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OLFACTORY ATTRACTANTS FOR MOSQUITO SURVEILLANCE AND CONTROL: 1-OCTEN-3-OL[1]

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ABSTRACT. The role of octenol as a mosquito attractant is still under investigation. When used alone, octenol has been a good attractant for only a few species. However, there appears to be a synergistic response of species of the genera *Aedes, Anopheles, Coquillettidia, Psorophora,* and *Mansonia* to the combination of octenol and CO_2. Further research is required to determine the potential role of this compound in mosquito management programs.

INTRODUCTION

1-octen-3-ol (octenol) is a volatile compound that has been isolated from many natural sources, mainly plants and fungi (Dijkstra and Wiken 1976). It has been identified in volatiles from clover (Honkanen and Moisio 1993) and alfalfa (Buttery and Kamm 1980). It is produced by numerous mold species (Kaminski et al. 1974) and mushrooms (Wurzenberger and Grosch 1982), and is known to be formed when grain is contaminated by mold (Kaminski et al. 1973). Octenol has also been isolated from animals (invertebrates and vertebrates). Hall et al. (1984) found octenol to be present in the breath of oxen. Raymer et al. (1985) reported that octenol is apparently produced by microorganisms in the anal sac of wolves. Pierce et al. (1989) reported that 2 species of grain beetles, *Oryzaephilus surinamensis* (Linn.) and *O. mercator* (Fauvel) produce octenol for use as an aggregation pheromone.

Chemically speaking, octenol is an 8-carbon mono-unsaturated alcohol that has an asymmetric center and therefore 2 optical isomers with a terminal double bond. Mushroom-produced 1-octen-3-ol was found to be the (R)-(−)enantiomer (Wurzenberger and Grosch 1984). (Z)-2-octenol is also characteristic of some molds (Kaminski et al. 1972) and mushrooms (Tressl et al. 1982). Oxen- and grain beetle-produced octenol were predominantly the (R)-(−)enantiomer; both enantiomers have been shown to be attractive to grain beetles (Pierce et al. 1989) and tsetse (Hall et al. 1984).

Since octenol's isolation from the breath of oxen by Hall et al. (1984), field tests have demonstrated that it serves as a powerful attractant for certain species of tsetse flies (Vale and Hall 1985). It has been successfully used as an attractant in tsetse control programs in Zimbabwe and other parts of Africa (Torr 1994). Workers in the USA have found that octenol's effect on the response of certain host-seeking mosquitoes (Takken and Kline 1989; Kline et al. 1990a, 1990b, 1991a, 1991b), ceratopogonids (Kline et al. 1994), tabanids (French and Kline 1989), and oestrids (Anderson 1989) is similar to that of tsetse.

The objective of this paper is to review the current status of our knowledge concerning the responses of mosquitoes to octenol, the potential use of this compound in mosquito surveillance and management programs, and to make recommendations for additional research.

FIELD STUDIES

After octenol was first reported as a mosquito attractant by Takken and Kline (1989), a series of field studies was conducted in a wide variety of ecological habitats. The basic experimental design described by Takken and Kline (1989) has been used in these field studies to determine the range of species that will respond to octenol either alone or in combination with other attractants. Field studies have been conducted in estuarine ecosystems in Florida and Georgia, such as mangrove swamps and brackish salt marshes (Kline et al. 1990b, 1991b; Takken and Kline 1989). Other field studies included cypress and red maple freshwater swamps (Takken and Kline 1989, unpublished data), phosphate-mined areas in Florida (Kline et al. 1990a), irrigated riceland in Arkansas (Kline et al. 1991a), tire dumps in Louisiana, and livestock holding areas in Colorado (Kline et al., unpublished data).

At each site the traps were baited either with octenol alone or in combination with carbon dioxide (CO_2). Butanone, lactic acid, and mixed phenols were used in one study with estuarine-associated mosquito species (Kline et al. 1991b). The dispensing system used to release octenol was adapted from that described by Hall et al. (1984). Octenol was released from microreaction

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[1] Mention of a commercial or proprietary product in this paper does not constitute an endorsement of this product by the United States Department of Agriculture.

vials (5 ml; Supelco, Bellefonte, PA) fitted with plastic lids and neoprene septa using a wick (Dills 15-cm pipe cleaner) system. Each lid had a 12-mm hole bored into the top. Two different release rates were obtained by modifying the wick release system. A low-level release rate (2–5 mg/h) was achieved by fashioning the wick so that its length (doubled over) was a few millimeters longer than the distance from the bottom of the vial to the septum positioned within the lid. This configuration allowed the wick to remain in contact with the permeable septum, releasing the octenol. High-level octenol release (37–43 mg/h) was achieved by allowing the doubled-over wick to protrude through a 1.5-mm hole and rise *ca.* 2 cm above the septum. Both configurations are illustrated in Kline et al. (1991a). Both high and low release rates were monitored daily by weighing (to nearest 0.001 g) all vials directly before and after each trapping period. Vials were affixed near the trap entrance, and when used in combination with CO_2, they were affixed adjacent to the CO_2 release point.

When CO_2 was used, it was metered from a 9-kg compressed gas cylinder at the desired flow rate using a double-stage pressure regulator. Gas flow was monitored using a compact flow meter (Gilmont Instruments, Great Neck, NY) and was delivered to its release point *ca.* 5 cm from the trap entrance via polyethylene tubing. To assure accurate flow calibration, micrometering valves (series M, Nupro, Willoughby, OH) were used in place of the standard open–shut valves found on the pressure regulators. Carbon dioxide was used either at 200 or 500 ml/min or both, depending on what the project cooperators normally used as a standard in their mosquito-surveillance programs.

Thirty-five species of mosquitoes were collected during these studies. These included *Aedes aegypti* (Linn.), *Aedes albopictus* (Skuse), *Aedes atlanticus* Dyar and Knab, *Aedes dorsalis* (Meigen), *Aedes dupreii* (Coq.), *Aedes flavescens* Theobald, *Aedes infirmatus* Dyar and Knab, *Aedes sollicitans* (Walker), *Aedes taeniorhynchus* (Wied.), *Aedes triseriatus* (Say), *Aedes trivittatus* (Coq.), *Aedes vexans* (Meigen), *Anopheles atropos* Dyar and Knab, *Anopheles crucians* Wied., *Anopheles quadrimaculatus* Say, *Coquillettidia perturbans* (Walker), *Culex erraticus* (Dyar and Knab), *Culex nigripalpus* Theobald, *Culex opisthopus* Komp, *Culex pilosus* (Dyar and Knab), *Culex pipiens* Linn., *Culex quinquefasciatus* Say, *Culex salinarius* Coq., *Culex tarsalis* Coq., *Culiseta inornata* (Theobald), *Culiseta melanura* (Coq.), *Mansonia dyari* Belkin, Heinemann and Page, *Mansonia titillans* (Walker), *Psorophora ciliata* (Fabricius), *Psorophora columbiae* (Dyar and Knab), *Psorophora ferox* (Von Humboldt), *Psorophora howardi* Coq., *Psorophora mathesoni* Belkin and Heinemann, *Wyeomyia mitchellii* (Theobald), and *Wyeomyia vanduzeei* Dyar and Knab. Data for the 23 most abundant species encountered are summarized in Table 1. These data and those for the other species collected indicate several basic trends:

1) CO_2 resulted in a response in all the species studied; the pattern of response to increasing CO_2 levels varied from species to species.
2) Very few species consistently respond in large numbers (i.e., response to octenol alone is $\geq 5\%$ of that obtained to CO_2 alone) to octenol alone; exceptions were *Ae. sollicitans, Ae. taeniorhynchus, Ae. triseriatus, Cx. salinarius,* and *Ma. titillans.*
3) The combination CO_2 + octenol causes a synergistic increase (i.e., greater than the combined response obtained by CO_2- and octenol-alone-baited traps) in the collections of many mosquito species, especially species of *Aedes, Anopheles, Psorophora, Coquillettidia,* and *Mansonia*; this response appears to vary geographically, seasonally, and according to the physiological state of the mosquito, especially for *Anopheles* spp.
4) *Culex* spp. generally show little response to octenol alone or in combination with CO_2; any increase tends to be an additive effect; *Cx. salinarius,* as noted above, is an exception to this generalization.
5) Lactic acid and mixed phenols alone and in combination with octenol attract *Ae. taeniorhynchus*; the effect is additive.
6) Octenol, at the release rates tested, tends to repel *Cs. melanura.*

These general trends have also been observed in the only other published research field study on the response of mosquito species to octenol. Kemme et al. (1993) conducted field studies near Queensland, Australia, and reported that 4 estuarine-associated species (*Aedes vigilax* (Skuse), *Aedes funereus* (Theobald), *Culex annulirostris* Skuse, and *Culex sitiens* Wied.) were collected in numbers large enough for a statistical analysis. These investigators concluded that octenol alone (5 mg/h) was only slightly attractive for the *Aedes* spp. Few *Culex* were collected by the octenol-baited traps. Using CO_2-alone-baited (200 ml/min) traps, all 4 species were collected in significantly increased ($P < 0.05$) numbers. The largest collections of *Ae. vigilax* and *Ae. funereus* were made using CO_2 + octenol-baited traps. The increase relative to CO_2 alone was statistically significant only for *Ae. vigilax* owing to the high variability of the data. Collections of both *Culex* species decreased with the addition of octenol to CO_2-baited traps.

Table 1. Field studies conducted to determine the responses of natural populations to octenol alone and in combination with carbon dioxide.

		Response index[2]						
Species	Study[1]	Low octenol	High octenol	Low octenol + 200 CO_2	High octenol + 200 CO_2	Low octenol + 500 CO_2	High octenol + 500 CO_2	Reference
Ae. albopictus	LA—Sept. 1987a	0.19		1.77				Unpublished
	LA—Sept. 1987b	0.01		1.36				Unpublished
Ae. dorsalis	CO—June 1989		0.01		1.21			Unpublished
Ae. infirmatus	SI—Aug. 1989a	0.5	<0.01	2.38	0.75			Unpublished
	SI—Aug. 1989b	0.07		2.70				Unpublished
	VB—Apr. 1993	0.02		1.77				Unpublished
Ae. sollicitans	SI—Aug. 1989a	0.07	0.09	2.58	1.65			Unpublished
	SI—Aug. 1989b	0.11		2.75				Unpublished
Ae. taeniorhynchus	ENP—Oct. 1986	0.90		5.90				Takken and Kline (1989)
	LSWR—Nov. 1986	1.67		16.00				Takken and Kline (1989)
	ENP—Mar. 1987	0.09		1.59				Kline et al. (1990b)
	ENP—Oct. 1987	0.10		2.26				Kline et al. (1990b)
	SI—Aug. 1989a	0.05	0.03	1.94	1.03			Unpublished
	SI—Aug. 1989b	0.07		2.35				Unpublished
Ae. triseriatus	LA—Sept. 1987a	0.55		3.18				Unpublished
	LA—Sept. 1987b	0.02		1.76				Unpublished
Ae. trivittatus	CO—June 1989		0.00		1.38			Unpublished
Ae. vexans	LA—Sept. 1987b	0.00		0.41				Unpublished
	CO—June 1989		0.01		1.01			Unpublished
An. atropos	LSWR—Oct. 1986	0.00		58.00				Takken and Kline (1989)
	ENP—Mar. 1987	0.01		5.49				Kline et al. (1990b)
	SI—Aug. 1989a	0.00	0.00	0.98	0.81			Unpublished
	SI—Aug. 1989b	0.00		2.13				Unpublished
An. crucians	ENP—Oct. 1986	0.00		32.00				Takken and Kline (1989)

Table 1. Continued.

Species	Study^1	Response index^2						Reference
		Low octenol	High octenol	Low octenol + 200 CO$_2$	High octenol + 200 CO$_2$	Low octenol + 500 CO$_2$	High octenol + 500 CO$_2$	
An. crucians	LSWR—Oct. 1986	0.30		6.60				Takken and Kline (1989)
	ENP—Mar. 1987	<0.01		1.50				Kline et al. (1990b)
	ARK—July 1987	0.00*				0.80		Kline et al. (1991a)
	ARK—July 1988	0.00		1.75		0.90		Kline et al. (1991a)
	ARK—Aug. 1988	0.00	0.09	3.18	2.40			Kline et al. (1991a)
	PKC—Sept. 1987		0.02			0.91		Kline et al. (1990a)
	PKC—May 1988		0.01/0.01*				1.27	Kline et al. (1990a)
	SI—Aug. 1989a	0.00	0.00	0.96	0.37			Unpublished
	SI—Aug. 1989b	0.00		2.02				Unpublished
	VB—Apr. 1993	0.00		7.10				Unpublished
An. quadrimaculatus	LSWR—Oct. 1986	0.46		9.46				Takken and Kline (1989)
	ARK—July 1987	0.01*				2.32		Kline et al. (1991a)
	PKC—Sept. 1987		0.02				0.91	Kline et al. (1990a)
	PKC—May 1988		0.00				0.13	Kline et al. (1990a)
	ARK—July 1988	0.00		1.02		0.77		Kline et al. (1991a)
	ARK—Aug. 1988	<0.01	<0.01	1.55	0.66			Kline et al. (1991a)
Culex (*Melanoconion*) spp.	ENP—Oct. 1986	0.00		0.54				Takken and Kline (1989)
	ENP—Mar. 1987	0.08		1.02				Kline et al. (1990b)
	ARK—July 1987	0.17*				1.48		Kline et al. (1991a)

Table 1. Continued.

Species	Study[1]	Response index[2]						Reference
		Low octenol	High octenol	Low octenol + 200 CO_2	High octenol + 200 CO_2	Low octenol + 500 CO_2	High octenol + 500 CO_2	
Cx. (*Melanoconion*) spp.	ENP—Oct. 1987	0.00		2.27				Kline et al. (1990b)
	PKC—Sept. 1987		0.12*			0.69		Kline et al. (1990a)
	PKC—May 1988		0.30/0.30*			0.50		Kline et al. (1990a)
	ARK—July 1988	0.00		0.71		0.74		Kline et al. (1991a)
	ARK—Aug. 1988	0.07	0.03	1.59	1.48			Kline et al. (1991a)
Cx. nigripalpus	ENP—Mar. 1987	0.01		0.97				Kline et al. (1990b)
	ENP—Oct. 1987	0.01		1.95				Kline et al. (1990b)
	PKC—Sept. 1987		0.07*				1.20	Kline et al. (1990a)
	VB—Apr.	0.00		0.77				Unpublished
Cx. pipiens	CO—June 1989	0.02		0.50				Unpublished
Cx. salinarius	ARK—July 1988	0.00		3.13		4.25		Kline et al. (1991a)
	ARK—Aug. 1988	0.07	0.03	2.58	4.14			Kline et al. (1991a)
	PKC—Sept. 1988		0.19/0.12*				1.34	Kline et al. (1990a)
	SI—Aug. 1989a	0.07	0.02	1.26	0.94			Unpublished
Cx. tarsalis	CO—June 1989	<0.01		0.75				Unpublished
Cq. perturbans	ENP—Oct. 1987	0.00		7.50				Kline et al. (1990b)
	ARK—July 1987	0.00*				3.40		Kline et al. (1991a)
	ARK—July 1988	0.00		2.53		2.06		Kline et al. (1991a)

Table 1. Continued.

Species	Study[1]	Response index[2]						Reference
		Low octenol	High octenol	Low octenol + 200 CO_2	High octenol + 200 CO_2	Low octenol + 500 CO_2	High octenol + 500 CO_2	
Cq. perturbans	ARK—Aug. 1988	0.00	0.00	2.47	5.47			Kline et al. (1991a)
	PKC—Sept. 1988		0.23*				9.49	Kline et al. (1990a)
	PKC—May 1988		0.07/0.07*				3.91	Kline et al. (1990a)
Ma. titillans	PKC—May 1988		0.32/0.37*				4.97	Kline et al. (1990a)
Ps. ciliata	ARK—July 1987	0.00*				2.62		Kline et al. (1991a)
Ps. columbiae	ARK—July 1987	0.02*				3.22		Kline et al. (1991a)
	ARK—July 1988	0.00		3.44		2.25		Kline et al. (1991a)
	ARK—Aug. 1988	0.01	0.04	1.65	1.74			Kline et al. (1991a)
	SI—Aug. 1989a	0.00	0.00	5.33	2.67			Unpublished
	SI—Aug. 1989b	0.40		3.00				Unpublished
Ps. ferox	VB—Apr. 1993	0.00		1.28				Unpublished
Ps. mathesoni	ARK—July 1987		0.00*			0.89		Kline et al. (1991a)
Wy. mitchellii	ENP—Oct. 1986	0.10		1.29				Takken and Kline (1989)
	ENP—Mar. 1987	0.20		0.50				Kline et al. (1990b)
	ENP—Oct. 1987	0.06		0.64				Kline et al. (1990b)

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[1] LA = Louisiana tire dump; CO = Colorado livestock area; SI = salt marsh at Sea Island, GA; VB = mangrove swamp at Vero Beach, FL; ENP = mangrove area at Everglades National Park, near Flamingo, FL; LSWR = Lower Suwannee Wildlife Refuge where there are freshwater swamps and salt-marsh habitats; ARK = irrigated ricelands near Stuttgart, AR; PKC = phosphate mining pits near Bartow, Polk County, FL.

[2] Response index = mean number of mosquitoes collected by an octenol-baited trap divided by mean number of mosquitoes collected by either the 200 or 500 ml/min CO_2 standard, or both (an * means that the octenol treatment was compared to a 500 ml/min CO_2 standard).

PRACTICAL APPLICATION

Bonvechio (1991) described the only known operational use of octenol by a mosquito abatement district. The Collier Mosquito Control District, located in southwest Florida, now uses 25 New Jersey (NJ) light traps baited with octenol for routine surveillance of mosquito populations. These NJ traps with photoelectric switches are placed in various locations throughout the Collier Mosquito Control District. The traps are wired to 110-volt AC power and equipped with a 7.5-watt light bulb. Octenol is placed in microreaction vials as described above except that a standard pipe cleaner cut in half is used as the wick through the rubber septum. One microreaction vial is attached with Velcro (Velcro Fasteners, Manchester, NH) to each NJ trap near the fan entrance. Each microreaction vial needs to be refilled approximately every 4 wk. No CO_2 is used. So far 21 mosquito species have been collected, including *Ae. taeniorhynchus, Cx. nigripalpus,* and *Ps. columbiae,* which are the main pest species for Collier Mosquito Control District. Other species collected include *Ae. atlanticus, Ae. infirmatus, Aedes fulvus pallens* Ross, *Ae. triseriatus, Ae. aegypti, Ae. sollicitans, An. crucians, An. quadrimaculatus, An. atropos, Cx. erraticus, Cs. melanura, Cq. perturbans, Ma. titillans, Ps. ferox, Uranotaenia lowii* Theobald, and *Wy. mitchellii.* The use of octenol in combination with the 7.5-watt light bulb resulted in a reduction in the large amounts of unwanted insects when a 25-watt bulb is used and the need to use CO_2, which district personnel found to be time consuming and expensive. Fewer mosquitoes were collected by the octenol technique than with the 25-watt bulb, but daily and seasonal trends were the same (Bonvechio, personal communication). These traps proved their usefulness during the St. Louis encephalitis epidemic year of 1990, providing valuable information on the relative abundance of *Cx. nigripalpus* in the area. This response of *Cx. nigripalpus* seems to contradict the findings reported above, but this species may be responding to the 7.5-watt light more than to the octenol. In a study conducted in Sarasota County *Cx. nigripalpus* responded in very large numbers to small wattage light traps (J. R. Wood, unpublished data) even if not baited with CO_2.

RECOMMENDATIONS FOR ADDITIONAL FIELD STUDIES

The following concerns need to be addressed to truly determine the potential of octenol for surveillance or control of mosquito species:

1) The precise mechanism of octenol as a kairomone in the host-seeking behavior of mosquitos remains unknown. Based on published tsetse work (Bursell 1984), it is expected that octenol does not activate mosquitoes to flight, but once they are airborne it may elicit upwind anemotaxis, but not induce alighting responses. Also the horizontal distance from which octenol elicits an upwind response is probably greater than that for CO_2. These hypotheses need to be tested.
2) The effect(s), if any, that chronological and/or physiological age may have on mosquito response to octenol needs to be determined.
3) The impact, if any, species complexes have on our interpretation of mosquito responses to octenol.
4) The response of some species and not others to octenol. Is it due to a lack of specific octenol receptors in some species, or limits set by release rates tested so far? The natural release rate by oxen is 0.043 mg/h (Hall et al. 1984). In these field studies the release rate used was either 4 or 40 mg/h.
5) It needs to be determined if over the course of a mosquito season that the response rate of a particular mosquito species to a specific release rate of octenol is constant. If not, why not?

CONCLUSION

Unlike CO_2, which appears to be a universal attractant for mosquitoes, octenol (based on current methods of testing) appears to be an attractant for only a few mosquito species. The limited research that has been conducted and reported on octenol as a mosquito attractant indicates a potential use in mosquito management programs for those species that do respond to it. The synergistic response of species, such as *Ae. taeniorhynchus,* to octenol and CO_2 especially provides optimism that a control strategy based on semiochemicals can be devised.

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