Automated Identification of Flying Insects by Analysis of Wingbeat Frequencies

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ABSTRACT A microcomputer-based instrument was designed to record and analyze flight movements of individual insects flying through a light beam. In an experiment with *Aedes aegypti* (L.) and *A. triseriatus* Say, species and sex of individuals were identified correctly with an accuracy of 84% using wingbeat frequency measurements. This technology may be useful in automated monitoring of flying insects in the field.

KEY WORDS wingbeat frequency, insect flight monitor, harmonics, *Aedes aegypti*, *Aedes triseriatus*

IN ECOLOGICAL RESEARCH, acquisition and processing of abiotic data such as temperature, precipitation, and solar radiation can be readily automated due to the availability of instrumentation for measuring these variables on a continual basis. Automated acquisition of biological field data is more difficult. In particular, the lack of instrumentation for measuring insect flight activity is a major limitation to progress towards a better understanding of the impact of movement and dispersal on the population dynamics of pest species (Stinner et al. 1983).

Wingbeat frequencies of flying insects can be useful for identification and may provide the basis for automated acquisition of flight activity data. Wingbeat frequencies are inversely proportional to the sizes of insects, ranging from <10 Hz for large butterflies to >1,000 Hz for small flies (Sotavalta 1947). Insects of dissimilar size, such as a bumble bee and a mosquito, have very different wingbeat frequencies and can be differentiated simply by listening to the flight sounds they produce. Sotavalta (1947) compiled a list of wingbeat frequencies for >200 species using his musical gift of perfect pitch to record the pitches he heard when listening to the flight sounds produced by free-flying insects. Since then, insect wingbeat frequencies have been measured in the field by several methods using sound, radar, and optics (Schaefer 1976, Greenbank et al. 1980, Schaefer & Bent 1984). The instrumentation used in this study is based upon an optical tachometer developed by Unwin & Ellington (1979). This device works by using the minute changes in the intensity of ambient light reflected from an insect's beating wings to produce an amplified electrical signal at the same frequency as the insect's wingbeat.

A few studies have indicated that wingbeat frequencies are useful for differentiating among insects of similar size and morphology. Reed et al. (1942) found that the average difference in wingbeat frequencies of two races of *Drosophila pseudoobscura* Frolova was highly significant. Belton & Costello (1979) analyzed tape recordings of females of 13 western Canadian species of mosquitoes and found that wingbeat frequencies of the species overlapped but each was distinct from at least 5 others and in 3 species, from 9 others. Sawedal & Hall (1979) used wingbeat frequencies to assess the morphological similarity among 23 different species of chironomids belonging to three subfamilies. They found that there were significant differences among wingbeat frequencies even for species within the same genus.

The objectives of the study reported here were to design and build a relatively simple and inexpensive instrument for measuring wingbeat frequencies and harmonics, and to determine if these data could be used to identify the species and sex of two species of mosquitoes automatically.

Materials and Methods

Instrumentation. An insect flight monitor that automatically records fluctuations in light intensity caused by individual insects flying through a beam of light was constructed (Fig. 1). The sensor component of this instrument was based on the optical tachometer designed by Unwin & Ellington (1979) and was mounted in a lighttight box at right angles to the beam of a flashlight. Insects were kept in clear plastic cages (10 by 10 by 12 cm) that were placed at the intersection of the light beam and the field of view of the sensor. Flying insects caused part of the beam to be reflected into the sensor. This signal was modulated due to changes in the angle of the wings during each wingbeat. The output from the sensor was fed into a microcomputer (IBM PC) equipped with an analog-to-digital converter (Tecmar LabTender) under the control of

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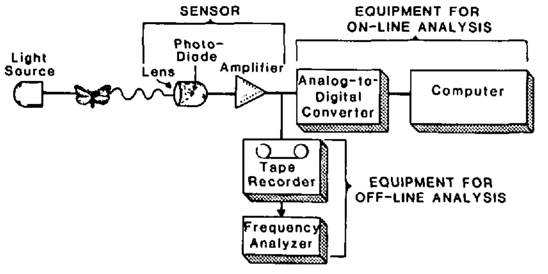

Fig. 1. Schematic diagram of equipment for recording and analyzing insect wingbeat frequencies.

a program we wrote to simulate a digital oscilloscope. When triggered by a change in light intensity, indicating that an insect was flying in front of the sensor, the computer sampled the output voltage of the sensor at a preset rate and recorded these samples as a string of numbers that could be plotted versus time (Fig. 2A). A frequency spectrum of the signal (Fig. 2B), consisting of the wingbeat frequency and several harmonics, was produced using a fast Fourier transform (Cooper 1981).

Experimental Procedure. Freshly emerged laboratory-reared *Aedes aegypti* (L.) and *A. triseriatus* Say adults were segregated by species and sex and placed in four transparent plastic cages (10 by 10 by 12 cm) with nylon mesh tops. Fifteen insects placed in each cage were kept at 22 ± 1°C throughout the experiment and were fed with cotton balls soaked in 10% honey solution placed on top of the nylon mesh. At least 12 recordings of the flight movements of individuals in each of the four groups were made on days 1, 2, 4, 6, 8, and 10 of adult life by placing cages in front of the sensor and setting the digital oscilloscope program to begin recording whenever an insect flew through the light beam. Each digital recording contained 512 samples taken at a rate of 10,000 samples per second (i.e., data were collected for 0.0512 s for each insect flying through the beam).

Analysis. Wingbeat frequencies and amplitudes of the first four harmonics were measured by performing a fast Fourier transform on each recording. These measurements plus the sex and species of the mosquitoes producing the recordings were submitted to discriminant analysis (Proc DISCRIM, SAS Institute 1982, 381–396). Discriminant functions defined by this program can be used to identify the species and sex of a mosquito given spectral characteristics (wingbeat frequency and amplitudes of harmonics) of the signal it produces. Three discriminant functions were derived using different subsets of spectral characteristics: 1) wingbeat frequencies alone, 2) wingbeat frequencies plus relative amplitudes of the first four harmonics, and 3) wingbeat frequencies plus absolute amplitudes of the first harmonic and relative amplitudes of harmonics 2, 3, and 4. Wingbeat frequency data were also submitted to analysis of variance and Duncan's multiple range test (Proc ANOVA, SAS Institute 1982, 119–137).

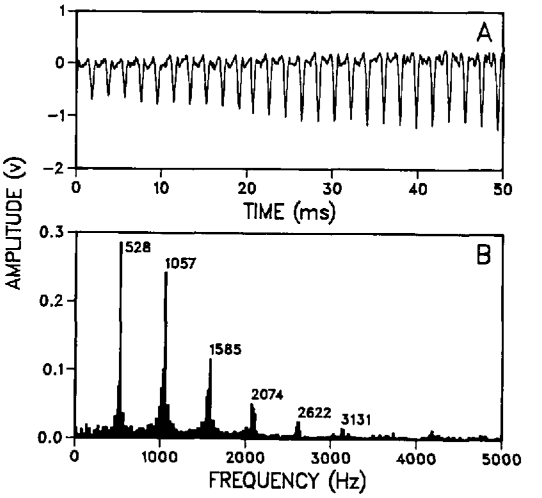

Fig. 2. Signal produced by the wingbeats of a female *A. aegypti* (A) and its Fourier transform (B).

Results

On all of the days that recordings were made, mean wingbeat frequencies had the same rank order. The group containing the largest individuals, *A. triseriatus* females, had the lowest frequency and the group containing the smallest individuals, *A. aegypti* males, had the highest frequency (Fig. 3). The wingbeat frequencies of all four groups of mosquitoes increased during the first 4 days of adult life. When the data for all ages were pooled (Fig. 4), there was significant variation in wingbeat frequency among the four groups ($F = 466$; df = 3; $P < 0.0001$). Although there was overlap in the wingbeat frequencies of individuals among the four groups, the mean for each group was significantly

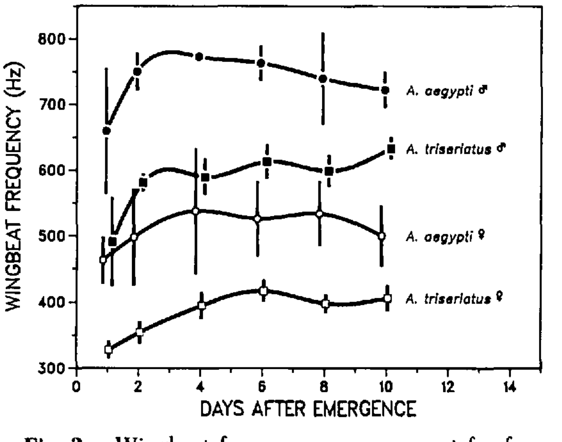

Fig. 3. Wingbeat frequency measurement for four groups of *Aedes* mosquitoes. Vertical lines represent ±1 SD.

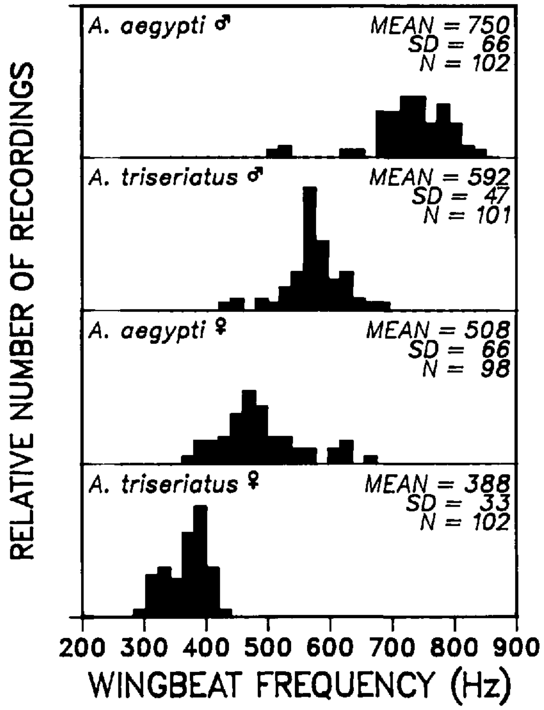

Fig. 4. Distribution of wingbeat frequencies for four groups of *Aedes* mosquitoes. The mean wingbeat frequency for each group is significantly different from all others ($\alpha = 0.05$; Duncan's multiple range test).

different from all others ($\alpha = 0.05$; Duncan's multiple range test). Spectral data from this first set of recordings were submitted to discriminant analysis.

Identification of individuals from a second group of separately reared insects made with the discriminant function based on wingbeat frequency alone was correct 84% of the time (Table 1). Accuracy of identification was not improved when discriminant functions were based on wingbeat frequency plus other spectral characteristics. The function derived using the wingbeat frequency plus relative amplitudes of the first four harmonics had an accuracy of 81% and the function derived using wingbeat frequency plus absolute amplitude of the first harmonic and relative amplitudes of harmonics 2, 3, and 4 had an accuracy of 82%.

Discussion

Our study documents the feasibility of developing an instrument that automatically detects and identifies insects flying through a light beam. The ability to differentiate among morphologically similar insects using wingbeat frequency as a character was demonstrated in the laboratory. We used a microcomputer equipped with appropriate hardware and software to monitor the output from a phototachometer. This instrument was pro-

Table 1. Results of identifications made using a discriminant function based only on wingbeat frequencies

Actual species and sex	(n)	% classified as: *A. triseriatus* ♀	*A. aegypti* ♀	*A. triseriatus* ♂	*A. aegypti* ♂
A. triseriatus ♀	(15)	100	0	0	0
A. aegypti ♀	(14)	57	43	0	0
A. triseriatus ♂	(15)	0	7	93	0
A. aegypti ♂	(13)	0	0	0	100

Overall accuracy of identification was 84%.

grammed so that light reflected from the wings of an insect flying through a light beam triggered the recording of 512 light intensity measurements within ca. $1/20$ s. Spectral analysis of recordings from males and females of two species of *Aedes* suggested that wingbeat frequencies could be used to differentiate among the four groups. In a test of this hypothesis, a discriminant function based on wingbeat frequencies alone was used to identify the species and sex of individual mosquitoes. Identifications were 100% correct for *A. triseriatus* females and *A. aegypti* males, 93% correct for *A. triseriatus* males. Only 43% of *A. aegypti* females were identified correctly. However, this is significantly greater accuracy than the expected 25% from random guesses ($\chi^2 = 14.57$; df = 3; $P < 0.005$). Measurement of wingbeat harmonics did not improve the accuracy of identification.

For field applications, it may be desirable to use a low-powered red or infrared laser as a light source, thereby allowing a considerable distance between the light source and detector. The wingbeat frequencies of insects flying through the volume of space defined by this beam could be measured and recorded along with the current time and values of selected abiotic variables.

Although the experiment reported in this article was performed at a constant temperature, it is known that wingbeat frequencies are a function of temperature for many species (Reed et al. 1942, Sotavalta 1947, Sawedal & Hall 1979, Unwin & Corbet 1984). Therefore, it would be necessary to determine the functional relationship between wingbeat frequency and temperature for species under study and measure ambient temperature at the time of each wingbeat measurement.

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References Cited

Belton, P. & R. A. Costello. 1979. Flight sounds of the females of some mosquitoes of western Canada. Entomol. Exp. Appl. 26: 105–116.

Cooper, J. W. 1981. Introduction to Pascal for scientists. Wiley, New York.

Greenbank, D. O., G. W. Schaefer & R. C. Rainey. 1980. Spruce budworm (Lepidoptera: Tortricidae) moth flight and dispersal: new understanding from canopy observations, radar, and aircraft. Mem. Entomol. Soc. Can. 110.

Reed, S. C., C. M. Williams & L. E. Chadwick. 1942. Frequency of wing-beat as a character for separating species races and geographic varieties of *Drosophila*. Genetics 27: 349–361.

SAS Institute. 1982. SAS user's guide: statistics. SAS Institute, Cary, N.C.

Sawedal, L. & R. Hall. 1979. Flight tone as a taxonomic character in Chironomidae (Diptera). Entomol. Scand. Suppl. 10: 139–143.

Schaefer, G. W. 1976. Radar observations of insect flight. *In* Insect flight. Symp. R. Entomol. Soc. London 7.

Schaefer, G. W. & G. A. Bent. 1984. An infra-red remote sensing system for the active detection and automatic determination of insect flight trajectories (IRADIT). Bull. Entomol. Res. 74: 261–278.

Sotavalta, O. 1947. The flight-tone (wing-stroke frequency) of insects (Contributions to the problem of insect flight 1.). Acta Entomol. Fenn. 4: 1–114.

Stinner, R. E., C. S. Barfield, J. L. Stimac & L. Dohse. 1983. Dispersal and movement of insect pests. Annu. Rev. Entomol. 28: 319–335.

Unwin, D. M. & S. A. Corbet. 1984. Wingbeat frequency, temperature and body size in bees and flies. Physiol. Entomol. 9: 115–121.

Unwin, D. M. & C. P. Ellington. 1979. An optical tachometer for measurement of the wing-beat frequency of free-flying insects. J. Exp. Biol. 82: 377–378.

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