Formosan Entomologist
Journal Homepage: entsocjournal.yabee.com.tw

【Research report】

水稻黑尾葉蟬對氨基酸及膽固醇之營養需求【研究報告】

林兩傳、侯豐男

*通訊作者E-mail :

Received: Accepted: Available online: 1981/03/01

Abstract

摘要

本文以人工合成飼料飼育法研究水稻黑尾葉蟬 (Nephotettix cincti-ceps) 對氨基酸及膽固醇之營養需求。由10種必需氨基酸中略去阿金氨酸 (arginine) 與苯初油氨酸 (phenylalanine)對此葉蟬之生長及成蟲羽化率不具無有害之效應，但除去甲硫氨酸 (methionine) 則引起100 ％ 若蟲死亡率。本蟲可用含有23種氨基酸之飼料飼育，但無法在僅含10種必需氨基酸之飼料存活。 以成群略去天門冬氨酸 (aspartic acid)，天門冬氨酸胺 (asparagine)，麩氨酸 (glutamic acid) 與麩氨酸胺 (glutamine)，或膀胱氨酸 (cystine) 與還原膀胱氨酸 (cysteine)；或膀胱氨酸，還原膀胱氨酸與甲硫氨酸等各群，致使葉蟬發育不良。由13種非必需氨基酸中除去普羅林(prol-ine) 並無顯著影響葉蟬之生長。 膽固醇以5mg/100ml之濃度最適於葉蟬之生長及發育，經定量分析第五齡若蟲蟲體，發現以含有5mg/100ml膽固醇之飼料所飼者，其體內膽固醇含量為1.789ug/mg 乾重，其與在水稻苗生長者之含量相似。

Key words:

關鍵詞:

Full Text: PDF(0.53 MB)

中華昆蟲 1 (41–53)
Chinese J. Entomol. 1 (41–53)

DIETARY REQUIREMENTS OF *NEPHOTETTIX CINCTICEPS* FOR AMINO ACIDS AND CHOLESTEROL

Lian-chang Lin and Roger F. Hou
Department of Entomology
National Chung Hsing University
Taichung, Taiwan
Republic of China

ABSTRACT

Dietary requirements of the rice green leafhopper, *Nephotettix cincticeps*, for amino acids and cholesterol were studied by feeding it on a holidic diet. Deletion of arginine or phenylalanine among 10 'essential' amino acids exhibited no detrimental effect on growth and adult emergence of the leafhopper, but omission of methionine caused 100% mortality of the nymphs. The leafhoppers can be reared on a diet containing 23 amino acids, but they are unable to live on the diet with 10 'essential' amino acids only.

Group deletion of aspartic acid, asparagine, glutamic acid, and glutamine; or cysteine and cystine; or cysteine, cystine, and methionine from diets supported poor development. Deprivation of proline from 13 'non-essential' amino acids did not markedly affect leafhopper growth.

Cholesterol was found suitable for the leafhopper growth and development at 5mg/100ml. Analysis of the tissue cholesterol revealed that the leafhoppers fed on the diet containing 5mg/100ml cholesterol have a content, *i.e.*, 1.789μ/mg dry wt., similar to those insects from rice seedlings.

INTRODUCTION

Nutritional requirements of plant-sucking insects, especially aphids and planthoppers, have been extensively studied (Mittler, 1976; Hou, 1977; Koyama and Mitsuhashi, 1979). Some dietary components may exert specific functions in this group of insects, for example, certain amino acids as phagostimulants or ovipositional factors; cholesterol as an imaginal moulting factor; folic acid as a wing-morph factor, etc. (Mittler, 1967; Mitsuhashi and Koyama, 1974; 1977; Hou and Brooks, 1975; Srivastava and Auclair, 1975). However, little is known about dietary requirements of the rice green leafhopper, *Nephotettix cincticeps*, which is one of the major vectors of rice diseases in Far East Asia. Since a holidic diet and feeding method has been developed for rearing this insect (Hou and Lin, 1979), it is of particular interest to study whether some dietary components are as important in leafhopper growth and development as they are in other animals. This report presents investigations on requirements of the rice green leafhopper for cholesterol and individual or groups of amino acids. Analysis of tissue cholesterol as affected by different dietary levels was conducted to elucidate the optimal dietary concentration.

MATERIALS AND METHODS

Insects and the diet

Stock cultures of *N. cincticeps* were maintained for several months on rice seedlings *which were water-cultured in a 9-cm Petri dish at 25° ± 2°C in the laboratory, offspring* of these insects were used for the experiments. Newly hatched nymphs were used in the feeding tests.

Composition of the standard diet is shown in Table 1. The feeding diets were prepared according to procedures described by Hou and Brooks (1975). Deletion of individual dietary ingredients or components in groups from the standard diet was conducted to examine their nutritional effect. Feeding tests were carried out in a growth chamber (Sherer Cel-8, Kysor Industrial Corp., Michigan, USA) with 25° ± 1°C and 85 ± 5% R.H. The criteria used for estimating nutritive value of various diets were comparisons of cumulative nymphal mortality, nymphal period, and adult emergence. Each treatment was replicated three times, and the data were analysed by the Duncan's multiple range test at 5% level.

Table 1. Composition of the standard diet for the rice green leafhopper, *Nephotettix cincticeps.*

I.	Amino Acids	mg/100 ml
	L-alanine	40
	L-arginine (Free base)	160
	L-asparagine	120
	L-aspartic acid	40
	L-cysteine (Free base)	20
	L-cystine	2
	γ-amino butyric acid	8
	L-glutamic acid	80
	L-glutamine	240
	Glycine	8
	L-histidine	32
	DL-homoserine	320
	L-isoleucine (D-Allo)	80
	L-leucine	80
	L-lysine HCl	80
	L-methionine	40
	L-phenylalanine	40
	L-proline	40
	L-serine	40

L-threonine	80
L-tryptophan	40
L-tyrosine	8
L-valine	80
II. B-Vitamins & Lipogenic Factors	
Biotin	0.05
Calcium pantothenate	2.50
Folic acid	0.25
Nicotinic acid	5.00
Pyridoxine HCl	1.25
Riboflavin	0.25
Thiamine HCl	1.25
Choline chloride	25.00
i-Inositol (meso)	25.00
III. L-Ascorbic acid	100.00
IV. Trace Metals	
$CuCl_2 \cdot 2H_2O$	0.051
$FeCl_3 \cdot 6H_2O$	0.534
$MnCl_2 \cdot 4H_2O$	0.100
$CaCl_2$	0.444
Zn-sequestrene	0.160
V. Cholesterol Dispersion	
Cholesterol	5.0
Lecithin (Vegetable)	5.0
Tween 80	10.0
CI. Others	
Na citrate	10.0
$MgSO_4 \cdot 7H_2O$	242.0
KH_2PO_4	250.0
Sucrose	5000.0
VII. pH	6.8 adjusted with 1.75 M KOH

Amino acids and vitamins were purchased from the Sigma Chemical Co., St. Louis, Mo., U. S.A.; Cholesterol, lecithin and Tween 80 were obtained from the ICN Nutritional Biochemicals Corp., Clevelnad, Ohio, U. S.A.

Quantitative analysis of cholesterol concentration of the leafhopper tissues

The standard curve was made using the method described by Zlatkis and Zak (1969). The working standard of cholesterol was prepared by dissolving 100mg dry cholesterol in 100ml glacial acetic acid, and then further diluting to 0, 20, 40, 60, and 80mg/100ml. Color reaction was obtained by pipetting 0.1ml of each working standard into 2ml o-phthalaldehyde reagent (50mg o-phthalaldehyde in 100ml glacial acetic acid), mixing well and then adding slowly 1ml concentrated H_2SO_4 (A.R.) from the top. After 10 minutes, absorvance at 550nm was read against a reagent blank by a spectrophotometer (Beckman Model BD-GT 1405). A linear regression equation ($Y = 0.01505 + 0.01909X$) was found to express the correlation between optical density and cholesterol concentrations.

The extraction technique for tissue cholesterol was modified from the methods by Saxena and Gohain (1974) and Zaidi and Khan (1972). Newly moulted 5th instar nymphs were collected from those fed on various cholesterol concentrations and on rice seedlings. The insects were first washed with distilled water and dried over filter papers, they were then macerated and homogenized with 3ml double distilled water, and centrifuged at 5,500 rpm for 30 min. The supernatant was discarded, and the residue was dried with an air blower. The dried homogenates were weighed and extracted with 2ml chloroform/methanol mixture (2 : 1, v/v) in a tube; the supernatant, regarded as the choloesterol extract, was then further centrifuged at 7,000 rpm for 30 min. Each extract was reduced to dryness in a water bath (70°C), and residue was analysed by the same method as used for the standard.

RESULTS AND DISCUSSION

Requirements for amino acids

Nutritional requirement for each of the 10 'essential' amino acids was examined by individual deletion. Table 2 shows that adult emergence of the leafhopper is not markedly affected by individual omission of arginine or phenylalanine, and that deletion of leucine does not exhibit any significant difference in adult emergence compared to the complete diet. On the other hand, single deletion of other essential amino acids retarded nymphal growth and caused higher mortality to some extent.

It was reported that only histidine, isoleucine, and methionine were essential for *Myzus persicae*, whereas alanine, cystine, histidine, methionine, phenylalanine, proline, serine, and tyrosine were indispensable for rearing *Aphis fabae* (Dadd and Krieger, 1968; Leckstein and Llewellyn, 1973). Since we tested only the 10 amino acids essential for the rate, effects of some 'non-essential' amino acids, *e.g.*, alanine, cystine, serine, and tyrosine on growth of the leafhopper are uncertain. The omission of proline showed that this amino acid is not required for adult emergence, but that without proline the nymphal period is slightly prolonged (Table 3).

High mortality of the 1st instar nymphs occurs on single deletion of methionine, tryptophan, histidine, and valine, causing poor adult emergence later (Table 2). Koyama

Table 2. Effects of single deletion of 10 'essential' amino acids from a diet containing 23 amino acids on *Nephtettix cincticeps*

Amino acid deleted	Initial insects tested	Cumulative nymphal mortality (%)					Nymphal period (days)	Adult emergence[c] %
		1st	2nd	3rd	4th	5th		
Arg	29	6.9	20.4	34.8	41.2	47.6	21.6	52.4 abc
His	36	17.0	28.9	42.4	60.2	80.3	22.5	19.7 f
Ile	37	13.5	29.6	40.9	50.1	56.2	24.0	43.8 cd
Leu	47	12.7	27.5	38.2	48.2	52.8	23.1	47.2 bcd
Lys	32	12.5	30.4	43.8	51.4	60.3	24.6	39.7 d
Met	39	62.0	78.5	92.4	100.0	–	–	0.0 h
Phe	36	16.4	39.5	34.2	41.7	48.8	22.4	51.2 abc
Thr	42	7.2	21.5	38.6	51.8	58.1	23.8	41.9 d
Trp	39	52.4	42.0	51.4	60.6	71.1	23.2	28.9 e
Val	34	38.2	56.9	66.0	79.7	90.8	25.1	9.2 g
None[a]	37	5.4	21.3	32.8	39.6	45.5	21.2	55.5 ab
Check[b]	40	2.5	6.0	9.0	24.5	40.8	20.3	59.2 a

a/ On complete diet.

b/ On rice seedlings.

c/ Means followed by the same letter are not significantly different at 5% level.

Table 3. Effects of group deletions of some amino acids on growth and development of *Nephotettix cincticeps*

Amino acids deleted	Initial insects tested	Cumulative nymphal mortality(%)					Nymphal period (day)	Adult emergence c/ (%)	
		1st	2nd	3rd	4th	5th			
10 'essential' amino acids	42	64.2	100.0	–	–	–	–	0	c
13 'non-essential' amino acids	33	54.6	82.4	100.0	–	–	–	0	c
γ-Aminobutyirc acd & dl-homoserine	39	18.0	29.6	38.4	46.4	55.2	24.6	44.8	b
asp, asn, glu, & gln	38	31.5	59.4	89.2	100.0	–	–	0	c
Cysteine & cystine	39	28.4	42.5	81.0	92.5	100.0	–	0	c
Cysteine, cystine & methionine	38	39.8	58.4	80.5	100.0	–	–	0	c
Proline	38	10.6	22.6	30.4	38.2	46.4	22.8	53.6	ab
None a/	37	5.4	21.3	32.8	39.6	45.5	21.2	55.5	a
Check b/	40	2.5	6.0	9.0	24.5	40.8	20.3	59.2	a

a/ On complete diet. b/ On rice seedlings. c/ Means followed by the same letter are not significently different at 5% level.

and Mitsuhashi (1975) found that only methionine and cystine (or cysteine) were essential for the smaller brown planthopper, *Laodelphax striatellus*, based on nymphal period and adult emergence in the first generation. Srivastava and Auclair (1975) reported that leucine, methionine, phenylalanine, tryptophan, and valine were phagostimulatory to *Acyrthosiphon pisum*. Methionine, in particular, was reported as a phagostimulant for *M. persicae* and an essential amino acid for protein synthesis in *A. pisum* (Leckstein and Llewellyn, 1974). Omission of methionine in the present study resulted in 100% mortality, even though we added $MgSO_4 \cdot 7H_2O$ to supplement the sulphur source. Similarly, Hou (1976) found that the aster leafhopper, *Macrosteles fascifrons,* needs methionine and valine for nymphal survival. It is thus clear that four of the 10 'essential' amino acids, *viz*., methionine, tryptophan, histidine, and valine are particularly required by this leafhopper.

Only arginine and phenylalanine among the 10 'essential' amino acids can be eliminated from the diet without affecting both growth and development of *N. cincticeps* (Table 2). Seven of these amino acids could be deleted from a diet for *M. persicae* (Dadd and Krieger, 1968), and intracellular symbiotes were suggested to be involved in the biosynthesis of these omissible amino acids in the aphid (Mittler, 1971). It seems that the symbiotes in this leafhopper are unable to do so.

The effects of group deletions on growth and development of the leafhopper are shown in Table 3. The insects failed to moult and to complete their development when given 10 'essential' or 13 'non-essential' amino acids only. Jyotsna (1976) reported that the bug, *Dysdercus similis,* reared on 'essential' or 'non-essential' amino acids only could not reach maturity. Deprivation of sulphur-containing amino acids, *i.e.*, cystine, cysteine, and methionine, halted adult emergence. Turner (1971) reported that this group of amino acids is essential for the growth of the cotton aphid, *Aphis gossypii.* Omission of aspartic acid, glutamic acid, asparagine, and glutamine as a group prevented the leafhoppers from normal growth. Dadd and Krieger (1968) also found that the growth of *M. persicae* was inhibited by the deletion of these amino acids. Auclair *et al.* (1957) revealed that aspartic acid, glutamic acid, asparagine, and glutamine are found in some aphid-susceptible pea plants in relatively high amounts. Sogawa (1972) pointed out that these four amino acids could strongly enhance the sucking response of *Nilaparvata lugens.*

Deletion of γ-aminobutyric acid and dl-homoserine as a group retarded the growth of *N. cincticeps* (Table 3). Retnakaran and Beck (1968) indicated that omission of dl-homoserine did not reduce the average weight gain of nymphs in *A. pisum.* However, Srivastava and Auclair (1975) reported that this amino acid is phagostimulatory to the same species.

It is concluded that *N. cincticeps* requires most of the amino acids formulated in our diet for normal growth and development, although some particular amino acids can be omitted from the mixture without markedly detrimental effect.

Requirement for cholesterol

To determine the optimal concentration, dispersions containing various amounts of cholesterol were made up, and the final volume of each dispersion that was to be added to the basal diet remained the same, *i.e.*, 10ml/100ml diet. .

Table 4. Effect of various cholesterol concentrations on growth and deveoopment of *Nephotettix cincticeps*

Cholesterol conc. (mg/100 ml)	Initial insects tested	Cumulative nymphal mortality (%)					Nymphal period (days)	Adult emergence[b] (%)	
		1st	2nd	3dr	4th	5th			
0	38	10.6	16.9	30.8	58.6	84.5	31.8	15.5	c
2.5	34	11.8	20.6	32.8	40.2	51.3	23.8	48.7	b ab
5.0	30	13.3	23.0	35.4	48.4	48.4	22.1	51.6	ab
10.0	34	26.6	48.7	67.9	85.7	89.9	22.6	10.1	c
15.0	34	32.2	58.0	100.0	–	–	–	0.0	d
20.0	38	68.2	100.0	–	– –	–	–	0.0	d
Check[a]	44	2.3	6.3	8.8	19.3	40.8	19.8	59.2	a

a/ On rice seedlings.

b/ Means followed by the same letter are not significantly different at 5% level.

The effect of cholesterol at various concentrations on survival and development of the leafhopper is shown in Table 4. It is indicated that a range of 2.5 to 5mg/100ml is optimal for the leafhopper in terms of adult emergence. Hou and Brooks (1975) reported the same results in rearing *M. fascifrons*. Sterols, especially cholesterol, have a variety of physiological functions in insects and need to be supplied in the diet because insects generally lack the cholesterogenic pathway which has been proposed for microorganisms and vertebrates (1978). However, some aphids can be reared through many generations on chemically defined diets completely lacking sterols (Dadd and Mittler, 1966; Ehrhardt, 1968a; Srivastava and Auclair, 1971; Akey and Beck, 1972), and other hemipterous bugs can grow and mature normally on the sterol-free diets (Srivastava and Auclair, 1970). The poor adult emergence and prolonged nymphal period when fed on cholesterolfree diet has revealed that *N. cincticeps* requires a dietary supply of sterol for its normal growth. Akey and Beck (1972) found that addition of cholesterol to the diet is beneficial for growth and larviposition of *A. pisum*. Koyama (1973) was able to improve the growth of *Inazuma dorsalis* and *N. cincticeps* when cholesterol suspension was supplied separately. Our results further confirm this observation.

The 15.5% adult emergence resulted from cholesterol-free diet is possibly due to a carry-over of cholesterol from the mother and/or a lecithin compensation as mentioned by Hou and Brooks (1975). Ehrhardt (1968b) presented evidence that the associated symbiotes can synthesize sterols for the aphid, *Neomyzus circumflexus*. Clayton (1970) stated that those insects self-sufficient in sterol requirements depend upon some form of symbiotes for their supply. Houk *et al.* (1976) and Griffiths and Beck (1977) have proved that the symbiotes harboured in *A. pisum* are able to synthesize cholesterol *in vivo*. More recently, Noda *et al.* (1979) have reported that the yeastlike symbiotes may provide 24-methylenecholesterol to *L. striatellus*. Although the rice green leafhopper also harbours bacteroid and rickettsia-like symbiotes (Mitsuhashi and Kono, 1975), it seems unlikely that these microorganisms may synthesize enough sterols for their host.

The nymphs died before the 3rd or 4th instar and never reached the adult stage if fed on diet containing more than 15mg/100ml cholesterol (Table 4). We found that the diet containing 15 or 20mg/100ml cholesterol is rarely accepted by the leafhoppers. It is thought that high levels of dietary cholesterol can possibly change the physical properites of diets, so that the yound nymphs are incapable of taking up the fluid.

In the determinations of the cholesterol content of the insects by a colorimetric method, we used the color reagent, o-phthalaldehyde, instead of ferric chloride, because the former is more sensitive in reacting with cholesterol (Zlatkis and Zak, 1969). Cholesterol content of 5th instar nymphs fed on diets containing various concentrations of cholesterol is given in Table 5. We found that the more cholesterol that occurs in the diet, the more cholesterol is detected in the insect tissues. The dietary cholesterol at 5mg/100ml makes the cholesterol content of the insects similar to that of insects fed on rice seedlings. From the data in Table 4 and 5, it is reasonable to conclude that cholesterol at 5mg/100ml is suitable for feeding *N. cincticeps* on holidic diets and that the body cholesterol is built up in accordance with exogenous supply in the food.

Table 5. Cholesterol content of the 5th instar nymphs of *Nephotettix cincdiceps* fed on the diets containing various cholesterol concentrations

Dietary cholesterol (mg/100 ml)	Tissue cholesterol content (μg/mg dry wt.)
0	1.314
2.5	1.664
5.0	1.789
10.0	1.862
Check*	1.800

* Fed on rice seedlings.

ACKNOWLEDGEMENTS

This study was supported by a research grant number NSC-66B-0409-04 (11) from the National Science Council (NSC), Republic of China. We thank Ms. S. H. Lin of NSC for valuable suggestions throughout this work, and Dr. Marion A. Brooks, University of Minnesota, for her critical reading of this manuscript.

REFERENCES

1. AKEY, D. H. and S. D. BECK. 1972. Nutrition of the pea aphid, *Acyrthosiphon pisum*, requirements for trace metals, sulphur, and cholesterol. *J. Insect Physiol.*, 18, 1901–1904.
2. AUCLAIR, J. L., J. B. MALTAIS and J. J. CARTIER. 1957. Factors in resistance of peas to the pea aphid, *Acyrthosiphon pisum* (Harr.). II. Amino acids. *Can. Ent.*, 89, 457–464.
3. CLAYTON, R. B. 1970. The chemistry of nonhormonal interactions: Terpenoid compounds in ecology. In *Chemical Ecology* (E. SONDMEIMER and J. B. SIMEONE, ec.), pp. 235–280. Academic Press, New York.
4. DADD, R. H. and D. L. KRIEGER. 1968. Dietary amino acid requirements of the aphid, *Myzus persicae*. *J. Insect Physiol.*, 14, 741–764.

5 DADD, R. H. and T. E. MITTLER. 1966. Permanent culture of an aphid on a totally synthetic diet. *Experientia*, 22, 832–833.

6. DOWNER, R. G. H. 1978. Functional role of lipids in insects. In *Biochemistry of Insects* (M. ROCKSTEIN, ed.), pp. 57–92. Academic Press, New York.

7. EHRHARDT, P. 1968a. Die Wirkung verschiedener Spurenelemente auf Wachstum, Reproducktion und Symbionten von *Neomyzus circumflexus* Bucket. (Aphidae, Homoptera, Insecta) bei kundtlicher Ernahrung. *Z. Vergl. Physiol.*., 58, 47–75.
8. EHRHARDT, P. 1968b. Nachweis einer durch symbiontische Mi kroorganismen bewirkten Sterinsynthese in kunstlich ernarhten Aphiden (Homoptera, Rhynchota, Insecta). *Experientia*, 24, 82–83.
9. GRIFFITHS, G. W. and S. D. BECK. 1977. In vivo sterol biosynthesis by pea phid phid symbiotes as determined by digitonin and electron microscopic autoradiography. *Cell Tiss. Res.*, 176, 179–190.
10. HOU, F.–N. 1976. Artificial rearing and nutrition of the aster leafhopper, *Macrosteles fascifrons* (Stal) (Homoptera: Cicadellidae). Ph.D. Thesis, University of Minnesota, 160pp.
11. HOU, R. F. 1977. Dietary requirements for sugars in plant-sucking insects. *Bull. Soc. Ent. (Taiwan)*, 12, 47–53.
12. HOU, R. F. and M. A. BROOKS. 1975. Continuous rearing of the aster leafhopper, *Macrosteles fascifrons*, on a chemically defined diet. *J. Insect Physiol.*, 21, 1481–1483.
13. HOU, R. F. and M. A. BROOKS. 1977. Effects of cholesterol on growth and development of the aster leafhopper, *Macrosteles fascifrons* (Stal) (Hemiptera; Deltocephalidae). *Appl. Ent. Zool.*, 12, 248–254.
14 HOU, R. F. and L.–C. LIN. 1979. Artificial rearing of the rice green leafhopper, *Nephotettix cincticeps*, on a holidic diet. *Entomologia exp. app.*, 25, 158–164.
15. HOUK, E. J., G. W. GRIFFITHS and S. D. BECK. 1976. Lipid metabolism in the symbiotes of the pea aphid, *Acryrthosiphon pisum*. *Comp. Biochem. Physiol.*, B, 54, 427–431.
16. JYOTSNA, S. 1976. Amino acid requirements of the bug, *Dysdercus similis* Freeman (Hemiptera: Heteroptera). *Experientia*, 32, 144–145.
17. KOYAMA, K. 1973. Rearing of *Inazuma dorsalis* and *Nephotettox cincticeps* on a synthetic diet. *Jap. J. appl. Ent. Zool.*, 17, 163–166. (In Japanese)
18. KOYAMA, K. and J. MITSUHASHI. 1975. Essential amino acids for the growth of the smaller brown planthopper, *Laodelphax striatellus* Fallen (Hemiptera: Delphacidae). *Appl. Ent. Zool.*, 10, 208–215.
19. KOYAMA, K. and J. MITSUHASHI. 1979. Essential trace metals for the nymphal growth of the smaller brown planthopper, *Laodelphax striatellus* Fallen. *Jap. J. appl. Ent. Zool.*, 23, 173–177. (In Japanese)
20. LECKSTEIN, P. M. and M. LLEWELLYN. 1973. Effect of dietary amino acid on the size and alary polymorphism of *Aphis fabae*. *J. Insect Physiol.*, 19, 973–980.
21. LECKSTEIN, P. M. and M. LLEWELLYN. 1974. The role of amino acids in diet intake and selection and utilization of dipeptides by *Aphis fabae*. *J. Insect Physiol.*, *20, 877–885.*
22. MITSUHASHI, J. and Y. KONO. 1975. Intracellular microorganisms in the green rice leafhopper, *Nephotettix cincticeps* Uhler (Hemiptera: Deltocephalidae). *Appl.*

Ent. Zool., 10, 1–9.

23. MITSUHASHI, J. and K. KOYAMA. 1974. Folic acid as a dietary factor affecting the wing morph of the planthopper, *Laodelphax striatellus* (Hemiptera, Delphaciae). *Entomologia exp. appl.*, 17, 77–82.
24. MITSUHASHI, J. and K. KOYAMA. 1977. Effects of amino acids on the oviposition of the smaller brown planthopper, *Laodelphax striatellus* (Hemiptera, Delphacidae). *Entomologia exp. appl.*, 22, 156–160.
25. MITTLER, T. E. 1967. Effect on aphid feeding of dietary methionine. *Nature*, 214, 386.
26. MITTLER, T. E. 1971. Dietary amino acid requirements of the aphid *Myzus persicae* affected by antibiotic uptake. *J. Nutrition*, 101, 1023–1028.
27. MITTLER, T. E. 1976. The value of artificial feeding techniques in aphid-host-plant studies. *Symp. Biol. Hung.*, 16, 173–180.
28. NODA, H., K. WADA and T. SAITO. 1979. Sterols in *Laodelphax striatellus* with special reference to the intracellular yeastlike symbiotes as a sterol source. *J. Insect Physiol.*, 25, 443–447.
29. RETNAKARAN, A. and S. D. BECK. 1968. Amino acid requirements and sulfur amino acid metabolism in the pea aphid, *Acyrthosiphon pisum* (Harris). *Comp. Biochem. Physiol.*, 24, 611–619.
30. SAXENA, S. C. and R. GOHAIN. 1974. Quantitative variation in total cholesterol in adult and developmental stages of *Lasioderma serricorne* (F.). *Bull. Grain Tech.*, 12, 196–198.
31. SOGAWA, K. 1972. Studies on the feeding habits of the brown planthopper. III. Effects of amino acids and other compounds on the sucking response. *Jap. J. appl. Ent. Zool.*, 16, 1–7. (In Japanese)
32 SRIVASTAVA, P. N. and J. L. AUCLAIR. 1970. Responses of the large milkweed bug, *Oncopeltus fasciatus* (Hemiptera: Lygaeidae) to chemically defined diets. *Entomologia exp. appl.*, 13, 208–216.
33. SRIVASTAVA, P. N. and J. L. AUCLAIR. 1971. An improved chemically defined diet for the pea aphid, *Acyrthosiphon pisum. Ann. Ent. Soc. Am.*, 64, 474–478.
34. SRIVASTAVA, P. N. and J. L. AUCLAIR. 1975. Role of single amino acids in phagostimulation, growth, and survival of *Acrhthosiphon pisum. J. Insect Physiol.*, 21, 1865–1871.
35. TURNER, R. B. 1971, Dietary amino acid requirements of the cotton aphid, *Aphis gossypii,* the sulfur containing amino acids. *J. Insect Physiol.*, 17, 2451–2456.
36. ZAIDI, Z. S. and M. A. KHAN. 1972. Cholesterol in the haemolymph of *Dysdercus cingulatus* Fabr. (Hemiptera: Pyrrhocoidae). *Curr. Sci.*, 41, 787.
37. ZLATKIS, A. and B. ZAK. 1969. Study of a new cholesterol reagent. *Analy. Biochem.*, 29, 143–148.

水稻黑尾葉蟬對氨基酸及胆固醇之營養需求

林丙傳　　　侯豐男

國立中興大學昆蟲學研究所

本文以人工合成飼料飼育法研究水稻黑尾葉蟬（*Nephotettix cincticeps*）對氨基酸及胆固醇之營養需求。由10種必需氨基酸中略去阿金氨酸（arginine）與苯初油氨酸（phenylalanine）對此葉蟬之生長及成蟲羽化率不具無有害之效應，但除去甲硫氨酸(methionine)則引起100％若蟲死亡率。本蟲可用含有 23 種氨基酸之飼料飼育，但無法在僅含 10 種必需氨基酸之飼料存活。

以成群略去天門冬氨酸（aspartic acid)，天門冬氨酸胺（asparagine），麩氨酸（glutamic acid）與麩氨酸胺（glutamine)，或膀胱氨酸（cystine）與還原膀胱氨酸（cysteine）；或膀胱氨酸，還原膀胱氨酸與甲硫氨酸等各群，致使葉蟬發育不良。由 13 種非必需氨基酸中除去普羅林（proline）並無顯著影響葉蟬之生長。

胆固醇以5 mg／100 mℓ之濃度最適於葉蟬之生長及發育，經定量分析第五齡若蟲蟲體，發現以含有5 mg／100mℓ胆固醇之飼料所飼者，其體內胆固醇含量爲1.789 μg/mg 乾重，其與在水稻苗生長者之含量相似。