one year (investigation into the infection rate and circadian rhythm). When engorged flies were dissected, the intestines containing blood were smeared onto filter paper on which the details of the capture were marked. Of 458 samples 404 (88%) could be identified; four of these were from two different hosts.

Normally the collectors avoided being bitten, so if gorged flies containing fresh blood were observed, they were not used.

**Blood Analysis**

The complement fixation test (CFT), production of antisera and control antigens, and the processing of samples have been described in detail by Staak *et al.* (1981). The enzyme-linked immunosorbent assay (ELISA) using absorbed antisera was adopted from Münstermann (1984).

The three step test system applied has been described previously (Staak *et al.*, 1986). Briefly, test material and controls were eluted with carbonate buffer (0·05 M, pH 9·6), and 50 µl were pipetted into the wells of microtitre F-plates. This was followed by incubation at 37°C for 60 minutes. Subsequently, 0·2% bovine serum albumin in carbonate buffer was added for neutralization of polystyrene material not covered by antigen. Rigorous washing with PBS plus 0·02% Tween 20 between the various reaction steps was strictly observed. The same buffer was used for the dilution of all following reagents. According to the classification (CFT) of samples into 'ruminants', 'suids', and 'others', parallel sets of individual samples were set up in numbers matching the number of antisera (raised in rabbits) available per reaction class: three times for 'suids', eight times for 'ruminants', 12 times for 'others'. Antisera against 'suids' and 'ruminants' were cross-absorbed with competing antigens for further specification of samples. Antisera against 'others', including man, were normally not cross-absorbed except for specific purposes. In this case, anti-man, anti-baboon (*Papio anubis*), and anti-chimpanzee (*Pan troglodytes*) were cross-absorbed and included in the test. Antisera at working dilution were added to the test plates, which were then incubated for 30 minutes at 37°C. Goat-anti-rabbit-IgG-PO (Biogenzia-Lemania, The Netherlands) at working dilution was used for second antibody. After another incubation period, substrate (5-amino-hydroxy-benzoic acid plus $H_2O_2$) was added to all microtitre wells, followed by a final incubation. Results were read either visually or by photometer (Multiskan, Titertek).

Forty-seven samples which had been identified as 'human' were additionally tested by using anti-chimpanzee and anti-baboon sera after cross-absorption with competing antigens.

## RESULTS

Results are presented in Tables 1–3. Regarding the host preferences, there was no significant difference between the two species of *Chrysops* ($\chi^2 = 0{\cdot}77$; 2 *df*), both of which showed an identical preference for humans, with about 90% of blood meals taken on man. The range of non-human hosts was of great variety and included hippopotamuses and wild ruminants (antelopes, principally *Tragelaphus scriptus*, and buffalos) as hosts of some importance. The difference between the two *Chrysops* species regarding the preference for hippopotamuses or wild ruminants was not significant ($\chi^2 = 1{\cdot}76$; 1 *df*), nor was that for rodents or reptiles ($P = 0{\cdot}375$, calculated according to the Fisher test).

None of the 47 primate samples tested with cross-absorbed antisera has been found to be of simian origin.

Tables 2 and 3 demonstrate that those *Chrysops* which took their blood meal from hippopotamuses were almost all caught in Moetche and Missama; only two *Chrysops*, one caught

in Mouala and one in Moutalango, were found to have taken their blood meal from hippopotamuses from other places.

## DISCUSSION

The existence of non-human hosts for *C. silacea* and *C. dimidiata* has sometimes been assumed (Gordon *et al.*, 1950) but has never been confirmed. One of the most interesting findings of this study is the identification of non-human hosts for these two species. The proportion of non-human blood meals is probably underestimated because the catching method used would tend to select flies particularly attracted to man.

Furthermore, it is of interest to note that domestic animals have not been found within the range of hosts for both species. This demonstrates that the flies prefer taking blood meals from man, with the occasional exception of rodents, even though they have a choice of dogs, cats and small ruminants (sheep and goats) in the villages. The identification of three human–rodent mixed samples is of special interest. Regarding the samples from rodents, they could probably originate from the Gambian rat (*Cricetomys*) or the Cane rat (*Thryonomys*), both of which are abundant around the villages. But even if the *Loa loa* would adapt itself to these rodents, they would constitute a reservoir of minor interest.

Blood meals taken from large animals were probably taken in areas with a complete absence of man. Unlike *C. langi* and *C. centurionis*, which hunt in the forest canopy and are probably the main vectors of simian filariasis (Duke, 1972), *C. silacea* and *C. dimidiata*, though living in the canopy, leave the tree tops to take their blood meals in the open areas where their hosts are present (Crewe and O'Rourke, 1951). This occurs in villages, which are always situated in man-made clearings, and also on great rivers, where herds of hippopotamuses might be present. Buffalos that live in the savanna areas surrounded by the forest may well have been the source of the blood meals from 'wild ruminants'.

None of the 47 specifically tested blood meals was derived from apes or monkeys, which might explain the absence of simian loiasis in man within the study area (Noireau, unpubl. paper). Concluding their observations on *C. silacea* and *C. dimidiata* in the Cameroons, Gordon *et al.* (1950) suggested that 'at Kumba the forest canopy is their normal habitat and that when man is not available monkeys are their chief source of blood'. Results achieved here are in contradiction with these suggestions, but they may explain the existence of a barrier between human and simian filariasis by the feeding behaviour of the vectors. Formerly, the separation of both infections has only been explained by the difference between the periodicity (diurnal and nocturnal) of both filariases and by differences in the circadian rhythm of the vectors (Duke and Wijers, 1958; Duke, 1972).

The villagers confirmed the presence of hippopotamuses in the rivers (Mpoukou, Lélali and Lékoumou, Fig.), but in all cases the herds of hippopotamuses were found at least 10 km away from the villages. These observations indicate that *Chrysops* are able to fly over larger distances than has been observed in marking-releasing-recapturing experiment (the maximum distance was estimated to be 3·2 km after six days; Beesley and Crewe, 1963). If the flies had taken their blood meals a maximum of two days before being caught, this would mean that they are able to fly over a distance of 5 km per day at tree top level in order to complete a non-sufficient blood meal.

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