http://ansinet.com/itj

ITJ

ISSN 1812-5638

INFORMATION TECHNOLOGY JOURNAL

ANSInet
Asian Network for Scientific Information
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

Information Technology Journal 6 (8): 1264-1269, 2007
ISSN 1812-5638

Automated Motion Tracking of Insects Using Invariant Moments in Image Sequence

[1,3]N. Ravi Kumar, [2]T.N. Janakiraman, [2]Hemalatha Thiagarajan and [3]K. Subaharan
[1]Department of Computer Applications, National Institute of Technology, Trichy-620015, India
[2]Department of Mathematics, National Institute of Technology, Trichy-620015, India
[3]Central Plantation Crops Research Institute, Kasaragod-671124, India

Abstract: In this study, we present a new approach to track multiple insects especially the *Opisina arenosella* larva which is elastomeric in nature. This study demonstrates the use of image processing techniques for segmenting insects, a detailed procedure for separating the head and tail sections from the larva trunk to minimize the errors due to deformation, invariant moment features to establish correspondence across frames to arrange insect parts in order. This study automates the host searching behavior of parasitoid *Goniozus nephantidis* which are used for biological control programs.

Key words: Motion tracking, larva, parasitoid, invariant moments, correspondence

INTRODUCTION

Insect behavioral studies are time consuming. The surface prospected by a parasitoid looking for host (larva) has always been considered to be an important feature of host and parasitoid associations. For most insect parasitoids, the last step of host-finding behavior is predominantly performed by walking (Wajnberg, 1994). The study of the surface searched by female parasitoids looking for the host is aimed at understanding their strategy to prospect this particular environment. The effective host searching behavior of parasitoids makes them an ideal candidate for bio control program. Various attempts have been made to understand the walking path of parasitoids. Using a stochastic model, simulating the walking path of isolated parasitoid females during their searching behavior (Mark, 2005) have shown efficient ways of discovering new hosts. Thomas *et al.* (1999) studied parasitoid host searching ability using empirical evidence. In all these, collecting observations from behavioral experiments by humans is laborious and error-prone. The automation of the experiment eliminates the errors in timing and reduces the experimental bias.

A number of automated systems have been developed for quantifying specific behavioral parameters in genetically-tractable organisms. Spruijt *et al.* (1998) explained the use of statistical classifiers and neural networks in animal behavior recognition. Some systems have been designed to observe multiple animals at low magnification and track the position of each animal over time (Dhawan *et al.*, 1999) and (de Bono and Bargmann, 1998). Subsequently, pattern recognition techniques are applied to allow an objective description of functional behaviors based on the features extracted from the image sequences and manually registered behavioral states. Lucas *et al.* (2002) developed computerized video tracking software which gives general purpose solutions. However behavioral studies include larvae also and the body or trunk of larva is soft and flexible. Measuring the workspace of larva is an elastomeric process. Hussaini *et al.* (2003) studied olfactory response of *Drosophila* larva using image processing techniques. Jason *et al.* (2003) studied dynamic analysis of larval locomotion in *Drosophila* chordotonal organ mutants using Dynamic Image Analysis System (DIMS). Free living warm *Caenorhabditis elegans* phenotypes' features were extracted and classified using computer vision and digital image analysis techniques. The characteristics of the worm's movement, posture and texture information were extracted from a 5 min image sequence (Geng *et al.*, 2004). However each system is capable of measuring some specific behavioral parameters, but there is no automated system that is designed to analyze the behavior of the host (larva) as well as parasitoid simultaneously.

The fundamental problem in tracking multiple objects from frame to frame is correspondence between frames. A general approach to tracking is to establish correspondences between points, or sets of points, in successive frames. Structural constraints on the relative positions of object points are imposed by the shape of the objects being tracked. Typically this problem is solved by extracting various features of an object and matching them across the frames. Hence feature extraction is one of the major processes.

Corresponding Author: N. Ravi Kumar, Central Plantation Crops Research Institute, Kasaragod-671124, India

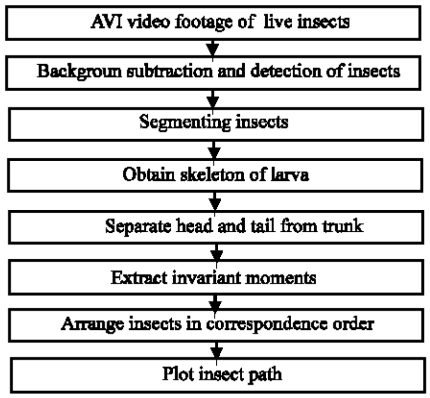

Fig. 1: Block diagram of multiple insect behavioral analysis

In this study, we present new automatic methods for feature extraction based on morphological operations and invariant moments. From image geometry, the joined coconut black head caterpillar *Opisina arenosella* larva and parasitoid *Goniozus nephantidis* were separated and larva body parts head, trunk and tail and parasitoid were classified using invariant moments. This approach can be divided into several well-defined stages, as shown in Fig. 1.

MATERIALS AND METHODS

Insect cultures and video footage: The cultures of *Opisina arenosella* and *Goniozus nephantidis* for this experiment, were maintained in the Entomology laboratory at Central Plantation Crops Research Institute (CPCRI), Kasaragod, Kerala, India. *G. nephantidis* culture maintained on *O. arenosella* was used for the study. This study was carried out in January 2007. The parasitoids were cultured by placing the adult female wasp and larvae of *O. arenosella* in a vial of 2.5×10 cm plugged with cotton. Drops of 50% honey (in water) placed on the wax coated paper served as food. All the cultures were maintained at 25°C and 65-70% RH with a constant dim light illumination. The size of the parasitoid was 3 mm and the VII instar larva (approximately 15-25 mm). The movements of the host and its parasitoid were recorded at 25°C between 9:00-16:00 h, by the JVC TK C 1380 Charge-Coupled Device (CCD) video camera, which was connected to HP Pavilion Media Centre CPU, Pentium D processor 2.80 GHz, with Windows XP Professional (SP 2) and 1024 megabytes of memory. The camera was fixed vertically above the Petri dish. The data was captured in 576×720 resolution, 30 frames per second and stored in windows media video format. Each experiment (batch) was recorded for 5 min. All video footages were converted into AVI format using Adobe Premier Pro 2.0 version, 30 frames per second, truecolor image type, square pixels (1.0) and 'indeo5' video compression rate. Backlight was used to create maximum contrast between the insect and the substrate (Lucas *et al*., 2002).

Segmentation of larva and parasitoid: The segmentation process started with background subtraction. The static background (Petri dish) was subtracted from every frame of the video footage. The RGB image was converted into grayscale image. The sequential algorithm for component labeling was used to remove unwanted small objects fewer than 80 pixels (Jain *et al*., 1995), producing a new binary image. Each labeled region was an insect (Fig. 2).

In host searching behavioral experiments, the parasitoid tries to reach the host and in the process touches the host (larva). The segmentation algorithms consider the joined insects as a single region in the binary image, implying a single insect. Separating touching objects in an image is a difficult image processing operations. We applied a simple geometrical approach to separate joined insects based on the image geometry.

Two boundary points were identified on the bounding box of the joined insect. These two points were used to divided the boundary into two parts X and Y. The sets X and Y contained sampled points of respective boundary. For each point x_i in X, we search every point y_j in Y to find the one closest to x_i. We calculated the distance W_{ij} between x_i and y_j as well as the outer angle θ_i (the angle between each pair of segments x_i-x_{i-2} and x_i-x_{i+2}). First we check if $\theta_i < 90°$, if the answer is yes, then x_i is considered a possible starting point of the second insect body touching the first. If the answer is no for all points, we compare W_{ij} to the previous distance $W_{i-1,j-1}$ between x_{i-1} and y_{j-1}. If $W_{ij} > W_{i-1,j-1}+\alpha$, for a pre-specified α, then x_i will still be considered a possible starting point. The search continues until the first possible starting point of the insect body touching the first insect is found. We repeat the same process for the set Y. We have found that at most one set will have a possible starting point with θ smaller than 90°. Assuming x_m is found to be the real starting point and y_n is its closest sampled point in the other set, we can keep locating division points which are W_{mn} (the distance between x_m and y_n) pixels away from the next points y_k (k>n) until the interior boundary is reached again. These division points are connected to form a dividing line and then pixels of the joined object on the dividing line are removed to separate the insects.

Larva head and tail separation: As the *Opisina* larva is highly deformable in nature, many conventional image matching and tracking algorithms do not apply to this

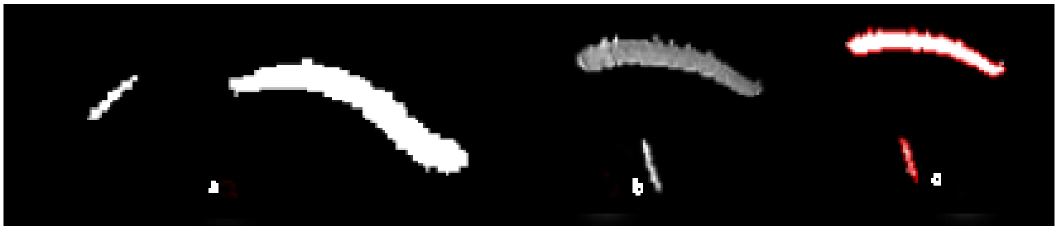

Fig. 2: Segmented insects, (a) Binary image of larva and parasitoid (small insect), (b) Gray scale image of identified insects and (c) Segmented insects' boundary in red color

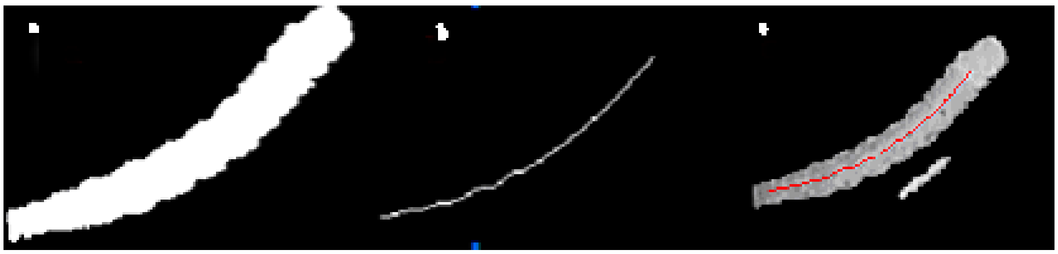

Fig. 3: (a) Binary image of larva, (b) Skeleton of binary image and (c) Skeleton was superimposed on the larva

problem. We have separated the head and tail sections from the larva trunk to minimize the errors due to deformation, so that detailed motion information of the larva could be extracted. In this process the larva alone was taken separately from the image. The larval skeleton was obtained by applying a skeletonizing algorithm described by Zhang and Suen (1984). Sometimes the obtained skeleton had Spur pixels due to noise in the larval boundary. This leads to wrong identification of the head, trunk and tail portions of the larva. Spur pixels on the skeleton were eliminated by thinning, so that we get a clean skeleton (Fig. 3b). The two end points of the skeleton are potential head and tail locations. To find these endpoints, we fixed an approximate mid point of the skeleton based on the bounding box. A 3×3 window was moved along the skeleton in both directions to reach the end points. We stored all skeleton pixel locations in an array. We identified two points on the skeleton that were 12 pixels interior from each end point, to isolate the head and tail sections from the rest of the body. To do this, the normal direction to the tangent of the skeleton was computed at each of the identified points. The line traversed in the normal direction at the chosen two points, is taken as the separation line between the head/tail and the rest of the body. The end sections are separated from the trunk by deleting binary pixels along these separation lines.

Basic theory of invariant moment functions: The image analysis system involves automatic recognition of an object in a scene regardless of its position, size and orientation. A number of techniques have been developed to derive features from an image, which are invariant under translation, scale change and rotation (Hu, 1962; Hsu *et al*., 1982; Bamieh and Figueiredo, 1986). In particular, the invariant properties of regular moment functions have attracted many users to use them as pattern features in object recognition. Hu first published his classic paper on pattern recognition in 1962, by deriving a set of regular moment invariants based on combinations of regular moments using algebraic invariants. He derived a set of invariant moments which have the desirable property of being invariant under image translation, scaling and rotation. If an image can be thought of as a two-dimensional discrete density function I (x,y), then the moment of order (p + q) is defined as:

$$M_{pq} = \sum_x \sum_y x^p y^q I(x,y) \text{ for } p, q = 0, 1, 2 \qquad (1)$$

Here, we assumed for simplicity the region of interest to be defined as $x_1 \le x \le x_2$ and $y_1 \le y \le y_2$ and with p, $q \in N_0$ as order indices, (x, y) as Cartesian coordinates, I is a non-negative intensity function with bounded and compact support so that summation within the available image plane is sufficient to gather all the signal information. To make these moments invariant to translation, one can define central moments of image I as (2)

$$\mu_{pq} = \sum_{x1}^{x2} \sum_{y1}^{y2} (x-\bar{x})^p (y-\bar{y})^q I(x,y) \qquad (2)$$

Where, $\bar{x}$ and $\bar{y}$ are the coordinates of the image centroid given by $\bar{x} = m_{10}/m_{00}$, $\bar{y} = m_{01}/m_{00}$ These moments are made invariant to scale change as proposed by Hu (1962) by normalizing the image intensity to unity (3):

$$\eta_{p,q} = \frac{\mu_{p,q}}{(\mu_{00})^{(p+q+2)/2}} \text{ for } p + q = 2, 3 \quad (3)$$

Rotation invariance can be achieved by combining the moments based on the theory of algebraic invariance (ϕ_1-ϕ_7), as shown by Hu (1962).

$$\phi_1 = \eta_{20} + \eta_{02}$$
$$\phi_2 = (\eta_{20} - \eta_{02})^2 + 4\eta_{11}^2$$
$$\phi_3 = (\eta_{30} - 3\eta_{12})^2 + (3\eta_{21} - \eta_{03})^2$$
$$\phi_4 = (\eta_{30} + \eta_{12})^2 + (\eta_{21} + \eta_{03})^2$$
$$\phi_5 = (\eta_{30} - 3\eta_{12}) (\eta_{30} + \eta_{12}) [(\eta_{30} + \eta_{12})^2 - 3 (\eta_{21} + \eta_{03})^2] + (3\eta_{21} - \eta_{03}) (\eta_{21} + \eta_{03}) [3 (\eta_{30} + \eta_{12})^2 - (\eta_{21} + \eta_{03})^2]$$
$$\phi_6 = (\eta_{20} - \eta_{02}) [(\eta_{30} + \eta_{12})^2 - (\eta_{21} + \eta_{03})^2] + 4\eta_{11} (\eta_{30} + \eta_{12}) (\eta_{21} + \eta_{03})$$
$$\phi_7 = (3\eta_{21} - \eta_{03}) (\eta_{30} + \eta_{12}) [(\eta_{30} + \eta_{12})^2 - 3 (\eta_{21} + \eta_{03})^2] - (\eta_{30} - 3\eta_{12}) (\eta_{21} + \eta_{03}) [3 (\eta_{30} + \eta_{12})^2 - (\eta_{21} + \eta_{03})^2]$$

The first one, ϕ_1, is roughly proportional to the moment of inertia around the image's centroid, if the pixels' intensities are interpreted as physical density. The last one, ϕ_7, is skew invariant, which enables it to distinguish mirror images of otherwise identical images.

Feature extraction: Features play a crucial role in classifying objects in a given image. This helps to establish correspondence between objects in successive frames of a video footage. Here we have three spatial and one temporal observation to recognize the insects. One was that the larva's head was darker than the tail. Second clue was, the parasitoid was darker than the larva's head. Further, we have extracted seven invariant moments (ϕ_1-ϕ_7) of larva and parasitoid.

We also computed larva area, length, width at center and head/tail, breadth, eccentricity and lengths of major/minor axes of best-fit ellipse, height and width of minimum enclosing rectangle (MER), ratio of MER width and height, ratio of the larva area to MER area, angle change rate, head/tail/center brightness, local head/tail/center movement relative to centroid and head-centroid-tail angle. The area, angle change rate and movement features were calculated for the head, tail and trunk.

RESULTS AND DISCUSSION

Experiments have been performed on real video footages acquired with a fixed video camera. We have analyzed a total of 18,000 frames (Five experiments). Larva and parasitoid were segmented from the image in the video footage. When the insects were joined together, the above discussed geometrical procedure separated them apart successfully (Fig. 4).

While skeletonizing the larva, the skeleton had branches (Fig. 5a). These branches were produced because of the noise in the image. We dilated the larva image to make the boundary uniform and then applied the skeletonzing algorithm to produce a clean skeleton. When the larva walks, the head moves more frequently than the tail (having to do with foraging behavior). To get the detailed analysis of the larva, it was cut into three parts head, tail and trunk using the procedure discussed earlier (Fig. 5b).

After separating head and tail from the larva's trunk, to track these parts the correspondence was established between these objects in successive frames using invariant moments (Table 1).

The computed invariant moments ϕ_1, ϕ_3 and ϕ_4 values were used for matching across the frames. We achieved better invariant moments when the image dimension was square and the background was black. In all the frames the tail had maximum ϕ_1, followed by the head. To classify between trunk and parasitoid, we used ϕ_1 and the quantity ϕ_3/ϕ_4, which is contrast invariant (Hupkens and Clippeleir, 1995). After establishing correspondence, the centroids of head, tail, trunk and parasitoid were arranged in order and plotted, to trace the path (Fig. 6).

Larva body length and area were calculated by counting the number of pixels on the skeleton and binary image respectively. We also measured the motion of the head with regard to the centroid across the frames. The tail movement was also measured. These measurements calculated how much the individual body parts move relative to the rest of the body. The rates of change of the angle were calculated for head, tail and trunk regions of larva as well as parasitoid. Some times the larva moves backwards when the parasitoid comes right opposite the larva. This reversal in motion was an interesting characteristic. To recognize this, we stored previous 15 centroids of the tail. A reversal was detected when the new centroid was closer to any of the 14 previous centroid locations than to the most recent past. A reversal in motion is a manifestation of the threat to the larva by the parasitoid.

Table 1: Invariant moments for three frames

Frames	ϕ_1	ϕ_2	ϕ_3	ϕ_4	ϕ_5	ϕ_6	ϕ_7	ϕ_3/ϕ_4
Frame 2								
Trunk	5.0469	10.2193	18.4874	19.9884	39.7638	26.5897	39.5600	0.924906
Parasitoid	5.4919	11.3643	24.5025	23.2909	47.3716	29.2214	47.7763	1.052020
Head	5.8814	15.1376	19.7155	22.1611	43.1502	30.0069	44.3794	0.889644
Tail	6.4469	14.3351	22.7119	24.1694	47.6902	31.8420	48.5642	0.939696
Frame 3								
Trunk	5.0351	10.1876	18.3806	20.0891	40.4362	28.7968	39.5063	0.914954
Parasitoid	5.3429	10.9690	27.0261	23.6752	49.1678	29.1859	50.3905	1.141536
Head	5.9335	15.6360	19.9446	23.5440	45.4036	32.6035	47.5597	0.847120
Tail	6.4996	14.5699	24.1495	24.5944	49.9681	31.9028	49.1566	0.981911
Frame 4								
Parasitoid	5.1712	10.5489	21.6722	21.2061	42.6452	26.4807	47.4348	1.021980
Trunk	5.2254	10.6153	19.4933	21.5762	42.8880	27.5066	42.3824	0.903463
Head	5.9266	15.9552	20.1427	23.4381	45.3902	32.6556	46.4843	0.859400
Tail	6.2953	14.0020	21.3807	23.8407	46.4674	31.5480	48.2835	0.896815

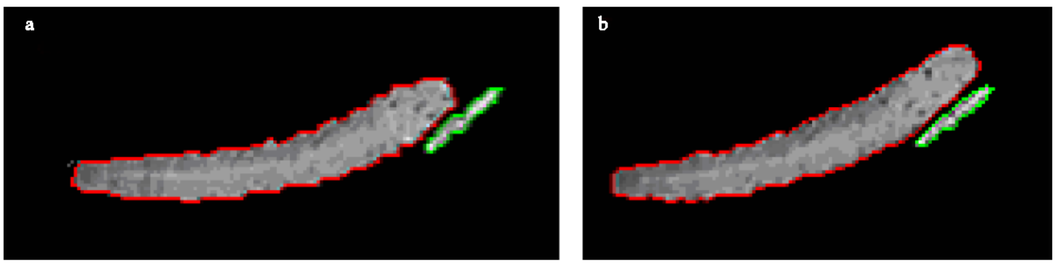

Fig. 4: (a) Joined larva and parasitoid were separated and (b) red boundary identifies larva and green boundary represents parasitoid

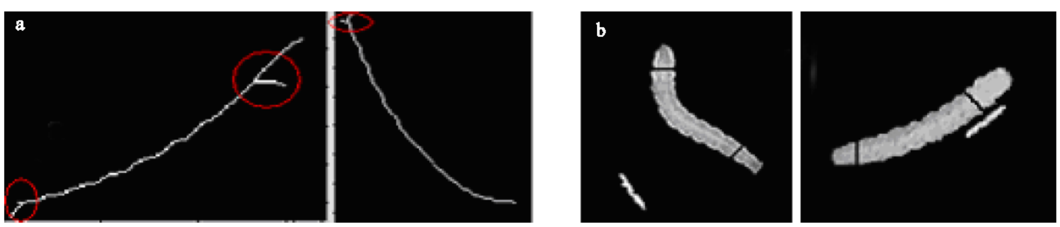

Fig. 5: (a) Branches in the skeleton caused by noise and (b) Larva was separated into head, tail and trunk

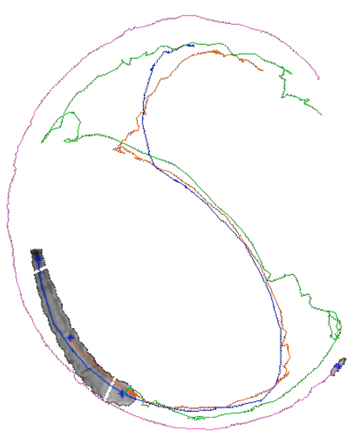

Fig. 6: Traced path: Blue color represents tail path, Green color represents head path, Red color represents trunk path and Magenta color represents parasitoid path

CONCLUSION

We have presented a new approach to track multiple insects, especially the *Opisina arenosella* larva, which is elastomeric in nature. This study demonstrates the use of image processing techniques for segmenting insects, geometrical method for separating joined insects. A detailed procedure for separating the head and tail sections from the larva trunk to minimize the errors due to deformation is presented. The usage of invariant moment features to determine the correspondence across frames was established. This study automates the host searching behavior of parasitoid *G. nephantidis* which are used for biological control programs. The clues would aid in evaluating and/or designing pest management strategies.

The goal of developing a new approach for measuring the larva and parasitoid movements is to understand the dynamism in an image sequence. However, the quantitative analysis of the parasitoid track has wider applications in insect behavioral studies. The researcher proposes to use this technique, in future behavioral studies of parasitoids in different environments.

ACKNOWLEDGMENTS

The first and fourth authors thank the Director, Central Plantation Crops Research Institute (CPCRI), Kasargod, under Indian Council of Agricultural Research (ICAR), for supporting the research work.

REFERENCES

Bamieh, R. and De Figueiredo, 1986. A general moments/attributed-graph method for the three dimensional object recognition from a single image. IEEE J. Robotics Automation, 2: 31-41.

de Bono, M. and C.I. Bargmann, 1998. Natural variation in a neuropeptide Y receptor homolog modifies social behavior and food response in *C. elegans*. Cell, 94 (5): 679-689.

Dhawan, R., D.B. Dusenbery and P.L. Williams, 1999. Comparison of lethality, reproduction and behavior as toxicological endpoints in the nematode *Caenorhabditis elegans*. J. Toxicol. Environ. Health, 58 (7): 451-462.

Geng, W., P. Cosman, C.C. Berry, Z. Feng and W.R. Schafer, 2004. Automatic tracking, feature extraction and classification of *C. elegans* phenotypes. IEEE Trans. Biomed. Eng., 51 (10): 1811-1820.

Hsu, Y.N., H.H. Arsenault and G. April, 1982. Rotational invariant digital pattern recognition using circular harmonic expansion. Applied Optics, 21 (22): 4012-4015.

Hu, M.K., 1962. Visual pattern recognition by moment invariants. IRE Trans. Inform. Theory, IT-8: 179-187.

Hupkens, Th. M. and J. de Clippeleir, 1995. Noise and intensity invariant moments. Pattern Recog. Lett., 16 (4): 371-376.

Hussaini, A., K. Parathasarathy, A. Vibhanshu and O. Siddiqi, 2003. A rapid method for measuring olfactory response of *Drosophila* larvae. Curr. Sci., 85 (10): 857-859.

Jain, R., K. Rangachar and B. Schunck, 1995. Machine Vision. New York, McGraw-Hill.

Jason, C.C., M.M. Miller, S. Wing, D.R. Soll and D.F. Eberl, 2003. Dynamic analysis of larval locomotion in *Drosophila* chordotonal organ mutants. doi/10.1073/pnas. 2535546100, PNAS, 100 (26): 16053-16058.

Lucas, P.J.J., N.A.J. Spink and R.A.J. Tegelenbosch, 2002. Computerised video tracking, movement analysis and behaviour recognition in insects. Comput. Electron. Agric., 35 (2): 201-227.

Mark, A.W., 2005. Odor-modulated navigation in insects and artificial systems. Chemical Senses 30 (Supplement 1): i287-i288.

Spruijt, B.M., M.O.S. Buma, M.O.S. Van Lochem and J.B.I. Rousseau, 1998. Automatic behavior recognition: What do we want to recognize and how do we measure it?. Proceedings of Measuring Behavior. 2nd International Conference on Methods and Techniques in Behavioral Research, Groningen.

Thomas, S.H. and P. Gienapp, 1999. Exploitation of the host's chemical communication in a parasitoid searching for concealed host larvae. Ethology, 105 (3): 223-232.

Wajnberg, E., 1994. Intra-Population Genetic Variation in *Trichogramma*. In: Biological Control with Egg Parasitoids, Wajnberg, E. and S.A. Hassan (Eds.). CABI, pp: 245-272.

Zhang, T.Y. and C.Y. Suen, 1984. A fast parallel algorithm for thinning digital patterns. Commun. ACM, 27 (3): 236-239.