

Bioacoustics of *Acanthoscelides obtectus* (Coleoptera: Chrysomelidae: Bruchinae) on *Phaseolus vulgaris* (Fabaceae)

Author(s): Anastasia Wanjiru Njoroge, Hippolyte Affognon, Christopher Mutungi, Uwe Richter, Oliver Hensel, Barukh Rohde and Richard W. Mankin

Source: Florida Entomologist, 100(1):109-115.

Published By: Florida Entomological Society

DOI: <http://dx.doi.org/10.1653/024.100.0116>

URL: <http://www.bioone.org/doi/full/10.1653/024.100.0116>

BioOne (www.bioone.org) is a nonprofit, online aggregation of core research in the biological, ecological, and environmental sciences. BioOne provides a sustainable online platform for over 170 journals and books published by nonprofit societies, associations, museums, institutions, and presses.

Your use of this PDF, the BioOne Web site, and all posted and associated content indicates your acceptance of BioOne's Terms of Use, available at www.bioone.org/page/terms_of_use.

Usage of BioOne content is strictly limited to personal, educational, and non-commercial use. Commercial inquiries or rights and permissions requests should be directed to the individual publisher as copyright holder.

Bioacoustics of *Acanthoscelides obtectus* (Coleoptera: Chrysomelidae: Bruchinae) on *Phaseolus vulgaris* (Fabaceae)

Anastasia Wanjiru Njoroge^{1,2,*}, Hippolyte Affognon³, Christopher Mutungi^{1,4}, Uwe Richter², Oliver Hensel², Barukh Rohde⁵, and Richard W. Mankin⁶

Abstract

Acanthoscelides obtectus (Say) (Coleoptera: Chrysomelidae) is an economically important pest of common bean, *Phaseolus vulgaris* L. (Fabaceae), in the tropics and subtropics. The larvae are cryptic and spend most of their development time inside the bean seeds. Their presence is almost imperceptible except for circular emergence holes created by the last instar larvae in preparation for their exit as adults. We considered a hypothesis that readily available acoustic detection devices can be used to detect larvae and adults in stored beans. Laboratory experiments were conducted in an anechoic chamber to characterize the sounds of movement and feeding and estimate whether they could be distinguished from background noise in storage environments. The larvae produced low-amplitude insect sound impulses frequently occurring in trains (bursts) of 2 or more impulses (mean = 3.6). The adults produced lower-amplitude impulses, although at a higher rate than the larvae, and there were significantly fewer impulses per burst. These features of the acoustic signals were useful for real-time detection of *A. obtectus* infestation and discrimination from background noise in stored common beans in sub-Saharan Africa. The use of such technology in the future, especially if its costs can be reduced further, may contribute to efforts to alleviate hunger and poverty in the region.

Key Words: acoustic detection; grain storage; sub-Saharan Africa; common bean

Resumen

Acanthoscelides obtectus (Say) (Coleoptera: Chrysomelidae) es una plaga económicamente importante del frijol común *Phaseolus vulgaris* L. (Fabaceae) en los trópicos y subtropicos. Las larvas son crípticas y pasan la mayor parte de su tiempo de desarrollo dentro de las semillas de frijol. Su presencia es casi imperceptible a excepción de los agujeros circulares de salida creados por las larvas del último instar cuando salen como adultos. Se consideró una hipótesis de que los medios acústicos fácilmente disponibles pueden ser utilizados para detectar larvas y adultos en frijoles almacenados. Se realizaron experimentos de laboratorio con una cámara anecoica para caracterizar sus movimientos y sonidos de alimentación y estimar si se podían distinguir del ruido de fondo en entornos de almacenamiento. Las larvas producían impulsos sonoros de insectos de baja amplitud que ocurrían frecuentemente en trenes (ráfagas) de 2 o más impulsos (promedio = 3.6). Los adultos mostraron impulsos de baja amplitud, aunque a una tasa mayor que las larvas y hubo significativamente menos impulsos por ráfaga. Estas características de las señales acústicas fueron de utilidad para la detección en tiempo real de la infección por *A. obtectus* y la discriminación del ruido de fondo en frijol común almacenado en el África subsahariana. El uso de tal tecnología en el futuro, especialmente si se pueden reducir aún más sus costos, puede contribuir a los esfuerzos para aliviar el hambre y la pobreza en la región.

Palabras Clave: detección acústica; almacenamiento de grano; África Sub-sahariana

Acanthoscelides obtectus (Say) (Coleoptera: Chrysomelidae: Bruchinae) is of Mesoamerican origin (Oliveira et al. 2013) and is a serious post-harvest and field pest species of wild and cultivated common beans (*Phaseolus vulgaris* L.; Fabaceae) in the tropics (Alvarez et al. 2005; Keals et al. 2000; Paul et al. 2010; Thakur 2012). Beans and other edible legumes are a key source of dietary protein throughout much of the world. In Kenya, common bean is the most important

food legume and second to maize (*Zea mays* L.; Poaceae) as a staple (Wagara et al. 2011). *Acanthoscelides obtectus* and *Zabrotes subfasciatus* (Boheman) (Coleoptera: Chrysomelidae) overlap in both niche and range, frequently co-occurring in bean stores. Of these 2 species, *A. obtectus* is reportedly more widely distributed in eastern and southern Africa (Masolwa & Nchimbi 1991; Ngamo & Hance 2007; Mutungi et al. 2015) and has a high predominance in bean stores of Uganda, Zim-

¹International Centre of Insect Physiology and Ecology, P.O. Box 30772-00100, Nairobi, Kenya; E-mail: anjoroge@icipe.org, annwanjiru608@gmail.com (A. W. N.), cmutungi@icipe.org (C. M.)

²University of Kassel, Faculty of Agricultural Engineering, Nordbahnhofstraße 1a, 37213 Witzenhausen, Germany; Email: judge@uni-kassel.de (U. R.), agrartechnik@uni-kassel.de (O. H.)

³International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), BP 320, Bamako, Mali; E-mail: H.Affognon@cgiar.org (H. A.)

⁴Department of Dairy and Food Science and Technology, Egerton University, P.O. Box 536-20115, Egerton, Kenya

⁵Department of Electrical and Computer Engineering, University of Florida, Gainesville, FL 32611, USA; E-mail: barukh94-work@yahoo.com (B. R.)

⁶US Department of Agriculture, Agricultural Research Service, Center for Medical, Agricultural, and Veterinary Entomology, Gainesville, FL 32608, USA; E-mail: Richard.Mankin@ars.usda.gov (R. W. M.)

*Corresponding author; E-mail: anjoroge@icipe.org, annwanjiru608@gmail.com (A. W. N.)

babwe, and the eastern highlands of Tanzania (Giga et al. 1992; Msolla & Misangu 2002).

In Africa, the economic importance of *A. obtectus* cannot be underestimated, with many small-scale farmers in Africa relying on the production and sale of beans as an important source of household income. Farmers respond to infestations by selling their commodity at harvest, when market prices are at their lowest. Infestations cause dry weight losses of between 10 and 40% in less than 6 mo, and up to 70% damage rates have been recorded in the same time period (Paul et al. 2009). Postharvest bean damage causes significant financial loss to African small-scale farmers; Mishili et al. (2011) estimated a 2.3% decrease in price per insect emergence hole in 100 beans. All the larval instars are voracious feeders and develop at the cost of legume proteins. Heavily infested beans are often reduced to empty shells.

Acanthoscelides obtectus has a short life cycle, just 3 to 4 wk, and has a high reproductive potential. It can therefore give rise to several generations per year under favorable conditions (Soares et al. 2015). The females deposit eggs in clusters inside the pods in the field or on the shelled stored bean seeds (Godrey & Long 2008). One larva generally infests each seed; however, multiple infestations sometimes occur. In such cases, late instar larvae enter the seed through the same burrow initially excavated by the first. The final instars excavate a chamber just below the seed testa for pupation to take place. The presence of a final instar or pupa can be visibly detected by a small window composed almost entirely of testa, for easy emergence of the adult. After eclosion, the adult chews a hole in the seed coat and pulls itself out of the seed, ready to mate.

As seen with many other postharvest pests of stored grain, *A. obtectus* infestation begins in the field, where adults lay eggs in dried bean pods. By late harvest, the damage to the beans can be so extensive that there is sometimes no harvest at all (Schmale et al. 2001, 2003; Velten et al. 2007). The best preventative measure is to plant seeds free from weevils, but careful visual inspection of every single seed is not practicable. Even if the planting seed is clean, the nearby bean fields must also be clean in order to ensure that the harvested crop will be free of weevils. This is a difficult task for bean farmers; therefore harvested beans often are infested. The internal mode of life of *A. obtectus* protects it from temperature and humidity variation, and enables it to be carried unnoticed during trade. Beans with undetected early-instar larvae move across geographic boundaries in import–export consignments, and pose a great phytosanitary threat in new ecological niches due to the absence of natural enemies.

Acoustic detection is a promising method for detecting insect larvae inside stored product grain kernels. The first studies on acoustic detection of pests were based on detection inside kernels by using low-frequency microphones and phonograph cartridges that transmitted signals to earphones or speakers, which were coupled with mechanical counters or strip chart recorders (Adams et al. 1953; Bailey & McCabe 1965; Street 1971; Vick et al. 1988). Next followed a series of studies on the use of high-frequency (up to 40 kHz) piezoelectric sensors combined with powerful amplifiers, used to detect a variety of stored product insect pests, including *Sitotroga cerealella* Olivier (Lepidoptera: Gelechiidae), *Callosobruchus maculatus* (F.) (Coleoptera: Chrysomelidae), *Rhyzopertha dominica* (F.) (Coleoptera: Bostrichidae), *Sitophilus oryzae* Schoenherr (Coleoptera: Curculionidae), *A. obtectus*, and *Z. subfasciatus* (Webb et al. 1985; Hagstrum 1988; Shade et al. 1990; Litzkow et al. 1990).

Adult and immature stages of stored product insect pests vary considerably in size and in the amplitudes and rates of sounds they produce (Arnett 1968; Mankin et al. 1997). However, most of the movement and feeding sounds produced by these insects are in the form of groups (trains) of short, 3 to 10 ms broadband sound impulses whose

spectra and temporal patterns can be analyzed to enable their classification as insect sounds and distinguish them from background noise (Mankin et al. 2011). Frequency and time patterns of sound impulses emitted by *Prostephanus truncatus* (Horn) (Coleoptera: Bostrichidae) and *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae) in stored maize were described by Njoroge et al. (2016).

Attempts to automate acoustic monitoring of postharvest insect pests in storage facilities began in the last century in mainly in the 1980s and 1990s (Vick et al. 1990) and continue (Eliopoulos et al. 2015; Kiobia et al. 2015). The performance of a laboratory acoustic device and an acoustic probe in the detection of infestation within grain bulks was tested in a field study in the cereal production area of western France (Leblanc et al. 2011). Mankin & Hagstrum (2011) summarized information about devices that have been developed for detection and that may be scaled up for commercial use, including a microphone system for insect detection in bag stacks in Zimbabwe.

Although Andrieu & Fleurat-Lessard (1990) studied the type of sensor that can be used to identify *A. obtectus*, little research has been done on its automated detection. In the present study, experiments were carried out to characterize the spectral and temporal patterns of sound impulses produced by *A. obtectus* in dry common beans under laboratory conditions. Sound signals of separate groups of the larvae and adults of this pest were recorded in an acoustically shielded chamber.

Materials and Methods

SAMPLE PREPARATION

The initial stock of *A. obtectus* was obtained from infested pesticide-free dry common beans bought 6 mo prior to the experiments. The common beans were a Rosecoco cultivar of *P. vulgaris*, cultivated in Kenya and procured from farmers through traders at Nyamakima Market, Nairobi. This first population of *A. obtectus* was reared and kept in a dark chamber under a 12:12 h L:D photoperiod at 27 to 28 °C and 70 ± 5% relative humidity (RH). The colony was maintained in multiple glass jars fed on 1 kg Rosecoco beans of 15% moisture content. No other food or water was provided.

For acoustic measurements, 100 *A. obtectus* adults were randomly selected from the 2nd generation and introduced into 3 sets of 1.45 L glass jars. Each set had 3 jars. Two hundred grams of cleaned, previously frozen common beans were put in each glass jar, and the adults were allowed to oviposit. Common beans with single eggs on the seed coat were selected using a microscope. These beans were placed in 100 g jars and held in an environmental chamber at 30 ± 1 °C and 70 ± 5% RH. Each day the jars were checked for eggs from which larvae had hatched and penetrated into the beans. The majority of larvae hatched on the 7th day, and the neonate larvae penetrated into the beans on the 8th day. After 15 d, the sample beans were dissected, and presence of final instars was ascertained by their morphological characteristics and by the presence of exuviae in the galleries, as described by Pfaffenberger (1985). Fifty infested bean seeds were isolated from this stock and (15th day) acoustic recordings of larval signals were collected. After 26 d, 50 bean seeds bearing adults were used for acoustic recordings of the adult signals. To minimize the effect of death-feigning behavior, recording commenced 10 min after placing the sample in the acoustic shielding chamber. For uninfested control samples, 100 g of undamaged beans were selected randomly from the initial sample lot.

RECORDING SETUP AND PROCEDURE

All insect sound recordings were carried out in a portable noise shielding chamber constructed as described in Njoroge et al. (2016) to

reduce the possibility of false positives due to vibrational and impact background noises. This chamber was kept in an isolated quiet room at ambient temperature (22–25 °C), with fluorescent lighting supplemented by ambient sunlight from nearby windows. Fifty common bean seeds infested with *A. obtectus* larvae or adults were monitored in a stainless steel container (13 cm in diameter, 4.5 cm deep) by using the method described in Njoroge et al. (2016). Signals were collected at the upper surface of the container by a 1.3-cm-diameter microphone and (20 dB) preamplifier system (Model 378B02, PCB Piezotronics, Inc., Depew, New York). The signals were amplified 10× additionally by using a 4-analog-, 8-digital-input measurement device (imc C-SERIES [CS-3008-N], imc Meßsysteme GmbH, Frankfurt, Germany). All signals were saved at a 20 kHz sampling rate with 16 bit resolution. The measurement device was configured and operated using an integrated software package (imc STUDIO, imc DataWorks, LLC, Novi, Michigan). Six 2 h recordings were collected from separate *A. obtectus* adult and larval treatment samples, and saved in a .ccv (curve configuration file) format.

SIGNAL PROCESSING

The signals recorded in .ccv format were converted to .wav (wave audio files) format by using a custom program written in MATLAB (Release 2012b, The MathWorks Inc., Newton, Massachusetts). After file conversion, the signals were band-pass filtered between 0.2 and 10 kHz, and several sections were screened from each 2 h recording with Raven Pro 1.5 Beta Version software (Cornell Lab of Ornithology, Ithaca, New York; Charif et al. 2008). It was determined from the screening that the spectral and temporal patterns of the sounds produced by both larvae and adults were very similar to those that had been observed in a previous study of sounds produced by stored product insects in maize (Njoroge et al. 2016).

A noise-free 3 min section of the filtered recording from each sample was selected at random for further characterization by using the insect signal analysis software program DAVIS (Digitize, Analyze, View, Insect Sounds) (Herrick et al. 2013; Jalinas et al. 2015). Because of the similarities between the signals in this study and those characterized in Njoroge et al. (2016), exploratory analyses were conducted to determine whether spectra of the *A. obtectus* larval or adult impulses successfully matched with any of the profiles (Broadband, HighF, MidF1, MidF2, or LowF) used previously for characterization of different movement and feeding activities of *P. truncatus* and *S. zeamais* larvae and adults. The profiles had been named based on the relative positions of energy peaks: Broadband with multiple peaks from 1 to 8 kHz, HighF with a band of high energy between 4 and 5.5 kHz, MidF1 with a peak near 3 kHz and a smaller peak between 3.5 and 5.2 kHz, MidF2 with a band at 3 kHz, and LowF with a peak between 0.5 and 0.7 kHz.

To perform each comparison, a spectrum first was constructed from a 512-point time slice centered on the peak of each impulse. The impulse then was provisionally classified as 1 of the 5 insect sound profile types based on the profile from which it had the smallest least squares difference (Mankin et al. 2011). Impulses whose spectra failed to match any profile within a preset least squares threshold were to be classified as noise impulses.

In the initial analysis process, all but a small percentage, <1 to 2% of signals per file, were found to match successfully with either the Broadband, HighF, MidF1, or MidF2 profile. No spectra were found to match with the LowF profile. It should be noted that if substantial numbers of impulses had been rejected as noise, additional profiles could have been considered for classification, but they were found not to be necessary. Consequently, we continued with temporal pattern analysis by applying the above 4-profile classification scheme in the DAVIS program, which then classified each impulse as a member of 1 of the 4 insect sound profile types.

Groups (trains) of 2 or more insect sound impulses separated by intervals <200 ms were classified as insect sound bursts. The beginning and end times, types of each burst, and the count of impulses in each burst were saved in a spreadsheet for subsequent analysis as in Njoroge et al. (2016) and Jalinas et al. (2015). The type of burst was classified as the predominant type of impulse in the burst, or in case of a tie, was classified as the type of the 1st impulse that occurred in that burst. In addition, part of the analysis was conducted by summing insect sound impulses of all profiles together, thus enabling consideration of rates of bursts and counts of insect sound impulses combined over all profiles.

STATISTICAL ANALYSES

All data were analyzed using Stata SE Data Analysis and Statistical Software Version 12 (StataCorp 2011). Analysis of variance (ANOVA) was performed to compare mean rates of bursts and number of impulses per burst among larval and adult treatments. Because these signals were collected in an anechoic chamber, there were only a few isolated impulses and no bursts detected in the controls; consequently, the control results were not analyzed in detail below.

Results

SPECTRAL PROFILES

In comparisons of spectra of each detected impulse in each recording against the 4 insect sound profiles described in the methods, adults produced impulses that matched the Broadband, HighF, MidF1, and MidF2 profiles, whereas larvae produced impulses that matched the Broadband, MidF1, and MidF2 profiles. The total counts of impulses of each type detected in recordings from larvae and adults in the study are shown in Fig. 1. The number of insect sound impulses per recording varied between 2 and 151 for the adults, and between 1 and 40 for the larvae, compared among all 3 min recordings.

LARVAL AND ADULT IMPULSE CHARACTERISTICS

Examples of sounds produced by moving and feeding adults and larvae are shown in Figs. 2 and 3. The similarities and differences among signals displayed in Fig. 2A and B were observed also in many other recordings screened in the preliminary analyses us-

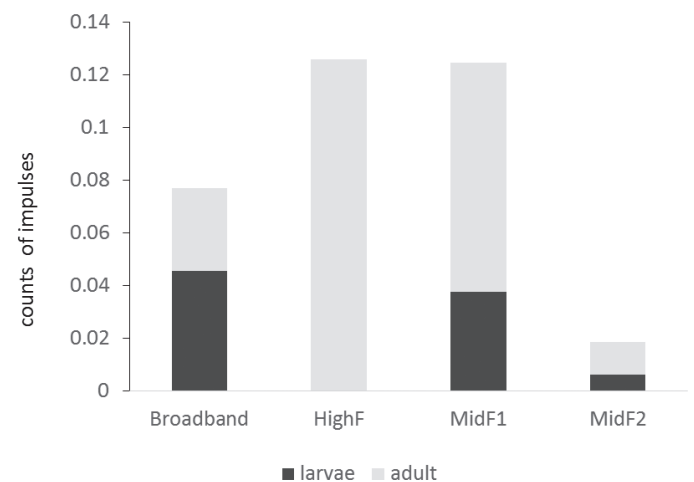


Fig. 1. Total counts of impulses of each profile type detected in recordings from the larvae and adults of *Acanthoscelides obtectus*.

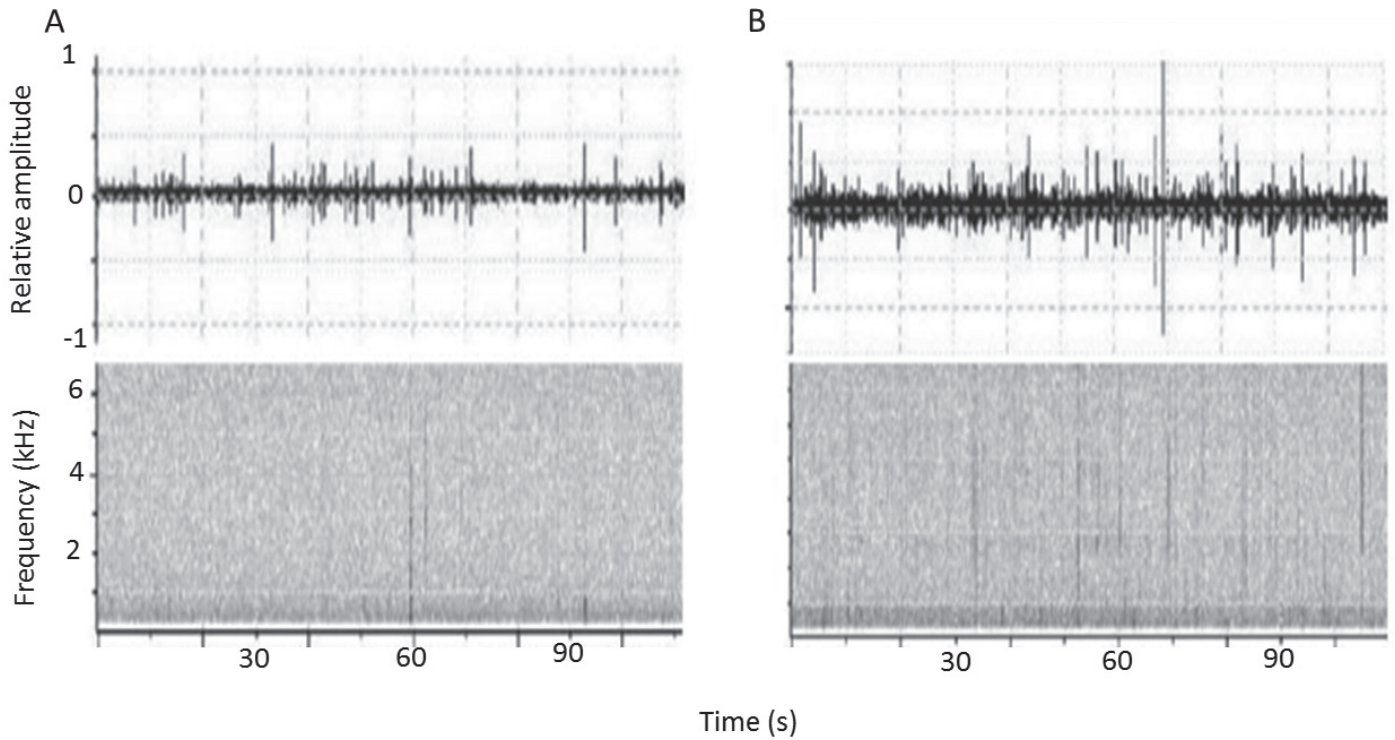


Fig. 2. Oscillograms and spectrograms of signals recorded from beans infested with A) adults and B) larvae of *Acanthoscelides obtectus*. Darker shade in spectrogram indicates greater energy at specified frequency and time.

ing Raven Pro 1.5 Beta Version software (see methods). The adult record (Fig. 2A) contained primarily HighF or MidF1 impulses (Fig. 1) with lower amplitudes than those observed with larvae. The larval record (Fig. 2B) consisted primarily of Broadband or MidF1 impulses

(Fig. 1) occurring in bursts with greater numbers of impulses than those observed with adults (Table 1). Bursts produced by larvae often consisted of multiple impulses of the same profile and similar amplitudes, separated by short intervals, e.g., as in Fig. 3B (c and d).

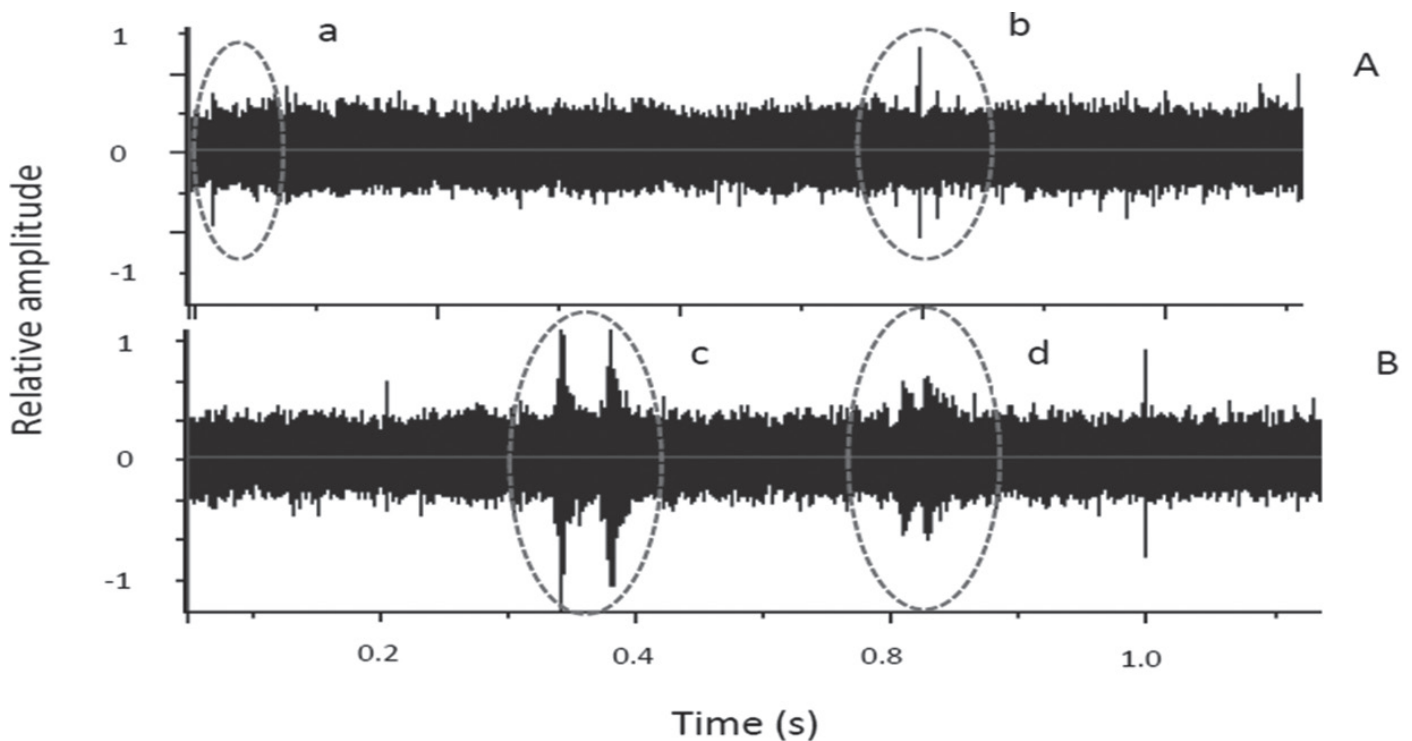


Fig. 3. Oscillograms of a 1 s period of signals recorded from beans infested with A) adults and B) larvae of *Acanthoscelides obtectus*. Signals enclosed by a dashed oval indicate bursts of the adults (a and b) and larvae (c and d).

IMPULSE-TRAIN AND BURST ANALYSES

Table 1 shows the combined burst rates and impulses per burst produced by the last instar larvae and adults of *A. obtectus*. There were significant differences ($P \leq 0.05$) between larvae and adults for the combined rates of bursts and numbers of insect sound impulses per burst. The combined counts of impulses per burst ranged from 2 to 20 with the majority being below 4.5.

MEAN RATES OF LARVAL AND ADULT IMPULSES

The mean rate of impulses from the larvae was subtracted from the mean of the adults, and the mean difference (expressed as mean \pm SEM) was 0.04 ± 0.03 impulses per s and indicated that there was no significant difference between the impulses of the larvae and the adults ($df = 11$, $t = 1.61$, $P = 0.14$). In contrast, the difference between the mean rates of bursts of the adults and the larvae was 0.09 ± 0.03 bursts per s and differed significantly between adults and larvae of *A. obtectus* ($df = 11$, $t = 2.11$, $P = 0.05$). This difference is supportive of a hypothesis that larvae, more often than adults, perform particular movement and feeding activities that result in bursts of sounds rather than individual sound impulses.

ANOVA was performed to test the effect of development stage on the rates of bursts and the counts of impulses per burst separately for Broadband, HighF, MidF1, and MidF2 burst types. The means were not significantly different between larvae and adults for any individual burst type (Table 2), although the mean combined rates of bursts and counts of impulses combined over all profile types were significantly different between larvae and adults.

Discussion

The results suggest that differences in sound-producing movement and feeding behaviors of *A. obtectus* larvae and adults can be assessed by measurements of differences in the rates of bursts and the counts of impulses per burst combined over a range of sound profile types. In addition, we observed that only adults produced signals of the HighF profile type; consequently, it was possible to distinguish larvae and adults by spectral as well as temporal pattern differences.

SPECTRAL AND TEMPORAL PATTERNS OF LARVAL AND ADULT SIGNALS

The differences in the spectral and temporal patterns of sounds produced by larvae and adults of *A. obtectus* correlate well with aspects of previous studies conducted on other postharvest insect pests that reported effects of insect size and stage on acoustic signal production (Rajendran 2005; Mankin et al. 2011; Njoroge et al. 2016). However, the results differ from numerous previous studies that found adult stages of insects to be producers of greater rates and louder sounds than the larvae. For example, Hagstrum et al. (1990) reported that *Rhyzopertha dominica* (F.) (Coleoptera: Bostrichidae) adults moving on the outside of the grain kernels produced 37-fold more sounds than larvae feeding inside the grain. In a study with *Tribolium castaneum*

Table 1. Mean (\pm SE) combined rates of bursts and combined counts of impulses per burst for adults and larvae of *Acanthoscelides obtectus*.

Stage	Rate of bursts (no. bursts per s)	No. impulses per burst
Adult	$0.023 \pm 0.007a$	$2.24 \pm 1.00a$
Larva	$0.014 \pm 0.011b$	$3.62 \pm 1.42b$

Entries in the same column followed by the same letter are not significantly different (Bonferroni, $P > 0.05$).

Table 2. Analysis of the effect of development stage on the mean rates of bursts and mean counts of impulses per burst of Broadband, HighF, MidF1, and MidF2 burst types.

Parameter	Error mean square	F	P
Rate of bursts (s^{-1})			
Broadband	0.0012	0.33	0.594
HighF	0.0207	4.79	0.123
MidF1	0.0090	3.42	0.138
MidF2	0.0008	1.04	0.365
Number of impulses per burst			
Broadband	0.053	0.03	0.865
HighF	0.809	4.00	0.116
MidF1	58.62	1.07	0.360
MidF2	2.597	0.74	0.438

(Herbst) (Coleoptera: Tenebrionidae), adults produced 80-fold more sounds than larvae (Hagstrum et al. 1991). Work by Pittendrigh et al. (1997) and Hickling et al. (2000) considering rates of sounds produced by *S. oryzae* in grain, as well as work by Shade et al. (1990) with *C. maculatus* larvae in cowpeas (*Vigna unguiculata* [L.] Walp.; Fabaceae), found that sound rates increased with instar. Studies on *S. oryzae* and *T. castaneum* adults showed that both species were more readily detectable than the smaller species *Cryptolestes ferrugineus* (Stephens) (Coleoptera: Laemophloeidae) or *Oryzaephilus surinamensis* (L.) (Coleoptera: Silvanidae), whereas *R. dominica* was intermediate to them (Hagstrum & Flinn, 1993; Mankin et al. 2011).

In such studies, the larvae were observed to produce signals, even if at lower rates or amplitudes than adults. Feeding events from the early instar through to the last instar have been detected previously in cowpea weevil, and the rate of feeding events has been found to be directly proportional to the population of larvae present per seed (Shade et al. 1990). Other research efforts have indicated that a considerable amount of larval time is spent just feeding. For instance, Vick et al. (1988) showed that, in grain samples, *R. dominica*, *S. oryzae*, and *S. cerealella* larvae spent 61 to 90% of their time feeding and thus producing sounds. Therefore, the unusual result of relatively louder acoustic signals of *A. obtectus* larvae compared with adults possibly can be explained by observations of the adult behavior. It has been shown previously, for example, that some insect females react to host deprivation by retarding egg maturation (Sadeghi & Gilbert 2000) or by delaying oviposition (Asman & Ekborn 2006). During our experiments, we transferred the insects from their mother culture to a new set of beans before acoustic measurements. The females may have perceived this transfer as disturbance or host deprivation and their adaptive response could have been to postpone egg laying.

Another factor possibly leading to relatively silent behavior of *A. obtectus* adults is that the beetle can be aphantous. Females at emergence contain sufficient energy to develop and lay eggs without feeding. The females may enter reproductive diapause and maintain quiescence for extended periods after landing on their host, before the second mating occurs and oogenesis resumes (Huignard & Biemont 1978). This lower level of activities could result in the larger mean number of impulses per burst observed with the *A. obtectus* adult signals in this work (Table 1). Quiescence observed with *A. obtectus* could also be attributed to reaction to light changes during recording in the acoustic chamber. This phenomenon is expected during acoustic recordings in laboratory conditions but should not be a problem in detecting *A. obtectus* in beans stores or natural habitats.

The feeding of adults has attracted little research attention in the past but acoustic recordings can help to better understand their activity patterns as well as the patterns of other Bruchinae. With the exception

of *Bruchus pisorum* (L.) (Coleoptera: Chrysomelidae) whose females are known to be able to reproduce only after feeding on pollen of their host plant *Pisum sativum* L. (Fabaceae), little is known about the feeding of other Bruchinae adults. Females of most Bruchinae species are able to oviposit without feeding due to accumulated body fat reserves (Godrey & Long 2008). Feeding of *A. obtectus* has almost never been observed. Like other Bruchinae, *A. obtectus* adults are also weak flyers.

Finally, the relatively low acoustic signal rates recorded from the *A. obtectus* adults compared with larvae could also be due to their tendency to feign death when being disturbed. Some insects become quiet when they are disturbed, and the time needed for them to return to normal activity after a disturbance must be taken into account when they are monitored (Arnett 1968; Mankin et al. 2011).

APPLICATION OF ACOUSTIC DETECTION METHODS IN THE CONTROL OF *A. OBTECTUS* INFESTATIONS

There is considerable need to eavesdrop on *A. obtectus* infestations because, from the moment when the 1st instar bores into the seed, it feeds, grows, and molts into successive instars and there is no visible sign of insect presence on the infested seed. The only sign of infestation is manifested when the prepupa gnaws a circular hole on the already damaged bean seed to facilitate adult emergence. Such hidden infestations can be moved inadvertently across geographic boundaries and pose phytosanitary threats in new ecological niches. Acoustic technology can serve as a means for detection at ports of entry/exit whereby larvae-infested consignments can be separated from clean consignments with accuracy and precision.

Previous research has documented the magnitude of postharvest losses and the importance of controlling various postharvest pests in sub-Saharan Africa (Njoroge et al. 2014; Affognon et al. 2015; Mutungi et al. 2015). This study of adult and larval sounds recorded on stored beans provides insights on timely detection of postharvest insect pests. Understanding the behavior of bean beetles and the characteristics of the signals they emit during feeding and locomotion can be useful for pest surveillance in storage warehouses using acoustic technology.

It is of interest that we were able to distinguish between the larvae and adults of *A. obtectus* based on both spectral and temporal patterns so as to improve the automation of the detection of these stored product insects. Ultimately, automatic signal classification efforts (Table 1) also may help distinguish the larval and adult stages when they exist together in grain stores, especially if engineers are able to adapt existing automated acoustic systems for the detection of this prevalent pest in African stores. Such results possibly also can be extrapolated to detection and management of other bean beetles like *Callosobruchus* species that cause postharvest loss of other pulses and legumes (Shade et al. 1990). There are ongoing efforts to reduce the cost and increase the automation of insect acoustic detection instruments, e.g., Kiobia et al. (2015), Njoroge et al. (2016), and Mankin et al. (2016). If such efforts achieve success, they may contribute to broad scale United Nations initiatives (Desmond-Hellmann 2016) to alleviate hunger and poverty and improve public health in sub-Saharan Africa.

Acknowledgments

We gratefully acknowledge the financial support for this research by the Deutsche Gesellschaft für Internationale Zusammenarbeit, GmbH (GIZ) under the Reduction of Postharvest Losses and Value Addition in East African Food Value Chains (RELOAD) project (project number 12.1433.7-001.00 contract number 81161243). A. W. N. was supported by a RELOAD project scholarship awarded by the International Centre for Insect Physiology and Ecology (icipe). B. B. R. was sup-

ported by a National Science Foundation Graduate Research Fellowship, DGE-1315138, and by a teaching assistantship from the University of Florida. Mention of a trademark or proprietary product is solely for the purpose of providing specific information and does not constitute a guarantee or warranty of the product by the United States Department of Agriculture and does not imply its approval to the exclusion of other products that may also be suitable. The United States Department of Agriculture is an equal opportunity employer.

References Cited

- Adams RE, Wolfe JE, Milner M, Shellenberger JA. 1953. Aural detection of grain infested internally with insects. *Science* 118: 163–164.
- Affognon H, Mutungi C, Sanginga P, Borgemeister C. 2015. Unpacking postharvest losses in sub-Saharan Africa: a meta-analysis. *World Development* 66: 49–68.
- Alvarez N, McKey D, Hossaert-McKey M, Born C, Mercier L, Benrey B. 2005. Ancient and recent evolutionary history of the bruchid beetle, *Acanthoscelides obtectus* (Say), a cosmopolitan pest of beans. *Molecular Ecology* 14: 1015–1024.
- Andrieu AJ, Fleurat-Lessard F. 1990. Insect noise in stored foodstuffs. Compact Disk, INRA Ed., Versailles, France.
- Arnett Jr RH. 1968. Measurement of adventitious sounds produced by some stored product insects. *Annals of the Entomological Society of America* 61: 1170–1172.
- Asman K, Ekborn B. 2006. Responses of ovipositing moths to host plant deprivation: life history aspects and implications for intercropping. *Agricultural and Forest Entomology* 8: 213–219.
- Bailey SW, McCabe JB. 1965. The detection of immature stages of insects within grains of wheat. *Journal of Stored Products Research* 1: 201–202.
- Charif RA, Waack AM, Strickman LM. 2008. Raven Pro 1.3 User's Manual. Cornell Laboratory of Ornithology, Ithaca, New York.
- Desmond-Hellmann S. 2016. Progress lies in precision. *Science* 353: 731.
- Eliopoulos PA, Potamitis I, Kontodimas DC, Givropoulou EG. 2015. Detection of adult beetles inside the stored wheat mass based on their acoustic emissions. *Journal of Economic Entomology* 108: 2808–2814.
- Giga DP, Ampofo JKO, Nahdy S, Negasi F, Nahimana M, Msolla SN. 1992. On-farm storage losses due to bean bruchids and farmers' control strategies. Occasional Publications Series No. 8. Centro International de Agricultura Tropical, Cali, Colombia.
- Godrey LD, Long RF. 2008. Dry Bean Weevils. UC IPM Pest Management Guidelines: Dry Beans. University of California Agriculture and Natural Resources Publication 3446, <http://ipm.ucanr.edu/PMG/r52300411.html> (last accessed 6 Dec 2016).
- Hagstrum DW. 1988. Acoustical detection and estimation of *Rhyzopertha dominica* (F.) larval populations in stored wheat. *Florida Entomologist* 71: 441–447.
- Hagstrum DW, Flinn P. 1993. Comparison of acoustical detection of several species of stored-grain beetles (Coleoptera: Curculionidae, Tenebrionidae, Bostrichidae, Cucujidae) over a range of temperatures. *Journal of Economic Entomology* 86: 1271–1278.
- Hagstrum DW, Vick KW, Webb JC. 1990. Acoustical monitoring of *Rhyzopertha dominica* (F.) (Coleoptera: Bostrichidae) populations in stored wheat. *Journal of Economic Entomology* 83: 625–628.
- Hagstrum DW, Vick KW, Flinn PW. 1991. Automated acoustical monitoring of *Tribolium castaneum* (Coleoptera: Tenebrionidae) populations in stored wheat. *Journal of Economic Entomology* 84: 1604–1608.
- Herrick NJ, Mankin RW, Dosunmu OG, Kairo MTK. 2013. Ecology and detection of the red palm weevil, *Rhynchophorus ferrugineus* (Coleoptera: Curculionidae), and related weevils for the protection of palm tree species in the United States, pp 269–283 *In Proceedings of the Palm Pest Mediterranean Conference, Nice, France, 16–18 Jan 2013*.
- Hickling R, Lee P, Velea D, Dennehy TJ, Patin AI. 2000. Acoustic system for rapidly detecting and monitoring pink bollworm in cotton bolls, pp. 984–987 *In Dugger P, Richter D [eds.], 2000 Proceedings of the Beltwide Cotton Conference, San Antonio, Texas, 4–8 Jan 2000, Volume 2. National Cotton Council, Memphis, Tennessee*.
- Huignard J, Biemont JC. 1978. Comparison of four populations of *Acanthoscelides obtectus* (Coleoptera: Bruchidae) from different Colombian ecosystems. *Oecologia* 35: 307–318.
- Jalinas J, Güerri-Agulló B, Mankin RW, Lopez-Follana R, Lopez-Llorca LV. 2015. Acoustic assessment of *Beauveria bassiana* (Hypocreales: Clavicipitaceae)

- effects on *Rhynchophorus ferrugineus* (Coleoptera: Dryophthoridae) larval activity and mortality. *Journal of Economic Entomology* 108: 444–453.
- Keals N, Hardie D, Emery R. 2000. Taxonomy and control of bruchids in pulses. In Regan K, White P, Siddique K. [eds.], *Pulse Research and Industry Development in Western Australia*, 2000. Agriculture Western Australia Bulletin No. 4405.
- Kiobia DO, Tumbo SD, Cantillo J, Rohde BB, Mallikarjunan PK, Mankin RW. 2015. Characterization of sounds in maize produced by internally feeding insects: investigations to develop inexpensive devices for detection of *Prostephanus truncatus* (Coleoptera: Bostrichidae) and *Sitophilus zeamais* (Coleoptera: Curculionidae) in small-scale storage facilities in sub-Saharan Africa. *Florida Entomologist* 98: 405–409.
- Leblanc M-P, Gaunt D, Fleurat-Lessard F. 2011. Experimental study of acoustic equipment for real-time insect detection in grain bins—assessment of their potential for infestation risk prediction during long term storage periods. *IOBC WPRS Bulletin* 69: 79–88.
- Litzkow CA, Webb JC, Vick KW. 1990. Piezoelectric apparatus and process for detection of insect infestation in an agricultural commodity, US Patent 4937555A.
- Mankin RW, Hagstrum D. 2011. Acoustic monitoring of insects, pp 263–269 In Hagstrum DW, Phillips TW, Cuperus G [eds.], *Stored Product Protection*, Publication S156-22. Kansas State University Press, Manhattan, Kansas.
- Mankin RW, Shuman D, Coffelt JA. 1997. Acoustic counting of adult insects with different rates and intensities of sound production in stored wheat. *Journal of Economic Entomology* 90: 1032–1038.
- Mankin RW, Hagstrum DW, Smith MT, Roda AL, Kairo MTK. 2011. Perspective and promise: a century of insect acoustic detection and monitoring. *American Entomologist* 57: 30–44.
- Mankin RW, Rohde B, McNeill S. 2016. Vibrational duetting mimics to trap and disrupt mating of the devastating Asian citrus psyllid insect pest. *Proceedings of Meetings on Acoustics* 25: 010006.
- Masolwa PE, Nchimbi S. 1991. Distribution patterns of bean bruchids *Zabrotes subfasciatus* (Boh.) and *Acanthoscelides obtectus* (Say) in some parts of Tanzania. *Bean Research* 6: 68–71.
- Mishili F, Temu A, Fulton J, Lowenberg-DeBoer J. 2011. Consumer preferences as drivers of the common bean trade in Tanzania: a marketing perspective. *Journal of International Food and Agribusiness Marketing* 23: 110–127.
- Msollla SN, Misangu RN, 2002. Seasonal distribution of common bean (*Phaseolus vulgaris* L.) bruchid species in selected areas in Tanzania, pp.1-5 In *Proceedings of the Bean Seed Workshop*, Arusha, Tanzania, 12–14 Jan 2002.
- Mutungu C, Affognon HD, Njoroge AW, Manono J, Baributsa D, Murdock LL. 2015. Triple-layer plastic bags protect dry common beans (*Phaseolus vulgaris*) against damage by *Acanthoscelides obtectus* (Coleoptera: Chrysomelidae) during storage. *Journal of Economic Entomology* 108: 2479–2488.
- Ngamo LST, Hance T. 2007. Diversité des ravageurs des denrées et méthodes alternatives de lutte en milieu tropical. *Tropicultura* 25: 215–220.
- Njoroge AW, Affognon HD, Mutungu CM, Manono J, Lamuka PO, Murdock LL. 2014. Triple bag hermetic storage delivers a lethal punch to *Prostephanus truncatus* (Horn) (Coleoptera: Bostrichidae) in stored maize. *Journal of Stored Products Research* 58: 12–19.
- Njoroge AW, Affognon H, Mutungu C, Rohde B, Richter U, Hensel O, Mankin RW. 2016. Frequency and time pattern differences in acoustic signals produced by *Prostephanus truncatus* (Horn) (Coleoptera: Bostrichidae) and *Sitophilus zeamais* (Motschulsky) (Coleoptera: Curculionidae) in stored maize. *Journal of Stored Products Research* 69: 31–40.
- Oliveira MRC, Corrêa AS, de Souza GA, Guedes RNC, de Oliveira LO. 2013. Mesoamerican origin and pre- and post-Columbian expansions of the ranges of *Acanthoscelides obtectus* Say, a cosmopolitan insect pest of the common bean. *PLoS One* 8: e70039.
- Paul UV, Lossini JS, Edwards PJ, Hilbeck A. 2009. Effectiveness of product from four locally grown plants for the management of *Acanthoscelides obtectus* (Say) and *Zabrotes subfasciatus* (Boheman) (both Coleoptera: Bruchidae) in stored beans under laboratory and farm conditions in northern Tanzania. *Journal of Stored Products Research* 45: 97–107.
- Paul UV, Hilbeck A, Edwards PJ. 2010. Preharvest infestation of beans (*Phaseolus vulgaris* L.) by *Acanthoscelides obtectus* Say (Coleoptera: Bruchidae) in relation to bean pod maturity and pod aperture. *International Journal of Pest Management* 56: 41–50.
- Pfaffenberger GS. 1985. Description, differentiation, and biology of the four larval instars of *Acanthoscelides obtectus* (Say) (Coleoptera: Bruchidae). *The Coleopterists' Bulletin*. 1: 239–256.
- Pittendrigh BR, Huesing JE, Shade RE, Murdock LL. 1997. Monitoring of rice weevil, *Sitophilus oryzae*, feeding behavior in maize seeds and the occurrence of supernumerary molts in low humidity conditions. *Entomologia Experimentalis et Applicata* 83: 225–231.
- Rajendran S. 2005. Detection of insect infestation in stored foods. *Advances in Food and Nutrition Research* 49: 163–232.
- Sadeghi H, Gilbert F. 2000. The effect of egg load and host deprivation on oviposition behaviour in aphidophagous hoverflies. *Ecological Entomology* 25: 101–108.
- Schmale I, Wäckers FL, Cardona C, Dorn S. 2001. Control potential of three hymenopteran parasitoid species against the bean weevil in stored beans: the effect of adult parasitoid nutrition on longevity and progeny production. *Biological Control* 21: 134–139.
- Schmale I, Wäckers FL, Cardona C, Dorn S. 2003. Combining parasitoids and plant resistance for the control of the bruchid *Acanthoscelides obtectus* in stored beans. *Journal of Stored Products Research* 39: 401–411.
- Shade RE, Furgason ES, Murdock LL. 1990. Detection of hidden insect infestations by feeding-generated ultrasonic signals. *American Entomologist* 36: 231–234.
- Soares MA, Quintela ED, Mascarin GM, Arthurs SP. 2015. Effect of temperature on the development and feeding behavior of *Acanthoscelides obtectus* (Chrysomelidae: Bruchinae) on dry bean (*Phaseolus vulgaris* L.). *Journal of Stored Products Research* 61: 90–96.
- StataCorp 2011. *Stata Data Analysis and Statistical Software: Release 12*. Stata-Corp LP, College Station, Texas.
- Street Jr WM. 1971. A method for aural monitoring of in-kernel insect activity. *Journal of the Georgia Entomological Society* 6: 72–75.
- Thakur DR. 2012. Taxonomy, distribution and pest status of Indian biotypes of *Acanthoscelides obtectus* (Coleoptera: Chrysomelidae: Bruchinae)—a new record. *Pakistan Journal of Zoology* 44: 189–195.
- Velten G, Rott AS, Cardona C, Dorn S. 2007. Effects of a plant resistance protein on parasitism of the common bean bruchid *Acanthoscelides obtectus* (Coleoptera: Bruchidae) by its natural enemy *Dinarmus basalis* (Hymenoptera: Pteromalidae). *Biological Control* 43: 78–84.
- Vick KW, Webb JC, Weaver BA, Litzkow C. 1988. Sound detection of stored-product insects that feed inside kernels of grain. *Journal of Economic Entomology* 81: 1489–1493.
- Vick KW, Webb JC, Litzkow CA. 1990. Insect detection using a pitfall probe trap having vibration detection. US Patent 5005416 A.
- Wagara IN, Mwang'ombe AW, Kimenju JW, Buruchara RA, Kimani PM. 2011. Reaction of selected common bean genotypes to physiological races of *Phaeoisariopsis griseola* occurring in Kenya. *African Crop Science Journal* 19: 343–355.
- Webb JC, Calkins CO, Vick KW. 1985. New system hears insects chewing. *Agricultural Research* 33: 13–15.