Response of Tabanidae (Diptera) to different natural attractants

Stjepan Krčmar, Alma Mikuska, and Enrih Merdić

Department of Biology, JJ Strossmayer University, Lj. Gaja 6, HR-31000 Osijek, Croatia

Received 11 April 2006; Accepted 30 May 2006

ABSTRACT: The response of female tabanids to natural attractants was studied in the Monjoroš Forest along the Nature Park Kopački rit in eastern Croatia. Tabanids were caught in canopy traps baited with either aged cow, horse, sheep, or pig urine and also in unbaited traps. Tabanids were collected in a significantly higher numbers in traps baited with natural attractants compared to unbaited traps. The number of females of *Tabanus bromius*, *Tabanus maculicornis*, *Tabanus tergestinus*, and *Hybomitra bimaculata* collected from canopy traps baited with cow urine and traps baited with other natural attractants differed significantly. Females of *Haematopota pluvialis* were also collected more frequently in canopy traps baited with aged cow urine than in those with aged horse urine, but this difference was not significant. However, the number of females of *Haematopota pluvialis* collected from canopy traps baited with other natural attractants (sheep and pig urine) differed significantly when compared with aged cow urine baited traps. Canopy traps baited with aged cow urine collected significantly more *Tabanus sudeticus* than did traps baited with aged pig urine. Finally, the aged cow urine baited canopy traps collected 51 times more tabanids than unbaited traps, while aged horse, aged sheep, and aged pig urine baited traps collected 36, 30, and 22 times as many tabanids, respectively, than unbaited traps. ***Journal of Vector Ecology* 31 (2): 262-265. 2006.**

Keyword Index: Tabanidae, canopy traps, natural attractants, Croatia.

INTRODUCTION

Synthetic attractants in traps are used in many parts of the world to collect female tabanids. Studies of attractants help to understand the host-finding process, and synthetic attractants may be valuable tools in monitoring and suppressing pest species (Nilssen 1998). Efficacy of tabanid traps is mainly increased by the addition of chemicals that mimic natural host odors (Hall and Wall 2004). For example, carbon dioxide is a well-established attractant for a large variety of haematophagous Diptera (Doskočil and Chvála 1974, Roberts 1971, 1975, Vale and Hall 1985, Kline et al. 1991, Leprince et al. 1994, Mohamed-Ahmed and Mihok 1999). Furthermore, octenol is an important attractant for blood-feeding Diptera, especially tabanids, because it increases trap catches several times (French and Kline 1989, Hayes et al. 1993, Foil and Hribar 1995, Gibson and Torr 1999). Also, ammonia has a positive effect on catches of tabanids (Hribar et al. 1992, Kristensen and Sommer 2000). Besides these synthetic attractants, some natural attractants such as the aged urine of cows or African buffalo, horses, and rhinoceros are known mainly for tsetse species (Glossinidae) and Tabanidae (Okech and Hassanali 1990, Madubunyi et al. 1996, Mihok et al. 1996, Krčmar et al. 2005). So far, however, little is known about the presence of key attractants in the aged urine of other insect species, so it is still not clear if urine from pigs, sheep, or other mammals is useful for collecting tabanids. Therefore, this study considers the effects of natural attractants such as aged cow, horse, pig, and sheep urine on catching tabanids in eastern Croatia.

MATERIALS AND METHODS

This study was carried out in the Monjoroš Forest (UTM CR 37), approximately 2 km NE from the Nature Park Kopački rit in eastern Croatia. The Monjoroš Forest is a floodable region located along the east bank of the Danube River. The largest part of the Monjoroš Forest lies between 78 and 82.5 m above sea level. In spite of its prevalent lowlands, the structure of the Monjoroš Forest is very complex. The forest is mostly composed of white willow, black poplar, and common oak. Higher terrains are overgrown with white willow forests and black poplar. Tabanids were collected by five black and white linen canopy traps constructed according to the design of Hribar et al. (1991). The traps were baited with either aged cow, horse, sheep, or pig urine. The urine from cows, horses, and pigs was collected mostly before the evening feeding at the pound, but sheep urine was collected in the morning hours in the paddock. The urine from these animals was left for two weeks at the ambient temperature (20-25° C) in 2.5 liter plastic bottles before it was used in experiments. The cows, horses, and pigs were stall-fed with leguminous plants and hybrid corn, while sheep were only feed on the pasture with different leguminous plants. A total of 25 samplings was made from the end of May to the end of August 2005 (on May 1, June 10, July 8, and August 6). The daily trapping period was between 7 a.m. and 7 p.m. Canopy traps were placed 700 m apart, about 8 m from the forest edge. Attractants were rotated among traps every day. There was always one canopy trap without an attractant that served as a control. Fresh attractants were

added at the beginning of each trapping period. Each time, traps were filled separately with 40 ml of aged urine of the above-mentioned domestic animals. Attractants were dispensed from glass vials with Styrofoam corks and a 10 cm cotton wick protruded from the center of the cork to the outside. The cotton wick was replaced when the attractants were replenished. All attractants dispensers were placed 30 cm below the top of the canopy traps. All trapped flies were preserved in ethanol. Identification and nomenclature followed Chvála et al. (1972) and Chvála (1988). A Chi-square test was used to test the null hypothesis that tabanids would be caught in the same proportion with all four natural attractants.

RESULTS

A total of 2,940 specimens was collected belonging to 19 species of tabanids grouped into the genera: *Chrysops, Atylotus, Hybomitra, Tabanus,* and *Haematopota* (Table 1). *Tabanus bromius* comprised 38.5% of the tabanids collected. *Haematopota pluvialis* followed with 34.0%. These two species represented 72.5% of the tabanids collected, while the remaining 17 species made up the remainder (Table 1). The χ^2 analyses of the trapping data showed that each of the natural attractants significantly increased the number of tabanids collected, in comparison to the number of tabanids collected in unbaited canopy traps ($\chi^2 = 2519.03$, $P < 0.05$; $\chi^2 = 703.24$, $P < 0.05$; $\chi^2 = 569.70$, $P < 0.05$; $\chi^2 = 396.68$, $P < 0.05$). The majority of tabanids, 36.4%, were collected from canopy traps baited with aged cow urine, 26% were obtained with aged horse urine, 21.4% were obtained with sheep urine, while 15.5% were obtained from traps baited with pig urine. Less than 1% of the total collected was obtained from traps without an attractant (Table 2). According to this data the aged cow urine baited canopy traps collected 51 times more tabanids than unbaited traps, while aged horse, aged sheep, and aged pig urine baited traps collected 36, 30, and 22 times as many tabanids, respectively, than unbaited traps. Canopy traps baited with cow urine collected significantly more *Tabanus bromius, Tabanus maculicornis*, *Tabanus tergestinus,* and *Hybomitra bimaculata* females then did traps baited with other natural attractants: aged horse, sheep, and pig urine (Table 3). The number of females of *Haematopota pluvialis* collected from cow urine baited canopy traps and horse urine baited canopy traps did not differ significantly ($\chi^2 = 0.66$ $P > 0.05$). Whereas specimens of *Haematopota pluvialis* collected from cow urine baited canopy traps and traps baited with other natural attractants (sheep and pig urine) differed significantly ($\chi^2 = 29.02$ $P < 0.05$; $\chi^2 = 21.56$

Table 1. Tabanids collected by canopy traps baited with aged cow, horse, sheep, and pig urine, and with unbaited control traps.

Species	Control traps	Cow urine	Horse urine	Sheep urine	Pig urine	∑
Tabanus bromius L.	4	443	299	265	120	1,131
Haematopota pluvialis (L.)	14	308	288	188	203	1,001
Tabanus maculicornis Zetterstedt	3	114	66	77	49	309
Tabanus sudeticus Zeller	0	62	67	49	40	218
Tabanus tergestinus Egger	0	49	17	21	18	105
Hybomitra bimaculata (Macquart)	0	48	8	6	1	63
Hybomitra ciureai (Séguy)	0	19	3	6	2	30
Chrysops viduatus (F.)	0	7	3	10	8	28
Chrysops relictus Meigen	0	5	1	3	7	16
Atylotus loewianus (Villeneuve)	0	1	4	2	2	9
Tabanus bovinus L.	0	2	2	0	2	6
Haematopota italica Meigen	0	2	2	0	1	5
Haematopota subcylindrica Pandellé	0	1	3	1	0	5
Haematopota pandazisi Kröber	0	4	0	0	0	4
Chrysops caecutiens (L.)	0	1	1	0	1	3
Hybomitra ukrainica (Olsufjev)	0	0	0	0	2	2
Tabanus autumnalis L.	0	1	0	1	0	2
Hybomitra acuminata (Loew)	0	1	0	1	0	2
Hybomitra solstitialis (Meigen)	0	1	0	0	0	1
Totals	21	1,069	764	630	456	2,940

Table 2. Total number of Tabanidae collected monthly by five canopy traps during 2005.

Attractants/Month	June	July	August	∑
Cow urine	445	411	213	1,069
Horse urine	271	428	65	764
Sheep urine	247	314	69	630
Pig urine	222	204	30	456
Control	8	11	2	21
Totals	1,193	1,368	379	2,940

$P < 0.05$). Furthermore, canopy traps baited with cow urine collected significantly more *Tabanus sudeticus* females than did traps baited with pig urine ($\chi^2 = 4.74$ $P < 0.05$). However, the number of collected females of *Tabanus sudeticus* did not differ significantly when comparing cow urine with horse and sheep urine ($\chi^2 = 0.18$ $P > 0.05$; $\chi^2 = 1.52$ $P > 0.05$). The response of the other species to natural attractants was not analyzed because of small sample sizes. The largest proportion of tabanids, 87.1% was caught during June and July (Table 2). Few tabanids were collected in August because of a long rainy period.

DISCUSSION

Aged cow urine baited canopy traps appeared to be the best collection methods for tabanids (Table 2). This study confirmed the established role of aged cow urine as a tabanid attractant (Okech and Hassanali 1990, Madubunyi et al. 1996). Also, the majority of species was collected in canopy traps baited with aged cow urine (Table 1). Moreover, the present study showed that aged cow urine is a very effective attractant for collecting *Tabanus bromius, Tabanus maculicornis, Tabanus tergestinus,* and *Hybomitra bimaculata* in the study area (Table 3). Madubunyi et al. (1996) also found that aged cow urine significantly increased the catch of some species from the family Glossinidae. However, the results with aged cow urine and aged horse urine are very similar and not significant for *Haematopota pluvialis*. Furthermore, the results with aged cow, aged horse, and aged sheep urine are very similar and not significant for the *Tabanus sudeticus*. Also, Krčmar et al. (2005) showed that aged horse urine baited traps collected significantly more females of *Tabanus sudeticus* than did traps baited with synthetic attractants. Bacteria convert trace chemicals in the urine to phenolic compounds that attract some species of tsetse, as well as most tabanids (Madubunyi et al. 1996). Fresh urine is not attractive (Okech and Hassanali 1990). Unfortunately, the efficiency and use of natural baits such as aged horse urine for other dipteran families is still very poor (Krčmar et al. 2005) and little has been published regarding the use of sheep and pig urine as attractants for tabanids. However, the response of some Glossinidae species, such as *Glossina austeni,* towards the pig urine was studied in Zanzibar, where catches of these flies did not increase significantly (Vreysen et al. 2000). On the other hand, the positive response of some Glossinidae species towards urine was observed in Kenya (Brightwell and Dransfield 1997). Likewise, when dispensed at the known natural doses of ox odour, a synthetic blend of acetone, carbon dioxide, 1-octen-3-ol, various phenols, and butanone is about half as effective as natural ox odor in the collection of Glossinidae species (Bursell et al. 1988, Hargrove et al. 1995, Torr et al. 1995).

Unfortunately, these data are not comparable with our prior results for Tabanidae. When traps were baited with aged horse urine, Krčmar et al. (2005) collected seven times the number of tabanids. The present study showed a much better effect of aged horse urine because baited traps collected 36 times more tabanids than unbaited traps. Because the effect of only aged horse urine was investigated, more extensive field studies using the aged urine of other species such us pig and sheep may be useful. The use of aged urine of domestic animals (cow, horse, sheep, and pig) in traps could result in the reduction of tabanids in certain geographic regions.

Table 3. The χ^2 - test values for some of the most abundant tabanids collected by canopy traps baited with cow urine and traps baited with other natural attractants.

Species/Attractants	Horse urine	Sheep urine	Pig urine
Tabanus bromius	27.94*	44.74*	185.30*
Tabanus maculicornis	12.80*	7.16*	25.92*
Tabanus tergestinus	15.50*	11.20*	14.34*
Hybomitra bimaculata	28.56*	32.66*	45.08*

* Significant differences ($P < 0.05$).

REFERENCES CITED

Brightwell, R. and R.D. Dransfield. 1997. Odour attractants for tsetse: *Glossina austeni, G. brevipalpis* and *G. swynnertoni*. Med. Vet. Entomol. 11: 297-299.

Bursell, E., A.J.E. Gough, A. Beevor, A. Cork, D.R. Hall, and G.A. Vale. 1988. Identification of components of cattle urine attractive to tsetse flies, *Glossina* spp. (Diptera: Glossinidae). Bull. Entomol. Res. 78: 281-291.

Chvála, M. 1988. Family Tabanidae. In: Á. Soós, (ed.), *Catalogue of Palaearctic Diptera. Athericidae - Asilidae*. Akadémiai kiadó, Budapest, 5: 97-171.

Chvála, M., L. Lyneborg, and J. Moucha. 1972. *The horse flies of Europe (Diptera, Tabanidae)*. Entomological Society of Copenhagen, Copenhagen, 499 pp.

Doskočil, J. and M. Chvála. 1974. Specific reactivity of horse flies (Diptera, Tabanidae) to traps baited with carbon dioxide. Folia Fac. Sci. Nat. Univ. Purk. Brun. 15: 51-55.

Foil, L.D. and L.J. Hribar. 1995. Evaluation of tsetse attractannts as baits for horse flies and deer flies (Diptera: Tabanidae) in Louisiana. Fla. Entomol. 78: 129-133.

French, F.E. and D.L. Kline. 1989. 1-octen-3-ol, an effective attractant for Tabanidae (Diptera). J. Med. Entomol. 26: 459-461.

Gibson, G. and S.J. Torr. 1999. Visual and olfactory responses of haematophagous Diptera to host stimuli. Med. Vet. Entomol. 13: 2-23.

Hall, M. and R. Wall. 2004. Biting flies: their role in the mechanical transmission of trypanosomes to livestock and methods for their control. In: I. Maudlin, P.H. Holmes, and M.A. Miles (eds.), *The trypanosomiases*. CAB Publishing 33: 573-583.

Hargrove, J.W., M.T.P. Holloway, G.A. Vale, A.J.E. Gough, and D.R. Hall. 1995. Catches of tsetse flies (*Glossina* spp) (Diptera: Glossinidae) from traps and targets baited with large doses of natural and synthetic host odour. Bull. Entomol. Res. 85: 215-227.

Hayes, R.O., O.W. Doane, JR. G. Sakolsky, and S. Berrick. 1993. Evaluation of attractants in traps for greenhead fly (Diptera: Tabanidae) collections on a Cape Cod, Massachusetts, salt marsh. J. Am. Mosq. Contr. Assoc. 9: 436-440.

Hribar, L.J., D.J. Leprince, and L.D. Foil. 1991. Design for a canopy trap for collecting horse flies (Diptera: Tabanidae). J. Am. Mosq. Contr. Assoc. 7: 657-659.

Hribar, L.J., D.J. Leprince, and L.D. Foil. 1992. Ammonia as an attractant for adult *Hybomitra lasiophthalma* (Diptera: Tabanidae). J. Med. Entomol. 29: 346-348.

Kline, D.L., J.R. Wood, and J.A. Cornell. 1991. Interactive effects of 1-octen-3-ol and carbon dioxide on mosquito (Diptera: Culicidae) surveillance and control. J. Med. Entomol. 28: 254-258.

Kristensen, P.K. and C. Sommer. 2000. Ammonia and 1-octen-3-ol as attractants for *Haematopota pluvialis*, *Hybomitra expollicata* (Diptera: Tabanidae) and *Morellia* spp. (Diptera: Muscidae). J. Med. Entomol. 37: 984-985.

Krčmar, S., L.J. Hribar, and M. Kopi. 2005. Response of Tabanidae (Diptera) to natural and synthetic olfactory attractants. J. Vector. Ecol. 30: 133-136.

Leprince, D.J., L.J. Hribar, and L.D. Foil. 1994. Responses of horse flies (Diptera: Tabanidae) to Jersey Bullocks and Canopy traps baited with ammonia, octenol and carbon dioxide. J. Med. Entomol. 31: 729-731.

Madubunyi, L.C., A. Hassanali, W. Ouma, D. Nyarango, and J. Kabii. 1996. Chemoecological role of mammalian urine in host location by tsetse, *Glossina* spp. (Diptera: Glosssinidae). J. Chem. Ecol. 22: 1187-1199.

Mihok, S., S.K. Moloo, J.O. Odeny, R.A. Brett, J.G. Rakwar, E. Munyoki, J. Kiilu, and C. Kyorku. 1996. Attractiveness of black rhinoceros (*Diceros bicornis*) to tsetse flies (*Glossina* spp) (Diptera, Glossinidae) and other biting flies. Bull. Entomol. Res. 86: 33-41.

Mohamed-Ahmed, M. M. and S. Mihok. 1999. Responses of *Glossina fuscipes fuscipes* (Diptera: Glossinidae) and other Diptera to carbon dioxide in linear and dense forests. Bull. Entomol. Res. 89: 177-184.

Nilssen, A.C. 1998. Effect of 1-octen-3-ol in field trapping *Aedes* spp. (Diptera, Culicidae) and *Hybomitra* spp. (Diptera, Tabanidae) in subarctic Norway. J. Appl. Entomol. 122: 465-468.

Okech, M. and A. Hassanali. 1990. The origin of phenolic tsetse attractants from host urine studies on the proattractants and microbes involved. Insect Sci. Appl. 11: 363-368.

Roberts, R.H. 1971. Effect of amount of CO_2 on collection of Tabanidae in Malaise traps. Mosq. News 31: 551-558.

Roberts, R.H. 1975. Relationship between the amount of CO_2 and the collection of Tabanidae in Malaise traps. Mosq. News 35: 150-154.

Torr, S.J., D.R. Hall, and J.L. Smith. 1995. The responses of tsetse flies (Diptera: Glossinidae) to natural and synthetic ox odour. Bull. Entomol. Res. 85: 157-166.

Vale, G.A. and D.R. Hall. 1985. The role of 1-octen-3-ol, acetone and carbon dioxide in the attraction of tsetse flies, *Glossina* spp. (Diptera: Glossinidae), to ox odour. Bull. Entomol. Res. 75: 209-217.

Vreysen, M.J.B., K.M. Saleh, Z.R. Zhu, and F.W. Suleiman. 2000. Responses of *Glossina austeni* to sticky panels and odours. Med. Vet. Entomol. 14: 283-289.