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OCCURRENCE OF *CULICOIDES* SPP. (DIPTERA: CERATOPOGONIDAE) IN TUNISIA, WITH EMPHASIS ON THE BLUETONGUE VECTOR *CULICOIDES IMICOLA*

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Summary:

Following the bluetongue (BT) outbreaks in Tunisia from 1999 to 2002, BTV (bluetongue virus) serotype 2 was isolated; however, no entomological investigation was performed. In the study presented here, we assessed the *Culicoides* species populations (particularly *C. imicola*) in proximity to the BT outbreaks locations, both as a retrospective analysis and to update the list of *Culicoides* species present in Tunisia. The insects were caught using light traps and the species identification was performed according to the standard entomological methods. This study revealed the presence of significant numbers of *C. imicola* in all the tested locations. In addition, we reported a new *Culicoides* species for the Tunisian fauna *C. punctatus*

KEY WORDS : bluetongue, vector, *Culicoides*, *Culicoides imicola*, Tunisia.

Résumé : PRÉSENCE DE DIFFÉRENTES ESPÈCES DE *CULICOIDES* (DIPTERA : CERATOPOGONIDAE) EN TUNISIE, EN PARTICULIER DE *CULICOIDES IMICOLA*, VECTEUR DE LA FIÈVRE CATARRHALE OVINE

*Suite à l'incursion de la fièvre catarrhale ovine (*bluetongue*) en Tunisie en 1999, le sérotype 2 du virus responsable a été isolé, toutefois, aucune investigation entomologique n'a été entreprise. Dans cette étude, nous avons évalué les populations de* Culicoides *(en particulier* C. imicola*) à proximité des foyers de la maladie, comme approche rétrospective et pour mettre à jour la liste des espèces de* Culicoides *présentes dans le pays. Les insectes ont été capturés en utilisant des pièges lumineux. Cette étude a permis de détecter un grand nombre de* C. imicola *dans tous les sites de piégeage. De plus, nous rapportons la présence d'une nouvelle espèce pour la faune tunisienne* C. punctatus.

MOTS CLÉS : *fièvre catarrhale ovine, vecteur,*Culicoides, Culicoides imicola, *Tunisie*

Bluetongue virus (BTV) causes an infectious, non-contagious, insect-borne disease of ruminants (Osburn, 1994). The clinical signs include fever, depression, excessive salivation, nasal discharge, facial oedema and ulceration of the oral mucosa (Parsonson, 1990). BTV is transmitted by haematophagous biting midges of the genus *Culicoides* (Diptera: Ceratopogonidae) with *C. imicola* Kieffer, 1913 considered the main vector of the disease in Africa, Middle East, southern Asia and southern Europe (Mellor & Boorman, 1995; Mellor & Wittmann, 2002). However, other species are potential vectors in other geographical areas: *C. obsoletus* Meigen, 1818 and *C. pulicaris* Linne, 1758 in European countries (Mellor & Pitzolis, 1979; Mellor & Wittmann, 2002; Caracappa *et al.*, 2003; Savini *et al.*, 2005; Carpenter *et al.*, 2006), *C. bolitinos* Meiswinkel, 1989 in South Africa (Gerdes, 2003) and *C. brevitarsis* Kieffer, 1917 in Australia (Standfast *et al.*, 1985).

In Tunisia, large BT outbreaks of serotype 2 were reported from 1999 to 2002 (Hammami, 2003). The first such outbreak occurred during autumn 1999 in the eastern part of the country along the coast. The overall morbidity and mortality rates were 8.35 % and 5.5 % respectively (Hammami, 2003). In 2000, 72 outbreaks were reported between June and October affecting 6,120 sheep in the eastern and central parts of the country. Subsequently, vaccination campaigns in sheep flocks were undertaken and allowed to control the disease. It is notable that data on *Culicoides* species in Tunisia is incomplete. In fact, fewer studies have been undertaken in comparison to other Mediterranean countries. A study by Chaker (1981) from mud samples after larval eclosion reported the presence of 22 *Culicoides* species, with the most abundant species were *C. circumscriptus*, *C. coluzzii*, *C. longipennis*, and *C. puncticollis*. However, no *C. imicola* was detected. The presence of *C. imicola* in Tunisia was reported only in 2005 and the study was limited to the Monastir governorate (Chaker *et al.*, 2005). The aim of this study was to determine whether *C. imicola* was present in other regions of Tunisia.

MATERIAL AND METHODS

During this study, insect trapping were performed in locations where previous BT outbreaks occurred, focussing on the governorates of

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Ariana and Sousse. The trappings were undertaken in Borj El Amri, Sidi Thabet, El Alam and Sousse districts in October 2004 and in Belvedere and Sidi Thabet districts in November 2005. The weather conditions were similar for all the trapping nights. Insect trappings were performed using light traps (Onderstepoort, South Africa) with an UV lamp (8 W) and a fan supplied by a battery (12 V). Light traps were set outdoors by the enclosure where the livestock was confined, as *C. imicola* are abundant around livestock holdings (Mellor & Wittmann, 2002). Two traps were placed in each farm and the numbers of specimen were cumulated. The traps were switched on before sunset and off after sunrise. The collection container was removed in the morning and taken to the laboratory and the insects were conserved in 70 % ethanol at room temperature. All specimens of the genus *Culicoides* were subsequently separated and identified according to their wing pattern. The identification was further confirmed by the use of the dichotomic keys (Chaker, 1981; Delécolle, 1985). For the identification of some *Culicoides* species, it was necessary to dissect the midges and make microscopic slide preparations of their body parts (Wirth & Marston, 1968).

RESULTS

This study allowed the trapping of 1,724 *Culicoides* specimen (1,546 in 2004 and 178 in 2005) (Table I). The captured specimens were classified into fourteen species. The most abundant species was *C. imicola* Kieffer, 1913 (76 %), which was detected in all the locations. *C. newsteadi* corresponded to 9.2 % and was detected in 4/5 locations. *C. paolae* (1.6 %) and *C. circumscriptus* (5.5 %) were detected in 4/5 locations. The other species were present at lower densities: *C. kingi* (0.8 %), *C. sahariensis* (0.3 %), *C. puncticollis* (0.3 %), *C. saevus* (2.3 %), *C. jumineri* (1.6 %), *C. odiatus* (1.3 %), *C. pseudopallidus* (0.3 %), *C. kurensis* (0.3 %) and *C. cataneii* (0.7 %). This study allowed the detection of a new *Culicoides* species to the Tunisian fauna, *C. punctatus* Meigen, 1804 trapped in 2005 in Sidi Thabet and corresponded to one specimen.

The highest insect density was reported in Borj El Amri (57 %) followed by El Alem (21 %). Only Sidi Thabet location was sampled in 2004 and 2005 (Table I) and was characterized by a similar trap density in both trap catches.

DISCUSSION

During this study, 14 *Culicoides* species were trapped in Tunisia, which is less than the previously reported number of 22 by Chaker and co workers (Chaker *et al.*, 2005). This might be due to the trap type and the period of study. Long term trapping is recommended for a better assessment of the number of *Culicoides* species present in the

	Sites of trapping in October 2004				**Sites of trapping in November2005**	
Species	Borj EL Amri	Sidi Thabet	El Alem	Sousse	Sidi Thabet	Belvédère
C. imicola	807	106	297	63	35	2
C. newteadi	62	19	2	16	59	
C. paolae	15		5	4	2	
C. circumscriptus	28	2	4		59	1
C. kingi	12		1			
C. sahariensis	5					
C. puncticollis	1		2		2	
C. saevus	9		31			
C. jumineri	14	11	1		5	
C. odiatus	15	2	4			
C. pseudopallidus			2		3	
C. kurensis	3				1	
C. cataneii	3				7	1
C. punctatus					1	
Total of trapping	974	140	349	83	174	4
No. of *Culicoides* spp. captured	12	5	10	3	10	4
No. of *Culicoides* specimens captured		1,546			178	
No. of *C. imicola* specimens captured		1,273			37	

Table I. – Number of *Culicoides* specimens trapped for each species and in each location.

country. The number of specimen differed between locations and might be an indicator of the establishment and the dynamics of the settled *Culicoides* species. However, only a very low number of insects were trapped in Belvedere near the zoological park of Tunis, which may be explained by the intensive insecticide treatment in the city.

C. imicola was trapped in high numbers in all locations and further north then previously reported. *C. imicola* populations are likely to be settled in Tunisia and seem to have played a major role in BT outbreaks. In fact, we were unable to detect other potential BT vectors such as *C. obsoletus* Meigen, 1818 (a BTV vector in the Mediterranean area), which was reported previously reported in Algeria and Morocco (Remm, 1988; Baylis *et al.*, 1997). However, we were able to detect *C. pulicaris* and *C. kingi*, the former is a potential BTV vector (*Baylis et al.*, 1997), whilst the later is a vector of a related orbivirus, the Epizootic Haemorrhagic Disease Virus (EHDV) (Mellor *et al.*, 1984). Consequently, a further investigation of the population dynamics and the vectorial capacity of these *Culicoides* species may be warranted to assess their role in the local BTV transmission.

This study allowed the identification of a new *Culicoides* species for the Tunisian Fauna: *C. punctatus*. Despite the fact that this species is not a potential vector, it is likely to be newly introduced in the country probably due to climatic conditions. Therefore, regular entomological surveys are important to detect the various BTV vectors and to understand the mechanisms of orbiviruses perpetuation in an endemic area.

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