KFRI Research Report 62

SEARCH FOR NATURAL RESISTANCE TO THE INSECT PEST, *Hyblaea purea* IN TEAK

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PEECHI, THRISSUR

December 1989 Pages: 32

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ABSTRACT

A search was made in Kerala for teak *(Tectona grandis)* clones resistant to attack from the teak defoliator, *Hyblaea puera.* Extensive areas of plantations, natural forests, and three seed orchards representing 31 plus trees were examined during periods of defoliator outbreak. It was found that many isolated trees were left distinctly unattacked amid totally defoliated trees. Detailed investigations revealed that this is not due to genetic resistance but due to, what may be called, phenological resistance. Tender foliage is essential for the initial establishment and survival of the teak defoliator. Phenological resistance is resistance to attack due to asynchrony between the flushing time of the tree and insect population cycles. Early flushers had a greater chance of escape from defoliation but the escape is circumstantial and not consistent over years so that it is of little practical utility. No instance of genetic resistance to the defoliator was discovered in teak clones of Kerala. The search must continue in teak clones of other origin. Evidence was obtained to indicate genetic resistance in the related species, *Tectona hamiitoniana.* The study has also led to standardisation of methods for screening trees for defoliator resistance and development of an artificial diet for rearing the insect in the laboratory.

Key words : Teak defoliator, *Hyblaea puera,* Pest resistance, Phenology, Artificial diet.

1. INTRODUCTION

Insect outbreaks are a regular annual feature in teak plantations throughout Kerala as well as other parts of India. Previous studies at this Institute (Nair *et a/,* 1985) revealed that in 4-9 year old plantations about 44 per cent of the potential volume increment go unrealised because of insect defoliation. Of the two well-known pests - *Hyblaea puera* Cramer, commonly called the teak defoliator and *Eutectona machaeralis* (Walker), commonly called the teak skeletoniser, the former which causes repeated defoliation during the early part of the growth season accounted for most of the damage. The latter insect which generally feeds on older foliage late in the season had no significant impact on growth. It has thus become clear that large benefits can be accomplished by preventing or reducing the damage caused by *H. puera.*

Past attempts to control the defoliators laid stress on silvicultural cum biological methods aimed at augmenting the activity of natural enemies, mainly the parasitoids (Beeson, 1941). The suggested measures included retention of strips of pre-existing natural forests between plantations as a natural enemy reserve and protection of certain plant species within the reserves as well as within the plantations (as an understorey) to sustain the natural enemies. Although we have harped upon this method for a long time, the efficacy of this approach was not tested due to various reasons, and recent research on the infestation pattern (Nair and Sudheendrakumar, 1986) suggested that it may not succeed due to the migrant nature of the pest and the high degree of aggregation of outbreak populations (Nair, 1988). Aerial application of insecticides was also tested by some workers (Basu-Choudhury, 1971; Singh, 1980), but there are several obvious limitations to the use of chemicals in extensive forest areas. Use of resistant trees is an approach that holds promise because it does not involve recurring costs and is environmentally safe. The present project was therefore undertaken to explore this possibility.

Forest tree breeding for insect resistance is still in its infancy world-wide, although many successes have been achieved in breeding agricultural crops resistant to pests. There are several programmes under way in many countries, but no spectacular achievement has so far been reported. In some cases, as in bark beetle damage to pines, no appreciable resistance has been found, but in many tree species evidence has been obtained for insect resistance with a genetic basis (Gerhold, 1966).

Existence of natural resistance in teak to the defoliator has been suggested by many foresters in informal discussions. Presence of unattacked individual trees as well as groups of trees within plantations have been indicated based on their casual field observations as well as our own. We suspected that many of the instances were probably results of escape from insect attack due to one of several reasons like difference in flushing time (generally *Hyblaea* moths show marked preference for tender foliage for egg laying) or abrupt collapse of an expanding pest population due to parasitism or disease. On the other hand, there was some laboratory evidence of difference in susceptibility to the teak skeletoniser, *E. machaeralis* among 8 clones of *Tectona grandis* (FGI, Teli, Kurseong, Barkot + 3, Barkot + 4, Laos, New Guinea and Togi) and one clone of *T. hamiltoniana* (Kedharnath and Singh, 1966).

In the light of the above observations, a critical evaluation of possible "resistance" against *H.* puera and its genetic basis was considered essential. This was attempted by organising a field search for phenotypically resistant trees during natural pest outbreaks and followIng up the lead by subsequent observations and experimentation. Briefly the objectives were to search for phenotypically resistant trees, and (2) to test the heritability of phenotypic resistance, if found.

In addition to research directly relevant to the search for resistance, some invstigations were carried out to gather background information useful for this study. These included (1) study of the flushing habits of teak saplings and (2) development of an artificial diet for laboratory rearing of *H. puera.* The results of these studies are presented in Appendices 1 and 2.

2. METHODS

2.1. Search for resistant trees in plantations and natural forests

To detect resistant trees, it is necessary to make observations during the progress of a defoliator outbreak or within a short interval thereafter as the trees generally put forth a new flush of leaves within about a fortnight.

With about 70,000 ha of teak plantations distributed all over Kerala and the unpredictability of the exact time of occurrence of defoliation, it was not feasible to make direct observations in all plantations or even In represenfafive areas within a short period. Two methods were therefore used to detect resistant trees in plantations and natural forests - (1) questionnaire survey and (2) direct observation by project party in selected areas.

2.1.1. *Questionnaire Survey*

The questionnaire survey was organized with the co-operation of the Forest Department. A reporting form (Table 1) was sent to all Forest Range Officers in Kerala. covering 92 territorial Ranges, with an endorsement letter from the Chief Conservator of Forest (Development).

Table 1 Resistance reporting form

Form for reporting resistance to defoliators in teak

With the collaboration of the Forest Department, the Kerala Forest Research Institute is conducting a search this year to locate teak trees resistant to insect defoliators. Between May and August, during natural incidence of defoliation, please keep an eye for any tree or stand of any age which is 'free of defoliation amidst adjoining insect-attacked trees. If you come across such trees in a plantation or natural forest, please fill in the form below and post it directly to the address given. We will rush to the site, examine the tree to confirm resistance, collect budwood material, raise grafted seedlings and try to breed pest-resistant teak. We shall be grateful for your active participation in this search.

Address to which the report is to be sent	The Entomologist Kerala Forest Research Institute - 680653

1 Reported by (name and designation) :

2 Division

3 Range

4 Location

5 Natural forest or plantations :

6 Single tree or stand

7 Any other relevant information

2.2.1. *Direct Observations*

Direct observations were made by visiting as many plantations and natural forests as possible. Plantations in the areas listed below were observed during the outbreak season from May to July 1983. In each area, roadside plantations were observed while travelling slowly by jeep along the plantations. Where necessary, closer observations were made on foot. The areas covered were the following:

1. *Southern Forest Circle* : Plantations and natural forest along the roadside from Kulathupuzha to near Shenkotah via Thenmala, proceeding to Konni via Achencoil. Extensive strips of teak plantations are located in this area, particularly along Achencoil-Konni, where there is an unbroken stretch of plantations extending over 30 km on either side of the plantation road.

2. *Central Forest Circle* : Plantations covered included those on either side of a 3 km stretch of road at Pothuchady (Peechi Range, Trichur Div.); along the roadside from Thumburmuzhy to Vazhachal, (Vazhachal Div.); at Kaprikatt (Kodanad Range, Malayattur Div.); at Palappilly, (Palappilly Range, Chalakudy Div,); Rama Varrna plantations covering a stretch of about 5.km on either side of the road (Pariyaram Range Chalakudy Div.); and extensive contiguous plantations in Parambikulam Division covering about 20 km along plantation roads. In addition, teak trees growing in a mixed forest under seminatural conditions within the KFRl campus at Peechi were also observed.

3. *Northern Forest Circle:* Extensive plantations in Nilarnbur Division, covering a length of about 30 km along plantation roads.

In general, the three main teak plantation areas in Kerala - Achencoil-Konni, Parambikulam and Nilambur were well covered in these observations.

2.2. *Search for resistant trees in seed orchards*

The Genetics Division of KFRl had earlier established three teak seed orchards in Kerala - at Arippa, near Kulathupuzha in the Southern Forest Circle; at Palappilly, near Trichur in the Central Forest Circle; and at Nilambur in the Northern Forest Circle (Venkatesh *et a/,* 1986). The clones represent plus trees selected from various parts of Kerala. The Nilarnbur orchard was established in 1979 with 15 clones, out of which 13 are from Nilambur and 2, from Konni. The Palappilly orchard, established in 1981 contains 20 clones - 8 from Nilarnbur, 3 from Konni and 9 from Arienkavu. The Arippa orchard, also established in 1981 has 25 clones-12 from Nilambur, 3 from Konni and 10 from Arienkavu. The orchards were raised from grafts planted at 8 m quincuncial design in such a way that no two ramets of the same clone were planted adjacently. In each orchard about 20 plants of each clone fell within our study area.

All the orchards were kept under observation during May, June and July 1983. When defoliation occurred, each tree in the orchard was visually for defoliation intensity and a score asssigned as follows.

Table 2 Defoliation scores

Score	Defoliation level*	mid-point of defoliation %
0	Nil	—
1	< 5%	2.5
2	6-50%	28.0
3	51-95%	73.0
4	96-100%	98.0

*% Leaf loss compared to fully leaved condition

To evaluate the comparative susceptibility of the clones, the mean percentage defoliation for each clone was calculated after converting the scores to defoliation percentage using the rnidvalue af the score class (Table 2). From this the mean defoliation of the plot was calculated. The mean value for each clone was then expressed as a percentage of the plot mean for making comparisons.

2.3. *In situ evaluation of candidate resistant trees*

Fifty six candidate resistant trees selected and marked at Kulathupuzha, Konni, Neriamangalam, Peechi, Parambikulam and Nilambur in 1983 were kept under observation in late 1983 and early 1984 for flushing] variability and incidence of *Hyblaea* infestation. To examine possible variations in the flushing time, the level of flushing of the selected tree and three surrounding trees (selected at random during each observation) were rated visually, into one of six score classes (Table 3). To compare between trees, a flushing index (Fi) was calculated using the formula Fi=Fst-Fnt where, Fst is the Flushing score of the Selected Tree and Fnt is the mean Flushing score of the 3 Neighbouring Trees.

Table 3 Flushing scores

Score	Flushing status
0	No tender leaves present (either leafless or with old leaves)
1	Partial budhreak (i. e. in some branches only), with new leaves less than one-fourth full size
2	Full budbreak, with new leaves less than one-fourth full size
3	Partial flushing, with fully formed tender leaves
4	Full flushing, with fully formed tender leaves
5	Flushing complete, most leaves mature

The intensity of defoliation due to *Hyblaea* was also scored in the selected tree and three neighbouring tree using 5 score classes as given in Table 4 (Unlike in the case of seed orchards, the score represented equal intervals and the scores were used for comparison directly, without converting them to defoliation percentage). To compare between trees, a susceptibility index (Si) was calculated using the formula Si=Sst-Snt where Sst is the Susceptibility score of the Selected Tree and Snt is the mean Susceptibility score of 3 Neighbouring Trees. The negative of Si was taken as Resistance Index (Ri)

Table 4 Scoring of intensity of defoliation

Score	Defoliation level (% leaf loss)
0	Nil
1	1-25
2	26-50
3	51-75
4	76-100

2.4. *Field cage and laboratory evaluation of resistance*

Selected trees showing the greatest promise of resistance were tested for absolute resistance under cage and laboratory condition. In February-April 1985, crown branches were collected from the selected trees, brought to the laboratory and suitable buds were grafted onto 1-2 year old teak stumps as described by Venkatesh *et a/,* (1986). Twelve clones, including some susceptible checks were included in the tests. The graftlings were maintained in polvthene bag containers until December 1985 when they were planted in a randomized block design inside a large screened field cage at Peechi. The floor of the cage was about 6.5 m X 4.5 m and the planting was done in two rows of 3 blocks each, leaving a 2 m wide central path. Each block had one ramet each of 12 clones planted at 1 m *X* 1 m spacing. Thus there were 72 plants in all, 6 of each clone. The plants were watered regularly for about 6 months until the monsoon arrived. In mid-June, about 3 weeks before the release of insects, all leaves except the terminal two pairs were plucked off so that the plants were comparable with respect to the quality and quantity of foliage. Thirty 3rd instar larve (field collected from Nilambur) were released by placing the larvae individually onto the foliage of each plant and follow-up observations were made on larval establishment and feeding.

In one set of laboratory trials, detached leaves of the selected clones were kept in closed plastic containers and 10 third instar larvae were released per leaf. The leaf petiole was wrapped in wet cotton to provide moisture.

For each clone, one of the first and second terminal pair of leaves was used. Eleven clones were tested simultaneously, using two leaves per clone, and 10 larvae per leaf, all the 220 larvae originating from a laboratory culture. Feeding intensity was assessed visually after 24 hours.

In another laboratory trial, a multiple choice test was conducted by presenting all the clones together to a group of larvae. Leaf pieces, 1.5 cm x 1 cm, were cut from a tender leaf of each clone. One end of the leaf piece was placed within a folded strip of filter paper and stapled leaving 1cm of the leaf area exposed, and the filter paper was moistened with water before the larvae were released. Using 11 clones, the 11 strips of leaves were arranged in a circle inside a glass petri dish. Twenty five 3rd instar larvae were released in the centre of the petri dish and the dish covered with a stretched muslin cloth followed by the lid. Three replications of the experiment were set up simultaneously. After 24 hrs the feeding damage was scored visually into 6 classes representing the following percentage of leaf area loss: 0=0%; 1=<24%: 2=26-50%: 3=51=75%; 4=76--99%; 5=100%. Using the midpoints of these score classes, the mean leaf loss in the 3 replicates was calculated.

3. RESULTS AND DISCUSSIONS

31. *Preliminary selection of candidate 'resistant' trees*

3.1.1. *Observation in plantations and natural forests*

Questionnaire survey

In response to the questionnaire (Table I), information was received on occurrence of possibly resistant trees at three places Neriamangalam Range (Munnar Division), Vellikulangara Range (Chalakudy Division) and Nilambur Range (Old Amarambalam section) (Nilambur Division). Reports were also received from several Ranges indicating that no resistant trees were located - these included Mannarappara (Konni), Palappilly, Kothamangalam, Kalady, Thodupuza, Marayur, Sholayar, Aralam (Iritty), Manantody and Tamarassery. The questionnaire did not ask for making any special effort to locate resistant trees and it was not mandatory to return the questionnaire if no resistant tree were sighted. This was done to ensure that only outstanding trees which made their presence felt by conspicuously escaping defoliation were brought to our attention.

We made follow-up observation in the three places mentioned above. At Vellikulangara two trees reported to be resistant, but in our observations they were not distinguishable from the surrounding trees. At Nilambur a patch of trees in the 1946 plantation at Old Amarambalam were reported to be resistant but our observations revealed that during that period infestation in that area was generally patchy indicating that it was a chance escape. At Neriamangalam, observations showed that 3 trees in the 1964 plantation at Valara (19 Year old) had totally escaped defoliation and stood in sharp contrast to the surrounding trees. These trees had fully mature old leaves when observed on 7 July 1983 while other trees had younger leaves representing a new flush which had come up after the first wave of defoliation in June. These 3 trees included in the first round of selection were marked for further follow-up observation in the following year.

Direct observations

Direct observations during the outbreak period in July 1983 revealed that in many areas some trees were left distinctly unattacked amid totally defoliated trees. (Fig. 1). Generally such trees had comparatively older leaves indicating earlier flushing. In addition to these isolated trees, there were fairly large patches of plantation which did not suffer defoliation. It was known from earlier studies (Nair *et a/,* 1985) that during the early phases of population outbreak of *H. puera* the distribution of attack is patchy, the attack spreading to more extensive areas during subsequent waves of infestation. Patches of unattacked trees were therefore ignored and only individual unattacked trees in the midst of attacked trees were selected for further observations. Based on preliminary observations, 56 trees were selected from the different localities as shown in Table 5.

Table 5 : Preliminary selection of potentially 'resistant' trees'

Location	No. of trees selected
Kulathupuzha	3
Konni	11
Neriamangalam	3
Peechi	17
Parambikulam	17
Nilambur	5
Total	56

Among these trees, some had suffered partial attack, but all were conspicuously different from the surrounding trees; all trees which escaped defoliation in a given locality were however, not selected; the selected trees constituted only a sample of such trees.

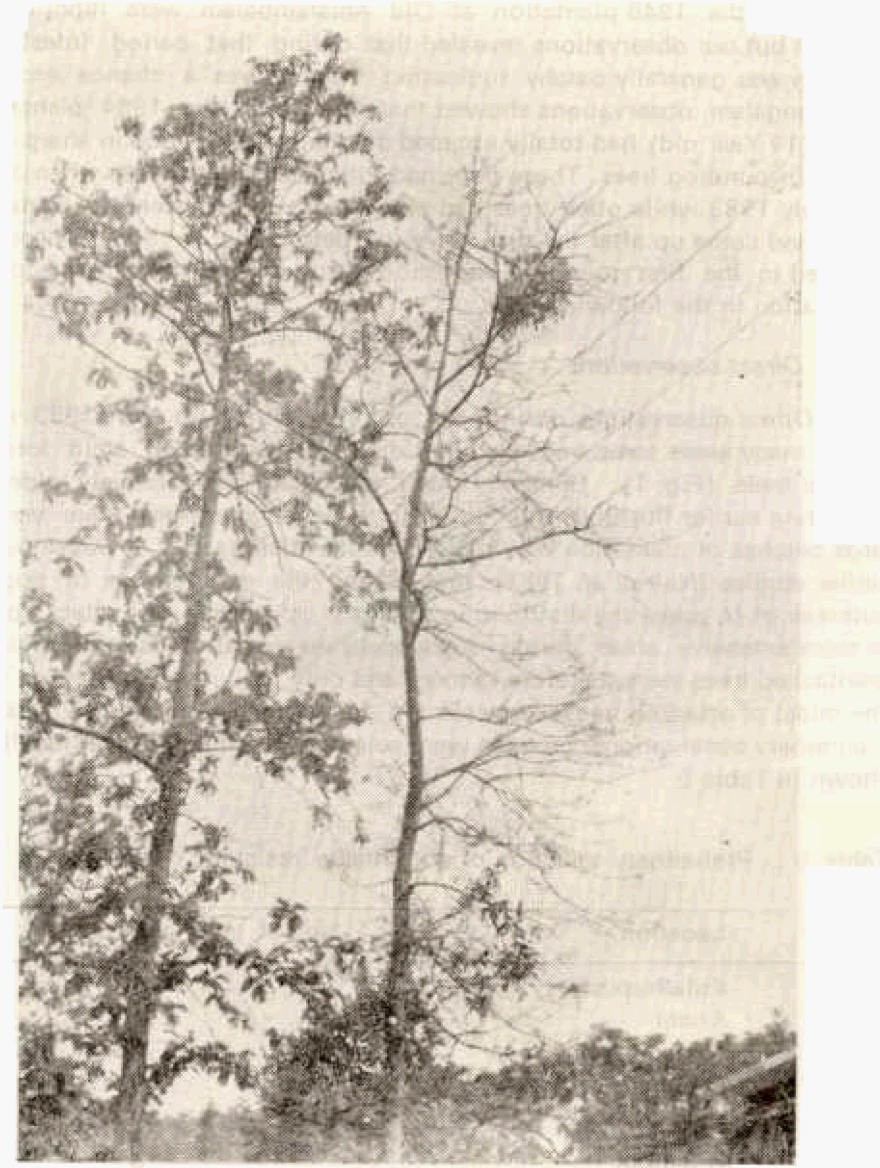

Fig. 1. An unattacked teak tree in the midst of heavily attacked trees.

Although observation did extend to the natural forest, no trees were selected as the 'resistant' status of trees in the natural forest were uncertain because generally the trees occurred in isolation and their escape from defoliation may have been due to chance alone.

3-1.2. Observations in Seed Orchards

Palappilly

Practically no infestation occurred in this orchard in 1983. The plants were fully flushed by 2 July and no defoliation had occurred when observed on 7 June. When examined again in June, a mild infestation had occurred over one corner of the orchard. In view of the general absence of infestation no information on potential resistance was obtained from this orchard.

Nilambur

In this 15-clone orchard, observations were recorded on 13 May and 14 July 1983, following *H.puera* infestation. The defoliation score ranged from 0 to 3 in May and 1 to 3 in July. The comparative susceptibility of the clones is shown in Fig. 2. It may be seen that defoliation ranged from 78 to 133 percent of the plot mean and that none of the 'clones showed worthwhile 'resistance'.

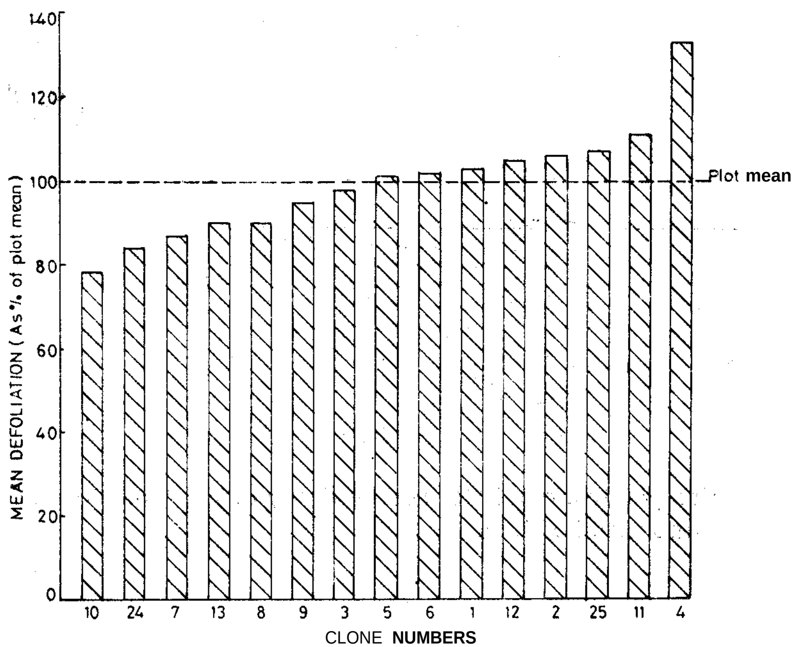

Fig. 2. Comparative defoliator susceptibility of 15 clones in the Nilambur teak seed orchard

Arippa

In this 25-clone orchard, observations were recorded on 27 May and 1 July 1983, following *H. puera* infestation.The trees were in full flush when examined on 10 May. The defoliation score ranged from *0* to 2 in May and 1 to 2 in July which shows that more than 50 percent defoliation did not occur in any tree. Fig. 3 shows the comparative susceptibility of the clones. In general it ranged from 50 to 175 percent of the plot mean. The exceptions were clones 8 and 17 which showed a defoliation score of 21% of the plot mean at one extreme and clone No. 33 which gave a score of 274 percent of the plot mean at the other extreme (Fig. 3). Based on these observations, clones 8 and 17 (both of Nilambur origin) were rated as potentially 'resistant' and were marked for follow up obervations.

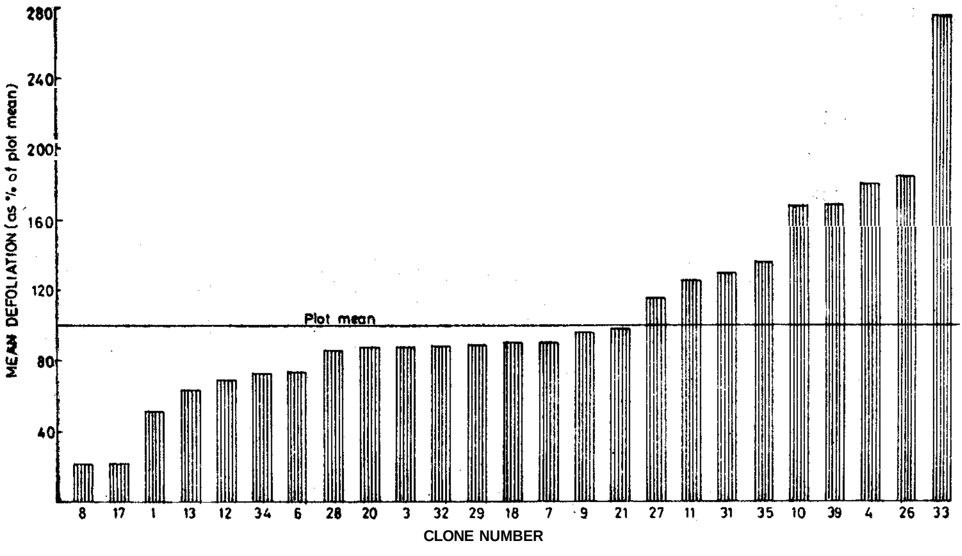

Fig. 3. Comparative defoliator susceptibility of 25 clones in the Arippa seed orchard

Between the Nilambur and Arippa orchards 10 clones were common. A comparison of the susceptibility rating of these clones between the two orchards (Table 6) did not show any consistent trend suggesting that susceptibility may be a circumstantial character.

Table 6 : Susceptibility rating of clones common to Nilambur and Arippa orchards

Clone No.	Mean Defoliation as % of plot mean	
	Nilambur	Arippa
1	52	103
3	85	98
4	171	133
6	73	102
7	91	87
a	21	90
9	96	95
10	167	78 .
11	125	111
12	68	105

3.2 In situ evalution *of* candidate ‘resistant trees'

3.2.1 Trees in plantations

The resistance index (Ri) of the candidate trees is given in Table 7. An index of 4 represents the maximum resistance to defoliation: this score is obtained when the defoliation score of selected tree (Sst) is O (no defoliation) and the mean score of the neighbouring trees is 4 (76-100% leaf loss), so that Sst-Snt=0-4= -4. Therefore Ri=4. An index of —4, on the other hand, denotes high susceptibility. In Table 7 the trees are arranged in decreasing order of resistance except for 10 trees at Parambikulam, 3, at Neriamangalam and 2, at Peechi, for which no index was calculated as there was no incidence of attack in these locations. It may be seen that the resistance index of the candidate trees ranged from 3 to —4; out of 41 trees scored 24 had a positive index, 8 negative and 9 neutral. This shows that although the trees under observation were selected primarily for defoliation resistance in 1983, some of them turned out to be more susceptible to damage than the surrounding trees (Ri<0) and some, as susceptible as them (Ri=0). Explanations for this descrepancy will be discussed later.

A study of the flushing index (Fi) revealed that it ranged from —4.5 to 4.5, the negative values indicating late flushing and positive values early flushing, compared to the neighbouring trees For each tree Fi was available for several observation dates; it was equal to zero during the early part of the growth season before flushing had started as well as later when flusning was complete, and reached a maximum (absolute) at some point of time when the flushing of the selected tree differed most from that of the neighbouring trees, The maximum absolute value of Fi for each tree is given in Table 7 along with the date on which it was recorded. Out of the 56 trees scored, 18 trees had a negative Fi (late flushers), **35** had a positive Fi (early flushers) and **3** had Fi equal to zero. Thus the candiate resistant trees included both late flushers and early flushers. the latter predominating.

Table **7** : Resistance index and flushing index of candidate tre es

Tree No.	Place	Resistance index (Ri)	Observation date for Fi	Flushing index (Fi)*
1	Nilambur	+3.0	Mar 15 84	4.0
2	Nilambur	+3.0	Mar 15 84	4.0
3	Nilambur	+ 3.0	Mar 15 84	4.0
32	Kulathupuzha	+2.0	Feb 1784	3.0
44	Konni	+2.0	Apr 10 84	-3.0
27	Peechi	+2.0	Feb 04 84	3.0
27A	Peechi	+2.0	Mar 19 84	-1.7
30	Peechi	+2.0	Mar 1984	0.5
13	Parambikulam	+1.7	Mar 13 84	0.0
14	Parambikulam	+1.3	Apr 10 84	1.7
12	Parambikulam	+1.0	Apr 10 84	-1.3
34	Kutathupuuha	+1.0	Feb 17 84	4.0
37	Konni	+1.0	Apr 10 84	-4.5
38	Konni	+1.0	Mar 15 84	2.0
41	Konni	+1 .0	Mar 15 84	3.0
20	Peechi	+1.0	Feb 0484	3.0
28	Peechi	+1.0	Feb 04 84	1.0
21	Peechi	+0.7	Feb 04 84	-0.3
36	Peechi	+0.5	Feb 04 84	1
1	Parambikulam	+0.3	Apr 1084	2.7
33	Kulathupuzha	+0.3	Mar 15 84	0.3
35	Konni	+0.3	Mar 15 84	0.7
4	Nilambur	+0.3	Mar 15 84	0.3
5	Nilambur	+0.3	Mar 15 84	0.3
3	Parambikulam	0.0	Apr 10 84	1.3
39	Konni	0.0	Apr 10 84	4.5
40	Konni	0.0	Apr 10 84	-1.5
43	Konni	0.0	Mar 15 84	2.0
45	Konni	0.0	Mar 15 84	0.7
46	Konni	0.0	Mar 15 84	0.7
22	Peehci	0.0	Mar 19 84	-1.0

Tree No.	Place	Resistance index (Ri)	Observation date for Fi	Flushing idex (Fi)*
23	Peechi	0.0	Mar 19 84	-0.3
29	Peechi	0.0	Dec 31 83	-0.7
2	Parambikulam	—0.7	Apr 09 84	-0.3
4	Parambikulam	—1.0	May 03 84	0.5
24	Peechi	—1.0	Mar 19 84	0.2
25	Peechi	-1.0	Mar 19 84	1.0
27B	Peechi	-1.0	Dec 31 83	2.7
33	Peechi	-1.0	Mar 19 84	0.0
42	Konni	—1.7	Apr 10 84	2.0
26	Peechi	—4.0	Feb 04 84	-3.3
5	Parambikulam		May 03 84	0.7.
6	Parambikulam		Apr 10 84	0.7
7	Parambikulam		Apr 10 84	2.3
8	Parambikulam		Apr 11 84	1.3
9	Parambikulam		May 03 a4	0.5
10	Parambikulam		Apr 10 84	-1.3
11	Parambikulam		Apr 10 84	3.3
15	Parambikulam		Apr 10 84	0.7
16	Parambikulam		May 03 84	-1.0
17	Parambikulam		Mar 12 84	0.0
1	Neriamangalam		Oct 10 83	-1.0
2	Neriamangalam		Oct 10 83	-3.0
3	Neriamangalam		Oct 03 83	-3.0
35	Peechi		Feb 04 84	1.0
37A	Peechi		Feb 04 84	3.0

* Positive scores represent early flushers and negative scores late flushers.

It might be expected that trees which flush either early or late in comparison to the surrounding trees may escape defoliation. If it were so, we should expect a strong correlation between the degree of resistance and the absolute value of Fi (i. e, irrespective of whether it is positive or negative). In Fig. 4 the absolute value of Fi are plotted (y-axis) aginst the Ri values from 0 to 3. The linear correlation coefficient (r) between the Fi and Ri worked out to 0.5175, yielding a coefficient of determination (r^2) of 0.2678. This indicates that only about 27 percent of the variation in resistance is explained by phenological variation. A study of Fig. 4 will show that the lack of high correlation between Ri and the absolute value of Fi is due to the large variability of Fi at low

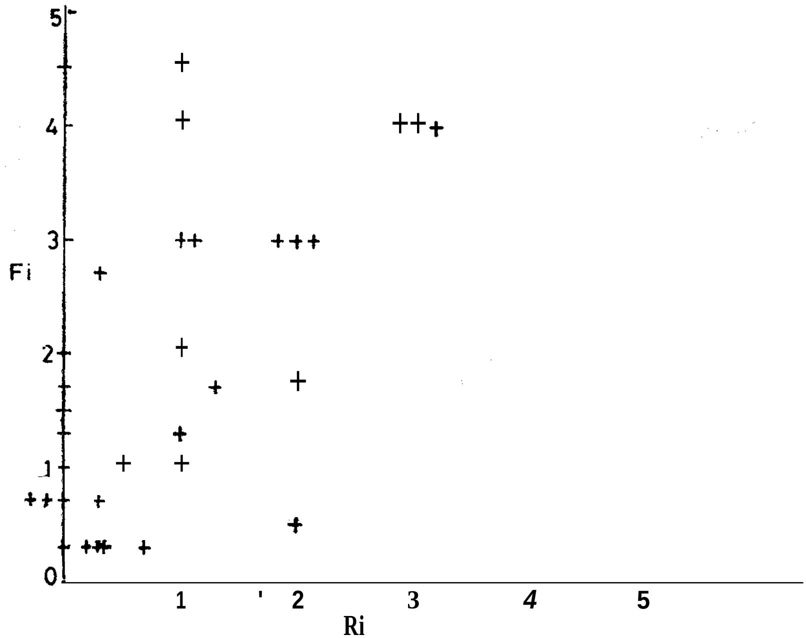

Fig. 4. Scatter diagram showing the relationship between Resistance index (Ri) and Flushing index (Fi) of selected trees.

The major conclusion from these results can be summarised as follows. (1) All trees identified as resistant in 1983 were not found to be resistant in 1984. (2) The more resistant trees included both early flushers and late flushers. although the former predominated; (3) only 27 percent of variation in resistance was explained by variation of phenology. Several explanations of these phenomena are possible at this stage, but we shall postpone the discussion unti additional experimental results are presented.

Based on the evaluation of resistance in 1984 season, 10 trees were selected from among the 56 trees including 3 trees from Nilambur, 1 from Kulathupuzha, 1 from Konni, 3 from Peechi and 2 from Parambikulam (Table 7) which had a resistance index between 1.3 to 3. To accommodate potentially resistant trees for which no resistance index could be arrived at, and to give better area representation, the final selection was modified to include at least one and not more than two trees from each locality. Thus the final selection included the following **10** trees: Tree Nos. 2 and **3** from Nilambur; **32** from Kulathupuzha; 44 and 37 from Konni; 27A and 30 from Peechi; 13 and 9 from Parambikulam and 1 from Neriamangalarn.

3.2.2 *'Teli' variety in Karnataka*

The 'teli' variety of teak in the North Canara District of Karnataka is credited to resist defoliator attack due to early flushing (Kaushik, **1956).** On site observations were made between 19-29 July, 1984 in two 'teli' plantations- a 1928 plantation (75 ha) at Hudsa. in the Gund Range of Haliyal Division and a 1952 plantation at Virampilly in the Virnoli Range of the same Division. The plantations were visited in July after insect outbreak was noticed in the Nilambur plantations in late April to June. However, it was found that no outbreak had occurred in plantations in the Haliyal Division either in the 'teli' or in adjacent normal varieties of teak, until that time. Information gathered from the local staff showed that 'teli' teak is susceptible to *Hyblaea,* but because of early leaf maturity, the damage intensity is less than in normal variety. The 'teli' variety flushes about a month in advance of the normal and is distinguishable by leaf characteristics. Unlike in the normal, the leaves of 'teli' teak have a shiny white undersurface and a smooth, oily ('teli') upper surface. The seeds are also large and of light-weight.

Isolated trees with similar characteristics were also located subsequently in some plantations in Kerala; one such tree in the KFRI Campus at Peechi was used for further testing for resistance.

3.2.3 *Trees in Seed Orchard*

As detailed in section 3.1.2 ,clones 3 and 17 in the Arippa Seed Orchard were also selected for *in situ* evaluation of resistance in 1984. Observations showed that all the saplings in the Arippa orchard were clean-stripped by *Hyblaea* in 1984.

3.3 *Field cage evaluation of selected trees*

Out of the 10 clones selected for further screening, sufficient number of grafts could be raised only for 7 clones; these included Tree Nos. I and 2 from Nilambur, 32 from Kulathupuzha, 44 from Konni, 27A and 30 from Peechi and 9 from Parambikulam. Grafts of two ordinary (susceptible) clones, one each from Nilambur and Peechi, were included for comparison. In addition, an early flushing clone from Peechi, and a clone similar in leaf characteristics to the 'teli' variety of teak as well as a related species, *Tectona hamiltoniana* were included (all grafted at the same time as the selected 'resistant' clones), thus making up a total of 12 clones.

The first screened cage-test was carried out on 4 July 1986 as described in Section 2.4 and the results are presented in Table 8. Although 30 healthy third instar larvae were released per seedling, very few survived. A large number of larvae were found dead; most are believed to have died due to infection by a nuclear polyhedrosis virus (Sudheendrakumar *et a/,* 1988) although some instances of parasitisation were also noted. The larvae, it may be recalled, were field-collected from infested trees at Nilambur. Very few live larvae were present on the 3rd day of release (Table 8). It may be seen, however, that initial establishment of larvae occurred on all the tested clones with a mean number of 2 to 5 larvae per plant. Since heavy mortality occurred subsequently, the leaf damage was minimal and was not measured. However, the data on initial establishment suggest that there was no difference in susceptibility (i. e. acceptance) between clones.

Table 8: Establishment of artificially released 3rd instar larvae on selected teak clones inside screened cage.

Serial No. of clone	Place of origin and code No.	*Mean No. larvae/plant on 2nd day of release	No. of uninfested plants out of 6	Mean No. of larvae/plant on 3rd day of release
1	Parambikulam, HR 9	5 5 ± 1.5	0	0.5
2	Kulathupuzha, HR 32	3 7 ± 2.4	1	0.2
3	Konni, HR 44	2.0 ± 2.2	2	0.3
4	Nilambur, HR 1	3.2 ± 2.2	1	0.5
5	Nilambur, HR 2	4.3 ± 4.3	1	1.8
6	Peechi, HR 27A	3.5 ± 2.2	0	0.8
7	Peechi, HR 30	3.2 ± 2.6	1	0.7
8	Peechi, HS 27B	2.8 ± 2.3	1	0
9	Nilambur, HS 3	3.3 ± 3.7	4	0.3
10	Introduced ***T.*** *hamiltoniana*	3.0 ± 2.6	2	0.2
11	Peechi, Teli-like, 27C	3.7 ± 3.2	1	0.3
12	Peechi, E. F. 30B	3.5 ± 2.6	0	0.7

Mean and standard deviation of 6 replicates; 30 third instar larvae were released on each plant on 4th July 1986.

The experiment was repeated on 17 June 1987, this time using larvae collected from a local field infestation at Peechi. Unfortunately, incidence of disease repeated itself and no useful data could be gathered.

Further testing was therefore carried out under laboratory conditions using laboratory reared larvae.

3.4 *Laboratory screening of selected clones*

Table 9 shows the results of feeding test on detached leaves of selected clones (see section 2.4 for methods). One susceptible clone (Peechi HS 278) was not included in these tests as leaves of comparable age and texture were not available The results show that feeding occurred on all clones except in *7'. hamiltoniana.* While in most clones about half of the medium-sized leaves were consumed by the 10 larvae, in *T. hamiltoniana* feeding was practically nil, although some nibbling was made. In addition, except for one attempted folding of a cut edge of the leaf blade on *T. hamiltoniana* the larvae did not construct the characteristic feeding shelters.

Table 10 shows the results of the multiple choice feeding test on leaf pieces of the **11** clones (see section 2.4 for methods). No feeding occurred on *T. hamiltoniana,* confirming the results of the whole leaf test. Clone No.2 (Kulathupuzha HR 32) showed the next least damage, followed by clone numbers 3, 11, 12 and 9.

Table 9: Feeding intensity on detached leaves of selected teak clones by *Hyblaea* larvae, in laboratory trial.

Clone No. & Place		Intensity of feeding (as indicated by extent of leaf area lost)*	
		1st pair of leaf	2nd pair of leaf
1	Parambikulam, HR 9	+ +	+ +
2	Kulathupuzha, HR 32	+ +	+ +
3	Konni, HR 44	+ + +	+ + +
4	Nilambur, HR 1	+ +	+ + +
5	Nilambur, HR 2	+ +	+ + +
6	Peechi, HR 27A	+ + + +	+ + +
7	Peechi, HR 30	+ + +	+ + +
9	Nilambur HS 3	+ + +	+ + +
10	*Tectona hamiltoniana*	Nil	Very little
11	Peechi, Teli 27C	+ +	+ + + +
12	Peechi. EF 30b	+ + +	+ + +

Ten 3rd insiar larvae were released per leaf and the feeding intensity was rated visually. Each + sign indicates consumption of one fourth portion of the leaf.

Table 10: Feeding preferences of 3rd instar *Hyblaea* larvae on leaf pieces of selected clones in multiple choice test

Clone No. & Place		Feeding score in three replicates			Mean % leaf loss due to feeding
		1	2	3	
1	Parambikulam, HR 9	3	0	1	25.3
2	Kulathupuzha, HR 32	0	1	0	4.2
3	Konni, HR 44	1	1	1	12.5
4	Nilambur, HR 1	6	1	1	41.5
5	Nilambur, HR 2	1	4	1	37.8
6	Peechi, HR27A	1	2	1	21.2
7	Peechi, HR30	1	2	2	29.8
9	Nilambur HS 3	1	0	2	17.0
10	*Tectona hamiltoniana*	0	0	0	0
11	Peechi, Teli 27C	1	1	1	12.5
12	Peechi, EF 30B	1	1	1	12.5

Attempts were made to use leaves of comparable age and texture for these experiments, but the possible influences of subtle differences due to leaf age are unknown. Since at least some feeding occurred on all leaves except that of *T. hamiltoniana,* it is safe to conclude that none of the selected clones have absolute non-preference type of resistance against *Hyblaea* and that *T. hamiltoniana* is the least preferred.

4. GENERAL DISCUSSION AND CONCLUSIONS

The present study has shown, as earlier casual observations has suggested that isolate trees or groups of trees often escape attack of the teak defoliator. Detailed investigations of this phenomenon have revealed that this is not due to genetic resistance but due to what may be called 'phenological resistance'. Under normal conditions, the moths lay eggs only on tender leaves and the newly hatched larvae survive only if such leaves are available for initial establishment and feeding. Phenological resistance is resistance to attack due to asynchrony between the flushing time and insect population cycles.

That the observed resistance is not a genetic character is shown by the following observations - (1) graftling prepared from 10 trees that had escaped defoliation under natural conditions were readily attacked when artificially exposed to the insect; (2) Many trees which had escaped defoliation in the first round of selection for resistance, were found attacked in the second year under natural conditions.

The characteristics of phenological resistance, the conditions under which it manifests itself and its practical usefulness must now be considered. One of the chief characteristics of phenological resistance brought out by this study, is its nonrepeatability, in other words, its variability between years. Not all trees which were rated resistant in 1983, and for which rating could be made again in 1984 (Table 7). only about 17 repeated the performance and an equal number were either more susceptible than the surrounding trees or similar to them. As noted earlier, the 'resistant' trees included both early flushers and late flushers, although the former predominated. Phenological difference explained only about 27% of the variation in resistance. Based on the insight gained during this study as well as on knowledge of the behaviour and population dynamics of the teak defoliator obtained from other investigations (Nair *et a/,* 1985, and unpublished observations) the following explanations are offered to account for the observed phenomena.

Any given population of trees exhibit considerable variation in flushing time. During the early part of the growth season, there is a period of time when trees have putforth a new flush of leaves while other trees are still in various stages of leaf shedding. As the season advances, more and more trees begin to flush. During this period (generally March to late April at Nilambur) the teak defoliator is generally not prevalent, and due to reasons not fully understood yet, the insect builds up suddenly, generally from late April to late May (at Nilambur). When the outbreak occurs, trees which possess predominantly tender leaves at that time are attacked and heavily defoliated while trees which hold only mature leaves are left unattacked or scarcely attacked and stand out conspicuously as "resistant" trees. To illustrate, consider three trees, A, B, and C which begin to flush in the first week of March, April and May, respectively. If the insect outbreak occurs in the last week of May, tree *C* which is full of tender foliage will be heavily attacked. Tree B which has a good proportion of tender leaves at this time will also suffer damage, either due to primary attack or due to secondary attack by middle-aged larvae dispersing from adjacent 'C' trees (see Nair and Sudheendrakumar. 1986), Tree A, which possesses only mature leaves will however, be left unattacked. If another wave of attack follows two or three weeks later trees B and C which by now had reflushed after the defoliation will be attacked again but more distinctly, tree A will again escape and stand out to as 'resistant'. However, if in another year the first population outbreak occurs in the last week of March, tree A will suffer defoliation. If a second wave of attack occurs again after two weeks trees A and B will suffer defoliation. Tree C which flushes later may escape defoliation as population outbreaks usually subside after one or two peaks due to build up of natural enemies. Thus, depending upon the time of insect population outbreaks differents sets of trees - either early, mid or late flushers may escape defoliation in different years. Thus the releationship between flushing time and resistance to attack will not be consistent. The occurrence of repeated outbreaks, at unpredictable times further complicates the relationship. During the early phase of outbreak development, when the insect populations are small and highly aggregated, large patches of plantations may also escape infestation simply by chance, although they are likely to get infested subsequently. Much of the phenological variation may be genetical, but edaphic factors may also influence phenology and lead to a mosaic of attacked and unattacked patches within a larger area.

This kind of resistance which is circumstantial and dependent on non-genetic factors has been termed "pseudoresistance" (Painter, 1951). It may also be called "ecological resistance" (Panda, 1979). Similar resistance due to preference of insects to fresh tender leaves and variation in the flushing time between clones has also been noted in some other forest tree species, eg. in spruce against the nunmoth, *Lymantria monacha,* in pine against in pine beautymoth, *Padolis flammea,* in oak against oak-roller moth, *Tortrix viridana,* all in Europe (Viviani, 1966; Schonborn, 1966).

Although attempts have been made to select and propagate early flushing clones as a means of protection against insect in all the above examples, there is the risk of the insect adapting to the altered phenology of the host in course of time. In the case of teak, although the early flushing trees had a greater chance of escape, they did suffer damage at times, and we must await more information on the temporal dynamics of teak defoliation, especially the chain of event leading to sudden outbreak during the earlier part of the growth season before we can make a judgement on the practical usefulness of phenological resistance. If the development of outbreak is dependent on pre-monsoon wind systems which concentrate and transport the moths positively over long distances as some observations suggest, there is scope for using early flushing clones to obtain practical protection against attack.

We must now ask whether the search we have made is sufficient *to* conclude that there exists no genetic resistance against *H. puera* in teak. Our search was mostly confined to seed orchards and plantations in Kerala. The search in natural forests has not been exhaustive because of the difficulty in distinguishing resistance from chance escape, under natural forest conditions. However, we can conclude that in teak of Kerala origin the chances of finding clones that are genetically resistant to *H. puera* is remote. The search must continue in teak of other origin from India as well as from other countries such as Indonesia and Burma. Heritable, within-species resistance against pests has been found at least in some forest tree species, eg. in chestnut varieties against chestnut gall wasp, and in Jaffrey-Coulter pine hybrids against pine reproduction weevil, and in a review of the current basic knowledge on the subject Gerhold (1966) commented that it can be discovered in almost any species, if only, the search is diligent enough. The present study has yielded valuable information on the commonly observed "resistance", which can be used profitably in planning the approach to resistance screening in teak. Since large efforts are needed for conducting such a search, we think it is best to link it to a broader tree improvement programme for teak. Preliminary screening can be carried out most profitably where a live clone bank representing a wide variety of genotypes is available, by artificially releasing the larvae on graftlings.

Evidence obtained in this study shows that the closely related species, *Tectona harniltoniana* is genetically resistant to teak defoliator attack. Although some larvae established themselves initially when placed on *T. hamiltoniana* leaf in screened cage experiment, little feeding was noticed in laboratory experiments. Observations made on a few trees in the Peechi and Nilambur campuses also showed that although initial establishment of some larvae occurred on these during periods of outbreaks, the damage caused was negligible and few insects, if any, completed development, suggesting an antibiosis type of genetic resistance (Painter, 1951). As the wood quality of this species is poor, it cannot replace *T. grandis.* But the possibility of interspecific hybridisation to transfer the resistance of *T. hamiltoniana* to *T. grandis* without secrificing the wood quality and productivity of the latter needs to be explored.

In conclusion, this study has shown that the commonly observed escape of some teak trees from defoliator attack is not due to true resistance, but due to asynchrony between their flushing time and the insect population cycles. Such escape is circumstantial and inconsistent over years. No instance of heritable defoliator resistance was discovered in teak clones from Kerala.

REFERENCES CITED

Basu-Choudhury, T. C. (1971). Interim report on the Konny aerial spraying project. Appendix *in:* K. G. Vasudevan (1971). *Working Plan for Nilambur Forest Division 1967-68 to 1976-77,* Govt. of Kerala, 236p.

Beeson, C. F.C. (1941). *The ecology and control of the forest insects of India and the neighbouring countries.* (1961 Reprint) New Delhi, Govt. of India, 767p.

Gerhold, H.D. (1966). In quest of insect-resistant forest trees. pp. 305-318 *in: Breeding Pest-Resistant Trees* (Gerhold *et al.* eds) Pergamon Press, Oxford.

Kaushik, R. C. (1956). Teli variety of *Tectona grandis. Proc. 9th Silv. Conf..* Forest Research Institute, Dehra Dun. Part 2 : 92-94.

Kedharnath. S. and Singh, P. (1975). Studies on natural variation in susceptibility of *Tectona* to leaf skeletonizer, *Pyrausta machaeralis* (Lepidoptera: Pyralidae). FAO/IUFRO Symp. on Forest Diseases & Insects Delhl, April 1975. (Cyclostyled).

Nair, K. S. S. (1988). Pest management in Indian Forestry: How to bridge the gap between theory and practice. *Proc. National Symp. on IPC-Progress & Perspectives,* Trivandrum, Oct. 1987 : 32-34.

Nair, K. S. S. and Sudneendrakumar. V. V. (1986). The teak defoliator, *Hyb/aea puera:* Defoliation dynamics and evidence for short-range migration of moths. *Proc Indian Acad. Sci.* (Anim. Sci.) 95 (1): 7-21.

Nair, K.S. S., Sudheendrakumar, V. V.Varma, R. V. and Chacko, K. C. (1985). Studies on the seasonal incidence of defoliators and the effect of defoliation on volume increment of teak. *KFRI Research Report* No. 30, Kerala Forest Research institute, 78p.

Painter, R. H. (1951). *Insect Resistance in Crop Plants.* Macmillan, New York, 520p.

Panda, N. (1979). *Principles of host Plant Resistance to Insect Pests.* Hindustan Publishing Company, Delhi, 386p.

Schonborn, A. (1966). The breeding of insect - resistant forest trees in Central and Northwestern Europe. pp. 25-27 *in: Breeding Pest Resistant Trees* (Gerhold *et a/,* eds.), Pergamon Press, Oxford.

Singh, P. (1980). Aerial spraying of chemicals to control teak defoliators *Proc. 2nd Forestry Conf.,* Dehra Dun, India, 9p. (Cyclostyled.)

Sudheendrakumar, V. V., Mohamed Ali M.I. and Varma R. V. (1988). Nuclear polyhedrosis virus of the teak defoliator *Hyblaea puera. J. Invertebr. Pathol.* 51(3)*:* 307-308.

Venkatesh, C. S., Koshy, M. P.. Chacko, K. C. and Indira, E. P. (1986). Genetic improvement of teak *(Tectona grandis* L. f.) in Kerala. *KFRI Research Report* No. 13, Kerala Forest Research Institute, 21p.

Viviani, W. (1966). Selection of forest trees for insect resistance in Southern Europe. *in: Breeding Pest Resistant Trees* Gerhold *et a/.* eds.). Pergamon Press, Oxford.19 -23

APPENDIX I

Flushing habits of teak saplings

In our attempts to rear *H. puera* in the laboratory to standardise methods for testing for resistance, nonavailability of sufficient quantities of tender teak leaves was often a handicap. It was noticed that during the period October-March tender leaves were seldom present on trees, but saplings and coppice growth produced new leaves throughout the year. Teak saplings were therefore raised within the campus to obtain a continuous supply of tender leaves. This opportunity was utilised to study the effect of watering on the production of new leaves, and to record data on leaf growth parameters.

Materials and methods

The study was carried out at Nilambur during the year 1983-84. The saplings were raised in an open area in ground-level beds. The beds were prepared by digging trenches 60 cm wide and 60 cm. deep and refilling thern with the excavated soil mixed with sand and dried-powdered cow-dung in the ratio 7:2:1. Three clones of teak collected from 3 different localities in Nilambur and grafted on root - shoot stumps were used. There were 6 rows of plants, each row consisting of 15 plants (5 each of the 3 clones). The spacing was 2 m between plants and 3 m between rows. The grafts were about 3 months old when planted out on 26 December 1983 and the experiment started about 45 days later. Two rows were used for the experiment – one row of plants was watered daily during the experimental period and the other row left without watering. Fortnightly observations were taken on the total number of leaves per plant as well as the number of tender leaves. Leaves which retained a soft texture as judged by feeling with hand were treated as tender. To study the growth of leaf, batches of new leaves from plants in the untreated rows were tagged and observed until they were shed. The length from the base of the petiole to the tip of leaf was measured, at intervals, until full growth was attained.

Results and discussion

The total number of leaves per seedling and the number of tender leaves, in the two groups – watered and control, during the 2-month period from 9 February to April are shown in Fig. 1.1. Watering increased the total number of leaves per sapling and maintained it more or less constantly throughout the observation period. The number of tender leaves was also consistently higher in watered plants.

The progress of growth of new leaves is shown in Table 1.1. Starting between mid-May and Mid-June the growth of three batches of newly developing leaves were followed.

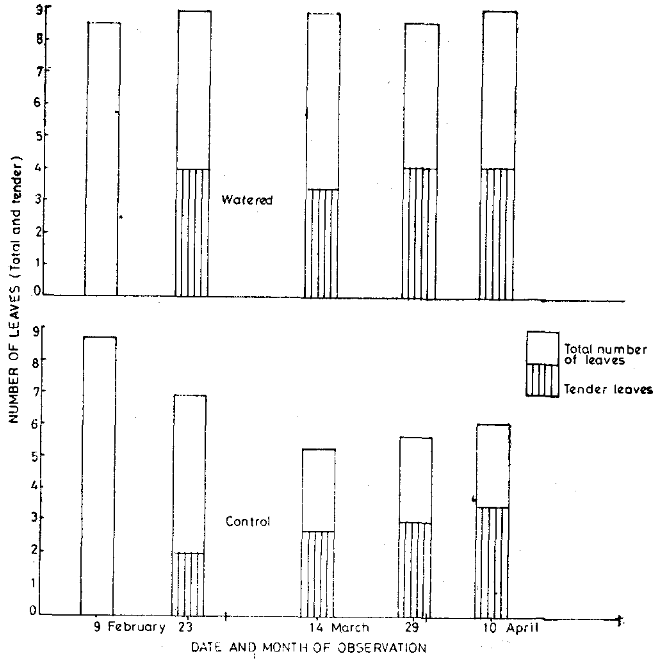

Fig. I. 1. Leaf growth in teak saplings. Mean of 15 saplings representing 3 clones; no record of tender leaves during the first observation on 9 February.

Table I. 1: *Progress of growth of new leaves in teak saplings*

Date of 1st observation	No. of saplings in the batch	Leaf length (cm) at specified intervals (mean and SD)					
		1st day	8th day	15th day	18th day	26th day	34th day
15 May	5	1.5 ± 0.5	4.1 ± 1.2	—	—	42.8 ± 0.7	47.3 ± 2.7
23 May	8	2.0 ± 0.6	—	—	43.8 ± 4.6	44.6 ± 4.7	—
11 June	13	1.5 ± 0.5	5.6 ± 2.5	25.0 ± 9.9	—	52.4 ± 4.6	—

The period taken for full development (from outbreak growth to loss of tenderness) ranged from 26 to 34 days, showing a mean of about 30 days. The length of the mature leaf ranged from 38 to 61cm, with a mean of about48 cm. The 'eaves under observation were shed during October-November. Thus the life span of the leaf is estimated at about 5 months.

Conclusions

The conclusions from this study are:

1. Watering increases the production of leaves in saplings
2. The normal development period of a teak leaf is about a month and the normal life span about 5 months.

APPENDIX II

Laboratory rearing of Hyblaea puera and development of an artificial diet

As a part of this project attempts were made to culture *H. puera* in the laboratory to ensure availability of sufficient larvae for conducting various experiments on evaluation of resistance. The major' handicap while rearing the larva? on teak leaves was the high rate of mortality due to bacterial and viral diseses. In addition, maintaining the larvae on leaves also demand considerable manpower and time. Attempts were therefore made to standardise an artificial diet using cheap and easily available ingredients.

Materials and methods

Test diets

The test diets were based on that developed by Nagarkatti and Prakash (1974) for rearing *Heliothis armigera* (Hub) (Lepidoptera: Noctuidae) Necessary modifications were made based on repeated trials to suit the requirements of *H. puera.* The comoosition of two diets D**1** and D 2 developed after priliminary trials and evaluated in this study are given in Table II.1

To prepare the teak leaf powder, fully expanded first and second pair of terminal leaves were collected from flushing branches and dried in a hot air oven at 60°C for 12 hrs. After removing the major veins the leaves were powdered in a blender. The fine dust obtained after seiving out the coarser particles was used All the other ingredients were commercially available.

Diet preparation

The ingredients of the diets are given in Table II.1. Agar was added to half the required quantity of distilled water in a beaker, stirred with a glass rod and brought to boil. The beaker was then removed and kept immersed in hot water to prevent solidification of agar.

Kabuligram, leaf powder, yeast extract, sorbic acid, casein/casein hydrolysate, and methyl-para-hydroxybenzoate were added to the other half of the distilled water in a blender jar and blended. To this was added the multivitamin mineral mixture, ascorbic acid, streptomycin and formalin and the mixture was further blended.

Agar solution cooled to 60°C was then slowly poured to the above mixture and blended.

The diet was poured while hot into sterilised glass tubes (7.5cm x 2 cm) up to about one third of the tube. This quantity of diet was sufficient for complete development of a larva. One litre of the diet was sufficient to prepare about 100 such tubes. The tubes were then covered with paper and allowed to cool at room temperature.

Test insects

To collect eggs, mated female moths were released into 17 cm x 7 cm glass bottles and covered with muslin cloth. Diluted honey was provided as food on cotton swabs. Eggs were mostly laid on the muslin cloth which made collection of eggs easy. As a measure to prevent mortality due to diseases, eggs were surface sterilized by dipping the muslin cloth containing the eggs in a 1% solution of sodium hypochlorite for 15 min. The cloth was then dipped in distilled water for 10 min with 2-3 changes and then kept over a blotting paper for draining off water. It was then placed in a glass bottle for hatching of eggs. The newly emerged larvae were allowed to feed on fresh teak leaves sprayed with a 200 ppm solution of streptomycin sulphate for 1-2 days before transferring to the diet.

Transfer of larvae

Larvae initially established on tender teak leaves were transferred to the diet when they were 3-4 days old (Second instar). Using a fine camel hair brush a single larva was transferred to each tube containing the diet. The tubes were then closed with cotton plugs. A set of 75 tubes were maintained for each diet. An equal number of larvae maintained on teak leaves formed the control. For this, five larvae each were maintained in 17cm x 7cm glass bottles. The bottles were changed once in two days and fresh teak leaves sprayed with antibiotic solution were provided every day. The rearing was carried out at room temperature (24-26°C and RH 81-92%) during August-September.

The pupae were removed from the rearing containers within 24 hrs of pupation and transferred to clean bottles for emergence.

The parameters measured in the study to evaluate the suitability of the diets were (1) larval period (2) pupal period (3) percent pupation, (4) pupal weight and (5) percent emergence of normal adults.

The data were analysed statistically using analysis of variance and mean comparison test (Snedecor and Cochran, 1967).

Results

The performance of the two diets D1 and D 2 (Table II.1) in comparison to teak leaf with respect to the parameters studied is shown in Table 11.2.

Table II 1. Composition of artificial diets evaluated for rearing *H. puera*

	Ingredients	Quantity of ingredients	
		Diet 1	Diet 2
1	Agar[1]	20 g	20 g
2	Kabuligram flour (*Cicer arietnurn)*	100 g	100 g
3	Casein (purified)[1]	30 g	—
4	Casein hydrolysate[1]	—	3 g
5	Yeast extract powder[1]	10 g	10 g
6	Teak leaf powder	20 g	20 9
7	Multivitamin and mineral mixture[2]	2 caps	2 caps
8	Vitamin E[3]	400 mg	400 mg
9	Ascorbic acid[4]	3.5 g	3.5 g
10	Sorbic acid	1 g	l g
11	Mythyl-para-hydroxybenzoate[5]	1.5 g	1.5 g
12	Streptomycin sulphate[6]	0.25 g	0.25 g
13	Formaldehyde 10%	2 rnl	2 ml
14	Distilled water	1000 ml	1000 ml

1- Supplied by Hymedia Ltd.;

2 - Becadexamine' capsule by Glaxo Ltd. containing Vitamin A 5000 IU, Vit D_3 400 IU, Vit. E 15mg. Vit. B_1 5 mg, Vit. B_2 5 mg, Vit. B_6 2 mg. Vit.B_{12} 5 mg, Vit. C 75 mg. Nicotinamide 45 mg, D-Panthenon 5 mg, Folic acid 100 mg. Ferrous Fumerate 50 mg, Dibasic calcium phosphate 70mg, Copper sulphate 0.1 mg, Dibasic calcium phosphate 70mg. Copper sulphate 0.1 mg. Manganese sulphate 0.01 mg, Potassium iodide 0.025mg. Manganese oxide 0.15mg. and Zinc sulphate 50mg.;

3 - Evion by Merck (2 caps);

4 - Supplied by Polypharm;

5 - Supplied by Romali;

6 - Ambystryn by Sarabhai Chemicals

Table II 2. Comparative performance of two artificial diets and teak leaf (control) evaluated for rearing *H. puera*

Treatment	Larval period (days)		Pupal period (days)		Total development period		Percent pupation	Pupal weight (mg)		% emergence out of pupae	% emergence out of initial no of larvae
	Mean	SD	Mean	SD	Mean	SD		Mean	SD		
Control (Leaf)	11.9^{a}	± 0.86	7.3^{a}	± 0.59	$19\ 3^{a}$	± 1.08	27^{a}	180.5^{b}	+ 34.99	59.3^{a}	
Diet-D1	13.5^{b}	± 0.98	6.7^{b}	± 0.72	20.1^{a}	± 1.25	56^{b}	223.4^{a}	± 43.42	78^{b}	44^{b}
Diet-D2	14.3^{c}	± 0.76	7.3^{a}	± 0.65	21.6^{b}	± 1.08	44^{ab}	207^{a}	± 27.6	33^{a}	16^{a}

Figures superscribed by the same letters in each column are not significantly different at 5% level of significance.

Survival in terms of percent emergence of the moth was highest (44%) in diet D 1. Even on teak leaf, the natural food, only 18% of the larvae survived to the adult stage, although 27% had pupated. This was possibly due to wandering off of larvae from the leaf. Cut teak leaf has a tendency to loose turgidity quickly even when the petiole is wrapped in moist cotton. In addition some larvae died due to bacterial or viral infection. On diet D2 although 44% pupated a larger percentage died as pupae. On both the diets the total developmental period was slightly higher than on leaf, but for diet D1 this difference was not significant. The insects reared on both the diets showed higher pupal weight than those reared on leaf. A comparison of all the parameters studied among the two diets and teak leaf (Table II. 2) will show that diet D1 was equal or better than teak leaf with respect to total developmental period, percent survival and weight of the insect. A slightly longer developmental period although not statistically significant was compensated by a significantly higher pupal weight. Between the two diets, those reared on D 2 had a significantly longer developmental period and significantly lower survival rate.

The general trend obtained in this experiment was noticed in several subsequent rearings on diet D1 showing that it is a satisfactory artificial diet. Several continuous generations were maintained on diet D 1 in the laboratory and no malformation was noticed.

Discussion

The basic composition of the diet used in our experiment was similar to that developed by Nagarkatti and Prakash (1974) for *Heliothis armigera* which in turn was similar to the diet developed by Dang et al, (1970) for *Chilo zonellus.* The alterations we made consisted of addition of casein and teak leaf powder; replacement of yeast with yeast extract, as well as changes in the quantity of various ingredients. These changes were made on an empirical basis and no attempt was made to arrive at the optimal composition or optimal relative concentration of the ingredients. We made a series of diets using various combinations of the ingredients before arriving at the two combinations used in diet D 1 and D 2. Apparently addition of teak leaf powder increased the acceptability of the diet to the larvae by arresting their tendency to wander away from the diet. Neither dried teak bark powder nor teak saw dust gave this effect.

The only difference between diet D1 and D2 was use of casein in diet D1 (30g) and casein hydrolysate (3g) in diet D2. Only a small quantity of casein hydrolysate was used since kabuligram powder was present as the chief protein source. The result showed that presence of hydrolysed protein in the form of casein hydrolysate was disadvantageous.

The diet D1 developed in this study was suitable for rearing *H. puera* for several repeated generations, but the composition was empirical. Apparantly there is scope for simplifying the diet further by eliminating inessential components. The Present diet may be taken as the base diet and improvement attempted. General observations showed that storing the diet for a period of 2 to 3 months in a refrigerator had no deleterious effect on survival of the insects.

It must be noted that we used 3 - 4 days old larvae that were initially fed on teak leaves. Freshly hatched larvae often failed to establish in the diet, but the reason remains unknown. It appears that the texture of the diet is important in initial larval establishment. The newly hatched larvae established more readily when the surface of the diet was scratched with a sterile needle. Further trials are needed to arrive at an optimal diet cornposition and to standardise the physical conditions of the diet to facilitate the establishment and survival of newly hatched larvae. In spite of the precautions taken some larvae died due to bacterial or viral infection, in our experiments. Much of this infection must have occurred before the larvae were placed on the diet, This can be prevented if conditions are standardised for direct transfer of newly hatched larvae on to the diet.