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THE ECOLOGY AND ECONOMIC POTENTIAL OF WILD SILKMOTH *ANAPHE PANDA* (BOISDUVAL) (LEPIDOPTERA: THAUMETOPOEIDAE) IN THE KAKAMEGA FOREST

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DECLARATION BY CANDIDATE

This thesis is my original work and has not been presented for a degree in any other University

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DEDICATION

This thesis is dedicated to the All Mighty God and to my late daughter Nzouetchou Mbahin Venceslas Williams.

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Km^2: Kilometer squared

KSh: Kenyan shilling

K W S: Kenya Wildlife Service

mm: Millimeter

N: Newton

Na$_2$CO$_3$: Sodium carbonate anhydrous

P: Probability

RH: Relative humidity

SE: Standard error

SEM: Scanning Electron Microscope

SPSS: Statistical Packages for Social sciences

USA: United States of America

$ US: United States dollars

χ^2: Chi-square

γ: Mean

μ: Population mean number in a unit of plot

σ^2: variance

°C : Degree Celsius

%: Percentage

10': Ten second

DEFINITION OF FABRIC TERMS AND TYPES

Cleanness defects: These are categorized into three general groups: super major defects; major defects and minor defects. Defects are: waste, slug, bad casting, split ends and large loop.

Denier: The measure denier expresses the size of silk thread. A denier is the weight of 450m length of silk thread divided into 0.05 g units.

Hand spinning wheel: Is a spinning apparatus that is operated manually, one hand to drive the wheel and other to feed cocoons.

Raw silk: Silk fiber as it comes from the cocoon and is covered with a protective layer called silk gum or sericin. The silk gum is dull and stiff.

Raw silk classification: Raw silk is divided into three categories according to their sizes: 1st category (18 denier and below); 2nd category (19-33 denier) and 3rd category (34 denier and above). *A. panda* raw silk belongs to the 3rd category.

Silk reeling: Is the process by which a number of cocoon filaments are reeled together to produce a single thread.

Spun silk: consists of several fibers of silk spun into narrow threads. It is often made from the broken cocoons from which moths have emerged.

Warp: the set of strands or fibers that are placed length-wise in the loom.

ABSTRACT

The superiority of silk as a textile fibre has been recognized from time immemorial, the luxurious look; sleek feel and lustre of silk fabric are unquestionably inimitable. The demand for silk is constantly increasing in the world market and this provides excellent opportunities for any producer country to diversify and optimize any source of production. African countries that enjoy congenital climate for rearing wild silkmoths have great scope and opportunity to promote sericulture. Studies were carried out during 2005-2007 on the ecology and economic potential of wild silkmoth *Anaphe panda* (Boisduval) which occurs in the Kakamega Forest, western Kenya. Silkworms feed on *Bridelia micrantha* (hochst). Egg clusters were present from mid-October to mid-May; silkworms appear from December to September; pupae are present from mid-April to January and adults occur from early October to April. Eggs hatch after 40 to 55 days depending of the temperature. Seven larval instars occur with a growth prodigious from 3 mg at 1st instar to more than 3,000 mg at the 7th instar. The duration of larval development depends on temperature and ranged from 83 to 118 days. The duration of the pupal stage ranged from 107 to 178 days depending on the brood. Pupal and adult sex ratios are not even. A moth life span ranged from 4 to 7 days. The factor that contributes most to egg mortality appears to be egg parasitism, and in the Kakamega Forest, eggs were mainly parasitized by two chalcids: *Telenomis gowaeyi* Grahan and *Pleurotropis telenomis* Lima. Eggs in the mixed indigenous forests (forest with indigenous and exotic species) seemed to be more affected than those of the indigenous forest (forest with indigenous species only), and the infection rate was also significantly different ($P = 0.0025 < 0.05$) between the mixed indigenous and indigenous forests. Higher mortality rate was observed from the 1st to 4th instar, but a highly significant difference was observed between the unprotected and the protected silkworms. Nevertheless, the lowest mortality rate was observed from the 5th to 7th instars. High mortality rate of silkworms was observed in the mixed indigenous forest compared to the indigenous forest. The survival rate observed during the larval developmental period was significantly higher for the protected than for the unprotected silkworms. Protection with net sleeves seemed to minimize the instantaneous risk and effectively increased the survival of the silkworm. The tachinid fly *Exorista cardinalis* Fabr and the ichneumon wasp *Cryptus leucopygus* Granenhorst were found to be parasitoids at the larval stage. *A. panda* cocoon nests were found to be infested by various dipteran and hymenopteran parasitoids. Geographical information systems (GIS) and Poisson distribution revealed that distribution of the host plant *B. micrantha*; cocoon nests and egg-clusters were not uniformly distributed in the indigenous and mixed indigenous forests. A bamboo structure was found on *A. panda* fine structure filament, and this structure is characteristic and unique as compared with all other cocoon filaments of lepidopteran insects. By processing *A. panda* cocoon nests into silk shirts, the net income was multiplied twenty four times. Therefore, wild silkmoth farming could be a supplementary activity carried out by farmers for income generation while at the same time conserving biodiversity.

CHAPTER ONE

1 GENERAL INTRODUCTION

1.1 Background

Non-mulberry sericulture is universally known as forest or wild sericulture. Silkmoths existing in wild conditions are known as wild silkmoths. In Kenya according to Kioko *et al*. (1999a), wild silkmoth habitats and natural ecosystems, such as the Kakamega Forest have suffered severe encroachment as a result of the growing human population and the demand for agricultural land. According to Kamugisha *et al.* (1997), the population density in Kakamega District varied from 336 to 1746 people per Km^2 in 1997 with an average density of 452 people. This region includes large areas of uninhabited forest, thus it may be considered as one of the most densely populated regions in Kenya. The rate of growth for the decade before 1989 for the district was over 3% per annum and was 2.9% during the previous decades (Haupt, 2000). Blackett (1994) reported an average of 600 people per km^2, ranging from 300-870 people per Km^2. In 1991, the population of the thirteen sub-locations adjacent to the forest was 155,594 people, with an average density of 713 per Km^2 (Blackett (1994).

Due to the continually growing population size, the pressure on the forest is increasing, because for the local people, the forest plays an important role in satisfying their daily needs and income generation: firewood (Plate: 1.1), house building material, medicinal plants (Plate: 1.2), charcoal burning (Plate: 1.3) and butterfly collection (Kokwaro, 1988; KIFCON, 1994).

Plate 1.1: Firewood collection in Kakamega Forest

Plate 1.2: Medicinal roots collected from Kakamega Forest

Plate 1.3: Charcoal burning in Kakamega Forest

However, despite all these forest resources, Western Kenya remains one of the poorest Provinces in Kenya with a high level of rural poverty of 60% (Daily Nation, 2005; The Standard, 2005).

Wild silk production is an eco-friendly, agro-based venture with a great potential for environmental amelioration, employment generation, artisan's development and export earnings (Kioko *et al.*, 2000). The highest diversity of the African wild silkmoths is in the family Lasiocampidae. An earlier survey on the diversity of wild silkmoths in East Africa recorded about 33 species in 17 genera of this family (Kioko *et al.*, 2000). Despite this high diversity, only a few of these species have so far been utilized for wild silk production in East Africa.

In Kakamega Forest (Plate 1.4), five species of wild silkmoths namely *Anaphe panda* Boisduval, *Pachmeta contraria* Walk, *Lechriolepsis pulchra* Aur., *Mimopacha bryki* Aur. and *Epiphora vacuna* Westw were identified by Kioko *et al.* (1999a) and recommended for wild silk production. *A. panda* (Boisduval) showed the most potential since it had a huge silk-nest (Plate 2.3) that is communally weaved by 20-105 silkworms larvae. A ground survey by Kioko *et al.* (1999a) confirmed the availability of the wild silkworm host plant; *Bridelia micrantha* (Hochst) (Plate 2.5). A case study on the distribution of *B. micrantha,* one of the host's plants feed on by larvae of *A. panda* showed that it had been recorded in 103 localities in Kenya and was widely distributed in East Africa (Kioko *et al.*, 1999a). A questionnaire distributed to local villagers near the Kakamega Forest (the only remaining equatorial rain forest in Kenya) responded positively to the potential of initiating local wild silk production activities as an extra source of income. In the Musembe community living adjacent to the forest, *B. micrantha* was found in 84% of the farms and 98% of the people confirmed their interest in pursuing wild sericulture (Kioko *et al.*, 1999a).

The *Anaphe* species are widely distributed in the intertropical regions of continental Africa such as Nigeria, Uganda, Kenya, Cameroon, Congo and Togo. The important species used in the production of *Anaphe* silk are *A. infracta* Walsingham, *A. venata* Butler, *A. panda* Boisduval, *A. reticulata* Walker, *A. carteri* Walsingham, *A. moloneyi* Druce and *A. ambrizia* Butler. The closely related species *Epanaphe carteri* Walsingham and *Epanaphe* (Hypsoides) *vuilleti* Joan are exploited in Cameroon (Jolly *et al.*, 1979).

The *Anaphe* are polyphagous moths. Out of the twenty-two of their host plants so far recorded, *Albizzia fastigiata, Sterculia tragacantha, S. setigera,* S. *rhinopetala,* S. *oblonga, Cordia milleni, Tamarindus indiicus* and *Zizyphus mucronata* are found in Nigeria; *B. micrantha, Cynometra alexandri* and *Triumfetta macrophylla* in Uganda; *Z. jujuba* and *A. fastigiata* in Cameroon and Congo.

Cynometa milleni and *B. micrantha* are the most preferred host plants by *A. moloneyi* and *A. infracta,* and *A. veneta* respectively. Because in nature the food plants are scattered over a large area, *B. micrantha,* which flourishes in virtually all tropical climatic conditions with very little attention can be raised from seed or seedlings or better from cuttings which propagate very rapidly and be ready for use as a host plants for *A. panda* in about a year (Jolly *et al.*, 1979; Gowdey, 1953)

Anaphe silkmoths, in the family Thaumetopoeidae comprise several species within the African continent. There are a number of records of *Anaphe* silk having been utilized for exportation in several parts of Africa (Ashiru, 1986, Gowdey, 1953). In Uganda *Anaphe* silk was one of the country's exports between 1910-1945 (Ashiru, 1986). However, there are some constraints facing the production of *Anaphe* silk. These include lack of sufficient silkmoth populations in the wild, lack of local know-how on silk farming practices and rearing techniques and insufficient knowledge on the ecology of *A. panda* (Kioko *et al*., 1999a). Hence, this study was undertaken to explore the biology and economic potential of wild silkmoth *A. panda* in the Kakamega Forest of western Kenya.

1.2 Importance of Kakamega Forest

Kakamega Forest (Plates: 1.4 and 2.6) is bio-geographically important as it is situated at the edge of several regional vegetation zones. It supports several Guineo-Congolian plant species at their eastern most African distribution limit, and White (1983) classified it as a transitional rain forest. The natural vegetation is tropical rainforest (KIFCON, 1994) and as a consequence, the rainforest spectrum is the forest's dominant vegetation class. There are over 150 documented species of woody trees, 90 species of dicotyledonous herbs, 80 species of monocotyledonous herbs, and a further 62 species of ferns totalling to about 380 identified species of vascular plants (KIFCON, 1994; Mutangah *et al.*, 1992). *B. micrantha* (Hochst) (Plate 2.5) the host plant of *A. panda* is found there (Kioko *et al.*, 1999a). Kakamega Forest has a diverse fauna exhibiting a high degree of endemism and rarity, and it is estimated that 10% to 20% of the fauna in general may be endemic (KIFCON, 1994).

Plate 1.4: A view of buffer zone in Kakamega Forest

Plate 1.5: Species of bird and primate found Kakamega Forest

Kakamega Forest has a high diversity of insects and it is particularly known for its butterfly diversity that is estimated to be about 400 species (Emmel and Warren, 1993; Larsen, 1991; Karanja and Chege, 1985). In 1980, 92 species of butterflies were recorded in the Kakamega Forest (Angwin, 1980; Clifton, 1980) of which, 5 species were Papilionidae, 8 Pieridae and 64 Nymphalidae.

In 1991 a total of 219 butterfly species were recorded during 13th – 16th August expedition (Emmel and Warren, 1993) comprising of 7 species of Papilionidae, 28 species of Pieridae and 115 species of Nymphalidae (Karanja and Chege, 1985). In addition, there are over 350 species of birds (Bennun and Njoroge, 1999) that reside in the forest, and several primate species (Plate 1.5) are also present. Opportunities for scientific studies are abundant in Kakamega Forest and are of crucial importance for the conservation of this unique ecosystem.

1.3 Statement of the problem

In order to propose an alternative strategy that takes into account the interactions between the diversity of resources and the subsistence means, several studies on wild silkmoths have been carried out at the International Centre of Insect Physiology and Ecology (icipe) Nairobi, Kenya. Similar studies are required for another wild silkmoth (*A. panda*) that is economically viable. Unfortunately, the population of wild silkmoths in Africa is declining due to over-consumption of silkmoth larvae, which are favored by some communities for food (Raina *et al.*, 2000; Kioko, 1999a; 2000; Ashiru, 1988b; Oberprieler, 1994; Munthali and Mughogho, 1992). Furthermore, the reduction in the species composition and natural population density of wild silkmoths may also be due to changes in the ecosystem, climate and depletion of the plants caused by extensive deforestation. The rapidly increasing human population density has also created pressure on land use, resulting in human colonization of the forestland for human dwelling and cultivation which has greatly changes in the biotic components (fauna and flora). The other reasons for depletion of these wild silkmoths may be their restricted level of distribution due to the narrow range of preferred food plants, low survival rate and increasing attack by natural enemies (parasites, predators and diseases) (Raina *et al.*, 2000).

However, the demand for silk is constantly increasing in the world market (Table 1.1) providing excellent opportunities for any producer country to diversify and optimize any source of production. There is a steady growth in silk consumption both in producing and consuming countries. Silk consumption in Japan, U.S.A and Europe, the three major silk consuming countries together, have contributed about 50% of raw silk product in the world.

Judging from the consumption trends, silk demand is growing annually by about 3 to 5%. However, despite the growth in demand and production, the contribution of silk to the world textile fibres is only about 0.2%, and this situation has remained unaltered since the last three decades. Because of insufficient production in the wild, lack of research and development, local know how on silk farming practices and rearing technology, parasitism and predation aspects, *A. panda* which has great potential for wild silk production is unexploited.

Table 1.1: Trends in world silk consumption (in tonnes)

Country	1982	1995-1996	1997-1998
Japan	20700	22350	13925
India	3500	15082	16445
China	7500	10000	12000
S. Korea	800	2047	2272
Europe	4530	16015	13363
U.S.A	5500	24499	23421
Rest of world	10970	13653	11739
Stock variation	-	2555	2796
Total	53500	106201	95961

Source: ITC Silk Review 1997

Despite their economic importance for rural household, little research has been carried out on these silkmoths. Hence, the current research was undertaken to provide insight on the population dynamics and economic potential of *A. panda*. It is hoped that this knowledge will be useful for sustainable production of wild Anaphe silk in areas adjacent to the Kakamega Forest.

1.4 Justification

The prospect for development of silk industry in Africa is very bright. Sericulture is a labor-intensive agro-based undertaking, which can provide employment to millions of people living in rural areas. African countries can use it as a venture for generating income and employment for rural households. The world trend in silk production (Table 1.2) and consumption are also quite favourable.

The investment requirements are low and the gestation period to generate income is very short. The demand is increasing and the production in major silk-producing countries is gradually diminishing due to various reasons such as: diversification, affluence and high cost of labor (Raje, 1999). Hence, African countries that enjoy congenital climate for cultivation of mulberry and rearing wild silkmoths have great scope and opportunity to promote sericulture. However, efforts to introduce and promote sericulture in the African continent have not been very successful and are riddled with many problems namely, lack of research and development support and suitable infrastructure, low awareness of the potential returns of silkworm farming and inexperience with sericultural techniques (Raina, 2000; 2004).

Table 1.2: Trends in world raw silk production (includes all types) in tonnes

Country	1998	1999	2000	2001	2002	2003	2004 (P)	Share (%)
China	57500	56956	61648	64567	68600	94600	102560	81.65
India	15544	15214	15857	17351	16319	15742	16500	13.14
Brazil	1821	1554	1389	1485	1607	1563	1512	1.2
Thailand	900	1000	955	1510	1510	1500	1420	1.13
Uzbekistan	1500	923	1100	1260	1260	950	950	0.76
Vietnam	862	780	780	2035	2200	750	750	0.6
Japan	1080	650	557	431	394	287	263	0.21
Korea S.	210	200	165	157	154	150	150	0.12
Others	1572	1250	1952	1692	3814	1500	1500	1.19
Total	80989	78530	84403	90488	95858	117042	125605	100

Source: ISC- 2005; P: Provisional

Research findings by Kioko *et al.* working in Kakamega Forest (1999a) have shown that *A. panda* cocoons have good characteristics for wild silk production. However, there are some constraints facing the production of *Anaphe* silk. These include lack of sufficient population in the wild, lack of know how of silk farming practices and rearing techniques, predators and parasites. To help curb some of these problems, the current study undertook to investigate the different factors affecting the population dynamics of *A. panda.* The study also focused on the mass production of *A. panda* cocoon nests and it profitability in the Kakamega Forest.

Moreover, the introduction of wild silk production in Kakamega Forest may offer an important economic benefit to households in this district. Silk production enterprise will also enhance conservation of the wild silkmoth, *A. panda* and their habitats for silk production. The final goal will enhance the conservation and utilization of a biodiversity resource.

The global silk demand is growing annually by about 3 to 5%. With this trend in production and consumption, silk will never be available in larger quantities and international supplies will remain limited in future, unless new production bases are created and nurtured. This world market scenario has thrown open a very good opportunity for the African countries to rapidly increase their silk production. All these developments have indicated that the availability of silk in the world market has continued to be in limited quantity for a long time.

According to Jolly *et al.* (1979), it would be necessary to expand the world non-mulberry silk production globally by 20,000 metric tons over ten years. Some of the leading mulberry silk producing countries appear to have reached saturation point-attribute due to the acute scarcity of labor and the increasing cost of production. There is therefore an opportunity for the developing countries like Kenya, Uganda, Madagascar and Cameroon to contribute in the production of raw silk annually for the developed world market and especially since the high quality of the wild silk has drawn the attention of silk users (Jolly *et al.*, 1979).

Tropical forests are being altered and destroyed at a rapid rate, yet the impact on biodiversity and ecosystem services are only beginning to be understood (Daily, 1997; Lawton and May, 1995). Kenyan forests are disappearing at a rate of 167 ha per year and Kakamega Forest specifically is disappearing at a rate of 245 ha per year (Emmerton, 1991). This forest in particular is at a risk since it is considered a "high potential" land area, which is particularly desirable for conversion into human uses. The Kakamega region of western Kenya remains one of the most populated rural areas in the world with 268 people per square kilometre (Wass, 1995). The loss of butterfly habitats is responsible for the decline and extinction of many species (Mattoni, 1990). Heightened environmental awareness, population reduction, and provision of alternative livelihoods to the forest adjacent communities would remove pressure from the forest resources and provide the opportunity for the preservation of the forest biodiversity.

1.5 Hypotheses

a. Biotic and abiotic factors strongly influence *A. panda* population dynamics.
b. There is a significant difference in the survival rate of the protected and unprotected silkworms with net sleeves.
c. Mass production of *A. panda* cocoons is economically profitable for communities living adjacent to the Kakamega Forest.

1.6 Objectives

1.6.1 General objective

To study the ecology and economic potential of wild silkmoth *A. panda* in the Kakamega Forest.

1.6.2 Specific objectives

a. To study the biology and identify biotic and abiotic factors that contributes significantly to the variation of *A. panda* population in the wild.
b. To study the effect of protecting early developmental stages of *A. panda* with net sleeves on its population growth.
c. To assess the economic profitability of *A. panda* silk farming for the communities lives around Kakamega Forest.

CHAPTER TWO

2 LITERATURE REVIEW

2.1 Historical perspective of wild silkmoths

The history of silk is as long as that of civilization itself. From its origin in China in about 2,200 B.C, the silk industry has had an adventurous course of evolution, becoming established with time in many parts of the world. Tradition credits His-ling-shi, the 14 year old bride of the China Emperor Huang Ti, with discovery of the potential of the cocoon and the invention of the first silk reel. China successfully guarded the secret until AD 300, when Japan and later India, penetrated the secrecy (Robert De Gregorio, 1997). The superiority of silk as a textile fibre has been recognized from time immemorial, the luxurious look; sleek feel and lustre of silk fabric are unquestionably inimitable. References in the Old Testament (The Bible, 1971) indicate that silk was known in biblical times in western Asia, from which it was presumably known to the Greek Islands of the Aegean Sea. When Darius III, king of Persia, surrendered to Alexander the Great, he was clothed in such silken splendour that Alexander was completely overshadowed and demanded as spoils the equivalent of US $7 million in silk. Caravans carried silk on camelbacks from the heart of Asia to Damascus, Syria, the marketplace where East and West met. Silk became a valuable commodity in both Greece and Rome. The Roman statesman and general Gaius Julius Caesar restricted silk to his exclusive use, the purple Roman stripes and his favoured officials. Despite this restriction, the use of silk in Rome spread in the paroxysm era. Until AD 550 all silk woven in Europe was derived from Asiatic sources. About that time, the Roman emperor Justinian I sent two Nestorian monks to China, where due to the risk on their lives, they stole mulberry seeds and silkworm eggs, and brought them to Byzantium (Robert De Gregorio, 1997). Thus, the Chinese and Persian silk monopolies ended. By the 12th and 13th centuries, Italy become the silk centre of the west, but at 17th century France challenged Italy's leadership and the silk looms were established in the Lyons area from that time until today (Robert De Gregorio, 1997).

The "Golden age" of the wild silkworm industry was during the years 30 of the Meiji era (1897). In those days, about fifty-two percent of the seven hundred farmers in the district of Ariake (China) reared wild silkworms in their fields, which occupied an area of about 3,000 ha. Wild silkworms' industry production was estimated at about 8,500,000 cocoons (Nakajima, 1980).

According to Raina *et al.*, (1999), the natural silk is broadly classified into two types: silk of plant and animal origin. The natural silk of plant origin is obtained from silk cotton tree and floss-silk tree. The natural silk of animal origin is broadly of two types: mulberry and non-mulberry. Mulberry silkmoth is a domesticated type, whereas non-mulberry is universally known as "wild type", which is also found in semi-domesticated form. Further, the non-mulberry variety of silk is classified as insect and non-insect type. Insect type of silk is named as Eri, Muga, Anaphe, Fagara, Coan and Tasar. The non-insect silk is named as mussel and spider silk and is obtained from mollusca and spider, respectively. The insect type of silk is again classified as commercial and non-commercial. Eri, Muga, Anaphe, Fagara, Coan and Tasar are commercial type of insect silk; whereas non-commercial type of silk is obtained from the weaver ants and green lacewing fly. The genera *Tasar*, *Eri*, *Muga* and *Anaphe* are the principal non-mulberry silk producers in the tropical or temperate regions. Others include *Fagara*, *Coan*, Mussel, Spider and *Gonometa* (Jolly *et al.*, 1979).

In the earlier days of the last century, an entomologist in Uganda (Gowdey, 1953) estimated, apparently on the evidence of local observation, that in the whole of Africa, *Anaphe* moth produced about two million kilograms of nests annually. According to Pinhey (1975), larvae of Thaumetopoeidae stay continuously together, often moving on trees or on the ground in columns. When fully-grown, they form a community cocoon, which is strongly welded together with silk. Others merely sew leaves together with silk to form their loose shelter.

2.2 Non – mulberry sericulture in sub-Saharan Africa

According to Bene *et al.* (1976), nearly 10 million hectares of the world's tropical forest is destroyed every year. The annual rate of forest loss in Africa has now reached 0.7%. The consumption of fuelwood doubled in the last 30 years of the 20th century and continues to rise at an annual rate of 0.5% (Clavreul, 2005). There is a danger that the short term benefit may override disadvantages, especially regarding the indigenous populations of forest. Non-mulberry sericulture holds a great promise for the world forestry as a supplementary activity (Jolly *et al.*, 1975). It can help arrest forest destruction, because, it permits gainful utilization of its vast natural wealth.

There are a number of records of *Anaphe* silk having been utilised in several parts of Africa and even in Uganda. *Anaphe* silk was one of Uganda's exports between 1910-1945 period (Mugenyi and Semakula, 1998). In Nigeria, silk weaving was practised before the colonial era and the early explorers found indigenous people weaving silk and cotton (Ashiru,1975a).

In Africa, most of the wild silkmoths belong to Saturniidae, Lasiocampidae, and Thaumetopoeidae families. The potential of the African indigenous silkmoth species for wild silk production has been documented in Uganda (Kato, 2000; Akai and Nagashima, 1999; Akai, 1999; Akai *et al.*, 1997; Iseki, 1985; Gowdey, 1953), Nigeria (Ashiru,1975a, b; 1986; 1988a, b; 1991), Kenya (Mbahin *et al.*, 2008; Ngoka *et al.*, 2008; Raina *et al.*, 1999; 2000; 2007; Raina, 2000; 2004; Ngoka, 2003; Kioko *et al.*, 1999a, b; 2000; Kioko, 1998), Botswana (Hartland-Rowe, 1992), Zimbabwe (Chikwenhere, 1992), Cameroon (Gérin, 1957; Malzy and Par, 1955), Madagascar (Razafimanantosoa *et al.*, 2006; Peigler, 1993), Angola (Rougeot, 1962) and South Africa (Veltman *et al.*, 2004; 2002); Van den berg, 1990; Delport *et al.*, 2006.

The quality of the African species of silkmoths is good; they give strong silk of high commercial value (Raina, 2004; Schultze, 1914). Cocoons of *Gonometa* sp. have been reported to occur on the African savanna where larvae feed on the mopane tree, *Colophospermum mopane* Kirk ex j. Leo (*Caesalpiniaceae*), a resiniferous tree (Hartland-Rowe, 1992). Cocoons are collected in the field and they give silk of a soft texture and of beige color (Peigler, 1993). A study by Hartland-Rowe (1992) indicated that in Botswana, collection and processing of this silk can offer a viable source of employment and income to people living in the villages. In Zimbabwe, Chikwenhere (1992) reported that from 1986 to 1987, rural families collected 430 tons of the wild silkmoth cocoons and this became a source of employment in the rural areas. *Gonometa* larvae also have urticating spines and are found feeding on *Acacia* species such as *A. hockii* and *A. mearnsii* and podo trees in East Africa (Ngoka *et al.*, 2008) and on *A. tortilis, A. nilotica, A. elatior,* and *A. mellifera* (Ngoka, 2003). In the Arabuko Sokoke forest the larvae of the wild silkmoth *Argema mimosae* feed on *Shweinfurthii, Sclerocarya birrea, Ozoroa insignis* (Ngoka *et al.*, 2008). Akai *et al.* (1997) detected needle like bristles 2 – 3 mm along all over the cocoon, and their role is to offer protection against enemies such as birds and other vertebrates. *Anaphe* silk of southern and central Africa is produced by silkworms of the genus *Anaphe*: which include *A. moloneyi* Druce, *A. panda* Boisduval, *A. reticulata* Walker, *A. ambrizia* Butler, *A. carteri* walsingham, *A. venata* Butler and *A. infracta* Walsingham. They spin cocoons in communes, all enclosed by a thin layer of silk. Local tribes in Nigeria collect them from the forest and spin the fluff into a raw silk that is soft and fairly lustrous (Ashiru, 1986). The fabric is elastic and stronger than that of the mulberry silk (Jolly *et al.*, 1979).

The large Thaumetopoeidae family is composed of *Thaumetopoea processionea* Hubner, *T. apologetica* Strand, *A. reticulata* Walker, *A. panda* (Boisduval), and *Epanaphe clarilla* Aurivillius moths (Pinhey, 1975). According the same author, Thaumetopoeidae silkmoths show highly gregarious community habits (Plates 2.1 & 2.2). About nine species are known in southern and central Africa. Butler (1878) was one of the pioneers to observe and describe *Anaphe* sp. This species is better known for its socio-economic importance. The order Lepidoptera to which the wild silkmoths, *A. panda* belong contains a large number of edible species (Ashiru, 1988b). These moths have large fleshy larvae, which are highly nutritious. Oliveira *et al.* (1976) reported that one hundred grams of caterpillars would provide 76% of the individual's daily protein requirement and more than 100% of the daily requirements of many of the vitamins and minerals. Larvae are eaten by peasant farmers in Nigeria (Ene, 1963; Golding, 1942) and contain substantial quantities of crude protein and mineral elements such as iron, phosphorus, calcium and magnesium (Ashiru, 1986). Cocoons (Plate 2.3), covering sheath are used to produce silk which is woven to produce silk in Nigeria (Ashiru, 1986; Ene, 1965).

2.3 Wild silkmoths in Kenya

There are over 60 wild silkmoth species in East Africa and their high potential has not been utilized (Raina, 2000; 2004; Kioko 1998). Despite this richness and abundance, only a few of these species have so far been utilized for wild silk production in East Africa. Studies by Kioko (1998) showed that *Gonometa* sp. occurred in Mwingi District and Sultan Hamud in Makueni District; the host plants being *Acacia elaitor* and *Acacia senegal*. According to Ngoka (2003), *Gonometa* sp. was also found to occur in Kamaguti in Uasin Gishu District; the host plants being the indigenous *Acacia hockii* and exotic wattle tree (*Acacia mearnsii*). The chorion structure *of Argema mimosae* (Kioko *et al.*, 1999b), developmental cycle of *Gonometa postica* (Ngoka *et al.*, 2008), and spatial distribution of the silk cocoon nests and egg-clusters of the silkmoth *A. panda* and its host plant *B. micrantha* in the Kakamega Forest (Mbahin *et al.*, 2008) has been studied.

Plate 2.1: *Anaphe panda* larvae procession

Plate 2.2: *A. panda* larvae resting before moulting

Plate 2.3: *A. panda* dry cocoon nests

2.3.1 Population dynamics of *A. panda*

Although *A. panda* egg clusters (Plate 2.4) can contain from 200 to 300 eggs (Gowdey, 1953), or 250 to 350 eggs (Jolly *et al.*, 1979), the population of wild silkmoth fluctuates. This is because there are many mortality factors at work throughout the life cycle, all of them varying in time and space. Some of these factors are biotic (parasites and predators) and may also include bacterial and viral diseases as well as the availability of food (Hartland-Rowe, 1992). The other factors are abiotic and these are mostly climatic and include temperature, humidity, precipitation and weather. According to Hartland-Rowe (1992), climatic factors play an important role in regulating the population density of many species of silkmoth. This not only regulates the timing of the moth emergence; the induction and termination of diapause but they also have great indirect impact by regulating the timing of the availability of food. However, the impact of parasitoids and predators on the larval stage is often far more severe than that of climatic factors. Although loss of *Anaphe* larvae due to diseases is negligible (Jolly *et al.*, 1979), the silkworm has been reported to suffer from muscardine (Rolet and Par, 1948), and is attacked by the tachinid fly, *Exorista cardinalis* Fabr and the ichneumon wasp, *Cryptus leucopygus* Granenhorst (Jolly *et al.*, (1979). These two larval parasitoids are responsible for 30-35 % of the larval mortality. According to Jolly *et al.* (1979), the egg is also parasitized by two chalcids (*Telenomis gowaeyi* Grahan and *Pleurotropis telenomis* Lima). Generally, *Anaphe* is vulnerable to many parasitoids and predators (Jolly *et al.*, 1979; Gowdey, 1953). Among the predators, birds cause considerable mortality.

2.3.2 Economic potential of *Anaphe* culture

In Africa, during the last three decades, there has been a decrease in the production of traditional export crops, as well as the unsustainable exploitation of indigenous forests and agricultural land. Among the many activities that might assist the poor to escape their vicious cycle of poverty, are the raising of bees and silkworms referred to as of apiculture and sericulture respectively (Raina, 2000; 2004; Raina *et al.*, 1999). In East Africa biodiversity surveys in the indigenous forests indicate that there are at least 60 identified different silkmoth species in the wild. Preserving and utilising these silkmoths for the production of silk fibres would generate income for the resource-poor farmer, as well as conserve the threatened natural forests (Kioko *et al.*, 2000). In countries where rural communities depend on subsistence farming, wild silkmoth farming can be a supplementary activity for income generation while at the same time conserving biodiversity.

According to Jolly *et al.* (1979), by applying the available knowledge and filling in the crucial gaps with well-focused research, a large increase in raw silk production can be realized in a number of countries where global potential of non-mulberry sericulture exists. As long as humans desire for silk garments continues, the demand for sericulture activity remains. Silk is the "queen" of textiles and the naturally produced animal fiber. The silk industry is having increased demand than natural production can satisfy, hence, the necessity of seeding. This involves the release in the nature of moths that have been reared in captivity and the release of cocoons placed in the net-sleeves.

Plate 2.4: *A. panda* egg-cluster on the host plant *B. micrantha*

Plate 2.5: *A. panda* host plant *B. micrantha* (Hochst)

Plate 2.6: A view of Kakamega Forest western Kenya

Unfortunately, *Anaphe* silkmoth is univoltine (Akerele, 1970; Gowdey, 1953) and undergoes diapauses at the pupal stage.

One hectare of plantation can accommodate about 2,200 plants of *B. micrantha* with 3.0 x 1.5 m spacing with a possible yield of 25-27 metric tons of leaves. According to Jolly *et al.* (1979) this is enough to sustain 6,500-7,000 layings eggs to produce 700-800 kg of cocoons, equivalent to 340-360 kg of unprocessed silk. At only Ksh 200 per kg of cocoon nests, a farmer can raise KSh 140,000 per year (Raina, 2000).

CHAPTER THREE

3 GENERAL MATERIALS AND METHODS

3.1 Study area

This research was carried out in two-study sites named: Ikuywa (indigenous forest) and Isecheno (mixed indigenous forests) within Kakamega Forest (Fig. 3.1 and Appendix 1) (Kakamega forest, 2000).

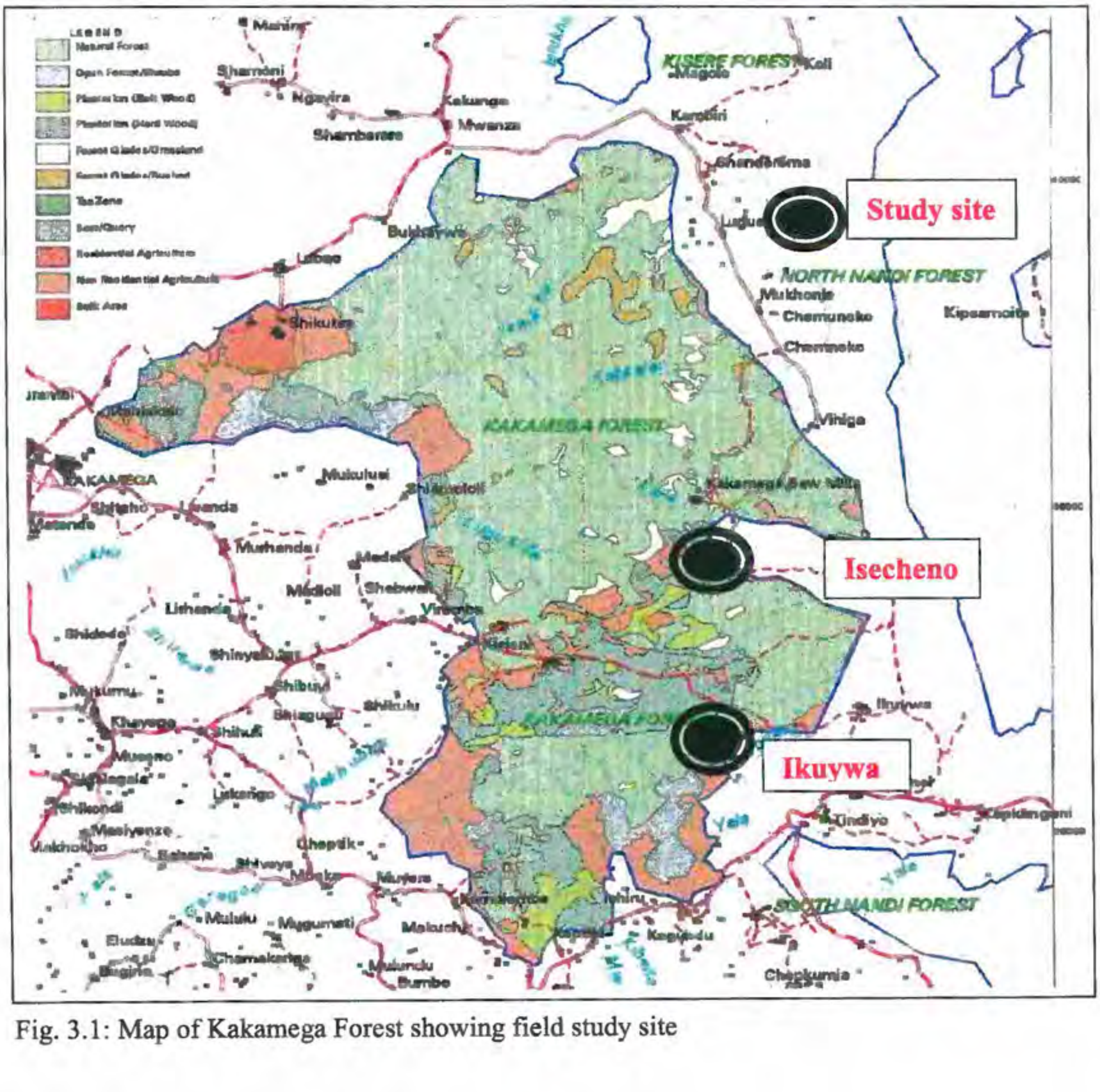


Fig. 3.1: Map of Kakamega Forest showing field study site

Research work was aimed at investigating the ecology and economic potential of the wild silkmoth *A. panda* (Boisduval). The Kakamega Forest is located in Kakamega and Vihiga Districts of Western Kenya (Appendix: 1). The forest, along the north-eastern edge of Lake Victoria Basin extends between Latitudes 0° 10' and 0° 21' North and 34° 47' and 34° 58' Longitude East. The entire Kakamega Forest covers an area of approximately 265 Km^2 (Muriuki and Tsingalia, 1990)

Its terrain varies greatly with altitude, and ranges between 1520-1680 meters above sea level (Kokwaro, 1988). The forest was first gazetted as a trust land in 1993, although two small Nature reserves, Yala and Isecheno Forest totaling about 700 ha had been established within the Forest Reserve in 1967 under the management of the Kenya Forestry Department (KIFCON, 1994). From 1986, nearly 4,000 ha of the northern portion of the forest, Buyangu or the Kakamega Forest National Reserve, along with the adjacent 471 ha comprising Kisere Forest, were formally excised and placed under the management of the Kenya Wildlife Service (K W S). These two blocks consist almost entirely of the indigenous forests (KIFCON, 1994). Mutangah *et al.*, (1992) recorded the Buyangu block (K.F.N.R) as having the highest tree density of all the Kakamega Forest plots. The Kakamega Forest complex lies mostly to the west of the Kakamega Town about 40 km north of Lake Victoria. It comprises several separate blocks of forest: Kakamega (13,878.2 ha), Bunyala (825.6 ha), Malava (722.8 ha), Kabiri (3,691.3 ha), Lirhanda hill (52.7 ha), Kisere (471.4 ha), Yala river (289.5 ha), Isecheno (415 ha), Ikuywa (380 ha) and Buyangu (3,997.5 ha). The entire Kakamega Forest area has relatively flat to gently undulating topography except for a few steep hills like the Buyangu to the north and the Lirhanda to the south. To the east, it is bound by the Nandi

Escarpment, which rises up to 2,200 meters above sea level. The terrain is undulating, with often steep-sided river valleys.

The main vegetation in the Kakamega Forest is rainforest vegetation type. There are also a variety of other habitats including indigenous forests, colonising forests, disturbed forests and areas of planted forests, glades and riverine forests (Mutangah *et al.*, (1992).

Kakamega Forest is classified as a tropical rainforest situated in a fairly wet area of Kenya with an average annual rainfall of 2,025 mm (Emmerton, 1991). However, it has been reported that the average annual rainfall may be decreasing due to the deforestation (Kokwaro, 1988). Zimmerman (1972) had recorded over 3,500 mm annual rainfall during 1963, indicating that, the has been substantial fluctuations from year to year.

3.2 Biology of *A. panda*

3.2.1 The adult stage

Anaphe panda population for the experiments were set up by collecting live *A. panda* cocoon nests which were used to start the initial breeding stock. Fresh healthy cocoons selected for moth's emergence were divided in two groups. The 1st group was placed in well-ventilated insect rearing cage (2m x 2m x 2m) in the field (Plate 3.1), and the 2nd group was dispatched in the net-sleeves (1.5 x 1.5 x 2 m) attached to the host plant *B. micrantha* in the field (Plate 3.2). The two groups of cocoon nests were observed until moth emergence (Plate 3.3).

Plate 3.1: *A. panda* cocoon nests in the insect rearing cage

Plate 3.2: *A. panda* cocoon nests in the net sleeve on *B. micrantha*

Plate 3.3: *A. panda* silkmoth emerge from cocoon nests

The following data were recorded: wing expanse, sex ratio, life span, and general behavior of the first day emerged moths. Sex ratio was calculated and recorded by placing a single cocoon nest in a small net-sleeve (60cm x 40cm x 30cm) in the field.

To obtain data on fecundity and mating time of silkmoths, 10 pairs of virgin silkmoths were put together for observation in the net-sleeve measuring (60 x 40 x 30 cm) and attached on the host plant *B. micrantha* (Plate 3.4). After copulation in the net sleeve, the egg laying period and egg fecundity (total egg production) were observed and recorded. Fecundity represents the number of eggs deposited plus the mature eggs present in the oviduct after death (Southwood, 1968; Klomp, 1966).

3.2.2 Egg stage

In the field, a protective shell of the egg cluster was observed and color of the egg recorded. The number of perforations by parasitoids on the cover shell of egg clusters was counted. The average diameter and weight of a random egg cluster were determined. The number of eggs in an egg-cluster was counted from freshly laid eggs obtained from the female moths in the net sleeve. The total number of eggs produced (fecundity) by a random sample of female moths was determined by counting the number of egg laid and those encounter after dissection. The incubation period was determined from freshly laid eggs obtained from the female moths in the net sleeve and the exact period of incubation was recorded.

Plate 3.4: Silkmoths mating in the small net sleeve

Plate 3.5: Digital hygrothermometer for recording temperature and RH

Plate 3.6: Rain gauge for recording rainfall

3.2.3 Larval stage

The numbers of larval instars were counted and their body colors were described. The duration of the larval development in the field under fluctuating weather conditions was observed through the rearing of one hundred and eighty (180) cohorts of more than three hundred larvae each of *A. panda* on the *B. micrantha* host plant until they spun cocoons. Each sample of *B. micrantha* was having two experimental areas: one with a net-sleeve measuring 1.5 x 1.5 x 2 m for one cohort of larvae and another unprotected area for another cohort of larvae. Determination of the number of instars was done by direct observation of cast exuviae of the larvae (Plate 3.9). This is a method that was used by Ashiru (1988a) and Schmint *et al.* (1977).

A recording digital hygrothermometer (Plate 3.5) was used to record daily maximum; minimum temperatures and relative humidity. Rain gauge (Plate 3.6) was also used for recording the rainfall data.

3.2.4 Pupal stage

The pupa is obtect (has all its appendages encased against the body) and enclosed in a cocoon nest in the field. The wall of this cell is sometimes reinforced by the use of other materials spun together with silk threads. The average number of pupae per cocoon nest were counted and recorded by dissecting cocoon nests (Plate 3.7). Electronic balance was used to determine the average weight of the cocoon nest. To determine pupal period, marked cocoon nests were kept in enclosed net-sleeves measuring (60cm x 40cm x

30cm) till moth emergence. The pupal period was determined by marking cocoon nests at the date of spinning and observing them until the date the moths emerged.

3.3 Parasitism and predation aspects

In order to develop a sustainable and ecological mass production of *A. panda* cocoons, it would be necessary to identify, evaluate and compare the impact of natural enemies on the developmental stages. *A. panda* silkworms are usually reared in the fields where they are exposed to unfavourable natural conditions such as birds, insects, pathogens and severe weather. Mortality factors of larval stages at each instar were determined by rearing 180 cohorts (ninety-four protected and eighty-six unprotected) each consisting of more than three hundred larvae on the host plant. Counts of the surviving larvae in each host plant were made after each moult. The natural enemies of egg, larva and adult stages were also determined by visual observations twice a week on the host plant *B. micrantha*. The natural enemies of pupal stage were recorded by observing, collecting and checking the cocoons in the field. Exposed eggs collected from the field were kept in the tubes for eventual emergence of egg parasitoids and their identification was made at the ICIPE biosystematics' unit and the National Museum of Kenya.

3.3.1 Survival analysis of silkworm's

Survival analysis was used for the protected and unprotected larval stages of *A. panda* on *B. micrantha* during the rainy and dry seasons.

Plate 3.7: Open *A. panda* cocoon nest

Plate 3.8: *A. panda* silkworms preserved in Bouin's fixation solution

Plate 3.9: Observation of cast exuviae of *A. panda* silkworms after moulting

The numbers of individuals that entered a particular stage in each generation were determined from the successive samples of the populations. The incidence rate or instantaneous risk represents the theoretical probability for a larva to die at a particular instar. The incidence rate that is the number of deceased larvae during a specific instar and period in the specified population was computed as outlined by Claves *et al.* (2004) and Statacorp (2004). The maximum-likelihood proportional hazards models were estimated (Lin and Wei, 1989; Muller and Wang, 1994; Gutierrez *et al.*, 2001). The Cox proportional hazard model was used to find out how independents variables like habitats, protection, brood years affected positively or negatively the hazard (Cox and Oakes, 1984; Cox, 1972).

One hundred and eighty (180) cohorts of *A. panda* silkworms which completed the seven larval stages on the host plant *B. micrantha* were investigated during the survival analysis study. The unprotected cohort of larvae which disappeared or the cohort who died before the end of the experiment were investigated but they were not included in the analysis. Counts of the surviving larvae on each host plant were made and recorded after each moult.

3.3.2 Histopathological parameters

Random sample of larvae were observed with light microscope and analysis of abnormal microscopic changes in the silk gland structure of silkworm were made. Infected larvae were sampled and preserved in glass vial (7.5 x 2.5 cm) with Bouin's fixation (Gretchen, 1962) (Plate 3.8). Freezing method (Gretchen, 1962) was used to make a block for the dissection of the parasitized larvae. Tissues were from the posterior, middle and anterior silk gland sections. Fibroin is secreted in the posterior section and transferred by peristalsis to middle section, which acts as a reservoir. Here it is stored as a viscous aqueous solution until required for spinning. The majority of the sericin is created within the walls of the middle section. Microtome was used for block cutting and stained with different stains (Giemsa stain or Fuchsin basic). Light microscope was used to observe tissues and silk gland slides.

3.4 Spatial distribution of *B. micrantha*, cocoon nests and egg-cluster

Information is required on spatial distribution of wild populations to assist in developing management plans for conservation and their sustainable utilisation for income generation. The distribution of *A. panda* silkmoth egg-clusters, cocoons nests and the host plants *B. micrantha* was assessed in two different habitats (indigenous forest and mixed indigenous forest) using the procedures outlined by Mbahin *et al.* (2008). The analytical procedure to determine the distribution of forest insects within a tree has been also outlined by Morris (1955). The "basic population" refers to a population with respect to a basic unit, which will permit conversion of the results to absolute population numbers. Embree (1965) considered the only common unit of measurement for basic populations to be the whole tree in the population dynamics study of winter moth. There are different methods to assess population numbers for the various stages (Southwood, 1968; Klomp, 1966).

3.5 Economic profitability of *A. panda* sericulture

Sericulture is a new industry being developed in Africa. The technologies will enable small landholders to increase their economic well being through self-reliance (Raina *et al*., 2000). The profitability on any enterprise is the key to its success and to this end, the evaluation of the sustainability of the product should be addressed. The evaluation study included: production system, socio-economic profiles, resource endowments and technological needs of the rural farmer. The net income gained by the household, by selling cocoon nests, or process the cocoon nests to raw silk, silk cloth, or silk shirts was determined. The net added-value was calculated at each level of the processing and the cost structure plus optimal profit calculated.

3.6 Data analysis

T-test and 3-ways analysis of variance (ANOVA) (SAS, 2003) were used to compare between the mean biological parameters of the two blocks: "Isecheno block (mixed indigenous forest) and Ikuywa block (indigenous forest)". Chi-square (χ^2) was used to compare between the expected and observed number of *B. micrantha*, cocoon nests and egg-clusters in the two blocks. ArcView GIS 3.2 (Garnin©, 2002) software was used to map georeference data. Isecheno and Ikuywa transect surface plots were mapped using a computer Surface Mapping System Surfer 6.0 (Surfer 6.0, 1995). ΣSigmat plot 2000 (SPSS, 2000) software was used to draw all the graphs. Cox proportional hazard model was used to analyze larvae survivor data (Cox and Oakes, 1984; Cox, 1972). Test of equality of survivor functions was done with Log-rank test (Lacatos and Lan, 1992;

Hsieh, 1999). The Newman-Keuls test (Miller, 1981) with unequal sample sizes was used for multiple comparisons test between experimental sites. Correlation and multiple regression analysis using the maximum-likelihood proportional hazards models (Statacorp, 2004) were used to analyze factors that are largely responsible for the observed changes in population. Poisson distribution (Dale, 1989) and negative binomial distribution (Anscombe, 1948) were used to calculate random occurrences. The degree of significance was indicated conventionally as follow:

*: Significant ($P < 0.05$)

**: Highly significant ($P < 0.001$)

^{ns}: Non significant ($P > 0.05$)

CHAPTER FOUR

4 BIOLOGY OF WILD SILKMOTH *ANAPHE PANDA* (BOISDUVAL)

4.1 Introduction

Anaphe panda (Boisduval) (Lepidoptera: Thaumetopoeidae) like other lepidoptera undergoes complete metamorphosis (change in form during post-embryonic development) consisting of four distinct stages namely: egg or ovum (the embryonic stage), the caterpillar (larva) or silkworm (the principal feeding and growing stage), the chrysalis stage or pupa (a dormant transition stage) and the adult or imago (the principal dispersal and reproductive stage). Hence, this research was undertaken in the Kakamega Forest to study the biology and life history stages of *A. panda*, whose larval stage feeds on *B. micrantha* (Hochst) (Euphorbiaceae).

4.1.1 Egg or ovum

Eggs of *A. panda* are usually laid in clusters (Plate 4.2). The female clings tightly to the underside of the leaf of the food plant and starts rubbing the tip of her abdomen to construct a covering layer using the golden brown hairs found on her abdominal extremity. The egg laying continues for 10-12 hours. The female moth usually glues her egg cluster to the back of the foliage of the food plant *B. micrantha*. The mass of eggs is protected by the golden brown hairs, which come from the anal segments of the female. By turning circular abdominal contractions on the freshly laid eggs, the female moth removes these golden brown hairs from her anal tuft covering the eggs. The freshly laid eggs are soft and creamy white and covered with a purplish brown secretion that cements the eggs to one another and to the surface of the leaf on to which the eggs are fastened. When fresh, this substance is soft, and becomes hard when dry. Moths start laying eggs from mid-November until mid-May and egg hatching is completed in July.

4.1.2 The caterpillar (larva) or silkworm

When tiny silkworms (Plate 4.20E) of *A. panda* first emerge from the eggs, they eat their eggshell and thereafter begin to feed on the host plant *B. micrantha.* At first, they prefer the underside of the leaf (Plate 4.3). As they grow, they migrate to other leaves but all the 1st instar (stage) larvae feed exclusively on the underside of *B. micrantha.* The early instars relish tender leaves, and the advanced stages prefer medium to mature leaves. The rate of feeding is minimal before and after the moult (ecdyse) (Plate 3.9), but reaches its peak between these two periods. Silkworms have soft and cylindrical bodies. They are green from 1st to 3rd instar and grow dense white hair from 4th to 7th instar. The silkworms (Plate 4.20F) have rounded, hardened head capsules and the chewing mandibles. They feed in groups during their larval period. They feed from the margin to the centre of the leaf, but they never eat the midrib. The younger larvae rest on the leaf, while the older ones attach themselves on the bark of *B. micrantha.* They move in a line (Plate 2.1) following a thread of silk laid down by the front most silkworm (captain). For protection from enemies like ants and spiders, all the feeding leaf surfaces and resting areas are preminary coated with silk. As they feed, they outgrow their exoskeleton and to provide room for further growth, they undergo six moults (ecdyses). Each time they moult the old exoskeleton cracks along the back and the caterpillar crawls out. The newly formed exoskeleton, which lies beneath the old one stretches and hardens soon after the old one is shed.

4.1.3 The chrysalis or pupa

When silkworms reach the mature size, they seek a protected place where they can pupate. Silkworms spins together a tight cocoon (Plate 4.20G) of silk, which it secretes from modified salivary glands and spins using a spinneret located below the mandible. Plant material may be used for support when spinning the cocoon. In the cocoon nest, when the caterpillar is ready to pupate, it moults one last time into the virtually inactive stage called the pupa (Plate 4.5). *Anaphe* pupa is obtect (has all its appendages encased against the body) and the exoskeleton is shiny brownish. Beneath the exoskeleton, substantial biochemical and anatomical changes occur, as larval features are replaced by adult structures such as antennae, proboscis, wings, and sex organs. The pupal shape is cylindrical with the cephalic end rounded. Male pupae have the genital opening ventromedially on the six abdominal segments. The genital aperture of the female pupa is present ventromedially on the anterior margins of the fifth and six abdominal segments.

4.1.4 The adult or imago

The emerging moth (Plate 4.1a, b) breaks through the pupal exoskeleton and drags itself out.

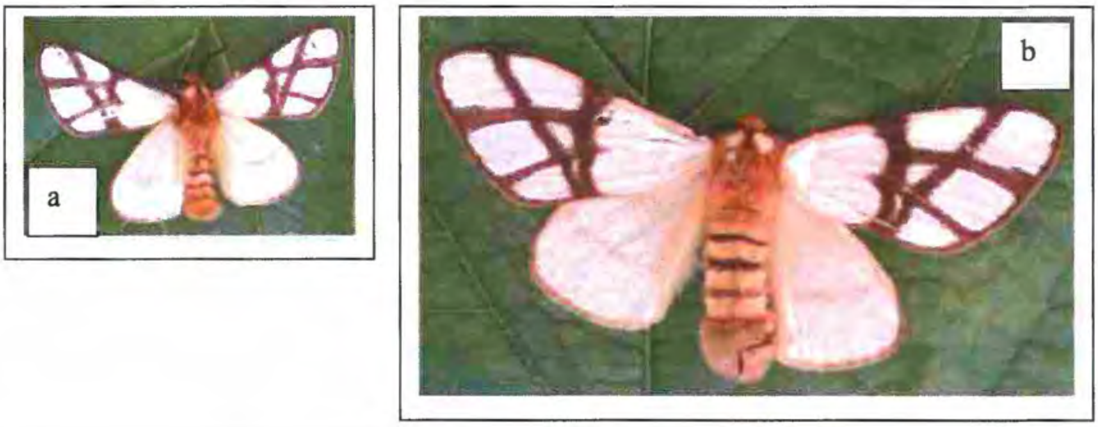


Plate 4.1: Male (a) and female (b) of *A. panda* adults moths

Plate 4.2: *A. panda* egg cluster on *B. micrantha* leaf

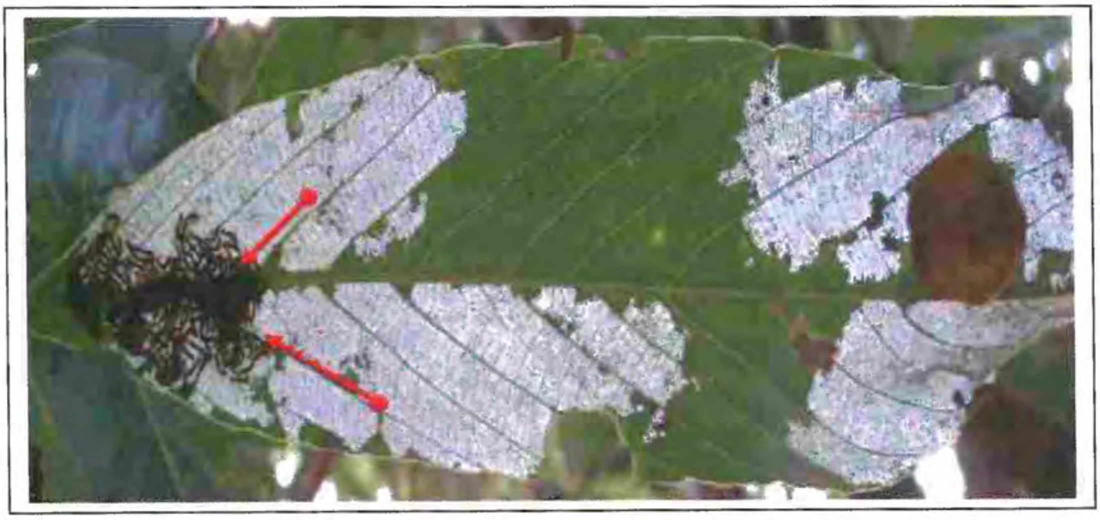


Plate 4.3: 1st instar *A. panda* silkworms feeding on underside of *B. micrantha* leaf

The cocoon of *A. panda* has a trap door (Plate 4.4) that pops open when the moth pushes against it. Before emergence, the moth secretes a colourless brown fluid that helps moisten the exit channel. Once out of the cocoon, the moth crawls onto it or some other support and begins to pump blood into the veins of the wrinkled wings. Expansion and hardening of the wings may take an hour or more, after which the moth takes flight to seek a mate. Males usually perform courtship behaviour and secrete pheromones to make the female receptive for mating. Pheromones may be secreted from wings, abdomen, and legs, and distributed by means of tufts of special hair-scales on those regions.

If the female is receptive to the male, she permits him to clasp the tip of her abdomen with the flaplike valves of his genitalia. The aedoeagus (penis) is inserted through her ostium bursae and into the ductus bursae and copulation may last several hours (Plate 4.6), after which the two moths part. The female starts laying eggs (Plate 4.8) as soon as they decouple.

Information on sex ratio is also required for contribution of life table (Pottinger and LeRoux, 1971) as well as in the development of population models and in the analysis of generation survival (Morris, 1963). As pointed out by Miller (1963), the sex ratio of an insect may refer to the various developmental stages. Differential mortality can result in the change of the sex ratio. The sex ratio of the moth is of primary importance in the determination of the population size and its potential for increase (Zwolfer, 1934).

Plate 4.4: Trap doors on the cocoon nest of *A. panda* silkmoth

Plate 4.5: *A. panda* silkmoth pupae

Plate 4.6: *A. panda* mating activity

4.2 Materials and methods

The initial population for the experiment was set up by collecting fresh *A. panda* cocoon nests to initiate the breeding stock. The collected *A. panda* cocoon nests were enclosed in 2 x 2 x 2 m net-sleeves cages in the field (Plate 3.1) and 1.5 x 1.5 x 2 m net-sleeves attached on *B. micrantha* (Plate 3.2). The two groups were observed until the moths emerged.

The wing span was determined by measuring with Vernier calliper the maximum distance between the tips of the forewing and hind wing after spreading the moths. This was the measurement from the apex of the forewing or hind wing and its attachment to the thorax. The length and the width of forewing and hind wing, the length of antenna and the diameter of head capsule of moths emerging from the breeding cages were measured. This experiment was repeated similarly in the consequent years (2005-2007).

The sex ratio of the breeding populations was calculated from *A. panda* silkmoth rear from the breeding cages and net sleeves in both blocks: Indigenous forest (Ikuywa block) and the mixed indigenous forest (Isecheno block).

A survey was carried out each year during the dry season (November-February) up to the rainy season (March-May) which was the period of the moth's emergence. Sex ratio of one *Anaphe* cocoon nest was made by placing a random sample of a single cocoon nest in the net sleeve measuring (60cm x 40cm x 30cm) and attached on *B. micrantha* (Plate 4.7).

Plate 4.7: Cocoon nest in the net sleeve for moth emergence

Plate 4.8: The *A. panda* silkmoth laying egg-cluster on *B. micrantha* leaf

Plate 4.9: Weighing of *A. panda* egg-cluster

A chi-square (χ^2) test of proportion "with Yate's correction for continuity; (Zar, 2005; David, 1995; Dyke, 1995; Yates, 1934)" was performed on the recorded sex ratio to find out if the sample data (silkmoth) came from a population having a 1:1 ratio of male to female. The Yates correction is routinely used when the degree of freedom (DF) is equal to 1 and it is not applicable when DF>1.

To determine female productivity , pairs of freshly emerged female and male moths were isolated from the net sleeve and enclosed in the small net sleeve (60 x 40 x 30 cm) attached on *B. micrantha* (Plate 3.4) for mating and oviposition. One hundred and-three (103) replications "Isecheno block: (54) and Ikuywa block (49)" of the paired moths were used in three years. The total number of eggs by egg-cluster laid in the net-sleeve were recorded. The life span of both male and female moths was also recorded for comparison.

Fecundity is a measure of the total egg production (Southwood, 1968), and according to Klomp (1966), fecundity represents the number of eggs oviposited plus the mature eggs present in the oviducts after death of the female. To determine the fecundity of the females, oviducts of sixty-six dead females enclosed in the small net sleeve (60 x 40 x 30 cm) were dissected during the study and the total number of mature eggs was counted and added to those of the egg cluster. The total number of eggs produced by a female was determined by adding the number of eggs of the egg-cluster to mature eggs obtained through dissection of the dead female. The dead females were preserved in 70% alcohol for at most a month before dissection under a microscope.

For egg size, one hundred eggs obtained from one hundred *A. panda* moths were measured using a stereomicroscope with an ocular micrometer. The mean length of the major and minor egg axis were compared using analysis of variance (Proc anova, SAS Institute, 2003). A Vernier calliper was used to determine the average length and width of eighty-three freshly laid egg clusters. The weight of the eggs together with golden brown hairs was determined by electronic balance (Plate 4.9). The mean of the weight was were compared between the two blocks of the forest using the Student-Newman-Keuls (SNK) procedure (SAS Institute, 2003). The micropyle (the opening that admits the sperm that fertilizes the egg nucleus), and aeropyles (microscopic holes in the shell which allow gas exchange between the developing embryo and the atmosphere) were examined and photographed using a scanning electron microscope (Joel JSM-T 330 A).

One hundred and ninety five egg-clusters laid daily (Plate 4.8) were marked to determine the exact incubation period. Daily observations on hatching were made after thirty-five days. Egg clusters were also divided into two batches according to the season: dry season from mid-October to February (earlier brood) and rainy season from March to mid-May (later brood). Daily temperatures and relative humidity were recorded by a digital hygrothermometer (Plate 3.5).

The larval period was determined during the three years period by following one hundred and eighty (180) cohorts of *A. panda* silkworms on *B. micrantha* until they span cocoon. The rearing was done by protecting the entire larval stage of ninety-four (94) cohorts with net sleeves (Plate 4.10), and unprotected another eighty-six (86) cohorts (Plate 4.11).

Plate 4.10: Larval stage of *A. panda* in the net sleeve

Plate 4.11: Unprotected 7th instar silkworms of *A. panda*

Plate 4.12: Marked *A. panda* cocoon nest kept in the enclosed net sleeve

The number of instars was determined by visual observation of exuviae (Plate 3.9) (Ashiru, 1988a; Schmidt *et al.*, 1977).

Younger silkworms rest about two days on the leaf, and older ones (from three to four days) attach themselves on the bark of *B. micrantha* before moulting (Plate 2.2). Hence, observations were made daily around 10h 00 am to record if the larvae had cast their skin (exuviae) (Plate 3.9). The total number of observed moults was used to determine the number of intars. These replicates were also used to identify the possible mortality factors during the larval stage.

To determine the quantity of leaves eaten by one silkworm, the number of silkworms that hatched was counted and recorded at the end of each instar with the help of a binocular lens and tally counter. The number of midribs (the larvae feed from margin to the centre of leaves and they never eat the midrib) were counted and removed after every four days from 1st to 4th instar, and also after every two days from 5th to 7th instar. Knowing the average weight of *B. micrantha* leaves, the numbers of leaves eaten during the instar and the number of larvae at the end of the instar; the mean consumption per larva was calculated and recorded.

The average length of feeding and resting period by instar was investigated by following in two years (2006 – 2007) twenty five cohorts of silkworms. At each instar, the feeding and resting periods were recorded.

Plate 4.13: Length measurement of female *A. panda* pupa with Vernier calliper

Plate 4.14: Weighing sample of male *A. panda* pupa using balance

Plate 4.15: Weighing sample of female *A. panda* pupa using balance

To determine the pupal period, spinning dates of 100 cocoon nests were noted and kept in enclosed net sleeve (60 x 40 x 30 cm) in the field until moths emerged (Plate 4.12). The length and width of male and female pupae randomly sampled from the two blocks was determined by a Vernier calliper (Plate 4.13). The weight of male (Plate 3.14) and female pupae were also obtained (Plate 4.15) by use of an electronic balance, Mettler PJ 360 DeltaRange®.

The sex ratio of the pupae was also investigated (calculated) by dissecting a randomly sampled cocoon nests from the two blocks at the pupal stage (Plate 4.5). A chi square (χ^2) test of proportion "with Yate's continuity correction; (Zar, 2005; David, 1995; Dyke, 1995; Yates, 1934)" was also performed on the obtained sex ratio to know if the sample data (pupae) came from a population having a 1:1 ratio of male to female at the pupal stage.

The time of emergence of moths was readily determined by direct observation and also by the marked pupae kept in the net sleeve attached on *B. micrantha* (Plate 4.12) in the field.

The Kolmogorov-Smirnov test procedure for discrete data (Smirnov, 1939a, b; Kolmogorov, 1933) was used to test the null hypothesis that each tree crown levels of *B. micrantha* (lower, middle and upper) are equally desirable by *A. panda* moth for resting. A total of sixty-nine moths were used for assessing goodness of fit of the observed and expected cumulative frequency distribution.

4.3 Results

4.3.1 Adult

The moth emergence commences at dusk and in the early evening. The wings of the moth are creamy white (Plate 4.1a, b). The forewing has two broad bands in the middle joining at posterior margin to form a V and two sub parallel bands from the outer arm of the V to the lateral margin. The lateral margins of both fore and hind wings are bordered with dark brown bands, less pronounced in the hind wings. The abdominal segments are marked with rings of golden brown hairs. The posterior extremity of the body of the female is covered with golden brown hairs.

4.3.1.1 Wing span; length of fore, hind wings and antennae, and diameter of head capsule

The mean wingspan of forewing and hind wing, the mean length of forewing, hind wing and antennae, and the mean diameter of head capsule of *A. panda* are summarize in table 4.1. There was a highly significant difference ($P < 0.001$) between all the variable for both male and female (Table 4.1). Repartition by years and type of forests (indigenous and mixed indigenous forest) and sex are summarized in both tables 4.3 and 4.4. Significant difference was observed between the two sexes and through out the three years, and also between the two forests habitats (Tables 4.3 and 4.4).

Table 4.1: Mean (±SE) wing span of fore and hind wings (cm); length of fore, hind wing and antennae (cm), and diameter of head capsule (mm) of *A. panda*

Variables	Males (n=2701)	Females (n=3088)
Forewing span (cm)	2.347±0.216	3.019±0.251**
Hind wing span (cm)	1.789±0.262	2.176±0.208**
Forewing width (cm)	1.137±0.106	1.448±0.138**
Hind wing width (cm)	1.142±0.167	1.623±0.163**
Length of antennae (cm)	0.865±0.06	0.8±0.07**
Diameter of the head (mm)	0.451±0.071	0.562±0.064**

**: Highly significant difference between males and females; n: Number of moths

4.3.1.2 Adult sex ratio

During the three years study period, the proportion of male to female moths was determined from the samplings which were carried out in the indigenous (Ikuywa block) and mixed indigenous (Isecheno block) forests. After performing the χ^2–test on the sex ratio, the obtained value was compared to the critical value ($\chi^2_{0.005,\ 1} = 7.879$). Values above 7.879 indicated that the sex ratios deviated significantly from the unit (1) and this indicated that sample data did not come from a population having 1:1 ratio of male to female (Table 4.2). If the obtained sampling value is below 7.879, the sample data comes from a population having 1:1 ratio of male to female. Field observation showed that sex ratio of moth's changed daily, but the pattern of emergence at the beginning of the moth emergence period (November-December) was very similar for both sexes with male moths being the majority. However, during the later period of the moth's emergence (March-April), the sex ratio changed from the male dominance to the female dominance.

Table 4.2: Adult sex ratios of *A. panda* from the indigenous and mixed indigenous forests in Kakamega (years 2005 - 2007)

Years	Isecheno block (mixed indigenous forest)						Ikuywa block (indigenous forest)					
	Observed moths		Sex ratio		Expected Sex ratio	χ^2	Observed moths		Sex ratio		Expected Sex ratio	χ^2
	m	f	m	f	m:f		m	f	m	f	m:f	
2005	238	526	1:	2.21	1:1	107.812^{ns}	391	407	1:	1.04	1:1	0.282*
2006	442	424	1:	0.959	1:1	0.334*	611	645	1:	1.055	1:1	0.867*
2007	391	492	1:	1.258	1:1	11.325^{ns}	628	594	1:	0.946	1:1	0.891*

* Moth sampling coming from a population having 1:1 ratio of male to female, m: male, f: female

$^{ns:}$ Moth sampling not coming from a population having 1:1 ratio of male to female.

Nevertheless, the general trend of emergence remained synchronized for both sexes throughout the emergence period. During the three years sampling period in the indigenous forest, the sex ratio did not deviate significantly from the unit (one), whereas in the mixed indigenous forest, sex ratio deviated significantly from the unit (one) in 2005 and 2007.

The moth adult sex ratio changed throughout the season. The sex ratio of moths changed daily, but the pattern of emergence at the beginning of the season was very similar for both sexes. However, when male moths were the majority at the end of moth emergence, the sex ratio changed from male dominance to female dominance; but the general trend of emergence remained synchronized for both sexes throughout the emergence period. The sex ratio for a random single cocoon nest did not deviate significantly from the unit if it came from the indigenous forest.

Table 4.3: Mean (±SE) of fore and hind wings expanse (in cm), length of fore and hind wings (in cm) of male and female *A. panda* from different types of forests (indigenous and mixed indigenous) and years in Kakamega

Years	Isecheno block (mixed indigenous forest)						Ikuywa block (indigenous forest)					
	Sex	n	Fw span	Fw width	Hw span	Hw width	Sex	n	FW span	FW width	Hw span	Hw width
2005	m	238	2.307±0.144	1.107±0.114	1.671±0.347	1.364±0.165	m	391	2.448±0.248	1.170±0.111	1.774±0.331	1.387±0.149
	f	526	2.968±0.260	1.429±0.104	2.2±0.213	1.613±0.201	f	407	2.971±0.346	1.441±0.121	2.204±0.171	1.617±0.169
2006	m	442	2.319±0.233	1.131±0.116	1.804±0.278	1.45±0.186	m	611	2.331±0.219	1.133±0.112	1.841±0.217	1.428±0.152
	f	424	2.956±0.284	1.424±0.186	2.176±0.230	1.624±0.179	f	645	3.090±0.174	1.146±0.115	2.155±0.181	1.637±0.160
2007	m	391	2.326±0.184	1.157±0.095	1.765±0.210	1.457±0.202	m	628	2.349±0.214	1.124±0.093	1.797±0.234	1.408±0.157
	f	492	3.052±0.199	1.462±0.112	2.183±0.211	1.628±0.136	f	594	3.04±244	1.457±0.179	2.154±0.243	1.614±0.144

Fw: Fore wing, Hw: Hind wing, m: Male, f: Female; n: Number of males or females moths

Table 4.4: Mean (±SE) length of antennae (in cm) and diameter of head (in mm) of male and female *A. panda* from different types of forests (indigenous and mixed indigenous) and years in Kakamega

Years	Isecheno block (mixed indigenous forest)				Ikuywa block (indigenous forest)			
	Sex	N	Antenna	Head capsule	Sex	n	Antenna	Head capsule
2005	m	238	0.857+0.065	0.436+0.063	m	391	0.865+0.049	0.461+0.078
	f	526	0.823±0.067	0.555±0.072	f	407	0.783±0.076	0.558±0.072
2006	m	442	0.877±0.043	0.442±0.070	m	611	0.847±0.077	0.453±0.070
	f	424	0.792±0.064	0.58±0.05	f	645	0.813±0.062	0.563±0.059
2007	m	391	0.878±0.052	0.452±0.059	m	628	0.868±0.058	0.454±0.080
	f	492	0.8±0.080	0.555±0.069	f	594	0.786±0.069	0.566±0.064

m: Male, f: Female; n: Number of males or females moth

4.3.1.3 Oviposition

In the Kakamega Forest, the egg-laying period of *A. panda* lasted approximately seven months (from mid-October to mid-May). Coupling started soon after emergence and lasted 12 to 24 hours in the net sleeves (Plate 4.6). The male moth fluttered its wings and approached the female, which raised its abdominal extremity. As soon as they decoupled, the female started laying eggs. The female clung tightly to the underside of the leaf of *B. micrantha* and started rubbing the tip of the abdomen (Plate 4.8) and constructed a covering layer by means of the golden brown hairs on its abdominal extremity. The eggs were then laid in clusters. The number of eggs in an egg cluster ranged from 318 to 461 in the mixed indigenous forest and from 347 to 604 in the indigenous forest. The mean number of eggs in the egg cluster by year is summarized in table 4.5.

Table 4.5: Mean number (± SE) of *A. panda* eggs by egg cluster from indigenous and mixed indigenous forests in Kakamega (years 2005 - 2007).

Years	Isecheno block (mixed indigenous forest)		Ikuywa block (indigenous forest)		t-test
	n	Means number of egg	n	Means number of egg	
2005	11	392.909±45.150	11	485±75.750	**
2006	18	395.167±40.736	16	485.313±89.901	**
2007	25	399.88±33.169	22	486.182±80.885	**

**: Highly significant difference between the two forests; n: Number of egg clusters

Highly significant difference ($P < 0.001$) was found between the indigenous and the mixed indigenous forest, whereas by years in the same habitat no significant difference ($P > 0.05$) was found.

4.3.1.4 Fecundity

The unlaid eggs ranged from 135 to 236 (mean 200.83±25.34) in the mixed indigenous forest, whereas in the indigenous forest it ranged from 5 to 106 (mean 64.12±36.54). The mean number of unlaid eggs (after dissection) was significantly higher in the mixed indigenous forest. In the indigenous forest however, the mean number of laid eggs was significantly higher (Table 4.6). The same trend was observed during the three years of the study period and no significant difference found between years. These also was, highly significant difference was found between the mean average of laying eggs and those obtained by dissection in the indigenous and the mixed indigenous forests (Table 4.6).

Table 4.6: Total mean number (±SE) of eggs by egg clusters; mean mature eggs in ovary for both the indigenous and mixed indigenous forests in Kakamega

Years	Isecheno block (mixed indigenous forest)			Ikuywa block (indigenous forest)			t-test
	n	Egg cluster	Eggs in ovary	n	Egg cluster	Eggs in ovary	
2005	7	370.43±38.89	208.43±16.81	7	528.57±36.45	78.29±30	**
2006	11	380.73±37.24	200±29.27	11	532.27±64.04	56.82±44.20	**
2007	15	396±30.96	197.93±26.39	15	500.87±83.25	62.87±33.57	**

ns: No significant difference between the average in the two habitats; n: Number of egg clusters

4.3.1.5 Adult longevity

Moths undertook short flights during the daytime as a result of disturbance, mostly due to heat of the sun. Highly significant difference was found between male and female moths life span in both habitats (Table 4.7). Nevertheless, moth's life span seems to be longer in the indigenous forest. By year, significant difference was found to exist between males in the mixed indigenous forest, whereas no significant difference was found to exist between females.

Table 4.7: Adult life span (in days) of *A. panda* from indigenous and mixed indigenous forests in Kakamega (years 2005 - 2007)

Years	Isecheno block (mixed indigenous forest)					Ikuywa block (indigenous forest)				
	n	Male	n	Female	t-test	n	Male	n	Female	t-test
2005	51	4.545±1.036	51	5.182±0.982	**	50	5±0.943	60	6.25±0.639	**
2006	65	3.88±1.013	51	5.091±0.701	**	60	5.25±1.209	64	6.25±1.032	**
2007	71	4.323±1.043	56	5±0.632	**	57	5.235±1.251	79	6.333±0.701	**

**: Highly significant difference between males and females; n: Number of males or females moths

4.3.2 Egg

4.3.2.1 *A. panda* egg size

The eggs are small (Plate 4.16a) and are usually discoidal and isodiametric (Table 4.8). No significant differences in egg size were found to exist between the two forests and also between the three years. However, a highly significant difference was found between the major and the minor axis in the two habitats and no significant difference was observed between years (Table 4.8). The surface of the egg is not smooth (Plate 4.16b) and the micropyle (Plates: 4.17a, b) was surrounded by petal-shaped cell prints (Plates 4.17a, b). The surface of the egg revealed an assembly of numerous knobs between which small aeropyles (Plates 4.17a, b and 4.18) were present. The knobs on the egg surface were large in size and approximately round in shape.

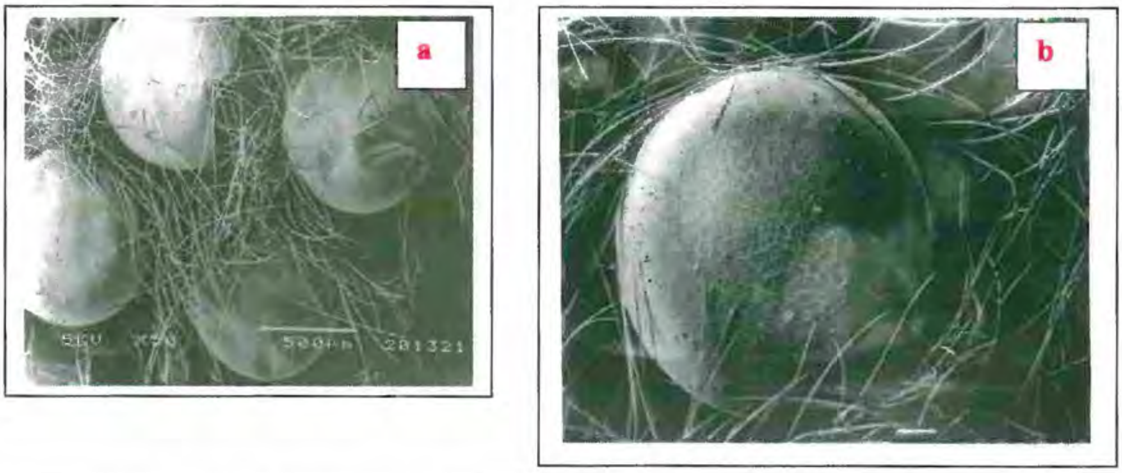


Plate 4.16a, b: Eggs of wild silkmoth *A. panda* in the Kakamega Forest

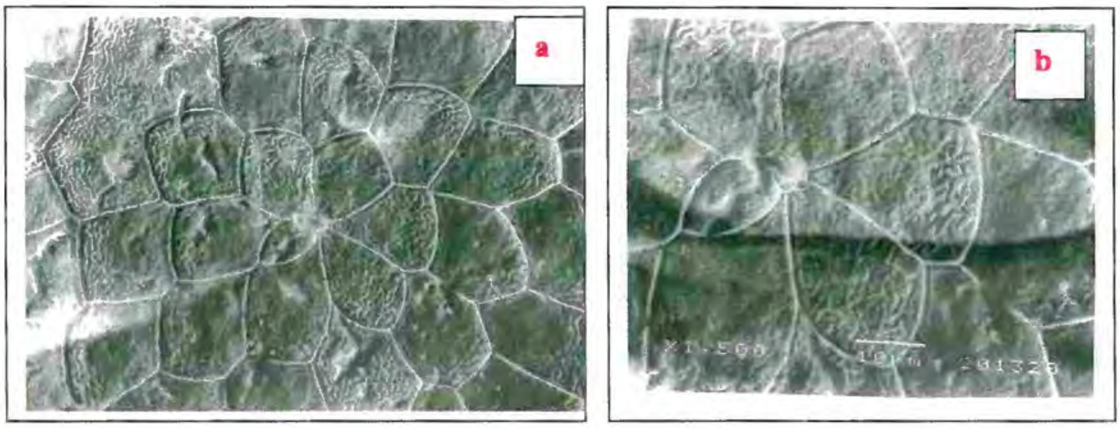


Plate 4.17a, b: Surface structure of micropyle regions of egg shells under (SEM)

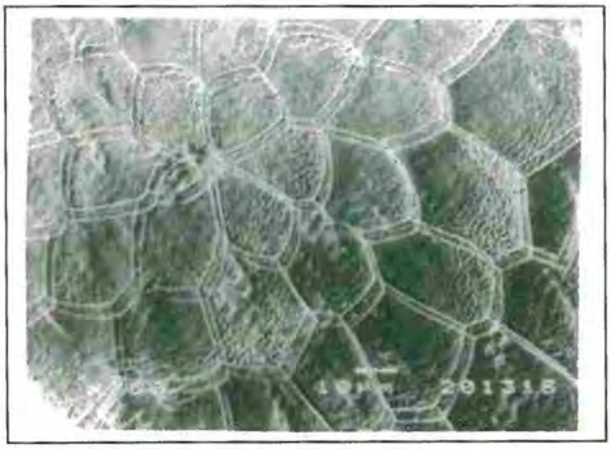


Plate 4.18: Cell prints surrounding micropyle opening of *A. panda* egg

Table 4.8: *A. panda* egg size mean length (mm) in the indigenous and mixed indigenous forests in Kakamega

Years	Isecheno block (mixed indigenous forest)			Ikuywa block (indigenous forest)			t-test
	n	Major axis (mm)	Minor axis (mm)	n	Major axis (mm)	Minor axis (mm)	
2005	15	0.957±0.014	0.61±0.008	15	0.963±0.007	0.6±0.009	ns
2006	15	0.961±0.003	0.6±0.009	15	0.963±0.008	0.61±0.004	ns
2007	20	0.959±0.005	0.62±0.006	20	0.961±0.003	0.6±0.005	ns

ns: Non significant difference between the two forests; n: Number of egg

4.3.2.2 *A. panda* egg cluster size and weight

The eggs of *A. panda* were laid in clusters on the back foliage of the host plant *B. micrantha*. Females started laying eggs from mid-October to mid-May. Eggs found on *B. micrantha* after June were parasitized. The length, width and weight of the egg cluster are summarized in table 4.9.

Table 4.9: *A. panda* egg cluster length (cm), width (cm) and weight (g) in the indigenous and mixed indigenous forests in Kakamega

Years	Isecheno block (mixed indigenous forest)				Ikuywa block (indigenous forest)			
	n	Length	Width	Weight	n	Length	Width	Weight
2005	9	1.71±0.21	0.58±0.08	0.21±0.06	11	1.79 ± 0.22^{ns}	0.63 ± 0.11^{ns}	0.25 ± 0.06^{ns}
2006	15	1.64±0.16	0.58±0.09	0.22±0.04	15	1.66 ± 0.2^{ns}	0.61 ± 0.14^{ns}	0.25 ± 0.07^{ns}
2007	16	1.66±0.34	0.64±0.15	0.26±0.08	17	1.81 ± 0.15^{ns}	0.71 ± 0.09^{ns}	0.29 ± 0.04^{ns}

ns: Non significant difference between egg cluster variables of the two habitats; n: Number of egg clusters

No significant difference was observed between the length, width and the weight of the egg clusters within the two habitats. Length, width and weight did not vary significantly between the years.

4.3.2.3 Incubation period of *A. panda* egg

As hatching progressed, it became obvious that eggs of the same egg cluster do not all hatch simultaneously and that hatching can extend over two days (Table 4.10). The incubation period ranged from 40 to 45 days during the 1st brood (dry season), and from 45 to 55 days during the 2nd brood (rainy season).

Table 4.10: Mean (±SE) incubation period in days for *A. panda* in the indigenous and mixed indigenous forests in Kakamega

Years	Isecheno block (mixed indigenous forest)				Ikuywa block (indigenous forest)			
	1st Brood		2nd Brood		1st Brood		2nd Brood	
	n	Incubation	N	Incubation	n	Incubation	n	Incubation
2005	12	41.75±1.82	8	49.88±3.91**	13	41.69±1.65	10	49.9±3.57**
2006	17	42.82±1.91	18	50.78±3.69**	16	42.19±1.97	15	49.87±3.58**
2007	16	42.13±1.86	18	50.39±3.15**	18	41.83±1.79	17	50.77±3.54**

n: Number of egg clusters, **: Highly significant difference between the two broods

In the same habitat, highly significant difference was found between the 1st brood (dry season) and the 2nd brood (rainy season), whereas no significant difference was found between the two habitats (indigenous and mixed indigenous forests).

Furthermore no significant difference was found between the incubation periods in the same habitat within years. A highly and negative correlation coefficient (r = - 0.951) was found between the mean daily temperature and the incubation period (Fig. 4.1).

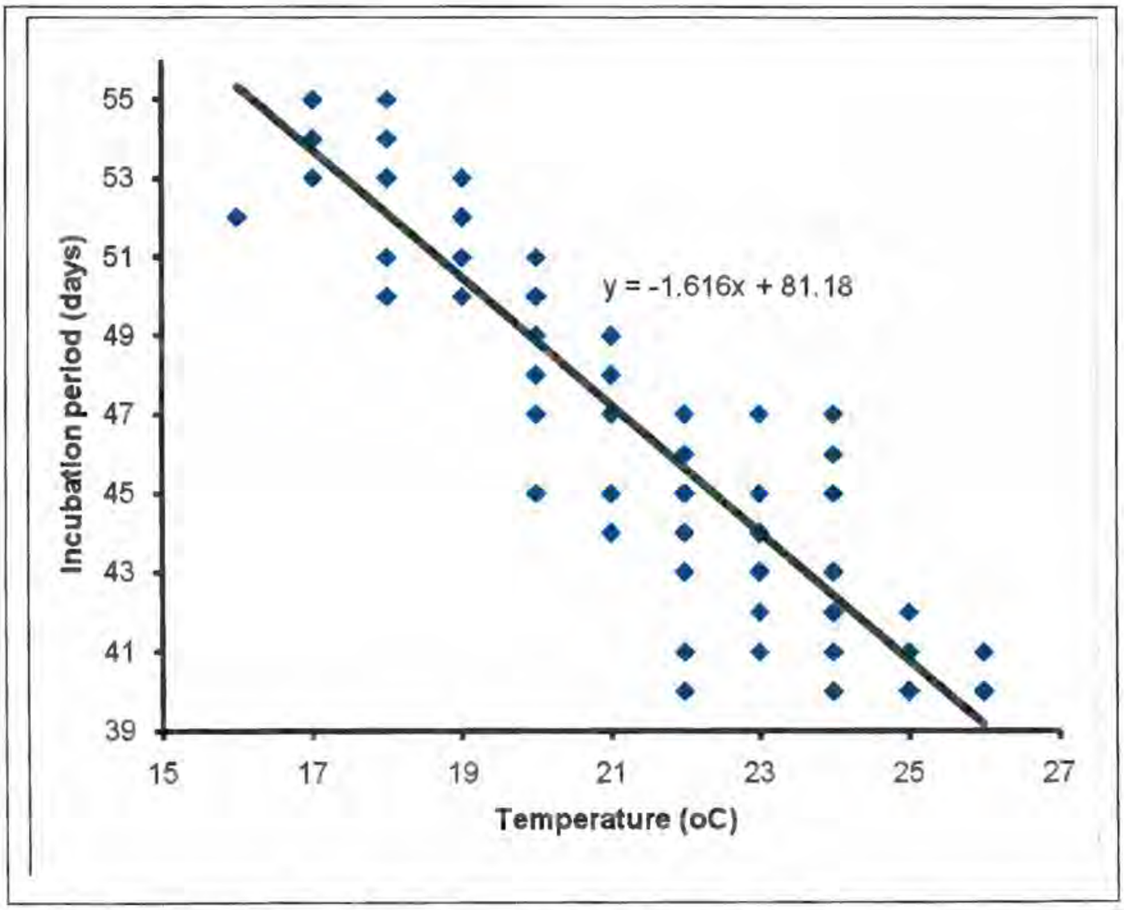


Fig. 4.1: Duration of egg stage of *A. panda* in the Kakamega Forest

4.3.3 Silkworm development

The direct observation of cast exuviae by larvae reared through the entire larval stage showed that six moults occurred from egg hatch to the time the silkworms pupated, hence seven instars. The seventh instar took between 83-86 days for the 1st brood (eggs hatched in the dry season) and 112-118 days for the 2nd brood (those hatched in the rainy season). From the egg hatches, young silkworms followed prodigious growth from 3 mg to more than 3,000 mg (Plate 4.20: E and F). The developmental velocity of the sampled silkworms is shown on fig. 4.2.

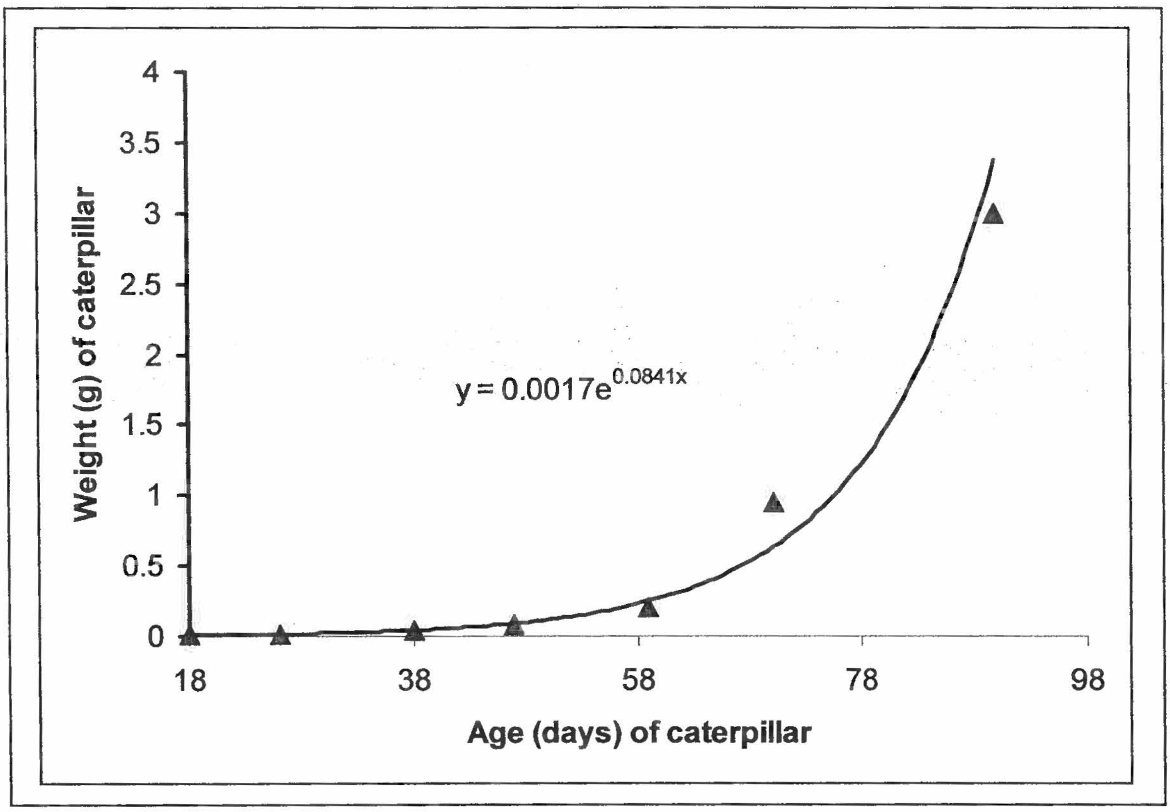


Fig. 4.2: Developmental velocity of *A. panda* silkworms in the Kakamega Forest

In the field, under fluctuating conditions, the first and seventh instars took the longest while the second and third instars took the shortest time to complete the development (Appendix: 2). The duration of the various feeding larval instars seems to be lengthened (delayed) by low temperatures (rainy season), while high temperatures (dry season) shortenened (accelerate) the duration of larval development (Table 4.12). The total developmental larval period by brood before moulting is shown in table 4. 11.

Table 4. 11: Mean (± SE) total developmental larval period (days); mean weight (g) and length (mm) for 7th instar *A. panda* silkworms in the indigenous and mixed indigenous forests in Kakamega

Years	Brood	Isecheno (mixed indigenous forest)				Ikywa (indigenous forest)			
		n	Period (days)	Weight (g)	Length (mm)	n	Period (days)	Weight (g)	Length (mm)
2005	1st	17	85.53±3.39	3.05±0.13	51.16±2.6	19	85.16±3.35^{ns}	3.09±0.1^{ns}	51.53±1.82^{ns}
	2nd	11	115±3.32	2.99±0.17	49.95±2.94	13	115.46±3.23^{ns}	3.08±0.11^{ns}	50±1.86^{ns}
2006	1st	18	85.33±3.05	3.02±0.14	49.53±4.98	15	85.27±3.01^{ns}	2.98±0.15^{ns}	49.69±3^{ns}
	2nd	15	115.07±3.20	2.96±0.16	49.65±2.8	12	115.33±3.52^{ns}	3.01±1.46^{ns}	50.22±2.44^{ns}
2007	1st	16	86±3.8	2.9±0.14	48.85±2.17	17	85.41±3.62^{ns}	2.97±0.15^{ns}	49.75±2.62^{ns}
	2nd	13	115±2.97	2.9±0.9	48.09±1.52	14	116.07±3.85^{ns}	2.97±0.18^{ns}	50.24±3.16*

Brood: 1st: Dry season, 2nd: Rainy season, n: Number of egg clusters, ns: Difference not significant between variables of the two habitats, *: Difference significant between variables of the two habitats.

In the same habitat, a highly significant difference was found between the 1st brood (dry season) and 2nd brood (rainy season) of the larval period, whereas no significant difference was found between the two habitats (indigenous and mixed indigenous forests). Furthermore, no significant difference was found between the weight and the length of the 1st brood and 2nd brood in the same habitat. By year, there was no significant difference between the period, weight and length respectively.

A simple positive linear correlation coefficient ($r = 0.9657$) was found between the mean length and the mean weight. This positive correlation implies that for an increase of the length, the weight also increases in the same way. There was a linear association between the magnitudes of the two variables. The relationship between the age and the length of the sampled silkworms is also shown in fig. 4.3 by a simple linear regression.

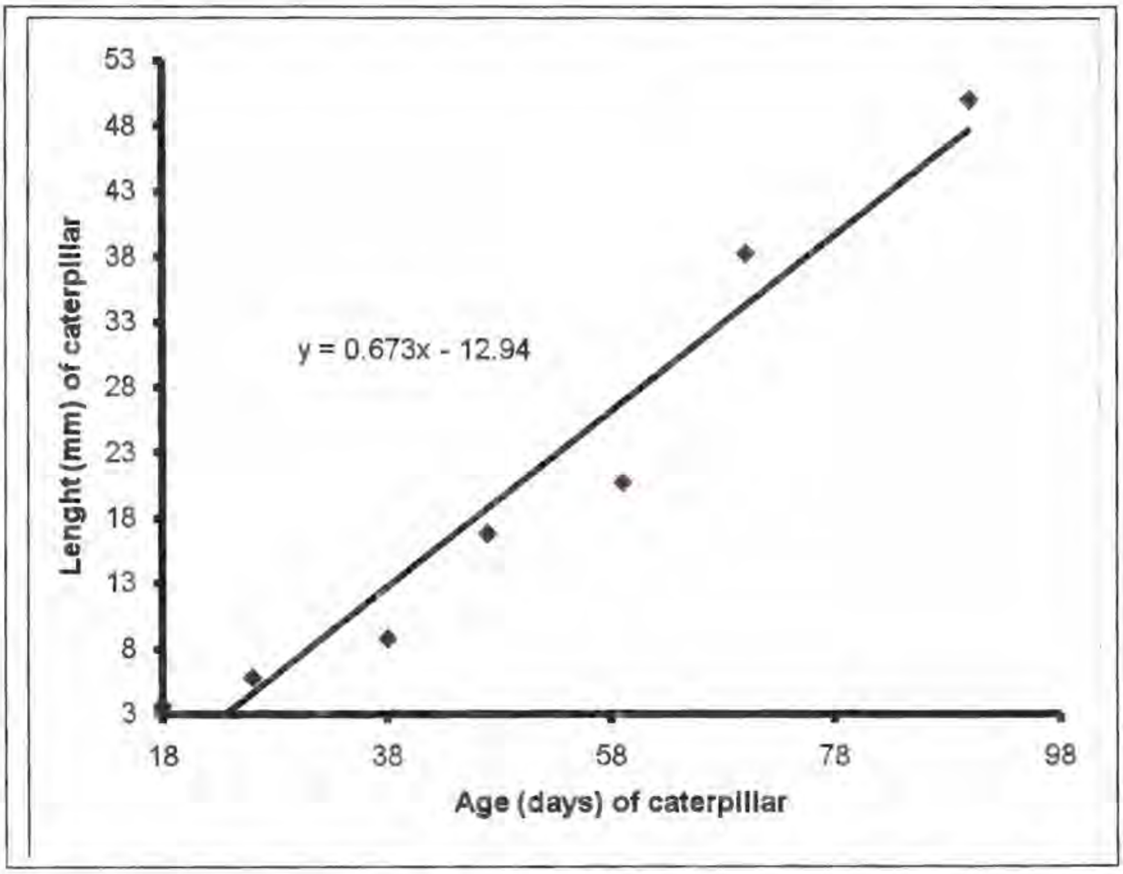


Fig. 4.3: *A. panda* silkworm's length as a function of age in the Kakamega Forest

From hatching to maturity, a single silkworm consumed nearly 45 g to 51g and 40 g to 45 g of foliage for the 1st and 2nd brood respectively. The average leaf consumption by instar is summarized in a Appendix: 3 and the total consumption by brood by year is shown in table 4.12.

Table 4.12: Average (± SE) leaf consumption (g) and total feeding time (days) of *A. panda* silkworms in the Kakamega Forest

		Isecheno (mixed indigenous forest)				Ikuywa (indigenous forest)			
Years	Broods	n	Feeding period (days)	Leaf consumption (g)	Resting period (days)	n	Feeding period (days)	Leaf consumption (g)	Resting period (days)
2006	1st	3	71.67±0.58**	48.67±2.52*	19.3±0.6	4	70.5±1.29**	48.75±1.71*	21±1.6^{ns}
	2nd	4	93.75±0.96	43.5±1.29	20±0.8^{ns}	3	93.33±1.53	44±1	22±1
2007	1st	2	71±1.41**	44.5±0.51^{ns}	19.5±0.7*	3	70.67±1.53**	44±1^{ns}	19.7±0.6*
	2nd	3	94±1	43±2.65	21.3±0.6	3	94.33±1.15	43.33±1.53	22±1

n: Number of cohorts, ns: Difference not significant between broods, *: Difference significant between broods, **: Difference highly significant between broods.

In the same habitat, highly significant difference was found between the 1st brood (dry season) and the 2nd brood (rainy season) of the total feeding period, whereas no significant difference was found between the two habitats (indigenous and mixed indigenous forests). Furthermore, no significant difference in leaf consumption was found in 2007 between the 1st brood and 2nd brood in the same habitat concerning leaf consumption. Furthermore, no significant difference in resting time was found between the 1st brood and 2nd brood in the same and between the habitats in 2006, whereas significant difference was observed in 2007. At the end of the seven instars, larvae wandered away from where they had been feeding and constructed their cocoons. Unprotected silkworms travelled between 5 to 25 metres to select suitable sites for their nest cocoon construction.

The total feeding and resting periods in days by instar did not vary significantly by year as show in table 4.13. However, significant difference was observed between the instars.

Table 4.13: Average (± SE) total feeding and resting time (days) of *A. panda* silkworms instars in the Kakamega Forest

Instars	2006 (n = 14)		2007 (n = 11)	
	Feeding periods	Resting periods	Feeding periods	Resting periods
1st	16.4±1.6	2.8±0.4	16.8±1.8	2.9±0.5
2nd	9.1±1.5	2.4±0.5	8.7±1.6	2.2±0.4
3rd	8.4±1.3	2.6±0.4	8.5±1	2.6±0.5
4th	8.9±0.5	2.4±0.5	9.1±0.5	2.4±0.4
5th	9.5±1.3	3.5±0.8	9.5±1.3	3.5±0.5
6th	11.9±2.9	4.9±0.9	12.2±3.5	5.1±0.8
7th	18.1±3.8	2.1±0.3	18.7±4	2±0.3

n: Number of cohorts

By year, no significant difference was observed between the feeding periods ($P = 0.398 > 0.05$), and also between the two resting time ($P = 0.388 > 0.05$). The data showed that silkworms spent approximately 80% of their life feeding and 20% resting and moulting.

4.3.4 Pupa

4.3.4.1 Pupal period

The pupal period for the larvae which spun between April and July (1st brood) ranged between 158 - 178 days, and 107 - 138 days for those which spun between August and October (2nd brood). Three months old 1st brood silkworm's larvae were found alive in the open cocoon nest (Plate 4.19).

Table 4.14: Mean (± SE) pupal period (days) of *A. panda* in the Kakamega Forest

Years	Brood	Isecheno (mixed indigenous forest)		Ikuywa (indigenous forest)	
		N	Pupal period (days)	n	Pupal period (days)
2005	1st	6	167.5±7.52**	9	167±7.7**
	2nd	7	122.14±10.49	8	126.25±11.88
2006	1st	10	168±6.06**	8	168±7.86**
	2nd	9	123.33±12.94	6	121.5±5.24
2007	1st	9	166.67±5.43**	8	169±8.33**
	2nd	9	122.33±10.69	11	124.64±11.94

n: Number of cocoon nests, **: Difference highly significant between the two broods

In the same habitat, highly significant difference in pupal period was found between the 1st brood (dry season) and 2nd brood pupae (rainy season), whereas there was no significant difference between the two habitats (indigenous and mixed indigenous forests) (Table 4.14). Furthermore, there was no significant difference between the three years of observation.

4.3.4.2 Pupal length and width

The mean length and width of male and female pupae from the two locations ranged from 1.8 to 2.7 (mean 2.11±0.17) cm and 0.6 to 1 (mean 0.73±0.07) cm for males' length and width respectively; whereas, that of the female pupae ranged from 2.1 to 3.1 (mean 2.4±0.15) and 0.6 to 1.3 (mean 0.82±0.08) cm for the females' length and width respectively. Highly significant differences ($P < 0.001$) where found between the lengths and also the widths of the two sexes. The weight of the male ranged from 0.23 to 0.9 g (mean 0.5±0.14) whereas that of the female pupae ranged from 0.27 to 1.16 g (mean

0.73±0.19) g. There were also significant differences between the weights of the two sexes. Fluctuation by year and by forest habitat which is summarized in table 4.15 shows that there was no significant difference between the three years of observation and also between the two broods.

Table 4.15: Mean (±SE) weight (g), length (cm) and width (cm) of *A. panda* pupae in the Kakamega Forest

Years	Sex	Isecheno (mixed indigenous forest)				Ikuywa (indigenous forest)			
		n	Weigh (g)	Length (cm)	Width (cm)	n	Weigh (g)	Length (cm)	Width (cm)
2005	m	42	0.52±0.15**	2.11±0.15**	0.73±0.06**	36	0.52±0.12**	2.12±0.2**	0.74±0.07**
	f	38	0.76±0.21	2.37±0.12	0.82±0.06	37	0.7±0.17	2.42±0.14	0.84±0.09
2006	m	49	0.51±0.15**	2.16±0.15**	0.75±0.08**	61	0.52±0.11**	2.08±0.16**	0.74±0.08**
	f	40	0.71±0.19	2.41±0.17	0.81±0.09	54	0.71±0.19	2.4±0.17	0.82±0.09
2007	m	62	0.47±0.14**	2.1±0.16**	0.72±0.06**	78	0.5±0.15**	2.09±0.18**	0.73±0.07**
	f	47	0.74±0.19	2.42±0.16	0.82±0.08	68	0.73±0.19	2.4±0.15	0.83±0.07

n: Number of *A. panda* pupae; **: Difference highly significant between the two pupae sex

4.3.4.3 Pupal sex ratio

Pupae which were sampled from the two forest habitats were sexed and the results are summarized in table 4.16. All the values of χ^2 –test (with Yate's continuity correction) obtained on the sex ratio of the pupa were less (Table: 4.16) compared to the critical value ($\chi^2_{0.05,\ 1}$ = 3.841). These results indicate that the sex ratios did not deviate significantly from the unit (that means: the sample pupae data come from a population having 1:1 ratio of male to female).

Table 4.16: Pupae sex ratios of *A. panda* from indigenous and mixed indigenous forests in Kakamega (years 2005 - 2007)

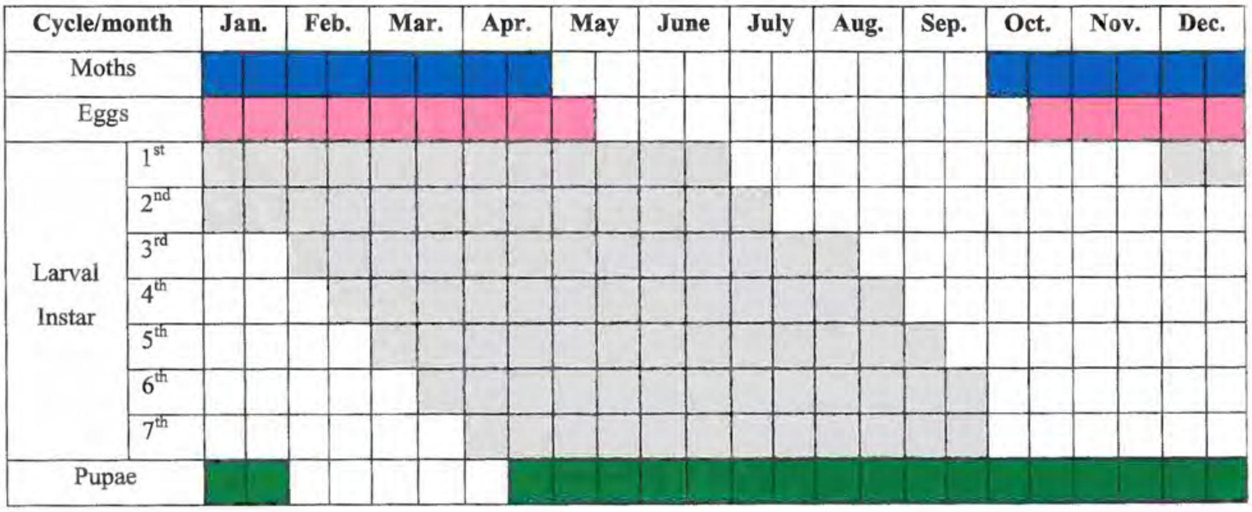
Years	Isecheno block (mixed indigenous forest)						Ikuywa block (indigenous forest)					
	Observed Pupae		Sex ratio		Expected Sex ratio	χ^2	Observed Pupae		Sex ratio		Expected Sex ratio	χ^2
	m	f	m	f	m:f		m	f	m	f	m:f	
2005	42	38	1:	0.905	1:1	0.213*	36	37	1:	1.03	1:1	0.027*
2006	49	40	1:	0.816	1:1	0.921*	61	54	1:	0.885	1:1	0.437*
2007	62	47	1:	0.758	1:1	2.073*	78	68	1:	0.872	1:1	0.692*

*: Pupae sampling coming from a population having 1:1 ratio of male to female, m: male, f: female

4.3.5 Life cycle of *A. panda* in the Kakamega Forest

Life stages of *A. panda* silkmoth are summarized in fig. 4.20. Moths are found in the forest from October to April (Fig. 4.4). They occur in peak numbers in late November to late February. Egg clusters can be seen from mid-October to early mid-May and larvae from early December to late September. Pupae are present in the field from mid-April to January, but new pupae are found in the forest from mid-April. February and March are the best months to harvest dry cocoon nests with minimal damage to fresh cocoons when moths have not emerged.

Plate 4.19: Three months old life silkworms since spinning in the cocoon nest



Cycle/month		Jan.		Feb.		Mar.		Apr.		May		June		July		Aug.		Sep.		Oct.		Nov.		Dec.	
Moths		🟦	🟦	🟦	🟦	🟦	🟦	🟦	🟦											🟦	🟦	🟦	🟦	🟦	🟦
Eggs		🟥	🟥	🟥	🟥	🟥	🟥	🟥	🟥	🟥											🟥	🟥	🟥	🟥	🟥
Larval Instar	1st	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨											▨	▨
	2nd	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨											
	3rd			▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨									
	4th				▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨								
	5th					▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨							
	6th						▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨						
	7th							▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨	▨						
Pupae		🟩	🟩						🟩	🟩	🟩	🟩	🟩	🟩	🟩	🟩	🟩	🟩	🟩	🟩	🟩	🟩	🟩	🟩	🟩

Fig. 4.4: Time of occurrence of the various stages of *A. panda* silkmoth in the Kakamega Forest (2005-2007).

4.3.6 *A. panda* behaviour in the Kakamega Forest

A. panda moths preferred lower and middle crown levels for resting (Table 4.17). Under the hypothesis that moths equally prefer the three tree crown levels (lower, middle, and upper); and the two distributions (observed frequencies and expected frequencies) are the same, a test statistic was calculated. In the Kolmogorov-Smirnov test, the largest difference between the cumulative observed and expected frequencies $|d_{max}|$ is the test statistic. Hence d_{max} = maximum $|d_i|$ = 10 and the critical value is: $(d_{max})_{0.05,3,69} = 9$. The Kolmogorov-Smirnov test rejected the hypothesis at the 5% significance level because d_{max} calculated $= 10 > d_{max}$ table $= (d_{max})_{0.05,3,69} = 9$. It therefore can be concluded that *A. panda* moths did not equally prefer the three tree crown levels (lower, middle, and upper) for resting.

Table 4.17: *A. panda* moth distribution in the three crowns level of host plant *B. micrantha* in the Kakamega Forest

Frequencies	*Bridelia micrantha* tree crown levels			Total
	Lower	Middle	Upper	
Observed frequencies	22	34	13	69
Expected frequencies	23	23	23	69
Cumulative observed frequencies	22	56	69	
Cumulative expected frequencies	23	46	69	
Difference between cumulative observed & expected freq.	1	10	0	

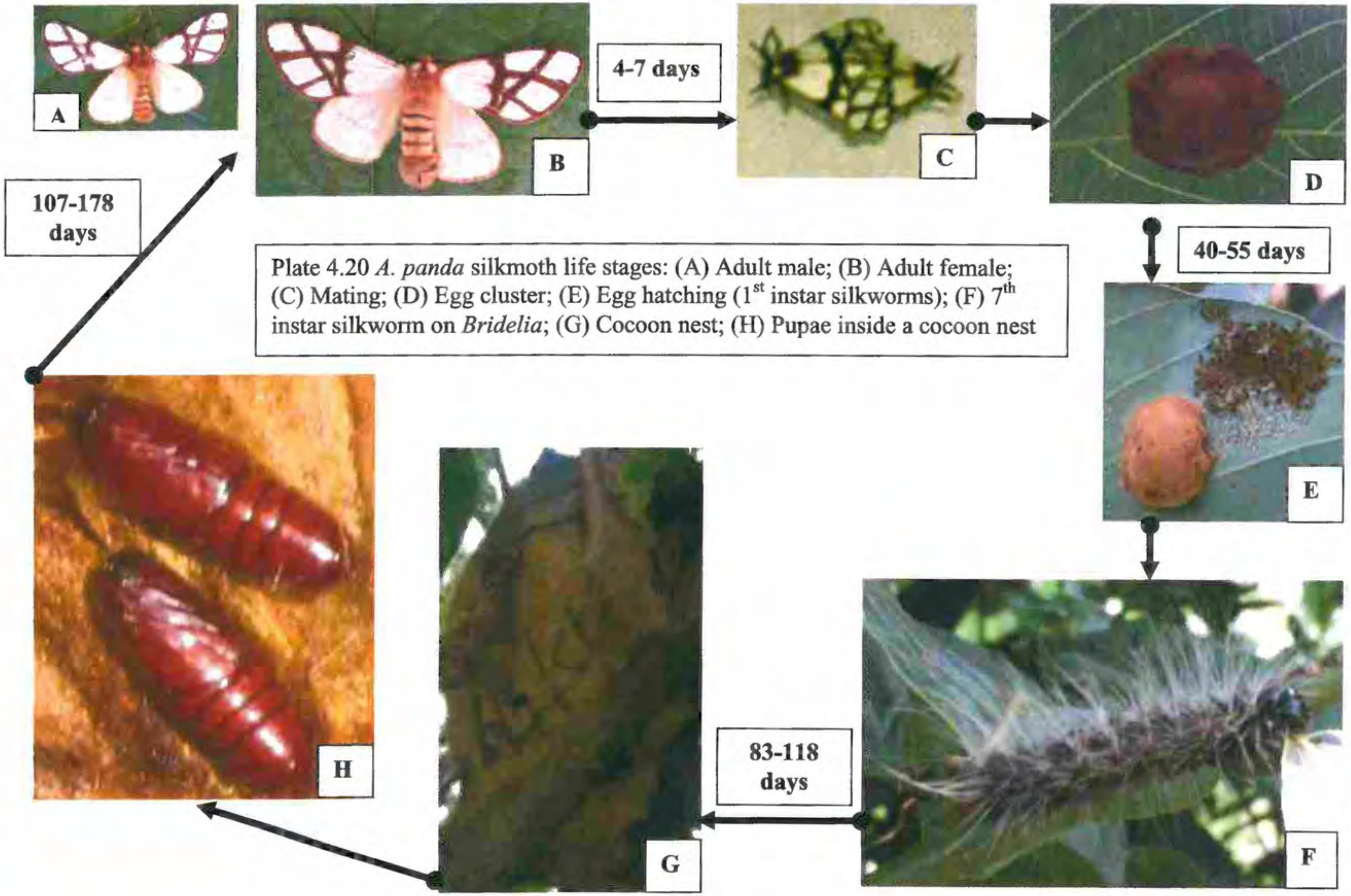


Plate 4.20 *A. panda* silkmoth life stages: (A) Adult male; (B) Adult female; (C) Mating; (D) Egg cluster; (E) Egg hatching (1st instar silkworms); (F) 7th instar silkworm on *Bridelia*; (G) Cocoon nest; (H) Pupae inside a cocoon nest

4.4 Discussion

4.4.1 Adult

The mean values of fore and hind wing expanses, lengths of antennae and head capsules of male and female moths differed for different generations in the same and between blocks of forests (Tables 4.2 and 4.3). Those differences were however not significant in both forests. There were highly significant differences ($P < 0.001$) between the means of the forewing expanse and the length of females and males. Mean forewing expanse and length of females were larger than that of the males. Highly significant differences ($P < 0.001$) were also observed between the means of the hind wing expanse and length; the mean length of antennae and the head diameter of females and males. Mean forewing expanse and length of females were larger than that of the males. The size of male and female seemed to be one of the explanations of the differences.

In the current study, it was found that the adult sex ratio differed significantly in the two different habitats (indigenous forest and mixed indigenous forests). In the indigenous forest during the three years, there was an equal sex ratio between males and females whereas in the mixed indigenous forest, this was not the case. This observation suggests that there were some factors which were responsible for a higher male mortality of *A. panda* pupae in the mixed indigenous forest. Contrary to this study's observation, Jolly et *al*. (1979) observed a ratio of male to female moths of 3:2 for *A. venata*. In the Kakamega Forest, females of *A. panda* moth occurred in predominantly higher numbers. The relative shortage of males at the end of the season can pose a problem of effective mating and fertilization of the eggs.

There was no significant difference in the longevity of *A. panda* males and females ($P = 0.324 > 0.05$) for the different generations. But, the difference between the two forest blocks was highly significant ($P < 0.001$). Generally, a moth seemed to have a higher life span in the indigenous forest compared to that of the mixed indigenous forest. There was also a highly significant difference between the longevity of the two sexes in both forest blocks ($P < 0.001$). Female perhaps lives longer because she must lay the maximum number of eggs before dying. Longevity seems to be related to temperature because moths may live longer during colder spells. Observations made during this study confirmed earlier findings by Jolly et *al.* (1979) who found that an adult *Anaphe* lived for nearly a week.

4.4.2 Egg

The female started laying eggs just after mating and these findings are similar to those reported by Ashiru (1975a). The egg laying continued for half a day to one and a half days. Jolly *et al.* (1979) observed egg laying from 8 to 20 hours for *Anaphe* sp. and Ashiru (1975a) recorded a whole day for *A. venata* egg laying behavior. The mating activity for *A. panda* lasts around twelve hours contrary to Jolly *et al.* (1979) who reported a mating activity of 5 to 24 hours for the same species. Depending on the health of the female, the number of eggs laid may vary from just a few to over hundred (Covell, 1984). In this study, observations showed that egg cluster content was from 350 to 539 eggs, contrary to Jolly et *al*. (1979) and Kioko et *al*. (1999a) who noted a cluster of 250-350 and 250-300 eggs respectively. The number of eggs per cluster did not vary significantly ($p > 0.05$) for different generations in the same habitat (block of forest). Nevertheless, there was a highly significant difference ($p < 0.001$) between the indigenous and the mixed indigenous forests. A positive trend in the mean number of eggs was observed from generation to generation in the mixed indigenous forest whereas in the indigenous forest, the trend was constant. This observation may suggest that due to conservation, there is a certain amelioration and evolution of the macro and microclimate of the mixed indigenous forest towards the indigenous forest equilibrium. During heavy rains, no egg laying took place and a possible explanation is that the cement gluing the eggs onto the substrate does not stick on wet surfaces. This glue furthermore does not dry under wet conditions and the eventual result is the declusterisation of egg clusters during heavy rains. The female seems to know instinctively which host plant to lay her eggs on, probably sensing chemical substances characteristic of that plant.

An average of 64.12±36.54 unlaid eggs were obtained after dissecting females in the indigenous forest, whereas this average was 200.83±25.34 in the mixed indigenous forest. Therefore, a highly significant difference in the unlaid number of eggs was found between the two-forest habitats. However, there was no significant difference between egg averages in the two forests. This finding shows that the number of unlaid eggs in the indigenous forest was much less (by about a third) than that in the mixed indigenous forest. According to Morris and Fulton (1970), differences in food quality can have an effect on fecundity.

The findings obtained from this study confirmed earlier reports by Jolly *et al.* (1979) who observed that *Anaphe* sp. eggs are discoidal and isodiametric, measuring 0.97 x 0.62 mm. Morphological analyses of the *Anaphe* eggshell indicated that the specialised area for sperm entry is distributed at the anterior pole region. Kioko *et al.* (1999b) made similar observations on *Argema mimosae* and *Gonometa* sp. eggshell, and Regier *et al.* (1980) on the eggshell of the domesticated silkmoth *Bombyx mori.*

The duration of egg stage depends on the prevailing climatic conditions. These findings confirm an earlier survey by Jolly *et al.* (1979) who reported that: the embryonic period may last a month or more (45 days) depending upon the climatic conditions. The fact that there was no significant difference between the two habitats means that temperature strongly influences the incubation period and this is confirmed by the high correlation coefficient between the mean temperature and incubation period. Furthermore, the negative sign of correlation coefficient means that an inverse relationship exists between the mean daily temperature and the incubation period. Hence, increase in temperature decreases the incubation period.

4.4.3 Silkworm development

The time between hatching from the egg and completion of larval stages varies. It may depend upon the availability of food, a favourable climate and other factors. Covell, (1984) made similar observation. The total (longest) larval period of *A. panda* is about 120 days (Table 4.11); this finding differs with the 140 days reported by Jolly *et al.*, (1979) for the *A. venata* total larval period. The general relationship between the developmental velocity, expressed in terms of weight and the number of days tends to be non-linear and it appears to be exponential. Similar observation was made by (Wigglesworth, 1965). From these experiments, it appears that the duration of the larval instars is dependent upon temperature; higher temperatures (dry season) causing shortening and lower temperatures (rainy season) prolonging the developmental period. This agrees with observations of a similar nature made by both Geertsema (1975) and Chapman (1969). The high correlation coefficient between weight and length has shown the measure of intensity of association between the two variables. According to Wigglesworth (1965), temperatures and nutrition might also have an effect on the number of larval instars.

The mean foliage consumption by a single *A. panda* silkworm fluctuates between 43 g – 49 g of leaves according to the season. However, this mean is less compared to 300 g of foliage consumed by another non-mulberry silk *A. mylitta*, (Jolly *et al.*, 1979). Notwithstanding this small average of foliage consumption per larva, *A. panda* (because of the high population per cohort) remains a defoliator, and a pest in the case of shortage of *B. micrantha* leaves. A case was reported from Cameroon (Gérin, 1957) where after a shortage of leaves on the host and alternative host plant, *A. venata* silkworms migrated to the cola-nut tree, and finally invaded and devoured crops (corn). It is apparent that the sixth and seventh instars are mainly responsible for the defoliation (Appendix: 3).

4.4.4 Pupa

A delay in emergence of moths was observed for the 1st brood of silkworm. A pupa was recorded extending its diapause over five to six months. This explains the significant difference in records between the two broods. Adult moths emerge from the 1st brood after 168 days and from the 2nd brood after 122 days. This indicates that the rate of development of the pupae differs and it is most probably influenced by intrinsic factors. Van den Berg (1971) investigating on *N. cytherea clarki* made a similar conclusion that pupae formed during the later (warmer) season were able to catch up in development with those formed earlier in the colder season. *Anaphe* silkworm is univoltime (Akerele, 1970; Gowdey, 1953) and undergoes diapause at the pupal stage.

More male pupae occurred in the two habitats from the random samplings. The data shows that the proportion of male to that of the female pupae is not approximately equal and that any fluctuations probably even out over the years. The differences in the sex ratio between the pupa and adult stages have been investigated by a number of authors (Miller, 1963); Stern and Smith, 1960). In the present study, proportion of male pupae was predominant perhaps because the sample population size was not high. Geertsema and Giliomee (1972) investigating on *Nudaurelia cytherea* (Fabr.) found a pupal sex ratio not significantly different from 1:1 ratio; with a sample population size above one thousand. In the light of this observation, it seems advisable to collect annual samples numbering above one thousand, if accurate determinations of the pupal sex ratio are to be determined.

4.4.5 *A. panda* behaviour

A. panda prefer middle and lower tree crown levels. Perhaps because of the larger amount of foliage being available there for oviposition, larvae feeding or for protecting themselves against predators. Most moths have been reported to mate during the night hours (Engelmann, 1970), but in Kakamega Forest, *A. panda* were observed many times in the net sleeve mating in daylight like other diurnal species such as *Oncopeltus fasciatus* (Caldwell and Dingle, 1965) and *Lucilia sericata* (Cousin, 1929) which mate nearly any time during the daylight or the carpenterworm moth *Prionoxystus robiniae* (Solomon and Morris, 1966) which also mate in bright daylight. These observations suggest that, daylight is essential for courtship in these species. This phenomenon is probably more widespread than is presently known and may he occurring in a number of species from different orders. A variety of stimuli ranging from acoustical, olfactory, and visual to mechanoreceptory, are employed by various species, and play a decisive role in the determination of a further courtship behavior of either sex; the sensory input will affect the response of the recipient animal. However, further work on *A. panda* oviposition will be required to understand the whole mechanism. According to Covell (1984), males are attracted to females of their species by both visual and chemical stimuli. Females usually secrete a pheromone into the air from special glands located toward the tip of the abdomen, which can be exposed at will. Males up to several miles away in large species may detect this pheromone with special sense organs on their antennae, and they fly upwind toward its source.

To develop wild silkmoth farming, the biology and life history information is very important. By understanding the life cycle, the species and its' host plants can be conserved and the community can be helped to utilize it for income generation.

CHAPTER FIVE

5 INFLUENCE OF BIOTIC AND ABIOTIC FACTORS ON THE LIFE HISTORY OF SILKMOTH *ANAPHE PANDA* (BOISDUVAL)

5.1 Introduction

An important issue in the studies on population dynamics is how biotic and abiotic factors determine population size. It is important to identify biotic factors such as natural enemies so that their regulatory effects can be deliberately reduced. It is important also to know why a forest insect like *A. panda* periodically develops high populations in certain well-defined stand types of forest and not in others. The factors affecting this species in any one place are of two types: those that cause a relatively constant mortality from year to year and contribute little to population variation, and those that cause a variable, though perhaps much smaller mortality and appear to be largely responsible for the observed changes in population (Morris, 1957). In order to develop a sustainable and ecological mass production system of *A. panda* cocoons, it was necessary to identify, evaluate and compare the impact of natural enemies on the developmental stages *A. panda*. In Asia; parasitoids that have been reported to attack wild sericulture are chalcidoid wasps attacking caterpillars (Peigler, 1989; Thangavelu *et al.*, 1988; Jolly *et al.*, 1979) and in Africa by Veltman *et al.* (2004) and Van den Berg (1990; 1974; 1970). A study carried out in U.S.A by Coffelt and Schultz (1992, 1993) revealed that new parasitoids records of wild silkmoth species still await identification.

In this study, the population dynamics of immature larval stage of *A. panda* populations were investigated with the aim of determining the rate of losses at each instar when larvae were protected and unprotected and also assessing the effect of biotic and abiotic factors that regulate the population. According to Jolly *et al.* (1979), the loss of *A. panda* silkworm due to diseases is negligible. The silkworms have been reported to suffer only from muscardine (Rolet and Par, 1948). However, they are vulnerable to many parasites and predators.

5.2 Materials and methods

In the two sampling sites (mixed indigenous and indigenous forests), a recording digital hygrothermometer (Plate 3.5) was used to record daily temperatures (maximum and minimum) and Relative humidity. A rain gauge (Plate 3.6) was also used for recording the rainfall data.

Eggs of *A. panda* moths collected from the field were kept in tubes, and regular weekly inspections of the collected eggs were undertaken and any egg parasitoids that emerged were collected and identified at the biosystematics unit at ICIPE. All parasites were preserved with 70% alcohol and stored in small glass vials with relevant data for eventual reference. Presence of egg parasitoids in the field was also checked by visual observation in the field. Each parasitoid was mounted on aluminium stubs with silver suspension, sputter coated with gold and examined and photographed using a scanning electron microscope (Joel JSM-T 330 A). The rate of egg parasitism was monitored by following

217 egg clusters in the field during the study period, and t-test was used for comparison between broods, forest habitats and years.

The mortality rate and the survival rate of the larva in the protected net sleeve and those unprotected was analysed by following up for three years, two hundred and twenty-one (221) cohorts of silkworms right from the beginning of the experiment. Only one hundred and eighty (180) cohorts of *A. panda* silkworm larvae on *B. micrantha* spun cocoons. The rearing was done by protecting and changing the tree branches during the entire larval stages of one hundred and five (105) cohorts with net sleeves, and one hundred and sixteen (116) unprotected. Only cohorts which completed all the seven larval instars with some survivors (180) that were considered for analysis. Any cohort without survivors at the end of the experiment was sampled but was not consider for analysis. During sampling, two egg clusters laid by the silkmoth and randomly selected on each one hundred and fifty (150) *B. micrantha* tree randomly selected with a canopy of 10 cubic feet were left to hatch. No more than two egg cluster were left by *B. micrantha* tree and independent to its geographical position. Two hundred and twenty-one egg clusters (221) hatched from the selected three hundred trees. Each *B. micrantha* sampling tree was divided into two experimental areas: one with a net-sleeve measuring 1.5 x 1.5 x 2 m for one cohort of larvae and another unprotected area of *B. micrantha* tree for another cohort of larvae. Area or branches were chosen randomly in function of the availability of food (leaves). The entire larval period of ninety-four (94) larval cohorts raised in the net sleeves (Plate 4.4), and another eighty-six (86) cohorts raised in the unprotected set up (Plate 4.5). Close observation on the possible causes of mortality twice a week and counts of the surviving silkworms in each experimental host plant were made during the time the larvae were just resting before each moult. The percentage of dead larvae (mortality rate) by instars was calculated as follows:

$$Mortality \quad rate = \frac{S_{ini} - S_{fin}}{S_{ini}} \times 100$$

With: S_{ini} = Number of surviving silkworms at the beginning of the instar and S_{fin} = Number of surviving silkworms at the end of the instar.

The incidence rate or instantaneous risk to die is the boundary of the expression below when $\Delta t \rightarrow 0$.

$$\frac{\Pr obability \ for \ a \ particular \ silkworm \ to \ die \ between \ \mathrm{t} \ and \ t + \Delta t \ (instar)}{\Delta t}$$

The "force of mortality" (incidence rate) at each instar was also be calculated as follows:

$$Incidence \quad rate = \frac{D_{per}}{S_{\exp}} \times 100$$

With: D_{per} = Number of dead silkworms in the specific instar and S_{exp} = Number of surviving silkworms exposed to risk in this specific instar.

A 3-way analysis of variance (ANOVA) (SAS Institude, 2003) was performed to determine whether significant interaction effect existed between the dependant and independent variables (number of silkworms pupating in each cohort): treatment (protected/unprotected); forest (indigenous/mixed); brood ($1^{st}/2^{nd}$) and years (2005/2006/2007). The maximum-likelihood proportional hazards models were estimated and were used to identify which factors (forest habitat, hatching period, protection by net sleeve, brood and years) positively or negatively affected the incidence rate (Cox, 1972; Cox and Oakes, 1984; Statacorp, 2004; Cleves *et al.*, 2004). A hazard ratio equalling one is taken to be the benchmark: if the hazard ratio is higher than one, the factor affects the hazard positively; if the hazard ratio is less than one, the factor contributes negatively to the hazard. Test of equality of survivor functions was done with Log-rank test (Lakatos and Law, 1992; Muller and Wang, 1994; Hsieh, 1999).

Two-sample sizes of twenty (20) fresh cocoon nests for each site were collected randomly from the field each year and kept in the breeding cage under field conditions to determine the emergence of parasites and parasitoids. All insects and other arthropods within the vicinity of the cocoon nests were observed, collected and identified.

Pathological signs observed on sick larvae were compared with those given by Hendry *et al.* (1967) and Tripconey (1970). To check for bacterial and fungal infection, the larvae were observed under a light microscope and histopathological changes in the silk gland tissues noted and recorded. Infected larvae were sampled and preserved in glass vials (7.5 x 2.5 cm) with Bouin's fixation (Gretchen, 1962). The freezing method (Gretchen, 1962) was used to make blocks for dissecting silk glands of parasitized larvae. Microtome was used for block cutting and stained with various histological stains (Giemsa stain or Fuchsin basic).

5.3 Results

5.3.1 Climatic change factors

The present work recorded the monthly rainfall and the number of rainy days in the experimental site at Isecheno (mixed indigenous forest) and Ikuywa (indigenous forest)

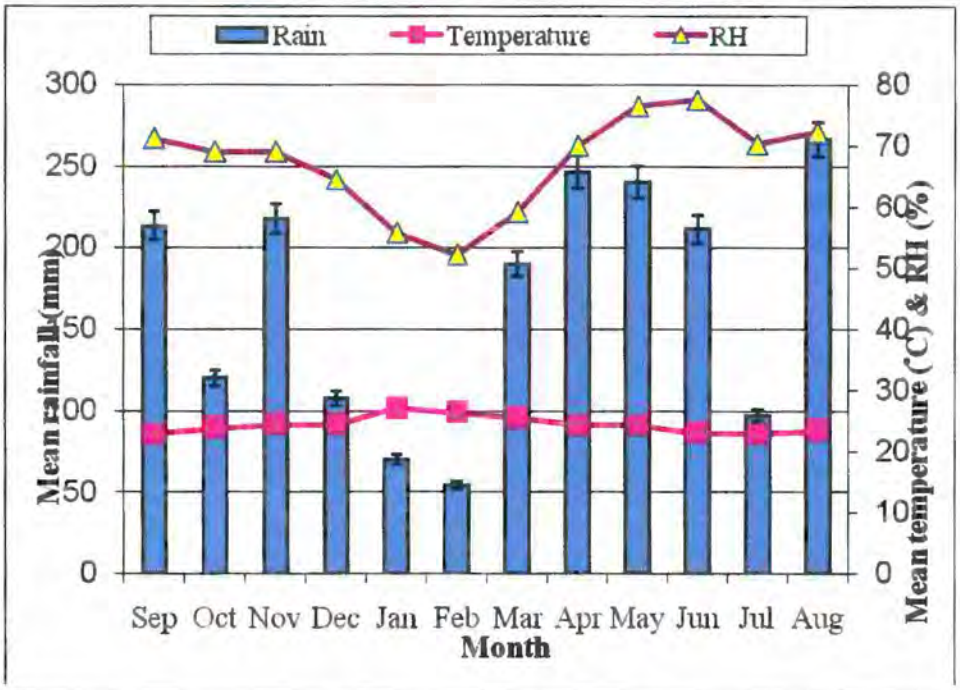


Fig. 5.1: Rainfall, temperature and relative humidity in Isecheno Forest block (2005-2007)

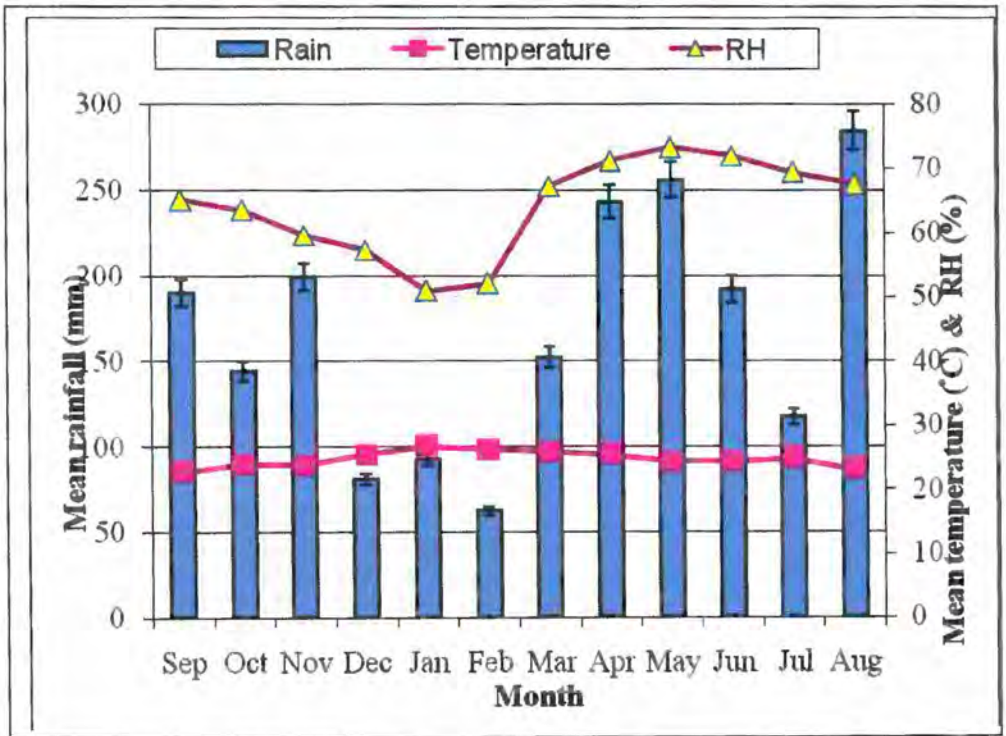


Fig. 5.2: Rainfall, temperature and relative humidity in Ikuywa Forest block (2005-2007)

in the Kakamega Forest. The rainfall was bimodal. The "long rains" were in March through June, with the "short rains" occurring in August and November (Fig. 5.1, and 5.2). From 2004 to 2007, the annual rainfall fluctuated from 1809 to 2658.7 mm in the mixed indigenous forest (Isecheno) (Apendix: 5.1), and from 1886.4 to 2246.6 mm in the indigenous forest (Ikuywa) (Appendix: 6.1). The driest months of the year were December, January and February (Fig. 5.1, and 5.2). The mean rainfall for three years is represented in fig. 5.1 and fig 5.2 for mixed indigenous and indigenous forest respectively.

The number of rainy days by year fluctuated between 196 – 219 days, and between 207 – 209 days in the mixed indigenous (Isecheno) and indigenous (Ikuywa) forests respectively (Appendix: 5.1 and 6.1).

In the mixed indigenous forest (Isecheno), the mean monthly temperature ranged from 15.5 °C to 36.8 °C (Appendix: 5.2), whereas at Ikuywa (indigenous forest), the mean monthly temperature ranged from 16.5 – 35.6 °C (Fig. 5.2). (Appendix: 6.2). The mean temperature for the three years study is represented in fig. 5.1 and fig 5.2 for mixed indigenous and indigenous forest respectively.

The mean monthly humidity ranged from 45.4 – 86.2 % in the mixed indigenous forest (Isecheno) (Appendix: 5.2), whereas in the indigenous forest (Ikuywa) the mean monthly maximum humidity ranged from 35.6 – 80.9 % (Appendix: 6.2). The mean humidity for the three years study is represented in fig. 5.1 and fig 5.2 for mixed indigenous and indigenous forest respectively.

5.3.2 Egg parasitoids

In the Kakamega Forest two chalcids: *Telenomis gowaeyi* (Plate 5.1a, b) and *Pleurotropis telenomis* (Plate 5.2a, b) were recorded from *A. panda* eggs. The photograph using a scanning electron microscope (SEM) also showed un-identified parasitoid (Plate 5.3a, b) that can be considered as a third parasitoid affecting *A. panda* egg in the Kakamega Forest. This new *A. panda* egg parasitoid still awaits identification from icipe Biosystematics' Unit. The main generation of parasitoids started emerging from the first brood of eggs around December.

Table 5.1: Percentage of un-hatched egg clusters observed in the mixed indigenous and indigenous forests of Kakamega.

Egg cluster	Ikuywa (indigenous forest)				Isecheno (mixed indigenous forest)			
	2006		2007		2006		2007	
	1st brood (n=25)	2nd brood (n=20)	1st brood (n=33)	2nd brood (n=35)	1st brood (n=17)	2nd brood (n=21)	1st brood (n=30)	2nd brood (n=36)
Un-hatch	2	5	3	6	3	8	6	14
%	8	25	9.1	17.1	17.7	38.1	20	38.9

n: Number of sampled *A. panda* egg clusters

These parasitoids probably concentrated on the eggs laid during the 2nd brood (rainy season). This could be one explaination of the high percentage of un-hatched egg clusters from the 2nd brood compared to the 1st brood (Table 5.1). The parasitism rate of 2nd brood eggs was significantly higher ($P < 0.001$) than that of the 1st brood. It also was significantly different ($P = 0.003 < 0.05$) between the two forest habitats.

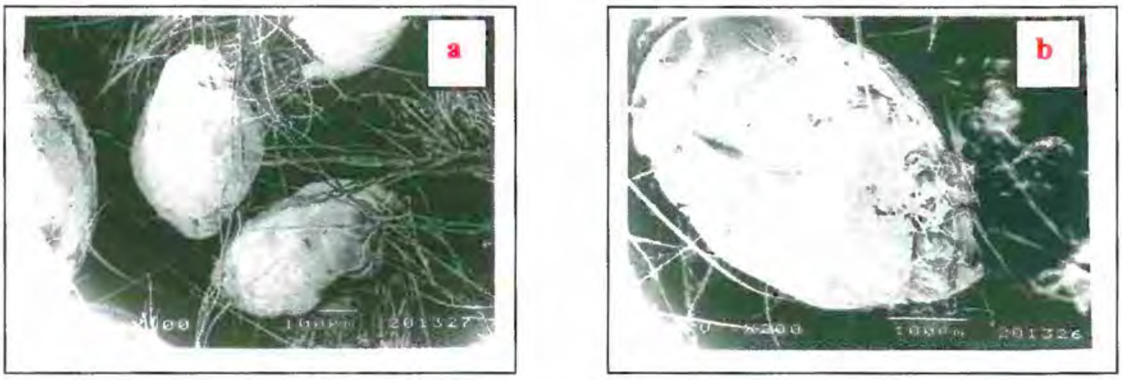


Plate 5.1a, b: SEM of *A. panda* egg parasitoid: *T. gowaeyi* collected from Kakamega Forest

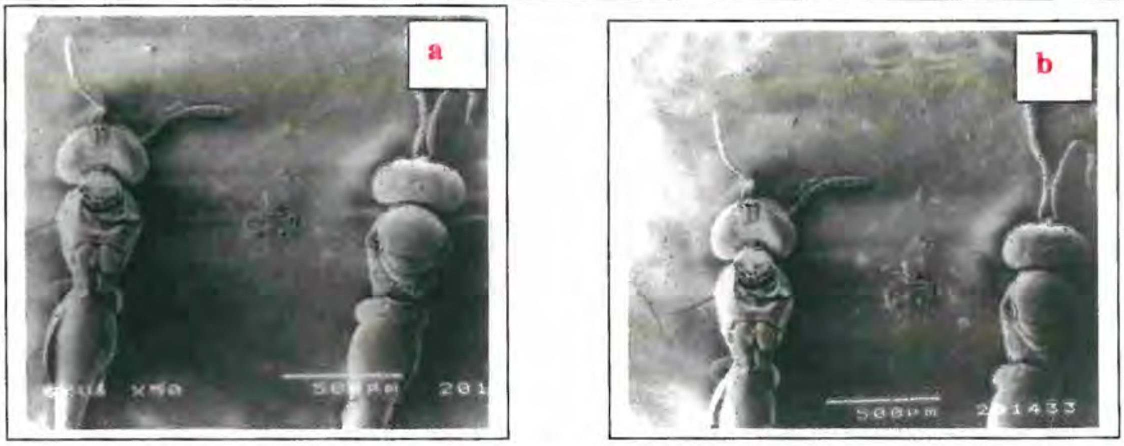


Plate 5.2a, b: SEM of *A. panda* egg parasitoid: *P. telenomis* collected from Kakamega Forest

Plate 5.3a, b: SEM of un-identified *A. panda* egg parasitoid collected from Kakamega Forest

However, between the two years there was no significant difference ($P = 0.503 > 0.05$). Furthermore, the number of perforations on the egg cluster protective shell (Plates 5.6 and 5.7) was easily observed and this also gave an indication on the quality of eggs and the outcome in terms of the numbers of hatching eggs. The head capsule of *P. telenomis* (Plate 5.4a), the thorax (Plate 5.4b) and the sting device (Plate 5.5a, b) were also observed. The latter is probably the one responsible for the damages observed on *A. panda* egg clusters in the field.

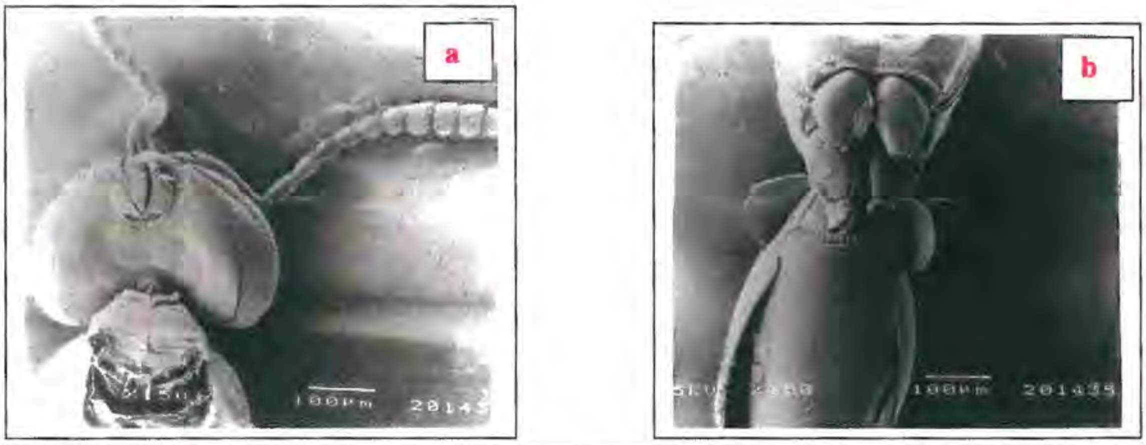


Plate 5.4a, b: SEM of head (a) and thorax (b) of *A. panda* egg parasitoid (*P. telenomis*)

Plate 5.5a, b: SEM of principal sting device of *A. panda* egg parasitoid (*P. telenomis*)

5.3.3 Silkworms

5.3.3.1 Silkworm parasitoids

During the investigations and especially during the rainy season, many silkworms were dying with body hanging down and others fell down after death. The tachinid fly *Exorista cardinalis* and the ichneumon wasp *Cryptus leucopygus* were observed flying around silkworms. Parasitized host larvae that stopped feeding and fell to the ground were frequently attacked by ants, which also attacked the parasitoid pupae. Parasitoid attacks were confined to the sixth and seventh instars of *A. panda* silkworms.

During the survival rate studies, the fifth, six and seventh *A. panda* larval instars were attacked by un-identified parasitoid (Plate 5.8). Its role was not clear, but it appeared that it was consuming of the cast exuviae after moulting.

5.3.3.2 Silkworm predators

In the Kakamega Forest, spiders and formicid ants, *Camponotus sp.* (carpenter ants) were observed in the present study preying on *A. panda* silkworms especially from 1st to 2nd instars. Birds posed a great threat to 3rd to 4th instars of the unprotected *A. panda* silkworms. The birds identified included: galliformes (Quails) from Phasianidae family, and ciconiniformes (Ibis) from Threskiornithidae family. It was observed that, *A. panda* silkworms developed protective white hairs during the 4th instar stage in addition to a coating with silk to all feeding leaf and resting areas and the silkworms camouflage colour that almost resembles the bark of their host plants and this served as a protective mechanism against the natural enemies.

Plate 5.6: *P. telenomis* parasitoid emerge from *A. panda* egg cluster

Plate 5.7: *A. panda* egg cluster protective shell with many perforations

Plate 5.8: Un-identified parasitoid of *A. panda* silkworms

5.3.3.3 Silkworm diseases

Silkworms which succumbed to the disease caused by viruses or bacteria were easily noticed as the disease concerned characteristically produced flaccidity and internal liquefaction. Sick silkworms frequently remained on branches and fell off when dead. Most silkworms that died hanged down from the branches or on the tree stems by their prolegs. Most silkworms which were attacked by this disease exhibited abnormal behaviour (eg. they did not eat with others). Frequently fifth, sixth and seventh instar silkworms which had not completed their feeding stage, were observed moving far down the tree bypassing the available food. Sick silkworms when touched frequently vomited a thick yellow to brown liquid, which was not the case with healthy silkworms. Silkworms succumbed to this disease when in their fifth, sixth, and seventh instars. Silkworms mostly succumbed in the middle phase of their particular instar, or stopped feeding. Most silkworms of the second brood died from the disease of this type during the rainy season.

5.3.3.4 Mortality of silkworms

The mortality rate was investigated for the protected and un-protected silkworms at each instar during this study. In the mixed indigenous forest, the mortality rate ranged from 1.2 to 97.1 per cent with means of 35±16.6 and from 0 to 28.1 per cent with means of 6.6±5 per cent for un-protected and protected silkworms respectively. In the case of the indigenous forest, it was from 1.3 to 72.1 per cent with means of 30±15.4 and from 0 to 26.3 per cent with means of 5.9±4.4 per cent for the un-protected and protected silkworms respectively. However, for both experiments, higher mortality rate was recorded for younger larvae (1st to 4th instars) whereas the lowest rate was recorded for the older silkworm (5th to 7th) instars (Appendix 7). As regards the brood, the mortality rate ranged from 0 to 63.3 per cent with means of 17.1±14.3 and from 0 to 97.1 per cent with means of 23.7±21.2 per cent for the 1st and 2nd silkworms broods respectively (Appendix 7). The mean mortality rate observed by years is summarized in table 5.2. Highly significant difference ($P = < 0.001$) was recorded between the protected and un-protected silkworms. Mortality rate was however significantly higher ($P = 0.023 < 0.05$) in the mixed indigenous forest compared to the indigenous forest. Furthermore, there was a highly significant difference ($P < 0.001$) between the 1st and 2nd brood mortality rate in the mixed indigenous forest.

Table 5.2: The mean (±SE) mortality rate (%) of *A. panda* silkworms in the Kakamega Forest.

Years	Brood	Protection	Isecheno (Mixed indigenous forest)		Ikuywa (indigenous forest)	
			Cohort (n)	Mortality rate (%)	Cohort (n)	Mortality rate (%)
2005	1st	0 (no)	63	31.6±10.7	70	20.2±9.6
		1 (yes)	56	8.7±6.4	63	5.1±4
	2nd	0 (no)	42	38.7±17.2	49	44.2±18.3
		1 (yes)	35	7.4±6.5	42	5.6±5
2006	1st	0 (no)	63	29.9±14.8	56	26.2±11.9
		1 (yes)	63	5.5±4.5	49	5.4±5
	2nd	0 (no)	56	43.8±19.6	42	32.6±19.5
		1 (yes)	49	6.5±5.1	42	6±4.3
2007	1st	0 (no)	56	30.8±13.3	63	27.3±9.1
		1 (yes)	56	5.9±3.1	56	5.3±2.7
	2nd	0 (no)	49	37.5±19.5	49	36±12.2
		1 (yes)	42	6.6±4	49	8.3±4.5

Protection=0: Un-protected silkworms; Protection=1: Protected silkworms

N.B: (n) Each of 180 cohorts is replicate 7 times and each replication represents one instar

Comparing by years show that, the mortality rate was highly significant ($P = 0.0007 < 0.001$) between 2005 and 2006, and also was significant ($P = 0.03 < 0.05$) between years 2005-2007, whereas between years 2006-2007, the difference was not significant ($P = 0.051 > 0.05$).

5.3.4 Pupal parasitoids

During the study period, 25% (81/323) of *A. panda* cocoon nests were moderately infested by various dipteran and hymenopteran parasitoids (Plate 5.10), compared to healthy pupae (Plate 5.9). However, wasps appeared to be a true pupal parasitoids because from July they were easily recorded on the cocoon nests with their ovipositor inside the cocoon (Plate 5.11). September was recorded to be the month when there was a high population of wasps in the field.

It was also observed that moths emerged with wasps from the fresh cocoon nests which were kept in the breeding cage (Plate 3.1). Wasps also emerged from the collected and stored cocoon nests which were waiting to be transformed into raw silk. Wasp larvae were observed in some cocoon nests when carrying out sampling during the pupae sex ratio experiment. Formicidae ants were also observed going in and out the cocoon nest using holes made by parasitoids on the shell of the cocoon nest especially in the mixed indigenous forest.

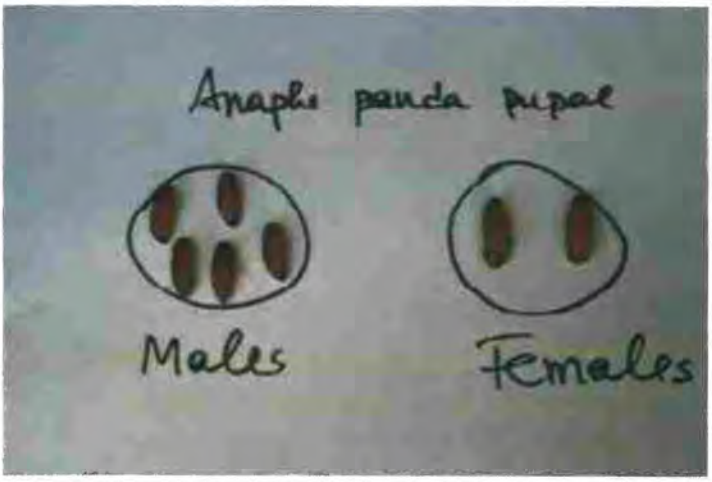


Plate 5.9: Healthy *A. panda* male and female pupae

Plate 5.10: *A. panda* cocoon nest infested by parasitoids

Plate 5.11: Wasp parasitoid on the *A. panda* cocoon nest

5.3.5 Adults predators

The *A. panda* moth life span ranged from 4-7 days. Several species of birds, mammals, spiders and insects were recorded praying *A. panda* adult moth. It was not easy to have reliable estimations of mortalities of adult moths caused by these predators. The birds identified included: galliformes (Quails) from Phasianidae family, and ciconiniformes (Ibis) from Threskiornithidae family. They were observed preying on both adult moths and silkworms. Black and red ants were found to be active predators of young silkworms. Cats were seen preying on *A. panda* adult moths on *B. micrantha* according to Musembe villagers. Pigs and chickens were also seen feeding on dead *A. panda* larvae that fell down from the host plants.

5.3.6 Abiotic factors

5.3.6.1 Egg stage

Bad weather like heavy rain (rain with wind and hailstorm) were responsible for the lost of many egg clusters. Generally, it destroyed the protective shell (golden brown hairs) of the egg cluster and the purplish brown secretion that cemented the eggs to one another and to the surface of the leaf on which the eggs are deposited.

5.3.6.2 Silkworm stage

In the farms, homestead and along the path sides, many cohorts of silkworms were burned or crushed by people those who thought that they were harmful. This happened when silkworms went down to the bottom of *B. micrantha* host plant before moulting or when they formed a long line to cross the road during the larval movement and colony establishment.

Food depletion was noted to be a mortality cause of silkworms. A canopy of 5 cubic feet could support one cohort of up to 300 silkworms. If more than two cohorts of silkworms were found on the same host plant (*B. micrantha*), many of them died of starvation, especially the smaller ones that had emerged from the eggs at a later stage (2nd brood). Bigger silkworms, usually the sixth and seventh instars were able to leave defoliated young *B. micrantha* host plants and find another host plant. Smaller instars usually did not move away from the host tree but dispersed within the tree itself. Mortality was probably hastened by the combined action of the sun, predators and diseases.

Disturbances also were noted to be a mortality cause of silkworms. Silkworms, especially the smaller instars had a tendency to drop using the silk thread onto the ground when disturbed. These younger silkworms were not able to return to the tree and perished either from food depletion, food deterioration, heat, ants or spider attacks.

5.3.7 Survival rate analysis

5.3.7.1 Egg survival

The factor that contributed most to the egg mortality appeared to be egg parasitism. The survival rate of the egg stage was not very stable particularly in the mixed indigenous forest. Parasitism had a significant effect in determining annual changes in the egg density. The general incidence rate of larvae (instantaneous risk for an egg cluster not to hatch) in the Kakamega Forest was 18%. This incidence rate fluctuated significantly according to the habitat and the brood (Table 5.3).

Table 5.3: Instantaneous risk or incidence rate (%) of *A. panda* egg clusters in the Kakamega Forest.

Years	Broods	Isecheno (mixed indigenous forest)		Ikuywa (indigenous forest)	
		n	Incidence rate (%)	n	Incidence rate (%)
2006	1st	20	15	27	7.4
	2nd	29	27.6	25	20
2007	1st	36	16.7	36	8.3
	2nd	50	28	41	14.6

n: Number of *A. panda* egg clusters at risk

The probability (instantaneous risk) for an egg cluster to be completely parasitized ranged from 15 to 28 per cent and from 7.4 to 20 per cent in the mixed indigenous forest and the indigenous forest respectively (Table 5.3). A highly significant difference ($P < 0.001$) was found between the survivor functions in the two habitats (Log-rank test for equality of survivor functions). The instantaneous risk was minimal in the indigenous forest compared to the mixed indigenous forest. High instantaneous risk was observed for the silkworms which hatched during the rainy season (2nd brood) compared to those of the dry season (1st brood). There was a highly significant difference ($P < 0.001$) between the survivor functions of the 1st and 2nd brood survivor silkworms.

5.3.7.2 Silkworm survival

No significant interaction effect was observed between protection and forest (F = 0.24; df = 3, 176; P = 0.6237), protection and brood (F = 2.34; df = 3, 176; P = 0.1276), protection and three years of study period (F = 0.01; df = 5, 174; P = 0.9944). Also no significant interaction effect was observed between forest and brood (F = 0.1; df = 3, 176; P = 0.7568), and between forest habitat and years (F = 0.54; df = 5,174; P = 0.5816). Between protection, forest and brood (F = 0.94; df = 7, 172; P = 0.4398) and between protection, forest, brood and years (F = 0.77; df = 23, 156; P = 0.7294) no significant interaction effect was observed. However, a positive relationship was observed between protection and forest and also between brood and years; whereas negative relationships were observed between protection and brood, protection and years, forest and brood, and between forest and years.

In this investigation, when silkworms were protected, the survivor function was above the non protected one (Fig. 5.3) and the instantaneous risk function was below the un-protected one (Fig.5.4). Silkworm population declined most rapidly during the larval period. Only 6068 silkworms survived from a hatching population of 104,210 when they were not protected by net sleeve (Fig. 5.4), whereas 16645 silkworms survived from a 138,951 hatching silkworms when they were protected until 7th instar stage (Fig. 5.4). The direct effect on the survival of the generation was demonstrated by the increase numbers of successive populations. There was a highly significant difference ($P < 0.001$) between the protected and the un-protected silkworms (Table 5.4). The incidence rate (instantaneous risk for a silkworm to die) of the protected and the unprotected silkworms by instar is summarized in table 5.4 and Appendix: 8.

Table 5.4: Instantaneous risk or incidence rate (%) of protected and unprotected silkworms by instars in the Kakamega Forest

Instar	Isecheno (mixed indigenous forest)				Ikuywa (indigenous forest)			
	N	Unprotected (%)	n	Protected (%)	n	Unprotected (%)	n	Protected (%)
1st	15198	24.3	12447	10.2	17213	22.4	13124	8.2
2nd	10065	11.2	10702	3.7	11838	9.9	11778	3.4
3rd	6971	6.7	9818	2.4	8711	6	10708	1.7
4th	5081	4.5	8786	1.1	6683	4.2	10083	1.1
5th	3852	3.2	8277	0.6	5217	3	9506	0.7
6th	3083	2.3	7951	0.4	4230	2.2	9126	0.4
7th	2557	1.9	7757	0.4	3511	1.7	8888	0.4

n: Number of silkworms at risk

The probability (instantaneous risk) for a silkworm to die varied from 0.3 to 28.1 per cent and from 0.3 to 27.6 per cent in the mixed indigenous forest and the indigenous forest respectively (Appendix 8). The survivor function was significantly higher ($P < 0.001$) in the indigenous forest compared to the mixed indigenous forest. In the mixed indigenous forest, the instantaneous risk for unprotected silkworms ranged from 1.5 to 28.1 per cent whereas for the protected silkworms, it ranged from 0.3 to 12.6 per cent (Appendix 8). Nevertheless, in the indigenous forest, variations ranged from 1.3 to 27.6 per cent and from 0.3 to 11.4 per cent for the unprotected and the protected silkworms respectively (Appendix 8). In the same habitat, a highly significant difference in variation ($P < 0.001$)

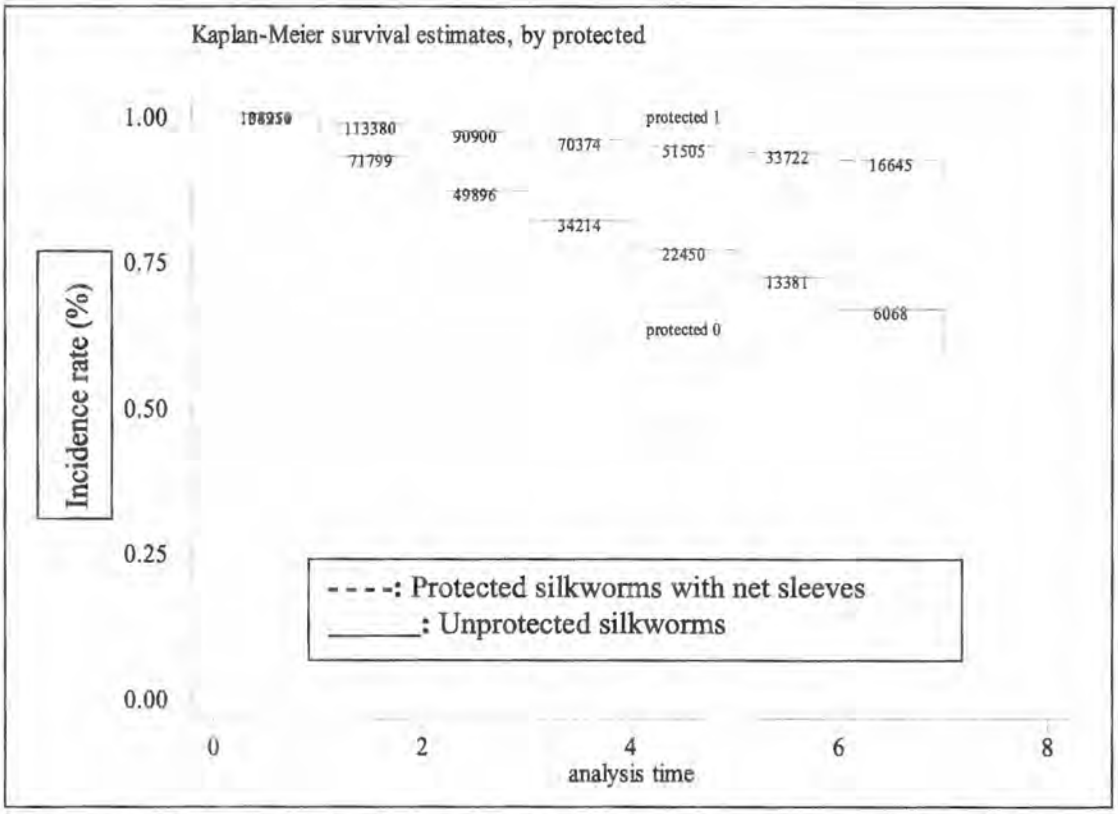


Fig. 5.3: Survivor function of *A. panda* silkworms in the Kakamega Forest

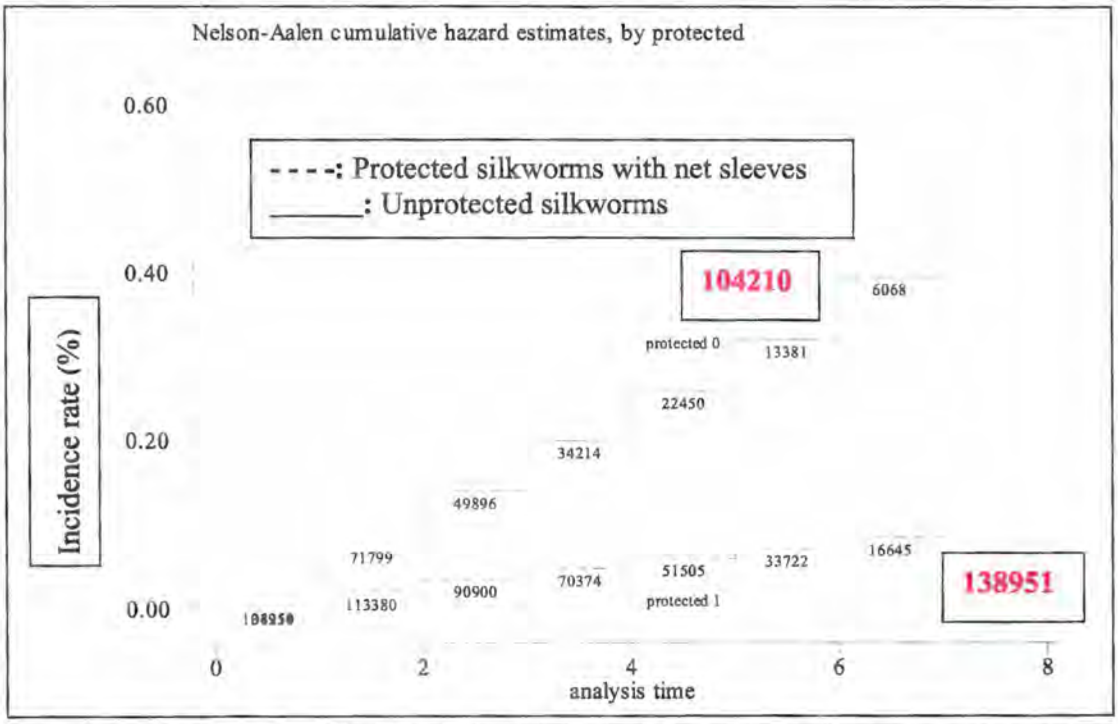


Fig. 5.4: Instantaneous risk function of *A. panda* silkworms in the Kakamega Forest

was recorded between the survivor functions (Fig. 5.3) of the protected (a long dash line) and the unprotected (solid line) silkworms cohort. Protection with net sleeves also minimized the instantaneous risk (Fig. 5.4). Instantaneous risk for silkworms to die also decreased when they were developing. Protection effectively increased the survival rate of silkworms (Fig. 5.3). There was also a highly significant difference ($P < 0.001$) between the survivor functions from 1st to 7th instars both for the protected and the unprotected silkworms. There was a highly significant difference ($P < 0.001$) between the survivor functions of silkworms in the indigenous and in the mixed indigenous forest (Appendix 9.1).

The instantaneous risk was minimized in the indigenous forest compared to the mixed indigenous forest (Appendix 9.2). There was a highly instantaneous risk for the silkworms which hatched during the rainy season (2nd brood) compared to those of the dry season (1st brood) (Appendix 10.2). There was also a highly significant difference ($P < 0.001$) between the survivor functions of the 1st and 2nd brood (Appendix 10.1).

In addition, there was a significant difference ($P < 0.05$) between the incidence rate in 2005, 2006 and 2007, whereas no significant difference ($P > 0.05$) was observed between 2006 and 2007 in the two habitats (indigenous and mixed indigenous forests).

The Cox proportional hazard ratio ranged from 0.21 to 1.34, but the factor for the year (0.96) seemed to be around the benchmark (Table 5.5).

Table 5.5: Cox proportional hazard ratio (± SE) of *A. panda* silkworms in the Kakamega Forest.

Factors examined	Cox hazard ratio	Effect on instantaneous risk
Protection by net sleeve	0.21±0.003	Negatively
Indigenous and mixed indigenous forests	0.85±0.01	Negatively
Brood (1st and 2nd)	1.34±0.016	Positively
Years (2005, 2006, 2007)	0.96±0.007	Negatively

The brood factor positively affected the incidence rate, whereas other factors namely protection and forest habitats negatively affected the incidence rate. However, the year factor (hazard ratio is equal to 0.96) had no significant effect on the hazard.

A highly significant difference was observed between the survivor rate of protected silkworms and those unprotected ($\chi^2 = 15.8$; $P < 0.000$), and also between the indigenous and mixed indigenous forest ($\chi^2 = 36.6$; $P < 0.000$) (Table 5.6).

Table 5.6: *A. panda* silkworm's cohorts failing to survive in the indigenous and mixed indigenous forests in Kakamega

Cohort of silkworms	Isecheno (mixed indigenous forest)		Ikywa (indigenous forest)		
	Protected	Unprotected	Protected	Unprotected	Total
Survive and spun cocoon	38	15	76	51	180
Fail to survive	10	23	2	6	41
Total	48	38	78	57	221

5.3.7.3 Pupal survival

Parasitoids did not appear to play a major role in reducing pupal numbers. Diseases sometimes claimed pupae, especially when the larvae that pupated were diseased. No incidence of pupal predation was observed during the investigation.

5.3.8 Histopathological changes in the tissue silk gland

Light microscope was not able to establish a clear difference between the tissue of sick and healthy silkworms; and also no clear difference were observed between the silk gland of sick and healthy silkworms.

5.4 Discussion

5.4.1 Climatic change factors

Between 2005 and 2007, mean monthly temperature ranged from 16.1 – 39.8 °C but the diurnal temperature ranged between a minimum of 11.1 °C and a maximum of 28.3 °C. This is in conformity with the reports by Muriuki and Tsingalia (1990) and Kokwaro (1988). The mean monthly humidity ranged from 45.4 – 81.4 % in the mixed indigenous forest (Isecheno) whereas the mean monthly maximum humidity ranged from 36.4 – 74.5 % in the indigenous forest (Ikuywa). These findings reveal more about environmental conditions in the Kakamega Forest namely: fine weather, with temperature of about 27° C, and a relative humidity (RH) below 53 % which may have contributed to the maximum *A. panda* moth emergence and the breaking of pupal diapauses. These are probably some external factors which may have influenced the live history of the moth. Ngoka, (2003) also found that the prevailing climate of an area had impact on the development of the wild silkmoth.

The protection of fifth, sixth and seventh instar silkworms with the net sleeve was found to be unnecessary, inappropriate and uneconomical because mortality factors acting at those levels are mainly diseases and abiotic factors like weather and human activities. The indigenous forest habitat seemed to be more suitable for the development of *A. panda* silkworms, compared to the mixed indigenous forest. The difference could be attributed to less disturbance and micro climate of the indigenous forest.

Protection and environmental micro climate (humidity, temperature and rainfall) were found to negatively influence the silkworm's instantaneous risk. Some *A. panda* poikilothermic organisms, the moths life history, activity, distribution and abundance are influenced by temperature (Hill *et al.*, 1999; Dennis, 1993). Generally, increased light and environmental heterogeneity usually lead to a rapid accumulation of species (Brown and Hutchings, 1997). The adult is the only one stage in the life cycle and the abundance of adults could depend on factors operating on the egg, larval or pupal stages of the life cycle (Pollard and Yates, 1993) and these factors may be related to weather in different ways. Pollard and Yates (1985) found that temperature and rainfall are likely to influence the survival of butterflies directly and indirectly through the effects on plant growth, disease, predation or other factors.

5.4.2 Egg parasitoids and egg survival

Eggs in the mixed indigenous forests seem to have been more affected by the weather and parasitoids than those of the indigenous forest, and the infection rate was also significantly different ($P = 0.0025 < 0.05$) between the mixed indigenous and the indigenous forests. The differences could be attributed to the presence of other insects in the indigenous forest which can efficiently control the population dynamics of the *A. panda* parasitoid *P. telenomis*. Those insects which regulate egg parasitoids seemed to be less efficient in the mixed indigenous forest compared to the indigenous forest. It also seemed that the time taken by *P. telenomis* to oviposit in different broods of *A. panda* eggs varied considerably. Nevertheless, further investigation will be required to know more about the life cycle and the mode of oviposition of this *A. panda* parasitoid. Eggs seem to face many challenges in the mixed indigenous forest and this explains why egg survival was highly variable between the indigenous and the mixed indigenous forests. This finding concurs well with Jolly *et al.* (1979) who reported that *T. gowaeyi* and *P. telenomis* are both egg parasitoids of *Anaphe* species.

5.4.3 Silkworms natural enemies

Two of the three *A. panda* silkworm parasitoids which were observed have been reported in the literature (Jolly *et al.*, 1979). The tachinid fly *Exorista cardinalis* and the ichneumonid wasp *Cryptus leucopygus* were found to be parasitoids of the silkworm developmental stage. This finding concurs well with the report by Jolly *et al.* (1979). According to Geertsema (1975) larvae are often parasitized when moulting. During this time, larvae remain quiescent and this probably facilitates oviposition by the parasitoid. According to Rolet and Par (1948), the *Anaphe* silkworm has been reported to suffer only from muscardine. However, in this study, it was not confirm on whether the sick silkworms that died, hanging down from the branches or on the tree stems died from muscardine or other diseases. Further investigations are required to determine the particular type of diseases risk (viral, bacterial or fungal) and identify the causative agent. Furthermore, it has been noted that silkworms which succumb to the diseases caused by viruses or bacteria are easily noticed because the sick silkworms characteristically produce flaccidity and internal liquefaction as describe by Tripconey (1970). Predation of younger silkworms of *A. panda* and diseases of the older instars were factors which made the largest contribution to the silkworm's mortality. More losses of fifth, sixth and seventh instars of *A. panda* silkworms seemed to be due to diseases and abiotic factors but not to predators. The mortality rate fluctuated by years probably due to climatic factors. Indigenous forests seem to be a good environment for the silkworms.

5.4.4 Silkworms survival analysis

Mortality factors exerted a heavy toll on silkworm numbers. Protection of silkworms with net sleeves seemed to minimize the instantaneous risk and effectively increased the survival. The instantaneous risk of silkworms to die (incidence rate) was high for larvae hatching during the rainy season (2nd brood) compared to the 1st brood (larvae hatch during the dry season). The differences could be attributed to predators and also to diseases due to the harsh climatic conditions during the rainy season.

No significant interaction was found between protection and forest, protection and brood, forest and brood, and protection and forest and brood. It appears from this result that the effect of these independent variables were independent for each other. Higher mortality rate was observed from the 1st to 4th instar silkworms, but a highly significant difference was observed between the unprotected and the protected larvae. Nevertheless, the lowest mortality rate was observed from the 5th to 7th instar, and also a significant difference was observed between the unprotected and the protected silkworms. These findings suggest that the use of net-sleeve for protection can be limited to the young silkworms from first to fourth instar. This approach presents many advantages: the fifth, sixth and seventh instar of *A. panda* silkworms are the principal feeding and growing stages and it is important for a silkworm to be free and have food. This also reduces work for the farmers because at those stages with the celerity of leaves consumption, net sleeve should be opened after three days to allow silkworms to migrate to other branches and then net sleeves can be put on those branches. Few net sleeves will also be needed because one net can be used for more than one cohort, and this is economically viable for the adoption of this technology. This approach is also good for silkworm's welfare in terms of movement and colony establishment because the quantity and quality of silk is strongly correlated to food quality and quantity reported by Ngoka (2008).

The findings suggest that the rate of survival of egg and larval stages largely determine the population sizes, hence the quantity of harvested silk cocoon nests.

5.4.5 Pupal parasitoids

A. panda cocoon nests were found to be infested by various dipteran and hymenopteran parasitoids and this confirms to earlier observations by Kioko *et al.* (1999a). In the cocoon nest, 7th instar silkworms were attacked by hymenopteran wasp parasitoids. Marsh (1937) suggested that a few Ichneumonid wasps specialize on pre-pupal larvae, being attracted by the smell of the freshly spun silk. They insert their ovipositors through the partially spun cocoons and lay eggs in the pre-pupal larvae. Such Ichneumonid parasitoids have only a narrow window of opportunity in which to locate and oviposit on the spinning larvae as once the cocoon has hardened, the ovipositor of the parasitoid cannot penetrate it. Further observations need to be carried out to confirm the stage at which the pupal parasitoids attack their host. This will help in developing a protective mechanism for the silkworms, hence the quality of silk cocoon nests. The attack of the pupa of wild sericulture by parasitoids of chacidoid wasps has been reported by (Peigler, 1989; Thangavelu *et al.*, 1988; Jolly *et al.*, 1979). Furthermore, 1st brood (larvae hatched during dry season) cocoon nests seem to be bigger in size than those of the 2nd brood (larvae hatched during rainy season). The presence of many diseases, particularly in the rainy season can be one of the explanations.

CHAPTER SIX

6 SPATIAL DISTRIBUTION OF COCOON NESTS AND EGG-CLUSTERS OF THE SILKMOTH *ANAPHE PANDA* (BOISDUVAL) AND ITS HOST PLANT *BRIDELIA MICRANTHA* (HOCHST) IN THE KAKAMEGA FOREST

6.1 Introduction

Four different types of vegetation are found in the Kakamega Forest: forest with only indigenous species (indigenous forest); mixed indigenous forest (mixed forest with both indigenous and exotic species); hardwood plantation (forest with exotic hard wood species only) and softwood plantation (forest with exotic soft species only). Exotic species are mainly Pines, Black Wattle and *Eucalyptus*. These alien species are preferred because they have been promoted as trees that mature faster and therefore give quicker return on investment. Ikuywa block (380 ha) is a typical indigenous forest (Plate: 6.1) whereas Isecheno block (415 ha) is a mixed indigenous forest (Plate: 6.2).

The insight on spatial distribution of wild silkmoths in the indigenous forest and mixed indigenous forest is one of the several challenges facing wild silk production in Western Kenya. Furthermore, information is required on spatial distribution of species to assist in developing management plans for conservation and their sustainable utilisation for income generation. The use of new technology like geographical information system (GIS) becomes necessary to analyse environmental changes with the aim to develop recommendations for a sustainable biodiversity management.

Plate 6.1: Indigenous forest (Ikuywa block)

Plate 6.2: Mixed indigenous forest (Isecheno block)

This chapter aims to answer the three following questions:

a. Can we identify any forest wide distributional patterns of *A. panda* silkmoth egg-clusters, cocoons nests and the host plants *B. micrantha*?
b. How are *A. panda* silkmoth egg-clusters, cocoons nests and the host plants *B. micrantha* distributed in different habitats (indigenous forest and mixed indigenous forests)?
c. Are *A. panda* silkmoth egg-clusters, cocoons nests and the host plants *B. micrantha* rare in the Kakamega Forest?

According to Watt (1968), information on the distribution of life stages of insects is required for population dynamics studies. For example knowledge on egg distribution also allows greater precision in the location of eggs in the minimum time (Pottinger & LeRoux, 1971). Furthermore, unbiased estimates of populations can only be obtained when distribution of the developmental stages sampled is known (Morris, 1955).

Zar (2005) reported that random distribution of objects/animals in the space is one in which each portion of the space has the same probability of containing an object. Consequently, the occurrence of an object in any portion of the space in no way influences the occurrence of any other objects in any portion of space. Rejection of the hypothesis of randomness (equidispersion) may result from one of the two situations: the population distribution may be underdispersed or: overdispersed. A Poisson distribution has a mean (μ) equal to the variance (σ^2). A distribution in which the mean (μ) equals the variance (σ^2) is equidispersion (random) distribution and $\sigma^2/\mu = 1$. When the variance (σ^2)

is greater than the mean (μ) there is overdispersion and $\sigma^2/\mu > 1$; if variance (σ^2) is smaller than the mean (μ) there is underdispersion and $\sigma^2/\mu < 1$. These two last cases might be an indicator that the Poisson's property; mean (μ) equals variance (σ^2) may be violated, and then the negative binomial distribution which is applicable to a wide range of biological data involving counts of organisms in units of space might be necessary to fit the Poisson distribution (David, 1995; Ludwig and Reynolds, 1988; Ross and Preece, 1985; Pielou, 1977; Geertsema, 1975; Klomp, 1966; Waters, 1955; Morris, 1954; Bliss and Fisher, 1953).

6.2 Materials and methods

Plots were chosen randomly in each block and about 8.6% of the total area of each block was sampled. Sixty-five plots of 5,000 m^2 each were chosen in Ikuywa block (Plate 6.3) and seventy-one of the same dimensions in Isecheno block (Plate 6.4). Sampling was carried out from February to April 2006. This period was chosen because there is high abundance of *A. panda* egg-clusters in the field, each egg-cluster containing between 250-560 eggs.

6.2.1 Spatial distribution of *A. panda* host plant *B. micrantha* (Hoecht)

In each plot, all *A. panda* host plant *B. micrantha* (Hochst) above 0.50 meter high were identified, checked and recorded using a global positioning system (GPS). A tape measure was used for determine the height.

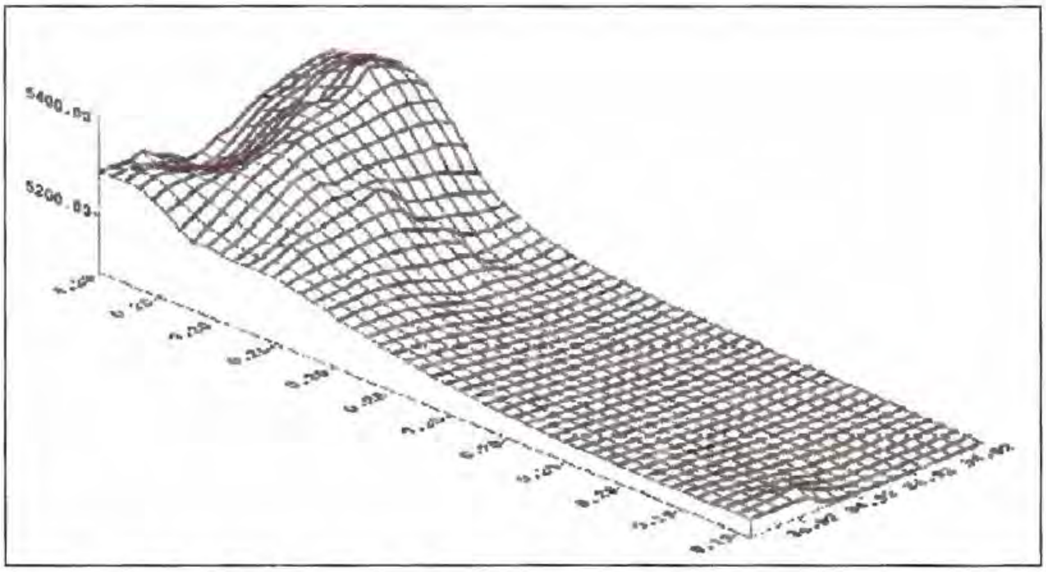


Plate 6.3: Transect sample plots in Ikuywa block (indigenous forest)

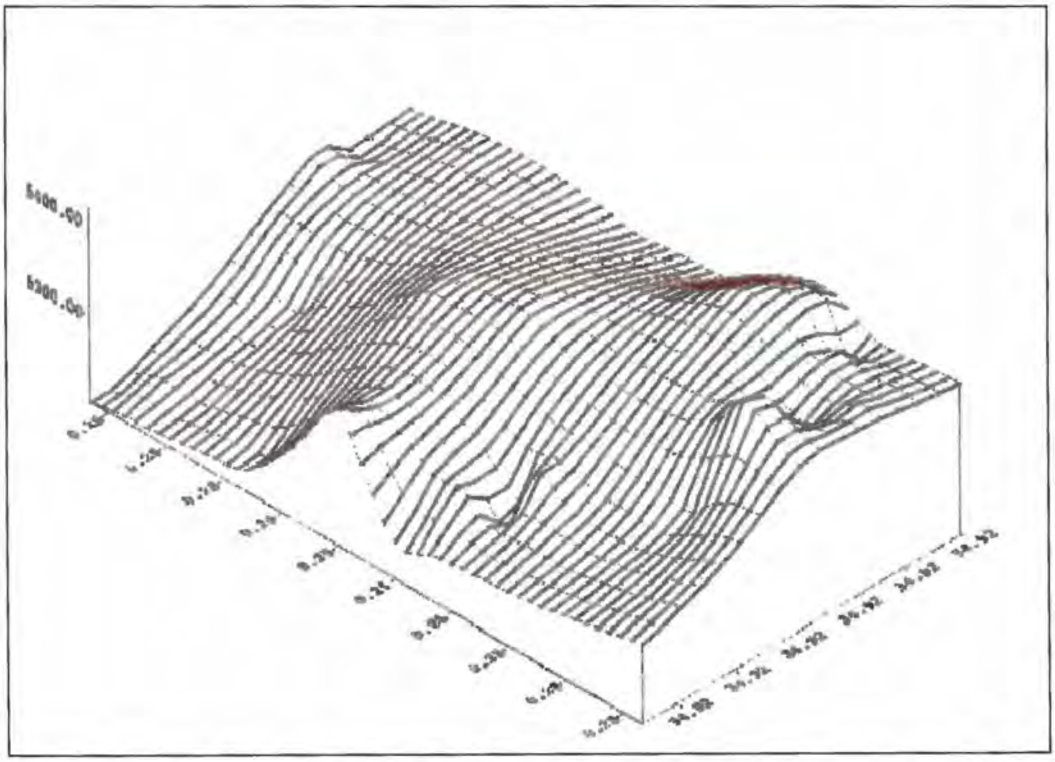


Plate 6.4: Transect sample plots in Isecheno block (mixed indigenous forest)

6.2.2 Spatial distribution of *A. panda* cocoon nests

Cocoon nests were checked not only on the *B. micrantha* host plant but also in several other places, such as the stones, cavities, holes or dead trees in each plot. The cocoons were also counted and their geographical point recorded using a global positioning system (GPS).

6.2.3 Spatial distribution of *A. panda* egg-clusters

In each plot, egg-clusters were checked on the living crown of the *B. micrantha* tree using a Binocular (Spectron 7*50 Multi-coated optics 123 M/1000 M) for the trees above 2.5 meters high. For the tallest *B. micrantha*, the part of crown level over 6 meters high was not checked for egg clusters. The crown was previously divided into three equal levels designated as lower (L), middle (M) and upper (U) respectively. Egg-clusters were identified, counted and recorded using GPS in four cardinal directions; viz North, South, East and West at the three crown levels of the canopy.

6.2.4 Flight range

The flight range: height average of the silkmoth egg-cluster have laid above the ground level was determined with a measuring tape and recorded.

6.2.5 Data analysis

With the assumption of homogeneous conditions in the two blocks of the Kakamega Forest, the Poisson distribution which is a important test in describing random occurrences was used to calculate the expected number of plots with *B. micrantha* trees, cocoon nests and egg-clusters. The following formula was used: $P(X) = \frac{\mu^x}{e^{\mu} X!}$ where P (X) is the probability of X occurrences in a unit of plot and μ is the populations mean number in a unit of plot. Chi-square (χ^2): $\chi^2 = \sum \frac{(observed\ frequency - \exp ected\ frequency)^2}{(\exp ected\ frequency)}$ was used to compare the observed and the expected number of plots where *B. micrantha* trees, cocoon nests and egg-clusters occurred.

The data were tested against the Poisson distribution (Dale, 1989) to determine whether *B. micrantha*, cocoon nests and egg-clusters of *A. panda* were randomly distributed in the indigenous and mixed indigenous forests. The observed and expected frequencies of the number of *B. micrantha*, *A. panda* cocoon nests and egg-clusters per 5,000 m^2 plot obtained from the two blocks of forest were incorporated into the negative binomial distribution (Gates and Ethridge, 1972; Anscombe, 1948). The observed and expected frequencies were compared by χ^2 and frequencies with small expectations (<5) were pooled as suggested by Bliss & Fisher (1953) and Cochran (1954). The degrees of freedom were *k-2* (*k* being the number of categories of the expected frequencies remaining after such pooling) (Zar, 2005). ArcView GIS 3.2 software (1999) and Surfer 6.0 (1995) were used to map geo-reference data. T-test (SAS Institude, 2003) was used to compare between the sampled data of Isecheno (mixed indigenous forest) and that of Ikuywa (indigenous forest).

6.3 Results

6.3.1 Mean numbers of *B. micrantha*, *A. panda* cocoon nests and egg clusters in the two blocks

The mean number of *A. panda* host plant (*B. micrantha*), cocoon nests and egg-clusters in Isecheno (mixed indigenous forest) and Ikuywa (indigenous forest) blocks are summarized in table 6.1. Differences were highly significant between the number of *B. micrantha* and egg-clusters by block, and also there were significant differences between the numbers of cocoon nests between the two blocks.

Table 6.1: Mean (± SE) number of *B. micrantha*, cocoon nests and egg-clusters per plot by block

Blocks (n=2)	Plots	*B. micrantha*		*Anaphe* cocoon		*Anaphe* egg-cluster	
	n	Mean	n	Mean	n	Mean	n
Isecheno (mixed indigenous)	71	7.28 ± 2.96**	517	0.99 ± 1.01*	70	1.37 ± 1.07**	97
Ikuywa (indigenous forest)	65	5.89 ± 2.16	383	1.31 ± 1.20	85	1.82 ± 1.27	118

**: Highly significant difference between blocks ($P < 0.001$). *: Significant difference between blocks ($P < 0.05$)

6.3.2 Spatial distribution of *B. micrantha* in the two blocks

The mean number of *B. micrantha* per plot was significantly lower ($P < 0.001$) in Ikuywa block (5.89) than in Isecheno block (7.28) (Table 6.1). The variance was 8.75 and 4.66 in Isecheno and Ikuywa blocks respectively (Table 6.2). The values of 1.20 and 0.79 for the ratio of variance to the mean were obtained for Isecheno and Ikuywa blocks respectively (Table 6.2). These values indicate that the observed figures (Table 6.3) were more variable than that expected from a Poisson distribution. From this table it can be seen that the discrepancies between the observed and the expected frequencies differ considerably.

Table 6.2: Ratio of the variance to the mean of *B. micrantha*, cocoon nests and egg-clusters by block

Blocks (n=2)	*B. micrantha*			*Anaphe* cocoon nest			*Anaphe* egg-cluster		
	Variance (σ^2)	Mean (μ)	σ^2/μ	σ^2	M	σ^2/μ	σ^2	μ	σ^2/μ
Isecheno	8.75	7.28	1.2	1.01	0.99	1.02	1.15	1.37	0.84
Ikuywa	4.66	5.89	0.79	1.44	1.31	1.10	1.62	1.82	0.89

Values for the ratio of the variance to the mean showed that *B. micrantha* were not randomly distributed throughout the two blocks. *B. micrantha* was found in all the sample plots in both blocks. The minimum number recorded per plot was 3 and 5 in Isecheno and Ikuywa blocks respectively (Table 6.3). The summed χ^2 values obtained for Isecheno block ($\chi^2 = 6.607$; $\chi^2_{0.05,\ 9} = 16.919$; P = 0.68) and Ikuywa block ($\chi^2 = 10.101$; $\chi^2_{0.05,\ 7} = 14.069$; P = 0.18) indicated that the negative binomial distribution agreed with the observed frequency of *B. micrantha* numbers (Table 6.3).

Consequently, spatial distribution of *B. micrantha* at a mean densities of 4.32 to 10.24 and 3.72 to 8.05 trees per 5,000 m^2 at the Isecheno and Ikuywa blocks respectively (Table 6.1) is non-random but overdispersed (contigious) in the mixed indigenous forest and underdispersed in the indigenous forest (Fig. 6.1).

Table 6.3: Observed and theoretical (Poisson distribution) frequencies of *B. micrantha* in plots in two blocks

Number of *Bridelia* per plot	Number of plots at Isecheno block			Number of plots at Ikuywa block		
	Observed ‡	Expected †	χ^2	Observed ‡	Expected †	χ^2
0	0	0.049	0.049	0	0.179	0.179
1	0	0.356	0.356	0	1.057	1.057
2	0	1.295	1.295	5	3.115	1.141
3	5	3.143	1.097	3	6.118	1.589
4	8	5.722	0.907	7	9.013	0.449
5	9	8.334	0.053	14	10.621	1.075
6	12	10.114	0.352	11	10.430	0.031
7	8	10.521	0.604	14	8.780	3.104
8	7	9.576	0.693	4	6.467	0.941
9	6	7.748	0.394	3	4.234	0.534
10	5	5.642	0.073	2	2.495	
11	3	3.735	0.735	1	1.336	
12^+	3	2.266		1	1.155	
13	2	1.269		-	-	-
14^+	3	1.229		-	-	-
Total	71	71	6.607	65	65	10.101

‡: Numbers of plots observed by survey †: Expected number of plots by Poisson distribution.

6.3.3 Spatial distribution of *A. panda* cocoon nests in the two blocks

Frequencies of in the number of cocoon nests obtained from the various plots at Isecheno and Ikuywa blocks are given in table 6.3. The mean cocoon nests per plot was significantly higher ($P < 0.05$) in Ikuywa (1.31) than in Isecheno (0.99) (Table 6.1). More plots without cocoon nests were recorded at Isecheno (39.44% (28/71) than in Ikuywa (21.54% (14/65) (Table 6.4). Furthermore, plots with more than five cocoons were 3.08% at Ikuywa whereas 0% was recorded at Isecheno.

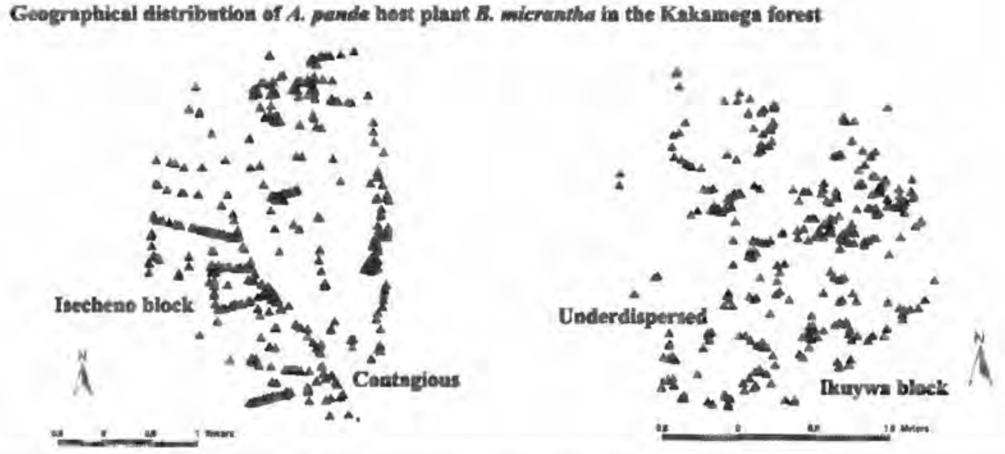


Fig. 6.1: Geographical distribution of *B. micrantha* at Isecheno and Ikuywa blocks

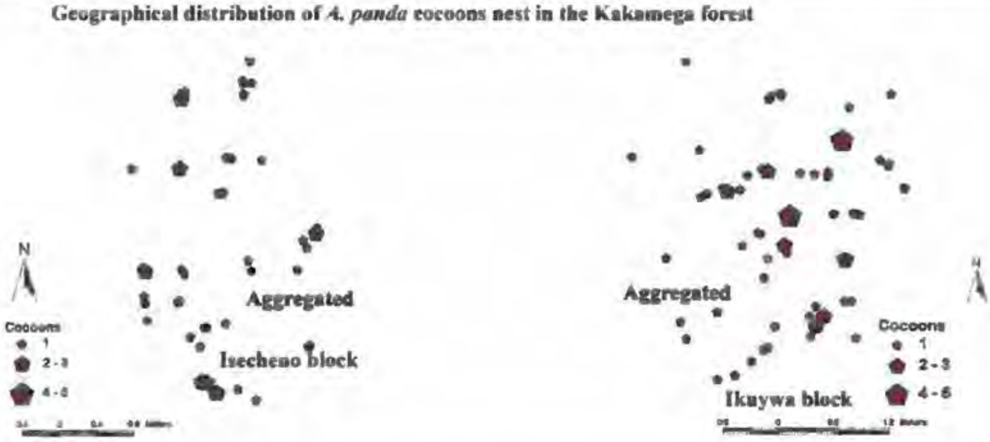


Fig. 6.2: Geographical distribution of *A. panda* cocoon nests at Isecheno and Ikuywa blocks

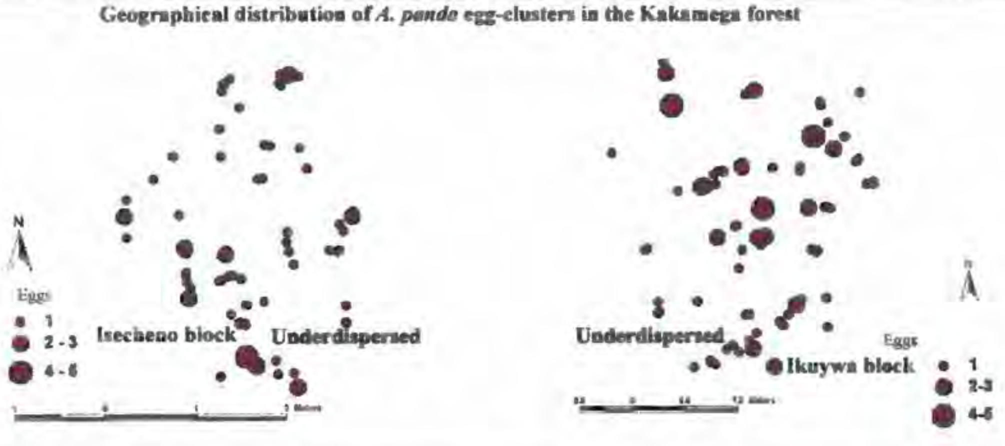


Fig. 6.3: Geographical distribution of *A. panda* egg-clusters at Isecheno and Ikuywa blocks

The variance of cocoon nests was 1.01 and 1.44 in Isecheno and Ikuywa blocks respectively (Table 6.2). The values of 1.02 and 1.1 for the ratio of variance to the mean were obtained for Isecheno and Ikuywa blocks respectively (Table 6.2). The values for the ratio of the variance to the mean showed that *A. panda* cocoon nests were not randomly distributed throughout the two blocks. The summed χ^2 values were ($\chi^2 = 0.598$; $\chi^2_{0.05,\ 1} = 3.841$; $P = 0.44$) and ($\chi^2 = 10.122$; $\chi^2_{0.05,\ 2} = 5.991$; $P = 0.01$) for Isecheno and Ikuywa blocks respectively (table 6.4). This indicates that the observed discrepancies between the observed and the expected frequencies in Isecheno block did not differ significantly while in the Ikuywa block; many plots had less cocoon nests than expected. This clumping is unlikely to have resulted from a random scattering of the cocoon nests in Ikuywa block. Hence, the spatial distribution of *Anaphe* cocoon nests, at mean densities of 0 to 2 and 0.11 to 2.51 cocoon nests per 5,000 m^2 in the mixed indigenous and indigenous forests respectively (Table 6.1) was not random but overdispersed (aggregated) (Fig. 6.2).

6.3.4 Spatial distribution of *A. panda* egg-cluster in the two blocks

The frequencies of the number of *A. panda* egg-clusters obtained for the various plots at Isecheno and Ikuywa are given in table 6.4. The average *A. panda* egg-cluster per plot of 5,000 m^2 was significantly lower ($P < 0.05$) in the Isecheno block (1.37) than in the Ikuywa block (1.82) (Table 6.1). More plots without egg-clusters were recorded at the Isecheno block (23.94%) than at the Ikuywa block (16.92%) (Table 6.4). Furthermore, plots with more than four egg-clusters were not observed at both Ikuywa and Isecheno blocks.

.Table 6.4: Observed and expected frequencies of *A. panda* cocoons nests and egg-clusters in plots in the two blocks

Number cocoons & egg-clusters/plot	Number of plots at Isecheno block (mixed indigenous forest)						Number of plots at Ikuywa block (indigenous forest)					
	Cocoons			Egg-cluster			Cocoons			Egg-cluster		
	Observed	Expected	χ^2	Observed	Expected	χ^2	Observed	Expected	χ^2	Observed	Expected	χ^2
0	28	26.490	0.086	17	18.110	0.068	14	17.579	0.729	11	10.580	0.017
1	23	26.117	0.372	24	24.742	0.022	33	22.988	4.361	18	19.207	0.076
2	14	12.874	0.098	19	16.901	0.261	9	15.030	2.419	16	17.434	0.118
3	5	4.231		9	7.697	0.221	4	6.552	0.994	12	10.550	0.199
4	1	1.043	0.042	2	2.629	0.676	3	2.142	1.619	8	4.788	0.083
5^+	0	0.245		0	0.920		2	0.709		0	2.439	
Total	71	71	0.598	71	71	1.248	65	65	10.122	65	65	0.493

Table 6.5: Distribution of *A. panda* egg-cluster at various geographic locations at different tree crown levels in the two blocks

Localization of egg-clusters & crown levels	Isecheno block (mixed indigenous forest)					Ikuywa block (indigenous forest)				
	North-eastern	South-eastern	North-western	South-western	Total	North-eastern	South-eastern	North-western	South-western	Total
Upper	3	2	2	1	8 (8.25%)	2	1	1	0	4 (3.39%)
Midle	15	11	5	6	37 (38.14%)	15	11	9	10	45 (38.14%)
Lower	21	11	12	8	52 (53.61%)	39	10	12	8	69 (58.47%)
Total	39 (40.21%)	24 (24.74%)	19 (19.59%)	15 (15.46%)	97	56 (47.46%)	22 (18.64%)	22 (18.64%)	18 (15.25%)	118

Values for the ratio of the variance to the mean showed that *A. panda* egg-clusters were not randomly distributed throughout the two blocks. The summed χ^2 values were (χ^2 = 1.248; $\chi^2_{0.05,\,1}$ = 5.991; P = 0.54) and (χ^2 = 0.493; $\chi^2_{0.05,\,2}$ = 5.991; P = 0.96) for Isecheno and Ikuywa block respectively (Table 6.4). This indicated that the spatial distribution of *A. panda* egg-cluster, at mean densities of 0.3 to 2.44 and 0.55 to 3.09 eggs per 5,000 m^2 respectively at Isecheno and Ikuywa blocks (Table 6.1) were not random. *A. panda* egg-clusters in those two blocks were underdispersed (Fig. 6.3).

6.3.5 *A. panda* egg-clusters localization in two blocks

Highly significant differences in the numbers of egg-clusters between crown levels were detected in both blocks. More egg-clusters were laid at the lower and middle crown levels than at the upper crown level (Table 6.5) of the canopy. Less number of egg-clusters was recorded at the upper level (8.25%) in Isecheno block and 3.39% in Ikuywa block, whereas more egg-clusters were laid at the lower level (53.61%) and 58.47% for Isecheno and Ikuywa blocks respectively (Table 6.5). Significant difference was observed between the lower and middle crown levels and a highly significant difference was recorded between the lower and upper crown levels. More egg-clusters were distributed in the North and South-eastern 64.95% (40.21% and 24.74%) and 66.1% (47.46% and 18.64%) directions of the crown levels as compared to the North and South-western 35.05% (19.59% and 15.46%) and 33.89% (18.64% and 15.25%) directions for Isecheno and Ikuywa blocks respectively (Table 6.5). No significant differences in the distribution of egg-cluster by geographical directions were found between the indigenous forest (Ikuywa block) and the mixed indigenous forest (Isecheno block).

6.3.6 *A. panda* flight range in Isecheno and Ikuywa blocks

The lowest and the highest measurements of the flight range were 1.2 - 5.3 m and 1.11 - 3.57 m for Isecheno and Ikuywa blocks respectively. The difference between the minimum and the maximum values obtained for Isecheno and Ikuywa blocks were 4.1 m and 2.46 m respectively. The mean flight range for Isecheno block was 2.06±0.89 m and for the Ikuywa block was 1.81±0.59 m and they were significantly different.

6.4 Discussion

6.4.1 Spatial distribution of *B. micrantha* in the two blocks

This study focused on the spatial distribution of *A. panda* host plant *B. micrantha*, cocoon nests and egg-clusters. Geographical information system and Poisson distribution revealed that distribution of *B. micrantha* at mean densities of 4.32 to 10.24 and 3.73 to 8.05 trees per 5,000 m^2 at Isecheno and Ikuywa blocks respectively was non-random. The spatial distribution of *B. micrantha* was overdispersed (clustered) at Isecheno block (mixed indigenous forest) and underdispersed at Ikuywa block (indigenous forest). This confirms the insufficient populations of *B. micrantha* in the wild and also supports the fact that the population of wild silkmoths in Africa is declining due to the deforestation (Kioko *et al.*, 1999a; Oberprieler, 1994; Munthali and Mughogho, 1992; Ashiru, 1988b).

6.4.2 Spatial distribution of *A. panda* cocoon nest and egg cluster in the two blocks

Similarly, the spatial distribution of cocoon nests at mean densities of 0 to 2 cocoon nests at Isecheno block and 0.11 to 2.51 cocoon nests at Ikuywa per 5,000 m^2 was not-random. Cocoon nests in those two blocks were overdispersed (contagious). Cocoon nests and egg-clusters were not also uniformly distributed in the mixed indigenous (Isecheno block) and the indigenous (Ikuywa block) forests. *B. micrantha*, cocoon nests and egg-clusters were recorded at different densities in the indigenous and the mixed indigenous forests. These findings confirmed the earlier survey by Kioko *et al.* (1999a) who reported that *B. micrantha* was abundantly distributed in western Kenya and 84% of the community members had these trees in varying numbers in their land. All the respondents confirmed to have seen *A. panda* silkworms and cocoon nests, and 16% had seen egg-clusters. Therefore, the introduction of wild silk production in the Kakamega Forest may offer important economic incentives to farmers in western Kenya to actively participate in conservation. In the Kakamega Forest, more than 12,400 hectares are suitable for starting a silkworm host plant plantation. This land can be utilized for the cultivation of *B. micrantha* (12 ha from Isecheno block and 7 ha from Ikuywa block) and the remaining from others blocks. According to Gowdey (1953) *B. micrantha* from cuttings grows fast and is ready for silkworm feeding in one-year period. The current study also revealed that, *A. panda* host plant (*B. micrantha*), cocoon nests and egg-clusters were not randomly distributed in the Kakamega Forest but were overdispersed or underdispersed according to the type of forest (indigenous or mixed indigenous forest).

6.4.3 *A. panda* egg-clusters localization in two blocks

Knowledge on egg distribution allows greater precision in the location of eggs using minimum time (Pottinger & LeRoux, 1971). Fewer egg-clusters were laid at the upper crown level of the tree canopy. However, more egg-clusters were laid at the lower and middle crown levels perhaps because of the larger amount of foliage being available there for oviposition and larval feeding. Geertsema (1975) made similar observations in the Vyeboom and Kluitjieskraal plantation in Cape-town. It appears from this study that *A. panda* tends to distribute its egg-clusters uniformly over the lower and middle crowns of *B. micrantha* with a preference to eastern location of the trees.

From the result of this study, we strongly recommend that (i) indigenous forests should be managed in a sustainable way, (ii) left alone or with a small amount of assistance through seeding and or the introduction of primary indigenous species, even the degraded indigenous forests will regenerate and sustain themselves and (iii) GIS data can be of tremendous importance for conservation, management and long-term monitoring of biodiversity.

CHAPTER SEVEN

7 ECONOMIC POTENTIAL OF WILD SERICULTURE: A CASE OF *ANAPHE PANDA* (BOISDUVAL) SILKWORM

7.1 Introduction

Since thousands of years ago, silk has been treasured as much as diamonds and pearls. This because it has its unique aroma that no man-made fibre can achieve. People all over the world enjoy wearing silk, which is a protein fibre produced by the silkworm. Silk enjoys immense interest and attraction to the public and has certain unique properties to offer; its chemical composition is close to that of human skin and could be one of the healthiest things to wear (Raina *et al.*, 1999). There are 18 amino acids in silk, of which glycine, alamine, serine and tyrosine are key components. These amino acids make silk a natural moisturizer, which is readily absorbed into the skin.

Wild silk constitutes about 10% of the total silk output in the world, but has a strong local and international demand (Raina, 2000; 2004). In order to meet this demand, it is consequently important for the producers of silk to come up with products of topmost quality. In addition, for the success of this initiative there is need to focus on the silk production process and the product quality. The production of raw silk fibre from cocoons is a crucial middle stage in the silk industry. In this regard analysis and testing of raw silk is essential for the production of quality silk fibre. To assure accuracy and replicability, textile testing should be carried out under carefully controlled conditions;

testing equipment must conform to specifications established in testing methodology, and fabric specimen must be of uniform size (Raina, 2004).

The profitability of any enterprise is the key to its success. *A. panda* sericulture practices were used as economic case studies to determine the factors influencing production and marketing, which in turn, influence economic profitability. To turn a profit from a commodity in the long term, money must be spent in the short term. This research also investigated the profitability of a commercial silk production system for farmers living adjacent to the forest as an additional income source

Wild silkmoth farming has its importance on socio-economic and ecological front. The scope of wild silkmoth farming includes creation of employment opportunity for people, perennial and assured income with nominal investment, conservation of the soil, ecosystem, forest and biodiversity.

Sericulture is one of the most labour intensive activities in the world. It involves long chains of interdependent operations that provide income for many people in both agriculture and industry. The basic stages of sericulture include host plant cultivation, leaf collection, silkworm rearing and cocoon reeling. Every stage requires specific skills, large labour inputs and some investment. In addition, each stage induces concurrent services and processing sub sector activities.

In Africa a recent survey indicated that common problems in implementing sericulture technologies include: unclear goals, lack of technology input, lack of training, lack of managerial autonomy and accountability, and financial constraints (Raina, 1998).

Wild silkworms produce silk with different cocoon filament sizes, microstructure, and chemical composition (Kato *et al.*, 1997; 1999b). The fine structure of cocoon filaments from various lepidopteran insects can be classified into two type's namely porous and compact filaments (Akai, 1988; Akai *et al.*, 1988). The former contains numerous tubular structures in all the filaments, but the latter has no such structure. *A. yamamai*, *A. pernyi*, *A. mylitta*, and all other Saturniidae insects spin the porous filament, *B. mori*, *B. mandarina*, and all other orders except Saturniidae spin the compact (Akai and Nagashima, 1999).

A study by Akai and Nagashima (1999), and Hiroshi (2000) revealed several characteristics of the cocoon filaments from *A. infracta, A. venata, and A. moloneyi* cocoon nests, but there is no information on the structural characteristics of the cocoon filaments from other African wild silkmoths like *A. panda.* Degumming, structural properties, and quality control of cocoon filament were studied.

7.2 Materials and methods

To observe the internal structure of the *A. panda* cocoon nest, a cross section of the cocoon nest was cut with the help of scalpel blade (Plate 7.1). Dry field cocoon nests of *A. panda* were collected in the Kakamega Forest and evaluated with respect to cocoon weight, size, and the number of small inner cocoons (Plate 7.2).

Plate 7.1: Cross section of *A. panda* cocoon nest

Plate 7.2: *A. panda* cocoon nests from Kakamega Forest

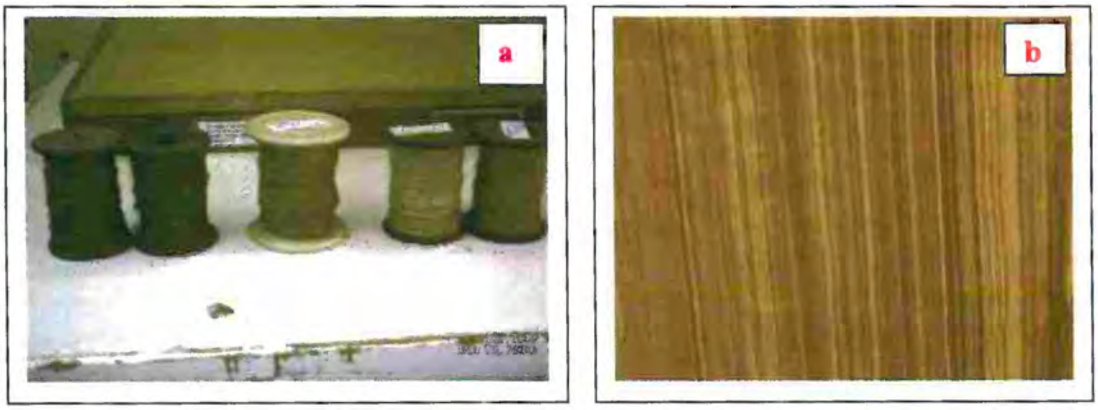


Plate 7.3a, b: *A. panda* silk thread bobbins (a); *A. panda* silk fabric (b)

This species of *Anaphe* which produces silk fibre of high quality has been reported in the Kakamega Forest (Mbahin *et al.*, 2008; Kioko *et al.*, 1999a; 2000). Three hundred and twenty three (323) *A. panda* cocoon nests were evaluated in three years.

To illustrate silk fibre and carry out fabric analysis, the quality of raw silk and fabric produced by *A. panda* silkworms were evaluated (Plates 7.3a, b). The cocoon filament from the connective silk shell (css), the very soft part of the cocoon nest was observed and photographed with a scanning electron microscope (SEM) (Joel JSM-T 330 A), and the fine structure was observed. The silk shell percentage was calculated as follows:

$$Silk\ shell\ percentage = \frac{Weight\ of\ the\ cocoon\ shell}{Weight\ of\ the\ whole\ cocoon} * 100$$

7.2.1 Degumming technology

The cocoons were first immersed in warm water to remove foreign particles and then degummed by the method shown in table 7.1. Alkaline solution (Na_2CO_3) (Hirao, 1998) and other methods (Raina, 2004; Kato *et al.*, 1999a; Ashiru, 1975b) can also be used. After degumming, the cocoons were washed with warm water at about 40 °C and extracted with ethanol for 5 days before using them as samples for analyzing the structural characteristics. These treated samples will be refereed as wild silk in this study.

Table 7.1: Degumming method of *A. panda* cocoon nests

Process	*Anaphe* middle-layer	*Anaphe* outer + inside-layer
Pre-treatment	95 - 98°C (hot water)	95 - 98°C (hot water)
Degumming	Na_2CO_3 (3g/l); 4 – 5hr; 98 - 100°C	Na_2CO_3 (4g/l); 3 – 4hr; 98 - 100°C ↓ (repeat) Na_2CO_3 (3g/l); 4 – 5hr; 98 - 100°C
Finishing	Hot water washing	Hot water washing

Na_2CO_3: Sodium carbonate anhydrous (aqueous solution)

7.2.2 SEM micrographs

The surface structure was observed and photographed with a scanning electron microscope (Joel JSM-T 330 A) with a magnification of 100 times.

7.2.3 Winding breaks

The samples (Plate 7.3a) skeins were fitted onto the winding frame/reel (Appendix: 11.1) and the end attached to a bobbin. Winding was carried out at an average speed of 165 meters/min for 20 minutes. The number of breaks that occurred were recorded.

7.2.4 Tenacity and elongation

A sizing reel of 1125 m in circumference (400 revolutions equal to 450 m) and a constant speed of 300 revolutions per minute are used to prepare the test sample. A serigraph (Appendix: 11.2) with a constant rate of extension (CRE) – speed (15 cm per min) was used to test the tenacity and elongation of the raw silk.

7.2.5 Cleanliness and neatness testing

Cleanliness imperfections are classified as super major defects, major defects and minor defects. Each defect carries penalty points and the difference of the total penalty point from 100 gives the test result. The kind and class to which each defect belongs was determined by comparing it with the standard photographs for cleanliness defects.

Neatness value was estimated in percentages. From 100 to 50 percent the estimate was made to the nearest 5 percent, while below 50 percent the estimate was made to the nearest 10 percent.

Inspection for both tests was done in dark room, from a position of 0.5 meters (2 feet) directly in front of the inspection panels.

7.2.6 Breaking load and elongation

Fabric samples (350 mm by 60 mm) were prepared (Plate 7.3b), five samples with warp yarns running in the direction of stress and the others with the stress in the filling direction.

The serigraph tension was equal to 1±0.25% of the probable breaking load. The average breaking time should be 20±3 seconds according to Kenya Bureau Standard (KS08 – 119, 1981). Breaking strength in each direction was calculated by dividing the sum of observed values of breaking load in Newton (N) by the number of observations.

7.2.7 Tear resistance

Five samples warp-wise and the other five weft-wise were prepared. The sample was clamped onto an Elmendorf tearing tester and the pendulum raised to the starting point and the pointer set against its stop. A slit approximately 20 mm was started on the sample specimen leaving 43.0±0.15 mm of fabric to be torn.

The pendulum stop was depressed, releasing the pendulum. The stop was held till the tear was completed and the pendulum caught on its return wing by the hand without disturbing the position of the pointer

Average force in N was thus calculated as follows:

$N = kg * 9.81$

7.2.8 Moisture Absorption

Moisture absorption was measured by the following method. The wild silk samples were held for 3 days in desiccators containing solutions of sulphuric acid and water the concentrations of which were adjusted to give 35%, 75% and 100% RH and weighed. The dry weight of the samples, air-dried to a constant weight was also measured and the per cent moisture absorption calculated.

7.2.9 Classification of A. panda cocoon filament

In cocoon classification, the mark for grading of cocoon filament length (m) and grading mark of cocoon reelability percent were added to give the final grade: A (over 90), B (88-89), C (86-87), D (84-85), E (below 83). Raw silk is divided into three categories according to their sizes: 1st category: 18 denier and below; 2nd category: 19-33 denier and 3rd category 34 denier and above. *A. panda* raw silk (70 denier and above) belong to the 3rd category. The grade is expressed in the following order in all categories of raw silk in I.S.A standard method: 4A, 3A, 2A, A and B (Yong-woo and Tee, 1999).

7.2.10 Profitability of A. panda sericulture venture

In this case study, two hectares (5 acres) of land with 1,350 *B. micrantha* trees spacing (4m x 4m) was simulated using data observed in the field. The net income gained by the household, when cocoon nests were sold, or processed to raw silk, silk cloth, or silk shirts were calculated. The net added-value was calculated at each level of processing and the cost structure plus optimal profit were also calculated.

Statistical comparison according to the changes of seasons and the years was analysed using chi-square (χ^2), and the degree of significance was determined with the help of Fisher's exact test (Statacorp, 2004).

7.3 Results

7.3.1 Structure and characteristic of *A. panda* silk cocoon nest

Seventh instar larvae of *A. panda* spin a compact silk filament and make a large silk nest. The cross section of the nest revealed an internal structure composed of a thick connective silk shell and numerous individual cocoons (Fig. 7.1).

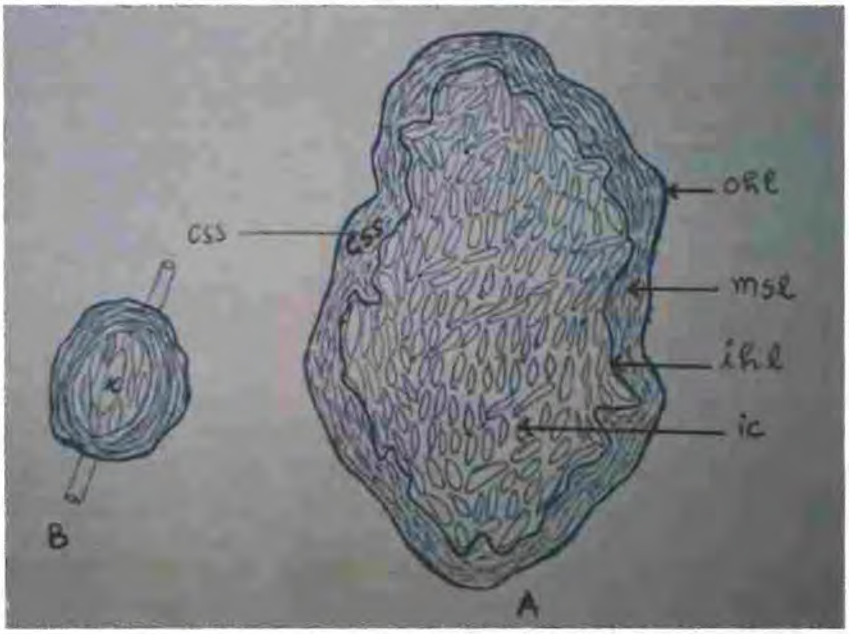


Fig. 7.1: A: The cross section of *A. panda* silk nest; B: small nest; Css: connective silk shell; Ohl: outer hard layer; Msl: middle soft layer; Ihl: inner hard layer; Ic: individual cocoon.

Plate 7.4: The middle soft layer of *A. panda* cocoon nest with many leaflets of silk (arrow)

The connective silk shell is made up of three layers, an outer hard layer (ohl) which was moderately soft, a middle soft layer (msl) which was very soft, and inner hard layer (ihl) which was a hide-like hard part of the cocoon (Fig 7.1). The middle soft layer has many leaves (Plate 7.4).

The 1st brood (silkworms hatch in dry season) cocoon nests seemed to be heavier and bigger than those of the 2nd brood (silkworms hatch in rainy season) (Table 7.2). There was a highly significant difference ($P < 0.001$) between the weights of cocoons coming from the 1st and 2nd brood, and also between the numbers of inner cocoons coming form 1st and 2nd brood of silkworms (Table 7.2).

Table 7.2: Means (±SE) weight and size of *A. panda* cocoon nests in the Kakamega Forest

Years	Brood	n	Cocoon weight (g)	Cocoon length (cm)	Cocoon width (cm)	Number inner cocoon
2005	1st	22	60.54±46.42**	12.23±4.5**	9.27±2.4*	233.3±154.1**
	2nd	32	24.05±18.25	8.42±2.8	7.81±2.7	59.9±33.1
2006	1st	62	69.83±46.43**	14.59±14.2**	10.04±3.4*	276±171.4**
	2nd	46	30.28±23.01	8.77±3.7	8.71±3.9	63.9±38.9
2007	1st	86	67.75±45.07**	13.95±12.5**	10.61±5.1**	269.9±173.9**
	2nd	75	23.34±17.44	8.91±4	7.86±3	62.1±36.6

n: Number of sampling *A. panda* cocoon nests; **: Highly significant difference between broods;
*: Significant difference between broods

The shell ratio varied from 0.27 to 0.50 and it fluctuated year to year. No significant difference ($P > 0.05$) was observed in the shell ratio between the 1st and 2nd brood of the year 2005, whereas in 2006 and 2007 highly significant differences ($P < 0.001$) were observed between the two broods (Table 7.3).

Table 7.3: Comparison of *A. panda* cocoon weight (g), cocoon shell weight (g) and shell ratio (%) in Kakamega Forest

Years	Brood	N	Cocoon weight (g)	Cocoon shell weight (g)	Shell ratio (%)
2005	1st	22	60.5±46.4**	22.5±6.1**	37.1±21.6^{ns}
	2nd	32	24.1±18.3	9.1±4.6	37.6±11
2006	1st	62	69.8±46.4**	28.3±11.2**	40.6±13.2**
	2nd	46	30.3±23.0	15.2±5.1	50.3±10.6
2007	1st	86	67.8±45.1**	24.1±8.3**	35.5±11.1**
	2nd	75	23.3±17.4	6.4±2.4	27.3±8

n: Number of sampling *A. panda* cocoon nests; **: Highly significant difference between broods;
^{ns}: No significant difference between broods

7.3.2 Structural properties of *A. panda* cocoon filament

An examination of the silk cocoons revealed that *A. panda* cocoons could be spun, were not reelable and gave a firm golden brown silk fibre (Plate 7.3a). Each cocoon filament had the appearance of a bamboo pole with a structure like bamboo joint that and be easily seem under SEM (Plate 7.5).

7.3.2.1 Surface structure of *A. panda* filament

Anaphe silk, from the structure revealed by SEM micrograph seemed to have relatively fine fibres. The size (thickness) of the cocoon fibres as estimated from the SEM

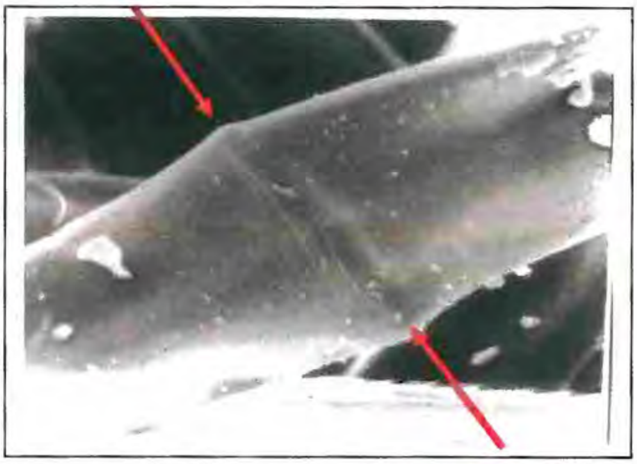


Plate 7.5: SEM of a brin of *A. panda* silk fibre showing the bamboo joint (arrows)

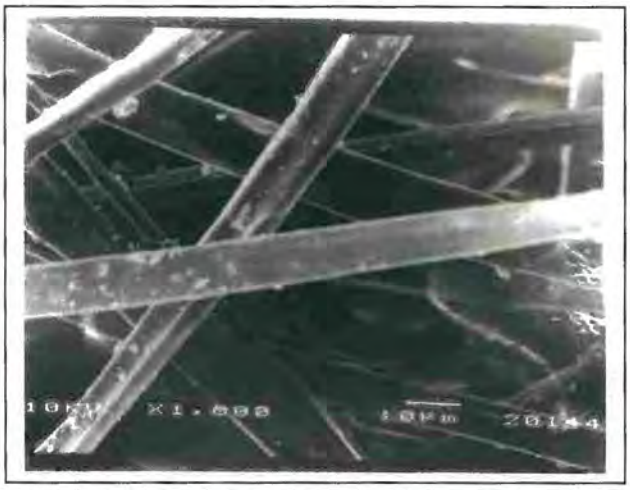


Plate 7.6: SEM of *A. panda* silk filament showing the smooth surface

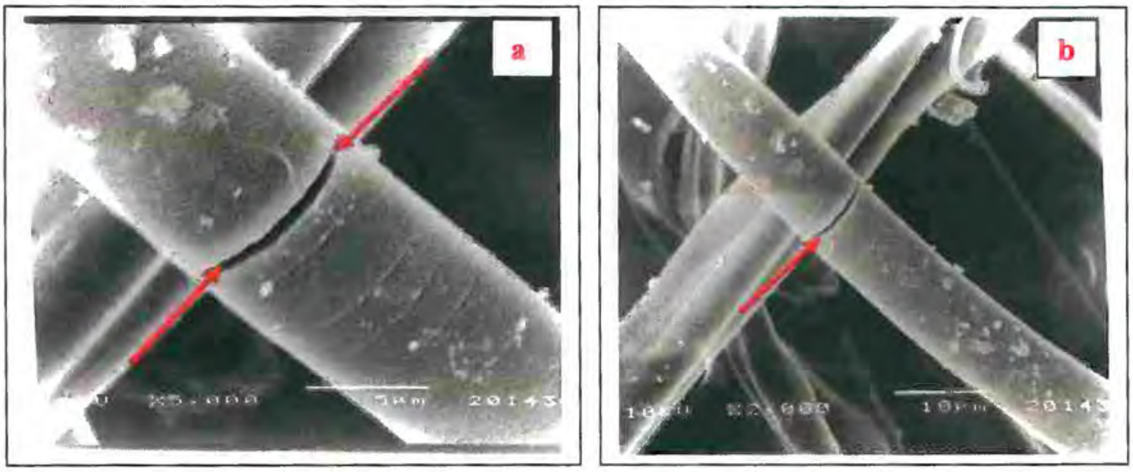


Plate 7.7a, b: SEM of *A. panda* brin showing a smooth surface with shallow grooves (arrows)

micrograph is about 20µm. The surface of *A. panda* silk seemed to be smooth (Plate 7.6) with some shallowing grooves (Plate 7.7a, b).

7.3.2.2 Winding break counts of *A. panda* filament

A. panda raw silk winding breaks varied from a minimum of 9 to maximum of 16 counts according to the sampled bobbins. This study showed that silkworm strains have varying characteristics in their spinning of silk thread. Spinning errors (by spinners) also become evident during this test.

7.3.2.3 Tenacity and elongation percentages of *A. panda* silk

Tenacity of *A. panda* silk thread was 116.63 *N* (12,958.88gms) and the elongation was 9.29mm (8.25%) which are within the ISA standards. Silkworm strains having the highest elongation count had the least winding breaks and vice versa.

7.3.2.4 Cleanliness and neatness percentages of *A. panda* silk cloth

The mean cleanliness and neatness of sampling *A. panda* silk strain is summarized in table 7.4.

Table 7.4: Mean cleanliness (%) and neatness (%) of *A. panda* silk

	Mean cleanliness (%)	Mean neatness (%)
A. panda silk fabric	34	10

A. panda silk failed these two tests because it has a hand spun therefore not even and smooth.

7.3.2.5 Fabric breaking load and elongation of *A. panda* silk

The average breaking load for a common warp yarn of *A. panda* is summarized in table 7.5, and it was above the required mean.

Table 7.5: Breaking load of *A. panda* silk

A. Panda silk strain	Warp	
	Force (N)	Elongation (mm)
A. panda silk	124.02	9.29

7.3.2.6 Tearing strength of *A. panda* silk

The mean tearing strength of a sampling common warp yarn of *A. panda* is summarized in table 7.6, and was also above the required mean

Table 7.6: Tearing strength (±SD) of *A. panda* silk

	Mean tearing strength (±SD)
	Warp
A. panda strain	392.00±2.842

7.3.2.7 Moisture absorption of *A. panda* silk

The moisture absorption of middle-layer, outer and inside-layer is summarized in table 7.7.

Table 7.7: Moisture absorption of degummed *A. panda* silk

Preserved humidity	Moisture absorption (%)		
	100%	75%	30%
A. panda (middle layer)	32.8	13.0	6.7
A. panda (outer & inside layer)	32.1	12.6	6.8

The outer and inner silk layers of *A. panda* showed slightly lower moisture absorption under high humidity conditions than the middle or inner layers. There was no significant difference under lower humidity between the middle layer and the outer plus inside layer.

7.3.2.8 Classification of the common *A. panda* raw silk (34 denier and coarser)

The major items to be considered when classifying of *A. panda* raw silk (above 70 denier) are summarized in table 7.8, and the result was above the mean required.

Table 7.8: Classification of *A. panda* raw silk

Major item	Winding (breaks)	Average neatness (%)	Cleanliness (%)	Elongation (%)	Tenacity (g)
A. panda silk	Class 3 (A)	B	B	Class 2 (B)	Class 1

7.3.3 Wild sericulture venture

Wild silkmoth farming can be a supplementary activity for income generation while conserving biodiversity where rural communities depend on subsistence farming. Before rearing starts, the rearing site and the surrounding area should be cleared of weeds and bushes to reduce shelter for pests and predators. The number of host plant *B. micrantha* must be at least 625 plants for one hectare (2.5 acres) of land at 4m x 4m interval. Planting can be done by cuttings or young tree from nursery in the beginning of the rainy season in March (it is highly recommended to be grown by cutting). Theoretically, and according to Jolly *et al.* (1979), one hectare of plantation can accommodate about 2,200 plants of *B. micrantha* with 3.0 x 1.5 m spacing with a possible yield of 25-27 metric tons of leaves, and this is enough to sustain 6,500-7,000 laid eggs to produce 700-800 kg of cocoons, equivalent to 340-360 kg of unprocessed silk. Wasp and ant nests should be removed from the existing host plants. Natural pesticides, such as neem, should be sprinkled around the based of host trees to prevent attack by ants and other arthropods. From mid-April to August fresh *A. panda* cocoon nests can be picked in the bush for seed and placed on *B. micrantha* trees in the farm. Moths emerge around October, mate and females lay egg clusters of 350-400 eggs on average. Approximately 75% of egg clusters survive to the incubation period (incidence rate of egg cluster 25%). To protect the young larvae from predators and parasites, fine net sleeves are required (Raina, 2000). One hectare plot containing 625 *B. micrantha* trees (with a canopy 10 cubic feet) should produce at least 1,750 cocoon nests (field observation in Kakamega Forest showed that one cohort of 350 silkworms complete all the larval instars on one *B. micrantha* with a canopy of 3.5 cubic feet). In February-March silk cocoons could be harvested from the farms (Table 7.9) and spun with manual hand loom (for value addition) or sent to the factory: Kakamega Silk & Honey marketplace (Plate: 7.8a).

Table 7.9: Calendar of activities for farming of *A. panda* in the Kakamega Forest

Months	Activity
January-February	Clearing field for maintenance
February-March	Harvesting
August- September	Clearing field and preparation of host plants
October-February	Moth emergence and egg production
December-July	Rearing

7.3.4 Profitability of wild silk venture

7.3.4.1 Selling of *A. panda* cocoon nests

One tree with a canopy of 10 cubic feet could support up to 350 silkworms. Two hectares of land of one thousand three hundred and fifty trees (of 10 cubic feet) each may at least yield a total of 3,500 cocoons (the mean weight of *Anaphe* dry cocoon nest is 58 g). The farmer can harvest at least 203 kg of dry *Anaphe* cocoon nests from two hectares of land per year. The value of a kg of cocoon nest is US $5, hence 203 kg worth in total US $1,015 for one cycle per year. The estimated value of one *A. panda* cocoon nest is US $0.29, and by year in the Kakamega Forest only one wild silkmoth crop can be produced, which yields an income of US $1,015/2 hectares/year. If considered this as a family enterprise using a family land of 5 persons, the income based on field data, and labour cost is shown in table 7.10.

Table 7.10: Calculated income in US dollars based on the data of a year (labour cost included)

Expenditure (A)		Income (B)	
Items	Cost	Items	Revenue
Maintenance of 2ha of 1,250 host plant 10 days@US $2/day	20	Sale of cocoon nests (203kg@US $5)	1,015
Nets for early instars rearing*0.5m/cohort for 3,500@0.17	595		
Labour for rearing	55		
Training	30		
Total	700		1,015

*: net sleeve can be reused in the second year season, or for two broods

$$Net\ income = (total\ B - total\ A) = 1{,}015 - 700 = US\ \$315$$

The household now get $315 extra income per year by selling dry cocoon nests ($63 per member of the household per year).

7.3.4.2 Selling of *A. panda* raw silk (spun silk)

The household can decide to process the wild cocoon nest into raw silk (spun silk) by buying one manual spinning wheel machines (Plate 7.8b). To produce 1 kg of *Anaphe* raw silk, 2,190 g (2.19 kg) of *Anaphe* cocoon nests are required. In one year, a two hectare plot of land with 203 kg of *Anaphe* cocoon nests will yield 92.8 kg of raw silk (spun silk). The estimated selling price of raw silk is US $30/kg (Raina *et al.*, 2007). The value added by the household is shown in table 7.11.

Table 7.11: Value added (US $) by transforming the cocoon nest to raw silk (spun silk)

Expenditure (A)		Income (B)	
Items	Cost	Items	Revenue
One manual spinning wheel (US $50@1)	50	Sale of raw silk (92.8kg@US $30)	2,784
cooking pot	50		
Chemicals, fire wood, water and electricity	50		
Labour (US $5@40 days)	200		
Total	350		2,784

$$Net\ income = Total\ income\ (B) - Total\ \exp enditures\ (A) = 2{,}784 - 350 = US\ \$2{,}434$$

The net amount of money they could gained by the household after processing the cocoon nest to raw silk is:

$$Net\ added\ revenue = Income\ raw\ silk - Income\ cocoon\ nest = 2{,}434 - 315 = US\ \$2{,}119$$

The household now get $2,119 extra income per year by processing dry cocoon nests to raw silk (spun silk). This translate into, $424 per a member of the household per year.

7.3.4.3 Selling of *A. panda* silk cloth

Instead of selling raw silk (spun silk), the household can decide to process it into silk cloth by buying one manual hand loom for weaving silk cloth (Plate 7.9). The standard width of cloth is 44 inches (112 cm), and to produce 1 meter of standard *Anaphe* cloth, 229 g (0.229 kg) of *Anaphe* thread silk (spun silk) or 7 bobbins of *Anaphe* thread will be required.

Plate 7.8a, b: Kakamega silk & honey marketplace (a) Manual spinning wheel machine for wild silk (b)

Plate 7.9: Handloom for wild silk cloth weaving

Plate 7.10: Electrical Singer tailoring machine for silk shirt and scarf making

The 92.8 kg of raw silk (spun silk) can fabricate 405m of silk cloth. The estimated selling price of *Anaphe* silk cloth (Plate 7.11) is US $15/meter. The value added on the household revenue is shown in table 7.12.

Table 7.12: Value added (US $) by transforming raw silk (spun silk) to silk cloth

Expenditure (A)		Income (B)	
Items	Cost	Items	Revenue
Manual hand loom weaving silk cloth	1,000	Sale of silk cloth (405 m@US $15)	6,075
Accessories	50		
Labour (US $10@60 days)	600		
Electricity	50		
Total	1,700		6,075

$Net\ income = Total\ income\ (B) - Total\ \exp enditures\ (A) = 6{,}075 - 1{,}700 = US\ \$4{,}375$

The net amount of money that can be gained by the household after processing the raw silk to silk cloth is:

$Net\ added\ revenue = Income\ silk\ cloth - Income\ raw\ silk = 4{,}375 - 2{,}434 = US\ \$1{,}941$

The household now get $1,041 extra income per year by processing raw silk (spun silk) to silk cloth. This translate into, $388 per a member of the household per year.

7.3.4.4 Selling of *A. panda* silk shirts.

If for example instead of selling the cloth, the household chooses to process it into shirts using one electrical Singer tailoring machine which has 35 stitches (Plate 7.10).

Plate 7.11: Wild silkmoth *A. panda* silk cloth

Plate 7.12: Others African silk cloths and threads

If 405 meters of *Anaphe* silk cloth is converted to silk shirts, it can give 175 shirts and the estimated selling price of an *Anaphe* silk shirt is US $50. The value added on the household revenue is shown in table 7.13.

Table 7.13: Value added (US $) by transforming silk cloth to silk shirt

Expenditure (A)		Income (B)	
Items	Cost	Items	Revenue
Electrical Singer tailoring machine	800	Sale of silk shirts (175@US $50)	8,750
Tailoring accessories	250		
Labour (US $15@10 days)	150		
Electricity	100		
Total	1,300		8,750

$$Net\ income = Total\ income\ (B) - Total\ \exp enditures\ (A) = 8{,}750 - 1{,}300 = US\ \$7{,}450$$

The net amount of money they could gain by the household after processing the silk cloth to silk shirts is:

$$Net\ added\ revenue = Income\ silk\ shirts - Income\ silk\ cloth = 7{,}450 - 4{,}375 = US\ \$3{,}075$$

The household now get $3,073 extra income per year by processing silk cloth to silk shirts. Hence, $615 per the member of the household per year. By processing *A. panda* cocoon nests (203 kg) to silk shirts, the net income are multiplied twenty four times (Fig. 7.2).

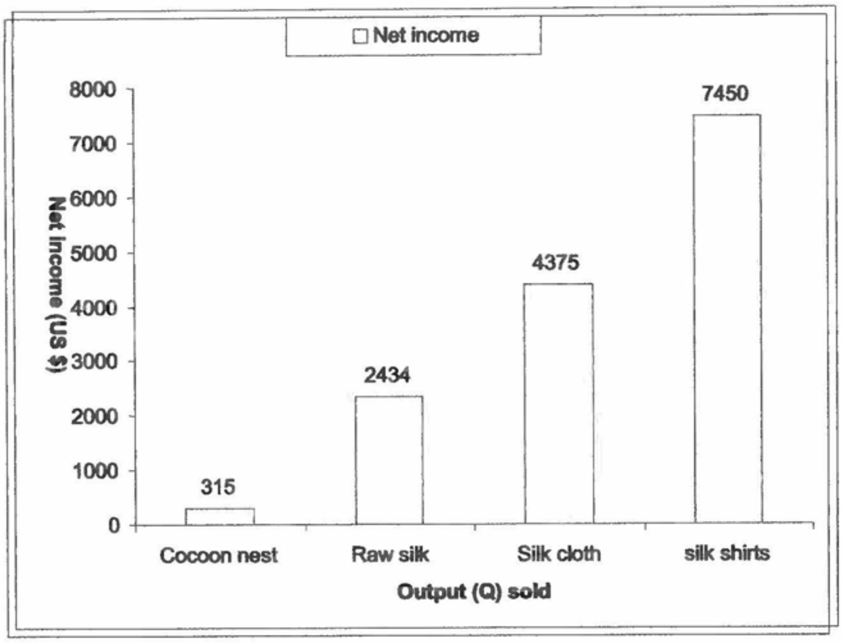


Fig.7.2: Evolution of net income when processing 203 kg of *A. panda* cocoon nests

7.4 Discussions

7.4.1 Structure and characteristic of *A. panda* cocoon nest

The dissection of *A. panda* cocoon nest has shown that the fine structure of cocoon filament is compact and this finding concurs well with findings by Akai and Nagashima (1999) who reported a similar structure working of *A. infracta*. Evaluation of dry cocoon nests revealed that 1st brood of *A. panda* cocoon nest seemed to be bigger than the 2nd brood. There was a highly significant difference between the cocoon nests from 1st and 2nd brood. The prevalence of predators and diseases during the rainy season could offer one explanation. Cocoon size or volume is a critical characteristic when evaluating raw silk materials and the size of the cocoon differs according to rearing season (Yong-woo and Tee, 1999). The most significant commercial feature of cocoons is weight. Cocoons are sold in the marketplace based on weight as this index signals the approximate quantity of raw silk that can be spun. The weight of the silk shell is the most consequential factor as this measure gives a forecast on the yield of raw silk yield. An outer hard layer (ohl) and an inner hard layer (ihl) were observed in this study. This observation concurs with the findings by Yong-woo and Tee (1999) who reported that low humidity during the mounting period makes the cocoon layer soft, while high humidity makes it hard. Being the only remaining rain forest in Kenya, Kakamega Forest is highly humid. A hard shell typically reduces reelability (during the cocoon spinning process), while a soft-shell may increase raw silk defects. In short, moderate humidity is preferred for good quality cocoons, and according to Yong-woo and Tee (1999), temperature near 25°C and relative humidity around 65% for silkworms is a strategy to improve *Bombyx mori* overall cocoon quality (higher silk content, longer filament, better reelability and lower percentage of defective cocoons). This study has shown that mean temperature in the Kakamega Forest fluctuates around this benchmark (25°C), whereas the mean relative humidity seems to deviate significantly from the benchmark (65%).

7.4.2 Structural properties of *A. panda* cocoon filament

A bamboo like structure was found on the *A. panda* fine filament of *A. panda*. This finding concurs well with results by Akai and Nagashima (1999) who reported a similar structure in the filament of *A. infrata*. Consequently, from this finding it can be stipulated that, the fine structure of the filament of all *Anaphe* species have a bamboo like structure. Akai and Nagashima (1999) concluded that this is a characteristic and completely unique structure found in *Anaphe* and doesn't occur in other lepidopteran insects. In the literature, there is no published information on the bamboo joint structure in cocoon filaments of insects. Furthermore, according to Akai and Nagashima (1999), such structures are created during the cocoon filament in the silk gland and it is speculated that these structures are traces of the rhythmical movement of liquid silk, which reflects the spinning behavior and speed. The properties of the silk fibre determine the performance of the fabric produced from it (Down, 1999). The fibre size of *A. panda* silk was about 20 μm and according to Komatsu (1980), wild silk fibres show large variation in fibre size because of wide variation in cocoon size.

7.4.3 Processing and quality control of *A. panda* raw silk

Quality control is not just about making judgment concerning the quality of a product, but also to enable the producer to take the necessary steps to improve the quality of the product throughout the manufacturing process. Silk is a natural fabric and for this reason, some irregularities do occur. *A. panda* raw silk winding breaks occurred mostly during spinning. Due to uneven nature of the yarn, the fine areas were not very strong and probably caused the breaks. The tenacity of the strain was within the ISA standards, because according to Lee (1999), the elongation percentage of raw silk must comprise between 18-23% of its original length. The cleanliness and neatness tests were only suitable for reeled silk. The spun silk was bound to fail these two tests because it was hand spun and therefore not even and smooth. The unevenness is taken as a defect and according to Aruga (1994) the observed defections on those two parameters can be attributed to the technique applied in the cooking and reeling of cocoons. The strength of the fibre structure is commonly regarded as the criterion of quality. One cannot overlook the production processes that silk undergoes. Damage may have occurred during the manufacturing process and hence had an effect on the strength either by weakening or strengthening the fibre and fabric ultimately. Eyre (1956) reported that, the production processes are important factors that may affect the strength of the textile. The mean tearing strength of the warp observed was within the ISA standards. Because *A. panda* cloth was hand spun its denier was above 70. The chemicals used for the degumming process may have had an affect a the strength of the silk fibre and consequently the fabric tearing strength. The change in any physical property and chemical composition of textile material will nearly always result in a change in strength. In this study it is significant to note that, the results of the tests carried out were within the international standard requirements.

7.4.4 Profitability of wild silk venture

Farmers could gain eight times by choosing to process cocoon nest to raw silk; fourteen times by transforming raw silk to cloth, and twenty four time by making shirts (final product) sale in the market. In this venture, farmers could acquire sericulture technology and this could provide extensive employment opportunities to women and youth in the village. Large scale wild silk cocoon production can be a lucrative activity if efforts are made by farmers to establish manual reeling centres in the areas where the cocoons are produced. The profitability of wild sericulture is strongly linked to the number of post-cocoon harvest processing units. With adequate training and transfer of information, small-holders and poor farming communities can undertake sericulture as an agro-based cottage industry. ICIPE is promoting this endeavour by introducing wild silkworm farming to rural farmers. Advances and applications in the complete sericulture technology, which range from silkworm egg-rearing to silk cloth production, will provide extensive employment opportunities and substantial income generation to resource-poor farmers.

A wise management of the natural resources and proper farm husbandry training to the farmers can conserve the habitat of the threatened silkmoth species and their utilisation for silk production can generate additional revenue for the household. In countries where rural communities depend on subsistence farming, wild silkmoth farming can be a supplementary activity for income generation while at the same time helping to conserve biodiversity. Silk farming as a business must be integrated at the local level. It must be taken as far up the value chain as possible (from farming, to fibre processing, spinning, weaving, textile production and finished products) to ensure that enough income is generated to substantially alleviate poverty among the rural poor. Hand woven textiles in particular lend themselves to the design elements as unique to Africa. By targeting the high end market and producing quality original hand loomed pieces in silk with designs inspired by the spirit of Africa, much value can be added to wild silk.

Wherever wild silk is produced, it is usually in relatively small quantities, and as a result it is unique in its own right. Right for the beginning, Africa must accept that it will never be able to compete with silk originating from Asia. Therefore, Africa needs to produce products that are uniquely African, with new ideas and designs and consequently distinct from the Asian silks.

CHAPTER EIGHT

8 GENERAL DISCUSSIONS, CONCLUSIONS AND RECOMMENDATIONS

8.1 General discussions

There was a steady increase in variability of *A. panda* silkworm from the first to the seventh instars. Klomp (1966) ascribed the increase in variability to the following causes: some partly genetic, attributable to sex as well as genetic differences within the sexes; others partly environmental. For instance, factors which induce a variable number of larval instars are differences in microhabitats (indigenous and mixed indigenous forests). Morris and Fulton (1970) also attributed to genetic divergence or a response to the environmental conditions. Harsh environmental conditions, such as winter, may curtail the embryos or larval growth and development for a certain period. This period of arrested growth, which can occur at any developmental stage, is called diapause. Development resumes when more favourable environmental conditions return.

According to Van den Berg (1968), sex ratio's obtained for pupae of some Saturniidae family can be identical for adults. The pupa sex ratio also differs from that of the emerging adults. In this study, males were dominant at pupa stage whereas females are slightly dominant at the adult stage in the mixed indigenous forest, but there was a ratio 50:50 in the indigenous forest. Similar conclusions were reported by Morris and Fulton (1970) working on *Hyphantria cunea* in Canada, and Mors (1942) in South Africa for *Lymantria monacha*. They observed that in the case of larval and pupal stages of the two insects, a slight excess of males occurred but this was subsequently offset by a slightly higher mortality of male pupae during diapause, resulting in an even 50:50 sex ratio for this moth. Differential mortality therefore resulted in the change of the sex ratio (Miller, 1963).

Differences in the sex ratio between the pupal and adult stages of *A. panda* were also investigated by Miller (1963). Stern and Smith (1960) concluded that polyhedrosis virus and other factors were responsible for a larger female mortality of *Colias philodice eurytheme* pupae because of the slower rate of development of females. In the case of *A. panda*, it seems to be the inverse and further studies are needed to determine what is responsible for the high mortality of male pupae.

It would also be important to work out the underlying genetic and endocrinological control mechanisms, which govern the biology of the various *Anaphe* populations. Because molting involves the activation of cerebral neurosecretory cell and the prothoracic glands before metamorphosis, the corpora allata presumably are inactivated. Abiotic factors apparently or probably may affect or synchronize these events in many individuals.

As regards of predation, birds were observed to pose a threat to *A. panda* silkworms and this finding concurs well with observations reported by Jolly *et al.* (1979). Nevertheless, birds were not the only key mortality factors because silkworm's mortality was also associated with Formicidae ants and spiders. Ants that were observed predating on *A.*

panda larvae have also been reported as predators by other researchers in wild silkmoth (Ngoka, 2003), and in tsetse fly predation (Rogers and Randolph, 1984; Carpenter, 1920; Fiske, 1920).

Higher mortality rates were observed for young larvae (from 1st to 4th instars), while lower mortalities rate were observed for older larvae (5th to 7th instars). This finding suggests that net sleeves should only be used to protect young larvae (from 1st to 4th instars); the fifth, sixth and seventh instars which are the principal feeding and growing stages of the silkworms should be left free because they do not need net sleeves protection. This approach can make the technology socially acceptable and economically viable for farmers. According to Gowdey (1953) the cardinal points in *Anaphe* silkworm rearing are sufficient food, ample shade and minimum disturbance.

Pleurotropis telenomis was found in this study to be one of the major *A. panda* egg parasitoid. This finding suggests that this parasitoid can have negative effect on silkworm farming. However, wherever *A. panda* silkworms become a pest by controlling the number of hatching *A. panda* eggs. There is therefore need to carry out further research work to determine the biology and life history of *P. telenomis*, and its effect on *A. panda* population in the field.

The mortality rate observed during the developmental period of *A. panda* silkworms in this investigation was significantly higher in the unprotected silkworms than in the protected silkworms. The highest mortality rate was recorded on unprotected silkworms reared in the mixed indigenous forest, whereas the lowest mortality rate was recorded in the indigenous forest on the protected silkworms. This is an indication that net sleeves acted as mechanical barriers to natural enemies especially from 1st to 4th instars. Silkworm mortality was not significant in most cases when net sleeves were removed during the 5th to 7th instars between the protected and unprotected cohorts. The impact of net sleeves as a protective device could be used effectively as a mechanical barrier from (1st to 4th) instar silkworms. By protecting the earlier larval instars (1st to 4th), the population is enhanced but other challenges remain. The challenge here is how egg clusters could be protected from egg parasitoids and how 5th to 6th instars silkworms could be protected from bacterial, fungal and/or viral diseases.

Parasitoids from Tachinidae and Ichneumonidae families were observed surrounding *A. panda* pupae. It is therefore possible that the way those parasitoids act could be fatal to *A. panda* moth population. Probably they lay their eggs on or in the silkworms. After some weeks, silkworms of these parasitoids start feeding and killing the silkworm's larvae. Parasitism was also observed on *Gonometa sp.* and Hartland-Rove (1992) proposed that parasitoids laid eggs in or on larvae and later the adult parasitoids emerged from the cocoons. Marsh (1937) suggested that a few Ichneumonid wasps specialize on pre-pupal larvae, being attracted by the smell of the freshly spun silk. The pupal parasitoids obtaining the current study leave unanswered question as to whether they laid eggs on larvae or pitched to lay eggs on freshly spun cocoon nests. Therefore, further work is required to explain this process.

The fact that the indigenous forest environment was more suitable for *A. panda* silkworms, the establishment of a *B. micrantha* plantation for the rearing of *A. panda* silkworms, farmers should mixed with other indigenous tree species and not with exotic trees species.

The spatial distribution of cocoon nests and egg clusters of the silk moth *A. panda* provided usefull information on one of the treasures of the Kakamega Forest. Unfortunately, many factors hindered the growth of wild sericulture in this region. These included insufficient populations in the wild, lack of well-researched and applicable information and poor extension services. Sericulture extension concerning wild silk farming would be more successful and better sustained if more information is built upon and the villages' interest also incorporated. Educating farmers on rearing techniques and in conservation of biodiversity would ultimately result in the improvement of wild silk production as a supplementary income generating activity. One way can be to educate local villagers adjacent to the Kakamega Forest on the importance of silkmoth population build up, the economic viability of wild sericulture and its potential to generate supplementary income for resource-poor farmers, reduce deforestation and conserve wild silkmoth species diversity. However, it should also be noted that farmers are free to follow the advice of extension or to ignore it. Thus, extension would only achieve its goal if it also meets the interests of farmers (Van den Ban and Hawkins, 1998). This study recommends utilization of Geographical Information System (GIS) for long term monitoring of biodiversity.

Based on SEM observations of *Anaphe panda* cocoon filaments, it can be concluded that the bamboo joint structure is characteristic and unique as compared with all other known cocoon filaments of lepidopteran insects. Such filament characteristics of *Anaphe* species are believed by Akai and Nagashima (1999) to provide a unique textile nature at the commercial level. Further studies are needed to determine why and how such a structure is probably created during the silk formation in the silk gland.

8.2 Conclusions

Africa has an advantage of being able to introduce new types of silk to the World market. Kenya has an incredible pool of art, craft, history and culture that can draw from and work into African creations. Africa has the exciting opportunity of being able to introduce completely new forms of wild silk and products to the world market. There is a need to create new products, which will attract new customers. This calls for exploration of new avenues of how wild silk can be utilized.

Designers around the word are always looking for new and interesting fabrics. Wild silk offers them the opportunity to be able to experiment with new and interesting natural materials. Every body is aware of the ever-growing move in the developed World towards "organic" products and natural fibres. There is need to capitalize on this. There is also need to educate the designers and consumers on the benefits that come with using or wearing wild silk. Producers should combine production under one regional banner and cooperative market. Silk producers like many other high value agricultural product producers can benefit from cooperative marketing. To effectively compete in the special market, the quality of all materials, from threads to the completed product must be consistently excellent. Thus, there must be quality control.

8.3 Recommendations

In the Kakamega forest, the *A. panda* female moth seemed to know instinctively which host plant to lay her eggs on (*B. micrantha*), probably sensing chemical substances characteristic of that plant. However, further works on what chemical attract the moth only on *B. micrantha* and also what repulse them from other trees (push and pull studies). For the mass production of *A. panda* cocoon nests, we strongly recommend the using of *B. micrantha*, during the reforestation campaign of the 12,000 hectares of buffer zone in the Kakamega forest.

To enhance the quality of cocoon nests, further observations need to be carried out to confirm the stage at which the pupal parasitoids attack their host. This will help in developing a protective mechanism for the silkworms or pre-pupal stages

To achieve this noble task, there is a need for collaboration between the private business sector and the scientific community to overcome bottlenecks in the production of silk threads. The quality, quantity, and price of reeled and spun silk will be major factors in attracting producers, artisans, weavers and other stakeholders in the silk industry. Despite the various problems facing silk industry, it enjoys immense interest and attraction to the public. Furthermore, silk has certain unique properties to offer; its chemical composition is close to that of human skin and could be one of the healthiest things to wear (Raina *et al.*, 1999).

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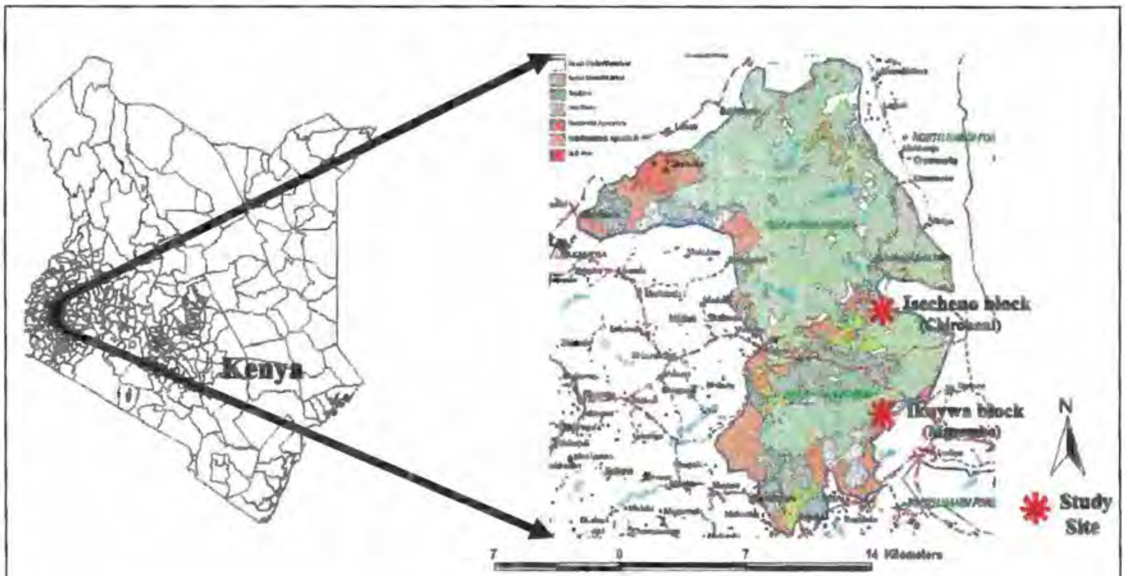
APPENDICES

Appendix 1: Position of Kakamega Forest in Kenya



Appendix 2: Mean (± SE) period (days) spent at each stage; and mean weight (g) and length (mm) of *A. panda* silkworms in the indigenous and mixed indigenous forests in Kakamega

Years	Instar	Brood	Isecheno (mixed indigenous forest)				Ikywa (indigenous forest)			
			n	Period (days)	Weight (g)	Length (mm)	n	Period (days)	Weight (g)	Length (mm)
2005	1st	1st	17	18.5±0.94	0.007±0.002	3.62±0.35	19	18.11±1.24	0.009±0.001	3.62±0.3
		2nd	11	21.81±0.6	0.009±0.002	3.69±0.34	13	21.15±0.69	0.007±0.001	3.46±0.28
	2nd	1st	17	9.06±0.75	0.017±0.002	5.8±0.6	19	9±0.88	0.016±0.003	5.58±0.65
		2nd	11	11.91±0.54	0.016±0.003	5.94±0.69	13	11.92±0.64	0.014±0.002	5.23±0.29
	3rd	1st	17	9.35±0.93	0.032±0.006	7.88±0.64	19	9.58±0.84	0.036±0.006	8.76±0.59
		2nd	11	12.73±0.47	0.032±0.006	8.26±0.41	13	12.92±0.49	0.033±0.007	8.59±0.56
	4th	1st	17	9.41±0.94	0.072±0.036	17.8±0.49	19	9.53±1.07	0.097±0.105	16.7±1.99
		2nd	11	14.42±0.98	0.067±0.11	18.2±0.45	13	15.54±0.88	0.074±0.009	17.4±0.91
	5th	1st	17	10.71±0.85	0.186±0.13	19.7±0.9	19	10.21±1.23	0.207±0.02	20.8±1.34
		2nd	11	15.91±1.58	0.197±0.03	20.1±1.42	13	16.23±1.17	0.235±0.23	22.2±1.36
	6th	1st	17	10.24±1.25	0.906±0.07	36.9±2.35	19	10.74±1.19	1.015±0.077	39.8±1.35
		2nd	11	15.64±0.67	0.854±0.01	37.6±0.48	13	16±0.71	0.95±0.097	38±2.52
	7th	1st	17	18.18±0.64	3.05±0.13	51.1±2.59	19	18±1.05	3.097±0.098	51.5±1.82
		2nd	11	22.18±0.87	2.99±0.17	49.9±2.94	13	21.69±0.95	3.082±0.112	50±1.86
2006	1st	1st	18	18.22±1.22	0.007±0.002	3.74±0.23	15	17.93±1.03	0.009±0.001	3.65±0.24
		2nd	15	21.67±0.62	0.007±0.001	3.35±0.35	12	21.5±0.9	0.008±0.002	3.43±0.38
	2nd	1st	18	9.11±0.76	0.017±0.002	5.89±0.75	15	9.07±0.59	0.017±0.002	5.81±0.68
		2nd	15	12±0.76	0.015±0.003	5.4±0.58	12	12±0.6	0.016±0.002	5.96±0.58
	3rd	1st	18	9.22±0.88	0.037±0.005	9.09±0.46	15	9.87±0.64	0.036±0.005	9.2±0.28
		2nd	15	12.73±0.7	0.034±0.001	8.87±0.58	12	12.83±0.94	0.035±0.006	9±0.425
	4th	1st	18	9.56±1.04	0.073±0.001	16.8±1.94	15	9.53±1.13	0.162±0.232	16.9±1.6
		2nd	15	15.33±1.18	0.069±0.011	16.1±1.51	12	15.5±0.8	0.073±0.009	16.4±1.68
	5th	1st	18	10.44±0.78	0.203±0.023	20.3±1.74	15	10.27±1.1	0.23±0.022	21.9±1.28
		2nd	15	15.73±1.28	0.208±0.029	20.8±1.82	12	15.92±1.31	0.2±0.026	20.1±1.62
	6th	1st	18	10.78±1.17	0.966±0.097	38.4±2.56	15	10.87±1.19	0.942±0.09	37.3±3
		2nd	15	15.73±0.96	0.966±0.098	37.8±3.11	12	15.83±0.84	0.962±0.085	38.7±2.26
	7th	1st	18	18±0.84	3.022±0.144	49.5±4.98	15	17.73±0.88	2.978±0.15	49.7±3
		2nd	15	21.87±0.92	2.965±0.158	49.7±2.8	12	21.75±0.87	3.014±0.146	50.2±2.44
2007	1st	1st	16	19.13±0.89	0.009±0.001	3.68±0.26	17	18.35±1.32	0.009±0.002	3.61±0.37
		2nd	13	21.62±0.65	0.008±0.002	3.5±0.35	14	21.57±0.76	0.009±0.001	3.72±0.28

2nd	1st	16	9.13±0.81	0.016±0.003	5.82±0.56	17	9.12±0.7	0.017±0.003	6.14±0.7
	2nd	13	12.08±0.28	0.015±0.003	5.59±0.75	14	11.93±0.83	0.017±0.003	6.12±0.73
3rd	1st	16	9.44±0.89	0.033±0.006	8.83±0.4	17	9.59±0.94	0.035±0.005	8.8±0.56
	2nd	13	12.92±0.28	0.035±0.008	8.85±0.58	14	12.79±0.8	0.037±0.006	9.05±0.48
4th	1st	16	9.56±1.26	0.096±0.11	16.1±1.72	17	9.59±1.06	0.071±0.008	15.7±1.84
	2nd	13	15.46±0.88	0.071±0.01	16.9±1.58	14	15.5±0.85	0.108±0.118	16.8±1.41
5th	1st	16	10.25±1	0.204±0.02	20.4±1.45	17	10.35±1.06	0.201±0.024	20.4±1.58
	2nd	13	15.92±1.38	0.23±0.023	21.9±1.55	14	16.43±1.02	0.202±0.023	20.4±1.6
6th	1st	16	10.25±1.39	0.931±0.08	38±2.4	17	10.29±1.26	0.932±0.093	38.0±2.35
	2nd	13	15.46±0.97	0.968±0.1	39.1±2.45	14	16.07±0.73	0.975±0.089	38.8±1.89
7th	1st	16	18.25±0.77	2.904±0.136	48.9±2.17	17	18.12±0.86	2.972±0.148	49.8±2.62
	2nd	13	21.54±1.05	2.898±0.094	48.1±1.52	14	21.79±1.05	2.974±0.176	50.2±3.16

Brood: 1st: Dry season (earlier), 2nd: Rainy season (later), n: Number of hatches egg clusters

Appendix 3: Average leaf consumption (g) and total feeding time (days) of *A. panda* silkworms in the Kakamega Forest

			Isecheno (mixed indigenous forest)			Ikuywa (indigenous forest)		
Years	Instars	Broods	n	Leaf consumption (g)	Total feeding time (days)	n	Leaf consumption (g)	Total feeding time (days)
2006	1st	1st	3	0.83±0.04	15.33±0.58	4	0.83±0.03	15±0.82
		2nd	4	0.74±0.02	17.75±0.96	3	0.75±0.02	17.67±1.15
	2nd	1st	3	0.97±0.05	8.33±0.58	4	0.98±0.03	7.75±0.96
		2nd	4	0.87±0.03	10.25±0.96	3	0.88±0.02	10.33±1.54
	3rd	1st	3	1.27±0.07	7±0.01	4	1.27±0.04	7.5±0.58
		2nd	4	1.13±0.03	9.25±0.01	3	1.14±0.03	9.67±0.58
	4th	1st	3	1.85±0.01	8.67±0.58	4	1.85±0.07	8.5±0.58
		2nd	4	1.66±0.05	9.25±0.5	3	1.67±0.04	9±0.01
	5th	1st	3	4.82±0.25	8.33±0.58	4	4.83±0.17	8.25±0.5
		2nd	4	4.31±0.13	10.75±0.5	3	4.36±0.01	10.67±0.58
	6th	1st	3	14.11±0.73	9±0.01	4	14.14±0.5	9.25±0.5
		2nd	4	12.62±0.37	14.75±0.96	3	12.76±0.29	14.33±1.53
	7th	1st	3	24.82±1.28	15±0.01	4	24.86±0.87	14.25±0.96
		2nd	4	22.19±0.66	21.75±0.96	3	22.24±0.51	21.67±1.15
2007	1st	1st	2	0.76±0.01	15±1.41	3	0.75±0.02	15.33±0.58
		2nd	3	0.73±0.04	18±1	3	0.74±0.03	18.33±1.15
	2nd	1st	2	0.89±0.01	8±1.41	3	0.88±0.02	7±0.01
		2nd	3	0.86±0.05	10±1	3	0.87±0.03	9.67±1.15
	3rd	1st	2	1.16±0.02	7.5±0.7	3	1.14±0.03	7.67±0.58
		2nd	3	1.12±0.07	9±1	3	1.13±0.04	9.33±0.58
	4th	1st	2	1.69±0.03	9.5±0.7	3	1.67±0.04	8.67±0.58
		2nd	3	1.63±0.1	9.33±0.58	3	1.65±0.06	9±0.01
	5th	1st	2	4.41±0.07	8±1.41	3	4.36±0.01	8.67±0.58
		2nd	3	4.26±0.26	10.67±0.58	3	4.29±0.15	10.33±0.58
	6th	1st	2	12.91±0.21	8.5±0.71	3	12.76±0.29	8.67±0.58
		2nd	3	12.47±0.77	15±1	3	12.57±0.44	15.33±1.15
	7th	1st	2	22.7±0.36	14.5±0.71	3	22.44±0.51	14.67±0.58
		2nd	3	21.93±1.35	22±1	3	22.1±0.78	22.33±1.15

Brood: 1: Earlier (dry season); 2: Later (rainy season); n: Number of cohorts

Appendix 4: Length (cm), width (cm) and weigh (g) of *A. panda* pupae in the Kakamega Forest

			Isecheno (mixed indigenous forest)				Ikuywa (indigenous forest)			
Years	Brood	Sex	n	Weigh (g)	Length (cm)	Width (cm)	n	Weigh (g)	Length (cm)	Width (cm)
2005	1st	m	22	0.53±0.17	2.14±0.17	0.74±0.06	19	0.54±0.09	2.11±0.2	0.73±0.07
		f	18	0.73±0.21	2.37±0.11	0.82±0.05	25	0.71±0.17	2.43±0.14	0.84±0.01
	2nd	m	20	0.51±0.14	2.08±0.12	0.72±0.07	17	0.49±0.15	2.14±0.21	0.74±0.06
		f	20	0.79±0.21	2.37±0.13	0.82±0.06	12	0.66±0.16	2.4±0.16	0.83±0.06
2006	1st	m	22	0.48±0.13	2.14±0.14	0.74±0.07	29	0.53±0.11	2.06±0.16	0.73±0.08
		f	14	0.67±0.2	2.44±0.17	0.81±0.09	24	0.71±0.17	2.42±0.21	0.83±0.12
	2nd	m	27	0.55±0.15	2.18±0.16	0.75±0.09	32	0.51±0.13	2.1±0.15	0.74±0.06
		f	26	0.74±0.18	2.39±0.16	0.8±0.09	30	0.72±0.2	2.4±0.14	0.82±0.06
2007	1st	m	26	0.47±0.17	2.09±0.16	0.72±0.06	36	0.51±0.14	2.06±0.16	0.73±0.06
		f	15	0.72±0.2	2.43±0.14	0.83±0.06	35	0.71±0.19	2.39±0.13	0.83±0.07
	2nd	m	36	0.47±0.12	2.11±0.17	0.72±0.07	42	0.49±0.16	2.11±0.19	0.73±0.07
		f	32	0.75±0.18	2.41±0.17	0.82±0.08	33	0.75±0.18	2.4±0.17	0.82±0.07

Brood: 1: Earlier (dry season); 2: Later (rainy season). Sex: m: male; f: female; n: Number of pupa

Appendix 5.1: Monthly rainfall (mm) and number of rainy days by year (2004-2007) in the mixed indigenous forest (Isecheno block)

Months	2004†		2005‡		2006‡		2007‡	
	Rainfall	Days	Rainfall	Days	Rainfall	Days	Rainfall	Days
January	104.6	11	100.9	10	56	5	18.5	5
February	69.4	8	75.9	7	54	6	18	7
March	95.8	13	225.8	16	345.5	16	91.5	6
April	320.6	24	169.7	17	297	19	197	12
May	167.2	20	343	22	337.5	22	113	16
June	158.4	27	146.5	19	329.5	19	278	15
July	76.1	20	117	20	97.5	15	123	12
August	236.4	27	363.5	23	199.2	18	227.2	20
September	212	22	247	13	181.5	15	206.3	17
October	133.8	20	134.9	13	91.5	11	109.7	12
November	127.5	14	59	6	466.5	28	112	7
December	106.8	13	13	4	203	22	4	1
Total	1808.6	219	1996.2	167	2658.7	196	1498.2	130

† Source: KEEP; ‡ Source: Our survey

Appendix 5.2: Mean monthly temperature: Temp. (°C) and relative humidity: RH (%) (2005-2007) in the mixed indigenous forest (Isecheno block)

Month	2005				2006				2007			
	Temp.		RH		Temp.		RH		Temp.		RH	
	d	n	d	n	d	n	d	n	d	n	d	n
January	35.4	19.7	48.9	55.6	39.8	22	50.2	57.3	25.8	19.3	62	59.9
February	32.5	23.1	45.4	54.3	28	22.6	48	55	29.4	22.1	54.1	55.6
March	26.3	24.6	62.2	60.7	27.8	22.4	64.8	62.9	28.8	22.5	50.4	53.3
April	26.1	22.4	72.5	77.2	27.5	19.3	75.7	78.8	27.3	22.7	59.7	55.5
May	27.7	19.4	76.9	79.2	29.8	20.9	84.6	86.2	27.7	19.8	66.5	64.6
June	25.2	20.5	77.4	78.1	26.3	19.9	76.4	77.7	25.6	19.4	79.7	81.2
July	27.2	17.3	72.6	68.1	29	17.7	70.4	69.7	28.3	18.2	69.1	68.7
August	27.1	15.9	74.3	78.9	26.5	23.8	68.6	66.7	26.3	22.4	71.6	68.2
September	25.4	15.5	75.6	77.9	26.9	23.5	67.3	63.4	25.7	22.5	74.8	69.3
October	27.6	16.1	71.2	81.4	27.7	23.4	61.6	61.1	26.7	20.5	62.1	61
November	29.6	17.4	65.8	73	25.5	24.8	68.1	68.6	26.4	20.2	60.1	63.4
December	32.4	19.5	53.7	65.8	25	20.6	69.9	68.3	28.3	24.5	66.7	70.2

Temp.: Temperature; RH: Relative humidity; d: day time; n: night time

Appendix 6.1: Monthly rainfall (mm) and number of rainy days by year (2004-2007) in the indigenous forest (Ikuywa block)

Months	2004†		2005‡		2006‡		2007‡	
	Rainfall	Days	Rainfall	Days	Rainfall	Days	Rainfall	Days
January	110.7	8	139.5	13	66.8	7	54.6	6
February	60.4	9	72.6	5	40.7	9	76.1	10
March	123.8	9	157.8	17	230.7	17	96	10
April	357	27	160.9	15	264.3	24	189.5	13
May	169.2	22	326.1	28	259.8	26	268.5	18
June	208.7	23	130.2	21	237.2	17	267.2	15
July	94.1	16	121.7	18	136.3	19	127.4	17
August	277.4	24	377.4	29	199.2	18	252.7	22
September	209.1	19	181.1	18	179.5	15	183	16
October	143.2	16	198	22	91.5	11	114	13
November	124.8	15	49.9	11	423.6	29	132	8
December	108	13	18	8	117	15	7	2
Total	1986.4	201	1933.2	205	2246.6	207	1768	150

† Source: BIOTA-Est Africa / KARI; ‡ Source: Our survey

Appendix 6.2: Mean monthly temperature: Temp. (°C) and relative humidity: RH (%) (2005-2007) in the indigenous forest (Ikuywa block)

Months	2005				2006				2007			
	Temp.		RH		Temp.		RH		Temp.		RH	
	d	n	d	n	d	n	d	n	d	n	d	n
January	32.9	22.6	35.8	41.2	35.2	23.4	36.4	39.9	23.1	22.8	69.9	82
February	31.5	21.8	37.1	39.7	33.8	23.3	39.7	42.6	23.2	23.6	71.4	81.5
March	33.7	21.1	60.1	69.8	32.6	20.4	60.1	69.8	23.1	23.9	70.5	73
April	30.4	22.5	64.7	70.2	29.2	23.3	65.3	72.1	22.9	23.4	73.7	80.9
May	27.6	23.1	70.3	71.9	25.6	23	71.9	72.1	23.4	23	73.4	80
June	26.2	22.1	72.5	70.6	25.6	23.1	74.5	70.3	24.3	22.6	73.7	69.6
July	25.3	23.7	72.2	65.5	25.6	24.3	72.5	67.4	25.5	24.1	70.3	66.8
August	25.1	16.5	71.6	63.2	26.5	23.8	68.6	66.7	25.2	21.1	69.3	65.2
September	24.8	17	70.1	63.7	26	22.7	65.1	61.3	24.4	20.2	67.7	62.5
October	26.7	17.7	68.7	62.6	27.7	23.4	61.6	61.1	26.5	19.9	63.2	60.6
November	30.1	17.8	54.3	55.9	23.8	23.2	63.8	64.4	25.5	22.6	58.3	61.7
December	35.2	20.2	40.5	38.9	21.3	25.1	71.6	78.2	27.1	23.3	67.4	71.3

Temp.: Temperature; RH: Relative humidity; d: day time; n: night time

Appendix 7: *A. panda* silkworm's mortality rate (%) by instars from protected and unprotected experiments in the indigenous and mixed indigenous forests in Kakamega (2005-2007).

Years	Instars	Brood	Protected	Isecheno (mixed indigenous forest)		Ikuywa (indigenous forest)	
				Cohort (n)	Losses (%)	Cohort (n)	Losses (%)
2005	1st	1st	0	9	41.2±7.9	10	35.2±5.1
			1	8	16±3.8	9	10.8±4.5
		2nd	0	6	63.7±20.5	7	61.3±10.5
			1	5	17.4±6.1	6	14.8±2
	2nd	1st	0	9	40.3±10.3	10	26±4.8
			1	8	13.2±5.1	9	8.3±4.4
		2nd	0	6	46.9±16.6	7	53.6±13.6
			1	5	11.9±2.2	6	10.3±2.5
	3rd	1st	0	9	34.9±8.5	10	20.9±6.1
			1	8	12.6±5	9	4.1±1.8
		2nd	0	6	41.8±9.5	7	49.4±16.7
			1	5	9.3±7.7	6	4.6±3.1
	4th	1st	0	9	31.8±7.4	10	17.9±7.5
			1	8	8.1±5	9	4.6±2.3
		2nd	0	6	38.4±8.7	7	45.8±18
			1	5	3.6±1.7	6	2.5±1.4
	5th	1st	0	9	26.4±8.3	10	15.9±7.1
			1	8	3±2.5	9	2.9±1.7
		2nd	0	6	30.2±9.5	7	37.3±18.3
			1	5	2.8±1.7	6	2.3±0.7
	6th	1st	0	9	24.2±7.6	10	13.2±6.2
			1	8	2.8±2.3	9	2.3±1.5
		2nd	0	6	26.8±7.8	7	31.9±15.7
			1	5	3.2±1.7	6	2.3±1.4
	7th	1st	0	9	22±8.5	10	12±5.3
			1	8	2.9±2.2	9	2.6±1.6
		2nd	0	6	23.6±5.4	7	30.3±15.7
			1	5	3.4±1.2	6	2.5±1.6
2006	1st	1st	0	9	46±13.3	8	35.6±11.4
			1	9	13.7±4	7	9.5±8.1
		2nd	0	8	65.6±19.7	6	43.9±21.5
			1	7	15±4.8	6	11.4±6.7
	2nd	1st	0	9	40.4±11	8	33.3±10.4
			1	9	7.3±3.7	7	6.9±5.5
		2nd	0	8	53.8±12.8	6	41.1±24.7
			1	7	7±2.7	6	9.5±3.3
	3rd	1st	0	9	31.1±13.9	8	30.3±11.8
			1	9	5.8±3	7	8.4±6.4
		2nd	0	8	49.2±16.3	6	36.3±17.7
			1	7	9.4±3.8	6	6.1±3.1
	4th	1st	0	9	29±10.5	8	24.1±10.3
			1	9	3.5±1.6	7	4±1.5
		2nd	0	8	44.8±17.3	6	33.6±20.8
			1	7	4.9±4	6	5.2±1.6
	5th	1st	0	9	24.6±13.2	8	22.9±10.8
			1	9	3.6±1.2	7	2.8±1.8
		2nd	0	8	34.7±13.2	6	24.8±16.4
			1	7	3.3±1.2	6	3.4±1.3

	6th	1st	0	9	21±11.1	8	20.9±9.4
			1	9	2.1±1.2	7	2.5±1.7
		2nd	0	8	27.9±11.5	6	24.1±15.6
			1	7	2.6±1.6	6	3.2±1.2
	7th	1st	0	9	17.3±9.2	8	16±8.8
			1	9	2.1±1	7	3.5±1.2
		2nd	0	8	30.5±17.5	6	24.7±17.3
			1	7	3.2±1	6	3.1±1.6
2007	1st	1st	0	8	41.2±14.1	9	37.2±6.8
			1	8	8.6±3.6	8	6.6±2
		2nd	0	7	51.8±25.2	7	47.8±12
			1	6	10.3±3.7	7	11.8±3.5
	2nd	1st	0	8	34.8±10.8	9	28.6±7.6
			1	8	6.6±3	8	6.5±3.4
		2nd	0	7	49.5±26.5	7	38±11.7
			1	6	11.1±5.5	7	12.4±5.4
	3rd	1st	0	8	34.9±13.7	9	24.7±6.8
			1	8	8.4±2.9	8	5.4±3.2
		2nd	0	7	38.7±11.2	7	38.9±5.3
			1	6	7.7±2.3	7	9.2±3.7
	4th	1st	0	8	26.1±12.6	9	29.1±6.2
			1	8	5.1±3.4	8	6.1±2.5
		2nd	0	7	31.8±14.5	7	33.9±8.2
			1	6	5.2±2.7	7	8.9±5.5
	5th	1st	0	8	28.2±13.3	9	25±11.4
			1	8	4.3±0.7	8	4.9±1.6
		2nd	0	7	32.7±17.6	7	32.6±15.2
			1	6	3.8±0.8	7	5.4±1.1
	6th	1st	0	8	23.1±9.4	9	24±9.5
			1	8	3.7±1.5	8	3.9±2.8
		2nd	0	7	30.3±14.4	7	30.1±13.2
			1	6	3.8±1.4	7	5.1±2.5
	7th	1st	0	8	27.5±13.6	9	22.3±8.2
			1	8	4±0.9	8	3.4±1.6
		2nd	0	7	28±15	7	30.6±11.5
			1	6	4.3±0.7	7	4.9±1.1

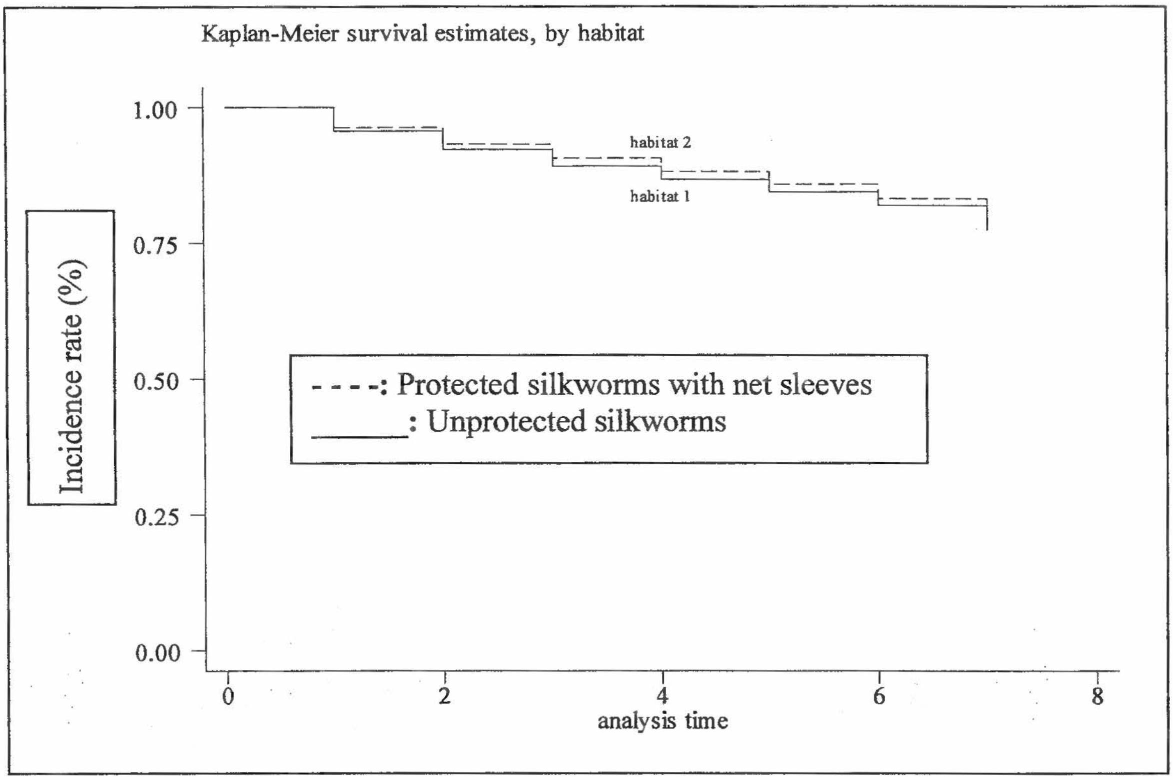
Brood: 1= Egg hatch in dry season, 2= Egg hatch in rainy season; Protected: 0= Larvae instar not protected by net sleeve, 1= Larvae instar protected by net sleeve.

Appendix 8: Incidence rate (%) of *A. panda* silkworms in the Kakamega Forest

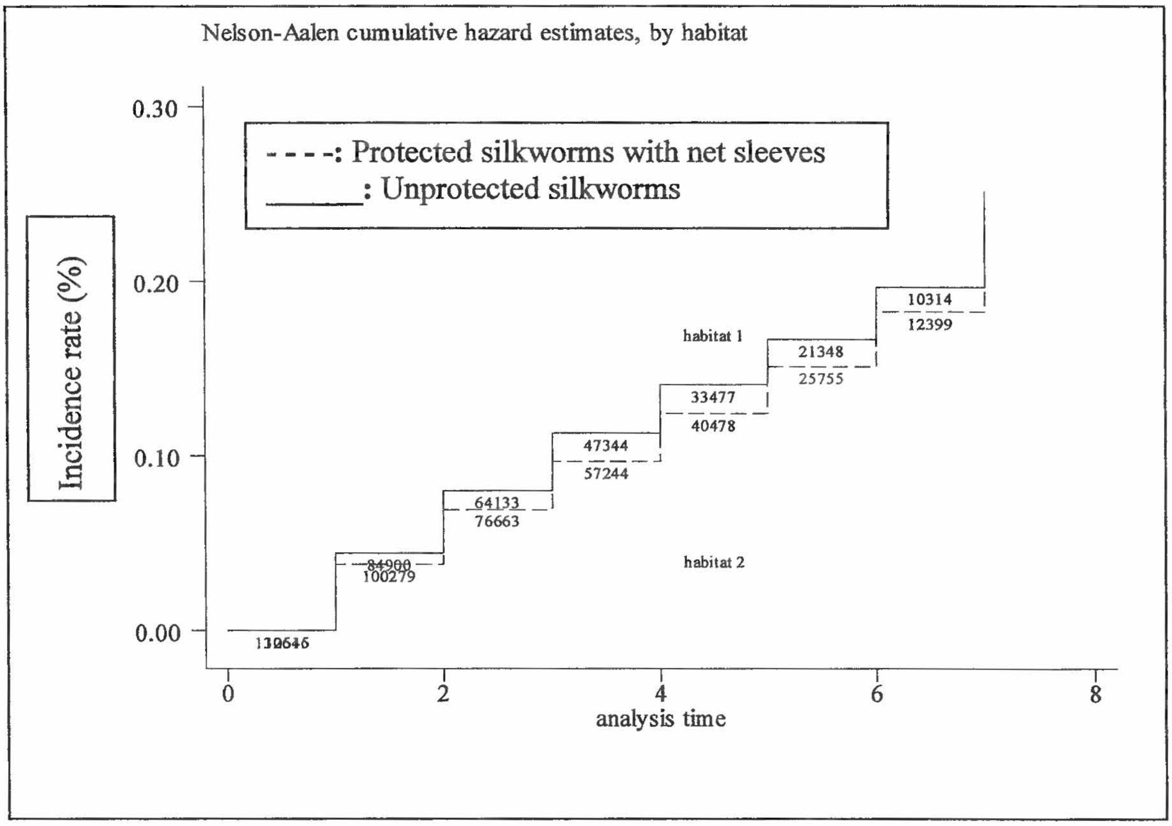
Years	Instars	Isecheno (mixed indigenous forest)					Ikuywa (indigenous forest)			
		Brood	n	Unprotected	n	Protected	n	Unprotected	n	Protected
2005	1st	1st	3745	22.6	2914	12.2	4025	20.4	3362	8.9
		2nd	1762	28.1	1030	12.6	2794	27.6	1689	11.4
	2nd	1st	2635	11.1	2461	5.2	2870	8.5	2963	3.4
		2nd	1024	12.3	854	4.9	1686	12.9	1423	4.2
	3rd	1st	1835	6.7	2155	3.2	2221	4.9	2666	1.3
		2nd	675	7.7	741	2.5	1073	8.1	1235	1.4
	4th	1st	1352	4.7	1869	1.7	1805	3.4	2554	1
		2nd	466	5.4	652	0.8	715	5.6	1160	0.6
	5th	1st	1010	3.2	1652	0.5	1505	2.4	2422	0.5
		2nd	324	3.7	627	0.5	489	3.8	1127	0.4
	6th	1st	805	2.5	1603	0.4	1271	1.7	2342	0.3
		2nd	246	2.8	611	0.5	362	2.8	1099	0.3
	7th	1st	651	2	1566	0.3	1108	1.3	2292	0.3
		2nd	193	2.3	593	0.4	285	2.3	1079	0.3
2006	1st	1st	2983	23.4	2689	10.6	2613	20.1	1985	6.4
		2nd	2352	27.6	2011	11.2	1957	23.1	1644	8.6
	2nd	1st	2025	10.8	2252	2.9	1923	9.4	1820	2.4
		2nd	1407	12.5	1662	3.1	1336	10.5	1474	3.9
	3rd	1st	1392	5.9	2104	1.8	1453	5.8	1739	1.9
		2nd	913	7.7	1573	2.5	958	6.4	1319	1.8
	4th	1st	1097	4.4	1937	0.8	1107	3.6	1606	1
		2nd	620	5.3	1394	1	711	4.3	1231	1.2
	5th	1st	825	2.9	1877	0.7	910	2.6	1518	0.4
		2nd	432	3.4	1323	0.6	539	2.6	1151	0.6
	6th	1st	671	2	1784	0.3	767	2.1	1477	0.3
		2nd	348	2.3	1272	0.4	451	1.9	1112	0.5
	7th	1st	562	1.5	1753	0.3	634	1.3	1466	0.4
		2nd	289	2	1253	0.4	387	1.5	1071	0.4
2007	1st	1st	2715	21.7	2485	6.7	3731	20.9	2657	5.5
		2nd	1638	25	1318	8.3	2093	24.6	1787	9.3
	2nd	1st	1907	9.8	2268	2.6	2662	9.2	2494	2.5
		2nd	1067	11.7	1205	4.4	1355	10.8	1604	4.7
	3rd	1st	1436	6.3	2180	2.2	2023	5.6	2341	1.4
		2nd	720	7	1065	2.2	983	7.2	1408	2.5
	4th	1st	1040	3.6	1965	1	1645	4.6	2247	1.2
		2nd	506	4.3	969	1.1	700	5	1285	1.7
	5th	1st	864	2.9	1886	0.8	1256	3.4	2124	0.9
		2nd	397	3.4	912	0.7	519	3.9	1164	1
	6th	1st	703	2.1	1802	0.6	992	2.7	1997	0.5
		2nd	310	2.5	879	0.5	387	3	1099	0.7
	7th	1st	613	2	1737	0.6	795	2.1	1933	0.4
		2nd	249	2	855	0.6	302	2.6	1047	0.6

n: Number of silkworms at risk

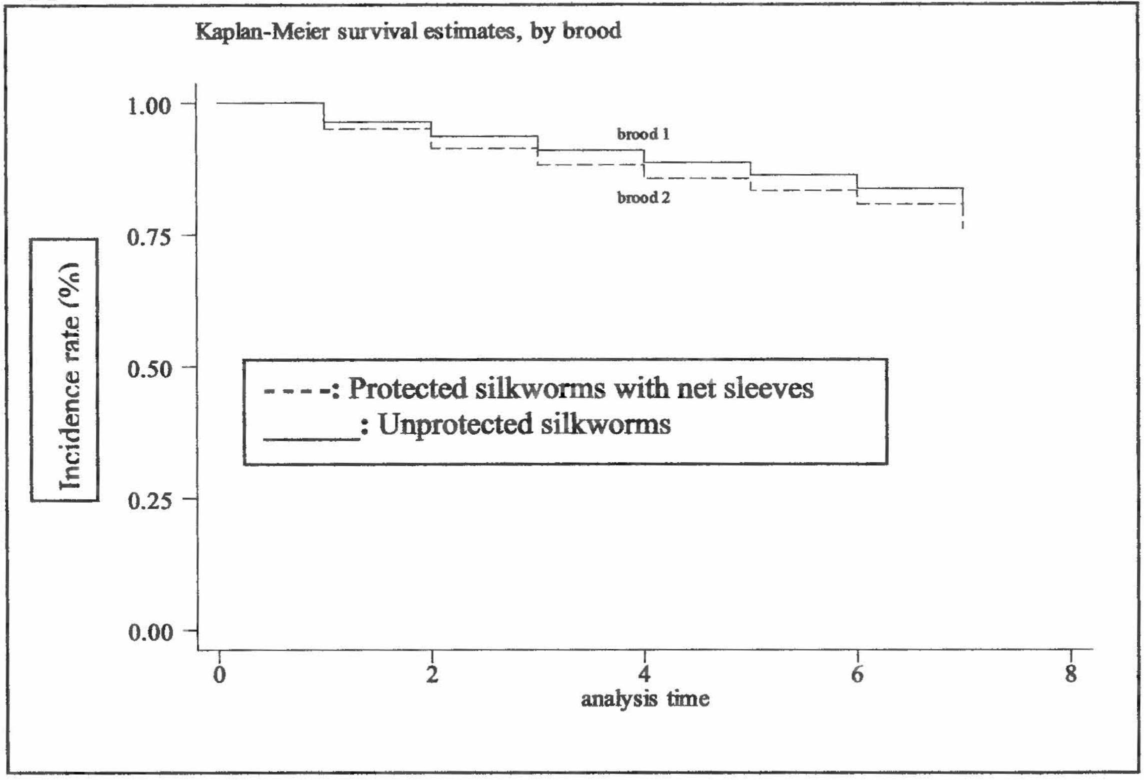
Appendix 9.1: Survivor function of *A. panda* silkworms by habitat in Kakamega



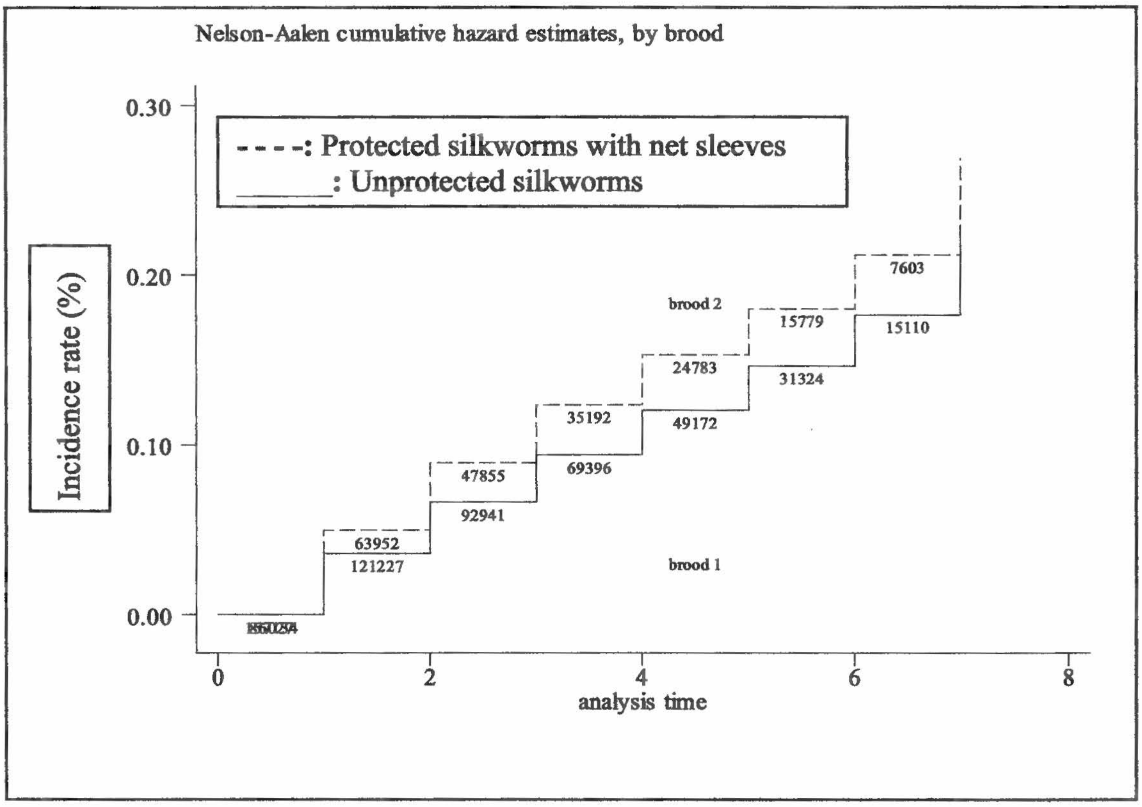
Appendix 9.2: Risk function of *A. panda* silkworms by habitat in Kakamega



Appendix 10.1: Survivor function of *A. panda* silkworms by brood in Kakamega



Appendix 10.2: Risk function of *A. panda* silkworms by brood in Kakamega



Appendix 11: Winding frame reel and serigraph for tenacity and elongation measuremement

AP 11.1: Winding frame/reel

AP 11.2: A serigraph for tenacity and elongation measurement