

## DESCRIPTION OF BREEDING SITES OF *CULICOIDES* SPECIES (DIPTERA: CERATOPOGONIDAE) IN TURKEY

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### Summary:

The aim of this study was to describe the breeding sites of *Culicoides* species in Konya province, Turkey. The samples taken from 11 different habitats in Konya province were examined for *Culicoides* species. The collected adult *Culicoides* specimens were reared in plastic buckets in our laboratory. Among 2,798 specimens reared, 18 species were identified. *Culicoides circumscriptus* Kieffer, 1918 was the most abundant species reared in the samples taken from mud rich in organic matters near the water reservoirs; *C. circumscriptus*, *C. nubeculosus* (Meigen), 1830 and *C. shaklawensis* Khalaf, 1957 in moist soil with organic matter; *C. gejgelensis* Dzhafarov, 1964 in moist soils, along watering channels and dripping waters; *C. festivipennis* Kieffer, 1914 and *C. circumscriptus* along sewage channel; *C. festivipennis* in reed sites and along garden watering channels; *C. circumscriptus* in rain pools; *C. odiatus* Austen, 1921 and *C. circumscriptus* in mud near the dams; *C. gejgelensis* and *C. kibunensis* Tokunaga, 1937 from sites along the stream. No *Culicoides* was reared in the samples obtained from livestock dung and tree holes. It was observed that *C. circumscriptus*, *C. festivipennis* and *C. shaklawensis* preferred mud rich in organic matters near the water reservoirs. *Culicoides imicola* Kieffer, 1913, which is the main vector of bluetongue, was not detected in Konya province. The others vectors of bluetongue, *C. obsoletus* gr. (Meigen), 1818, *C. schultzei* gr. (Enderlein), 1908 and *C. pulicaris* (Linnaeus), 1758 were only obtained in little numbers.

**KEY WORDS :** *Culicoides*, breeding sites, Konya province, Turkey.

*Culicoides* species are widely seen throughout the world (Blackwell, 2001). Some *Culicoides* species such as *C. imicola* Kieffer, 1913 and *C. obsoletus* gr. (Meigen), 1818 are known as vectors of a number of serious viral diseases such as bluetongue and african horse sickness (Braverman, 1994; Blackwell, 2001). The species belonging to the genus *Culicoides* may rear in various breeding sites (Mullen & Hribar, 1988). Some *Culicoides* species can rear in soil, mud rich in organic matter, swamps, marshes, mud around dams, water reservoirs, along the drainage channels and stream, tree holes, cow dung (Braverman *et al.*, 1974;

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### Résumé :

DESCRIPTION DES SITES DE DÉVELOPPEMENT LARVAIRE DES DIFFÉRENTES ESPÈCES DE *CULICOIDES* (DIPTERA : CERATOPOGONIDAE) EN TURQUIE

Le but de cette étude était de décrire les sites de développement larvaire des espèces de *Culicoides* dans la province de Konya, en Turquie. Les *Culicoides* adultes, prélevés au niveau de 11 habitats, ont été élevés dans des seaux en plastique dans notre laboratoire. Parmi les 2798 spécimens élevés, 18 espèces ont été identifiées. *Culicoides circumscriptus* Kieffer, 1918 est l'espèce la plus abondantes dans les échantillons de boue riche en matière organique à proximité des réservoirs d'eau; *C. circumscriptus*, *C. nubeculosus* (Meigen), 1830 et *C. shaklawensis* Khalaf, 1957 dans les sols humides riches en matière organique; *C. gejgelensis* Dzhafarov, 1964 dans les sols humides le long des canaux d'irrigation et des eaux d'égouttement; *C. festivipennis* Kieffer, 1914 et *C. circumscriptus* le long des égouts; *C. festivipennis* dans les zones de roseaux et le long des canaux d'irrigation de jardin; *C. circumscriptus* dans les réservoirs d'eau de pluie; *C. odiatus* Austen, 1921 et *C. circumscriptus* dans la boue près des barrages; *C. gejgelensis* et *C. kibunensis* Tokunaga, 1937 le long des rivières. Aucun *Culicoides* en provenance du fumier du bétail ou de trous d'arbre n'a été élevé. En définitive, *C. circumscriptus*, *C. festivipennis* et *C. shaklawensis* préfèrent les boues riches en matières organiques à proximité des réservoirs d'eau; *C. imicola* Kieffer, 1913, qui est le vecteur principal de la maladie de la langue bleue, n'a pas été détecté dans la province de Konya; *C. obsoletus* gr. (Meigen), 1818, *C. schultzei* gr. (Enderlein), 1908 et *C. pulicaris* (Linnaeus), 1758, qui sont également des vecteurs de cette maladie, n'ont été obtenus qu'en petite quantité.

**MOTS CLÉS :** *Culicoides*, site de reproduction, province de Konya, Turquie.

Lubega & Khamala, 1976; Bram, 1978; Braverman, 1978; Mullen & Hribar, 1988; Nevill *et al.*, 1988). Such great diversity of habitats of different species of biting midges makes it difficult to find out the immature stages. Immature stages of *Culicoides* species usually lives in 0-5 cm depth of the soil (Lubega & Khamala, 1976; Blackwell & King, 1997; Uslu & Dik, 2006). It was stated that *C. imicola* were reared in cow dung and rich in organic material without surface water (Braverman, 1973; Braverman *et al.*, 1974; Braverman & Galun, 1973), *C. schultzei* gr. (Enderlein), 1908 in between rows of tomatoes (Muradov, 1965), *C. obsoletus* gr. in stagnant water reservoirs and marsh-ridden parts with bushy vegetation (Dzhafarov, 1976), and *C. pulicaris* (Linnaeus), 1758 from small water logged areas in near lakes, bogs and small marsh-ridden places (Konurbayev, 1965; Dzhafarov, 1976).

Hribar (1990) stated that gar-extraction and salt-flotation was the most effective method for collecting larvae samples of biting midges.

In Turkey, the first systematic research on adult *Culicoides* species was carried out by Navai (1977). Afterwards, systematic and ecological studies on *Culicoides* species have been studied in different parts of Turkey (Jennings *et al.*, 1983; Dik & Dinçer, 1992; Dik, 1993; 1996; Yılmaz, 1994; Eren *et al.*, 1995; Yagci *et al.* 1999; Tilki & Dik, 2004; Dik *et al.*, 2006; Uslu & Dik, 2005; Uslu & Dik, 2006). No study has been found describing breeding sites of *Culicoides* species in Turkey. The purpose of this study was to describe the breeding sites of *Culicoides* species in Konya province, Turkey.

## MATERIALS AND METHODS

Konya province has a climate that is hot and dry in the summer, cold and rainy in the winter season, and is located at least 1,000 m above sea level and between latitudes 37.59 N and 32.34 E. Four districts of Konya (Centrum, Çumra, Kadınhanı, Sarayönü) were regularly visited every 15 days from April 2001 to April 2002, and the samples from different breeding sites were taken.

All samples were taken from ten different places, 10 cm in diameter and 5 cm deep for each breeding sites by using steel-made sampling tool, except of the materials obtained from tree holes using a metal spoon or scoop. The samples were put in nylon bags, brought to the lab and placed in the plastic buckets. The samples were kept for 30-35 days in the lab to allow rearing of adult *Culicoides*. In order to avoid drying out, the samples were humidified with sprayed distilled water every other day. The buckets were lighted up with 300 Watt bulb to capture the flying *Culicoides* specimens. The insects were caught by vacuum and transferred into a tube containing 70 % of ethyl alcohol. Big sized insects were removed by a brush and little sized insects were examined for identification of *Culicoides* under the light microscope. The *Culicoides* species were cleared in alcohol-phenol for a few days, and identified by using a Nikon SMZ-10 stereo microscope. In addition, the samples were analyzed to determine the amount of organic substances.

Chi-square test was used for the statistical analyses.

## RESULTS

*Culicoides* specimens of all samples were taken from 13 different breeding sites in Konya province, except from dung and tree holes. In this

study, 2,798 *Culicoides* specimens were reared in the lab; of these, 1,559 were females and 1,239 males.

The 18 identified *Culicoides* species were namely *Culicoides circumscriptus* Kieffer, 1918, *C. festivipennis* Kieffer, 1914, *C. shaklawensis*, *C. gejjelensis* Dzhafarov, 1964, *C. puncticollis* (Becker), 1903, *C. nubeculosus* (Meigen), 1830, *C. riethi* Kieffer, 1914, *C. longipennis* Khalaf, 1957, *C. odiatus* Austen, 1921, *C. flavipulicaris* Dzhafarov, 1964, *C. cataneii* Clastries, 1957, *C. maritimus* Kieffer, 1924, *C. parroti* Kieffer, 1922, *C. kibunensis* Tokunaga, 1937, *C. schultzei* gr., *C. subfasciipennis* Kieffer, 1919, *C. salinarius* Kieffer, 1914 and *C. pulicaris*.

The range of *Culicoides* species is shown in Table I. Out of the specimens collected, 2,283 breed in the samples taken from organic matter rich mud near the water reservoirs, 126 in moist soils with organic matter, 78 in rain pools, 71 in watering channels, 67 in sewage channel, 66 in dripping water, 49 in reed sites, 28 in garden watering channels, 23 in moist soils, five along the stream and two in mud near the dams.

*C. circumscriptus* and *C. festivipennis* were seen as dominant species especially in organic matter rich mud near the water reservoirs. *C. circumscriptus* was found in a wide variety of habitats. *C. maritimus* was detected only in organic matter rich mud near the water reservoirs. It was observed that *C. gejjelensis* was found in soils dripping water, watering channels and moist soils, *C. festivipennis* and *C. circumscriptus* in sewage channels, garden watering channels and rain pools, *C. festivipennis* in reed sites and in sewage channels, *C. odiatus* only in mud near the dams as dominant species.

The results of statistical analysis were shown in Table II. *Culicoides* specimens were collected in May (0.25 %), June (3.75 %), July (34.13 %), August (33.95 %), September (23.44 %) and October (4.50 %).

## DISCUSSION

18 *Culicoides* species found in the current study were mainly *C. circumscriptus*, *C. festivipennis*, *C. shaklawensis*, *C. puncticollis*, *C. nubeculosus* and *C. riethi*, and were reared in the samples taken from organic matter rich mud near water reservoirs. *Nubeculosus* group (*C. nubeculosus*, *C. puncticollis*, *C. riethi*) are able to reproduce especially in habitats with rich organic contents and organic content complications. The remaining *Culicoides* species possibly breed in completely different sites with completely different needs. *Culicoides circumscriptus*, *C. gejjelensis*, *C. shaklawensis* and *C. festivipennis* were cosmopolite species and grew in different breeding sites. While *Culicoides circumscriptus* and *C. gejjelensis* were seen in 10 out of

Breeding sites												
Species	Mud rich in organic matters near the water reservoirs	Moist soil with organic matter	Breeding sites					Mud near the dams	Sites along the stream	Totally (%)		
			Moist soils	Dripping water	Sewage channels	Reed sites	Garden watering channels				Watering channels	Rain pools
<i>C. circumscriptus</i>	1,201	62	3	7	20	2	6	1	47	1	—	48.25
<i>C. festiviipennis</i>	506	9	—	—	40	41	16	—	18	—	—	22.52
<i>C. sbaklavensis</i>	233	17	—	—	—	—	1	2	6	—	—	9.26
<i>C. puncticollis</i>	167	5	—	—	—	—	—	—	—	—	—	6.15
<i>C. nubeculosus</i>	56	22	—	—	1	—	1	—	—	—	—	2.86
<i>C. rietli</i>	47	4	—	—	—	—	—	—	—	—	—	1.82
<i>C. geigelenis</i>	33	4	19	56	2	3	3	56	5	—	3	6.58
<i>C. longipennis</i>	18	2	—	—	2	2	—	—	—	—	—	0.86
<i>C. flavipulcaris</i>	8	—	—	—	—	—	—	1	—	—	—	0.32
<i>C. maritimus</i>	3	—	—	2	—	—	—	1	—	—	—	0.21
<i>C. parroti</i>	3	—	—	—	—	—	—	—	—	—	—	0.11
<i>C. schulzei</i>	3	—	—	—	—	—	—	—	—	—	—	0.11
<i>C. subfascipennis</i>	2	—	—	—	—	—	—	—	—	—	—	0.07
<i>C. cataneti</i>	1	—	1	1	2	—	—	3	—	—	—	0.29
<i>C. odiatus</i>	1	—	—	—	—	1	—	7	2	—	—	0.43
<i>C. kabunensis</i>	1	—	—	—	—	—	—	—	—	—	2	0.11
<i>C. salinarius</i>	—	—	—	—	—	—	—	—	—	—	—	0.04
<i>C. pulcaris</i>	—	1	—	—	—	—	—	—	—	—	—	0.04
N	2,283	126	23	66	67	49	28	71	78	2	5	100

Table I. – Larval habitats for *Culicoides* species in Konya.

Species	Mud rich in organic matters near the water reservoirs	Moist soil with organic matter	Breeding sites					Mud near the dams	Sites along the stream	Totally (%)		
			Moist soils	Dripping water	Garden watering channels	Reed sites	Watering channels				Rain pools	
<i>C. circumscriptus</i>	1,201	62	3	7	20	2	6	1	47	1	—	48.25
<i>C. festiviipennis</i>	506	9	—	—	40	41	16	—	18	—	—	22.52
<i>C. sbaklavensis</i>	233	17	—	—	—	—	1	2	6	—	—	9.26
<i>C. puncticollis</i>	167	5	—	—	—	—	—	—	—	—	—	6.15
<i>C. nubeculosus</i>	56	22	—	—	1	—	1	—	—	—	—	2.86
<i>C. rietli</i>	47	4	—	—	—	—	—	—	—	—	—	1.82
<i>C. geigelenis</i>	33	4	19	56	2	3	3	56	5	—	3	6.58
<i>C. longipennis</i>	18	2	—	—	2	2	—	—	—	—	—	0.86
<i>C. flavipulcaris</i>	8	—	—	—	—	—	—	—	—	—	—	0.32
<i>C. maritimus</i>	3	—	—	2	—	—	—	—	—	—	—	0.21
<i>C. parroti</i>	3	—	—	—	—	—	—	—	—	—	—	0.11
<i>C. schulzei</i>	3	—	—	—	—	—	—	—	—	—	—	0.11
<i>C. subfascipennis</i>	2	—	—	—	—	—	—	—	—	—	—	0.07
<i>C. cataneti</i>	1	—	1	1	2	—	—	3	—	—	—	0.29
<i>C. odiatus</i>	1	—	—	—	—	1	—	7	2	—	—	0.43
<i>C. kabunensis</i>	1	—	—	—	—	—	—	—	—	—	2	0.11
<i>C. salinarius</i>	—	—	—	—	—	—	—	—	—	—	—	0.04
<i>C. pulcaris</i>	—	1	—	—	—	—	—	—	—	—	—	0.04
N	2,283 <sup>a</sup>	126 <sup>b</sup>	23	66 <sup>c</sup>	28 <sup>d</sup>	49 <sup>c</sup>	78 <sup>c</sup>	2 <sup>c</sup>	78 <sup>c</sup>	5 <sup>c</sup>	66 <sup>c</sup>	2,798

a, b, c, d, e: Breeding sites having in row different character are statistically important (P &lt; 0.05).

Table II. – Numbers of *Culicoides* spp. found in various breeding sites and statistically analyses.

13 different habitats (Table I), *Culicoides parroti*, *C. schultzei* gr., *C. subfasciipennis* and *C. maritimus* breed in the samples taken from organic matter rich mud near the water reservoirs. *C. pulicaris* and *C. salinarius* breed in only mud moist with organic matter and in garden watering channels, respectively. These species were not observed in other breeding sites.

*Culicoides* species were not reared in the samples obtained from the tree holes in this study. The reason for this might be that the year 2001, when the study was done, was dry in Konya with the resultant effect of dry tree holes. No *Culicoides* specimen was found in the samples taken from dung in this study either. The reason for this may be that the heap dung where the samples were taken was sprayed with insecticides according to farmers to prevent reproduction of flies. Many *Culicoides* species breed in organic matter rich mud near the water reservoirs. This result showed that the immature stages of *Culicoides* species need organic substances for growth. The reason why there were several species in this habitat was that breeding sites where the samples were taken had high organic matter, and that these habitats were water-saturated during summer.

Most of the *Culicoides* species were found in organic matter rich mud near the water reservoirs with statistically significant difference compared to the rest of the habitats. While no statistically significant difference was found among sewage channels, reed sites, watering channels, rain pools and dripping water, it was statistically significant among the rest of the habitats. There was statistically significant difference for the moist soil with organic matter compared to other habitats. Although no significant difference was found between the garden watering channels and in moist soils without organic matter, these two habitats were significant different from the rest of the habitats. A small number of *Culicoides* species was found in mud near the dam sites and along the stream with no statistically significant difference between the two, which was significantly different from the rest of the habitats (Table II).

*Culicoides imicola*, the main vector of bluetongue in Europe and Middle-East, was not reared in the samples taken from organic matter rich mud near the water reservoirs in spite of many *Culicoides* specimens (2,283 individuals) collected from the samples taken from the same habitat in this study. Other vector of bluetongue, *C. obsoletus* gr. was not reared in the samples taken from wet soil rich in organic matter and other breeding sites in this study. *C. imicola* was not found in any samples taken from all habitats because it is a tropical and sub-tropical species, and climatic conditions are not suitable for its breeding in Konya. In a study done to detect distribution of *Culicoides* species in Konya province, *C. imicola* was not detected,

however *C. obsoletus* gr. was found relatively rare (Dik & Diğer, 1992). *C. imicola* has not ever been seen in this region up to date. These findings supported that *C. imicola* could be reared in tropical and subtropical regions. Dik (1993, 1996) observed small numbers of *C. schultzei* gr., *C. pulicaris*, *C. obsoletus* gr. and *C. imicola* in Mediterranean and Aegean regions of Turkey and reported that these species are also rare in Turkey. On the other hand, *C. schultzei* gr. was caught in high numbers in Mediterranean region of Turkey (Jennings *et al.*, 1983). Tilki & Dik (2003) reported that *C. imicola* and *C. schultzei* gr. were relatively common species in Iskenderun located in eastern coast of Mediterranean Sea. It was concluded that this might be due to climatic conditions and geographical characteristics of the region.

As a vector of bluetongue, *C. pulicaris* breed in only mud moist with organic matter in this study. In addition to this, probable vectors of bluetongue as *C. schultzei* gr. have also a range of breeding sites. Three specimens of *C. schultzei* gr. were found in the samples taken from organic matter rich mud near the water reservoirs. A number of species may rear in different habitats as in different geographical regions with various climate and vegetation. On the other hand, finding breeding sites of vector species where they grow will make it easier to fight against these species.

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