

STUDIES ON SOME BIOLOGICAL AND
ECOLOGICAL ASPECTS OF PAPILIO POLYTES L.
(LEPIDOPTERA: PAPILIONIDAE) AND PAPILIO
DEMOLEUS L. (LEPIDOPTERA: PAPILIONIDAE)
ON RUTACEOUS HOST PLANTS

S U W A R N O

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PAPILIO DEMOLEUS L. (LEPIDOPTERA: PAPILIONIDAE)
ON RUTACEOUS HOST PLANTS

by

S U W A R N O

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KAJIAN TERHADAP BEBERAPA ASPEK BIOLOGI DAN EKOLOGI
***PAPILIO POLYTES* L. (LEPIDOPTERA: PAPILIONIDAE) DAN**
***PAPILIO DEMOLEUS* L. (LEPIDOPTERA: PAPILIONIDAE)**
PADA TUMBUHAN PERUMAH RUTECEAE

ABSTRAK

Kajian biologi dan ekologi ke atas *Papilio demoleus* L. dan *Papilio polytes* L. (Lepidoptera: Papilionidae) telah dijalankan di makmal dan di lapangan. Kesesuaian empat spesies tumbuhan [limau nipis, *Citrus aurantifolia* [(Christm.) Swingle]; limau purut, *Citrus hystrix* DC.; limau madu, *Citrus reticulata* Blanco dan daun kari, *Murraya koenigii* (L.) Sprengle] sebagai tumbuhan perumah rama-rama *P. polytes* dibandingkan dimakmal dan didapati *Papilio polytes* betina lebih memilih *C. reticulata* dan *C. aurantifolia* ($P < 0.01$) untuk bertelur. *Murraya koenigii* paling tidak digemari. Bahagian bawah daun *C. reticulata* dan *C. aurantifolia* adalah lebih sesuai berbanding *C. hystrix* dan *M. koenigii* ($F_{3,36} = 182.25$, $P < 0.01$). Larva instar yang muda (instar 1-3) lebih memilih makanannya berbanding larva yang lebih tua (instar 5). Kemampuan penghadaman dan pertumbuhan larva *P. polytes* lebih tinggi apabila dipelihara pada *Citrus* spp. berbanding pada *M. koenigii* dan kitar hidup paling pendek di perhatikan pada *C. reticulata* ($F_{3,116} = 4.25$; $P < 0.01$). Badan pupa dan dewasa serta rentang kepak dewasa adalah paling panjang ($F_{\text{pupa}} = 4.99$; $P < 0.01$; $F_{\text{panjang badan}} = 3.47$; $P < 0.05$; $F_{\text{rentang kepak}} = 2.90$; $P < 0.05$) pada *C. hystrix*. Kajian jadual hayat dan dinamik populasi *P. demoleus* dan *P. polytes* di lapangan, dilakukan selama setahun merangkumi musim kering (DS), musim lembap sekunder (SWS) dan musim lembap primer (PWS). Secara amnya *P. polytes* lebih bermandiri berbanding *P. demoleus*. Kedua-dua spesies bermandiri paling baik ketika PWS

(3.20% dan 3.93%, masing-masing) dan paling buruk ketika DS (1.74% dan 3.08%, masing-masing). Kemandirian *Papilio demoleus* lebih rendah dari *P. polytes* di setiap musim. Nilai-*k* bagi larva instar kelima adalah tertinggi (1.32 untuk *P. demoleus* dan 0.99 untuk *P. polytes*) dan pemangsa merupakan kunci mortaliti bagi peringkat tidak matang kedua-dua spesies. Perkembangan peringkat tidak matang kedua-dua spesies tidak dipengaruhi oleh musim, kecuali pada telur dan peringkat larva instar pertama (ujian Kruskal-Wallis, $P = 0.05$). Walau bagaimanapun, kelimpahan mereka adalah berbeza antara musim dan terendah ketika DS. Labah-labah *Oxyopes quadrifasciatus* merupakan ejen mortaliti paling penting bagi larva instar muda dan *Sycanus dichotomus* ialah spesies pemangsa utama bagi larva instar yang lebih tua. Bilangan telur dan larva yang diserang sangat bergantung kepada kepadatan kedua-duanya. Faktor-faktor abiotik menunjukkan pengaruh yang lemah terhadap kelimpahan peringkat tidak matang pada PWS dan boleh diabaikan pada DS dan SWS.

**STUDIES ON SOME BIOLOGICAL AND ECOLOGICAL ASPECTS OF
PAPILIO POLYTES L. (LEPIDOPTERA: PAPILIONIDAE) AND
PAPILIO DEMOLEUS L. (LEPIDOPTERA: PAPILIONIDAE)
ON RUTACEOUS HOST PLANTS**

ABSTRACT

Studies on the biological and ecological aspects of *Papilio demoleus* L. and *Papilio polytes* L. (Lepidoptera: Papilionidae) were carried out both in the laboratory and in the field. The suitability of four rutaceous species [*Citrus aurantifolia* [(Christm) Swingle], *Citrus hystrix* DC., *Citrus reticulata* Blanco and *Murraya koenigii* (L.) Sprengle] as host plants for *P. polytes* was compared. Females of *P. polytes* preferred *C. reticulata* and *C. aurantifolia* ($P < 0.01$) as oviposition sites. *Murraya koenigii* was the least preferred. More eggs were oviposited on the undersides of leaves of *C. reticulata* and *C. aurantifolia* ($F_{3,36} = 182.25$, $P < 0.01$). Younger instar larvae (instar 1-3) displayed higher selectivity for food. The larval performance of *P. polytes* was better when reared on *Citrus* spp. than on *M. koenigii*. The shortest life cycle of *P. polytes* was observed on *C. reticulata* ($F_{3,116} = 4.25$; $P < 0.01$). The pupal and adult body lengths as well as adult wing span were the longest ($F_{\text{pupa}} = 4.99$ and $F_{\text{body length}} = 3.47$; $F_{\text{wingspan}} = 2.90$; $P < 0.05$) on *C. hystrix*. Studies on life table and population dynamics of *P. demoleus* and *P. polytes* in the field were carried out for a year, in the dry season (DS), secondary wet season (SWS) and primary wet season (PWS). Survival of both species was the highest in the PWS (3.20% and 3.93%, respectively) and the lowest in the DS (1.74% and 3.08%, respectively). The survivorship of *P. demoleus* was lower than *P. polytes* in all seasons. The k -value of the fifth instar larva of both species was the highest. Except

in the egg and first larval instar (Kruskal-Wallis test, $P = 0.05$), the development of immature stages was not influenced by season. Their abundance however, varied among seasons and was the lowest in the DS. Predators *Oxyopes quadrifasciatus* was the most important mortality agent for young larval instars and *Sycanus dichotomus* for older larval instars. The number of parasitized and predated eggs as well as larvae depended strongly on the density of the immature stages. The abiotic factor weakly influenced the abundance of the immatures during the PWS and considered negligible during other seasons.

CHAPTER 1

GENERAL INTRODUCTION

1.1. Research background

The common mormon butterfly, *Papilio polytes* L. (Lepidoptera: Papilionidae) and the lime butterfly *Papilio demoleus* L. (Lepidoptera: Papilionidae) are tropical or subtropical papilionid butterflies. *Papilio polytes* is distributed from Southeast Asia to Southwestern Island of Japan (Corbet & Pendlebury, 1992; Nakayama et al., 2003; Nakayama & Honda, 2004), meanwhile *P. demoleus* is widely distributed in Southern Asia, Australia, Taiwan, Hainan, New Guinea, and Southeast Asia (Corbet & Pendlebury, 1992). Both *P. polytes* and *P. demoleus* are found throughout the Malaysian Peninsula, including Kedah, Langkawi, and Tioman Island (Corbet & Pendlebury, 1992).

In the past, the larvae of *P. polytes* and *P. demoleus* were only known to feed on various genera of rutaceous plants, such as *Aegle*, *Atalantia*, *Evodia*, *Glycosmis*, *Murraya*, *Poncirus*, *Toddalia*, *Triphasia* and *Zanthoxylum* in forests (Corbet & Pendlebury, 1992; Suguru & Hurao, 2000). However, more recently, both butterflies have been reported to have adapted to the urban/suburban environment and as a result they are common in neighbourhoods with gardens and villages where citrus is grown (Corbet & Pendlebury, 1992; Otsuka, 2001).

Citrus is an important tropical fruit, it has been traditionally cultivated in home gardens and modern plantations (Saljoqi et al., 2006) and is used in food industries, chemical and pharmaceutical industries, perfumes, cosmetics and aromatherapy (<http://users.kymp.net/citruspages/oils.html>). A number of insect pests attack citrus plants both in the nurseries as well as in the orchards inflicting heavy

economic losses. Some of the most serious pests of citrus includes: citrus caterpillars (*P. demoleus* and *P. polytes*), citrus psylla [*Diaphorina citri* Kuwayama (Hemiptera: Psyllidae)], citrus leaf miner [*Phyllocnistis citrella* Stainton (Lepidoptera: Gracillariidae)], citrus red scale [*Aonidiella aurantii* Maskell (Homoptera: Diaspididae)] and citrus whitefly [*Dialeurodes citri* Ashmnead (Hemiptera: Aleyrodidae)] (Atwal, 1976 cited by Saljoqi et al., 2006).

The lime butterfly, *P. demoleus* is the most notorious and destructive pest of citrus plantation throughout the world (Saljoqi et al., 2006). According to Badawi (1981), *P. demoleus* is a serious pest on citrus plantation in Saudi Arabia. This butterfly species is recognized also as a major pest throughout most of the Old World range, causing significant economic losses (Pakistan Agriculture Research Council, 2003; Malaysian Tropical Fruit Information System, 2004; Agribusiness Information Centre of India, 2005). Furthermore, Munir (2004) reported that *P. demoleus* and *P. polytes* are serious pest on many species of citrus in Pakistan.

Host-plant selection by the herbivore insect involves not only choosing the right species of plant, but also selecting an individual plant within that species that is, or will be, suitable for feeding, survival and development of immature stages. The importance of selection within the species is clearly indicated by a field study on butterfly oviposition (Bernays & Chapman, 1994). The factors determining differences in the establishment of different species of *Papilio* on different plants, according to Saxena (1969) cited by Saxena and Goyal (1978), can be arranged under the following six categories; (1) Orientation, determining arrival of the insect on a plant, (2) Oviposition, (3) Feeding, (4) Utilization of ingested food, (5) Growth, and (6) Egg-production. Dethier (1982) stated that most herbivorous insect species accept only a limited number of plant species as hosts. Their behavioral decisions to accept

or reject a particular plant species as oviposition substrate or food source are based largely on the perception of the chemical profile of the plant under evaluation.

Previous studies have reported that *P. demoleus* preferred *Citrus reticulata* Blanco and *Citrus aurantium* L. (Rafi et al., 1999a), *C. reticulata* and *Citrus aurantifolia* (Christm.) Swingle (Munir et al., 2005) and *Poncirus trifoliata* (L.) Raf. and *C. reticulata* (Saljoqi et al., 2006) for oviposition. Whereas, *P. polytes* preferred *Toddalia asiatica* Lam. and *Citrus depressa* Hayata for the same purpose (Nakayama et al., 2002). Munir et al. (2005) reported that the larvae of *P. demoleus* that were fed on *C. reticulata* and *C. aurantifolia* had shorter developmental period and higher survivorship compared to those fed on the other citrus species. Herbivore preferences and performance are strongly affected by host plant traits, natural enemies, and the tri-trophic interaction between plant, herbivores and their natural enemies (Espírito-Santo & Fernandes, 2002). Plants may influence herbivores indirectly, by controlling their growth and development, increasing their susceptibility to parasitoids and predators (Benrey & Denno, 1997), or even by producing volatile substances to attract natural enemies (Mc Call et al., 1993).

Natural enemies are very important component in the population dynamics of insect species. They influence total insect numbers and their impact is related to the density of their prey or their host. Their action is density dependent (Pedigo, 2002). Hirosie et al. (1980) and Watanabe (1981) reported that a population of *Papilio xuthus* L. (Lepidoptera: Papilionidae) fluctuated throughout the year but an outbreak never occurred. They found that natural enemies played a significant role in controlling the butterfly population. On the other hand, Diez et al. (2006) reported that the citrus leaf miner, *P. citrella* and its parasitoids exhibited similar levels of population fluctuation throughout their entire study period.

Climatic factors are very important factors because they influence the insect population fluctuations. The relationship between these abiotic factors and insect abundance could be positive or negative (Peng et al., 2006).

A life table study is one of the most important components to understanding the population dynamics of butterflies (Carrey, 1993 *cited by* Southwood & Henderson, 2000; Xiushan et al., 2006). Such studies help to assess mortality rates at various stages in the life cycle (Xiushan et al., 2006). At the same time, the life tables are necessary to identify the key factors that influence the population size (Chi & Yang, 2003; Choate & Rieske, 2005; Yu et al., 2005; Chi & Su, 2006; Stefanescu et al., 2006; Xiushan et al., 2006; Yang & Chi, 2006).

Based on field observations at several citrus plantations in Penang, Taiping, and Kedah it is established that both *P. demoleus* and *P. polytes* have potentially become serious pests in local citrus nurseries and orchards. Meanwhile, *P. polytes* is also found on *Murayya koenigii* (L.) Sprengle in the neighbourhoods and at the edges of forests. In the past, several studies have been conducted on the biology and ecological aspects of *P. demoleus* (Yunus & Munir, 1972; Badawi, 1981; Matsumoto & Noerdjito, 1996; Rafi et al., 1999a; 1999b; 2001; Matsumoto, 2002; Munir et al., 2005; Homziak & Homziak, 2006; Saljoqi et al., 2006), but such studies on *P. polytes* are rather limited (Nakayama et al., 2002; Munir, 2004). A thorough knowledge of the pest and its natural enemies is essential in developing a control strategy that is effective. This thesis presents information on biological aspects, such as oviposition preference, life cycle, feeding preferences, feeding behaviour and larval nutrition indices of *P. polytes* on selected rutaceous plants and the ecological aspects, such as life table, population dynamics and natural enemies of *P. demoleus* and *P. polytes* reared on *C. reticulata* in different seasons.

To develop of efficient strategies for controlling *P. demoleus* and *P. polytes* will require the knowledge of its biological relationships with various host plants and the ecological aspects of both butterflies. Based on the studies of its biological and ecological aspects, suitable hosts for both *Papilio* species will be found.

1.2. Research objectives

The general objectives are to observe some biological aspects of *P. polytes* in relation to three commercial rutaceous host plants (*Citrus aurantifolia*, *C. hystrix*, and *C. reticulata*) and a partially domesticated rutaceous species, *M. koenigii* (curry leaf), and to compare some ecological aspects of *P. polytes* and *P. demoleus* in different seasons. The specific objectives of this research are as follows:

- (1). To determine the growth and life cycle of *P. polytes* on the four rutaceous host plants.
- (2). To investigate the oviposition preference, feeding preference and feeding behaviour of *P. polytes* reared on four rutaceous host plants.
- (3). To determine the efficiency of food utilization by *P. polytes* larvae on the four rutaceous host plants.
- (5). To construct and compare the life tables of *P. polytes* and *P. demoleus* in different seasons.
- (6). To study the abundance of both *P. polytes* and *P. demoleus* and their relationship to natural enemies in different seasons.
- (7). To determine the effects of climatic factors on the abundance of immature stages of *P. polytes* and *P. demoleus*.

CHAPTER 2

LITERATURE REVIEW

2.1. Common mormon butterfly (*Papilio polytes* L.)

2.1.1. Biology of *P. polytes*

Papilio polytes commonly known as swallowtail butterfly belongs to the family Papilionidae and the subfamily Papilioninae. The male of this species is monomorphic while the female is polymorphic (Corbet & Pendlebury, 1992; Otsuka, 2001; Salmah et al., 2002; Munir, 2004). The wings of male *P. polytes* are black and yellowish white. There are a few small pale yellow spots on the distal margin of the forewing (Corbet & Pendlebury, 1992; Otsuka, 2001). The males are smaller in size than the females (Corbet & Pendlebury, 1992). For instance Munir (2004) reported that the average wing span of male *P. polytes polytes* is 77.6 ± 1.57 mm which is shorter than the female wing span of 89.2 ± 3.53 mm.

The female *P. polytes* has four forms which are *cyrus*, *polytes*, *romulus* and *theseus* (Clarke & Sheppard, 1971). The *cyrus* form is similar to the male, differing in that it always has strongly marked red crescents. The three other forms have a periphery of orange-red spots on the hind wings and sometimes it also bears a small, white spot in the middle of the hind wing (Clarke & Sheppard, 1971; Otsuka, 2001). The form *theseus* and *polytes* are mimics of the unpalatable papilionid, *Pachliopta aristolochiae* Fabr. (Lepidoptera: Papilionidae) whereas the form *romulus* mimics the other unpalatable papilionid, *Pachliopta hector* L. (Lepidoptera: Papilionidae). According to Corbet and Pendlebury (1992) the forms *cyrus* and *polytes* can be found in the Malaysia Peninsula. The wingspan of females (all forms) ranged from 90-115 mm (http://en.wikipedia.org/wiki/Papilio_polytes).

Similar to other lepidopterans, *P. polytes* undergoes complete metamorphosis. The stages of the cycle include egg, larva, pupa, and adult (Corbet & Pendlebury, 1992; Pedigo, 2002; Johnson & Triplehorn, 2005). The egg is spherical, yellowish, 1.00-1.08 mm in diameter and 0.96-1.00 long (Salmah et al., 2002). Munir (2004), described the eggs of *P. polytes* as light pale yellow to slightly shiny with rough surface of corion. It is spherical in shape with a diameter of 1.056 mm. The female of *P. polytes* lays its eggs singly on the tip or on the lower surface of the leaf of a host plant (Salmah et al., 2002; Munir, 2004). After three to four days the eggs hatch and first instar larvae emerge. A larva comes out from an egg by making a small hole. The first instar larva is very small (Salmah et al., 2002). According to Munir (2004) the body length of the first instar larva of *P. polytes f. polytes* is 4.5 mm.

There are five larval instars in *P. polytes*. The first and second instars are dark grey to dark brown in colour and there are small spines on the dorsolateral sides of the bodies. The head size of both instars is small, glossy pale green and reddish towards the mandibles (Salmah et al., 2002; Munir, 2004). The first instar larva begins feeding by consuming its egg shell. The egg shell is believed to contain plenty of nutrients and is probably necessary for the larval growth. Later instars consume the leaves of the host plant and grow very rapidly during the process (Corbet & Pendlebury, 1992).

The third larval instar has dark orange colour on the dorsal side of its thorax with short spines. The larva appears to be slightly greasy. In the fourth instar, the dorsal side of the abdomen becomes greenish grey, whereas the dorsal side of the thorax is blackish grey (Salmah et al., 2002). In swallowtail caterpillars, the first three instars often resemble bird droppings, while the last two instars look very much like snakes (Corbet & Pendlebury, 1992; Otsuka, 2001). The skin of the fourth instar

becomes velvety, blackish green, and the anterior portion is broader than the abdominal segments. The dorsal line is indistinct while the other characteristics are similar to the third instar (Munir, 2004). The final instar is quite different from the previous four instars in shape, length, colour, and activity. The larva becomes green with three black lines appearing on the lateral part of the abdomen (Salmah et al. 2002; Munir, 2004). The larval duration of *P. polytes* is 17 - 19 days when reared on *Clausena excavata* Burm. and 14 - 19 when reared on *Citrus sinensis* (L.) Osbeck (Salmah et al., 2002). Munir (2004) found that the larval duration of *P. polytes f. polytes* varied between 14 - 17 days, depending on the temperature and host plants.

The pupal stage is a non-feeding period, during which the adult is formed in the chrysalis. The colour of *P. polytes* pupa is green or brown. Pupal colour in *P. polytes*, *P. demoleus*, *P. xuthus* L. and *P. polyxenes* Fabr. depends upon variations in environmental conditions (Smith, 1978; Hiraga, 2006). In the chrysalis, the body of the caterpillar transforms into an adult butterfly. The wings are now fully formed. Antennae and the chewing mouthparts of the caterpillar are also transformed into the sucking mouthparts of the butterfly in the chrysalis (Corbet & Pendlebury, 1992; Pedigo, 2002; Johnson & Triplehorn, 2005). Pupal duration lasts for 11-13 days under 24°C and 63-68% relative humidity (Salmah et al., 2002).

Many species of butterflies in nature exhibit pupal colour polyphenism to mimic the colours of a variety of pupation sites as a survival mechanism against insectivorous birds (Hassel et al., 1998; van Dyck et al., 1998; Hiraga, 2006). Bird predation is the major mortality factor of the pupal stage of the swallowtail butterflies in the field (Stefanescu, 2004).

2.1.2. Ecology of *P. polytes*

Members of the genus *Papilio* are the largest in number in the subfamily Papilioninae. There are 14 species of *Papilio* distributed throughout the Malaysian peninsula (Corbet & Pendlebury, 1992). In general, the sexes are similar in colour, except that females of *Papilio memnon* L. and *P. polytes* are polymorphic (Corbet & Pendlebury, 1992; Otsuka, 2001).

Papilio polytes feeds on rutaceous plants in Southeast Asia and Southwestern Island of Japan (Nakayama et al., 2002; 2003; Murakami et al., 2003; Nakayama & Honda, 2004; Omura & Honda, 2005). According to Corbet and Pendlebury (1992) *P. polytes* is found throughout the Malaysian peninsular, including Kedawi and Tioman Island, but it is strictly confined to the plains. This species is distributed from Sri Langka, India, China and through to the Archipelago of the Molucas and the Lesser Sunda Islands (Figure 2.1).

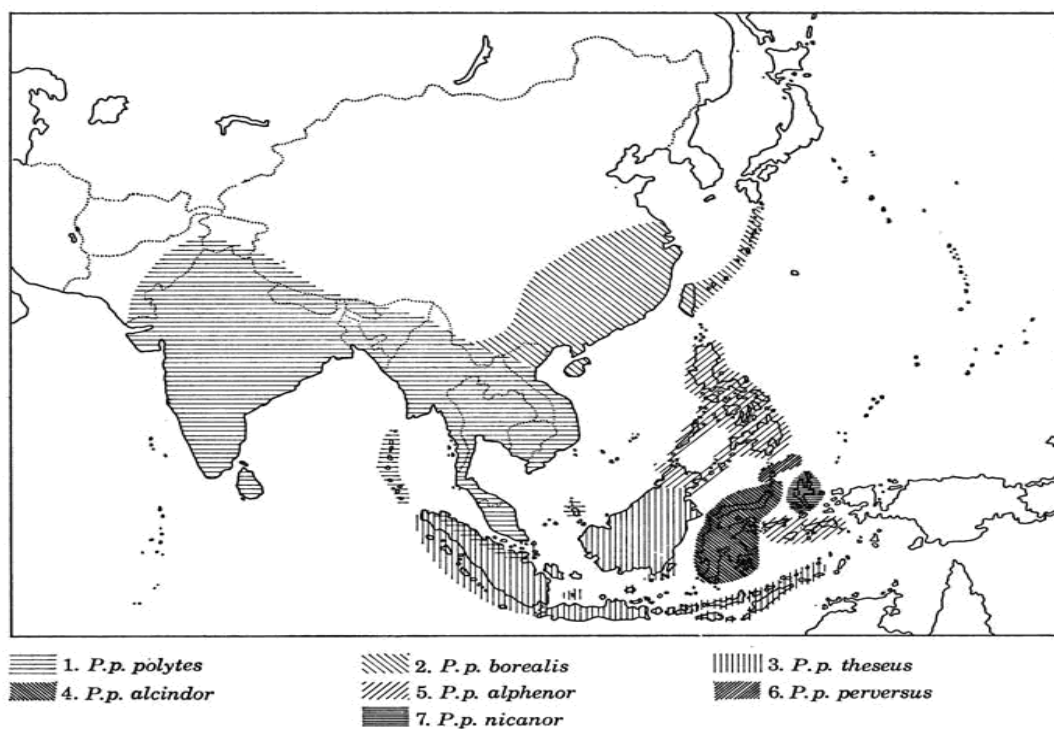


Figure 2.1. The distribution of the races of *Papilio polytes* (Clarke & Sheppard, 1971, p. 443)

Papilio polytes has been reported to become urbanised recently. It is common in neighbourhoods with gardens and in villages where *Citrus* are grown (Corbet & Pendlebury, 1992). Salmah et al., (1997), studied the population fluctuations of *P. polytes* for a period of one and a half years in Sipisang Area Kayu Tanam, West Sumatra. She found that the population of *P. polytes* was higher in January, June and December than in other months of the year. The number of individuals caught in home gardens of local inhabitants was higher than those in open areas and rubber plantations. In India (Rajasekhar, 1995) and in Lower Sindh, Pakistan (Munir, 2004) *P. polytes* was reported to be abundant from March to November.

The survivorship of *P. polytes* varies in each life stage. Generally, the mortality in the egg stage, younger larval instars (1, 2 and 3) and the pupal stages is higher than in older larval instars (3 and 4) (Munir, 2004). More specifically, mortality of *P. polytes f. polytes* was the highest in the first larval instar, followed by second larval instar, eggs, and the pupal stage, whereas mortality of *P. polytes f. stichius* was the highest in the second larval instar, followed by first larval instar, pupa, and egg stages (Munir, 2004).

The mortality of eggs was higher due to predators, parasitoids, infertility and dropping-off from the leaves. The major cause of mortality for larvae and pupae is predation and parasitoids, respectively (Munir, 2004). Ants, spiders and praying mantis are the common predators of the egg and larval stages of *P. polytes* and *P. demoleus* in the field, whereas the most important predators during the larval stages are the garden lizards and birds (Munir, 2004).

2.2. Lime butterfly (*Papilio demoleus* L.)

2.2.1. Biology of *P. demoleus*

Papilio demoleus belongs to the family Papilionidae and is a beautiful black yellow butterfly; on the upper side of both wings of this butterfly there are large yellow marking and a series of yellow submarginal spot (Corbet & Pendlebury, 1992; Otsuka, 2001). Male and female do not differ in colour but can be distinguished by a red spot with an anterior blue lunule on the hind wing of the female. In the Malaysian peninsula, subspecies *malayanus* of *P. demoleus* occurs (Corbet & Pendlebury, 1992; Matsumoto, 2002).

Females of *P. demoleus* lay their eggs singly on the upper and under surfaces of leaves and petioles or stems of their host plants (Corbet & Pendlebury, 1992; Salmah et al., 2002; Munir, 2004). The eggs of the lime butterfly are spherical, yellow with 1.00-1.10 mm in diameter and 0.90-1.05 long (Salmah et al., 2002). Munir (2004) further described the eggs of *P. demoleus* as shiny, yellow green in colour, with diameter 0.98-1.12 mm. After 3-5 days these eggs hatch and first instar larvae emerge (Salmah et al., 2002). The first instar larva of *P. demoleus* escapes the eggshell by making a small hole as described earlier for *P. polytes*. Their abdomens and heads are dark brown in colour (Salmah et al., 2002). According to Munir (2004) the first instar larva of *P. demoleus* is dark to light brown with yellowish white head.

There are five larval instars in *P. demoleus*. Munir (2004) observed that the head colour of the first and second instars is shiny brown, with prothorax yellowish brown in colour. The colour of the 3rd-4th, and 8th-9th, abdominal segments of both first and second instars *P. demoleus* larvae is yellowish milky and brownish yellow, respectively (Munir, 2004). The dorsal side of the body is covered with minute dark brown spines (Salmah et al. 2002; Munir, 2004).

The colour and shape of the third instar of *P. demoleus* is similar to that of the 2nd instar but the 3rd instar larva is larger in size. It is yellowish brown with some milky white marking on its lateral sides (Salmah et al. 2002; Munir, 2004). A pair of small white patches appears on each lateral side of the 7th and 8th abdominal segments and run to the dorsal side. The dorsal side of the body is covered by dark brown minute spines (Munir, 2004).

The fourth larval instar of *P. demoleus* is bigger in body size than the 3rd, instar is black (Salmah et al., 2002) or dark brown (Munir, 2004) in colour with a shiny brown head. The white marking on the lateral sides of the body, extends from the tip to first segment of the abdomen (Salmah et al., 2002; Munir, 2004). According to Salmah et al. (2002) the spines on the dorsal body of the larva disappeared after the third moult. However, Munir (2004) reported that the spines are present in the fourth instar larva.

Unlike *P. polytes*, the fifth instar larva of *P. demoleus* is quite different from its previous instars in shape, length, and colour. The fully grown 5th instar larva is smooth without any spine and generally its dorsal and lateral sides are green to yellowish green. Its ventral side near the legs, is milky white in colour (Salmah et al., 2002; Munir, 2004). There are two black bands on the dorsal side of the thorax with an accessory eye on the mesothorax. Black markings appear on the first and second segments of the abdomen in the 5th instar larva.

The average larval duration of *P. demoleus* is 13.5 days when reared at 26-28°C (Dahelmi et al., 2008) with a developmental range of 12-20 days (Salmah et al., 2002). According to Munir (2004) the larval duration of *P. demoleus* varies from 8 to 13.5 days at a temperature of 17.5-37°C.

Fully matured 5th instar larva stops feeding and rests for 1-3 hours after which it starts to search for a suitable place to pupate (Munir, 2004). The prepupal period ranges between 1-2 days (Salmah et al. 2002; Munir, 2004) with an average duration of 12.2 days (Dahelmi et al., 2008). According to Salmah et al. (2002) the range of the pupal period of *P. demoleus* is 13-14 days, while Munir (2004) reported it is 6-20 days at temperatures ranging between 17.5-37.5°C. The colour of the pupa depends on the background coloration of substrates where it is attached to (Smith, 1978; Munir, 2004; Hiraga, 2006).

2.2.2. Ecology of *P. demoleus*

Papilio demoleus L., commonly known as the lime or citrus butterfly, is a tropical/subtropical butterfly found throughout southern Asia to Australia (Corbet & Pendlebury, 1992; Suguru & Hurao, 2000). In Asia particularly, this butterfly is common in Sri Lanka, Pakistan, India, Taiwan, China, Japan and the Lesser Sunda islands (Corbet & Pendlebury, 1992). This butterfly species invaded Java, Borneo, the Philippines and Sumatra Island in the beginning of the 1970s apparently facilitated by deforestation and increased availability of citrus groves, (Matsumoto & Noerdjito, 1996). In Java *P. demoleus* had already established itself in 1994 and had spread all over the island (Matsumoto & Noerdjito 1996). Recently, *P. demoleus* was reported from Hispaniola Island, Dominican Republic (Guerrero et al., 2004), and from Puerto Rico (Homziak & Homziak, 2006).

Populations of *P. demoleus* occur from lowlands to low mountainous areas about 700 m above sea level, however, the species is more abundant in lowlands (Suguru & Hurao, 2000). *Papilio demoleus* is one of the commonest butterflies that frequents forest edge, urban areas, home gardens, and citrus orchards (Matsumoto &

Noerdjito, 1996; Suguru & Hurao, 2000; Matsumoto, 2002). It prefers open habitats to thick shadow rain forests as it is used to dominate (Matsumoto, 2002). In Pakistan, this butterfly was commonly found in Rawalpindi and Islamabad from April to October (Rafi et al., 1991). According to Suguru and Hurao (2000) *P. demoleus* also survives in extreme conditions in hot and dry deserts.

Changing environmental factors affect the population of citrus butterfly differently at different times and situations. Rafi et al. (2001) reported the egg mortality of *P. demoleus* varied from 44 to 61% in different generations and times. Factors such as failure to hatch, parasitoids, predators and eggs dropping off to the ground due to either the effect of rain or other host pests are responsible for egg mortality. Based on his finding, Rafi et al. (2001) also reported that the egg mortality of *P. demoleus* was higher than the larval (5-12%) and pupal (26-36%) mortalities. Predation on young larvae and pupal parasitism by *Pteromalus puparum* L. (Hymenoptera: Pteromalidae) are the major factors for the decrease in number of larvae and pupae, respectively. According to Munir (2004) the highest mortality of *P. demoleus* occurred in its pupal stage, followed by the egg stage, and then in the first and fourth instars larval stages.

2.3. Insect-plant interaction

2.3.1. Oviposition preferences of insects

Finding and choosing a host plant for oviposition is a challenging task for female herbivorous insects, and the decision made by the female may have far-reaching consequences for her fitness and that of her off-springs. Under natural conditions, ovipositing insects experience many external stimuli (e.g. visual and olfactory cues), their own internal physiological stimuli and a series of

environmental constraints (e.g. availability of host plant) (Bernays & Chapman, 1994; Badenes et al., 2004). Oviposition site selection is crucial for the successful development of the larvae (Singer, 1984). Optimal oviposition theory predicts that oviposition preference should correlate with host plant suitability for offspring development (Amwack & Leather, 2002), although experimental studies do not appear to support such a positive correlation between preference and performance (Scheirs & De Bruyn, 2002). Previously, Price (1991) developed the theory whereby vigorous plants that grow faster and ultimately reach a larger than average size should be preferred by the herbivore. Both plant biomass and nutritional quality are supposed to be higher in vigorous plants (Heisswolf et al., 2005).

Oviposition preferences by female on host plants are different and affected by many factors, from the host plant species (Jallow et al., 2001; Greenberg et al., 2001, 2002; Ulmer et al., 2002; Smyth et al., 2003) to the chemical compound of host plants (Nishida et al., 1990; Nishida, 1995; Ono et al., 2000a; 2000b; Konstantopoulou et al., 2002; Nakayama et al., 2002; 2003, Murakami et al., 2003; Nakayama & Honda, 2004; Omura & Honda, 2005; Li & Ishikawa, 2006; Derksen et al., 2007). In a single host plant species, preference of females ovipositing their eggs are affected by plant phenology (Ulmer et al., 2002; Smyth et al., 2003; Potter et al., 2004), plant parts (Amwack & Leather, 2002; Catta-Petra & Zucoloto, 2003), plant texture and shape (Jiang & Cheng, 2003; Maher & Thiery, 2003) and plant nutritional quality (Scriber & Slansky, 1981; Suzuki, 1998; Greenberg et al., 2001; 2002; Ide, 2003; Jiang & Cheng, 2003; Scheirs et al., 2003). Lundgren et al. (2008) stated that the females of *Orius insidiosus* (Say) (Heteroptera: Anthocoridae) were not attracted to constitutively produced plant volatiles when selecting oviposition

sites but rely on proximal cues (thickness of external plant tissues and trichome densities).

Generally, insect herbivores prefer the leaves more than other parts of plants for egg laying and feeding. Based on several reasons female *P. xuthus* (Nishida et al., 1990; Nishida, 1995), *Ascia monuste* L. (Lepidoptera: Pieridae) (Barros-Bellanda & Zucoloto, 1999; Catta-Preta & Zucoloto, 2003), *Spodoptera exigua* Hübner (Lepidoptera: Noctuidae) (Greenberg et al., 2001; 2002), *Mamestra configurata* Walker (Lepidoptera: Noctuidae) (Ulmer et al., 2001) prefer to lay their eggs on the underside more than the upper side of leaves and other plant parts. However, some lepidopteran species such as *Helicoverpa armigera* Hübner (Lepidoptera: Noctuidae) (Jallow et al., 2001), and *Erionota thrax* L. (Lepidoptera: HesperIIDae) (Okolle, 2007), prefer the upper side of leaves. Mascarenhas and Pitre (1997) and Ulmer et al. (2002) reported that some noctuids Lepidoptera lay their eggs on leaves which have more trichomes.

The full flower of the host plant *Brassica napus* L. was preferred more for egg laying by female *M. configurata* than the preflower and pods (Ulmer et al., 2002). Smyth et al. (2003) reported that for oviposition, female of *Crocidolomia pavonana* Fabr. (Lepidoptera: Pyralidae) preferred the flowering and preflowering stages more than stage 4 (base of the stem and base of all leaves still visible from above, 7-8 weeks after planting) and 5 (similar to stage 4, innermost heart leaves growing upright and not yet forming a firm center, 9-11 weeks after planting) of *Brassica olearacea* L., *B. juncea* (L.) Czern. and *B. rapa* L.

Nitrogen and water content of the leaves are the determining factors for oviposition and feeding preference of many insects. Butterflies and moths prefer more young leaves over old leaves (Sappington et al., 2001; Ulmer et al., 2001;

Greenberg et al., 2001; 2002; Okolle, 2007) because young leaves have higher nutritive value (Wheeler, 2001; Ide, 2003). Most studies have found that Lepidoptera oviposit their eggs on host plant leaves which have a higher water and nitrogen content (Greenberg et al., 2001; 2002; Hodar et al., 2003; Jallow et al., 2003; Moreau *et al*, 2006a; 2006b; 2006c)

Most hypotheses on the evolution of oviposition behaviour agree that females choose plant species, individual plant or parts of a plant that will maximize the larval growth and survival of their off-springs (off-spring performance) (Thompson & Pellmyer, 1991; Potter et al., 2004). Papilionid butterflies mostly feed selectively on the limited number of plants belonging to family Rutaceae. Although the choice of food plants is determined at the oviposition and larval feeding stages, the fate of the larvae are dependent mostly on the gravid females selection of oviposition site (Nishida, 1995).

Eighty percent of *Papilio* species feed on rutaceous plants. Preference and adaptation of *P. polytes* on several rutaceous plant species differs. Oviposition response and larval survivorship of *P. polytes* on *C. depressa* was strong and similar to *Fagara ailanthoides* (Siebold & Zucc.) Engl., moderate on *Evodia meliifolia* Benth. and weak on *Orixa japonica* Thumb. Adaptation of *P. polytes* to *Citrus*, *Fagara*, and *Orixa* plants is quite similar to those of *Papilio protenor* L. and *P. xuthus* L. (Murakami et al., 2003).

Oviposition preferences by females of *P. polytes* are also affected by important host factors, such as shape, colour, and size. Meanwhile, chemical cues also play a major role in recognition at the final step of female oviposition and host selection in butterflies (Saxena & Goyal, 1978; Renwick & Chew, 1994; Konstantopoulou et al., 2002). Oviposition response in *P. polytes* is not induced by a

single chemical component but by a complex synergistic action of compounds (Nakayama et al., 2002; 2003; Murakami et al., 2003; Nakayama & Honda, 2004; Nishida, 2005). Thus primarily adults, and to some degree larvae must recognise the best food available and choose them for oviposition or feeding (Bernays & Chapman, 1994; Hodar et al., 2003).

2.3.2. Food quality and larval performance

Oviposition site selection in plant-insect interaction is assumed both in ecological and evolutionary terms to be determined by the quality of the oviposition site and the offspring performance (Mayhew, 1997; Rank et al., 1998). Plant leaf nitrogen concentration and water content are generally considered as an indication of food quality (Scriber & Slansky, 1981). Nitrogen is a constituent of amino acids, proteins, and chlorophyll, and is a critical nutrient for plants which, in turn, is critical for polyphagous insects (Wang et al., 2006).

Larval feeding preference studies have revealed a correlation with larval performance. Larvae of Lepidoptera prefer host plants with higher food quality because larval performance, survivorship, abundance, and female fecundity and distribution are strongly affected by larval food quality (Rodriguez & Moreita, 2002; Stefanescu et al., 2006; Moreau et al., 2006a; 2006b; 2006c). Gyertych et al. (2005) reported that the larval performance of gypsy moth, *Lymantria dispar* L. (Lepidoptera: Lymantriidae) increased with increased food quality.

Fecundity indirectly depends on the larval diet and has a positive correlation with pupal weight and pupal size (Rodriguez & Moreita, 2002; Moreau et al., 2006c). *Papilio demoleus* prefers the young leaves of *C. aurantifolia* and *C. reticulata* for oviposition and as larval food (Munir et al., 2005). On both species of citrus, Munir

et al. (2005) reported that the larval duration of *P. demoleus* was shorter, and the survivorship and growth index values of larvae are higher than the scores on some other host plants.

Performance values of penultimate larval instar are significantly greater on more suitable host plants. These values based on five categories are usually measured to evaluate efficiency of food utilization and larval development and relationship with larval food quality. The values includes relative consumption rate (RCR), relative growth rate (RGR), efficiency of conversion of digested food (ECD), efficiency of conversion of ingested food (ECI), and approximated digestibility (AD) (Scriber & Slansky, 1981; Simpson & Simpson, 1990).

When food quality is low the efficiency of food utilization by the Lepidoptera larvae will decline. For example, the ECI value will decline with lower plant nitrogen, followed by a decrease in ECD value. The decline in AD, especially for leaf chewers are often associated with less selective feeding by later instars, resulting in the consumption of a higher proportion of indigestible fibre (Simpson & Simpson, 1990).

Food quality is also the main factor that determines the distribution and abundance of many phytophagous insects (Hodar et al., 2002). The larval stage is more abundant on hosts that support faster growth (Kursar et al., 2006). The amount and the quality of the food ingested by larvae strongly influence the storage of resources that are allocated for reproduction (Slansky & Rodriguez, 1987; Amwack & Leather, 2002).

2.4. Life tables

The abundance of an insect species in a population or generation in the field is always changing. These changes and the factors that cause them can be enumerated in expressing the result in the form of a life table (Varley et al., 1973). As agreed by Yang and Chi (2006), the life table studies provide the most comprehensive portrayal of survival, development, and reproduction capabilities of a population under varying conditions. Life table studies are important in understanding population dynamics of a species (Carrey, 1993 *cit.* Southwood & Henderson, 2000; Xiushan et al., 2006); such tables are necessary to identify the key factors that influence the population size (Xiushan et al., 2006).

Life tables have long been used by actuaries for determining life expectancy (Southwood & Henderson, 2000) and presented in several formats (Krebs, 1989). Constructing a life table is often a simple method for keeping track of births, deaths, and reproductive output in a population of interest (Krebs, 1989; Southwood & Henderson, 2000). According to Southwood and Henderson (2000) there are two basic methods for constructing life tables which always apply to insect life, depending on the species and population structure. First, the age-specific (or horizontal, or cohort) life table which is based on the fate of a real cohort in which the population may be stationary or fluctuating. Second, the time-specific (or vertical, or current) life table is based on the fate of an imaginary cohort calculated by determining the age structure at one instant time.

In age specific life table, successive observations of the cohort are needed, while in time-specific life table the age-grouping of the individuals at a single time is needed. The age-specific life table is more widely applicable to determine an insect's life expectancy. However, on *Aphids* and mosquito groups the time specific life table

is more relevant because of the necessity of distinguishing various instars. The time-specific life table have been most widely applied to vertebrate populations (Southwood & Henderson, 2000). Studies on insect life tables could be undertaken in the laboratory (Chi & Yang, 2003; Zied et al., 2003; Baniameri et al., 2005; Yu et al., 2005; Chi & Su, 2006; Ellers-Kirk & Fleischer, 2006; Yang & Chi, 2006) or in the field (Hirose et al., 1980; Watanabe, 1981; Feeny et al., 1985; Choate & Rieske, 2005; Xiushan et al., 2006).

The method of data collecting is very important to generate information on the life table of an insect species. There are four general methods of collecting data viz. age at death, colony size, age at death for several cohorts, and age structure (Krebs, 1989). Generally, data collected for life table studies contain information on mortality, natality, survival, emigration, immigration and age structure since all of these factors can affect a population (Varley et al., 1973; Krebs, 1989; Southwood & Henderson, 2000).

Mortality in insects is influenced by biotic (such as predators, parasitoids, and pathogens) and abiotic factors including rainfall, temperature, wind, etc. (Varley et al., 1973; Yamamura, 1999; Choate & Rieske, 2005). Usually mortality in many insect species occurs before the adult stage is reached (Chi & Yang, 2003). Each factor causes mortality at different life stages of different species of insects. Some factors may cause mortality only in the egg stage or larval stage, while other factors could cause mortality at both stages (Stilling, 1988; Yamamura, 1999; Choate & Rieske, 2005; Yu et al., 2005).

After observing 12 generations of *P. xuthus* in three years, Hirose et al. (1980) reported that most of the observed eggs' mortality in every generation was due to parasitism by several parasitoids primarily *Trichogramma papilionis*

Nagarkatti (Hymenoptera: Trichogrammatidae). In addition, predation of eggs by the anthocorid bug, *Orius sauteri* Poppius (Heteroptera: Anthocoridae) and the salticid spiders was noted. The data of 13 generations of *P. demoleus* showed that the mortalities of eggs and first instar larvae were higher than mortalities in the older instars Munir et al. (2007). Responsible mortality factors included egg infertility, egg falling off, predators and parasitoids. Mortality of the first instar larvae in every generation was very high (50-70%) but its causes remained unknown. However, 30-60% of second to fifth instar larval mortality was mainly caused by predators. In the pupal stage of *P. xuthus*, parasitism by pteromalid wasp, *P. puparum*, was usually high throughout the year as compared to mortality caused by other factors (Hirose et al., 1980).

Variable predation rates and vulnerabilities of preys have been observed in many studies on predator-prey interactions. For example, Clement and Yeargan (1997) reported that all stages of the predatory *O. insidiosus* (Heteroptera: Anthocoridae) fed on eggs and first instar larvae of green clover worm, *Platytepan scabra* Fabr. (Lepidoptera: Noctuidae). The fourth instar nymph and adult *O. insidiosus* could prey upon the second instar caterpillar, but none of the immature or adult *O. insidiosus* was able to prey upon the third and fourth instar larvae of *P. scabra*.

2.5. Population dynamics

Berryman & Turchin (2001) and Canto-Silva & Romanowski (2003) stated that understanding the population ecology was extremely necessary to solve many problems in insects managements, such as critical environmental issues in conservation biology, natural resources management, and pest control. The strength

and manner of endogenous (density dependent) vs. exogenous (density-independent) forces determine the insect population dynamics (Royama, 1992 *cit.* Ylioja et al., 1999; Berryman et al., 2002; Pedigo, 2002; Berryman, 2004).

Many density-dependent factors affect the population abundance, e.g., natural enemies (Varley et al., 1973; Feeny et al., 1985; Hassel et al., 1998; Berryman & Turchin, 2001; Zalucki et al., 2002; Gomes-Filho, 2003), host plant quality (Varley et al., 1973; Hassel et al., 1998; Zanuncio et al., 1998; Ylioja et al., 1999), competition (Varley et al., 1973; Hassel et al., 1998) and maternal effect (Fowler, 2005).

Natural enemies regulate the populations of most herbivores. Prey population may grow infinitely when there are no predators. On the contrary, predators can extinguish as well when there are no preys (Varley et al., 1973; Hassel et al., 1998). In case of presence of both predator and prey, the rate of prey hunting predators is proportional to prey density (Berryman & Turchin, 2001).

The population dynamics of some forest Lepidoptera are strongly related to the abundance of their insect parasitoids (Berryman, 1996). Based on 11 years of observations, Morris (1959) found that population of black headed budworm, *Acleris variana* Fernald (Lepidoptera: Tortricidae) was very much determined by the abundance of its larval parasitoids. The black headed budworm population has already declined as a result of heavy parasitization. Meanwhile, according to Menge (1992), Power (1992) and Strong (1992) a balance between natural enemies and plant quality or quantity influences the population ecology of herbivores.

The density of eggs and larvae of *Phytobia betulae* Kangas (Diptera: Agromyzidae) was positively associated in the quality and quantity of host plants (Ylioja et al., 1999). Unpalatable or poor plant quality of most plant tissue limits the

populations of many insects (Hunter et al., 1997). In case of *P. betulae*, Ylioja et al. (1999) found that yearly abundance of the fly was greater on individual tree that grows most rapidly since the tree provides more food and/or space.

Ehrlich (1984) stated that the distribution and abundance of nutritional resources, primarily food plants for larvae and liquid nourishment for adult is the most important factor influencing the structure of butterfly populations. Food limitation affects the intra-specific competition within insect populations. In *A. monuste orseis* (Lepidoptera: Pieridae) population for example, intraspecific competition for food during the larval phase leads to serious problems for larval and adult performances such as prolonged larval duration, high larval mortality and reduced adult weight and number of eggs per female (Barros-Bellanda & Zucoloto, 2002). Meanwhile Corbitt and Bryning (1996) stated that the larvae from a competition showed a high approximate digestibility (AD), lower in efficiency of conversion of digested food (ECD) and efficiency of conversion of ingested food (ECI). It was indicating that these larvae were less selective in feeding. In *Ips typographus* L. (Coleoptera: Curculionidae) population competition for host substrate was the main factor regulating its populations (Okland & Berryman, 2004).

Climate affects insect as individuals and determine the changing insect population (Varley et al., 1973). Some Lepidoptera species exhibit conspicuous population fluctuations that are synchronous over large geographical areas, suggesting that climate may play a major role in the timing and occurrence of these population changes (Jones et al. 2003, Haukioja 2005). Weather such as temperature (Walther et al. 2002; Helmuth et al. 2005; Reynolds et al., 2006; Yamamura et al., 2006), rainfall (Sivapragasam et al., 1988; Pedigo, 2002; Kobori & Amano, 2003), and humidity (Pedigo, 2002) act on the population indiscriminately.