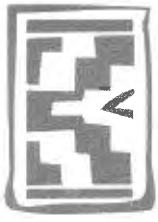

RESEARCH COMMUNICATION

Discovery of a *Culicoides imicola*-free zone in South Africa: preliminary notes and potential significance

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ABSTRACT

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In December 1993, a light-trap survey was made of the *Culicoides* found at eight horse stables and dairies in the sandy dune field west of Port Elizabeth, South Africa. While it was notable that *Culicoides* numbers were low (4 749) and that the diversity was poor (15 species), the most remarkable fact to emerge, was that *C. imicola*, the only proven vector of the virus of African horse sickness (AHS), was entirely absent. Though not abundant, *C. bolitinos*, a sister species of *C. imicola*, was overwhelmingly dominant (91,7%). Its larvae and pupae develop exclusively in the dung of cattle, but it is a species that is not implicated in the transmission of animal viruses. Elsewhere in South Africa, a frost-free climate, good rainfall and a plentiful supply of livestock would normally lead to the development of large foci of *C. imicola*. That this is not the case in the Port Elizabeth (P.E.) area is most likely owing to the winds inhibiting adult flight and the sandy soils being nutrient-poor and too well-drained to sustain *Culicoides* larvae. Studies are needed to confirm that sandy soils cannot sustain *C. imicola*. If so, the sandy coastal areas hold promise for quarantining against AHS.

Keywords: African horse sickness, AHS, *Culicoides imicola*-free zone, potential significance, sandy soils

INTRODUCTION

Culicoides biting midges (Diptera: Ceratopogonidae) are among the most plentiful insects that attack domestic livestock. For example, in the hotter, low-lying regions of the AHS-endemic Mpumalanga lowveld of South Africa, 500 000 *Culicoides* can be caught at a horse stable on a warm summer night (Meiswinkel 1995).

In the process of sucking blood, female *Culicoides*, 0,5–3,0 mm in size, may transmit diseases. In Africa, the most important of these are caused by the viruses of bluetongue (BT), African horse sickness (AHS), Akabane (AKA) and equine encephalosis (EE).

Although 112 species of *Culicoides* occur in South Africa (Meiswinkel 1995), the virus of AHS has regularly been isolated from field populations of *C. imicola* only (Nevill, Erasmus & Venter 1992). The single isolation from the uncommon *C. gulbenkiani* reported by Meiswinkel, Nevill & Venter (1994), appears to have been in error (B.J. Erasmus, personal communication 1995). While Du Toit (1944) did not identify the *Culicoides* used in his original studies on the transmission of AHS and BT to species, there seems little doubt that he was working with *C. imicola* (Venter, Meiswinkel, Nevill & Edwardes 1996a).

In South Africa, *C. imicola* is the most common and prevalent of all *Culicoides* species. In a countrywide study (Venter, Nevill & Van der Linde 1996b) it was found on 33 of 34 farms surveyed, and comprised 71% of >3 000 000 midges captured. This superabundance of *C. imicola* appears to be largely manmade, as the maintenance of cattle, horses, sheep and other livestock on irrigated pastures suits the needs of this midge (Meiswinkel 1995). Therefore, the occurrence of *C. imicola* in large numbers throughout

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much of the farming community in South Africa, means that unvaccinated horses are continually in danger of contracting AHS.

Since 1719, South Africa has experienced regular outbreaks of AHS. The country therefore stands classified as an enzootic zone, with the result that free trade in live animals, semen and embryos is disallowed, and participation in overseas equestrian events is complicated as horses have to undergo lengthy quarantine periods.

In the case of AHS, the European Community (EC) is developing the concept of regionalization to facilitate South Africa's reacceptance into the international arena. This entails the selection of low-risk zones, known to be relatively free of disease, for quarantine purposes. The Cape Peninsula, to some extent, fulfils the requirements, but must subscribe to a restrictive protocol which includes a ban on vaccination, strict monitoring of animals into and out of the zone, constant surveillance of insect populations, obligatory notification of mortalities, and mandatory post-mortems (Bosman 1994).

Until vaccination of horses against AHS is made compulsory for all horse owners in South Africa, and is accepted by the international community, this protocol will remain necessary. *Culicoides* surveys in the south-western Cape have shown that not only is *C. imicola* present at all sites sampled, but that at > 20% of these sites, it is also the dominant species (Nevill, Venter, Edwardes, Pajor, Meiswinkel & Van Gas 1988; Nevill, Venter, Le Roux & Barnard 1994).

New data, indicating that an *imicola*-free area does exist in South Africa, are presented here. This area is the sandy dune field found along the coast immediately west of P.E., and from which no cases of AHS have been reported this century. Possible reasons for the absence of *C. imicola* are advanced, and its significance discussed.

MATERIALS AND METHODS

Culicoides were collected at the height of summer, with a single UV light-trap per site and set on most nights from 21–31 December 1993. Eight plots or farms were sampled. The majority are situated in the Sardinia Bay area west of P.E. (Fig. 1). Four of the sites housed only horses, two only cattle, and two had both (Table 1). The weather conditions prevailing at each site on the day and night of trapping are summarized in Table 1. Each specimen of 4 749 *Culicoides* captured was identified and sexed (Table 1).

RESULTS AND DISCUSSION

In South Africa, the concurrence of a number of factors causes large populations of *Culicoides* midges to develop. These factors occur in the P.E. area, and are:

- A frost-free regime
- Good rainfall (average 613 mm/annum and falling year-round)
- Extensive irrigation of kikuyu pastures
- The presence of large groups of animals providing blood for biting *Culicoides*

Although these preconditions are to be found in the P.E. dune field, it can be seen from Table 1 not only that *C. imicola* is absent, but also that *Culicoides* numbers are low and that species diversity is poor. The most immediate reasons appear to be:

- Windy circumstances. Only 1 d in 5 is calm (Marker 1988). Windspeeds of more than 3m/s are known to suppress flight in *Culicoides* (Walker 1977).
- Sandy soils. Their poor nutrient status and excellent drainage qualities would impact negatively on both the feeding and survival of moisture-dependent *Culicoides* larvae.

The extent of the sandy area west of P.E., i.e. the potential *imicola*-free zone, is shown in Fig. 1. To the north and north-east of P.E. are found the Alexandria dune fields, the largest in South Africa.

Dominance of dung-inhabiting (coprophilic) *Culicoides*

As regards species composition, Table 1 shows that the great majority (92,5%) of the *Culicoides* caught in the P.E. area, belong to two species of the subgenus *Avaritia*, i.e. *C. bolitinos* Meiswinkel, 1989 (91,7%), and *C. gulbenkiani* Caeiro, 1959 (0,8%). Their immature stages are known to develop in the dung of cattle throughout South Africa (Nevill *et al.* 1988; Meiswinkel 1989). Neither species has been shown to transmit AHS. The close association between *C. bolitinos* and cattle, and the ability of the former to develop reasonable populations, implicates it as a potential vector of bluetongue. This, however, remains to be investigated.

The P.E. area is only one of three sites known in the country where the number of dung-inhabiting *Culicoides* dominates those of other species (Meiswinkel & Braack 1994; Venter & Meiswinkel 1994). Not only is dung largely independent of the soil type on which it is deposited, but it also carries its own suite of micro-organisms on which *Culicoides* larvae, of a few species of the subgenus *Avaritia* only, will feed.

It seems improbable that the currently recorded average of 600 *Culicoides* per light-trap collection would be exceeded if sampling were to be done at any other time of the year. These data show clearly that the dominant *Culicoides* in the dune fields are coprophilic; their population levels are therefore determined

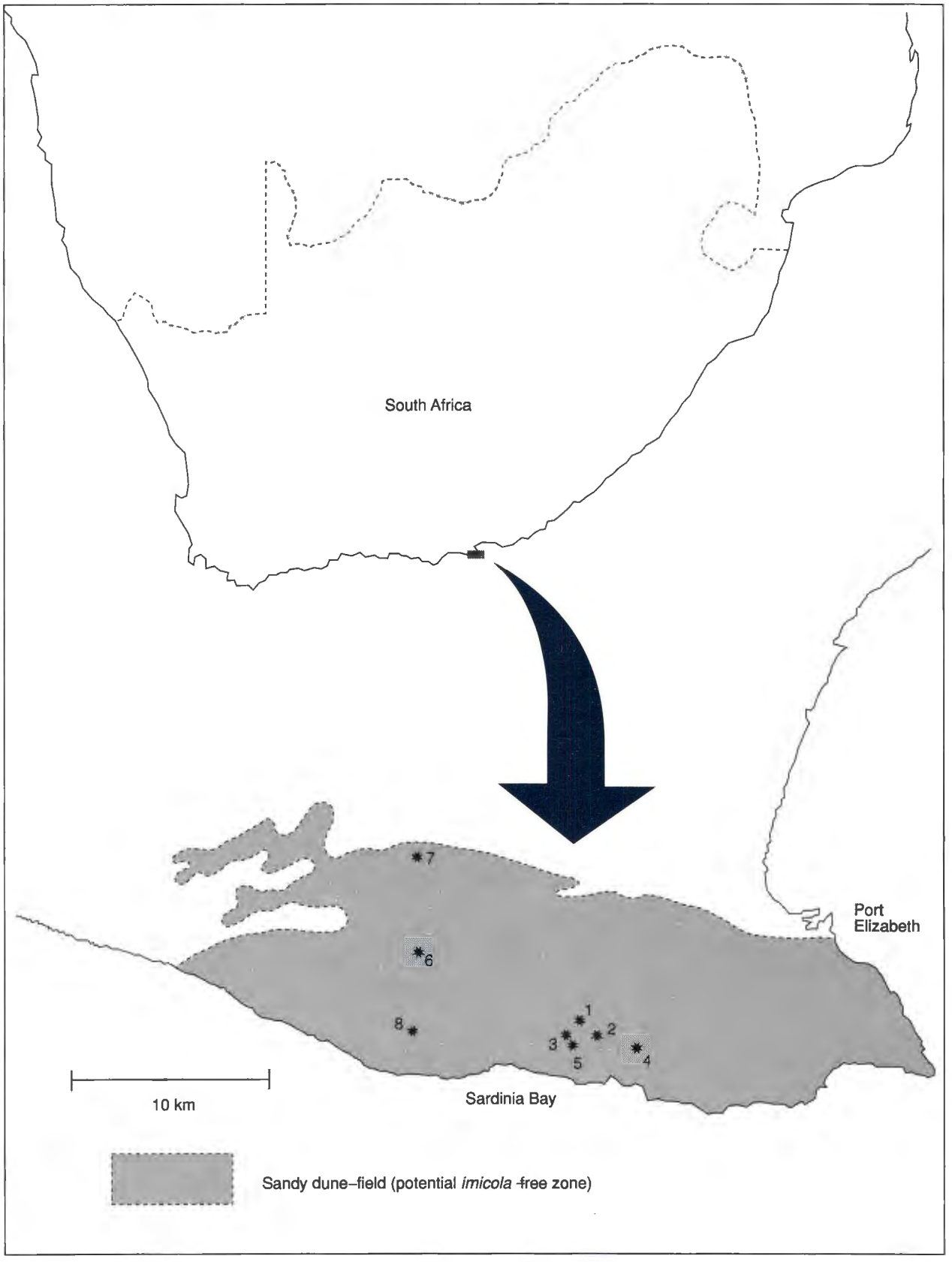

FIG. 1 Location of eight horse stables and dairy farms sampled for *Culicoides* midges on the sandy coastal strip south-west of Port Elizabeth, South Africa

TABLE 1 Numbers and species of *Culicoides* midges collected at eight horse stables and dairy farms in the sandy dune field west of Port Elizabeth (21.xii.1993–31.xii.1993)

Site No.	1	2	3	4	5	6	7	8	Males	Total (Males and females)	%
Site	Hustlers', Sardinia Bay	El Paso, Sardinia Bay	G. & P. Scallen, Farm Broadlands, Sardinia Bay	David Emslie, Farm Avonlea, Sardinia Bay	B. & N. Bartlett, Under Milkwoods, Sardinia Bay	Yellow Sea Dairy, P.E.	Fairview Racecourse, Greenbushes, P.E.	Bushy Park Dairy, Sardinia Bay			
	6 horses	2 cattle, horses	9 horses	24 horses	30 horses and cattle	160 Jersey herd, blacklight	360 horses	± 400 cattle			
	21.xii.1993	22.xii.1993	23.xii.1993	26 xii. 1993	28.xii.1993	29.xii.1993	30.xii.1993	31.xii.1993			
C. bolitinos	2 916	1	416	218	635	95	1	16	56	4 354	91,72
C. pycnostictus	2	1	2	4	86	16	3	16	16	146	3,08
C. nivosus	1		1	5	20	6		9	1	43	0,91
C. leucostictus					9				1	10	0,21
C. zuluensis						3			18	21	0,44
C. gulbenkiani	1					24			14	39	0,82
C. magnus	1			1		61	19	1	13	96	2,02
C. neavei	1					12	1		8	22	0,46
C. angolensis						1				1	0,02
C. onderstepoortensis				2	1	1			1	5	0,11
C. sp. # 69						1				1	0,02
C. bedfordi	2							3	2	7	0,15
C. similis				1						1	0,02
C. tropicalis	1									1	0,02
C. accraensis group		1		1						2	0,04
Total	2 925	3	419	232	751	220	24	45	130	4 749	
Weather conditions	Warm/calm	Very windy	Coolish/calm	Cool/misty/calm	Coolish/calm	Intermittently windy	Windy	Windy			

by the amount of dung available. This, in turn, is governed by cattle density. While it remains to be investigated, the precise role played by horse-dung in the maintenance of local *Culicoides*, appears to be negligible.

Paucity of "groundwater" *Culicoides*

Twelve of the remaining 13 species (7,4%) of *Culicoides* found in the P.E. area were "groundwater" breeders, i.e. their larvae and pupae, develop on the edges of water bodies. These include lakes, streams, drainage furrows and overflows from leaking cement dams. The low prevalence and low diversity of "groundwater" *Culicoides*, a group that includes *C. imicola*, suggests a lack of these larval habitats. This could be a result of the sandy substrate not remaining moist for sufficiently long periods (10–20 d). As noted above, rapidly draining, and therefore rapidly drying, soils would adversely affect moisture-dependent *Culicoides* larvae. Also, if the soil is poor in micro-organisms, conditions would be further exacerbated for the larvae of species such as *C. imicola* which depend upon prey organisms in organically enriched situations that are permanently moist, but not waterlogged. As moisture is essential for "groundwater" *Culicoides* larvae, it follows that aridity in the surface layer of sandy soil must depress overall species abundance and diversity.

The total number of "groundwater" *Culicoides* (355 specimens), and the number of species (12), is the lowest recorded at any group of sites yet sampled in South Africa over the past 20 years. Even the cold eastern Free State has more than double this number of species (Venter & Meiswinkel 1994). This reinforces the notion that it is not climate but organically poor, single-origin, sandysoils that are responsible for the short *Culicoides* species list in the P.E. area. While wind would also depress *Culicoides* numbers (as can be seen on certain nights as noted in Table 1), it would not affect overall species diversity.

CONCLUSION

Culicoides bolitinos was the most common species collected in the sandy dune field. Its role in the transmission of AHS is unknown, but it is not considered to be a vector, since it also dominates in the eastern Free State where *C. imicola* is rare and AHS virtually absent (Venter & Meiswinkel 1994). The dependence of *C. bolitinos* on the blood and dung of the African buffalo (*Syncerus caffer*), the blue wildebeest (*Connochaetes taurinus*) and cattle throughout Africa, indicates an evolutionary history linked to the Artiodactyla, and it is therefore unlikely that *C. bolitinos* is involved in the transmission of an orbivirus to equids (Perissodactyla). However, there is little doubt that in the P.E. area, *C. bolitinos* will disperse away from cattle to feed on horses. If there is concern regarding its potential to transmit AHS, it is worth noting that *C. bolitinos* could be eliminated from specific areas by the exclusion of cattle. There is strong evidence that the disappearance of dung-producing herbivores is accompanied by local extinction of coprophilic *Culicoides* species (Meiswinkel 1992).

In a survey that sampled *Culicoides* at 34 sites throughout South Africa, Venter *et al.* (1996b) showed that two of every three *Culicoides* midges captured, were *C. imicola*. Indeed, in only one of 959 collections analysed, was *C. imicola* found to be absent. This was at Rhodes, a small high-altitude enclave in the southern Drakensberg, but sampled for one night only. Therefore, the absence of *C. imicola* in the frost-free P.E. area is all the more unusual when it is taken into account that *C. imicola* is the most common *Culicoides* found in association with livestock in other frost-free areas of South Africa (Venter *et al.* 1996b).

As discussed above, the significant disparity in the numbers of dung-inhabiting and "groundwater" *Culicoides*, is the reverse of the norm in South Africa. This suggests that edaphic conditions do play as decisive a role as climate in determining the distribution of "groundwater" *Culicoides*, including *C. imicola*. The "sandy soil" factor has not previously been considered in studies on the epidemiology of AHS. These data suggest that soil type should be investigated as an important geographical barrier defining the limits of *C. imicola*, and thus perhaps, of AHS.

If the absence of *C. imicola*, and the suppression of *Culicoides* midges, in general, could be demonstrated in the sandy dune fields along the southern coastline of South Africa, it would mean that such areas could serve as natural quarantine stations. They would also make ideal equestrian venues, as valuable, unvaccinated horses, whether stabled at night or not, would not be exposed to *C. imicola*.

The greater P.E. area will be more intensively surveyed in the near future to establish whether the *imicola*-free zone is a reality and, if so, to determine its exact size. The survey will also investigate the absence of *C. imicola* throughout the year.

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