application. The same treatment also showed a strong effect on the movement of the labium hypoparynx maxillary complex (15.3/min), without any effect on antennal movement. Treating the top of the maxillary palps with clerodin had a weak effect on the complex structure, but the rate of movement became higher 10 min after application (10.9/min). Treatment of antennae with clerodin showed a very weak effect on the rate of mandibular movement and other mouth parts, and had no effect on the antennae themselves.

The movement of both mandibles was characterized as horizontal, while the maxillary palps always responded with irregular movements. Antennal movements were characteristically tremors.

The results obtained from the present investigation are in full agreement with the antifeeding and behavioural studies of the previous topical application: the most sensitive action site of chlordimeform is the hypopharynx and for clerodin it is the gustatory sensilla basiconica located on the top of both maxillary palps (ANTONIOUS and SAITO, 1983).

The physiological explanation of the observed movement in the various responding mouth parts and antennae is that the chemoreceptors of the treated targets showed different sensitivity to the applied materials. The evoked movements were integrated from the nerves of the affected chemoreceptors to the CNS which in return integrated this effect to the other peripheral nerves arising from it and innervated other organs.

The above argument supports the data reported by SHIMIZU et al. (1981 b) who reported that the antifeeding activity of chlordimeform results from continuous repetitive bursts of mandibular movements (CBMM) in *Mamestra brassicae*. They reported that this CBMM was not evoked when an aqueous solution of chlordimeform was placed topically on the antennae of prepupal insects.

REFERENCES

ANTONIOUS, A. A. and T. SAITO (1983) *Appl. Ent. Zool.* **18**: 22–31.

SCHOONHOVEN, L. M. (1968) *Ann. Rev. Entomol.* **13**: 115–136.

SHIMIZU, T., K. MATSUZAWA and J. FUKAMI (1981 a) *Appl. Ent. Zool.* **16**: 167–169.

SHIMIZU, T., K. MATSUZAWA and J. FUKAMI (1981 b) *Int. Pest Control* **4**: 102–109.

SHIMIZU, T., K. MATSUZAWA, S. YAGI and R. J. ROBBINS (1980) *Appl. Ent. Zool.* **15**: 352–355.

WAGO, H. and D. YAMAMOTO (1978) *Appl. Ent. Zool.* **13**: 84–90.

Sequence of Predatory Behavior of the Ladybeetle, *Coccinella septempunctata* L. (Coleoptera: Coccinellidae) on the Green Peach Aphid, *Myzus persicae* SULZER (Homoptera: Aphididae)[1]

Kiyoshi NAKAMUTA

Laboratory of Applied Entomology and Nematology, Faculty of Agriculture, Nagoya University, Nagoya 464, Japan

(Received May 4, 1983)

As ladybeetles are effective predators of aphids and scales (HÄMÄLÄINEN et al., 1975), many studies have been conducted on their feeding behavior (cf. HODEK, 1973). However, little information is available on the sequence analysis and the involvement of stimuli in the predatory behavior of coccinellids. Since each event in a behavioral sequence may be governed by different kinds of stimuli (CURIO, 1976), a description of predatory behavior is necessary to analyze the stimuli involved in that behavior.

Ladybeetles, *Coccinella septempunctata* L., were reared on an excess of daily food, the green peach aphid, *Myzus persicae* SULZER, under 25±3°C and 16 hr photophase from egg to adult. Aphids were reared on potted cabbage plants under the same conditions of temperature and light as the beetles.

To observe the predatory behavior of the ladybeetle, an observation apparatus composed of a gray vinyl chloride cylinder (30 cm in diameter and 5 cm in height) resting on a white paper overlaid a transparent acryl plate, was used. The top of the cylinder was covered with another transparent

[1] Appl. Ent. Zool. **18** (4): 559–561 (1983)

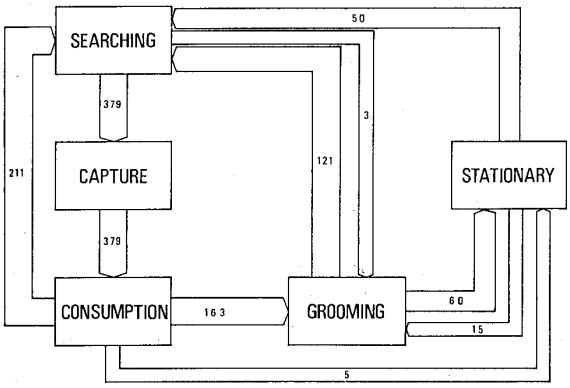

Fig. 1. Sequence of the predatory behavior of the ladybeetle, *Coccinella septempunctata*. Numbers in the arrows indicate the number of sequences observed.

acryl plate. The observation was conducted under 25±1°C during the light phase when the beetle is active. Ladybeetles used in the experiments were deprived of food for 24 hr beforehand.

Two hundred 3rd or 4th instar nymphs of the green peach aphid were distributed randomly in the observation arena. After one min, an adult *C. septempunctata* was introduced and the subsequent behavior was recorded. If prey were not encountered, the observation was terminated after 15 min, while if prey were encountered observation was continued until 60 min after the prey were no longer accepted by the ladybeetle. The form and sequence of movements of the ladybeetle, duration of each act and number of prey encountered were systematically recorded with audio tape. These observations were replicated eleven times with different ladybeetles.

The sequence and components of the predatory behavior are shown in Fig. 1. Three hundred and seventy-nine predations were observed in this experiment. The predatory behavior consisted of four distinct acts: searching, capture, consumption and grooming.

Searching: While searching for prey, the ladybeetle held its antennae parallel to the searching substratum and its maxillary palpi perpendicular to the substratum. It vibrated its maxillary palpi and sometimes turned its head from side to side. No ladybeetle was observed to directly orientate toward an aphid from a distance greater than several centimeters.

Capture: Upon making contact with a prey, the ladybeetle promptly captured it with its mandibles. Occasionally the predator used a combination of forelegs to aid in the mandibular capture (84/379 captures). Most of the initial prey contacts were made with the head and appendages of the ladybeetle, other contact sites being the forelegs and other parts. The capture efficiency (no. aphids captured/no. aphids contacted×100) was 98, 45 and 2% in the order of head, forelegs and other parts, respectively. The capture points of the aphid were the abdomen (73%), head (22%) and thorax (5%).

Consumption: A ladybeetle capturing a prey consumed the entire body. During consumption, they sometimes used their forelegs to aid in the manipulation. A predator consumed 3.9±3.9 (range 1–21) aphids during a single feeding bout. After a few replications of the sequence: searching—capture—consumption, the ladybeetle groomed itself or remained stationary (see Fig. 1). A few replications of this sequence have therefore been termed here a "feeding bout." In this experiment 98 feeding bouts were observed. A ladybeetle consumed 34.1±11.9 aphids (range 18–54) during an observation period (range 109–203 min). The handling time of a 3rd or 4th instar nymph of *M. persicae* with *C. septempunctata* was 48.8±19.9 sec.

Grooming: After consumption of a prey, the beetle immediately caught and consumed another if it was encountered or groomed the tibia of its forelegs with its mandibles, compound eyes and maxillary palpi. Grooming was observed both after prey consumption and after a feeding bout. It followed almost all feeding bouts (93/98); only in 5 did the ladybeetle remain stationary without grooming. However, the duration was much longer after a feeding bout (240±198 sec, $n=103$) than after the consumption of a prey (26±13 sec, $n=60$).

Stationary: Stationary was distinguished from other acts by its posture. During this time, the ladybeetle folded its legs and contacted its mouthparts and abdominal tip with the substratum.

Time spent for each component of the predatory behavior is shown in Table 1. Seventy-five percent of the time was expended on non-feeding activities (grooming and remaining stationary). The predator has to actually search for a prey because most aphid species are inactive. Since in this experiment sufficient aphids were contained in the arena, no searching time was needed. Banks (1957) reported that unfed coccinellid larvae spent more than 90% of the observation time in searching for a

Table 1. Time expended on components of predatory behavior of the ladybeetle, *Coccinella septempunctata*

	Component		
	Feeding bout[a]	Grooming	Stationary
Mean duration of a component (min)	3.2±3.3[b]	4.0±3.3	9.1±10.4
% Expended on each component	25.2±13.5	33.8±16.5	41.0±18.1
No. of components during observation	4.7± 1.7	5.2± 2.1	2.9± 1.2

[a] Feeding bout includes searching and capture, both of which were of short duration.
[b] Values are means±S.D.

prey when there was none.

It is suggested that the most frequent sequence of the predatory behavior of the ladybeetle is: searching—capture—consumption—searching and that grooming often appears after consumption (Fig. 1.). After grooming, the beetle will again search for a prey. However, if satiated it will simply become stationary.

The author is indebted to Drs. T. Saito and Y. Itô of this laboratory for their invaluable advice and critical reading of the manuscript. He also thanks Dr. T. Ono of Kinjo Gakuin University for his criticism of the manuscript.

REFERENCES

Banks, C. J. (1957) *Brit. J. Anim. Behav.* **5**: 12–24.
Curio, E. (1976) *The Ethology of Predation.* Springer-Verlag, Berlin, 250 p.
Hämäläinen, M., M. Markkula and T. Raij (1975) *Ann. Ent. Fenn.* **41**: 124–127.
Hodek, I. (1973) *Biology of Coccinellidae.* Academia, Prague, 260 p.

Preliminary Notes on Glycosidases in Two Cell Lines Derived from the Ovary of the Cabbage Armyworm, *Mamestra brassicae* (Lepidoptera: Noctuidae)[1]

Shigeru Kimura

Sericultural Experiment Station, Ohwashi, Yatabe, Ibaraki 305, Japan

Jun Mitsuhashi

National Institute of Agricultural Sciences, Kannondai, Yatabe, Ibaraki 305, Japan

(Received May 19, 1983)

Characterization of the enzymes in insect cell lines provides useful informations not only for clarifying the function of the enzymes, but also for distinguishing cell lines from each other. However, only a few attempts have been made to study the enzymes of cultured insect cells. The present paper reports the results of preliminary experiments on several glycosidases found in the cells and the culture media after the two cell lines were cultured.

The cell lines used in this experiment were NIAS-MB-19 and NIAS-MB-32, both derived from pupal ovaries of the cabbage armyworm, *Mamestra brassicae* (Mitsuhashi, 1977). In brief, each cell line was cultured in 30 m*l* of MM medium (Mitsuhashi and Maramorosch, 1964) containing 3% fetal bovine serum (FBS) at 25°C. After a week of cultivation, the cells were harvested, spun down, and both of the cells and the media were stored separately in a refrigerator until use. Amounts of cells used for each enzyme experiment were expressed as a protein concentration.

The enzyme was prepared as follows; the cells, separated from the culture medium by low speed centrifugation, were washed three times in a salined phosphate buffer solution (pH 7.0) in a cold room, and then homogenized with 4 m*l* of the above buffer by means of a sonicator for 30 sec. The homogenate was centrifuged at 10,000 g for 20 min,

[1] Appl. Ent. Zool. **18** (4): 561–563 (1983)