

## A REVIEW OF FEEDING STUDIES OF LYGUS SPP. WITH EMPHASIS ON ARTIFICIAL DIETS

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### ABSTRACT

Several researchers have reported varying degrees of success with diets for *Lygus* spp. Earlier efforts at developing diets for lygus were based on the assumption that the hemipteran feeding mechanism restricts ingestion to materials that were already liquids before feeding had begun. Therefore, the diets formulated for lygus bugs consisted only of ingredients that were soluble or that could be included as emulsions (i.e., lipids). Interestingly, simultaneous with the early diet work, several exquisitely detailed and very sophisticated histological studies showed that lygus bugs destroy solid plant tissues and remove particulate cellular contents, including storage vacuoles, plastids, nuclei, etc. In the mid-1970's some literature emerged describing lygus bugs as facultative entomophages, but diet researchers evidently assumed that lygus bugs, as well as other hemipterans, used only the "body fluids" of their insect prey. Although earlier diet work provided some excellent basic information about consumption and utilization rates, as well as some excellent basic nutrition information, none of these studies provided diet formulations that could be used in a practical lygus rearing system. It was not until a nutritionally complex diet slurry was described that rearing systems could be advanced. The feeding mechanisms of lygus are discussed here as they relate to application to artificial diet technology.

### INTRODUCTION

*Lygus* bugs continue to be among the most formidable pests of agriculture on a world-wide basis. One aspect of their success as pests is their ability to use a wide variety of plants as hosts (Hedlund and Graham 1987). The ability of lygus bugs, especially the western tarnished plant bug (WTPB), *L. hesperus* Knight and the tarnished plant bug (TPB) *L. lineolaris* (Palisot de Beauvois), to use such a wide variety of hosts is certainly related to their very efficient feeding apparatus. An appreciation for the interest generated by the lygus feeding apparatus can be gained even by a cursory survey of the extensive literature on this subject over the past six decades. The main purpose of this paper is to demonstrate that many aspects of lygus management depend on a thorough understanding of the feeding system used by these incredibly successful pests. It is especially important to understand the basics and nuances of the lygus feeding apparatus if mass rearing technology is to be applied to management strategies. This review is intended to show how a such an understanding of the lygus feeding mechanism is important in the development and

improvement of artificial diets for these insects. It is also intended to suggest that failure to apply such knowledge resulted in inability to develop a mass rearing system based upon artificial diet technology.

## OVERVIEW OF LYGUS FEEDING

Lygus bugs can increase their biomass from 0.050 mg to 12 mg (~240 x), and a female WTPB can produce 500 eggs (about 25 mg) in a 20-day period of reproductive activity (Cohen, unpublished data). To accomplish this biomass accretion, the feeding apparatus must undertake highly efficient ingestion, digestion, and absorption processes. As hemipterans, lygus bugs use a process of extra-oral digestion or solid-to-liquid feeding as described by Cohen (1990, 1995, 1998a, and unpublished data). Briefly, this process involves the insertion of mouthparts called stylets, which are paired structures that are highly modified mandibles and maxillae (Miles 1972). The mandibular stylets contain strong dentition and serve to penetrate and mechanically macerate tissues. The maxillary stylets are interlocking, and they form a functionally singular device with a pair of channels that run the entire length: a salivary canal and a food canal. The salivary canal is used to deliver watery secretions containing digestive enzymes that are injected into the food source and applied to propitious sites (Cohen 1990, Miles 1972). The actions of the mandibular stylets (mechanical maceration) and the saliva (chemical maceration) conspire to turn targeted plant or prey tissues into a highly concentrated, nutritious slurry. The slurry, containing dissolved and particulate nutrients, is sucked up the food canal into the buccal cavity and forced by swallowing action into the anterior gut. Peristalsis carries the slurry into the midgut where digestion continues via the action of ingested salivary enzymes and the secretion of enzymes produced within the gut cells (Cohen 1995, 1998a, b). Finally, absorption of the simple products of digestion takes place in the posterior midgut, where digested materials are transported via the hemolymph.

This type of feeding (extra-oral digestion or solid-to-liquid feeding) is not unique to mirids, such as lygus, nor to Hemiptera. This process is commonly used throughout Arthropoda (and in several other invertebrate groups), and its salient feature is that it permits very precise selection of nutrients that were highly concentrated in some part of the food source targeted by users of this feeding strategy. In the case of lygus bugs, foods such as nutrient rich, solid plant parts, especially fruit and floral tissues or meristematic cells, and insect tissues are turned into slurries rich in proteins, carbohydrates, and lipids; and they are ingested in a mixture of digestive enzymes that have already begun the digestive process. The hallmark of this system is efficiency of ingestion and speed of processing of nutrients. It cannot be stated strongly enough that the material ingested and selected by lygus bugs is not confined to (or mainly) plant liquids such as saps (xylem, phloem, or nectar), nor are lygus bugs confined to ingesting hemolymph when they take on the role of facultative predators.

## MORPHOLOGY OF THE FEEDING APPARATUS

There are no studies dedicated to the structure or morphological features of the mouthparts of lygus, although Hatfield and Frazier (1979) characterized the ultrastructure of the labial tip sensilla. Lygus mouthpart morphology is included, however, in general treatments of hemipteran feeding structure and function such as in the studies by Cobben (1978), Cohen (1990, 1998b), and Miles (1972). The salivary gland morphology was described by Baptist (1941) and Nishijima and Sogawa (1963). Lygus bugs have no

extraordinary structures or morphological features that distinguish them from other mirids or heteropterans, in general. However, it is important to note that lygus, typical of mirids, do not secrete a stylet flange or any other "salivary sheath" material (Cohen 1998c, 2000). This means that they must get leverage for stylet movements (twisting, penetration thrusts, and withdrawal) from counter forces of the apposed stylets and anchoring from mandibular grasping of the surface that they penetrate. The biomechanics of this action are very dynamic and deserve study. This will become increasingly clear as feeding activity within plants is discussed in the next section. It is also important to note that lygus bugs are typical of the majority of heteropterans in having a well developed pair of accessory glands whose function is to recycle water from food material in the gut back to the salivary glands. The result of this recycling is that the water, which is used to carry digestive enzymes from the salivary glands into the food source, is continuously re-used throughout the feeding bout. This is one of the telling features that lygus bugs are suited for solid-to-liquid feeding, not for feeding on dilute liquids. Secondly, the structure of the gut is a straight tube rather than a filter chamber or other special apparatus for concentrating dilute liquids as is found in true fluid-feeding Hemiptera.

### FEEDING ACTIVITY IN PLANTS

Several studies have been dedicated to understanding the damage to plant tissues attacked by lygus bugs (e.g., Flemion et al. 1954, Hori 1971, Tingey and Pillemer 1977). The consensus of these studies is that lygus bugs use new growth tissues (meristematic tissues) and flower or young fruit tissues preferentially over stems, leaves, or other less nutritive plant materials. There is less agreement on the cause(s) of feeding damage by lygus bugs, with some authors contending that the destruction is due to mechanical actions of the stylets (Flemion et al. 1954), while others attribute damage to the action of digestive enzymes (e.g., Strong 1970) or to a combination of mechanical and chemical maceration (e.g., Tingey and Pillemer 1977).

One of the most remarkably thorough studies of lygus feeding and histological interactions is that of Flemion et al. (1954). They used a variety of techniques to demonstrate the nature of lygus feeding, including high speed microscopic cinematography, histological sectioning, and electric shock to freeze the stylets *in situ* in host tissues. These authors described the rapid movements of stylets darting in between and inside cells, zig-zagging back and forth, and even turning back upon themselves making more than 180 degree bends. In a striking series of photomicrographs, they showed the collapse of tissues such as parenchyma cells in parsnip fruits. Although they made brief mention of the possibility of tissue destruction by salivary fluids and microbial contamination, their emphasis was that lygus caused damage mechanically by removing plant fluids, leading to a "collapse" of cells. Other authors such as Hori (1970, 1971), Strong (1970), Strong and Landes (1965), and Strong and Kruitwagen (1969) placed more emphasis upon digestive enzymes and what Tingey and Pillemer (1977) later referred to as "external digestion" (what I call extra-oral digestion or solid-to-liquid feeding).

What emerges from these studies is the understanding that lygus bugs use a maceration-type feeding to remove very nutritious, highly concentrated, originally solid materials from the most nutrient-rich parts of plants. However, there has been confusion about the true nature of lygus bug feeding because of the frequently repeated statements (sometimes by the same authors) that lygus bugs remove plant fluids. An example of this type of confusion can be seen in the following paragraph from Tingey and Pillemer (1977):

"Following maceration of plant tissues by combined action of the stylets and salivary secretions, lygus bugs withdraw considerable amounts of liquified plant material. Hori (1971b) reported that nymphs of *L. disponsi* withdrew an avg of 82.5 mg plant juice/bug over 5 stadia, while the withdrawal rate for adults was 7-13 mg plant juice/day. Strong (1970) reported that a single adult *L. hesperus* could ingest the liquified contents of an alfalfa bud in only 23 sec."

Clearly, Tingey and Pillemer express correctly that lygus bugs macerate plant tissues and remove liquified material. However, the use of the term "plant juice," which is repeatedly used in literature on lygus feeding, and general hemipteran feeding, suggests that lygus feeding is specific to and exclusively liquids within the plant tissues. This ambiguity is typical and is a major factor in the disappointingly slow progress in development of artificial diets for lygus as will be explained in the next section.

### ARTIFICIAL DIETS FOR NUTRITIONAL STUDIES OR FOR PRACTICAL PURPOSES?

The problem of lygus dietetics is further exacerbated by some underlying confusion about the purposes of artificial diets. Nutritionists use artificial diets as tools to help decipher the nutritional requirements of organisms. They use chemically-defined media (holidic diets) to control the exact composition of foods ingested by the organisms being studied. Once a complete diet is established, the researcher can then use a systematic deletion or replacement technique to determine the essentiality of given nutrients in a painstaking "one at a time" process of elimination. Because of difficulties in purifying macromolecules such as proteins, simple sub-units such as free amino acids, sterols, salts, etc. are generally used in development of defined diets.

Although defined diets are good research tools, they are not good diets for most organisms. Natural products are very complex, and the nutrient composition of even the most "simple" materials such as xylem and phloem saps and nectars have scores of components and myriad combinations and concentrations of these components. Work on phloem-feeding insects, especially aphids, was relatively successful with respect to both nutrition and practical dietetics, in part, because of the simplicity of saps compared to complex tissues that contain organelles, storage granules, cell membranes, and other complex substances. Unfortunately, assumptions that as so-called "liquid feeders" (described above), lygus bugs ingested the same materials utilized by aphids and other vascular sap feeders are common.

Starting with this assumption, early work on lygus diets (e.g., Auclair and Raulston 1966; Hori 1977; Raulston and Auclair 1968; Landes and Strong 1965; Vanderzant 1967) focused on defined diets based on fairly successful diets formulated for aphids or other plant sap feeders. Furthermore, all of these diets were presented as fairly dilute liquids, as their counterparts were presented to aphids and other sap feeders. Unfortunately, phloem or xylem feeders are not good models for diet development for insects that feed on more complex materials than plant saps. Therefore, there was little progress in developing diets for lygus bugs.

The principal nutrients of saps are sugars, free amino acids, minerals, and sparse amounts of lipids (Srivastava 1987). The digestive systems of true sap feeders are highly specialized for concentration of non-macromolecular components, and they tend to be void of enzymes for digestion and utilization of macromolecular structures (Miles 1987). The

assumption that lygus bugs somehow extracted liquids (i.e., nutrients that were already in solution) from plant tissues evidently lay behind all the pre-1980s dietary studies. In fact, even after the development of an excellent diet for WTPB (Debolt 1982), there was considerable misunderstanding of how the diet worked, with the confusion resulting from misunderstanding of the fundamental feeding biology of lygus bugs. A personal anecdote about the degree of confusion about lygus feeding will be informative and will substantiate my point.

When I arrived at the USDA, ARS, Biological Control of Insects Laboratory in Tucson, AZ in 1979 Dr. Jack Debolt had already succeeded in developing a diet for WTPB and had reared numerous continuous generations of healthy individuals. As an insect biochemist, I was asked to help identify the nutrients that were actually responsible for the excellent biological success of the WTPB provided with the Debolt diet, a complex mixture of lima bean meal, wheat germ, chicken eggs and a large number of defined nutrients. Because it was unassailably certain to me that the true nutrients had to be liquids (soluble liquids, in my mind), I used filtration systems and centrifuges to discard the solid "sludge" in the diet so that we could purify the nutritious liquids.

The more I freed the diet of solid "sludge," the more poorly the WTPB performed in bioassays. I also made extracts of natural diets such as green beans and beet armyworm hemolymph, all to no avail. Simultaneously, efforts made by Dr. Debolt to reduce the solid components of the diet also failed, and the highly successful rearing program (Debolt 1982, 1987, 1989, Debolt and Patana 1985, Patana 1982; DeGrandi-Hoffman 1994) based on this diet and serving as a basis for mass-rearing WTPB and its parasites continued with the original slurry-type diet. Over the past 20 years of my career, I developed an understanding first of how predaceous arthropods use solid-to-liquid feeding (Cohen 1998a) and, more recently, how phytophagous Heteroptera such as lygus bugs use this maceration feeding method to obtain highly nutritious slurries of food. This understanding makes sense of the major reasons for the excellent success of the Debolt diet for nurturing lygus bugs. The understanding also served as a basis for my developing a more simple diet for WTPB (Cohen unpublished), a diet used for over a year to support the WTPB colony at the USDA, ARS, Biological Control and Mass Rearing Research Unit at Mississippi State, MS.

Video microscopy of WTPB feeding on both the Debolt and the Cohen diets makes it evident that these insects use their stylets to mechanically rasp food particles that are too large to ingest. Using biochemical tests, Hori (1970a; 1970b) showed that *L. disponsi* Linnavuori possess salivary digestive enzymes capable of breaking down complex carbohydrates and proteins; Laurema et al. (1985) showed that *L. rugulipennis* Poppius produce a complex of digestive enzymes capable of hydrolyzing macromolecular components in plant tissues; Cohen (1990), Agusti and Cohen (2000) and Zeng and Cohen (2000) showed that both WTPB and TPB produce macromolecular digesting enzymes that could attack both plant and animal tissues. Unifying the histology/plant damage studies, the digestive enzyme studies, the anatomy of the feeding apparatus, and direct feeding observations on slurry-type diets, it is evident that lygus bugs are equipped to do much more than suck saps out of plants. Instead, it is evident that lygus bugs regularly feed on tissues (both of plants and animals) that are solid or semisolid and as such highly concentrated with appropriate nutrients, including lipids, proteins, and complex carbohydrates, as well as micronutrients, such as vitamins and minerals.

## THE NEXUS BETWEEN DIGESTIVE ENZYMES AND ARTIFICIAL DIETS

Cohen (1989, 1998a) discussed the importance of understanding the basics of the feeding process of a given insect for the development of a practical artificial diet for that insect. When faced with the difficult task of developing an artificial diet for an insect, several approaches can be taken, each with varying degrees of success. Among such techniques as analysis of the foods naturally eaten by a given insect, whole carcass analysis, trial and error using already established diets for related species (Cohen 1984, 1985, 1992), and analysis of digestive enzymes, the latter is probably the most telling about the kinds of macromolecules that an insect is predisposed to ingest (Cohen 1998a).

### THE IMPORTANCE OF COMPREHENSIVE FEEDING STUDIES

The emergence of lygus bugs as serious pests in numerous cropping systems worldwide and the problem of their development of resistance to pesticides mandates development of non-conventional management strategies. Such non-conventional methods, which include but are not limited to biological control, sterile insect techniques, lethal F<sub>1</sub> hybrids, pheromone manipulation, or genetic alteration of host plants require a thorough understanding of the target species' biology, especially feeding biology. Such methods often call for colonization and mass-rearing technologies which depend upon reliable means of rearing high quality insects for both experimental use or as penultimate products (such as in production of natural enemies for biological control). Because artificial diets are economical, require less labor than natural diets, are easier to standardize, and lend themselves to automation (Nordlund 1994, Nordlund and Greenberg 1996), mass rearing technology depends upon the availability of high quality artificial diets.

It has been pointed out (Cohen 1985, 1990, 1992, and 1998a) that success in developing artificial diets depends greatly upon a thorough understanding of the feeding system of a given species. It is evident from studies of the histology of lygus feeding damage and the preliminary studies on the nature of lygus digestive enzymes, that these mirids (like most Heteroptera) use a feeding strategy of macromolecular disruption. They use a typical lacerate and flush (as described by Miles (1972) feeding method, which depends on application of potently hydrolytic digestive enzymes that attack a variety of macromolecules (both in plants and in other insects). In fact, it is not overstating the case to say that the foundation of lygus feeding (and that of most Heteroptera and other taxa that use extra-oral digestion) is the macromolecular digestive enzymes with which these insects are equipped.

### UNDEFINED BUT PRACTICAL DIETS FOR LYGUS BUGS

What is meant by a practical diet is one that succeeds in producing high quality, healthy animals on a continuous basis. The degree of practicality increases as the cost and complexity of production decreases. The Debolt (1982) diet contains complete nutrient complexes such as lima bean meal and wheat germ (both seed derivatives with the incumbent nutrient richness), as well as supplementary nutrients such as casein hydrolyzate, vitamin mixture, salt mixture, soy lecithin (with soy oil), RNA, linoleic acid, and sugar. It is interesting to note that Vanderzant (1967), a pioneer in the use of wheat germ in insect diets, had used wheat germ in a lygus diet, but she prepared the diet with an aqueous extract of the wheat germ. The aqueous extract either did not contain the proper array of nutrients or they were too dilute to be of advantage in the resulting diet. The use

of lima bean meal further enhances the availability of plant proteins, plant lipids (such as phytosterols), and other plant nutrients such as mineral complements rich in potassium and magnesium.

In developing a simpler diet for WTPB, I felt that while the unrefined plant materials such as wheat germ and lima bean meal were very useful nutrients that could be included economically, there was possibly no need for the defined nutrients mentioned above. Instead, I chose to use nearly all undefined, whole foods that should supply the appropriate amounts of nutrients required by WTPB, and to satisfy the apparent requirement for animal protein (Wheeler 1976, Bryan et al. 1976, Debolt 1982), I used a supplement of the Cohen entomophage diet (Cohen 1998b, Cohen and Smith 1998). The results of extensive tests of the Cohen diet for WTPB (Cohen unpublished data) show that the Debolt diet is superior in terms of adult survival and total numbers of eggs that can be obtained for a given number of females. However, the Cohen diet is equal to the Debolt diet in egg weight, adult yield. Also, the Cohen diet is less expensive (about 1/8 the cost of Debolt diet ingredients) and simpler to prepare. Both diets, however, have the consistency of a slurry with fairly large particles (>500 microns), both have a very high nutrient concentration, and both offer the lygus bugs a choice of nutrients because of a fairly great heterogeneity. One final point is that the use of gel packets placed on top of the cage screen for oviposition (Patana 1982 and Debolt and Patana 1985) adds to the simplicity of the artificial diet-based rearing system, and it answers a nagging question about the ability to produce lygus bugs without fresh plant material.

These diets stand as examples of how well lygus bugs can be reared (including mass rearing) on an artificial diets. It is my contention that because an increased understanding of the feeding process reviewed here, progress in lygus dietetics, as well as in diets for other hemipterans that feed similarly, can be rapid and rewarding.

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#### LITERATURE CITED

- Agustí, N. and A. C. Cohen. 2000. Salivary and mid-gut digestive enzymes from *Lygus hesperus* and *L. lineolaris*. J. Entomol. Sci. (In Press).
- Auclair, J. L. and J. R. Raulston. 1966. Feeding of *Lygus hesperus* (Hemiptera: Miridae) on a chemically defined diet. Ann. Entomol. Soc. Amer. 59:1016-1017.
- Baptist, B. A. 1941. The morphology and physiology of the salivary glands of the Hemiptera-Heteroptera. Quart. J. Microsc. Sci. 83:91-139.
- Bryan, D. E., C. G. Jackson, R. L. Carranza, and E. G. Neemann. 1976. *Lygus hesperus*: production and development in the laboratory. J. Econ. Entomol. 69:127-129.
- Cobben, R. H. 1978. Evolutionary trends in Heteroptera. Part II. Mouthpart-structures and feeding strategies. Med. Land. Wageningen 78-5, H. Veeman and Zonen B. V.
- Cohen, A. C. 1984. Food consumption, food utilization and metabolic rates of *Geocoris punctipes* (Het.: Lygaeidae) fed *Heliothis virescens* (Lep.: Noctuidae) eggs. Entomophaga 29:361-367.

- Cohen, A. C. 1985. Simple method for rearing the insect predator *Geocoris punctipes* (Heteroptera: Lygaeidae) on a meat diet. *J. Econ. Entomol.* 78:1173-1175 .
- Cohen, A. C. 1989. Ingestion efficiency and protein consumption by a heteropteran predator. *Ann. Entomol. Soc. Am.* 82:495-499.
- Cohen, A. C. 1990. Feeding adaptations of some predatory Hemiptera. *Ann. Entomol. Soc. Am.* 83:1215-1223.
- Cohen, A. C. 1992. Using a systematic approach to develop artificial diets for predators. pp. 77-91. *In* T. E. Anderson and N. C. Leppla, (eds.) *Advances in Insect Rearing for Research & Pest Management*. Boulder. Westview Press. 519 pp.
- Cohen, A. C. 1995. Extra-oral digestion in predatory Arthropoda. *Annu. Rev. Entomol.* 40:85-103.
- Cohen, A. C. 1998a. Solid-to-liquid feeding: the inside(s) story of extra-oral digestion in predaceous Arthropoda. *American Entomologist.* 44:103-117.
- Cohen, A. C. 1998b. Artificial media for rearing entomophages. Docket # 0087.96, Serial # 08/699,815, U.S. Patent # 5,834, 177, November 10, 1998.
- Cohen, A. C. 1998c. Biochemical and morphological dynamics and predatory feeding habits in terrestrial Heteroptera. pp. 21-32. *In* M. Coll and J. R. Ruberson (eds.) *Predatory Heteroptera in agroecosystems: their ecology and use in biological control*. Thomas Say Publications in Entomology. Entomological Society of America, Lanham, MD.
- Cohen, A. C. 2000. Feeding fitness and quality of domesticated and feral predators: effects of long term rearing on artificial diet. *Biological Control* 17:50-54.
- Cohen, A. C. and L. K. Smith. 1998. A new concept in artificial diets for *Chrysoperla rufilabris*: efficacy of solid diets. *Biological Control.* 13:49-54.
- Debolt, J. W. 1982. Meridic diet for rearing successive generations of *Lygus hesperus*. *Ann. Entomol. Soc. Am.* 75:119-122.
- Debolt, J. W. 1987. Augmentation: Rearing, release and evaluation. *In*: R. C. Hedlund and H. M. Graham. (eds.) *Economic Importance of Biological Control of Lygus and Adelphocoris in North America*. pp. 82-87. U.S. Dept. Agric. Res. Serv. (ARS-64).
- Debolt, J. W. 1989. Encapsulation of *Leiophron uniformis* by *Lygus lineolaris* and its relationship to host acceptance behavior. *Entomol. exp. appl.* 50:87-95.
- Debolt, J. W. and Patana, R. 1985. *Lygus hesperus*. pp. 329-338. *In* *Handbook of Insect Rearing*. Vol. 1. P. Singh and R. F. Moore (eds.) Elsevier Science Publishers B. V. Amsterdam. 488 pp.
- DeGrandi-Hoffman, G., Diel, J., D. Li, L. Flexner, G. Jackson, W. Jones, J. Debolt. 1994. BIOCONTROL-PARASITE: Parasitoid-host and crop loss assessment simulation model. *Environ. Entomol.* 23:1045-1060.
- Flemion, F., M. C. Ledbetter, and E. Schaefer Kelley. 1954. Penetration and damage of plant tissues during feeding by the tarnished plant bug (*Lygus lineolaris*). *Contrib. Boyce Thompson Inst.* 17:340-357.
- Hatfield, L. D. and J. L. Frazier. 1979. Ultrastructure of the labial tip sensilla of the tarnished plant bug, *Lygus lineolaris* (P. De Beauvois) (Hemiptera: Miridae). *Int. J. Insect Morphol. & Embryol.* 9:59-66.
- Hedlund, R. C. and H. M. Graham. 1987. Economic importance and biological control of *Lygus* and *Adelphocoris* in North America. USDA, Technical Bulletin ARS-64. 95 pp.
- Hori, K. 1970a. Some properties of amylase in the salivary gland of *Lygus disponisi* (Hemiptera). *J. Insect Physiol.* 16:373-386.



- Hori, K. 1970b. Some variations in the activities of salivary amylase and protease of *Lygus disponsi* Linnavuori (Hemiptera, Miridae). *App. Entomol. Zool.* 5:51-61.
- Hori, K. 1970c. Some properties of proteases in the gut and in the salivary gland of *Lygus disponsi* Linnavuori (Hemiptera: Miridae). *Res. Bull. Obihiro Univ.* 6:318-324.
- Hori, K. 1971. Studies on the feeding habits of *Lygus disponsi* Linnavuori (Hemiptera: Miridae) and the injury to its host plants I. Histological observations of the injury. *App. Entomol. Zool.* 6:84-90.
- Hori, K. 1977. Uselessness of casamino acids for the rearing of *Lygus disponsi* Linnavuori (Hemiptera: Miridae). *Res. Bull. Obihiro Univ.* 10:743-747.
- Landes, D. A. and F. E. Strong. 1965. Feeding and nutrition of *Lygus hesperus* (Hemiptera: Miridae). I. Survival of bugs fed on artificial diets. *Ann. Entomol. Soc. Amer.* 58:306-309.
- Laurema, S., A. Varis, and H. Miettinen. 1985. Studies on enzymes in the salivary glands of *Lygus rugulipennis* (Hemiptera: Miridae). *Insect Biochem.* 15:211-224.
- Miles, P. W. 1972. The saliva of Hemiptera. *Advances in Insect Physiology.* 9:183-255.
- Miles, P. W. 1987. Feeding process of Aphidoidea in relation to effects on their food plants. In: *Aphids: Their Biology, Natural Enemies and Control.* A. K. Minks and P. Harrewijn, eds. Elsevier. Amsterdam. 321-339 pp.
- Nishijima, Y. and K. Sogawa. 1963. Morphological studies on the salivary glands of Hemiptera I. Heteroptera. *Res. Bull. Obihiro Zotech. Univ.* 3:512-521.
- Nordlund, D. A. 1996. Biological control, integrated pest management and conceptual models. *Biocontrol News and Information* 17:35-44.
- Nordlund, D. A. and S. M. Greenberg. 1994. Facilities and automation for the mass production of arthropod predators and parasitoids. *Biocontrol News and Information* 4:45-50.
- Patana, R. 1982. Disposable diet packet for feeding and oviposition of *Lygus hesperus* (Hemiptera: Miridae). *J. Econ. Entomol.* 75:668-669.
- Raulston, J. R. and J. L. Auclair. 1968. Responses of *Lygus hesperus* to chemically defined diets. *Ann. Entomol. Soc. Am.* 61:1495-1500.
- Srivastava, P. N. 1987. Nutritional physiology. In: *Aphids: Their Biology, Natural Enemies and Control.* A. K. Minks and P. Harrewijn, eds. Elsevier. Amsterdam. 99-130 pp.
- Strong, F. E. 1970. Physiology of injury caused by *Lygus hesperus* J. Econ. Entomol. 63:808-814.
- Strong, F. E. and D. A. Landes. 1965. Feeding and nutrition of *Lygus hesperus* (Hemiptera: Miridae). II. An estimation of normal feeding rates. *Ann. Entomol. Soc. Am.* 58:309-314.
- Strong, F. E. and E. Kruitwagen. 1969. Feeding and nutrition of *Lygus hesperus*. III. Limited growth and development on a meridic diet. *Ann. Entomol. Soc. Am.* 62:148-155.
- Tingey, W. M. and E. A. Pillemer. 1977. *Lygus* bugs: crop resistance and physiological nature of feeding injury. *Bull. Ent. Soc. Amer.* 23:277-287.
- Wheeler, A. G. 1976. *Lygus* bugs as facultative predators. In Scott, D. R. and L. E. O'Keefee, eds., *Lygus Bug: Host Plant Interactions.* Univ. Idaho Press, Moscow.
- Vanderzant, E. S. 1967. Rearing *Lygus* bugs on artificial diets. *J. Econ. Entomol.* 60:813-816.
- Zeng, F. and A. C. Cohen. 2000. Partial characterization of  $\alpha$ -amylase in the salivary glands of *Lygus hesperus* and *L. lineolaris*. *Comp. Biochem. And Physiol.* (In Press).