

A STUDY ON *PROXENUS HENNIA* SWINHOE AND *NYMPHULA RESPONSALIS*
WALKER AS POTENTIAL BIOLOGICAL CONTROL AGENTS OF
WATER LETTUCE (*PISTIA STRATIOTES* L.)

BY
KASNO

Thesis submitted in fulfilment of the
requirements for the degree of

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HADIAH

Certification of Examination of Thesis

We, the following members of the Thesis Examination Panel appointed by the Senate to examine the thesis entitled:-

"Study on Proxenus hennia Swinhoe and Nymphula responsalis Wlk. as potential biological control agents of Pistia stratiotes L."

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- (i) We met on Wednesday, 28 July 1982 and submitted the candidate, Encik Kasno Martosentono to an Oral Examination in accordance with the Provisions of Part (A) Clause 8(3) of the Higher Degree Programme Requirements:-

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and

- (ii) that we make the following individual recommendations:-

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Date: 28 Julai 1982

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RINGKASAN

Suatu pencarian binatang bertubuh ruas pemakan rumput air kiyambang (*Pistia stratiotes* L.) telah dilakukan di beberapa tempat dalam kawasan Negeri Pulau Pinang dan Kuala Kedah, Malaysia. Adapun matlamat dari pada pencarian tersebut adalah untuk menemukan sebarang binatang bertubuh ruas yang mampu mengawal pertumbuhan kiyambang tersebut. Di antara sebanyak binatang bertubuh ruas yang diperolehi, *Proxenus hennia* Swinhoe (Lepidoptera: Noctuidae) dan *Nymphula responsalis* Wlk. (Lepidoptera: Pyralidae) merupakan serangga yang terpilih untuk dikaji lebih lanjut.

Kajian cara hidup dan kemungkinan untuk menggunakan *P. hennia* dan *N. responsalis* sebagai makhluk hidup pengawal pertumbuhan kiyambang telah dilakukan dalam suasana makmal. Lamanya perkembangan sempurna dari pada *P. hennia* bermula dari peringkat dewasa adalah 28.53 ± 1.65 hari dengan masa peringkat ulat yang merusak 18.80 ± 1.45 hari. Dan satu putaran hidup dari pada *N. responsalis* adalah 32.92 ± 1.44 hari dengan masa peringkat ulat yang merusak 23.04 ± 1.31 hari. Dalam keadaan yang terkawal, seekor kupu *P. hennia* mampu menghasilkan telur sebanyak 620 butir dan kurang lebih 70% dari padanya mampu hidup sampai peringkat kepompong

sedang kupu *N. responsalis* mampu menghasilkan telur sebanyak 323 butir dan kurang lebih 80% dari padanya dapat berjaya sampai peringkat kepompong. Dalam suasana alamiah, jumlah ulat *P. hennia* dan *N. responsalis* amat sedikit padahal berdasar kajian makmaliah kemampuan hidupnya sampai 70% dan 80%. Hal ini menunjukkan bahawa peranan faktor penyebab kematian serangga adalah cukup besar. Oleh karena itu faktor penyebab kematian, seumpama pemangsa, parasit, dan iklim mustahak untuk segera dikaji.

Berdasar hasil kajian ini menunjukkan peranan dari pada ulat *P. hennia* dan *N. responsalis* adalah cukup tinggi dalam merusakkan kiyambang. Untuk menyebabkan kerusakan yang pantas bagi kiyambang yang bergaris tengah tajuk daun 8-10 cm memerlukan dua atau tiga ulat *P. hennia* atau *N. responsalis*, atau campuran keduanya.

Selain kemampuan merusak yang tinggi suatu makhluk hidup pengawal rumpai air harus tidak bersifat merusak tanaman berekonomi. *P. hennia* merupakan serangga yang sangat khas makan pada kiyambang dan *N. responsalis* tidak khas makan kiyambang tetapi ianya juga makan beberapa tumbuhan air bukan tanaman berekonomi misalnya: *Salvinia cucullata* Roxb. ex Bory, *Azolla pinnata* R.Br., *Lemna minor* Auct., *Eichhornia crassipes* (Mart.) Solms, *Limnocharis flava* (L.) Buch., dan tanaman darat berekonomi *Colocasia esculenta* (L.) Schott.

SUMMARY

A search for arthropods attacking water lettuce (*Pistia stratiotes* L.) was carried out, at some locations on Penang Island and Kuala Kedah, Malaysia. The objective of the search was to look for appropriate candidate biological control agents of this aquatic weed. Several arthropods were found attacking the weed, amongst them, were *Proxenus hennia* Swinhoe (Lepidoptera: Noctuidae) and *Nymphula responsalis* Wlk. (Lepidoptera: Pyralidae), both of which were selected for further studies.

The life histories and the possible use of *P. hennia* and *N. responsalis* as biological control agents of water lettuce were studied under laboratory conditions. The complete life cycle of *P. hennia* took 28.53 ± 1.65 days of which 18.80 ± 1.45 days were destructive to *Pistia stratiotes*. The complete life cycle of *N. responsalis* was 32.92 ± 1.44 days of which 23.04 ± 1.31 days were destructive. Under controlled conditions the adult female of *P. hennia* produced 620 eggs with 70 percent survival to the pupal stage and *N. responsalis* produces 320 eggs with 80 percent survival to the pupal stage. However, under field conditions the populations of *P. hennia* and *N. responsalis* were generally low: indicating

that natural factors have an important role, in their respective ability to survive. Therefore, these factors need to be studied further, e.g. predators, parasites and climate.

Studies on damage potential of the insects to water lettuce were carried out; two or three larvae of either or both of the insects cause significant decrease in the growth of water lettuce plants having a crown diameter of 8 - 10 cm.

Besides the high damage potential of the insects to the weed, *P. hennia* is monophagous on water lettuce, whilst *N. responsalis* is oligophagous, the larvae feeding on several non-economic aquatic plants, such as *Salvinia cucullata* Roxb. ex Bory, *Azolla pinnata* R.Br., *Lemna minor* Auct., *Eichhornia crassipes* (Mart.) Solms, *Limnocharis flava* (L.) Buch., as well and economic terrestrial plant *Colocasia esculenta* (L.) Schott.

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I. INTRODUCTION

Water lettuce (*Pistia stratiotes* L.) which belongs to the family Araceae is amongst the most noxious aquatic weeds in the world and is ranked third amongst the ten most important aquatic weeds in Southeast Asia (Soerjani et al., 1975).

Water lettuce is not indigenous to Asia but may have originated from East Africa (Ivens, 1968; Scheibelreichter & Apaloo, 1971). There are not many closely related economic plants to water lettuce. Amongst these are *Colocasia esculenta* (L.) Schott and *Amorphophallus campanulatus* (Roxb.) Bl. ex Dence which are edible plants and economically important.

The problems caused by water lettuce are not only limited to water management but also extend to rural health because the weed is the breeding site for *Mansonia* spp. and other mosquito larvae (Oemijati & Kadarsan, 1974).

Effort in the control of water lettuce has been mainly by mechanical and chemical means using herbicides, which because of their hazard to the environment (mainly to planktonic algae), is now greatly restricted (Anon., 1968). Conceptually, biological methods using natural enemies, have the potential to give long term control of the weed. Unfortunately such biological control techniques have not been explored much. Should biological control agents be successfully applied, they can provide long term control at comparatively

low cost with a minimum of adverse effects to the environment (Mangoendihardjo & Soerjani, 1978).

In recent times the concept of integrated pest control has found favour amongst entomologists and other people involved in pest control who would like to apply this method to aquatic weed management. However such management measures need many basic studies (like the biotic components associated with the pest) to be carried out. Biological control is an important component in integrated pest control because natural enemies are key mortality factors of pests (Sankaran, 1977), and a start in finding these enemies needs to be made if integrated control is to materialize.

Investigations on the biotic components associated with water lettuce are important basic information needed in deciding the kind of action that must be taken to control the weed. This study is a first step towards finding out.

1. promising arthropods which may be potential biological control agents of water lettuce
 2. the life histories and behaviour of the potential biological control agents
 3. ecological relationship between the biological control agents and water lettuce
 4. identification of their damage potential to water lettuce and
 5. host specificity of the biological control agents
- all of which can be used in the management of water lettuce in SouthEast Asian countries.

II. LITERATURE REVIEW

1. *Pistia stratiotes* L.

Water lettuce (*Pistia stratiotes* L.) which belongs to the family Aracae is a cup-like aquatic plant floating on slow-moving fresh water or rooting on muddy banks. It has a short stem and rosetted leaves which are green to yellow and either truncated or rounded. Their bases are thickened and very porous. There are a few white, oblique, short-penducled spathes in the centre of the rosette of leaves (Pancho & Soerjani, 1978).

The weed reproduces rapidly by both stoloniferous offshoots and seeds. Dense mats of the weed often cover the entire surface of ponds and lakes or canals and rivers of slow-moving water (Ivens, 1968; Mitchell, 1974; Pancho & Soerjani, 1978), and the plant is distributed throughout the tropics of both hemispheres (Pancho & Soerjani, 1978). It probably is not indigenous to Peninsular Malaysia (Ridley as quoted by Sculthorpe, 1967) and may have originated from East Africa (Ivens, 1968; Scheibelreichter & Apaloo, 1971).

Water lettuce is considered a serious weed (Holm *et al.*, 1969; Mitchell, 1974), and is ranked third amongst the ten most important aquatic weeds in Southeast Asia. The first and second being water hyacinth (*Eichhornia crassipes*(Mart.)

Solm) and water fern (*Salvinia molesta* D.S. Mitchell) respectively (Soerjani et al., 1975). However, according to Nemesthothy (1978), there have been some changes to this noxious plant list in Australia. Water lettuce, which was formerly classified as a noxious weed class A has now been changed to class B. This is due to the fact it is now so widely spread and it is probably beyond eradication. Weeds belong to class A need to be eradicated and class B to be managed properly, i.e. further spread prevented and small infestations eradicated.

Like other aquatic weeds, water lettuce has created serious problems as it interferes with fishing and boating, blocks canals, increase evapotranspiration, reduces aquatic production and, on the lowland areas, it competes with rice and other agricultural crops for space and nutrient.

Besides causing serious water management problems, water lettuce may act as a breeding site for other organisms such as arthropods and fish. In some cases where organisms living on water lettuce are beneficial to man, then water lettuce is likely to be useful but on the other hand when the organisms living on the plant are pests such as mosquitoes, then its status changes. It has been mentioned that water lettuce is a breeding site for mosquitoes especially of the genus *Mansonia* (Weldon & Blackburn, 1967; Holm et al., 1970; Cheong et al., 1972; Mitchell, 1974; Oemijati & Kadarsan, 1974;

Pancho & Soerjani, 1978) the adults of which are vectors of human filariasis. Water lettuce is the preferred anchoring site as well as source of oxygen for *Mansonia* larvae (Yap & Hanafi, 1976) but other mosquito species are generally not found breeding in water lettuce (Sangat & Adisoemarto, 1978).

2. Control of water lettuce

The importance of water lettuce has meant that it receives a lot of attention in any aquatic weeds management programme. When the weed occupies water bodies either alone or with other weeds, the control efforts must include management priorities based upon the degree of infestation and difficulties of controlling the weed concerned. On Kariba Lake water lettuce is given top priority (Holm et al., 1969). In general, aquatic weed control is carried out by physical, mechanical, biological, and chemical methods.

2.1. Physical control

This method currently embraces several physical principles or processes to disturb the weed growth. The commonly practised method is the use of fire to burn the slant mass after it has been dried with the help of kerosene also used as a fire starter. The ash produced can be partially utilized

as a fertilizer. Other physical methods, e.g. the use of heat, gamma ultra-shortwave and ultra-longwave rays which affect the plant growth at the atomic or molecular level (Koch & Hurle, 1978) are not practically developed yet.

2.2. Mechanical control

This control technique is to retard or kill the weed by the use of mechanical tools (weed cutter, dredger, harvester, etc.) to control weeds (Robson, 1974). In this case manual control by means of hand picking and cultural control in the irrigated rice fields are classified as mechanical control techniques. Manual control or hand picking was practised in the control of water lettuce in Egypt and Nigeria (Sculthorpe, 1967).

In cases where aquatic weeds are cut or chopped and left the chopped bio-mass, may create an increase in the biological oxygen demand (B.O.D.) and accelerate the eutrophication rate of the water body, followed by the growth of other plants or bloom of single and filamentous algae. Another method is removal of the plant mass from the water body, with or without being destroyed beforehand. Removal of the plants can be regarded as harvesting the plant if it is followed by the proper utilization of the mass. Nevertheless, in general, mechanical control is very expensive, unless proper

utilization of the removed plants appropriately follows (Soerjani, 1979). The combination of mechanical control of weeds and their utilization may reduce the cost, making it justifiable to implement (Soerjani, 1978).

2.3. Biological control

Biological control of pests is defined as the action of man in utilizing the potential of parasites, predators and pathogens in maintaining pest population at lower average than would occur in their absence (De Bach, 1964). This is called classical biological control. The wider view of biological control embraces such factors as host resistance, autosterilization, and genetic manipulation of species, or modern biological control (Price, 1972).

Biological control of pests as a technique has been applied in controlling animal pests (insects, rats, etc.) and plant pests (weeds). In principle there is little difference between the biological control of animal pests and weeds. Both involve natural enemies which act to suppress or maintain pest or potential pest species below economically injurious levels. But there are some differences in the biological control of the two pest groups. For one thing, with weed feeding organisms, a high degree of host specificity, preferably monophagy, is an absolute necessity, for

there cannot be the remotest chance that the controlling agent will develop an affinity for any plant of economic value. On the other hand, with entomophagous insects, oligophagy or even polyphagy may sometimes be advantageous, and certainly there is no hard and fast stipulation that an imported parasite or predator be narrowly specific (van den Bosch & Messenger, 1974).

Biological control of aquatic weeds has not been widely applied due to the fact that little information is available concerning the procedures and control techniques. Even though it is ecologically sound, it acts slowly and in the past has only received scant attention from investigators. Biological control techniques are considered safest amongst control methods but they also present difficulty in obtaining appropriate control agents. It usually needs a very lengthy investigation to achieve successful control (Muzik, 1970; Kassasian, 1971; Mercado, 1979) but when successful control is achieved the agents will provide perpetual control (Day, 1972).

There are only very few examples of successful biological control of weeds and most of these projects are on terrestrial weeds (De Bach, 1964; Anon, 1968; Andreas, 1976).

Recently there has been an increase in the awareness of the importance of biological control (Soerjani et al., 1977; Andreas, 1977; Soehardjan, 1979; Naito et al., 1979),

resulting in more personnel working in the field.

The procedure of biological control of weeds was described by many authors such as De Bach (1964) and Anon.(1968), and Andreas *et al.* (1976). The basic steps of the procedure are : (1) determination of the suitability of a weed for biological control; (2) exploration for natural enemies; (3) ecological observation; (4) determination of host specificity; (5) liberation and establishment; and (6) evaluation studies.

To consider the suitability of biological control of water lettuce, one has to study the status of the weed. Such as, whether water lettuce is a native or an introduced species and whether it is closely related to other plants of economic importance.

The second step in the work of biological control of weeds is the search for natural enemies. Ideally, the exploration of natural enemies of water lettuce must be carried out in the centre of origin of the weed. The reason for an exploration to be carried out in its native area is that in the process of introduction, water lettuce may have been freed from all of its natural enemies. In its native area, water lettuce, usually has natural enemies which have specific hyper-parasites and predators that can keep the population of its natural enemies low. However, by careful introduction, natural enemies of the weed shipped or carried to

other areas or countries may be established without these specific hyper-parasites and predators. When the new area is suitable for these natural enemies, some will be able to establish and may suppress the weed population. As earlier mentioned, Malaysia, or even Southeast Asia, is not where water lettuce originated.

Theoretically, any kinds of organisms which diminish the growth or reproduction of weeds may be used as biological weed control agents. Ideally, the search for weed biological control agents should encompass all organisms associated with the target weed. Phytophagous organisms that may be used as biological weed control agents include insects, mites, molluscs, fish, mammals and pathogens.

So far the potential role of plant pathogens as biological control agents of aquatic weeds have not been widely explored, in America (Freeman *et al.*, 1976; Conway & Freeman, 1976) and in Indonesia (Kasno *et al.*, 1981). *Cercospora rodmanii* F. is a fungus affecting water hyacinth and it has been established that it specifically attacks the weed in America (Freeman, 1976). A virus causing die-back of water lettuce occurs in Nigeria. The virus is transmitted by an aphid viz. *Rophalosiphum nymphaea* L. (Scheibelreiter & Apaloo, 1971; Bennett, 1973). So far the host specificity both virus and aphid has not been studied.

2.4. Chemical control

The use of herbicides to control aquatic weeds has received more attention than the other methods because its cost is usually lower and its application easier than mechanical control (Blackburn, 1974). Several herbicides have been used, i.e. glyphosate, diquat, 2,4-D amine (Bua-ngam & Mercado, 1977), atrazine, ipazine and diquat (Weldon & Blackburn, 1967; Blackburn, 1974).

The general problem in the use of herbicides is the regrowth of young weeds (Weldon & Blackburn, 1967). In the case of aquatic weeds, the multiple use of water makes it very difficult to make a general statement concerning control procedures and their efficacy. The residue of herbicides in the water may give adverse side effects on the quality of drinking water, on growth and survival of crop plants, fish and planktonic algae (Blackburn, 1974).

3. *Proxenus hennia* Swinhoe

Proxenus hennia belongs to the family Noctuidae of the order Lepidoptera. According to Maxwell-Lefroy & Howlett (1971) the general characters of Noctuidae are : antennae not dilated, hindwing with vein 8 anastomising, forewing with 5 from nearer 4 than 6. Moths with short

robust bodies, moderate antennae, which are pectinate in the males of a few, usually simple or ciliate. The forewing is stiff and narrow, the hindwing larger. Colour usually sombre.

Males are distinguished by many minor characters such as the pectination or ciliation of the antennae, the presence of scent diffusing hair-tufts on wings or legs, females are usually larger.

Proxenus hennia is probably the earlier synonym of *Caradrina hennia* Swinhoe. According to an examination of the genitalia of the adult of *Proxenus* it may be placed in the genus *Spodoptera* but this finding is as yet unpublished. The insect is distributed from North East Himalayas down to islands on the Sunda Shelf (Holloway, 1981, personal communication).

The life-history of all Noctuidae is uniform in general characters. The eggs are round and the micropyle is at the top, most are pearly white or dull green in colour, they are laid singly or in clusters on the food plant, with the clusters sometimes covered with hair. The typical larva is smooth with regularly disposed short hairs and dull brown or green colouring. Mostly larvae of Noctuidae are herbivorous.

Pupation takes place in the soil with no cocoon, but a case of consolidated earth, on the surface with a cocoon

and leaves, or more rarely, on plants in a cocoon. The imago is nocturnal. In some, reproduction is very rapid and the number of eggs laid totals hundreds and in some cases thousands.

Some species of *Proxenus* found in South Africa (Janse, 1940). *P. hennia* is reported from Malaysia and Indonesia (Mangoendihardjo & Nasroh, 1976).

4. *Nymphula responsalis* Wlk.

The insect belongs to the family Pyralidae of the order Lepidoptera. In India, *Nymphula* is universally distributed. The known larvae are aquatic, the body having tubular gills with tracheal tubes in them, O₂ being apparently obtained by transpiration through the thin gill, walls. Spiracles though present are closed and functionless. The larvae rolls a leaf and lives within being able to breathe in water or air.

Nymphula responsalis is found feeding on *Salvinia* sp. in India (Vergis, 1980).

In Kerala, the female of *N. responsalis* lays 50 - 100 eggs singly on the leaves of *Salvinia* sp., and these hatch in 5-6 days. The larval and pupal stage last 21 and 5-6 days respectively (Vergis, 1980).

III. MATERIALS AND METHODS

1. Study area

Plant materials and insects used in this study were collected from ponds, irrigation canals and drains of shoreland Sungai Pinang, Ginting, Telok Kumbar, Bayan Lepas areas of southwestern coastal Penang Island and at Nibong Tebal a district on the western mainland of Penang (Figure 1).

Sungai Pinang is located at the western side of Penang Island. Its northern and western part areas are forestland. The western part is a lowland with settlements bordering rubber and coconut plantations as well as rice fields. The area has been recognized as a mosquito breeding site. Water lettuce can be found on irrigation canals, fish ponds and drainage canals. The collection site was an unmanaged fish pond of Sungai Rusa, where water lettuce occurs throughout the year.

The southern part of Sungai Pinang borders with Ginting and south of Ginting is Telok Kumbar. These areas are more or less similar to Sungai Pinang in their agro-ecosystems. Collection was carried out at

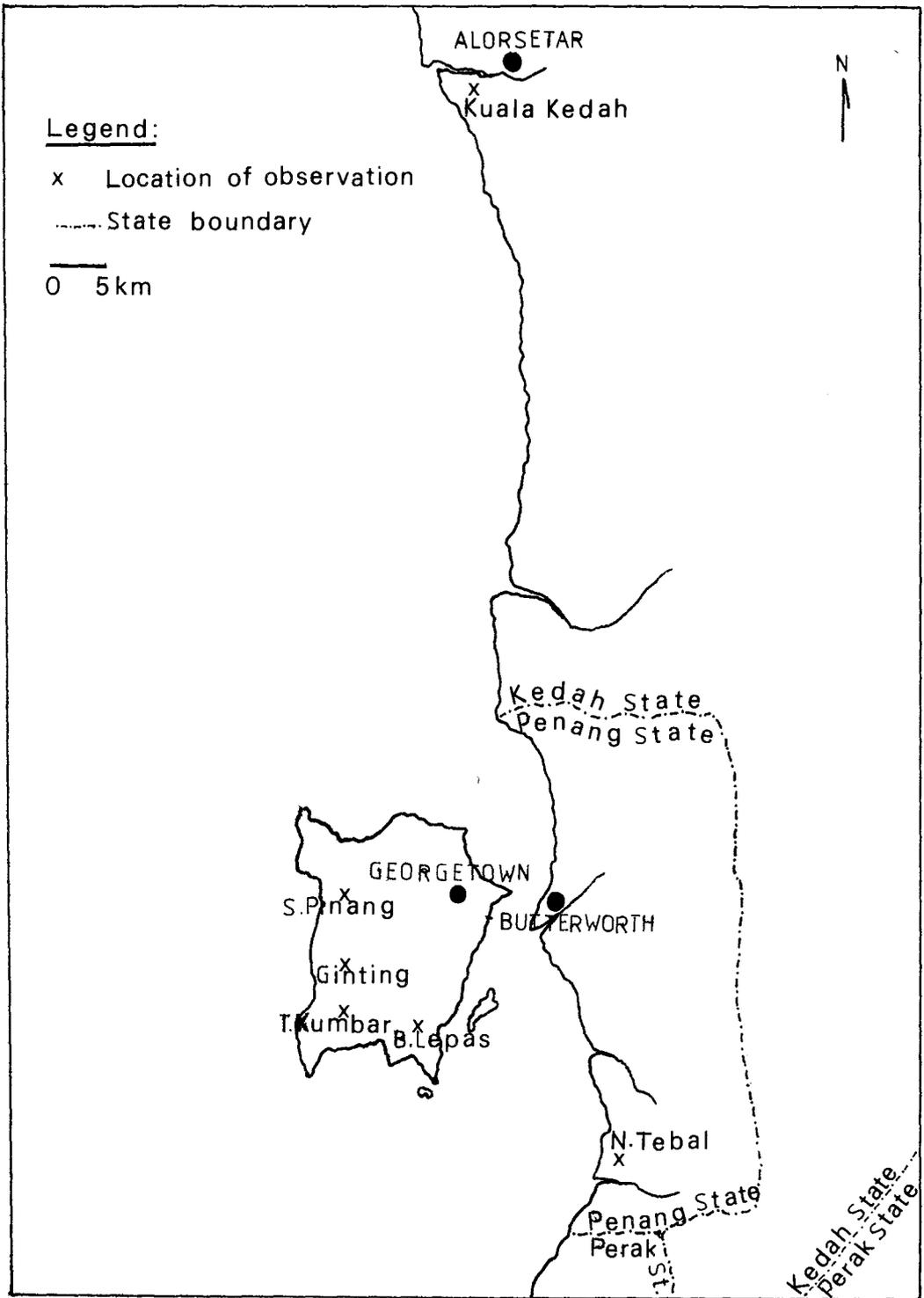


Figure 1. Study area

the main irrigation canals of the area.

Bayan Lepas is located at the southwestern tip of Penang Island, it borders Telok Kumbar on the western side and Bayan Baru (a satellite township) in its eastern side. Water lettuce occurs throughout the year in this area as well. The expansion of human settlement, industrial site and airport has reduced the areas infested by water lettuce considerably. Water lettuce occurring in the drains between rows of vegetable garden facing the entrance to the International Airport at Bayan Lepas was the source of our collection.

Nibong Tebal is located about 40 km south of Butterworth. The collection site was a 2 km long canal between rubber plantations and settlements, and it is about 5 km from the sea.

2. Field studies

Insects and plant materials used in this study were collected at some localities of Penang Island State mainly in the western coastal areas of the island and in Kuala Kedah, Malaysia. Sampling was done regularly using wooden frame sample plots measuring 15 x 30 cm² and aimed at observing the kinds of natural enemies and their potentials in controlling water lettuce.

The samples were then removed and brought back to the laboratory at Glugor in plastic bags for further observation.

2.1. Observation on the arthropods population of water lettuce

Before detailed investigations were carried out on the role of *P. hennia* and *N. responsalis*, casual observation were made on the arthropods inhabiting water lettuce. This was done by walking on the site of canals or ponds with pauses to watch arthropods infesting water lettuce. The presence of grasshoppers, leafhoppers and worms were visible. Some water lettuce was collected, for close observation of small arthropods. The occurrence of arthropods was recorded subjectively, by scoring each occurrence of the data for which are presented in tables 1 and 2.

2.2. Observation on population densities of *P. hennia* and *N. responsalis*

Population trends of *P. hennia* Swinhoe and *N. responsalis* Wlk. were determined from regular samplings at Bayan Lepas and Sungai Pinang at three-week intervals during January - March 1977. Since it was suspected that the insects preferred plants occurring in open rather than in shaded habitats, samples were collected from each

of two strata: Stratum I where water lettuce occurred as a single or most dominant species; Stratum II where water lettuce grew with other taller plants over shading it. Three or four samples were collected every three week period. The results are presented in tables 3, 4 and appendix 1.

Besides population density, estimates of larval density/damage relationship for both *P. hennia*, *N. responsalis* were made. The results of this observation are presented in table 5 and appendix 2.

2.3. Host specificity of *P. hennia* and *N. responsalis*

The objective of this study was to find out the alternate hosts of these insects by casual observations on plants growing within a radius of 10 m of water lettuce sites. The results are presented on sections 1.2.1 and 1.2.2 in a latter section.

2.4. Observation on natural enemies of *P. hennia* and *N. responsalis*

The aim was to investigate the occurrence of the natural enemies mainly parasites of both *P. hennia* and *N. responsalis*. Insects were collected from the field and reared in the laboratory in plastic containers covered with nylon mesh until they emerged as adults. Trapping the egg parasite was done by placing one night old eggs of the insects in the field off Ginting. The fresh eggs were placed on marked single water

lettuce plants among infested water lettuce and trap sites were replicated 20 times. The egg-traps were then collected and brought back to the laboratory after about 24 hours in the field. They were then reared and degrees of parasitism recorded after emergence of adults. The results of these observations are presented in table 6 (Chapter IV).

3. Laboratory Studies

The purpose of the laboratory work was to confirm the field observations. The collected water lettuce plants with insects feeding on them were then brought back to the laboratory for further observation. Insect rearing was done in round plastic containers containing water lettuce in screened cages. The size of the container was 5 cm in height and 15 cm in diameter. The size of the cage was 40 cm in height, 40 cm long and 40 cm wide. Insects used in the study were the first or second generation which were produced in laboratory cultures. Water lettuce plants were grown in outdoor plastic ponds having dimensions of 15 cm in height, 4 m long and 1.5 m wide. The insects were cultured on water lettuce growing in indoor screened cages. The laboratory conditions were 27°-31°C, and 48-67% of relative humidity. The laboratory work was limited to the study of the life histories, behaviour, host

ranges, and damage potentials of *P. hennia* and *N. responsalis* to water lettuce.

3.1. Life history and habit

Insects were released onto water lettuce plants grown in plastic containers containing tap water and initially, ten pairs of adults were released into each culture cage to allow the females to lay their eggs. Groups of eggs were taken on the next morning, and were then placed separately in plastic jars to allow them to hatch.

After hatching, five of the newly hatched larvae were transferred to round plastic jars having a dimension of 5 cm in height and 15 cm in diameter and they were then allowed to mature. This was replicated 10 times. Water lettuce used as the natural food for the insects was changed once every three or four days. Records were made on the larvae until they started pupating. When the larvae reached the prepupal stage, they were transferred into petridishes lined with two layers of wet filter paper to provide moisture. The mature larvae and pieces of leaf were put on the petridishes and they were allowed to pupate and emerge as adults.

3.2. Egg productivity

Twelve pairs of adults of the insects were allowed to mate, then each pair was placed in a round plastic jar containing water lettuce in 2 cm depth of tap water, the size of the jar was 13 cm in diameter and 7 cm in height. A solution containing 1% honey was spotted with fine brush on the water lettuce leaves to provide food for the adults. The jar was covered with white nylon mesh. The following morning the pair was placed in a new jar similar to the above and this was repeated till the death of the female moth. The measure of fecundity is presented in table 7 and figure 13.

3.3. Survival rate

The survival rate of eggs of *P. hennia* and *N. responsalis* was observed under laboratory conditions. Some egg groups found on the leaf of water lettuce were taken and transferred moistened petridish as so that each petridish contained a group of eggs covered with a glass dish. The eggs were counted under a binocular microscope. The number of eggs which failed to hatch were then recorded after hatching had completed. The results of egg mortality are presented in appendices 5 and 11.

Fifty eggs of *P. hennia* and *N. responsalis* were released on screen-caged water lettuce floating in a concrete pond. The cage measured 50 cm wide, 50 cm long, and 80 cm high. Eggs used for the test were about 10-12 hours old. Survival of *P. hennia* was recorded on the 5th, 8th, 12th, 16th, 20th and 22nd day and that of *N. responsalis* on the 6th, 10th, 14th, 18th, 22nd and 26th after the release of eggs. There were 24 cages and four of them were sampled at each time and, once sampled cages were removed from the experiment. The results are presented in tables 11 and 15, figures 14 and 18, and appendices 7 and 12.

3.4. Larval instars

The number of larval instars of *P. hennia* was established by measuring the size of head capsule of the larvae using Dyar's law (Chapman, 1939). Larvae were reared in groups in a plastic basin. Water lettuce served as their natural food. This was changed every three or four days. Ten larvae were sampled every day. They were then preserved in solution containing 4% of formaline. Each head capsule was then dissected and measured under a microscope using a calibrated micrometer (Figure 15).

Beside the method described above, continued individual observation was done on 15 larvae of *P. hennia* and 30 larvae of *N. responsalis*. Individual larvae were reared on small water lettuce plants clipped to a plastic petridish to facilitate subsequent observations. Clipping was done as follows: (1) A hole was made in each plastic dish with a red hot nail. (2) The roots of small water lettuce plants were supported by winding them with thread. The supported roots were then inserted into the hole in the dish and fixed in place with melted wax. (3) The clipped water lettuce was then floated in containers containing pond water. (4) Newly hatched larvae were then released onto the plant to allow them to grow (Figure 2). Observations were made every day till the larvae showed signs of pupating. The results are presented in tables 12 and 16, and appendices 8, 9 and 13.

3.5. Effect of crowding on larval behaviour of *P. hennia*

The purpose of this observation was to study cannibalism of *P. hennia*. This study was carried out by rearing a variable number of larvae in small glass tubes. The numbers were 5, 10, 15, 20, 25 and 30 newly

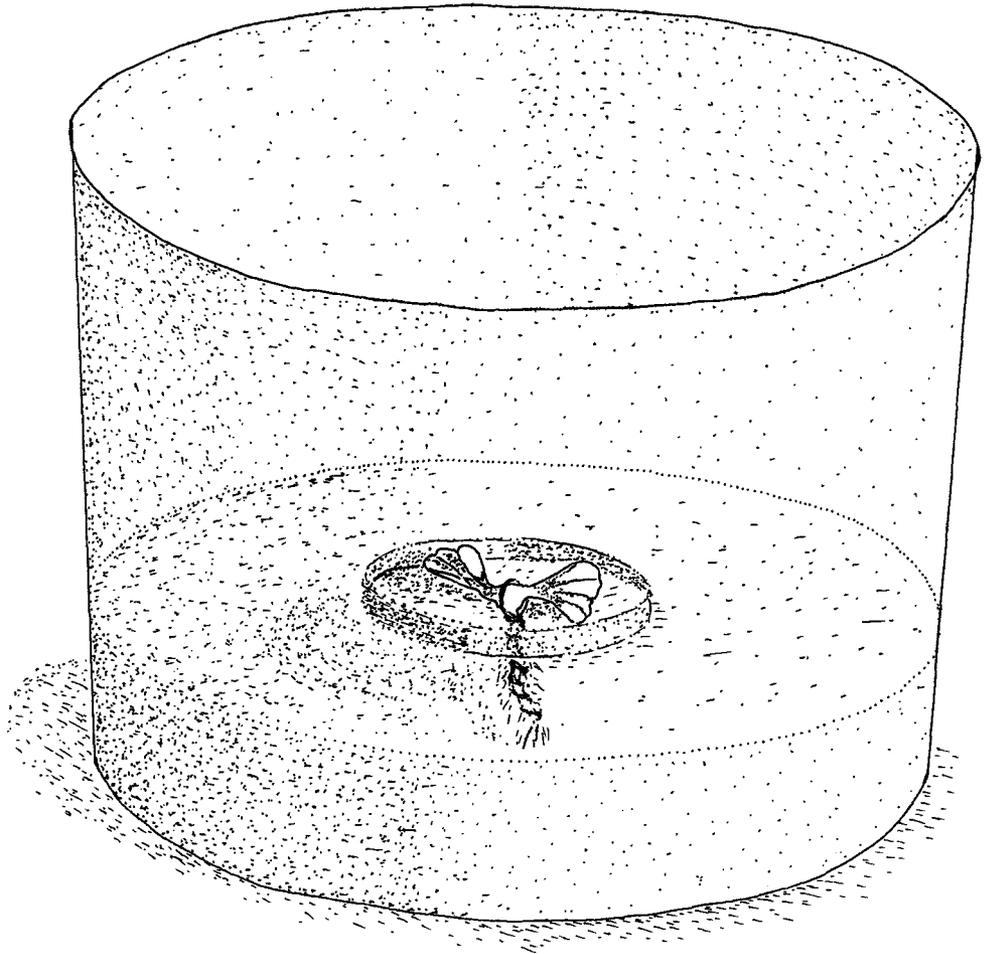


Figure 2. Single larvae of *P. hennia* or *N. responsalis* reared on small water lettuce to study the larval development