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TESTING HUMAN URINE AS A LOW-TECH BAIT FOR *ANASTREPHA* SPP. (DIPTERA: TEPHRITIDAE) IN SMALL GUAVA, MANGO, SAPODILLA AND GRAPEFRUIT ORCHARDS

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ABSTRACT

We evaluated the attractiveness of three aqueous dilutions of human urine (HU 50, 25, and 12.5%) to adults of pestiferous and nonpestiferous *Anastrepha* species (Diptera: Tephritidae) in small guava, grapefruit, mango, and sapodilla orchards with glass McPhail traps. As control treatments we used a commercially available hydrolyzed protein bait (Captor Plus®) and tap water. In the guava orchard, the three urine dilutions were as effective as hydrolyzed protein in attracting *A. fraterculus*. Also, when 25 and 50% urine were used, 93 and 96%, respectively, of the adults captured were females. In the grapefruit orchard, protein-baited traps captured significantly more *A. ludens* than urine-baited traps. In the mango orchard, both *A. obliqua* and *A. serpentina* were more attracted to hydrolyzed protein than to any other bait treatment. In the sapodilla orchard, traps baited with 50% urine surpassed all other treatments in the capture of *A. serpentina* and *A. obliqua*. Our findings indicate that human urine performs as well or better than hydrolyzed protein in certain types of orchards. They also support the notion that there is no "universal" *Anastrepha* bait. We conclude that human urine is a viable, low-tech alternative *Anastrepha* bait for subsistence or low income, small-scale fruit growers in rural Latin America.

Key Words: *Anastrepha*, Tephritidae, human urine, hydrolyzed protein, trapping, attractants

RESUMEN

Evaluamos el potencial atractivo de 3 diluciones acuosas de orina humana (OH 50, 25 y 12.5%) para adultos de especies plaga y no plaga de *Anastrepha* (Diptera: Tephritidae) en pequeños huertos de guayaba, toronja, mango y chico zapote utilizando trampas McPhail de vidrio. Los tratamientos control consistieron de proteína hidrolizada (Captor Plus®) y agua. En la huerta de guayaba, las tres diluciones de orina fueron igual de efectivas que la proteína hidrolizada en capturar *A. fraterculus*. A diluciones de 25 y 50% de orina, el 93 y 96%, respectivamente, de las capturas fueron hembras. En la huerta de toronja, las trampas cebadas con proteína capturaron significativamente más adultos que aquellas cebadas con orina. Un resultado similar se obtuvo en la huerta de mango donde se capturaron adultos de *A. obliqua* y *A. serpentina*. En la huerta de chico zapote, las trampas cebadas con orina al 50% superaron a todos los demás tratamientos capturando significativamente más adultos de *A. serpentina*. Nuestros resultados indican que la orina tiene un potencial atractivo similar o en algunos casos mayor que la proteína hidrolizada en ciertos tipos de huertos. También apoyan la noción de que no existe un cebo "universal" para *Anastrepha*. Concluimos que la orina humana representa una alternativa viable de baja tecnología para pequeños productores de bajo ingreso o subsistencia en áreas rurales de América Latina.

Translation provided by author.

In recent years, there has been renewed interest in developing more efficient baits and traps for monitoring economically important fruit flies (Diptera: Tephritidae). Even though most resources have been invested in developing Medfly (*Ceratitis capitata* [Wiedemann]) traps (e.g., Heath et al. 1995, 1996a, 1997; Epsky et al. 1995, 1999; Katsoyannos et al. 1999a, b), there have been some interesting trap developments for *Anastrepha* spp. (Heath et al. 1995, 1997; Thomas et al. 2001), *Rhagoletis* spp. (Liburd et al. 1998; Prokopy et al. 2000; Stelinski & Liburd 2001), and *Toxotrypana curvicauda* Gerstaecker (Heath et al. 1996b).

A common theme in the approach followed to develop this new generation of traps has been finding optimal combinations of visual and chemical elements and the desire to find appropriate substitutes for liquid-based traps such as the McPhail (Epsky et al. 1995; Epsky & Heath 1997). Given the need to monitor female numbers in adult fruit fly populations (Casaña-Giner et al. 2001), there has been renewed interest in evaluating protein- and plant-based attractants. Although some of the resulting female-targeted traps have shown promising results (Heath et al. 1997; Epsky et al. 1999; Katsoyannos et al. 1999a, b), they still have to overcome questions of cost and manageability (e.g., susceptibility of traps to dust or theft). Cost considerations are of paramount importance given the trend toward phasing out large-scale governmental support for fruit fly management, and the subsequent transfer to growers of the responsibility for funding management and eradication programs. The situation in Latin America is particularly critical because large quantities of fruit are still produced by subsistence or resource-poor, small-scale growers who cannot afford expensive monitoring and management tools (Aluja & Liedo 1986; Aluja 1996, 1999). Furthermore, market niches for organically grown fruit are continuously expanding. As a result, the need for biorational management schemes has become more critical than ever.

In the case of flies in the genus *Anastrepha*, the challenge for developing a substitute for the McPhail trap is particularly difficult. This liquid-based trap, developed in the early 1900s (McPhail 1937, 1939), is still widely used throughout Latin America in spite of its inefficiency and high cost (Aluja et al. 1989; Aluja 1999). However, it sometimes outperforms the recently developed dry-based substitutes (e.g., Heath et al. 1995, 1997) because adult flies are attracted to the humid environment in and around it, particularly during the dry season. The biggest challenge in developing an effective substitute to the McPhail trap is the occurrence of at least seven economically important *Anastrepha* species (*A. fraterculus* [Wiedemann], *A. grandis* [Macquart], *A. ludens* [Loew], *A. obliqua* [Macquart], *A. serpentina* [Wiedemann], *A. striata* [Schiner], and *A. suspensa* [Loew]; Aluja 1994), and the evidence that not all species respond with equal intensity to a single bait (Aluja et al. 1989). As discussed by Aluja (1999) and Aluja et al. (2001), the most effective *Anastrepha* attractant will likely end up being a complex aromatic bouquet containing native host fruit and food-based odors, as well as sexual pheromones. Formulating such a lure and assembling a trap that is easy to handle and also visually attractive to all the economically important *Anastrepha* species will take time. But even if such a trap design is ever produced, its cost may be prohibitive especially given the low purchasing power of the vast majority of fruit growers in Latin America. Therefore, developing cheap, low-tech baits and traps that are easily accessible to local growers should remain a high priority.

An inexpensive alternative fruit fly bait was studied by Hedström (1988) in Costa Rica. This author tested human urine (HU) in a guava orchard, and found that McPhail traps baited with this naturally occurring compound (50% dilution in water) captured 10 times more *A. striata* and *A. obliqua* adults than traps baited with the commercially available torula yeast. In a laboratory study with *A. ludens*, *A. obliqua*, *A. serpentina*, and *A. striata* adults, Piñero et al. (2002) found that responses toward human urine depended on previous diet (e.g., protein-fed adults responded weakly to baits), reproductive state (responses were always greater in sexually mature individuals than sexually immature individuals), and sex (female responses were greater than male responses, particularly for sexually mature individuals) and, importantly, such responses varied among species. More recently, Piñero et al. (2003) determined that McPhail traps baited with human urine captured a high proportion of sexually immature *A. obliqua* and *A. serpentina* females in a commercial mango orchard (cultivar Manila).

Here, we report the results of a study aimed at determining the attractiveness of three aqueous dilutions of human urine in glass McPhail traps. In an effort to make this study as useful as possible to subsistence or small-scale, resource poor farmers, we tested this naturally occurring compound in four types of environments (guava, grapefruit, mango, and sapodilla orchards), comparing its effectiveness against the commercially available hydrolyzed protein bait (Captor Plus®) and tap water.

MATERIALS AND METHODS

Study Sites

We worked in Cosautlán, Apazapan, and Tuzamapan (central Veracruz, México), in small, unsprayed guava (*Psidium guajava* [L.]), grapefruit (*Citrus paradisi* [Macfadyn]), mango (*Mangifera indica* [L.]), and sapodilla (*Manilkara zapota* [L.] P. Royen) orchards, during the period of September 1995 to July 1997. Exact location of study sites, orchard characteristics, and fly trapping dates for each orchard are shown in Table 1.

Bait Treatments

Five bait treatments were evaluated in McPhail traps: 50%, 25%, and 12.5% water dilutions of human urine (HU50, HU25 and HU12.5, respectively), hydrolyzed protein (Captor Plus®, Agroquímica Tridente S.A. de C.V. México, D.F.; 10 ml of protein per l of water), and tap water (control treatment). Each trap was baited with 200 ml of the particular bait. The HU50, HU25, and HU12.5 dilutions were prepared by mixing 100, 50 and 25 ml of human urine in 100, 150 and 175 ml of tap water, respectively. To avoid modifying bait pH values, we did not use Borax as a preservative.

The human urine stemmed from a single source (JP) because more donors could not make a commitment for the entire study period (1995-1997). The donor was a healthy 26-year old male who followed a strict diet free of coffee, alcohol, vitamin supplements, food condiments such as hot chilies, and who did not smoke. Food ingested included vegetables, fruits, rice, meat, chicken, and

TABLE 1. DESCRIPTION OF STUDY SITES IN THE STATE OF VERACRUZ, MEXICO.

Site	North Latitude	West Longitude	Altitude (meters above sea level)	Fruit tree species	Orchard Characteristics	Trapping Period
Cosautlán	19°20'	96°59'	1,298	Guava (*Psidium guajava* L.)	About 20 guava trees with patches of cow pasture. Coffee plantation nearby. Guava fruit present on trees.	19 September-24 October, 1995
San Pedro-Tuzamapan	19°24'	96°52'	922	Grapefruit (*Citrus paradisi* Macfadyn)	Mixed grapefruit-coffee orchard interspersed with fruiting orange trees. Grapefruit present on trees.	29 November, 1995-05 January, 1996
Apazapan	19°19'	96°42'	320	Mango (*Mangifera indica* L.) (cultivar Manila)	Mango orchard adjacent to a sapodilla orchard. Very low amount of mango fruit on trees and no sapodilla fruit present.	23 June-01 September, 1996
Apazapan	19°19'	96°42'	320	Sapodilla (*Manilkara zapota* [L.] P. Royen)	Sapodilla orchard adjacent to a mango orchard. Sapodilla fruit on trees was abundant and mango harvest was over.	12 June-27 July, 1997

occasionally fish. This diet was started 15 days prior to the initiation of the first set of experiments and maintained throughout the study period. The donating individual underwent a medical checkup to determine possible kidney damage or any metabolic disorder. His urine was chemically characterized by a local laboratory (Laboratorio Hernández-Blázquez, Coatepec, Veracruz, México) and the results (exact information in Piñero et al. 2002) indicated that urea and ammonia contents fell within the normal ranges for a healthy individual (normal ranges: 20-30 g/100 ml for urea and 0.5-0.9 g/100 ml for ammonia; Bell et al. 1961; Anonymous 1981). Even though we acknowledge that there can be variability in the chemical composition of human urine due to factors such as age and the quality and quantity of food ingested (Bell et al. 1961; Langley 1971; Anonymous 1981), we believe that the two components critical for the study (urea and ammonia) varied relatively little throughout the study because the human urine used was always provided by a single, healthy individual who also followed a strict diet. Furthermore, we note that variability in bait composition is a common problem faced by researchers even when buying commercially produced protein-based baits. Hence, we believe that all appropriate procedures were followed.

Trap Placement and Servicing Procedure

Five fruit trees of similar size and fruit load were selected within each of the four orchard types, except in the sapodilla orchard, where 20 trees were used (see below). Tree canopy size ranged between 4-6 m for grapefruit, guava and sapodilla and 10-12 m for mango. Five glass McPhail traps, each baited with one of the five different bait treatments, were placed at equidistant locations (over 2 m apart in all cases and up to 8 m in some) in the interior portion of each tree canopy. However, in the sapodilla orchard, only one trap was placed per tree because branches were too thin and, hence, trees would have not supported all five traps. Every three days traps were inspected, cleaned, and re-baited. This procedure was carried out 20 times in each one of the orchards. Trap positions were systematically rotated each servicing day. Even though we acknowledge that placing 5 traps in the same tree may have caused interaction between the baits, we believe that given the relatively large size of the tree canopies in which traps were hung, and the fact that trap positions within the canopy were systematically rotated every three days, such a possible effect was most likely negligible. Besides, as our goal was to test human urine in low-tech, resource poor scenarios, orchards were small and therefore the number of trees available to us, with the exception of the sapodilla orchard, was not enough to test each bait in different trees.

Species and sex were determined for all adults captured. All females of the predominant fly species captured by traps in each orchard were examined under a dissecting stereomicroscope to determine the presence or absence of developed ovaries (a measure of sexual maturity), by the methods of Martínez et al. (1995). Also, in each orchard, ten pH readings were taken at different intervals with a portable pH meter (Cole-Parmer Model 59000-20, Chicago, IL, USA) for each one of the bait treatments.

Statistical Analyses

Since McPhail traps commonly capture adults of up to 12 *Anastrepha* species, statistical analyses were conducted only on the predominant species in each orchard (one or two species are normally abundant; Aluja et al. 1996). One-way analyses of variance (ANOVA) were carried out on fly/trap/day (FTD) values, pooling males and females for each fly species. FTD values represent the total number of adults captured per trap per day (Celedonio-Hurtado et al. 1995). Data were square-root transformed (X + 0.5) to homogenize variances but in the figures we present untransformed mean (±SE) values. In all cases, ANOVAs were followed by Fisher-protected LSD separations of treatment means. For the most representative fly species in each orchard type, linear regression analyses were conducted to further determine the degree of relationship between the three concentrations of human urine evaluated and attractiveness to adult flies. Comparisons of the numbers of females versus males of a particular species and bait treatment were performed with nonparametric Mann-Whitney tests. All analyses were carried out at the 0.05% level of significance, with the software Statistica® (StatSoft 1999).

RESULTS

Bait pH Values

Since there were no differences in pH values for similar bait treatments among orchards (F = 0.86, df = 3, 178, $P > 0.05$), we report global pH values for each bait (i.e., pooled over all orchards). The ANOVA indicated that there were differences in the pH values among the bait treatments (F = 16.71, df = 4, 178, $P < 0.0001$). The mean (±SE) pH value of water (6.7 ± 0.15) was significantly lower than the mean (±SE) pH values of the three human urine concentrations (among which there were no significant differences; 7.58 ± 0.21, 7.87 ± 0.27, and 8.17 ± 0.26, for HU12.5, HU25, and HU50, respectively), and was significantly lower than that of hydrolyzed protein (7.25 ± 0.23).

Guava Orchard

Altogether, 105 adults of four *Anastrepha* species were captured. The proportion of each species in the sample was as follows: *A. fraterculus* (84.8%), *A. ludens* (9.5%), *A. striata* (3.8%) and *A. obliqua* (1.9%). Consequently, results reported next refer only to *A. fraterculus*. Traps baited with human urine (all three concentrations) and hydrolyzed protein captured similar numbers of adults of this species and more adult flies than water-baited traps (F = 5.05, df = 4, 295, $P <$ 0.001) (Fig. 1). A regression analysis confirmed the lack of association between the concentrations of human urine evaluated and the number of adult *A. fraterculus* captured by traps (F = 0.48, P = 0.50, R^2 = 0.04). Notably, 93% and 96%, respectively, of the adults captured by traps baited with either HU25 or HU50 were females (Table 2A).

Grapefruit Orchard

A total of 101 adults of four *Anastrepha* species were captured (*A. ludens* 71.3%, *A. obliqua* 25.7%, *A. fraterculus* 2%, and *A. distincta* 1%). Hence, what follows refers only to *A. ludens* because *A. obliqua* is not a pest of citrus. Traps baited with hydrolyzed protein captured the greatest number of adults of this species (F = 29.33, df = 4, 295, $P <$ 0.001). In turn, human urine-baited traps captured statistically similar numbers of adults, and captured more flies than water-baited traps (Fig. 2). A regression analysis further corroborated that captures were independent of the concentration of human urine evaluated (F = 0.58, P = 0.46, R^2 = 0.04).

Hydrolyzed protein attracted significantly more *A. ludens* females than males (Table 2B). Furthermore, numerically more sexually mature than immature *A. ludens* females were captured by traps baited with hydrolyzed protein.

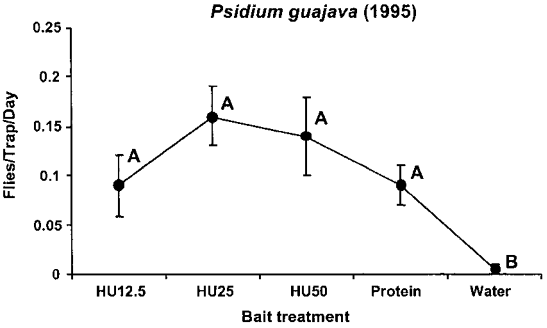

Fig. 1. Mean (±SE) number (fly/trap/day) of adult *A. fraterculus* captured by McPhail traps containing three dilutions of human urine (HU) (12.5%, 25%, and 50%), hydrolyzed protein, or water (control). Study was conducted in 1995 in Cosautlán, Veracruz, México, in an unsprayed non-commercial guava orchard. Means with different letters are significantly different (Fisher LSD test, α = 0.05).

TABLE 2. PROPORTIONS OF *ANASTREPHA* SPP. MALES AND FEMALES CAPTURED BY MCPHAIL TRAPS BAITED WITH EITHER THREE AQUEOUS DILUTIONS OF HUMAN URINE (HU) (12.5, 25, AND 50%) OR HYDROLYZED PROTEIN IN GUAVA, GRAPEFRUIT, MANGO AND SAPODILLA ORCHARDS.

Species	Treatment	N	% Females	% Males	P value[1]
A) *Psidium guajava*					
A. fraterculus	HU12.5	17	64.7	35.3	0.52
	HU25	28	92.9	7.1	0.03
	HU50	26	96.1	3.9	0.03
	Protein	17	82.3	17.7	0.10
B) *Citrus paradisi*					
A. ludens	Protein	57	91.0	9.0	0.01
C) *Mangifera indica*					
A. obliqua	Protein	13	61.5	38.5	0.90
A. serpentina	HU12.5	28	57.1	42.9	0.45
	HU25	18	61.1	38.9	0.45
	HU50	27	51.8	48.2	0.83
	Protein	124	49.2	50.8	0.83
D) *Manilkara zapota*					
A. serpentina	HU12.5	118	51.7	48.3	0.77
	HU25	97	60.8	39.1	0.08
	HU50	313	55.9	44.1	0.25
	Protein	183	54.1	45.9	0.47
A. obliqua	HU12.5	26	69.2	30.8	0.46
	HU25	24	62.5	37.5	0.23
	HU50	75	62.7	37.3	0.37
	Protein	37	70.3	29.7	0.02

[1] Comparisons of the numbers of males and females of a particular species were performed through Mann Whitney U tests.

Mango Orchard

In this orchard, 226 *Anastrepha* adults were captured by traps. *Anastrepha serpentina* was the most abundant species (87.1% of the total capture), followed by *A. obliqua* (11.1%), *A. ludens* (0.9%), and *A. alveata* (Stone) (0.9%). Results refer only to the first two species (we chose to include *A. obliqua* as this species is the common pest of mangos in the region). Traps baited with hydrolyzed protein captured more *A. obliqua* and *A. serpentina* adults than traps baited with any other bait treatment (F = 3.94, df = 4, 320, P = 0.004; F = 7.88, df = 4, 320, P < 0.001, respectively). Furthermore, there was no relationship between the number of adult *A. obliqua* and *A. serpentina* and the concentrations of human urine tested (F = 0.00, P = 1.00, R^2 = 0.00; F = 0.01, P = 0.96, R^2 = 0.00, respectively) (Fig. 3).

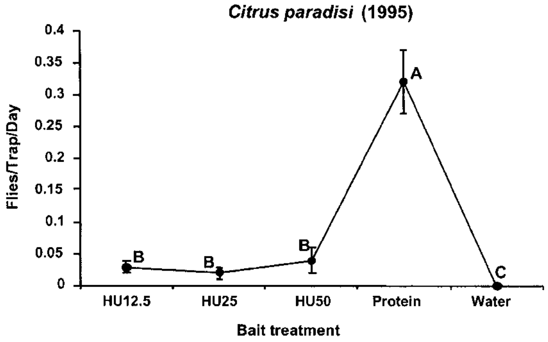

Fig. 2. Mean (±SE) number (fly/trap/day) of adult *A. ludens* captured by McPhail traps containing three dilutions of human urine (HU) (12.5%, 25%, and 50%), hydrolyzed protein, or water (control). Study was conducted in 1995 in Tuzamapan, Veracruz, México, in a mixed, unsprayed grapefruit/coffee orchard. Means with different letters are significantly different (Fisher LSD test, α = 0.05).

The three human urine-based treatments, as well as protein, attracted similar numbers of *A. serpentina* males and females (Table 2C). Interestingly, human urine-baited traps captured numerically more sexually immature than mature *A. serpentina* females (62.5% vs. 37.5%, 54.6% vs. 45.4%, and 69.2% vs. 30.8% HU12.5, HU25, and HU50, respectively) whereas traps baited with hydrolyzed protein captured a greater proportion of sexually mature (65.6%) than immature (34.4%) females.

Sapodilla Orchard

Overall, 876 *Anastrepha* adults were captured. Of these, 80.4% were *A. serpentina*, 18.3% were

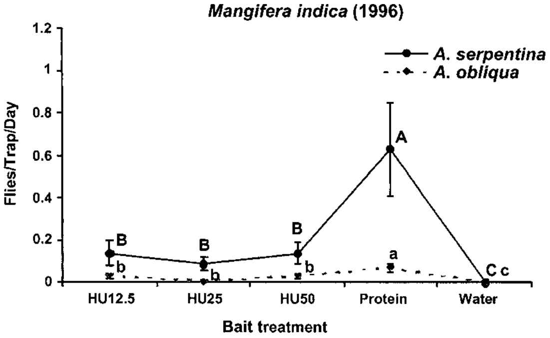

Fig. 3. Mean (±SE) number (fly/trap/day) of adult *Anastrepha serpentina* and *A. obliqua* captured by McPhail traps containing three dilutions of human urine (HU) (12.5%, 25%, and 50%), hydrolyzed protein, or water (control). Study was conducted in 1996 in Apazapan, Veracruz, México, in an unsprayed, commercial mango orchard (Manila cultivar). Means with different letters are significantly different (Fisher LSD test, α = 0.05).

A. obliqua and 1.3% were *A. ludens*. Data shown below refer only to the first two species. Traps baited with HU50 surpassed all other treatments in capturing *A. serpentina* and *A. obliqua* adults ($F = 10.19$, df = 4, 315, $P < 0.001$ and $F = 6.11$, df = 4, 315, $P < 0.001$, respectively). There were no significant differences among the other baits (i.e., hydrolyzed protein, HU25 and HU12.5), but each of these captured more flies than traps with water (Fig. 4).

Data summarized in Table 2D indicates that all bait treatments attracted similar numbers of *A. serpentina* males and females. In the case of *A. obliqua*, only protein-baited traps captured more females than males. HU25 was the only treatment attracting more sexually immature (75.5%) than mature (24.5%) *A. serpentina* females. HU12.5, HU50, and hydrolyzed protein attracted similar numbers of immature and mature *A. serpentina* females (58.1 vs. 41.9%, 56.2 vs. 43.8%, and 51.8 vs. 48.2% of sexually immature vs. mature females, respectively).

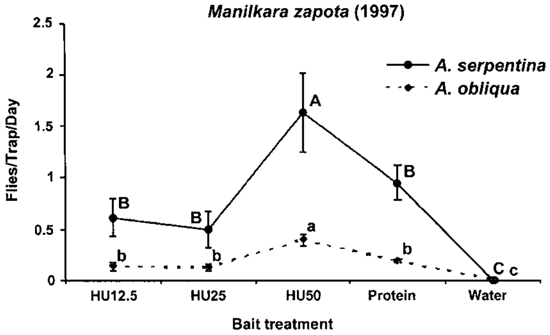

Fig. 4. Mean (±SE) number (fly/trap/day) of adult *Anastrepha serpentina* and *A. obliqua* captured by McPhail traps containing three dilutions of human urine (HU) (12.5%, 25%, and 50%), hydrolyzed protein, or water (control). Study was conducted in 1997 in Apazapan, Veracruz, México, in an unsprayed, commercial sapodilla orchard. Means with different letters are significantly different (Fisher LSD test, α = 0.05).

DISCUSSION

We found that human urine-baited traps were, in certain cases (i.e., guava and sapodilla orchards), similar or superior to protein-baited traps with respect to the total number of adult *A. fraterculus*, *A. obliqua* and *A. serpentina* captured. In other cases (i.e., mango and grapefruit orchards), traps baited with human urine captured fewer adults than traps baited with hydrolyzed protein. We also found that with one exception (sapodilla orchard), human urine concentrations tested did not vary significantly in their attractiveness to flies. The latter has important practical implications as a farmer could fill more traps with a small amount of human urine.

Our results partially confirm those obtained by Hedström (1988). That is, we also found that human urine is attractive to *Anastrepha* adults, but not to the extent this author did. Some possible explanations for the latter are: (1) differences in the nature of the commercially available baits used by Hedström (torula yeast) and us (hydrolyzed corn protein), (2) possible effect of bait aging since Hedström did not replace the urine contained in traps throughout his study, (3) the ecological characteristics of the study orchard and the fact that *A. striata* was the predominant species in Hedström's study (a species not evaluated here), (4) differences in population size, and (5), probable differences in attractiveness of the human urine used in both studies.

The favorable response to human urine shown by *A. fraterculus* adults in the guava orchard, and in some instances (e.g., sapodilla orchard) by *A. obliqua* and *A. serpentina*, suggests that individuals of these *Anastrepha* species could be responding to nitrogenous compounds such as ammonia present in human urine (Piñero et al. 2003). Given that ammonia plays an important role in attracting fruit flies (e.g., Morton & Bateman 1981; Bateman & Morton 1981; Mazor et al. 1987; Prokopy et al. 1992; Epksy et al. 1995; Heath et al. 1995), and that some ammonium salts (e.g., ammonium acetate or carbonate) and amines (e.g., methylamine, putrescine) are also known to attract adults of *A. suspensa* (e.g., Burditt et al. 1983; Thomas et al. 2001), *A. striata*, *A. obliqua* (e.g., Hedström & Jiménez 1988), and *A. ludens* (e.g., Robacker 1995; Robacker et al. 1996; 1997; Heath et al. 1997; Thomas et al. 2001), a study aimed at identifying the attractive elements of human urine to *Anastrepha* spp. is, in our opinion, warranted.

Bateman & Morton (1981) and Mazor et al. (1987) clearly demonstrated a close relationship between ammonia concentration and attraction of female *C. capitata* to protein-based baits. In our case, however, we did not find such an association as all three human urine dilutions were similarly attractive to adult *A. fraterculus*, *A. ludens*, *A. serpentina* and *A. obliqua* in the guava, grapefruit, and mango orchards. The notable exception was the sapodilla orchard, in which the less-diluted human urine (HU 50%) was significantly more attractive to *A. serpentina* and *A. obliqua* than any other human urine dilution. Robacker (1995) found results similar to current data. Liquid baits with different concentrations of ammonia and amines were similar in attractiveness over a range of over 10×, but became less attractive at very high concentrations. The latter can be explained by the fact that pH regulates emission of ammonia, amines, acids, and other ionizable compounds. At pH > 9, emission of ammonia increases greatly and can become repellent, depending on the concentration in the bait. Bateman & Morton (1981) and Mazor et al. (1987), also determined that adult females responded more strongly to baits with pH values between 7 and 8.5, a result later confirmed by Robacker et al. (1993), Robacker & Flath (1995) and Robacker & Bartelt (1997). We note, that such pH values coincided with the pH values found for the three human urine concentrations used in our study (average pH values of approximately 8 in all three cases).

In guava trees, HU25 and HU50 attracted significantly more *A. fraterculus* females than males and, in the sapodilla orchard, the same baits also attracted more females than males of both *A. serpentina* and *A. obliqua*. This agrees with previous results found for *A. serpentina* in a mango orchard (Piñero et al. 2003), and in a laboratory setting (Piñero et al. 2002). Given that Piñero et al. (2002) controlled the proportion of females and males in the experimental population, we are confident that our results in the field do not reflect a situation in which more females than males were present and as a consequence, more females were captured. Furthermore, in our study HU25 attracted a large proportion of sexually immature *A. serpentina* females when tested in the sapodilla orchard.

Interestingly, we found important differences in the response to human urine by *A. obliqua* and *A. serpentina* adults according to the type of orchard and other conditions prevalent where trapping was performed. For example, in the mango orchard (1996), traps baited with hydrolyzed protein captured the highest numbers of both *A. serpentina* and *A. obliqua* adults. In contrast, in the sapodilla orchard (1997) traps baited with HU50 captured more adults of both species than protein-baited traps. Since the two orchards are adjacent to each other, we believe that such differences might be due to variations in the type, abundance, and quality (i.e., nutritional value) of host fruits, both within and outside the orchards (e.g., presence of wild hosts). For instance, at the time the study was carried out in the mango orchard, there was very little fruit left on trees, and there was no fruit available in the contiguous sapodilla orchard. Thus, the higher capture of *A. serpentina* adults in protein-baited traps (when compared to urine-baited traps) may be explained in terms of protein hunger, since sapodilla fruit is an important protein source for *A. serpentina* adults (Jácome et al. 1999). In contrast, when trapping took place in the sapodilla orchard (in 1997), fruit was abundant and traps baited with hydrolyzed protein were not as attractive to adult *A. serpentina* as urine-baited traps. This clearly illustrates that the effectiveness of an attractant depends on the prevailing ecological conditions in a given orchard (Robacker 1992; Celedonio-Hurtado et al. 1995; Heath et al. 1997; Epsky et al. 1999), as well as on adult physiological state (Robacker 1991; Robacker et al. 1996; Rull & Prokopy 2000; Piñero et al. 2002). The latter aspects, in addition to others such as annual variations in adult fly populations in fruit orchards (Aluja et al. 1996), must be considered in the design of fruit fly monitoring systems and underscores the challenge faced by those trying to develop a replacement for the McPhail trap for use in orchards in which flies in the genus *Anastrepha* are predominant.

As a final point, we would like to address the economic benefit of using cost-free human urine. In Mexico, the value of a liter of a commercially available protein-based bait (e.g., Captor Plus®) is approximately USD $6.00 (10.9/1 US dollars/ Mexican peso). Considering that each trap is baited with 10-40 ml of bait diluted per liter of water, the cost of the bait per trap is USD $0.06-0.20. The latter multiplied by 52 (weeks in a year) raises the cost of the bait per trap per year to ca. USD $3.10-10.40 (average of $6.75). Considering that in Mexico a glass McPhail trap costs USD $4.00, and the placement of 10-20 traps in an orchard (USD $40.00-80.00), the total cost of trap placement and servicing per year would range between USD $46.75 and 86.75 or ca $510.00-945.00 Mexican pesos (not considering salaries). In the case of commercially available *Anastrepha* spp. synthetic lures (multi-component lure) and yellow plastic traps the cost in the US (not considering handling and shipping charges to Mexico) ranges between USD $3.30-4.50 and 8.50 per dispenser (lure) and trap, respectively (Great Lakes IMP, Vestaburg, MI; IPM Technologies, Portland, OR). All the latter would be unmanageable for a subsistence farmer (one that uses all his fruit for self-consumption) and hard to handle for a small-scale producer selling fruit locally. Therefore the alternative of using a cost-free bait like human urine becomes highly attractive.

In conclusion, we believe that human urine represents a low-tech alternative *Anastrepha* bait for subsistence or low income, small-scale fruit growers in rural areas in Latin America who cannot afford costly inputs such as commercial baits and traps. Human urine was capable of attracting adult flies of various species of *Anastrepha* and, at least in two orchards (guava and sapodilla), it performed as well as, or even better, than a commercially available protein bait. Even though in some instances human urine did not attract as many flies as hydrolyzed protein, qualitatively it proved to be equal or superior to this bait (i.e., it usually attracts more females than males and a large proportion of sexually immature females). A poor farmer would have access to a cost-free trap by simply reusing a two-liter plastic bottle of a soft drink or serum flask (see Salles 1996) and filling it with human urine diluted in water. There will sometimes be a trade off between cost and trap quality/efficiency, but for growers accustomed to regularly loosing between 60 to 100% of their harvest because of fruit fly damage, the benefit of a cheap, low-technology and relatively efficient trap would be very valuable. If such growers could be alerted in a timely fashion of an increasing influx of flies into their orchards or backyard gardens from surrounding native vegetation, they could protect their fruit by, for example, bagging it (Fang 1989). Further, if the bait is particularly attractive to sexually immature females, even a low capture rate could reduce fruit damage to the extent of allowing the production of a certain proportion of clean fruit (i.e., free of larvae) for local markets. One should keep in mind that the principal objective of peasant farmers in Latin America is not to produce fruit for export markets but for in-house consumption or local markets that do not demand blemish-free products.

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