INHERITANCE AND ALLELIC RELATIONSHIP OF GENES CONTROLLING RESISTANCE TO GALL MIDGE (*Orseolia oryzae* Wood Mason) IN SOME RICE CULTIVARS

THESIS

Submitted to the

Indira Gandhi Krishi Vishwavidyalaya, Raipur

in partial fulfilment of the requirements

For the Degree of

DOCTOR OF PHILOSOPHY

IN

AGRICULTURE

(PLANT BREEDING AND GENETICS)

By

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DEPARTMENT OF PLANT BREEDING AND GENETICS
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1998

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RAIPUR, 492 012 (M. P.)

Dated: 24.2.98

CERTIFICATE - I

This is to certify that the thesis entitled **"INHERITANCE AND ALLELIC RELATIONSHIP OF GENES CONTROLLING RESISTANCE TO GALL MIDGE (*Orseolia oryzae* Wood Mason) IN SOME RICE CULTIVARS "** submitted in partial fulfilment of the requirements for the degree of **DOCTOR OF PHILOSOPHY IN AGRICULTURE (Plant Breeding and Genetics)** of the Indira Gandhi Krishi Vishwavidyalaya, Raipur is a bonafide research work conducted by **Shri RAJEEV SHRIVASTAVA** under my guidance and supervision. The subject of the thesis has been approved by the student's advisory committee and the Director of Instructions.

No part of the thesis has been submitted for any other degree or diploma or has been published / published part, has been fully acknowledged. All the assistance and help received during the course of investigations have been duly acknowledged by him.

24/2/98

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RAIPUR, 492 012 (M. P.)

Dated: 7/12/98

CERTIFICATE - II

This is to certify that the thesis entitled **"INHERITANCE AND ALLELIC RELATIONSHIP OF GENES CONTROLLING RESISTANCE TO GALL MIDGE (*Orseolia oryzae* Wood Mason) IN SOME RICE CULTIVARS "** submitted by **Shri RAJEEV SHRIVASTAVA to the** Indira Gandhi Krishi Vishwavidyalaya, Raipur in partial fulfilment of the requirements for the degree of Ph.D. (Ag.) in the Department of Plant Breeding and Genetics has been approved by the Student's Advisory Committee and External Examiner after an oral examination on the same.

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Date - 24.2.98

(RAJEEV SHRIVASTAVA)

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Chapter - 1

CHAPTER - I

INTRODUCTION

India has largest area under rice in the World and ranks second in production next to China. These two countries account for over 55 percent of the total production from about 50 percent of the World's area under rice (Taimni, 1996).

In India, rice is cultivated in about 42 m ha with an annual production of 78.79 m.t. In Madhya Pradesh, it is grown in about 5.4 m ha with an annual production of about 6.5 m.t. accounting for nearly 14 per cent of national acreage and 8 per cent of national production. In Chhattisgarh, which is also known as 'rice bowl' of M.P. rice is cultivated as a major crop, covering an area of about 4 m ha and production of 5.3 m. t., which is 74.42 and 82.03 percent of State's area and production, respectively (Anonymous, 1994-95).

In India, rice production reached to 81.61 million tonnes during the year 1995-96, while it was only 30.81 million tonnes during 1950-51. The productivity of rice during the same period increased from 0.67 t/ha to 1.9 t/ha. Rice has contributed nearly 42.5 percent of the total food grain production (189.77 million tonnes) during 1995-96 (The Hindu, survey of Indian Agriculture, 1996).

With the advent of dwarf fertilizer responsive varieties of rice, farmers were encouraged to grow these high yielding varieties with modern technology. This promoted the domination of monoculture cultivation in larger areas, resulting into simultaneous replacement of the land-races and local cultivars on a large scale. Because of reduction in genetic variability, the genetic vulnerability of the crop to diseases and insect pests became a serious problem.

There has been a tremendous change in the status of several rice insect pests in the recent past. Thus, relatively minor insect pests in the past such as leaf folder, case worms, army worms and cutworms have gained major importance. Likewise gall midge, brown plant hopper (BPH) and green leaf hopper (GLH), besides stem borers have become major problem during last 3 decades.

Rice gall midge *(Orseolia oryzae* Wood Mason) is a significant pest in the irrigated and rainfed ecosystems of India, China, Thailand, Cambodia, Vietnam, Indonesia, Myanmar, Sri Lanka, Bangladesh, Lao PDR etc. causing 10-100 percent yield losses. (Prakasa Rao, 1972; Anonymous 1981, 1991; Lai *et al.,* 1984 and Tan *et al.,* 1993).

Similarly the related species *Orseolia oryzivora* Harris and Gagne, the African Rice gall midge, is a problem in Nigeria and several other African countries (Ukwungwu and Joshi, 1992 and Taylor *et al.*, 1995). Insect pressure is highest in wet seasons when the atmospheric relative humidity is consistently high. Periods of dry weather in the rainfed ecosystem reduce the insect pressure and are a limitation in screening of breeding material for gall midge resistance. Richharia (1972) estimated the losses to the extent of Rs. 2 crores annually in Madhya Pradesh only, whereas, globally the losses have been estimated to exceed US $ 550 million annually (Herdt, 1991).

In India, rice crop in important rice growing states like, Assam, Andhra Pradesh, Bihar, Orissa, Tamil Nadu, Tripura, Madhya Pradesh, Maharastra, Manipur, Kerala etc. is equally infested by this pest.

The character symptom of attack of *Orseolia oryzae* on rice plant is the formation of gall commonly called 'silver shoot', which is a modified leaf sheath. Galls arise as a cellular response to an irritant secretion of salivary glands containing `Cecidogen' which causes abnormal proliferation of cells at the site of feeding (Chiu-Foon, 1980), clustering of leaf blades at flowering (Rajamani *et al.*, 1979), infestation of terminal shoot apices, tender grains in panicle just at flowering,

abnormal twisting and curling of leaf sheaths, profusion of leaf primordial from panicle initiation to ripening etc.

The insect population becomes intense 6 to 8 weeks after the onset of the monsoon and declines later in the growing season largely due to parasitization. In dry season it is reported to survive on some grass hosts (Israel *et al.,* 1970). Being an internal feeder, chemical control of this pest is difficult. Therefore the use of resistant varieties is the most effective strategy to manage this pest.

Use of host plant resistance (HPR) is an inexpensive and environment friendly method for the control of pests. Although, many resistant genes for rice gall midge have been identified in rice germplasm, it has been difficult to develop varieties with long lasting or stable resistance due to the high degree of variability in the insect. Moreover resistant varieties, particularly those carrying single gene for resistance, often break down (become susceptible) due to inadequate screening and/or due to emergence of new biotype races. For effective deployment of resistance genes in different localities, knowledge on the biotype population structure occurring in a particular area and an effective and precise method of selection of the breeding material are essential.

Biotypic variability of gall midge appears to be the major factor for the variable reaction of resistant donors. The term `biotypes' is used in a provisional and operational sense to mean the different genotypes of an insect that display differential responses on a set of donors. One of the earliest indications of the existence of biotypes in Asian rice gall midge was the differential reaction of test donors in different countries (Fernando, 1972).

Since then, many regional, national and international collaborative studies have been undertaken to detect and define different biotypes of gall midge.

The earlier reports indicated the presence of at least six biotypes in India, four in China, two in Sri Lanka and one each in Thailand and Indonesia (Anonymous, 1981, 1991; Nair and Devi 1994; Prakasa Rao and Kittur, 1989; Kalode and Bentur, 1989 and Singh, 1996). These biotypes differ in their host range, presumably depending upon the resistant genes present in the cultivars. The presence of different insect biotypes is adjudged from the differential reactions of a defined set of crop varieties (differentials) to pest populations over a number of locations at a regional, state or country level. The existence of biotypes must be taken into account while breeding cultivars for gall midge endemic regions.

In Sixties many gall midge resistant sources were identified at Warangal. These were, Eswarkora, HR 42, HR 63, Ptb 18, Ptb 21 and Siam 29. Now these sources as well as their derivatives are used as the sources for resistance against gall midge.

Information on nature of genetic control of the trait is an important feature for using a new donor in any breeding programme. The available information regarding nature of genetic control of gall midge resistance is very much conflicting. For instance, Shastry *et al.* (1972), Sastry and Prakasa Rao (1973) and Sastry *et al.* (1975) reported involvement of three complementary genes for resistance, while Satyanarayan and Reddy (1972), Venkataswamy (1974) and Prasad *et al.* (1975) found only monogenic resistance by dominant gene. Khamboonruang (1973) reported resistance to be controlled by recessive gene(s). The possibility of polygenic control had also been expressed by Wickram Singhe (1980). However, extensive studies carried out in the Department of Plant Breeding and Genetics, Indira Gandhi Agricultural University, Raipur led to the identification of 5 genes for resistance viz., Gm-1, Gm-2, gm-3, Gm-4 and Gm-5 (Chaudhary *et al.*, 1985; Shrivastava *et al.*, 1993 and Shrivastava *et al.*, 1995).

To over come the problem's of break down of resistance arising due to development of new biotypes, it is necessary to have clear understanding of inheritance of resistance and also the allelic relationship of gene(s) present in donors. The allelic study provides information on genetic relationship among different sources of resistance, so that the breeder may not unconsciously incorporate the same gene for resistance, even though he may be using different parents (Athwal and Khush, 1974). For this, we need to identify new diverse donors. Carefully planned studies on allelic relationship of resistant gene(s) will be helpful in identifying new genes for resistance which is necessary not only for developing varieties with wider adaptability through gene pyramiding but also to have a check on genetic vulnerability of crops to the changed biotypes of the insect.

Keeping these considerations in view, the present study was carried out with following objectives:

1. To study the inheritance of gall midge resistance in some rice varieties.
2. To study allelic relationships of the genes present in test varieties with those carried by 4 known resistant varieties.
3. To study the linkage relationship of gall midge resistance with leaf pigmentation.

Chapter - 2

CHAPTER - II

REVIEW OF LITERATURE

The Asian rice gall midge *Orseolia oryzae* Wood Mason, is one of the most destructive pests of irrigated and shallow low land rice ecosystem in South and South - East Asia. The rice gall midge problem is causing increasing concern, especially because of the existence of different biotypes and the inability to control the pest *by* chemical use in areas, where management of water is not possible. During the last few years, as a result of extensive screening, a number of donors imparting resistance against different biotypes have been identified. Since good gall midge donors are available, a breeding programme using these donors is Likely to pay rich dividends. The mechanism of gall midge resistance to rice cultivars which has a great, bearing in formulating the future breeding programme has so far been not adequately worked out. Even our knowledge on bionomics of rice gall midge and species involved in alternate grass hosts is far from clarity. The aspects which are included here are related to its occurrence, economic importance, taxonomy, bionomics, biotypes, sources of resistance, mechanism of resistance and genetics of resistance etc.

2.1. OCCURRENCE :

In sixtees to eighties, the gall midge was considered to be a minor pest of rice with low damage. However, the advent of dwarf high yielding varieties and improved technology resulted in change in status of some insect pests and gall midge became one of the most important pest, causing extensive damage to rice crop.

Gall midge has covered a large area in South, South - East Asia, Africa and some Western countries. In Asia this pest has been reported in Bangladesh, Pakistan and Srilanka (Hill, 1975; Wickram Singhe, 1969), in Burma and Combodia (Ghosh, 1924; Grist and Lever, 1969) in China (Yen et al., 1941 ; Li and Chiu, 1951 and Barnes, 1956), in India (Cotes, 1890; Mani, 1973 and Reddy, 1967) and in Indonesia, Laos, Nepal, Thailand and Vietnam (Nguyen, 1922; Ladell, 1933).

In African countries an other species of this pest (*Orseolia oryzivora* Harris and Gagne) has been reported from Cameroon (Descamps, 1956), Ivory Coast, Mali, Senegal, Sudan, Nigeria, Upper Volta (Anonymous, 1963; Harris, 1960 ; Joyce, 1954 and Hill, 1975), New Guinea, Zambia (Alam *et al.,* 1985). This pest has also been reported from Texas in U.S.A. (Way and Wallace, 1989). So far Australia, Malaysia and Philippines are reported to be free from gall midge (Reddy, 1967; Hidaka *et al.,* 1974).

Available information showed that the gall midge was first observed by a magistrate of Monghyr district of Bihar in 1880. The first record regarding gall midge was published by Riley (1881), without any format description. Further, it was observed at Mysore (India) in 1901 (Sontakay, 1941). Since 1913, its occurrence has been reported from various parts of Bengal, Bihar, Madras, Orissa (Ghosh, 1921), Andhra Pradesh, Karnataka, Uttar Pradesh (Panda, 1976), Manipur (Singh, 1990) and (Sundarraju, 1984).

Earlier, incidence of this pest was a localised in Raipur, Durg, Balaghat, Rajnandgaon and Raigarh districts of Madhya Pradesh (Anonymous, 1938; Shrivastava *et al.,* 1988). Later on this pest has been reported from Jabalpur (Patel and Bhowmic, 1987) and Morena (Rajpoot *et al.,* 1993) districts.

The gall midge was considered as a pest of wet season and low land rice but it had also been reported to infest the winter paddy (Kalode and Kasivishwanathan, 1976), upland rice (Li and Chiu, 1951), deep water rice (Venugopal Rao, 1975), wild rice species (Reddy, 1967; Israel *et al.,* 1970; Hidaka *et al,* 1983) and weeds (Israel *et al.,* 1970).

Joshi and Ukwoungwu(1990) reported rice ratoons as potential host for African rice gall midge. They also reported that in general, damage on ratoons was higher than on main crop (Joshi *et al.,* 1984).

2.2. ECONOMIC IMPORTANCE :

Rice gall midge *(Orseolia oryzae* Wood Mason) is an important pest of rice, causing severe damage to rice crop in South and South - East Asian countries, including India, regularly. In India none of the rice growing state is escaped from the attack of this pest. In the regions, where rice is taken both during *kharif* and *rabi,* the intensity of occurrence of this pest is greatly increased.

In rice ratoon, the pest causes severe yield losses. Richharia (1972) estimated loss of Rupees two crores per annum in Madhya Pradesh alone.

In endemic area of Orissa, the losses due to gall midge were reported to the extent of about 80 percent (Panda, 1976). In North - Eastern Andhra Pradesh the average yield losses due to this pest in different susceptible varieties to be ranged between 33% - 88% in 1986 (Bentur *et al.,* 1987). The emergence of new biotype caused heavy infestation of gall midge in 31,000 hectors rice area of North - Eastern Andhra Pradesh, there by causing a loss of more than Rupees 150 million (Kalode and Bentur, 1988).

Globally, the losses due to gall midge have been estimated to exceed US $ 550 million annually (Hardt, 1991).

2.3. TAXONOMY :

The rice gall midge *Orseolia oryzae* Wood Mason belongs to order Diptera, family Cecidomyiidae. After its first reported occurrence in 1880 in Monghyr district of Bihar, it was named as Cecidomydia oryzae by Wood - Mason in 1881 (Cotes, 1890). The description given by Wood Mason (Culcutta Gazette, 1880) was not enough. Ghosh (1917) illustrated different stages of the life cycle of the fly along with its characteristics damage on rice. Subsequently, Felt (1921) identified it belonging to the genus *Pachydiplosis* and Kiffer named it *Pachydiplosis oryzae.* Mani (1934) for first time described the midge in detail and retained the same name. However, doubts were raised about the genus *Pachydiplosis,* since rice gall midge resemble more with species of *Orseolia,* Creating galls of several graminacious plants. Ultimately, Gagne (1973) shifted it to genus *Orseolia,* although authorship still remain in the name of Wood - Mason. So the rice gall midge in Asia is now nominated as *Orseolia oryzae* Wood -Mason.

Previously it was assumed that the rice gall midge occurring in Asia and Africa belonged to the species *Orseolia oryzae* Wood - Mason (Diptera; Cecidomyiidae). But later critical taxonomic studies of Harris and Gagne (1982) resolved that the African samples of rice gall midge collected from Senegal, Upper Volta, Ivory Coast, Nigeria and Sudan belonged to *Orseolia oryzivora* Harris and Gagne, and are distinctly different in their larval, pupal and adult stages from the Asian rice gall midge *Orseolia oryzae* occurring in India and other Asian countries (Prakasa Rao, 1984).

2.4. BIONOMICS:

The rice gall midge *(Orseolia oryzae* Wood Mason) is a minute mosquito like insect. The females are orange to orange brown, whereas the males are considerably smaller and pale brown. Under natural conditions adults emerge from galls at night or at early dawn and copulate immediately after emerging (Ghosh, 1921). The male dies within 12-18 hours after emergence, while the female survives for 3 days. Females start ovipositing on rice plants in the evening after emergence. Eggs start hatching 72 hours later at dusk (Ghosh, 1921).

The biology and life cycle of this insect have been described by Panda and Mohanty (1970), Perera and Fernando (1970) and Hidaka et. al.(1974). Panda (1976) revealed that gall midge adults started emerging in between 17.00 to 22.00 hrs. and peak emergence was between 22.00 to 23.00 hrs. Immediately after emergence mating

took place and oviposition occurred towards early morning. The eggs are usually laid on both sides of leaf blade and also on leaf sheath and ligule. Eggs are laid either singly or in groups on substrate. The unfertilized females also laid eggs, but such eggs did not hatch. The fertile eggs hatch within 48 hours after they are laid. The just hatched maggots usually penetrate inside the leaf sheath immediately after emergence between 3 to 5 hours. The female population of gall midge was almost twice to that male and the male mates with the female more than twice. Single female lays most of her 175 to 200 eggs on the first night after emergence. The different stages in the life cycle of gall midge are described below:

EGG -	Elongate, Reddish brown (0.44 mm X 0.25 mm), 72 hor. old egg hatched
	1 st instar (0.50 mm X 0.12 mm) 13 segmented body, 3 to 4 days.
LARVAE	2nd instar (1.5 mm X 0.4 mm),resemble to 1st instar larvae, 3 to 4 days.
	3rd instar (3.2 mm X 0.8 mm), 6 to 7 days
PUPA	Male Pupa smaller then female pupa 24 hors.
ADULT	Male one day
	Female three day.

The African rice gall midge (*Orseolia oryzivora* Harris and Gagne) lays eggs singly or in cluster on the both sides of leaf blade, the eggs hatched in 3 days and the lifecycle completed in 26 days. About one - third of the eggs laid by mated females failed to hatch. On an average, 408 eggs were laid by mated females in a mean life of 3 days. Unmated females Lived for 1 -3 days and laid 88 eggs. At the beginning of season, the male to female ratio was 1 : 2 , at middle, it was 1 : 1, but at the end of the season the ratio reach up to 1 : 3 respectively (Umeh and Joshi, 1993).

2.5. OVIPOSITIONAL PREFERENCE :

Rajamani and Chatterji (1986) reported, that there was no significant difference among the resistant and susceptible cultivars in the. number of eggs laid by the gall midge. They reported in rice plant irrespective of varieties, leaf blade was most preferred for egg laying. Even pigmented rice were equally preferred for egg laying and larval development. They also reported that rice cultivars were preferred more by insect for laying eggs than the alternate hosts.

2.6. ALTERNATE HOSTS :

Few alternate grass hosts, on which the midge maintained the life cycle during off season were *Cynodon dactylon, Paspalum scorbiculatum, Saccolepis interrupta, Panicum milia-ccum,*

Heteropgon opntrotus, Ischaemum rugosum. In these grass hosts the midge completed its life cycle much faster (9 - 15 days) during May - June and migrated to rice plants during *kharif*season and further repeated its generations (13 - 21 days) . Besides the rice gall midge, the weed hosts also harboured some other unidentified midges. The role of these unidentified midges on rice hosts was not known (Panda, 1976).

2.7. POPULATION FLUCTUATIONS IN THE FIELD :

Ten years (1961 - 1971) data of light trap catches at Cuttack confirmed the earlier observations of Ghosh (1921), Ramachandra Rao (1925) and Hegdekatti (1927), Wong *et al.* (1956) and Israel *et al.* (1964), that the gall midge occurred essentially in the monsoon season corresponds to the *kharif* crop (July November/December) and did not appear at light trap during *rabi*, except only once during May and June 1971. The periods of gall midge abundance varied from 30 to 70 days between early August and Novemberend.

Wickram Singhe (1969) and Fernando (1972) observed that gall midge attack in the field began with a low infestation rate in nurseries or in broadcast seedlings. Its incidence increased gradually up to 8 to 11 weeks later, which was the stage of maximum tillering in 4½ month, rice variety. Most of the productive tillers were formed

by the 10 th week and three after mainly tertiary unproductive tillers were produced. Gall midge attack after about the 10 th week from sowing had no adverse effect on yield.

Fernando (1972) revealed that the eggs, first instar larvae and adults were highly susceptible to change in relative humidity. The eggs required a relative humidity of over 90 percent for normal development, whereas the first instar larvae required a relative humidity of over 95 per cent, associated with moist surface for successful infestation of rice plants. The adult female required a relative humidity of over 70 percent, for normal longevity and egg laying.

Bentur *et al.* (1987) revealed that cloudy and humid weather favour gall midge multiplication. Prakasa Rao (1988) reported that continuous rains, cloudy and rainy weather favoured gall midge emergence and incidence. High relative humidity of 89 per cent and above prevailing at the base of the rice plant enhanced egg hatching and infestation.

2.8. NATURE OF DAMAGE :

Roy *et al.* (1971) reported that the maggots of the rice gall midge penetrated to the developing primordium of the rice plant by wringling down through the enveloping whorls of the leaf sheaths. In the

primoridium they fed and developed on the tender leaf sheath, leaf blade tissues, with the result that the free margins of the leaf sheaths became glued in the form of a hallow, long tubular gall or silver shoot. Affected tillers did not produce panicles and significant yield losses were caused.

According to Panda (1976), infestation by this insect resulted not only in the formation of silver shoot but also in the stunting of tillers. Inside the stunted tillers, the gall midge larvae were surviving and there were no sign of silver shoots.

The maggots feed internally on the growing tips of the tillers and transform them into tabular, onion leaf like structures called `Silver shoots'. Since the insect is endoparasitic, chemicals have not been found very effective in controlling it. The host plant resistance, this has been suggested to be the most logical approach to check the yield losses (Khush , 1977 and Heinrich and Pathak, 1981).

Prakasa Rao (1983) reported that the characteristic symptom of attack of *Orseolia oryzae* on rice plant was the formation of gall commonly called the silver shoot, which was a modified leaf sheath. Gall arose as a cellular response to an irritant secretion of salivary glands containing 'Cecidogen ' which caused abnormal proliferation of cells at the site of feeding (Chiu Foon, 1980). Clustering of leaf blade

at flowering (Rajamani *et al.*, 1979) infestation of terminal shoot apices, tender grains in panicles, abnormal twisting and curling of leaf sheaths, profusion of leaf primordia from panicles initiation to ripening etc. as a result of gall midge infestation have also been reported.

2.9. BIOTYPES :

In nature all living organism including insects have potential to adjust themselves in changing environment. Mutation and hybridization are the major ways of achieving this objectives. In case of varietal resistance this, however, leads to development of new biotypes capable of attacking a known resistant variety of the area. Generally, the biotypes may occupy definite geographical areas.

As early as 1894, Erickson in Sweden demonstrated that the fungi causing stem rust, *Puccinia graminis* varied in pathogenicity (Panda, 1979). He next established that in collections taken from various hosts, each had a characteristic, host range attacking certain species but not others.

The biotypic variation in insects is well known in Hessian fly on wheat and aphids in various crop plants. Recent studies in rice gall midge also showed the existence of different biotypes in this pest.

Earlier studies regarding differential reactions of rice varieties to gall midge population were made by Israel and Vedamoorthy (1953), Khan and Murthy (1955) and Roy *et al.* (1971). Other workers (Shastry *et al.*, 1972 ; Fernando, 1972; Pangprasert *et al.*, 1972; Prakasa Rao *et al.*, 1973 and Kalode and Kasiviswanathan, 1976) reported wide variation in the degree of gall midge incidence on the same varieties under field conditions, suggesting the possible intrinsic variations in the insect itself or admixture of biotypes at different locations.

W-1263 and Ptb-21 showed resistant reaction at Warangal (A.P.), but susceptible at Sambalpur and Cuttack and vice versa (Roy *et al.*, 1971).

Shastry *et al.* (1972) for the first time reported the existence of biotypes in gall midge in India, on the basis of multi location trials with differentials, such as, Eswarkora, Siam-29, Ptb-21, Levang-152, Muey Nahug-62M and their derivatives.

Joshi and Venugopal (1984) observed distinct morphological variations in adult gall midge *Orseolia oryzae* Wood - Mason collected from Hyderabad, Nagpur and Madurai. They found variation in size of trochanter and femur, the structure and shape of the kind tarsal segments and the body pigmentation.

On the basis of differential reaction of rice varieties Joshi and Vanugopal (1985) suspected the possibility of a different biotype of *Orseolia oryzae* at Madurai. Existence of at least three biotypes in India was reported by Roy *et al.* (1985).

Existence of a different biotype was suspected at Raipur, where Ptb group has shown susceptibility in variance to its behavior in Andhra Pradesh (Bentur *et al.*, 1987). The existence of different biotype at Raipur and Andhra Pradesh was confirmed by Kalode and Bentur (1988) and Reddy *et al.* (1989), they reported that Phalguna and Surekha showing resistant reaction at Raipur, were observed to be susceptible in Andhra Pradesh. Prakasa Rao and Kittur (1989) studied the incidence of gall midge in 21 varieties of rice at Raipur and Cuttack. They observed differential reaction at two locations, varieties Asha, Usha and Samridhi found resistant in Raipur, were susceptible at Cuttack whereas, Shakti, IR 36, IR 50, Daya and Pratap which were resistant at Cuttack were found to be susceptible at Raipur. They suspected this differential reaction was due to the existence of different biotypes at these locations.

Bentur *et al.* (1987) reported the breakdown of resistance of Surekha and Phalguna varieties in North - Eastern region of Andhra

Pradesh in 1986 and attributed to the emergence of new virulent biotype of gall midge.

Kalode and Bentur (1987) reviewed the situation related to biotypes in gall midge on the basis of the genes (Gm -1) and (Gm-2) defined by Chaudhary *et al.* (1985). They confirmed the existence of three biotypes in India. Later Kalode and Bentur (1988) observed that most of the known resistant donors became susceptible and they suggested that the new population of gall midge should be considered as biotype-4.

Gene	Biotype-1 AP & MP	Biotype-2 Orissa	Biotype-3 Ranchi	Biotype-4 Srikakulam
Gm -1 Eswarkora	R	S	R	S
Gm -2 . Siam 29/leaung 159	R	R	S	S

Reddy *et al.* (1989) found that at Srikakulam (A.P.) all the resistant sources and their derivatives became susceptible except Bangalei. This confirmed the existence of a new biotype in this district.

Recently, Bentur *et al.* (1994) studied the reaction of rice varieties to different biotypes (B_1, B_2 and B_4) of rice gall midge and found that twenty two varieties were resistant against all biotypes, 13

were resistant to B_1 and B_2 and IR 9828 - 2 -3 1 was resistant against B_1 and B_4.

Srinivas *et al.* (1994) studied the gall midge reaction in 11 varieties of 3 group of donors viz., Eswarkora, Siam-29 and Velluthacheera. They concluded that there was a shift in the gall midge reaction from biotype -1 to biotype -3 in the district.

Fifth biotype has been reported by Nair and Ambika Devi (1994) from Moncompu, Kerla. They evaluated 10 differentials in 3 major groups and susceptible check TN-1 all with known reactions to the biotypes. They observed that gall midge population at Moncompu tended to be different from four biotypes. They have been designated as biotype -5 with the characteristic R - R - S - S pattern for the four group of differentials.

Recently sixth biotype of gall midge has been reported from Manipur by Singh (1996). The Manipur biotype was previously supposed to be biotype-3, similar to each of Ranchi biotype. Singh (1996) found a new pattern of reaction to differential rice varieties. Only W-1263 and ARC-6605 of group I showed resistant (R) reaction, whereas differentials of the other three groups (i.e., Phalguna, ARC-5984 , CR-MR 1523, Velluthacheera, Aganni, Ptb -10 and T 1477) showed a consistent susceptible(s) reaction, thus depicting a new R - S - S - S *type*

response, which had not been previously reported in India. In supplementary studies, the Iroisemba population did not mate with any of the biotypes. Therefore, this be considered a distinct biotype-3 M, that is different from biotype-3 prevalent in Bihar (Ranchi) and Andhra Pradesh (Jagtiyal) (Singh, 1996).

2.9.1. GALL MIDGE BIOTYPES :

Differential reactions due to different biotypes are summarised as follows :

Differential	1	2	3	4	5	6
Eswarkora (W 1263)	R	S	R	S	R	R
Siam 29/Leuang 152	R	R	S	S	R	S
Ptb 21	R	R	R	R	S	S
TN-1	S	S	S	S	S	S

BIOTYPE-1.	Madhya Pradesh and Andhra Pradesh
BIOTYPE-2.	Orissa (Cuttack)
BIOTYPE-3.	Bihar (Ranchi)
BIOTYPE-4.	Srikakulam and Vijayanagram (A.P.)
BIOTYPE-5.	Moncompu , (Kerala)
BIOTYPE-6.	Manipur

Biotype -1 , which is prevalent in Andhra Pradesh and Madhya Pradesh can not attack plants carrying genes Gm-1, Gm-2 and Gm-4. Biotype-2 which exists in Orissa, can attack on Gm-1 but not to Gm-2 and Gm4. Biotype-3 exists in Bihar and it can not attack on Gm-1 and Gm-4 but can attack on Gm-2, whereas, biotype -4 which appeared in 1988 at Srikakulam and Vijayanagram district of Andhra Pradesh is capable of attacking both Gm-1 and Gm-2 but cannot attack on Gm-4. Biotype -5 which was reported in 1994 at Moncompu, Kerala can attack on plants with Gm-4, but can not attack plants with Gm-1 and gm-3. Recently reported biotype -6 from Manipur, can not attack plants with Gm-1 only, whereas, it can attack the plants with other genes Gm-2 and Gm-4.

A review of the available information indicated the presence of at least six biotypes in India, four in China, two in Srilanka and one each in Thailand and Indonesia (Anonymous, 1981,1991 ; Nair and Ambika Devi, 1994; Prakasa Rao and Kittur, 1989; Kalode and Bentur, 1989; Shaw *et al.,* 1981; Lai *et al.,* 1984; Kudagamage and Gunawardena, 1989 and Singh, 1990; 1996). These biotypes differed in their host range, presumably depending upon the resistance gene present in the cultivars. The present status of the biotype variation in Asian rice gall midge is presented in Table - 1.

Table - 1 : Asian rice gall midge (*Orseolia oryzae*) biotypes in different countries.

Differentials	China				India						Indonesia	Thailand	Sri Lanka B	Sri Lanka P
	I	II	III	IV	I	II	III	IV	V	VI				
Eswarkora (W 1263)	MR	S	R	S	R	S	R	S	R	R	S	R	S	S
OB 677 (Ptb/EK)	MR	R	S	S	-	-	-	-	-	-	R	S	R	R
Ptb 21	S	S	S	S	R	R	R	R	S	S	S	S	R	R
Siam 29	R	R	S	S	R	R	S	S	R	S	R	S	R	S
Leuang 152	R	R	S	S	R	R	S	S	R	S	R	S	R	R
TN -1	S	S	S	S	S	S	S	S	S	S	S	S	S	S
Dagivgi	R	R	R	R										
Dwkang #1	R	R	R	R										

R - Resistant, S - Susceptible & MR - Moderately resistant

2.10. SOURCE OF RESISTANCE :

The varietal differences in gall midge resistance were recorded by earlier workers like Tien (1922), Hegdekatti (1927), Li and Chiu (1951), Ramaiah and Rao (1953), Khan and Murthy (1955), Sen (1957) and Bhat *et al.* (1958). These workers identified rice varieties Ratnachudi, Jaddu 1061 and Red Halga- 244 as moderately resistant, whereas varieties Bentawal, Maskaty- 1315, Nato, Bainsa and White Halga-1690 were found to be highly resistant to the gall midge. Studies on variental differences in respect of resistance to gall midge were initiated at CRRI in early 1950 's. The infestations were relatively low in scented, low tillering varieties (Anonymous, 1963). Ou and Kanjanasoon (1961) found Muey Nahang variety to be highly resistant against gall midge in Lampang province of Thailand. The thai variety Leaung 152 was also identified as highly resistant to gall midge (Anonymous, 1970). Kovitvadhi (1963) found in multilocational tests, that four varieties viz., Muey Mahang-62, Muey Mahang-62 M, Muey Mahang Fang and Dokputdor were resistant to gall midge. Thomas and Chacko (1968) identified Ptb 10 as resistant source. Pongprasert *et al.* (1972) confirmed the resistance of Muey Mahng-62 M, Peykeo - P - 129 and Peykeo - E - 63 and a line CR-94-13 derived from Ptb 18 X ptb 21 had been used at IRRI to develop popular gall midge resistant varieties, such as IR 32, IR 36, IR 40 and IR 42 (Prakasa Rao, 1972). Workers

of All India Coordinated Rice Improvement Project at Warangal also screened a large collection of varieties against rice gall midge and identified 50 varieties to be resistant against gall midge, 43 of which were from Assam Collection (Mishra and Kulshreshtha, 1971).

Shastry and Seshu (1971) reviewed the progress of work done in India on gall midge resistance and listed W-13400, W-12708 and W-12709 to show fair degree of resistance to gall midge. At Warangal (A.P.) similar studies resulted in the identification of resistant donors like Eswarkora, HR-42, HR-63 and Siam-29. The resistance of Ptb-18 and Ptb-21 was confirmed by Venkataswamy (1974), and Shastiy and Sehu (1971). Balasubrimaniam and Purushothaman (1973) identified Ptb-15 as resistant line for this pest in Kerla.

Resistance of Ptb-10, Ptb-18, Ptb-21, Siam-29, Leuang-152, Eswarkora and W-1263 was further confirmed at Warangal (Krishnamurthy and Krishnamurthy, 1964; Shastry *et al.*, 1972; Kalode *et al.*, 1977; Kalode, 1980 and Reddy *et al.*, 1990). Some of these lines are now being utilized as donors in resistance breeding programme.

CR 94 - MR 1550 a high yielding, medium duration, short statured variety with red kernel was found resistant to this pest, derived from a cross of (Ptb-18 X Ptb-21) X IR-8 (Sastry and Prakasa Rao,

1975). In Orissa varieties CR 57 - MR 1511, CR 57 - MR 1510, CR 57 - MR 1523, CR 94 - MR 1550 and CR 94 - MR 1624 were reported to combine high yield potential and resistance to gall midge (Sastry *et al.*, 1975).

Kalode *et al.* (1977) found AC-1224, ARC-5984, ARC-6158 and Leuang-152 to be resistant at Cuttack. Sastry and Prakasa Rao (1976) ' reported that the varieties Ptb-10, Ptb-18, Ptb-21, Leaung-152, ARC-6158, ARC-5984, ARC-6221, JBS-446, MNP-30 and MNP-471 were resistant against gall midge. ADR-52, Velluthacheera, T-1425, T-1471, Ptb-19, Ptb-21, Chenni Nayakan, Chemban, T-1426, MR-1523, ARC-11704, T-10, T-16, T-141, T-1432, Karutha Vellathan, Parakulam . and Chennethi were found to possess multiple resistance against gall midge, BPH and WBPH (Kalode *et al.,* 1977).

High yielding gall midge resistant selections of RP-352 series derived from two sources of resistance Eswarkora and Ptb 18, released as gall midge resistant varieties, Shakti and RP 6 -17 proved resistant to this pest in Orissa and A. P. respectively (Panda, 1976). Cultures, CR 56-MR-1511-IW-1020-2, CR 157-992-25, CR 94-MR-1550, CR 57-MR-1523-1030, CR 57-MR-1511-1026, RPW 6-17, OR 78-19, OR 79-11 and Shakti were found to be resistant to gall midge in Orissa (Panda, 1979), Shakti later lost its resistance in Sambalpur and Bolangir.

Richharia (1979) reported that varieties, Bd-1142, Bd-105, Bd-1238, Bd-1292, Bd-8 and Bd-2157 were tolerant against gall midge.

High yielding gall midge resistant varieties Vikram, Surekha, Phalguna, Shakti, Asha, Usha, RP 9-4 in all parts of the country, because of the existence of more than one biotypes in India (Kalode, 1980). Reddy *et al.,* (1989) found variety Bangalei as resistant at Srikakulam (A.P.) against a new gall midge biotype-4 and used it as a donor parent.

The gall midge resistance has been successfully incorporated into modern high yielding varieties in India and elsewhere. These are Kakatiya (1972), Surekha (1976) and Phalguna (1977) in Andhra Pradesh, Vikram (1972) in Karnataka, Shakti (1973) and Samalei (1980) in Orissa, Asha,Ushaand Samridhi (1980) in M.P. (Prakasa Rao, 1983). Recently a large number of high yielding varieties having resistance against different biotypes of gall midge have been released under All India Coordinated Rice Improvement Project are Mahamaya and Abhaya.

In Thailand RD -4 and RD -9 (Weerapt *et al.,* 1977) and in, Srilanka, BG 400-1 and BG 276-5 (Wickram Singhe, 1980) were recommended for commercial cultivation in gall midge endemic areas of the respective countries. The IRRI's involvement in gall midge

resistance breeding programme is very much limited due to non-occurrence of the pest in Philippines (Khush, 1977).

At Manipur, Phovern and Moirangphov Knokg angbi varieties were found resistant by Singh (1990).

Harinkhere and Phophaly (1991) reported that WGL-28712, CR 95-181-2, OR 151-17, Surekha and Phalguna were resistant in M.P. Shobharani et al., (1992) reported ARC-6157, IET-8636, IET-8890 and IET-9576 as the promising gall midge resistant cultures.

Soewito (1992) reported that Tajum, Barito-40 and Krad-e-Pn-89 were the resistant varieties in Indonesia.

Kalojira and Patnai -23 were found resistant against gall midge, whereas Hashikalmi, Badshahbhog and Paijam -II were found moderately resistant in Bangladesh (Alam *et al.*, 1979).

In Nigeria, 88 Asian gall midge resistant varieties were screened against African rice gall midge (*Orseolia oryzivora* Harris and Gagne) and 24 of these were rated as high to moderately resistant under field conditions (Ukwungwu and Joshi, 1992).

Prakasa Rao (1983) reviewed the source of gall midge resistance used in hybridization programme in India and elsewhere, as follows.

Cuttack	Ptb-10, Ptb-18, Ptb-21, Eswarkora derivatives (W-1263, W-12708), ARC-984, ARC-6221, ARC-6158, Leuang-152, derivatives of Ptb-18 X Ptb-21 (CR-93, CR-94), derivatives of Siam-29 (RPW-6).
Warangal & AICRIP	Ptb-18, Eswarkora and its derivatives (W-1263, W-1251, W-1257) RP-9, RP-8, RP-7, Siam-29 and its derivatives (RPW -6).
Raipur	Eswarkora derivative (W-1263).
Bhubaneswar	Derivatives of Ptb-18 X Ptb-21 (CR-93, CR-94), derivatives of Siam-29 (RPW-6), derivatives of Eswarkora (W-1263). Leaung-152.
Thailand	Eswarkora, Ptb-18 X Ptb-21 (CR-94), Mueynawng-62 M.
Indonesia	Eswarkora, Ptb-18 X Ptb-21 (CR-94) and their derivatives.
Ceylon	Eswarakora and its derivatives (W-1263), Ptb-18 and its derivatives.
IRRI	Ptb-18 X Ptb-21 (CR-94).

2.11. GENETICS OF RESISTANCE :

The first study appears to have been made by Shastly *et al.*, (1972) who postulated involvement of three complementary recessive

genes (Gm-1, Gm-2 and Gm-3) in resistant reaction and the effect of their dominant counter parts were suppressed by a dominant inhibitory gene (I-Gm-1). Resistant parents used in the study were W-1263 and Ptb-21. Presence of three recessive genes for resistance in W-1263 has also been reported by Sastry and Prakasa Rao (1973).

Sastry *et al.* (1975) reported the involvement of one to four pair of genes in the expression of resistance to gall midge in different genotypes. They obtained in F_2 the segregating ratio of 3 S : 1 R, 15 S : 1 R, 63 S : 1 R based on the presence of 1, 2 and 3 genes for susceptible and resistant, respectively. The ratio of 27 S : 37 R and 81 S : 175 R were also observed, indicating the interaction of complementary genes. The segregating ratios observed F2 were confirmed in F_3. They assigned the different genes in various genotypes studied by them, based on the segregating pattern in F2 and F_3.

Prasad *et al.* (1975) reported resistance to be governed by one dominant gene. They observed differential reaction in the cross RPW 6-13 X Vijaya and its reciprocal, suggesting the influence of cytoplasm in the expression of resistance to gall midge. The influence of cytoplasm in the resistance was reported for the first time.

The possibility of polygenic control of resistance to gall midge was expressed by Wickram Singhe (1980).

Nugaliyadde and Kalode (1981) observed that the resistance in experimental line Ob-678 was controlled by dominant gene(s), whereas, both the parent of this line viz., Ptb-18 and Eswarkora were found to have recessive gene for resistance. Resistance of Ptb-21, IR-36, MR-1523, Eswarkora and Muey Nawng-62 M was also indicated to be due to recessive genes. Prakasa Rao (1983) reported that the resistance to gall midge is governed by major genes and it takes the form of antibiosis.

Sastry *et al.* (1984) suggested that two recessive genes in Phalugna, one dominant gene in RP-9-4, two to three dominant complementary genes in CR-57-MR-1523 and one dominant gene along with one inhibitory gene in variety Leb - Muey Nahng was responsible for imparting resistance against the pest. The presence of single dominant gene in RP-9-4 was later confirmed by Sastry *et al.* (1985).

• Satyanarayan *et al.* (1987) concluded that resistance was governed by two to four genes involving complementary and inhibitory action. The presence of modifier genes was also indicated.

Prakasa Rao (1988) successfully transferred the duplicate and complementary genes of IR-20 in culture CR 294-548. This culture also combined field resistance against BLB and flash flooding up to 5 days.

Tomar and Prasad (1992) reported that the resistance in Surekha, Phalguna, Rajendradhan-202, IET-7918, IET-6187 and BG 404-1 was governed by a single dominant gene, whereas a monogenic recessive gene was responsible in OB-677 and BKNBR 1008-21. Shobharani *et al.* (1992) also reported, a single recessive gene to be responsible for resistance to gall midge in ARC-6157. They also found dominant gene governed resistance in IET-8890/IET-9576.

Yujuan *et al.* (1994) reported that the reaction of varieties to gall midge biotypes differed distinctly. He reported that resistance against gall midge biotype-4 was governed by a single dominant gene which was non allelic to gene imparting resistance against gall midge biotype-1 (Gm-1).

Chaudhary (1988) reported that the gall midge resistance in Usha, Surekha, BD 6-1, BD-83 and IET-6286 was governed by a single dominant gene, whereas in Shakti resistance was due to a recessive gene. The resistance in Bd-105 (Badshahbhog), Bd-1192 (Banspatri) and Kalimoonch-64 appeared to be due to polygenes.

Sahu *et al.* (1990) confirmed that the resistance of Samridhi and Surekha to be due to the presence of one dominant gene and in Shakti, it was controlled by a single recessive gene. Sahu *et al.* (1990) also reported the recessive nature of resistance in ARC-5984.

Mishra (1990) reported the presence of single dominant gene for resistance in Asha mutant, ARC-6619 and WR 35-81; two dominant gene in NHTA -8 and RP 2068-16-4-5 and one recessive gene for resistance in ARC-5984 and CR 331-403-2-2. Sahu (1991) confirmed the monogenic resistance of Samridhi, Surekha and Bahawarkanak.

Arvind Kumar (1992) reported the presence of a single dominant gene in Samridhi, R 302 -111, R 335-2, R 435-707, R 296-260. Dharmendra Kumar (1992) also confirmed the monogenic dominance for resistance in cultivars Samridhi and R 302-111. Bose (1996) reported the presence of single dominant gene for gall midge resistance in CR 392-5088-2, CR 410-3225, Kakatiya, Samlei and Assamchudi, whereas in Shyamla a single dominant gene for resistance along with one inhibitory gene was indicated. Shrivastava *et al.* (1995) reported that Abhaya had a single dominant gene for resistance designated as Gm-4, ARC 5984 had a single dominant gene designated as Gm-5, and RP 2068-18-3-5 had a single recessive gene designated as gm-3.

2.12. ALLELIC TEST :

The varieties which are resistant today may become susceptible after some years because of development of new biotype(s). Therefore, it is very much essential to identify new strains for resistance from available germplasm, and their genetic analysis

to identify resistant genes present in them. Existence of more than one biotype requires identification of different sources of (differentials) resistance. These identified strains could provide a scope for developing superior elite resistant varieties by combining resistant genes from different sources, the varieties would stabilize their resistance and production as well. The identification of new resistant gene(s) could be done by allelic test.

The first report regarding allelic test was made by Sastry *et al.* (1976). They reported that the resistance conferring gene present in Ptb-21 was different from the one present in W-1263. Choudhary *et al.* (1986) reported that the dominant gene Gm-1 was present in Eswarakora derivatives (Usha, Samridhi and Bd 6-1) whereas dominant gene Gm-2 was present in Siam-29 derivatives (Surekha and IET-6286), and these were non allelic.

Kulkarni *et al.* (1986) reported on the basis of two crosses BPT-3301 X Wg-129327 and Wg-129798 X Wg-129147 that 3 dominant genes controlling resistance against this pest. Sahu (1987) reported that the resistance in Usha was conferred by a single dominant gene, Gm-1.

Sahu *et al.* (1990) found that recessive gene of ARC-5984 segregated independently of `Gm-1' and `Gm-2' and designated it as `gm-

3'. Mishra (1990) reported that dominant resistant gene of Asha mutant, R-296-421-2 and recessive gene of ARC-5984 and CR 331-405-2-2 were non allelic to Gm-1 and Gm 2.

Arvind Kumar (1992) found that the gene present in ARC-10658 allelic to Samridhi (Gm-1), whereas it was non allelic to dominant gene present in R 296-418-100 and ARC-6621. Dharmendra Kumar, (1992) reported that Gm-1 gene of Usha was non allelic to Gm-2 gene of Surekha. Tomar and Prasad (1992) reported that resistant gene of RPW 6-17, IET-7918 and IET-6187 was allelic to Gm-1 and non allelic to Gm-2. The resistance in Phalguna, Rajendradhan-202, W-1263, RPW-6-17, IR-36 and WGL-48684 was governed by Gm-2 and it was independent of Gm-1.

A new gene `Gm-4' for resistance to gall midge in rice variety "Abhaya" was reported by Shrivastava *et al.* (1993). Shrivastava *et al.* (1995) also reported a new recessive gene for gall midge resistance in RP 2068-18-3-5. This recessive gene was designated as `gm-3'. Variety ARC-5984, which was reported to possess a recessive gene `gm-3' for resistance (Sahu *et al.,* 1990) was subsequently observed to possess a single dominant gene, was designated as `Gm-5' by them. Bose (1996) studied the allelic relationship of resistant gene (Gm-2) of Surekha with genotypes. He found two of them CR 410-3225 and CR

392-5088-2 were allelic to Surekha and thus it was concluded by him that these genotypes possessed Gm -2 genes for resistance to gall midge.

The list of genes identified at Raipur are summarized in Table -2 :

Table - 2 : List of gall midge resistant genes, reported from I.G.A.U., Raipur (M.P.)-

Desig-nation of gene	Nature of gene	Donors in which gene identified	Source	Reported by
Gm -1	Dominant	Samridhi	Eswarakora	Chaudhary et al. (1985)
Gm -2	Dominant	Surekha/ Phalguna	Siam 21 leaung 152	Chaudhary et al. (1985)
gm -3	Recessive	RP 2068 -18-3-5	Velluthecheera	Shrivastava et al.(1995)
Gm -4	Dominant	Abhaya	Ptb -10	Shrivastava et al. (1993)
Gm -5	Dominant	ARC 5984	ARC 5984	Shrivastava et al. (1995)

2.13. MECHANISM OF RESISTANCE :

Various morphological characters such as hairiness of leaves, purple pigmentation and compactness of leaf sheath were reported to be associated with gall midge resistance (Israel *et al.*, 1964; Venkataswamy, 1974; Roy *et al.*, 1973 and Joshi *et al.*, 1984). Modder

and Alagoda (1971) observed no difference in preference for oviposition between resistant and susceptible varieties. The differences became evident when larva entered in to the growing point. In case of resistant varieties, they died without further development, indicating the presence of antibiosis.

Guru and Roy (1974) correlated amino acid content with gall midge resistance. High phenol accumulation in growing points before infestation and decrease in phenol content after infestation in susceptible varieties was observed at Cuttack, whereas increasing phenol content in resistant varieties was observed during early infestation (Anonymous, 1975).

The resistant varieties exhibited high degree of antibiosis against gall midge maggots, killing them before reaching pupation and also prevented silver shoot formation (Prakasa Rao, 1972; Narsimha Rao, 1970; Fernando, 1972 and Pongprasert *et al.*, 1972). However if parasitized maggots infested tiller in some resistant varieties, silver shoots manifested (Prakasa Rao, 1984). Therefore, the pupal population recovered from test entries reflected more reliably the relative build up of gall midge populations. Considerable differences in the test varieties viz., CR-57 - Mr 1523-1030, CR 93- MR 1626, Shakti, CR 93-MR -1543-1065 and CR 93-MR-1549-1070 in respect of percentage silver

shoots, on the one hand and number of pupae recovered in periodical samplings on the other, might be attributed to the above influence (Padhi and Prakasa Rao, 1978). Kalode (1980) observed that the number of eggs laid on resistant lines were less in comparison to susceptible lines. However no difference was observed in percentage of hatched eggs in both group of varieties.

Joshi and Venugopal (1984) found that the resistance to gall midge in IET-7009 was due to antibiosis. They found that the development of the insect was poor and an adults emerged.

Joshi and Venugopal (1985) reported that the growth of the galls formed by the Cecidomyiidae was found to be highly and positively correlated with plant height. Galls on the growing points of resistant rice varieties were only recipient, not externally visible and contained small dead larvae of *Orseolia oryzae* indicating the absence of some nutrient essential to *Cecidomyiidae* growth, or the presence of some inhibiting substance.

Rajamani and Chatterji (1986) found no significant difference in number of eggs laid by *Orseolia oryzae* on resistant as well as susceptible cultivars. Leaf blade was mostly preferred for egg laying in all varieties.

Satyanarayana *et al.* (1987) revealed that, there is no association between resistance and anthocyanin pigmentation of the leaf sheath.

Sain *et al.* (1988) reported incidence of gall midge was higher in irrigated rice than in rain fed upland rice. Sain and Kalode (1994) also reported antibiosis as the predominant resistance mechanism antibiosis affecting oviposition was noted in resistant varieties. They observed that the concentration of phenolic compound in the basal stem portion was higher in resistant rice cultivars as compared to the concentration in susceptible varieties.

2.14. LINKAGE OF GALL MIDGE RESISTANCE GENE :

Linkage was found to exist between gall midge resistance gene derived from Eswarakora and a gene controlling brown hull colour with a cross over value of 34.5 to 43.8 percent (Sastry *et al.,* 1975). Mishra (1985), Chaudhary (1988), Sahu and Sahu (1989), Dharmendra Kumar (1992) and Bose (1996) reported linkage of Gm-2 gene with the gene for purple leaf colour. Tomar (1986) proposed that Gm-2 gene to be Linked with inhibitory gene of purple leaf colour. It was confirmed by Arvind Kumar (1992), who reported that linkage was found to occur between gall midge resistance gene and the purple leaf colour inhibitory gene and also between gall midge resistance inhibitory gene and purple leaf colour controlling gene.

Chapter - 3

CHAPTER -

MATERIALS AND METHODS

3.1. EXPERIMENTAL SITE :

The present study was carried out at the Research farm of Department of Plant Breeding and Genetics, Indira Gandhi Agriculture University, Raipur (M.P.) during *kharif* 1994 and *kharif* and *rabi* of 1995, 1996.

3.2. GEOGRAPHICAL LOCATION AND CLIMATIC CONDITIONS:

Raipur is situated at 22°33' to 21° 14' N latitude and 82°6' to 81°38' E longitude at an altitude of 298.15 m above the mean sea level and have dry sub humid climate. The average rainfall is about 1385 mm received mostly during June to September. The minimum and maximum temperatures vary between 13.2°C to 42.3°C from January to May.

The climatic conditions during the crop period were favorable for normal crop growth, as well as for inset multiplication, as given in Appendix - I.

3.3. EXPERIMENTAL MATERIAL:

Twenty one genotypes were selected for this study. Out of these, 15 were resistant against gall midge. However genes responsible for resistance were not known. Six genotypes were used as testers to determine the allelic relationship. Out of 6 testers, 4 were

Table - 3 : List of varieties and their sources.

S.No	Parents	Parentage	Source
(a) Test varieties			
1.	Balam	Donor	Cuttack, CRRI
2.	AC 1224-2	Donor	Cuttack, CRRI
3.	NHTA-8	Donor	Cuttack, CRRI
4.	Riting	Donor	USA
5.	MNP-76	Donor	Cuttack, CRRI
6.	AC 169	Donor	Cuttack, CRRI
7.	ARC 6632	Donar	Assam
8.	AE 20	Donor	Cuttack, CRRI
9.	AC 424	Donor	Cuttack, CRRI
10.	Calrose-70	Donor	USA
11.	AC 630	Donor	Cuttack, CRRI
12.	ARC 15831	Donor	Assam
13.	MNP 709	Donor	Cuttack, CRRI
14.	AC 8070	Donor	Cuttack, CRRI
15.	Kataribhog	Land race (donor)	Madhya Pradesh
(b) Known resistant varieties			
16.	Samridhi	IR 22/W 1263	Madhya Pradesh
17.	Phalguna	IR 8/Siam 29	Andhra Pradesh
18.	Abhaya	CR 157-392/OR 57-21	Andhra Pradesh
19.	ARC 5984	Donor	Assam
(c) Known susceptible varieties			
20.	Shyamla	R 60-2713/IR 8	Madhya Pradesh
21.	MW-10	MTU 15/WAIKOKU	Andhra Pradesh

possessing known resistant genes, whereas the remaining two were susceptible. The resistant genotypes with unknown genetic constitution were crossed to each of the six testers during *kharif* 1994. Sufficient seeds could be obtained only in 41 combinations. The F_1's of these 41 crosses were grown during *rabi* summer 1994 - 95 to raise F2 generations.

During *kharif* 1995, additional crosses were attempted to complete the set. However, sufficient seeds could be obtained only in 31 crosses. The seeds of these 31 F_1's were grown during *rabi* 1995-96, advance to F_2. Thus, sufficient seeds of 72 cross combinations could be obtained. Simultaneously F_2 generation of 72 cross combination and F_3 generation of 38 cross combinations were raised.

Thus the experimental material consisted of 15 parental lines, 6 testers, 72 F_1's, 72 F2's and 38 F_3 generations.

3.4. SCREENING METHOD :

Raipur and its adjoining regions are supposed to be the 'hot spot' for the occurrence of gall midge in *kharif*, therefore this region is good for natural screening of gall midge. Gall midge infestation occurs mostly in late sown crops. Its pressure builds up after September. Therefore, to synchronies the tillering phase (stage at which gall midge infestation takes place) with the occurrence of gall midge, the sowing of parents, F_1, F_2 and F_3 generation's were delayed up to last week of July (27^{th} - 30^{th} July) and the transplanting was completed in the first week

of September (31 st August to 1st September).

Each screening material i.e. parents, F_1, F_2 and F3 populations were surrounded by two rows of susceptible purple leaf variety 'Shyamla'. These rows were used as 'spreader rows', Since the gall midge incidence is quite high on this variety.

During *kharif* 1994, 1995 and 1996 fifty plants of each parent were screened against gall midge. F_1's were planted on platforms of Department of Plant Breeding and Genetics during *kharif* 1995 and 1996, for their screening against gall midge. In *kharif* 1995, 41 F_2 were grown in the field. The seeds of fifty F_2 plants in each cross were randomly harvested to raise F_3 generation. During *kharif* 1996, the F_2 populations were grown family wise (as produce of single F_1 , plant) in rows, whereas F3 were grown progeny wise. Single plant progeny were grown in a row. Therefore for each cross fifty rows were grown in F_3. For each generation a spacing of 15 x 15 cm between plant to plant, and 20 x 20 cm between row to row were maintained.

To achieve maximum infestation of gall midge, continuous irrigation along with extra doses of nitrogen were applied in the field. Urea was applied @ 150 kg/ha in three split doses to encourage succulency as well as number of tillers per plant, Phosphorus was applied @ 35 kg/ha as basal, whereas, potassium fertilizer was not used at all in any year. Standard cultural practices recommended for rice cultivation were followed in each generation.

When the crop was 40 days old, electric lights (100 - watt bulbs) were installed at a height of 5 m. above ground level all over the field ,to attract the gall midge from surrounding areas.

The observations were recorded, classifying individual plants as resistant or susceptible. F3 progeny rows were classified on row basis, as breeding true for resistance, true for susceptible or segregating for resistant and susceptible. The presence of single silver shoot per hill was taken as criteria of susceptibility (Shastry *et al.,* 1972).

3.5. STATISTICAL ANALYSIS

3.5.1. TEST FOR GOODNESS OF FIT :

The Chi-square (χ^2) test is a valuable tool that aids the investigator in determining goodness of fit. The *X* test hypothesize about the equality of the observer frequency distribution and the corresponding theoretical frequency distribution.

i.e. Ho : Observed frequency distribution fits satisfactorily to the theoretical given ratio.

Its approximate test criteria for the category type of data is given :

$$\chi^2 = \sum_{i=1}^{n} \frac{(O-E)^2}{E}$$

Where,

0 = Observed frequency of the class,

E = Expected frequency of the respective class,

Σ = Summation of all classes.

The null hypothesis Ho is rejected, if observed χ is found to be greater than the theoretical χ and there by these difference is said to be significant. Otherwise, the null hypothesis is accepted and the difference between observed and theoretical frequency distribution are not significant.

3.5.2. TEST OF INDEPENDENCE :

Simultaneous segregation for two character's in F2 population was subjected to the test of independence (Panse and Sukhatme, 1967) in order to know whether, there was any association between the two characters.

If a,b,c and d are the frequencies of four phenotypic classes AB, Ab, aB and ab respectively, then the X value tests the independence of A and B variables. It is calculated by :

$$\chi^2 = \frac{(ad-bc)^2(a+b+c+d)}{(a+b)(a+c)(b+d)(c+d)}$$

for, 1 degree of freedom.

The value of χ is compaired against the theoretical value of χ^2 at a selected (5 or 1%) level of significance for 1 degree of freedom. Suppose the selected level of significance is 0.05, then the calculated value of χ was less than the theoretical value (3.841), is concluded that

these characters are independent and if the calculated value of χ was higher than 3.841, it is concluded that two characters were not independent.

3.5.3. DETECTION OF LINKAGE :

If in test of independence it was found that the two characters did not segregate independently, further test was made to detect linkage between the genes of the two characters following by the method described by Panse and Sukhatme (1967).

When the material under study was segregating simultaneously for two characters Aa and Bb, which in turn segregated in the ratios of I_1 : 1 and I_2 : 1 respectively, their joint segregation into four phenotypic classes AB, Ab, aB and ab would be in the ratio of $I_1 I_2$: I_1 : I_2 : 1 respectively.

The X value was calculated by taking deviations of observed frequencies in the four classes from their frequencies on the assumption of independence. The total χ value for 3 d.f., thus obtained was partitioned into three orthogonal components:

i) X^2 for first character Aa with 1 d.f.

ii) X for second character Bb with 1 d.f., and

iii) X^2 for linkage with 1 d.f.

The X values of these components were compaired against theoretical values for 1 d.f. at 5 % level of significance (p=0.05,

3.841;0.01, 6.635). If component 1 and 2 were observed to be non significant and 3 was significant the existence of linkage was indicated.

3.5.4. ESTIMATION OF LINKAGE :

Linkage in terms of cross over value between two genes was estimated by the Minimum discrepancy method given by .Haldane (1953) as described by Murty (1954).

The estimation procedure is described by taking an example, in which the gall midge resistance is governed by a single dominant gene (R) segregating in a ratio of 3 resistant : 1 susceptible in F2 generation. The second character of leaf colour is governed by two complementary factors A and B segregating in a ratio of 9 purple : 7 green in F_2. Suppose R locus is linked with A locus in repulsion phase with a cross over frequency of P. Thus, the F_1 genotypic constitution of linked genes would be

$$\frac{R.C.O _ Pa}{r \quad \sim \quad A}$$

This will produce four types of gametes with following proportions:

Genotype of gametes	Frequency
RA	$\frac{p}{2}$
Ra	$\frac{1-p}{2}$
rA	$\frac{1-p}{2}$
ra	$\frac{p}{2}$

The following genotypes from there four types of gametes will be obtained in F_2.

Gametes	RA p/2	Ra l-p/2	rA l-p/2	ra p/2
RA p/2	p2/4	$p-p^2/4$	$p-p^2/4$	$p^2/4$
Ra l-p/2	$p-p^2/4$	$1-2p+p^2/4$	$1-2p+p^2/4$	$p-p^2/4$
rA l-p/2	$p-p^2/4$	$1-2p+p^2/4$	$1-2p+p^2/4$	$p-p^2/4$
ra p/2	$p^2/4$	$p-p^2/4$	$p-p^2/4$	$p^2/4$

Thus the four F_2 genotypes will be obtained in the following proportions.

F_2 genotypes	A-R-	A-rr	aaR-	aarr
Frequency	$2+p^2/4$	$1-p^2/4$	$-p^2/4$	$p^2/4$
If p^2 = P				
the	2 + P/4	1-P/4	l-P/4	P/4
frequency				

Segregation at the B locus in combination with the above genotypes will lead to the following six genotypes.

		R-A-	R-aa	rrA-	rraa
		2 + P/4	l-P/4	1-P/4	P/4
	3	6+3 P	3-3 P	3-3 P	3P
	4	16	16	16	16
		RP	RG	SP	SG
	1	2+P	1-P	1-P	P
bb			....	---	---
	4	16	16	16	16
		RG	RG	SG	SG

where, R = Resistant, S = Susceptible, G = Green, & P = Purple

The four phenotypes will thus be produced in the following proportions :

Phenotype	Resistant Purple	Resistant Green	Susceptible Purple	Susceptible Green
Symbol				a_4
Frequency	6+3 P	6-3 P	3-3 P	1+3 P
	16	16	16	16

This will give the function :

$$(6+3P)^2 \quad (6-3P)^2 \quad (3-3P)^2 \quad (1+3P)^2 \qquad .(i)$$

where, 1, L, n3 = a_3+ 1, and n4 =

Differentiating the function (i) with respect to P and solving for P after equating to zero we get,

-6 6

Cross over value under Repulsion Phase = $\sqrt{P}$

Cross over value under Coupling Phase = 1 -

Chapter - 4

CHAPTER - IV

RESULTS

The present study was conducted with following objectives:

1. To study inheritance of gall midge resistance in some rice cultivars.

2. To study allelic relationships of the genes present in test varieties with those carried by 4 known resistant varieties.

3. To study the Linkage relationship of gall midge resistance with leaf pigmentation.

The study was conducted during *kharif* 1994, 1995 and 1996 and during *rabi* in 1995 and 1996 in the department of Plant Breeding and Genetics, Indira Gandhi Agricultural University, Raipur (M.P.). The weather conditions during all these years were congenial for gall midge infestation and the incidence of gall midge in susceptible varieties was observed to be 100 per cent. Thus, the valid conclusions could be drawn regarding the various aspects of the study.

The inheritance studies were carried our in fifteen test varieties (Table - 3), in which the resistant genes were not identified earlier. These varieties were crossed with two susceptible varieties

viz., Shyamla and MW-10. Since MW-10 did not possess any resistant gene, the crosses involving it as one of the parents, gave an idea of the number and nature of resistant gene(s) present in test varieties, whereas crosses with Shyamla provided additional information regarding inheritance of gall midge resistance and its association with purple leaf colour. The crosses between resistant test varieties and known resistant donors viz., Samridhi, Phalguna, Abhaya and ARC 5984 gave information on allelic nature of the resistant genes present in them.

The results are summarised under the following heads:

4.1. Gall midge reaction of the test varieties included in the study.

4.2. Inheritance of gall midge resistance in test varieties .

4.3. Allelic relationships among the genes for resistance present in the known and new resistant varieties.

4.4. Linkage of gall midge resistant genes with the genes controlling purple leaf character.

4.1. GALL MIDGE REACTION OF THE PARENTS :

Fifteen test varieties along with four known resistant and two susceptible varieties were screened in the field during *kharif* 1994, 1995 and 1996 (Table - 4) for there reaction against gall midge.

Table - 4 : Reaction of parental strain to gall midge during *kharif* 1994, 1995 and 1996

S.No.	Varieties	No of Plants														
		Total			Resistant			Susceptible			% Susceptibility			Reaction		
		1994	1995	1996	1994	1995	1996	1994	1995	1996	1994	1995	1996	1994	1995	1996
1.	Balam	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
2.	AC 1224-2	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
3.	NHTA-8	50	49	50	50	49	50	0	0	0	0	0	0	R	R	R
4.	Riting	50	50	48	50	50	48	0	0	0	0	0	0	R	R	R
5.	MNP 76	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
6.	AC 169	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
7.	ARC 6632	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
8.	AE 20	50	49	48	50	49	48	0	0	0	0	0	0	R	R	R
9.	AC 424	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
10.	Calrose 70	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
11.	AC 630	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
12.	ARC 15831	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
13.	MNP 709	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
14.	AC 8070	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
15.	Kataribhog	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
16.	Samridhi	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
17.	Phalguna	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
18.	Abhaya	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
19.	ARC 5984	50	50	50	50	50	50	0	0	0	0	0	0	R	R	R
20.	Shyamla	50	50	50	0	0	0	50	50	50	100	100	100	S	S	S
21.	MW 10	50	50	50	0	0	0	50	50	50	100	100	100	S	S	S

Samridhi, Phalguna, Abhaya and ARC 5984, the known resistant varieties, showed 100 percent resistant reaction during all three years. Similarly, all the test varieties Balam, AC 1224-2, NHTA-8, Riting, MNP-76, AC-169, ARC-6632, AE-20, ' AC-424, Calrose-70, AC-630, ARC-15831, MNP-709, AC-8070 and Kataribhog also showed 100 per cent resistance in every screening.

Both the susceptible varieties, MW-10 and Shyamla consistently showed susceptible reaction during the screening.

4.2. INHERITANCE OF GALL MIDGE RESISTANCE IN TEST VARIETIES :

The F_1 and F_2 generations of the crosses between the test varieties and the susceptible variety, MW-10 were screened for gall midge reaction. The results are presented in Table - 5. Reactions of F3 progenies for some of the crosses are also presented in Table - 5.

All the crosses showed resistant reaction in F_1 and F2 segregated in 3 resistant : 1 susceptible ratio. This indicated that resistance in each test variety was controlled by a single dominant gene. The segregation in F_3 generation of the available ten crosses confirmed the resistance being governed by a single dominant gene in test varieties, since progenies in F3 segregated in the expected ratio of 1 true breeding resistant, 2 segregating and 1 true breeding susceptible.

Table - 5 : Gall midge reaction of F_1, F_2 and F_3 progenies of the crosses between resistant test varieties and susceptible variety (MW-10).

S.No.	Cross Combination	F_1		F_2 (No. of Plants)						F_3 No. of Progenies			
		No. of plants	Reaction	R	S	Total	Ratio	X^2 Value	P Value	Pro.Reaction R: Seg: S	Ratio R:Seg:S	X^2 Value	P Value
1.	MW-10/Balam	13	R	442	150	592	3:1	0.036	0.90-0.80				
2.	MW-10/AC 1224-2	13	R	223	76	299	3:1	0.027	0.90-0.80	13-26-11	1:2:1	0.24	0.70-0.50
3.	MW-10/NHTA 8	13	R	429	153	582	3:1	0.515	0.50-0.30				
4.	MW-10/Riting	13	R	515	190	705	3:1	1.429	0.30-0.20				
5.	MW-10/MNP 76	13	R	376	134	510	3:1	0.441	0.60-0.50	10-24-16	1:2:1	1.525	0.30-0.20
6.	MW-10/AC 169	13	R	441	151	592	3:1	0.081	0.80-0.70	15-25-09	1:2:1	1.480	0.30-0.20
7.	MW-10/ARC 6632	13	R	339	121	460	3:1	0.417	0.60-0.50				
8.	MW-10/AE 20	10	R	354	125	479	3:1	0.306	0.70-0.50				
9.	MW-10/AC 424	13	R	240	77	317	3:1	0.084	0.80-0.70	13-23-14	1:2:1	0.360	0.70-0.50
10.	MW-10/Calrose 70	12	R	400	144	544	3:1	0.626	0.50-0.30	13-23-14	1:2:1	0.360	0.70-0.50
11.	MW-10/AC 630	13	R	358	125	483	3:1	0.199	0.70-0.50	11-29-10	1:2:1	1.320	0.30-0.20
12.	MW-10/ARC 15831	13	R	490	161	651	3:1	0.025	0.90-0.80	13-27-10	1:2:1	0.680	0.50-0.30
13.	MW-10/MNP 709	13	R	400	129	529	3:1	0.137	0.80-0.70	08-28-04	1:2:1	2.160	0.20-0.10
14.	MW-10/AC 8070	13	R	448	148	596	3:1	0.008	0.98-0.95	14-26-10	1:2:1	0.780	0.50-0.30
15.	MW-10/Kataribhog	13	R	186	78	264	3:1	2.909	0.10-0.05	14-26-10	1:2:1	0.780	0.50-0.30

R = Resistant, S = Susceptible & Seg. = Segregation

Table - 6 : Gall midge reaction of F_1, F_2, and F_3 progenies of the crosses between resistant test varieties and known resistant varieties.

S.No.	Cross Combination	F_1		F_2 (No. of Plants)						F_3 No. of Progenies			
		No.of Plants.	Reac-tion	R	S	Total	Ratio	X^2 Value	P Value	Pro.Re-action	Ratio	X^2 Value	P Value
1.	Samridhi/Balam	13	R	518	36	554	15:1	0.058	0.90-0.80	22-24-01	7:8:1	0.285	0.70-0.50
2.	Phalguna/Balam	13	R	755	51	806	15:1	0.632	0.50-0.30	22-24-04	7:8:1	0.285	0.70-0.50
3.	Samridhi/AC 1224-2	13	R	595	05	600							
4.	Phalguna/AC 1224-2	10	R	520	38	558	15:1	0.298	0.70-0.50	25-24-01	7:8:1	1.931	0.20-0.10
5.	Abhaya/AC 1224-2	13	R	550	30	580	15:1	1.077	0.30-0.20	21-27-02	7:8:1	0.600	0.50-0.30
6.	Samridhi/NHTA 8	8	R	327	0	327							
7.	Phalguna/NHTA 8	13	R	430	32	462	15:1	0.360	0.70-0.50	21-28-01	7:8:1	1.840	0.20-0.10
8.	Abhaya/NHTA 8	13	R	597	39	636	15:1	0.015	0.90-0.80	25-23-02	7:8:1	1.011	0.50-0.30
9.	Samridhi/Riting	13	R	540	32	572	15:1	0.419	0.70-0.50	22-25-03	7:8:1	0.005	0.95-0.90
10.	Phalguna/Riting	13	R	596	44	640	15:1	0.426	0.70-0.50	18-28-04	7:8:1	1.291	0.30-0.20
11.	Abhaya/Riting	13	R	487	38	525	15:1	0.874	0.50-0.30	20-29-01	7:8:1	2.245	0.20-0.10
12.	Samridhi/MNP 76	13	R	349	32	381	15:1	3.002	0.10-0.05	21-25-04	7:8:1	0.280	0.70-0.50
13.	Abhaya/MNP 76	13	R	418	01	419							
14.	ARC 5984/MNP 76	13	R	434	39	473	15:1	3.213	0.10-0.05				
15.	Samridhi/AC 169	13	R	637	53	690	15:1	2.411	0.20-0.10	18-29-03	7:8:1	1.331	0.30-0.20
16.	Phalguna/AC 169	13	R	362	31	393	15:1	1.799	0.20-0.10				
17.	Abhaya/ AC 169	13	R	448	35	483	15:1	0.818	0.50-0.30	20-29-01	7:8:1	2.245	0.20-0.10
18.	ARC 5984/ AC 169	13	R	385	31	416	15:1	1.025	0.50-0.30	22-26-02	7:8:1	0.445	0.70-0.50
19.	Samridhi/ARC 6632	10	R	321	30	351	15:1	3.160	0.10-0.05	22-27-01	7:8:1	1.605	0.30-0.20
20.	Phalguna/ARC 6632	10	R	468	35	503	15:1	0.430	0.60-0.50	22-24-04	7:8:1	0.285	0.10-0.05
21.	ARC 5984/ARC 6632	10	R	390	03	393							
22.	Samridhi/AE 20	8	R	452	0	452							
23.	ARC 5984/AE 20	10	R	395	34	429	15:1	2.055	0.20-0.10				
24.	Samridhi/AC 424	13	R	389	18	407	15:1	2.318	0.20-0.10				
25.	Phalguna/AC 424	13	R	407	33	440	15:1	1.173	0.30-0.20				
26.	Abhaya/AC 424	13	R	421	35	456	15:1	1.615	0.20-0.10	22-24-04	7:8:1	0.285	0.70-0.50
27.	Samridhi/Calrose 70	13	R	362	33	395	15:1	2.985	0.10-0.05				
28.	Phalguna/Calrose 70	13	R	422	04	426							
29.	Abhaya/Calrose 70	8	R	273	28	310	15:1	4.786	0.05-0.02	24-24-02	7:8:1	0.651	0.50-0.30
30.	ARC 5984/Calrose 70	13	R	489	36	525	15:1	0.330	0.70-0.50				
31.	Samridhi/AC 630	13	R	361	19	380	15:1	1.013	0.50-0.30	22-24-04	7:8:1	0.285	0.70-0.50
32.	Phalguna/AC 630	13	R	352	33	385	15:1	3.540	0.10-0.05				
33.	Abhaya/AC 630	13	R	362	28	390	15:1	0.575	0.50-0.30	23-24-03	7:8:1	0.102	0.80-0.70
34.	Samridhi/ARC 15831	12	R	337	28	365	15:1	1.626	0.30-0.20				
35.	Abhaya/ARC 15831	13	R	308	3	311							
36.	Phalguna/MNP 709	13	R	465	36	501	15:1	0.748	0.50-0.30				
37.	Abhaya/MNP 709	8	R	569	3	572							
38.	ARC 5984/MNP 709	13	R	463	30	493	15:1	0.022	0.90-0.80				
39.	Samridhi/AC 8070	13	R	348	19	367	15:1	0.720	0.50-0.30				
40.	Abhaya/AC 8070	13	R	413	34	447	15:1	1.403	0.30-0.20	20-26-04	7:8:1	0.445	0.70-0.50
41.	Phalguna/AC 8070	13	R	541	21	562	15:1	2.170	0.20-0.10				
42.	Phalguna/Kataribhog	13	R	341	2	343							
43.	Abhaya/Kataribhog	13	R	430	36	466	15:1	1.731	0.20-0.10	15-30-04	7:8:1	2.405	0.20-0.10

R = Resistant, S = Susceptible & Seg. = Segregation

4.3. ALLELIC RELATIONSHIPS AMONG THE GENES FOR RESISTANCE PRESENT IN THE KNOWN RESISTANT VARIETIES AND IN TEST VARIETIES :

Allelic test is an important tool to identify the diverse sources of resistance. This test determines whether the gene(s) present in different varieties are the same or different. These could be helpful in breeding programme for developing new strains with diverse source of resistance. Screening results for allelic test are presented in Table - 6. The results of different test varieties are summerised below :

4.3.1. Balam :

Allelic relationship of gene(s) present in Balam was studied by crossing Balam with Samridhi and Phalguna possessing Gm - 1 and Gm -2 genes, respectively. The F_2 progenies of these crosses segregated in 15 R:l S ratio, indicating the segregation of two independent dominant genes. The F3 populations of these crosses segregated in a ratio of 7 resistant : 8 segregating : 1 Susceptible, indicating the presence of a non- allelic gene in Balam, other than Gm -1 and Gm -2.

4.3.2. AC 1224-2:

The F_2 of AC 1224-2 crossed with Phalguna and Abhaya segregated in a 15 R : 1 S ratio, indicating the presence of single dominant resistant gene in AC 1224-2. F_2 and F3 generations of Samridhi X AC 1224-2 did not show any segregation,

suggesting that resistant gene present in AC 1224-2 is allelic to Gm -1 gene present in Samridhi.

4.3.3. **NHTA - 8 :**

The F_2 and F_3 of Samridhi X NHTA -8 did not segregate. All the plants of F_2 and F3 showed resistant reaction against gall midge, indicating that the dominant resistant gene present in NHTA - 8 is allelic to Gm -1. The F_2 of Phalguna X NHTA -8 and Abhaya X NHTA -8 segregated in a 15 R : 1 S ratio, and in F3 7 resistant, : 8 segregating : 1 susceptible ratio was observed. This indicated that the dominant resistant gene possessed by NHTA -8 is not allelic to Gm -2 and Gm -4 genes present in Phalguna and Abhaya, respectively.

4.3.4. **Riting :**

Riting was crossed with Samridhi, Phalguna and Abhaya. The F_2 population of these three crosses segregated in 15 R : 1 S ratio, indicating that the gene conferring resistance in Riting was different and was non allelic to the gene present in Samridhi (Gm -1), Phalguna (Gm -2) and Abhaya (Gm -4). The F3 progenies segregated in 7 R : 8 Seg. : 1 S ratio, which again confirmed the segregation pattern of two independent dominant genes observed in F_2.

4.3.5. MNP -76 :

The F_2 populations of MNP-76 X Samridhi and MNP-76 X ARC 5984, segregated in 15 R : 1 S ratio. The F_3 progenies segregated in 7 R : 8 Seg. : 1 S ratio, suggesting the presence of an independent

dominant gene in MNP 76, which was non allelic to Gm -1 gene of Samridhi and Gm -5 gene of ARC 5984. However the F_2 of MNP-76 X Abhaya did not show any segregation for resistance. This indicated that MNP-76 possessed the same gene as present in Abhaya i.e. Gm -4 for resistance.

4.3.6. AC-169 :

In F_2 of crosses of AC-169 with Samridhi, Phalguna, Abhaya and ARC-5984 a 15 R : 1 S ratio was observed. The F_3 progenies segregated in a well fitted ratio of 7R : 8 Seg. : 1 S. This segregation pattern indicated that the gene for resistance present in AC -169 was different from Gm -1, Gm -2, Gm -4 and Gm -5 genes present, in Samridhi, Phalguna, Abhaya and ARC-5984, respectively. The results thus, indicated that AC -169 possessed a new gall midge resistant gene, not reported till now.

4.3.7. ARC-6632 :

In F_2 of crosses of ARC -6632 with Samridhi and Phalguna a ratio of 15 R : 1 S was observed and F3 progenies segregated in a ratio of 7R : 8 Seg. : 1 S, which is expected on the basis of segregation of two non allelic dominant genes.

ARC-6632, when crossed with ARC-5984, did not show any segregation in F_2 and F_3 indicating that the gene present in ARC-6632 is allelic to Gm -5 present in ARC -5984.

4.3.8. AE -20 :

AE -20 was crossed with Samridhi and ARC -5984. The F_2 of Samridhi X AE -20 did not segregate whereas ARC-5984 x AE -20 segregate in a ratio of 15 R : 1 S, indicating that the gene possessed by AE -20 is allelic to Gm -1 gene of Samridhi and is non-allelic to GM-5.

4.3.9. AC -424 :

The F_2 population of crosses of AC -424 with Samridhi, Phalguna and Abhaya showed a ratio of 15 R : 1 S, indicating that the gene present in AC -424 *is* different from Gm -1, Gm -2 and Gm -4. The F_3 of Abhaya x AC-424 gave a ratio of 7 R : 8 Seg. : 1 S, confirming the non-allelic nature of resistant gene present in AC-424 to that of Gm-4 gene of Abhaya.

4.3.10. Calrose -70 :

The F2 generations of Samridhi X Calrose -70 Abhaya X Calrose -70 and ARC -5984 X Calrose -70 segregated in a ratio of 15 R : 1 S, suggesting that Calrose -70 possessed a different gene than Gm -1, Gm -4 and Gm -5. The F3 progenies of Abhaya X Calrose -70 showed a ratio of 7 resistant : 8 segregating : 1 susceptible, confirming the presence of non-allelic gene to Gm -4.

In F_2 Calrose -70 X Phalguna did not show segregation and all the plants were resistant against gall midge, indicating the presence of an allelic gene Gm -2 possessed by the Phalguna.

4.3.11. **AC -630** :

The F_1's of AC -630 crossed with Samridhi, Phalguna and Abhaya were all resistant and the F_2 population segregated in a 15 R : 1. S ratio. This segregation pattern indicated the independent segregation of two dominant genes. Thus, the dominant gene present in AC -630 is different than that of Samridhi (Gm -1), Phalguna (Gm -2) and Abhaya (Gm -4). The non-allelic nature of resistance gene present in AC- 630 to the gene present in Abhaya (Gm-4) was also confirmed in F3, which segregated in a ratio of 7 R : 8 Seg. : 1 S.

4.3.12. ARC -15831 :

The F_2 of Samridhi X ARC -15831, segregated in a 15 R : 1 S ratio, indicating the presence of two non allelic independent dominant genes. Whereas, in cross Abhaya X ARC -15831 the F_2 and F3 did not segregate indicating the presence of allelic genes. This revealed that ARC -15831 possessed a dominant gene allelic to Gm -4, gene present in Abhaya.

4.3.13. **MNP -709** :

Phalguna X MNP -709 and ARC -5984 X MNP -709 showed a ratio of 15 R : 1 S ratio in F2, suggesting the presence of a dominant gene in MNP -709 which was non allelic to Gm -2 and Gm -5. F_2 and F_3 generations of Abhaya X MNP -709 did not show any segregation, indicating that MNP -709 possessed a gene Gm -4, as present in Abhaya.

4.3.14. **AC -8070** :

The F_2 's of the Samridhi X AC -8070, Phalguna X AC -8070 and Abhaya X AC -8070 segregated in 15 R : 1 S ratio. This indicated that the dominant resistant gene present in AC -8070, was different than the genes present in Samridhi (Gm -1), Phalguna (Gm -2) and Abhaya (Gm -4).The F_3 progenies of Abhaya X AC-8070 segre-gated in 7 R : 8 Seg. : 1 S ratio, which again confirmed the presence of non-allelic gene in AC-8070 than Gm-4 gene of Abhaya.

4.3.15. **Kataribhog :**

The F_2 of Kataribhog X Abhaya segregated in 15 R : 1 S ratio and the F_3 progenies segregated in 7 resistant : 8 segregating : 1 susceptible ratio, which was expected on the basis of segregation of two non-allelic dominant genes. On the other hand in Kataribhog X Phalguna, all the plants were resistant in F_2 generation, indicating that gene responsible for resistance in Kataribhog is allelic to Gm -2 gene present in Phalguna.

4.4. LINKAGE OF GALL MIDGE RESISTANT GENES WITH THE GENES CONTROLLING PURPLE LEAF CHARACTER IN RICE :

The results related to Linkage studies are presented in four parts :

4.4.1. Inheritance of gall midge resistance in crosses of test varieties with Shyamla.

4.4.2. Inheritance of purple leaf colour in Shyamla.

4.4.3. Joint segregation of purple leaf colour and gall midge resistance.

4.4.4. Estimation of linkage values.

4.4.1. Inheritance of gall midge resistance in crosses of test varieties with Shyamla :

All the crosses of Shyamla with fourteen resistant test varieties showed green leaf colour as well as resistant reaction against gall midge in F_1, However, no common segregation pattern was observed in F2 for gall midge resistance. The number of plants observed as resistant and susceptible in F_2 and true breeding resistant, segregating and true breeding susceptible in F3 are presented in Table - 7. The segregation fitted in two ratios i.e. 3 R : 1 S in 5 cross combinations and 51 R : 13 S in 8 crosses (Table - 7). In one cross Shyamla X Kataribhog no definite ratio could be fitted. F2 progenies of crosses of Shyamla with AC 1224-2, NHTA-8, Riting, AC-169 and ARC-15831 segregated in a 3 R : 1 S ratio, indicating that the test varieties differed in one dominant gene with Shyamla for resistance against gall midge.

The 3 R : 1 S segregation could be explained by the hypothesis, that the resistant parent has one dominant gene (Gm A) as well as one inhibitory gene (I-Gm B), whereas Shyamla has one

Table : 7 Gall midge reaction of F_1, F_2 and F_3 progenies of the crosses between test varieties and susceptible variety Shyamla.

S. No.	Cross	F_1 Reaction	Reaction against gall midge in F_2 (No. of Plant)						F_3			
			Resistant R	Susceptible S	Total	Expected Ratio	X^2 Value	P Value	Progenies reaction	Expected Ratio	X^2 Value	P Value
1	Shyamla X Balam	GR	476	125	601	51:13	0.0870	0.80-0.70	27-21-02	37:26:1	2.050	0.20-0.10
2	Shyamla X AC1224-2	GR	422	126	548	3:1	1.1770	0.30-0.20				
3	Shyamla X NHTA-8	GR	307	92	399	3:1	0.8020	0.50-0.30				
4	Shyamla X Riting	GR	580	174	754	3:1	1.4860	0.30-0.20				
5	Shyamla X MNP-76	GR	473	122	595	51:13	0.0130	0.95-0.90				
6	Shyamla X AC-169	GR	265	77	342	3:1	1.1260	0.30-0.20				
7	Shyamla X AE-20	GR	376	96	472	51:13	0.0010	0.95-0.90				
8	Shyamla X AC-424	GR	346	86	432	51:13	0.0430	0.90-0.80				
9	Shyamla X Calrose 70	GR	147	25	172	51:13	3.5470	0.10-0.05				
10	Shyamla X AC-630	GR	256	65	321	51:13	0.0007	0.98-0.95				
11	Shyamla X ARC-15831	GR	167	64	231	3:1	0.9140	0.50-0.30				
12	Shyamla X MNP-709	GR	436	110	546	51:13	0.0090	0.98-0.95				
13	Shyamla X AC-8070	GR	203	46	249	51:13	0.5190	0.50-0.30	26-23-01	37:26:1	2.708	0.50-0.30
14	Shyamla X Kataribhog	GR	362	64	426	3:1	22.6130	<0.001				
						13:3	3.8830	0.05-0.02				
						51:13	7.3610	<0.001				

GR - Green Ressistaant

dominant gene (Gm B) as well as one inhibitory gene (I-GmB). Due to this segregation of inhibitory gene did not occur. Thus, it resulted in a 3 R : 1 S ratio. The proposed genotypes of the parents are as under:

Shyamla (Susceptible) : gmagma GmBGmB IGmBIGmB

Test variety (Resistant) : GmAGmA gmbgmb IGmBIGmB

(Genotype) : GmAgma GmBgmb IGmBIGmB

(Phenotype) : Resistant

Genotypes			**Frequency**	**Reaction**
GmA -	GmB -	IGmBIGmB	**9**	**R**
GmA -	gmbgmb	IGmBIGmB	**3**	**R**
gmagma	GmB -	IGmBIGmB	**3**	*S*
gmagma	gmbgmb	IGmBIGmB	**1**	S

Phenotypic ratio : 3 R : 1 S

In F2 populations of crosses of Shyamla with Balam, MNP -76, AE-20, AC -424, Calrose -70 , AC -630, MNP -709 and AC -8070, a ratio of 51 resistant : 13 susceptible was observed which indicated the involvement of 3 gene pairs. The F3 progenies of Shyamla x Balam and Shyamla x AC -8070 segregated in 37 R : 26 seg : 1 S ratio, which again confirmed the involvement of three gene pairs. In this case, one dominant gene (Gm A) in the gall midge resistant test varieties and another dominant gene (Gm B) along with its inhibitory gene (IGm B) in Shyamla were assumed.

According to this hypothesis, it can be explained as follows

Shyamla (Susceptible) : gmagma GmBGmB IGmBIGmB
Test **variety** (Resistant) : GmAGmA gmbgmb igmbigmb •

F_1 (Genotype) : Gmagma GmBgmb IGmBigmb
(Phenotype) : Resistant

Genotypes			Frequency	Reaction
GmA _	GmB _	IGmB_	27	R
GmA _	GmB _	igmbigmb	9	R
GmA _	gmbgmb	IGmB_	9	R
gmagma	GmB _	IGmB_	9	S
GmA _.	gmbgmb	igmbigmb	3	R
gmagma	GmB _	igmbigmb	3	R
gmagma	gmbgmb	IGmB _	3	S
gmagma	gmbgmb	igmbigmb	1	S

Phenotypic ratio : 51 R : 13 S

4.4.2. Inheritance of purple leaf colour in Shyamla :

Inheritance of purple leaf colour was studied in fourteen crosses (Table - 8). Shyamla a purple leaf gall midge susceptible variety was used as female parent in each cross, whereas the test varieties, which were all green in colour as well as resistant to gall midge, were used as male parents. In all the fourteen crosses the leaf colour was green in F_1, indicating that the green colour was dominant over purple colour. Shyamla x NHTA 8 and Shyamla x MNP 76 showed a ratio of 13 G : 3 P in F2, while in the rest of the crosses a ratio of 55 green : 9 purple ratio was observed. The F_2 ratio of 13 green : 3 purple was due to the presence of dominant genes Pl and C in purple leaf variety. Shyamla and an inhibitory gene IP1 and a dominant gene C in

Table : 8 Segregation of purple leaf colour in F_1 and F_2 of crosses involving test varieties with Shyamla.

S. No.	Cross	F_1 Reaction	F_2 (No. of Plant)					
			Green G	Purple P	Total	Expected Ratio	X^2 Value	P Value
1	Shyamla X Balam	G	510	91	601	55G:9P	0.579	0.50-0.30
2	Shyamla X AC1224-2	G	460	88	548	55G:9P	1.806	0.20-0.10
3	Shyamla X NHTA-8	G	327	72	399	13G:3P	0.131	0.80-0.70
4	Shyamla X Riting	G	661	93	754	55G:9P	1.863	0.20-0.10
5	Shyamla X MNP-76	G	496	99	595	13G:3P	1.741	0.20-0.10
6	Shyamla X AC-169	G	288	54	342	55G:9P	0.843	0.50-0.30
7	Shyamla X AE-20	G	413	59	472	55G:9P	0.953	0.50-0.30
8	Shyamla X AC-424	G	371	61	432	55G:9P	0.062	0.90-0.80
9	Shyamla X Calrose 70	G	142	30	172	55G:9P	1.624	0.20-0.10
10	Shyamla X AC-630	G	279	42	321	55G:9P	0.253	0.70-0.50
11	Shyamla X ARC-15831	G	194	30	224	55G:9P	0.292	0.70-0.50
12	Shyamla X MNP-709	G	471	75	546	55G:9P	0.048	0.90-0.80
13	Shyamla X AC-8070	G	220	29	249	55G:9P	1.202	0.30-0.20
14	Shyamla X Kataribhog	G	360	66	426	55G:9P	0.72	0.50-0.30

G = Green, P = Purple

NHTA-8 and MNP-76, the varieties with green colour. The 55 green : 9 purple ratio was due to two complementary genes Pl and C in the purple leaf variety and an inhibitory gene IP1 in green variety. IP1 gene inhibits Pl gene. The inhibitory gene IP1 is present in all the green leaved gall midge resistant test varieties. The F_1 and F_2 phenotypes and genotypes of these crosses would therefore be as follows:

Parent :	**Shyamla**	**Test varieties (NHTA 8 and MNP 76)**
Genotype	CC P1P1 iplipl	CC plpl IP1IP1
Phenotype	(Purple leaf)	(Green leaf)

F1 Genotype	CC Plpl IPlipl
Phenotype	(Green leaf)

F2 Genotype			**Frequency**	**Phenotype**
CC	Pl	IP1_	9	Green
CC	Pl_	iplipl	3	Purple
CC	plpl	IP1_	3	Green
CC	plpl	iplipl	1	Green

Phenotypic ratio : 13 Green : 3 Purple

In the rest of the twelve crosses of Shyamla with Balam, AC 1224-2, Riting, AC-169, AE-20, AC-424, Calrose-70, AC-630, ARC-15831, MNP-709, AC-8070 and Kataribhog, the F2 population segregated in 55 G : 9 P ratio. This can be explained by assuming that purple leaf colour was governed by two complementary genes (C and Pl) and one inhibitory gene IPl, which inhibits the action of Pl

gene. The inhibitory gene IP1 is present in all the green leaved test varieties. The genotype and phenotype of parent, F1 and F2 would be therefore as follows:

Parent	**Shyamla**	**Test variety**
Genotype	CC P1P1 iplipl	cc plpl IP1IP1
Phenotype	Purple leaf	Green leaf

Fl Genotype	Cc Plpl IPlipl	
Phenotype	Green leaf	

F2 Genotype	**Frequency**	**Phenotype**
C_ PL IPl_	**27**	**G**
C_ PL iplipl	**9**	**P**
C_ plpl IP1_	**9**	**G**
cc PL IPL	**9**	**G**
C_ plpl iplipl	**3**	**G**
cc Pl_ iplipl	**3**	**G**
cc plpl IP1_	**3**	**G**
cc plpl iplipl	**1**	**G**

Phenotypic ratio : 55 Green(G) : 9 Purple (P)

4.4.3. Joint segregation of gall midge resistance and purple leaf colour :

The joint segregation of gall midge resistance and purple leaf colour was studied in crosses of Shyamla with Balam, AC 1224-2, NHTA-8, Riting, MNP-76, AC-169, AE-20, AC-424, Calrose-70, AC-630, ARC-15831, MNP-709, AC-8070 and Kataribhog. The existence of linkage between the genes governing gall midge resistance and purple leaf colour was revealed in three out of fourteen crosses. These crosses

Table : 9 Summary results of joint segregation of gall midge resistance and purple leaf colour.

S. No.	Cross	Segregation in F_2				X^2 for independence	Existence of linkage
		For gall midge resistant		For purple leaf colour			
		Ratio	X^2 Value	Ratio	X^2 Value		
1	Shyamla X Balam	51:R	0.0087	55G:9P	0.5789	6.471	L
2	Shyamla X AC1224-2	3R:1S	1.177	55G:9P	1.806	0.004	-
3	Shyamla X NHTA-8	3R:1S	0.802	13G:3P	0.131	0.015	-
4	Shyamla X Riting	3R:1S	1.486	55G:9P	1.863	0.014	-
5	Shyamla X MNP-76	51R:13S	0.013	13G:3P	1.741	6.995	L
6	Shyamla X AC-169	3R:1S	1.126	55G:9P	0.843	1.014	-
7	Shyamla X AE-20	51R:13S	0.001	55G:9P	0.953	1.914	-
8	Shyamla X AC-424	51R:13S	0.043	55G:9P	0.062	2.82	-
9	Shyamla X Calrose 70	51R:13S	3.547	55G:9P	1.624	14.32	L
10	Shyamla X AC-630	51R:13S	0.0007	55G:9P	0.253	0.379	-
11	Shyamla X ARC-15831	3R:1S	0.914	55G:9P	0.292	0.251	-
12	Shyamla X MNP-709	51R:13S	0.009	55G:9P	0.048	3.333	-
13	Shyamla X AC-8070	51R:13S	0.519	55G:9P	1.202	0.033	-
14	Shyamla X Kataribhog	3R:1S	22.613	55G:9P	0.72	-	-
		13R:3S	3.883				
		51R:13S	7.361				

X2 Value Significant at 5% level 3.841 and at 1 % 6.635
L - Linkage exist.

were Shyamla x Balam, Shyamla x MNP-76 and Shyamla X Calrose-70 (Table - 9 , Appendix - II).

4.4.4. **Estimation of linkage values :**

Linkage values in terms of cross over percentages were estimated by minimum discrepancy method as described under Materials and Methods.

In first group of crosses viz., Shyamla x Balam and Shyamla x Calrose-70, 51 R : 13 S ratio was observed in F2 for gall midge resistance and 55 G : 9 P for leaf colour. In the cross Shyamla x MNP 76 a ratio of 51 R : 13 S was observed in F2 for gall midge resistance and 13 G : 3 P for leaf colour.

4.4.4.1. Estimation of linkage values in first group of crosses:

Segregation in the ratio of 51 R : 13 S for gall midge resistance and 55 G : 9 P for leaf colour was observed in F_2 of two crosses of Shyamla with Balam and Calrose 70. As explained earlier, the 51 R : 13 S ratio could be obtained due to action of dominant gene GmA present in green resistant varieties and another dominant gene GmB along with its inhibitory gene IGmB present in purple susceptible variety "Shyamla". Similarly segregation for leaf colour was due to two complementary genes C and Pl and one inhibitory gene I-P1. Therefore, the genotypes of Shyamla and other two parents for leaf colour and gall midge resistance would be as follows :

Genotypes :

Green Resistant	GmAGmA	gmbgmb	igmbigmb	GM reaction
(Balam, Calrose-70)	cc	plpl	IP1IP1	Leaf colour
Purple Susceptible	gmagma	GmBGmB	IGmBIGmB	GM reaction
(Shyamla)	CC	P1P1	iplipl	Leaf colour

Phenotype	Green Resistant				
Genotype	Gm A	Gm B	IGm B	C	Pl IP1
	gma	gm b	igm b	c	pl ipl

The possible linkages are :

i) GmA_ IP1
ii) GmA_ pl or c
iii) Pl or C_ I Gm B
iv) Gm B_ ipl
v) I Gm B_ ipl
vi) Gm B_ Pl or C

Linkage values were estimated by minimum discrepancy method (Appendix - III). Following functions of each linkage were obtained :

i) GmA - IP1

$(672 + 117\ P)^2$ $(208 - 117\ P)^2$ $(144 - 117\ P)^2$ $(117)^2$

n_1 n_2 n_3 n_4

ii) GmA - pl or c

$(711 - 39\ P)^2$ $(169 + 39\ P)^2$ $(105 + 39\ P)^2$ $(39 - 39\ P)^2$.

n_3 n_4

iii) **Pl - IGm B**

$$\frac{(695 + 9P)^2}{n_1} + \frac{(181 - 9P)^2}{n_2} + \frac{(117 - 9P)^2}{n_3} + \frac{(27 + 9P)^2}{n_4}$$

iv) GmB - ipl

$$\frac{(699 + .9P)^2}{n_1} + \frac{(181 - 9P)^2}{n_2} + \frac{(117 - 9P)^2}{n_3} + \frac{(27 - 9P)^2}{n_4}$$

v) **IGmB** - ipl

$$\frac{(708 + 27P)^2}{n_1} + \frac{(172 - 27P)^2}{n_2} + \frac{(108 + 27P)^2}{n_3} + \frac{(36 - 27P)^2}{n_4}$$

vi) GmB - Pl

$$\frac{(666 - 3P)^2}{n_1} + \frac{(214 + 3P)^2}{n_2} + \frac{(114 + 3P)^2}{n_3} + \frac{(30 - 3P)^2}{n_4}$$

As described in the "Estimation of linkage" under Materials and Methods, differentiating the function for above mentioned (i) to (vi) linkage combinations with respect to P (where P = p2 and p =C.O value) and then solving for `P' after equating them to zero ,the values of P for each type of linkage was worked out as follows:

i) GmA - IP1

$$P = \frac{-5.7435\, n_2n_3n_4 + 1.7435\, n_1n_3n_4 + 1.2307\, n_1n_2n_4}{n_2n_3n_4 + n_1n_3n_4 + n_1n_2n_4 + n_1n_2n_3}$$

ii) GmA - **pl** or c

$$P = \frac{18.2307\, n_2n_3n_4 - 4.3334\, n_1n_3n_4 - 2.6923\, n_1n_2n_4 +}{n_2n_3n_4 + n_1n_3n_4}$$

iii) Pl - IGm B

$$P = \frac{-77.6667\, n_2n_3n_4 + 20.1112\, n_1n_3n_4 + 13\, n_1n_2n_4 - 3\, n_1n_2n_3}{n_2n_3n_4 + n_1n_3n_4 + n_1n_2n_4 + n_1n_2n_3}$$

iv) GmB - ipl

$$P = \frac{-77.6667\, n_2n_3n_4 + 20.1112\, n_1n_3n_4 + 13\, n_1n_2n_4 - 3\, n_1n_2n_3}{n_2n_3n_4 + n_1n_3n_4 + n_1n_2n_4 + n_1n_2n_3}$$

v) IGmB - ipl

$$P = \frac{26.2223\, n_2n_3n_4 - 6.3703\, n_1n_3n_4 - 4\, n_1n_2n_4 + 1.3334\, n_1n_2n_3}{n_2n_3n_4 + n_1n_3n_4 + n_1n_2n_4 + n_1n_2n_3}$$

vi) GmB - Pl

$$P = \frac{222\, n_2n_3n_4 - 71.3334\, n_1n_3n_4 - 38\, n_1n_2n_4 + 10\, n_1n_2n_3}{n_2n_3n_4 + n_1n_3n_4 + n_1n_2n_4 + n_1n_2n_3}$$

By substituting the values of n_1, n_2, n_3 and n_4 the P and cross over values were computed for different crosses and are presented in Table - 10.

Table - 10 : P and cross over values for different possible linkages in first group of crosses.

Cross	P and C.O. values for possible linkage					
	GmA-Ipl	GmA-pl	Pl-IGmB	GmB-ipl	IGmB-ipl	GmB-Pl
Shyamla/Balam						
P	0.3690	-0.1238	1.8698	1.8698	-0.2899	-7.0237
C.O.	0.3925	-	-0.3674	1.3674	-	-
Shyamla/Calrose 70						
P	0.6413	-0.9570	5.4809	5.4809	-1.4935	-19.7413
C.O.	0.1991	-	-1.3411	2.3411	-	-

In both the crosses, the C.O. values were under acceptable Limit for only GmA - IP1 Linkage. The crossing over values for other linkages were either negative or more than 50 per cent, indicating the absence of linkage among other genes. The results, thus, showed that the gall midge resistant genes of Balam and MNP 709 were Linked with inhibitory gene IP1 with a cross over value of 39 per cent, while that of Calrose 70 is linked with IP1, with a cross over value of 19 per cent.

4.4.4.2. Estimation of linkage in Shyamla x MNP 76

In this cross F_2 showed a ratio of 51 R : 13 S for gall midge resistance and 13 G : 3 P for leaf colour. The genotypes and phenotypes of parents F_1 and F2 would be as follows :

	Genotypes :	
Green Resistant	GmAGmA gmbgmb igmbigmb	**GM reaction**
(MNP-76)	CC plpl IP1IP1	**Leaf colour**
Purple Susceptible	gmagma GmBGmB IGmBIGmB	**GM reaction**
(Shyamla)	CC P1P1 iplipl	**Leaf colour**

Phenotype	Green Resistant				
Genotype	Gm A	Gm B	IGm B	C	Pl IP1
	gma	gm b	igm b	C	pl ipl

for six possible linkages are presented in Appendix - IV. The functions derived due to different possible linkages were as follows :

i) **GmA** - IP1

$(156+ 39\ P)^2$ $(52 - 39\ P)^2$ $(48 - 39\ P)^2$ $(39\ P)^2$

n_1 n_2 n_3 n_4

ii) GmA - **pl**

$(169 - 13\ P)^2$ $(39+ 13\ P)^2$ $(35 + 13\ P)^2$ $(13 - 13\ P)^2$

n_1 n_2 n_3 n_4

iii) Pl - IGm B

$(165 + 3\ P)^2$ $(43 - 3\ P)^2$ $(39 - 3\ P)^2$ $(9 + 3\ P)^2$

n_3 n_4

iv) GmB - **ipl**

$(165 + 3\ P)^2$ $(43 - 3\ P)^2$ $(39 - 3\ P)^2$ $(9 + 3\ P)^2$

n_1 n_2 n_3 n_4

v) **IGmB** - ipl

$(168 - 9\ P)^2$ $(40+ 9\ P)^2$ $(36 + 9\ P)^2$ $(12 - 9\ P)^2$

n_2 n_3 n_4

vi) GmB - **Pl**

$(166 - P)^2$ $(42 + 3\ P)^2$ $(38 + 3\ P)^2$ $(10 - P)^2$

n_1 n_2 n_3 n_4

Differentiating these function and equating them to zero the P values for each linkage thus computed were as follows :

i) GmA - IP1

$$P = \frac{1.2307}{n_2n_3n_4 + n_1n_3n_4}$$

ii) GmA - pl

$$P = \frac{13\, n_2n_3n_4 - 3\, n_1n_3n_4 - 2.6923\, n_1n_2n_4 + }{n_2n_3n_4 + n_1n_3n_4 + n_1n_2n_4 + n_1n_2n_3}$$

iii) Pl - IGm B

$$P = \frac{-55\, n_2n_3n_4 + 14.3334\, n_1n_3n_4 + 13\, n_1n_2n_4 - 3}{n_2n_3n_4 + n_1n_3n_4 + n_1n_2n_4 + n_1n_2n_3}$$

iv) GmB - ipl

$$P = \frac{-55\, n_2n_3n_4 + 14.3334\, n_1n_3n_4 + 13\, n_1n_2n_4 - 3}{n_2n_3n_4 + n_1n_3n_4 + n_1n_2n_4 + n_1n_2n_3}$$

v) IGmB - ipl

$$P = \frac{-18.6667\, n_2n_3n_4 + 4.4445\, n_1n_3n_4 + 4\, n_1n_2n_4 - 1.3334}{n_2n_3n_4 + n_1n_3n_4 + n_1n_2n_4 + n_1n_2n_3}$$

vi) GmB - **Pl**

$$P = \frac{-166\, n_2n_3n_4 + 42\, n_1n_3n_4 + 38\, n_1n_2n_4 - 10\, n_1n_2n_3}{n_2n_3n_4 + n_1n_3n_4 + n_1n_2n_4 + n_1n_2n_3}$$

Table - 11: P and *cross* over values for different possible linkages in second group of crosses.

Cross	P and C.O. values for possible linkage ..... -					
	GmA-Ipl	GmA-pl	Pl-IGmB	GmB-ipl	IGmB-ipl	GmB-Pl
Shyamla/ MNP 76						
P	0.3688	-0.1064	1.7946	1.7946	0.2648	4.3840
C.O.	0.3926	-	-0.3396	1.3396	0.5146	-1.0938

In this cross again the cross over values were under acceptable limits for only GmA-IPl linkage. For other linkages these values were either negative or more than 50 per cent, suggesting the independent assortment of genes. The results thus indicated that the gall midge resistant gene of MNP 76 was linked with IP1 gene with a cross over value of 0.39 (Table - 11).

The linkage observed in 3 crosses indicated that only IP1 gene responsible for inhibition of purple leaf colour was linked with the independent dominant gene for the gall midge resistance. The following linkage values were obtained in different crosses :

Cross	C.O.value
1. Shyamla/Balam	0.39
2. Shyamla/MNP 76	0.39
3. Shyamla/Calrose 70	0.19

Chapter - 5

CHAPTER - V

DISCUSSION

Gall midge *(Orseolia oryzae* Wood Mason) a major destructive pest of rice crop has been reported to cause an seasonal global loss of more than US $ 550 million (Herdt, 1991). The maggots of gall midge feed internally in the growing meristems of the tillers and therefore the chemical control has not been found to be very effective against this insect. Its management by resistant host plants was found as the most effective approach (Khush, 1977; Heinrich and Pathak, 1981). Considerable progress has been made in developing resistant cultivars. Original donors, being land races, are poor in yield. The resistant genes present in donors are needed to be transferred in otherwise agronomically desirable lines. The knowledge of mode of inheritance of the gene(s), conferring resistance in the Lines, thus, is a prerequisite for their effective utilization in a breeding programme.

The existence of variability (biotypes) in gall midge has further complicated the situation. the occurrence of biotypes in gall midge was first suspected by Shastry *et al.* (1972).

A review of the literature indicated the presence of at least six biotypes in India, four in China, two in Srilanka and one each in Thailand and Indonesia (Anonymous, 1981, 1991; Shaw *et al.*, 1981; Lai *et al*, 1984; Prakasa Rao and Kittur, 1989; Kalode and Bentur, 1989; Kudagamage and GunaWardena, 1989; Nair and Ambika Devi, 1994 and Singh, 1996).

The presence of different biotypes is adjudged from the differential reaction of a defined set of crop varieties (differentials) to pest population over a number of locations at a regional, state or country level. The existence of biotypes must be taken in to account by the breeder while evolving the cultivars for gall midge endemic region (Katiyar *et al.*, 1995).

In order to deal with the variability in the insect it is necessary that a constant search is made for new sources of resistance so that they can be incorporated, when resistance provided by earlier ones is broken down by the new biotype (s) of the pest or to use the new gene(s) in some other manner, like pyramiding one over another and as a component in multilines. This information is precisely obtained by allelic tests.

Chaudhary *et al.* (1985) identified two dominant genes Gm-1 (in Samridhi, a derivative of W 1263) and Gm-2 (in Surekha.

derivative of Siam 29). This was further confirmed by Mishra (1985) and Chaudhary (1988). Sahu *et al.* (1990) reported a recessive gene being responsible for resistance in ARC 5984 and designated it as "gm-3". On the other hand, Shrivastava *et al.* (1995) found that the gene conferring resistance in ARC 5984 was dominant and designated it as `Gm-5' and the recessive gene of RP 2068-18-2-5 was designated as `gm-3'. Shrivastava *et al.* (1993) also identified `Gm-4', a dominant resistant gene present in cultivar `Abhaya'. This was further confirmed by Shrivastava *et al.* (1995).

The present study was under taken to identify the gene(s) for resistance present in some rice genotypes and also to identify new genes, if any, through allelic test. Twenty-one parents, seventy-two F_1 and F2 populations and available F3 populations were screened for their reaction against gall midge under natural infestation. The hundred per cent hill damage was observed in susceptible varieties (MW-10 and Shyamla) on plant basis (Table -2), indicating that the gall midge population was adequate to undertake genetic studies. All the fifteen test varieties viz., Balam, AC 1224-2, NHTA-8, Riting, MNP-76, AC-169, ARC-6632, AE-20, AC-424, Calrose-70, AC-630, ARC-15831, MNP 709, AC-8070 and Kataribhog, were observed to be resistant against gall midge biotype-1 existing in Raipur.

6.1. INHERITANCE OF GALL MIDGE RESISTANCE :

The F_1s of the crosses of fifteen test varieties with the susceptible parent MW-10 were resistant and segregated in a ratio of 3R :1 S, clearly indicating that the test varieties possessed a single dominant gene, responsible for gall midge resistance. Presence of single dominant gene for resistance had also been reported by Chaudhary *et al.* (1985), Mishra (1990), Tomar and Prasad (1992), Arvind Kumar (1992), Dharmendra Kumar (1992), Shrivastava *et al.* (1993), Shrivastava *et al.* (1995) and Bose (1996).

However, Mishra (1985) suspected that the resistance in variety 'Surekha' was either due to single dominant gene or one dominant and one recessive genes. The recessive gene, gm_3 governing resistance in variety RP 2068-18-3-5 was reported by Shrivastava *et al.* (1995).

5.2. ALLELIC TEST :

Allelic test helps to determine whether the resistant gene(s) present in different test varieties are the same a different. It also helps in identifying the new genes conferring resistance. In view of the known existence of different biotypes in gall midge in different parts of the country, this study is of great significance. It is likely to ensure the availability of alternate sources of resistance, in case of

breakdbwn of resistance due to sudden change in biotypes. Incorporation of more than one gene would help in avoiding the possibilities of breaking of resistance and would ensure stable production.

The inheritance and allelic studies, in relation to gall midge resistant gene(s) were initiated at Indira Gandhi Agricultural University, Raipur in 1982. Chaudhary *et al.* (1985) reported that genes for resistance present in Samridhi and Surekha were non-allelic and designated them as Gm-1 and Gm-2, respectively. Mishra (1990) reported the presence of Gm-1 gene for resistance in WR 35-8-1 and RP 2068-16-4-5 and Gm-2 in R 243-3041-1 and ARC 6619. He also reported that the dominant gene in Asha mutant-4, R 296-421-2 and the recessive gene of ARC 5984 were non allelic and were different from Gm-1 and Gm-2. Sahu (1991) further confirmed the presence of Gm-1 gene in Samridhi and also reported presence of Gm-2 gene in Bahawarkanak. Presence of Gm-2 gene was also reported by Dharmendra Kumar (1992) in R 435-107, R 435-756 and R 670. Shrivastava *et al.* (1993) observed that resistance conferring gene of Abhaya was non allelic to Gm-1 and Gm-2 and designated it as Gm-4. Shrivastava *et al.* (1995) reported that recessive gene for resistance in RP 2068-18-3-5 and dominant gene of ARC-5984 segregated independently of Gm-1, Gm-2 and Gm-4 and designated them as gm-3 and Gm-5,

respectively. Bose (1996) reported that CR 410-3225 and CR 392-5083-2 possessed Gm-2 gene for resistance.

For allelic test fifteen test varieties were crossed with four gall midge resistant varieties Samridhi, Phalguna, Abhaya and ARC 5984 which are known to possess Gm-1, Gm-2, Gm-4 and Gm-5 genes, respectively.

The results indicated that Gm-1 was present in test varieties AC 1224-2, NHTA-S and AE-20; Gm-2 in Calrose-70 and Kataribhog; Gm-4 in MNP-76, MNP-709 and ARC 15831 and Gm-5 was present in ARC 6632. There was an indication of the presence of a new dominant gene in AC-169,as the gene present in this variety was observed to be non allelic to all the genes reported so far except gm-3, which was not included in this study. Since, the gene present in AC-169 showed dominant action for resistance, it cannot be allelic to gm-3, which is a recessive resistant gene. It can be therefore inferred that AC-169 possessed a different dominant gene for resistance to gall midge. These results indicated that if a large number of resistant cultivars were tested, more and more new genes responsible for gall midge resistance could be discovered, widening the range of choice for producing multilines varieties, or for gene pyramiding. Summary of allelic test is presented in Table - 12.

Table - 12 : Summary of allelic tests.

SN.	Test varieties	Allelic nature of resistant genes to Gm-1	Gm-2	Gm-4	Gm-5	New	Resistant gene present
1.	Balam	NA	NA	--	--	--	
2.	AC 1224-2	A	NA	NA	--		Gm-1
3.	NHTA-8	A	NA	NA	--		Gm-1
4.	Kiting	NA	NA	NA	--	--	
5.	MNP-76	NA	--	A	NA		Gm-4
6.	AC-169	NA	NA	NA	NA	New	New
7.	ARC-6632	NA	NA	--	A		Gm-5
8.	AE-20	A	--	--	NA		Gm-1
9.	AC-424	NA	NA	NA	--	--	
10.	Calrose-70	NA	A	NA	NA		Gm-2
11.	AC-630	NA	NA	NA	--	--	
12.	ARC-15831	NA	--	A	--		Gm-4
13.	MNP-709	--	NA	A	NA		Gm-4
14.	AC-8070	NA	NA	NA	--	--	
15.	Kataribhog	--	A	NA	--		Gm-2

A - Allelic , NA - Non allelic

5.3. INHERITANCE OF GALL MIDGE RESISTANCE IN CROSSES OF TEST VERITIES WITH SHYAMLA :

No common segregation pattern was observed among crosses of 14 test varieties with Shyamla. Out of 14 crosses, five showed monogenic dominant ratio of 3 R:1 S; eight 51 R:13 S while in one cross, Shyamla X Kataribhog, the observed F_2 ratio did not fit well with of the ratios, 3 R:l S, 13 R:3 S and 51 R:13 S, based on the segregation of 1, 2 and 3 genes, respectively.

F_2 population of crosses of Shyamla with AC 1224-2, NHTA-8, Riting. AC-169 and and ARC-15831 segregated in 3 R:l S ratio, indicating the presence of a single dominant gene responsible for resistance against gall midge in these test varieties.

. These results are in conformity to those obtained from the crosses of these varieties with MW-10, the susceptible variety. The allelic studies indicated that the single dominant gene of AC 1224-2 and NHTA-8 is Gm-1, since these test varieties were observed to be allelic to Samridhi, which is known to possess Gm-1 gene (Chaudhary *et al.*, 1985) ARC 15381 showed allelic reaction with Abhaya and therefore it could be concluded that this genotype also had Gm-4 gene similar to that of Abhaya (Shrivastava *et al.*, 1993). The resistant gene present in Riting could not be determined, since it showed non allelic relationship with Samridhi, Phalguna and Abhaya the cross with ARC 5984 was not available. AC-169 did not show allelic relationship with any of the varieties possessed known resistant gene Gm-1 to Gm-5 and therefore presence of a new gene was indicated. Several earlier workers(Chaudhary *et al.*, 1985 ; Mishra, 1985 and Chaudhary, 1988) also reported monogenic segregation for gall midge resistance.

Shyamla had been reported to possess one dominant gene for gall midge resistance along with an inhibitory gene. The susceptibility of variety Shyamla is due to the inhibitory gene, although

it possess a dominant gene for resistance. The inhibitory gene interaction was observed in previous studies also (Mishra, 1985; Chaudhary, 1988; Arvind Kumar, 1992 and Bose, 1996).

The observations of 51 R : 13 S ratio in some crosses indicated 3 gene pair interaction. The 3 R : 1 S ratio, in a 3 gene system could be explained by the hypothesis, that the resistant parent has one dominant gene (Gm-A) as well as one inhibitory gene (I-Gm B), whereas, Shyamla has one dominant gene (Gm-B), as well as one inhibitory gene (I-Gm B). Due to presence of same inhibitory gene in both the parents, segregation of inhibitory gene did not occur. Thus it produced a 3 R : 1 S ratio.

The F_2 populations of crosses of Shyamla with Balam, MNP-76, AE-20, AC-424, Calrose-70, AC-630, MNP-709 and AC-8070 showed a ratio of 51 R : 13 S. This ratio could result from the segregation of 3 genes. Shrivastava (1992) reported that the Shyamla possessed one dominant gene for resistance and one dominant inhibitory gene, resulting into susceptibility of Shyamla to gall midge. The test varieties possessed one dominant gene (Gm A), for resistance. The observed ratio of 51 R : 13 S resulted from the segregation of two dominant genes for resistance and one dominant inhibitory gene.

It is therefore, obvious that by following appropriate breeding process the inhibitory gene of Shyamla can be deleted and a gall midge resistant purple leaf variety can be evolved, which is needed in view the wide scale occurrence of wild rice in Chhattisgarh region.

5.4. INHERITANCE OF PURPLE LEAF COLOUR :

For working out the association of gall midge resistance and purple leaf colour governing genes, it is necessary to understand the genetics of purple leaf colour. Purple leaf variety Shyamla, when crossed with green leaf varieties gave all green plants in F_1 in all the fourteen cross. Two crosses viz., Shyamla x NHTA-8 and Shyamla x MNP-76, shared a segregation of 13 green : 3 purple ratio in F2 * indicating the presence of a dominant gene `P1' responsible for producing purple anthocyanin colour in leaf lamina and one inhibitory gene I-P1 which inhibits the action of P1 gene.

In remaining twelve crosses of Shyamla with Balam AC 1224-2, Riting, AC-169, AE-20, AC-424, Calrose-70, AC-630, ARC-15831, MNP-709, AC8070 and Kataribhog the F2 population segregated in 55 green : 9 purple ratio, suggesting the involvement of 3 gene pairs in the expression of leaf pigmentation. The observed ratio of 55 G : 9 P could be expected on the basis of segregation of two dominant genes for pigmentation and one of its inhibitory gene. This result are in agreement with those reported earlier in the literature

on the inheritance of anthocyanin pigment (Nagao, 1951; Chandraratha, 1964; Takahashi, 1982 and Shrivatava, 1992). The gene `C' produces Chromogen forms anthocyanin pigment, while `Pl' gene spreads the anthocyanin pigments in leaf blade (Nagao, 1951; Dhulappanavar, 1973). The purple leaf variety Shyamla, therefore, possessed two complementary genes C and Pl while the inhibitory gene I-Pl is present in all the green test varieties under study.

5.5. ESTIMATION OF LINKAGE VALUES :

The joint segregation of purple leaf colour and gall midge resistance was studied in crosses of fourteen test varieties with Shyamla. The test of independence (Appendix - II) revealed that in three out of 14 crosses there was association between leaf pigmentation and gall midge resistance, whereas remaining 11 crosses no such dependence was observed. The pertaining of total X^2 into different components viz., X^2 for segregation of pigmentation, X^2 for segregation of gall midge resistance and X^2 for linkages, revealed the existence of linkage only in 3 crosses viz., Shyamla x Balam, Shyamla x MNP-76 and Shyamla x Calrose-70. In Shyamla x Balam and Shyamla x MNP-76 the cross over value was observed to be 0.39 whereas, in Shyamla x Calrose-70 it was 0.19. MNP-76 through allelic test was found to posses Gm-4 gene. This gene has already been reported to be linked with IP1 gene with a cross over value of 0.35 (Shrivastava, 1992), which is very

close to the 0.39 cross over value obtained in the present study. The gall midge resistant gene of Balam could not be ascertained from the allelic test, because of the non availability of its crosses with Abhaya (Gm-4) and ARC 5984 (Gm-5). However since the resistant gene of this test entry showed linkage with IP1 gene with a cross over value of a 0.39, it could be presumed to possess Gm-4. However it had to be confirmed by crossing it with Abhaya or any other variety possessing Gm4 gene. Allelic tests revealed that Calrose-70 possessed Gm-2 gene for gall midge resistance. It was observed to the linked with IP1 with a cross over value of 0.19.

Chapter - 6

CHAPTER - VI

SUMMARY, CONCLUSION AND SUGGESTIONS

The present studies were under taken during *kharif* 1994, 1995 and 1996 and *rabi* 1994 and 1995, in the department of Plant Breeding and Genetics, Indira Gandhi Agricultural University, Raipur (M.P.) with the following objectives :

1. To study the inheritance of gall midge resistance in some rice varieties.
2. To study allelic relationship of genes present in the test varieties with those carried by 4 known resistant varieties.
3. To study the linkage relationship of gall midge resistance with leaf pigmentation.

The experimental material consisted of 21 parents, 72 F_1 and F_2 populations and F3 progenies. Crosses were made during *kharif* 1994 and 1995. The F_1's were grown in *rabi* 1994 and 1995 to advance the generation. For screening against gall midge, the sowing of the parents, F_1, F_2 and F3 progenies were delayed till first week of August to synchronise with the build up of adequate pest population during active tillering phase of the crop. Heavy doses of nitrogen were applied to enhance the tillering, succulency and susceptibility of the crop plant against the gall midge. Artificial light was provided *during* night to attract the insect at the time of active tillering phase.

The F_1's were harvested individually. The F_2 populations were grown family wise (produce of single F_1 plant) in paired rows. The plant to plant distance of 15 cm and row to row distance of 20 cm were maintained. Fifty random F_2 plants were harvested individually to raise F3 generation. The progenies of a single plant harvested in F_2 were grown in a row. Each cross was surrounded by two row of purple leaf, gall midge susceptible variety, Shyamla to spread the insect population throughout the field. Individual plant were scored for susceptibility and the presence of single silver shoot per hill was taken as a criterion of susceptibility.

The X test was applied to test the goodness of fit, so as to derive appropriate result relating to mode of inheritance, and for detection of linkage between the genes governing purple leaf colour and gall midge resistance. Minimum discrepancy method was applied for the estimation of linkage values.

Based on the results obtained the following conclusions could be drawn :

1. The crosses of test varieties with susceptible variety MW-10 revealed that each variety viz., Balam, AC 1224-2, NHTA-8, Riting, MNP-76, AC-169, ARC-6632, AE-20, AC-424, Calrose-70, AC-630, ARC-15831, MNP-709, AC-8070 and Kataribhog, possessed a single dominant gene for gall midge resistance.

2. The segregating populations of test varieties and Shyamla showed interaction of three pairs of genes , one dominant gene present in test variety and another dominant gene responsible for gall midge resistance along with an inhibitory gene, which inhibited the resistant gene present in Shyamla.

3. Test varieties AC 1224-2, NHTA-8, and AE-20 possessed `GM-1' gene for gall midge resistance.

4. Test varieties Calrose-70 and Kataribhog possessed `Gm-2' gene for gall midge resistance.

5. Test varieties MNP-76, ARC-15831 and MNP-709 possessed `GM-4' gene for gall midge resistance.

6. Test variety ARC-6632 possessed `GM-5' gene for resistance against gall midge.

7. Test variety AC-169 possessed a new gene, other than known genes (Gm-1, Gm-2, gm-3, Gm-4 and Gm-5) for resistance against gall midge. It needs to be confirmed.

8. Segregation pattern in F_2 of crosses of pigmented variety, Shyamla with different green test varieties viz., Balam, AC 1224-2, NHTA-8, Riting, MNP-76, AC-169, ARC-6632, AE-20, AC-424, Calrose-70, AC-630, ARC-15831, MNP-709, AC-8070 and Kataribhog revealed that three genes C, Pl and IP1 were responsible for inheritance of purple leaf colour. Out of these

three genes two complementary gene C and Pl were present in pigmented variety Shyamla, whereas an inhibitory gene IPl was present in green leaf varieties.

9. In crosses of Shyamla with Balam, MNP-76 and Calrose-70 existence of a linkage between the genes for gall midge resistance and purple leaf colour was revealed.

10. In above crosses linkages were found between gall midge resistant gene's (Gm-2 or Gm-4) and the gene responsible for inhibiting the purple leaf colour (IPl) pigmentation.

11. In crosses Shyamla x Balam and Shyamla x MNP-76 (GM-4) a cross over value 0.39 was obtained which indicated that gene IPl and Gm-4 are located at a distance of 39 per cent map unit.

12. Since the cross over values of Balam and MNP-76 were the same i.e. 0.39 and MNP-76 possessed Gm-4 gene for gall midge resistance, the gall midge resistant gene present in Balam could also be presumed to be Gm-4.

13. In cross, Shyamla x Calrose-70 (Gm-2) the cross over value of 0.19 was obtained, which indicated the Gm-2 gene was located at 19 per cent map unit distance with respect to IPl gene.

SUGGESTIONS FOR FURTHER WORK :

The results of allelic studies revealed that variety AC-169 possessed a dominant gene for resistance against gall midge, which was non allelic to all the known resistant genes, used in this study. Therefore, the resistant gene of AC-169 appeared to be a new resistant gene. In India six biotypes of gall midge have been reported so far. The variety AC-169 should be tested against all these biotypes to establish that it is a new gene.

Present study revealed that Shyamla possessed a dominant gene for resistance, but due to presence of its inhibitory gene, it showed susceptible reaction. From segregating population, plants without inhibitory gene could be isolated enabling the evaluation of a gall midge resistant purple coloured variety.

In present study all the test varieties had single dominant gene for gall midge resistance. Degree of break down of resistance governed by oligogene or vertical resistance is very common. Therefore, it is necessary to transfer and combine as many genes as possible in a variety to overcome the problem of resistance being broken down.

It is necessary to screen local germ plasm, traditional varieties and wild relatives to search out further new genes for resistance. By designing appropriate experiments, the presence of non-allelic genes could be identified and the genetic nature of these donors be investigated.

INHERITANCE AND ALLELIC RELATIONSHIP OF GENES CONTROLLING RESISTANCE TO GALL MIDGE (*Orseolia oryze* Wood Mason) IN SOME RICE CULTIVARS

ABSTRACT

The present study was conducted in the Department of Plant Breeding and Genetics, Indira Gandhi Agricultural University, Raipur (M.P.) during *kharif* 1994, 1995 and 1996 and *rabi* 1995 and 1996 with the objectives to study the mode of inheritance of resistance; to establish the allelic relationship among the genes conferring resistance in selected genotypes and to study the linkage relationship between gall midge resistance and leaf pigmentation.

The experimental material consisting of 15 test varieties, 4 resistant varieties possessing known genes for resistance and two susceptible varieties.

The screening test revealed that all the 15 test varieties were resistant against gall midge. The remaining six varieties gave the expected reaction. 4 known resistant lines showing resistance whereas the remaining two varieties were susceptible.

Inheritance study revealed the presence of single dominant gene for gall midge resistance in each of 15 test varieties, viz., Balam, AC 1224-2, NHTA-8, Riting, MNP-76, AC-169, ARC-6632, AE-20, AC-424, Calrose-70, AC-630, ARC-15831, MNP-709, AC-8070 and Kataribhog.

Allelic studies revealed that the gene governing resistance in test varieties AC 1224-2, NHTA-8 and AE-20 was `Gm-1', in Calrose-70 and Kataribhog `Gm-2', in MNP-76, MNP-709 and ARC 15831 `Gm-4' and in test variety ARC-6632 it was `Gm-5' gene. There were indications that test variety `AC-169' possessed a now

dominant gene for gall midge resistance which was different from so far reported genes Gm-1, Gm-2, gm-3, Gm-4 and Gm-5.

Crosses of Shyamla with different test varieties revealed that Shyamla possessed a dominant gene for gall midge resistance, along with an inhibitory gene, which resulted in the susceptible reaction of Shyamla.

The study revealed that the inheritance of pigmentation in Shyamla was governed by two complementary gene's C and Pl, while an inhibitory gene IP1 was present in green leaf varieties.

In three crosses Shyamla x Balam, Shyamla x MNP-76 and Shyamla x Calrose-70, existence of linkage between gall midge resistant gene's (Gm-2 and Gm-4) and purple colour inhibitory gene (IP1) was observed.

Crosses Shyamla x Balam (Gm-4) and Shyamla x MNP-76 (Gm-4) showed a cross over value of 0.39 whereas in cross Shyamla x Calrose-70 (Gm-2) a cross over value of 0.19 was estimated in respect of IP1 gene.

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Appendices

APPENDIX - I

Monthly meteorological data (rainfall & relative humidity) during *kharif* 1994, 1995 and 1996

	Rainfall (mm)			RH (%)					
	1994	1995	1996	1994		1995		1996	
				I	II	I	II	I	II
August	336.2	352.6	473.9	94	80	93	76	93	78
September	206.6	108.6	198.0	93	72	91	70	93	70
October	39.2	28.4	68.0	92	61	92	57	91	57
November	2.6	8.2	0.0	92	44	90	39	90	38

I - Maximum Relative humidity (%)

n - Minimum Relative humidity (%)

APPENDIX - II

Test of independence and estimation of linkage for the joint segregation of leaf pigmentation and gall midge resistance in different crosses.

(i) Shyamla x Balam

Leaf colour	Gall midge reaction			X2	P
	R	S	Total	55G:9 P	value
Green	413	97	510	0.5789	0.50-0.30
Purple	63	28	91		
Total	476	125	601		

Gall midge resistance : 51 R : 13 S, X^2 = 0.087

for independence

$$\frac{[(413 \times 28) - (97 \times 63)]^2 \times 601}{476 \times 125 \times 510 \times 91}$$

6.471 *

The test of independence indicated that the two characters were not independent that is the existence of linkage was utilised.

	Ratio	Observed O	Expected E	$(O-E)^2$	$\frac{(O-E)^2}{E}$
GR	2805	413	411.57	2.034	0.005
GS	715	97	104.91	62.582	0.596
PR	459	63	67.34	18.908	0.281
PS	117	28	17.18	117.348	6.830 * *
Total	4096	601	601		7.712 * *

Estimation of total X^2

Source of variation	d.f.	X^2
GM reaction 51 R : 13 S	1	0.087
Leaf colour 55 G: 9P	1	0.578
Linkage	1	7.047 * *
Total	3	7.712 * *

Significant value shows existence of linkage.

X^2 value	d.f.	P	
		0.05	0.01
	1	3.843	6.635
	3	7.815	11.365

(ii) Shyamla x AC 1224-2

Leaf colour	Gall midge reaction			X^2	P
	R	S	Total	55 G:9 P	value
Green	354	106	460	1.806	0.20-0.10
Purple	68	20	88		
Total	422	126	548		

Gall midge resistance : 3 R : 1 S, $X^2 = 1.177$

X^2 for independence

$$= \frac{[(354 \times 20) - (106 \times 68)]^2 \times 548}{422 \times 126 \times 460 \times 88}$$

$$= 0.004$$

(iii) Shyamla x NHTA-8

Leaf colour	Gall midge reaction			X^2	P
	R	S	Total	13 G:3 P	value
Green	252	75	327	0.131	0.80-0.70
Purple	55	17	72		
Total	307	92	399		

Gall midge resistance : 3 R : 1 S, $X^2 = 0.802$

X^2 for independence

$$= \frac{[(252 \times 17) - (75 \times 55)]^2 \times 399}{307 \times 92 \times 327 \times 72}$$

$$= 0.015$$

(iv) Shyamla x Riting

Leaf colour	Gall midge reaction			X^2	P
	R	S	Total	55 G:9 P	value
Green	508	153	661	1.863	0.20-0.10
Purple	72	21	93		
Total	580	174	754		

Gall midge resistance : 3 R : 1 S, $X^2 = 1.486$

X^2 for independence

$$= \frac{[(508 \times 21) - (153 \times 72)]^2 \times 754}{580 \times 174 \times 661 \times 93}$$

$$= 0.014$$

(v) Shyamla x MNP-76

Leaf colour	Gall midge reaction			X^2	P
	R	S	Total	13 G:3 P	value
Green	404	92	496	1.741	0.20-0.10
Purple	69	30	99		
Total	473	122	595		

Gall midge resistance : 51 R : 13 S, $X^2 = 0.013$

X^2 for independence

$$= \frac{[(404 \times 30) - (92 \times 69)]^2 \times 595}{473 \times 122 \times 496 \times 99}$$

$$= 6.995 **$$

The test of independence indicated that the two characters were not independent that is the existence of linkage was utilised.

	Ratio	Observed O	Expected E	$(O-E)^2$	$\frac{(O-E)^2}{E}$
GR	663	404	385.23	352.31	0.914
GS	169	92	98.20	38.44	0.39
PR	153	69	88.90	396.01	4.45
PS	39	30	22.67	53.72	2.36
Total	1024	595	595		8.124

Estimation of total X^2

Source of variation	d.f.	X^2
GM reaction 51 R : 13 S	1	0.013
Leaf colour 55 G : 9 P	1	1.741
Linkage	1	6.370 *
Total	3	8.124 **

Significant value shows existence of linkage.

(vi) Shyamla x AC 169

Leaf colour	Gall midge reaction			X^2	P
	R	S	Total	55 G:9 P	value
Green	226	62	288	0.843	0.50-0.30
Purple	39	15	54		
Total	265	77	342		

Gall midge resistance : 3 R : 1 S, $X^2 = 1.126$

X^2 for independence

$$= \frac{[(226 \times 15) - (62 \times 39)]^2 \times 342}{265 \times 77 \times 288 \times 54}$$

$$= 1.014$$

(vii) Shyamla x AE 20

Leaf colour	Gall midge reaction			X^2	P
	R	S	Total	55 G:9 P	value
Green	333	80	413	0.953	0.50-0.30
Purple	43	16	59		
Total	376	96	472		

Gall midge resistance : 51 R : 13 S, $X^2 = 0.001$

X^2 for independence

$$\frac{[(333 \times 16) - (80 \times 43)]^2 \times 472}{376 \times 96 \times 413 \times 59}$$

1.912

(viii) Shyamla x AC 424

Leaf colour	Gall midge reaction			X^2	P
	R	S	Total	55 G:9 P	value
Green	302	69	371	0.062	0.90-0.80
Purple	44	17	61		
Total	346	86	432		

Gall midge resistance : 51 R : 13 S, X^2 = 0.043

X^2 for independence

$$\frac{[(302 \times 17) - (69 \times 44)]^2 \times 432}{346 \times 86 \times 371 \times 61}$$

2.82

(ix) Shyamla x Calrose-70

Leaf colour	Gall midge reaction			X^2	P
	R	S	Total	55 G:9 P	value
Green	128	14	142	1.624	0.2-0.1
Purple	19	11	30		
Total	147	25	172		

Gall midge resistance : 51 R : 13 S, $X^2 = 3.547$

X^2 for independence

$$[(128 \times 11) - (14 \times 19)]^2 \times 172$$

$$147 \times 25 \times 142 \times 30$$

$$14.32 * *$$

The test of independence indicated that the two characters were not independent that is the existence of linkage was utilised.

	Ratio	Observed O	Expected E	$(O-E)^2$	$(O-E)^2$ E
GR	2805	128	117.79	104.24	0.884
GS	715	14	30.03	256.96	8.556
PR	459	19	19.27	0.07	0.003
PS	117	11	4.91	37.08	7.553 * *
Total	4096	172	172		16.998 * *

Estimation of total X^2

Source of variation	d.f.	X^2
GM reaction 51 R: 13 S	1	3.547
Leaf colour 55 G: 9P	1	1.624
Linkage	1	11.827 * *
Total	3	16.998 * *

Significant value shows existence of linkage.

(x) Shyamla x AC 630

Leaf colour	Gall midge reaction			X^2	P
	R	S	Total	55 G:9 P	value
Green	224	55	279	0.253	0.50-0.30
Purple	32	10	42		
Total	256	65	321		

Gall midge resistance : 51 R : 13 S, $X^2 = 0.0007$

X^2 for independence

$$= \frac{[(224 \times 10) - (55 \times 32)]^2 \times 321}{256 \times 65 \times 279 \times 42}$$

$$= 0.379$$

(xi) Shyamla x ARC 15831

Leaf colour	Gall midge reaction			X^2	P
	R	S	Total	55 G:9 P	value
Green	139	55	194	0.292	0.50-0.30
Purple	19	11	30		
Total	167	64	231		

Gall midge resistance : 3 R : 1 S, $X^2 = 0.914$

X^2 for independence

$$= \frac{[(139 \times 9) - (55 \times 28)]^2 \times 231}{194 \times 37 \times 167 \times 64}$$

$$= 0.251$$

(xii) Shyamla x MNP-709

Leaf colour	Gall midge reaction			X^2	P
	R	S	Total	55 G:9 P	value
Green	382	89	471	0.048	0.90-0.80
Purple	54	21	75		
Total	436	110	546		

Gall midge resistance : 51 R : 13 S, $X^2 = 0.009$

X^2 for independence

$$= \frac{[(382 \times 21) - (89 \times 54)]^2 \times 546}{436 \times 110 \times 471 \times 75}$$

$$= 3.333$$

(xiii) Shyamla x AC 8070

Leaf colour	Gall midge reaction			X^2	P
	R	S	Total	55 G:9 P	value
Green	179	41	220	1.202	0.30-0.20
Purple	24	5	29		
Total	203	46	249		

Gall midge resistance : 51 R : 13 S, $X^2 = 0.519$

X^2 for independence

$$= \frac{[(179 \times 5) - (41 \times 24)]^2 \times 249}{203 \times 46 \times 220 \times 29}$$

$$= 0.033$$

(xiv) Shyamla x Kataribhog

Leaf colour	Gall midge reaction			X^2	P
	R	S	Total	55 G:9 P	value
Green	338	22	360	0.720	0.50-0.30
Purple	24	42	66		
Total	362	64	426		

Gall midge resistance

3 R : 1 S, $X^2 = 22.613$ **

13 R : 3 S $X^2 = 3.883$ *

51 R : 13 S $X^2 = 7.361$ **

Since the X values are highly significant, it shows some ther mechanism is working here.

APPENDIX-HI

Estimation of linkage value (p^2=P) by minimum discrepancy method , when the ratio for GM resistance 51 R : 13 S and for purple leaf colour 55 G : 9 P were obtained.

Possible Linkage.

(i) Gm A IPI

Gm A IP1 GmB IGmB C PI

gm A ip1 gm b igm b c p1

					Gm A IPl 2+P	GmA ipl 1-P	**gma IP1** 1-P	**gma ipl** P
GmB	**IGmB**	C	**Pl**	81	GR	PR	GS	PS
GmB	**IGmB**	C	**pl**	27	GR	GR	GS	GS
GmB	IGmB	c	**Pl**	27	GR	GR	GS	GS
GmB	**igmb**	C	**Pl**	27	GR	PR	GR	PR
gmb	IGmB	C	**Pl**	27	GR	PR	GS	PS
GmB	IGmB	c	**pl**	9	GR	GR	GS	GS
GmB	igmb	C	**pl**	9	GR	GR	GR	GR
gmb	IGmB	C	**pl**	9	GR	GR	GS	GS
GmB	igmb	c	**Pl**	9	GR	GR	GR	GR
gmb	IGmB	c	**Pl**	9	GR	GR	GS	GS
gmb	igmb	C	**Pl**	9	GR	PR	GS	PS
GmB	igmb	c	**pl**	3	GR	GR	GR	GR
gmb	IGmB	c	**pl**	3	GR	GR	GS	GS
gmb	igmb	C	**pl**	3	GR	GR	GS	GS
gmb	igmb	**c**	**Pl**	3	GR	GR	GS	GS
gmb	igmb	c	**pl**	1	GR	GR	GS	GS

Then frequencies of GR(Green Resistant), GS(Green Susceptible), PR(Purple Resistant) and PS(Purple Susceptible) classes will be :

GR= G72+117P, = 208-117P,
PR = 144 -117P and PS =177P

$$O = \frac{(672+117P)^2}{n_1} + \frac{(208-117P)^2}{n_2} + \frac{(144-117P)^2}{n_3} + \frac{(117P)^2}{n_4}$$

by differentiating above equation (i) we get,

$$0 = \frac{(2)(672+117P)(117)}{n_1} + \frac{(2)(208-117P)(-117)}{n_2} + \frac{(2)(144-117P)(-117P)}{n_3} +$$

by simplyfing it, P will be

$$P = \frac{-5.7435\ 112113114 + 1.7435\ n_1n_3n_4 + 1.2307}{n_2n_3n_4 + n_1n_3n_4}$$

(ii) GmA pl or c

Gm A	Pl	Gm B	IGm B	C	IP1
gm a	pl	gm b	igm b	c	ipl

					Gm A Pl 2+P	GmA pl 1-P	gma Pl 1-P	gma pl P
GmB	IGmB	C	Ipl	81	GR	GR	GS	GS
GmB	IGmB	C	Ipl	27	PR	GR	PS	GS
GmB	IGmB	c	Ipl	27	GR	GR	GS	GS
GmB	igmb	C	Ipl	27	GR	GR	GR	GR
gmb	IGmB	C	Ipl	27	GR	GR	GS	GS
GmB	IGmB	c	Ipl	9	GR	GR	GS	GS
GmB	igmb	C	Ipl	9	PR	GR	PR	GR
gmb	IGmB	C	Ipl	9	PR	GR	PS	GS
GmB	igmb	c	Ipl	9	GR	GR	GR	GR
gmb	IGmB	c	Ipl	9	GR	GR	GS	GS
gmb	igmb	C	Ipl	9	GR	GR	GS	GS
GmB	igmb	c	Ipl	3	GR	GR	GR	GR
gmb	IGmB	c	Ipl	3	GR	GR	GS	GS
gmb	igmb	C	Ipl	3	PR	GR	PS	GS
gmb	igmb	c	Ipl	3	GR	GR	GS	GS
gmb	igmb	c	ipl	1	GR	GR	GS	GS

Then frequencies of GR(Green Resistant), GS(Grecn Susceptible), PR(Purple Resistant) and PS (Purple Susceptible) classes will be :

GR = 711-39P, GS = 169+39P, PR = 105+39P and PS = 39-39P

$$0 = \frac{(711-39P)^2}{n_1} + \frac{(169+39P)^2}{n_2} + \frac{(105+39P)^2}{n_3} + \frac{(39-39P)^2}{n_4} \quad \text{(ii)}$$

by differentiating above equation (ii) we get ,

$$0 = \frac{(2)(711-39P)(-39)}{n_1} + \frac{(2)(169+39P)(+39)}{n_2} + \frac{(2)(105+39P)(39)}{n_3} + \frac{(2)(39-39P)(-39)}{n_4}$$

by simplifying above equation, the P Value will be

$$P = \frac{18.2307\, n_2n_3n_4 - 4.3334\, 111113114 - 2.6923\, n_1n_2n_4 + n_1n_2n_3}{n_2n_3n_4 + n_1n_3n_4 + n_1n_2n_4 + n_1n_2n_3}$$

(iii) Pl IGm B

Gm **A Gm B** Pl **IGm B C IP1**

gm a gm b pl igm **b c ipl**

					Pl Igmb 2+P	**Pl igmb** 1-P	**pl Igmb** 1-P	**pl igmb** P
GmA	**GmB**	**C**	**Ipl**	**81**	**GR**	GR	GR	GR
GmA	**GmB**	**C**	**ipl**	**27**	**PR**	PR	GR	GR
GmA	**GmB**	**c**	**Ipl**	**27**	**GR**	GR	GR	GR
GmA	**gmb**	**C**	**Ipl**	**27**	**GR**	GR	GR	GR
gmb	**GmB**	**C**	**Ipl**	**27**	**GS**	GR	GS	GR
GmA	**GmB**	**c**	**ipl**	**9**	**GR**	GR	GR	GR
GmA	**gmb**	**C**	**ipl**	**9**	**PR**	PR	GR	GR
gma	**GmB**	**C**	**ipl**	**9**	**PS**	PR	GS	GR
GmA	**gmb**	**c**	**Ipl**	**9**	**GR**	GR	GR	GR
gma	**GmB**	**c**	**Ipl**	**9**	**GS**	GR	GS	GR
gma	**gmb**	**C**	**Ipl**	**9**	**GS**	GS	GS	GS
GmA	**gmb**	**c**	**ipl**	**3**	**GR**	GR	GR	GR
gma	**GmB**	**c**	**ipl**	**3**	**GS**	GR	GS	GR
gma	**gmb**	**C**	**ipl**	**3**	**PS**	PS	GS	GS
gma	**gmb**	**c**	**Ipl**	**3**	**GS**	GS	GS	GS
gma	**gmb**	**c**	**ipl**	**1**	**GS**	GS	GS	GS

Then frequencies of GR(Green Resistant), GS(Green Susceptible), PR(Purple Resistant) and PS (Purple Susceptible) classes will be :

= 699+9P, GS = 181-9P, PR = 117-9P and = 27+9P

$$O = \frac{(699+9P)^2}{n_1} + \frac{(181-9P)^2}{n_2} + \frac{(117-9P)^2}{n_3} + \frac{(27+9P)^2}{n_4} \quad \text{(iii)}$$

by differentiating the above equation (iii), we get,

$$0 = \frac{(2)(699+9P)(9)}{n_1} + \frac{(2)(27+9P)(9)}{n_4}$$

by simplifying it , P will be

$$P = \frac{-77.6667\, n_2n_3n_4 + 20.1112\, 111113114 + 13\, n_1n_2n_4 - 3}{n_2n_3n_4 + n_1n_3n_4 \qquad n_1n_2n_3}$$

(iv) Gm B ipl

GmA	Gm B	IP1	Pl	IGm B	C
gm a	gmb	ipl	pl	igm b	c

				GmB IP1	GmB ipl	gmb IP1	gmb ipl
				2+P	1-P	1-P	P
GmA	IGmB	PIC	81	GR	PR	GR	PR
GmA	IGmB	Plc	27	GR	GR	GR	GR
GmA	IGmB	pl C	27	GR	GR	GR	GR
GmA	igmb	PlC	27	GR	PR	GR	PR
gma	IGmB	PlC	27	GS	PS	GS	PS
GmA	IGmB	pl c	9	GR	GR	GR	GR
GmA	igmb	Plc	9	GR	GR	GR	GR
gma	IGmB	Plc	9	GS	GS	GS	GS
GmA	igmb	pl C	9	GR	GR	GR	GR
gma	IGmB	pl C	9	GS	GS	GS	GS
gma	igmb	PlC	9	GR	PR	GS	PS
GmA	igmb	pl c	3	GR	GR	GR	GR
gma	igmb	Plc	3	GR	GR	GS	GS
gma	IGmB	pl c	3	GS	GS	GS	GS
gma	igmb	pl C	3	GR	GR	GS	GS
gma	igmb	pl c	1	GR	GR	GS	GS

Then frequencies of GR(Green Resistant), GS(Green Susceptible), PR(Purple Resistant) and PS (Purple Susceptible) classes will be :

GR = 699+9P, GS = 181-9P, PR = 117-9P and PS = 27+9P

$$0 = \frac{(699+9P)^2}{n_1} + \frac{(181-9P)^2}{n_2} + \frac{(117-9P)^2}{n_3} -- + \frac{(27+9P)^2}{n_4} \quad \text{(iv)}$$

by differentiating the above equation (iv), we get,

$$0 = \frac{(2)(G99+9P)(9)}{n_1} + \qquad \frac{(2)(27+9P)(9)}{n_4}$$

by simplifying it , P will be

$$P = \frac{-77.6667\, n_2 n_3 n_4 + 20.1112\, 111113114 + 13\, n_1 n_2 n_4 - 3\, 111112113}{112113114 + n_1 n_3 n_4 + n_1 n_2 n_4 +}$$

(v) IGm B ipl

Gm A Gm B IGmB IP1 C Pl

gm a gm b igm b ipl c

				Pl IGmB 2+P	Pl igmb 1-P	pl IGmB 1-P	pl igmb P
GmA	**GmB**	**C Pl**	**81**	**GR**	**PR**	**GR**	**PR**
GmA	**GmB**	**C pl**	**27**	**GR**	**GR**	**GR**	**GR**
GmA	**GmB**	**c Pl**	**27**	**GR**	**GR**	**GR**	**GR**
GmA	**gmb**	**C Pl**	**27**	**GR**	**PR**	**GR**	**PR**
gma	**GmB**	**C Pl**	**27**	**GS**	**PS**	**GR**	**PR**
GmA	**GmB**	**c pl**	**9**	**GR**	**GR**	**GR**	**GR**
GmA	**gmb**	**C pl**	**9**	**GR**	**GR**	**GR**	**GR**
gma	**GmB**	**C pl**	**9**	**GS**	**GS**	**GR**	**GR**
GmA	**gmb**	**c Pl**	**9**	**GR**	**GR**	**GR**	**GR**
gma	**GmB**	**c Pl**	**9**	**GS**	**GS**	**GR**	**GR**
gma	**gmb**	**C Pl**	**9**	**GS**	**PS**	**GS**	**PS**
GmA	**gmb**	**c pl**	**3**	**GR**	**GR**	**GR**	**GR**
gma	**Gmb**	**c pl**	**3**	**GS**	**GS**	**GR**	**GR**
gma	**gmb**	**C pl**	**3**	**GS**	**GS**	**GS**	**GS**
gma	**gmb**	**c Pl**	**3**	**GS**	**GS**	**GS**	**GS**
gma	**gmb**	**c pl**	**1**	**GS**	**GS**	**GS**	**GS**

Then frequencies of GR(Green Resistant), GS(Green Susceptible), PR(Purple Resistant) and PS (Purple Susceptible) classes will be :

GR= 708-27P, GS = 172+27P, PR = 108+27P and PS = 3G-27P

$$0 = \frac{(708-27P)^2}{n_1} + \frac{(172+27P)^2}{n_2} + \frac{(108+27P)^2}{n_3} + \frac{(36-27P)^2}{n_4} \quad \text{(v)}$$

by differentiating tbe above equation (v), we get,

$$0 = \frac{(2)(708-27P)(-27)}{n_1} + \frac{(2)(172+27P)(27)}{n_2} + \frac{(2)(108+27P)(27)}{n_3} + \frac{(2)(36-27P)(-27)}{\quad}$$

by simplifying it , P will be

$$P = \frac{26.2223\, n_2n_3n_4 - 6.3703\, n_1n_3n_4 - 4 + 1.333}{n_2n_3n_4 + n_1n_3n_4 + 111112114 +}$$

(vi) Gm B Pl

GmA IGm B IP1 C Gm B Pl

gm a igm b ipl c gm b pl

					GmB Pl 2+P	GmB pl 1-P	gmb Pl 1-P	gmb pl P
GmA	IGmB	**IPI**	C	81	GR	GR	GR	GR
GmA	IGmB	**Ipl**	c	27	GR	GR	GR	GR
GmA	IGmB	**ipl**	C	27	PR	GR	PR	GR
GmA	**igmb**	Ipl	C	27	GR	GR	GR	GR
gmA	**IgmB**	**Ipl**	C	27	GS	GS	GS	GS
GmA	IGmB	ipl	c	9	GS	GS	GS	GS
GmA	igmb	Ipl	c	9	GR	GR	GR	GR
gma	IGmB	**IPl**	c	9	GS	GS	GS	GS
GmA	igmb	ipl	C	9	PR	GR	PR	GR
gma	IGmB	**ipl**	C	9	PS	GS	PS	GS
gma	igmb	Ipl	C	9	GR	GR	GS	GS
GmA	igmb	ipl	c	3	GR	GR	GR	GR
gma	IGmB	ipl	**c**	3	GS	GS	GS	GS
gma	igmb	Ipl	c	3	GR	GR	GS	GS
gma	igmb	ipl	C	3	PR	GR	PS	GS
gma	igmb	ipl	c	1	GR	GR	GS	GS

Then frequencies of GR(Green Resistant), GS(Green Susceptible), PR(Purple Resistant) and PS (Purple Susceptible) classes will be :

GR = G66-3P, GS = 214+3P, PR = 114+3P and PS = 30-3P

$$0 = \frac{(666-3P)^2}{\ldots\ldots\ldots\ldots} + \frac{(214+3P)^2}{\ldots\ldots\ldots\ldots} + \frac{(114+3P)^2}{\ldots\ldots\ldots\ldots} + \frac{(30-3P)^2}{\ldots\ldots\ldots\ldots} \quad \text{(vi)}$$

by differentiating the above equation (vi), we get,

$$0 = \frac{(2)(666-3P)(-3)}{\ldots\ldots\ldots\ldots} + \frac{(2)(214+3P)(-3)}{\ldots\ldots\ldots\ldots} + \frac{(2)(114+3P)(3)}{\ldots\ldots\ldots\ldots} + \frac{(2)(30-3P)(-3)}{\ldots\ldots\ldots\ldots}$$

n_2 n_4

by simplifying it , P will be

$$P = \frac{222\ n_2n_3n_4 - 71.3334\ 111113114 - 38\ n_1n_2n_4 + 10}{112113114 + n_1n_3n_4 + n_1n_2n_4 +}$$

APPENDIX - IV

Estimation of linkage value (p^2=P) through minimum discrepancy method, when the ratio for GM resistance 51 R : 13 S and for purple loaf colour 13 G : 3 P were obtained.

Possible Linkage.

(i) **Gm A IPI**

GmA IPl GmB IGm B Pl

gm a ipl gm b igm b pl

				Gm A IPl 2+P	GmA ipl 1-P	gma IPl 1-P	gma ipl P
GmB	IGmB	Pl	27	GR	PR	GS	PS
GmB	IGmB	pl	9	GR	GR	GS	GS
GmB	igmb	Pl	9	GR	PR	GR	PR
gmb	IGmB	Pl	9	GR	PR	GS	PS
GmB	igmb	pl	3	GR	GR	GR	GR
gmb	IGmB	pl	3	GR	GR	GS	GS
gmb	igmb	Pl	3	GR	PR	GS	PS
gmb	igmb	pl	1	GR	GR	GS	GS .

Then the frequencies of GR (Green Resistant), GS (Green Susceptible), PR (Purple Resistant) and PS (Purple Susceptible) classes will be :

GR = 156+39P, GS = 52-39P, PR = 48-39P and PS = 39P

$$0 = \frac{(156+39P)^2}{n_1} + \frac{(52-39P)^2}{n_2} + \frac{(48-39P)^2}{n_3} + \frac{(39P)^2}{n_4} \quad \text{(i)}$$

differentiating the equation (i) we get,

$$O = \frac{(2)(156+39P)(39)}{n_1} + \frac{(2)(52-39P)(-39)}{n_2} + \frac{(2)(48-39P)(-39P)}{n_3} + \frac{(2)(39P)(39)}{n_4}$$

$$P = \frac{1.3334\, n_1 n_3 n_4 + 1.2307 - 4\, n_2 n_3 n_4}{}$$

(ii) GmA - **pl**

Gm A	Pl	GmB	IGm B	IPI
gm a	pl	gm B	igm b	ipl

				Gm A **Pl** 2+P	GmA **pl** 1-P	**gma Pl** 1-P	**gma pl** P
GmB	**IGmB**	IPI	27	GR	GR	GS	GS
GmB	IGmB	ipl	9	PR	GR	PS	GS
GmB	igmb	Ipl	9	GR	GR	GR	GR .
gmb	IGmB	Ipl	9	GR	GR	GS	GS
GmB	igmb	ipl	3	PR	GR	PR	GR
gmb	IGmB	ipl	3	PR	GR	PS	GS
gmb	igmb	**IPl**	3	GR	GR	GS	GS
gmb	igmb	ipl	1	PR	GR	PS	GS

Then the frequencies of GR (Green Resistant), GS (Green Susceptible), PR (Purple Resistant) and PS (Purple Susceptible) classes will be :

GR = 169-13P, GS = 39+13P, PR = 35+13P and PS = 13-13P

$$0 = \frac{(169-13P)^2}{n_1} + \frac{(39+13P)^2}{n_2} + \frac{(35+13P)^2}{n_3} + \frac{(13-13P)^2}{n_4} \quad \text{(ii)}$$

differentiating the equation (ii) we get,

$$O = \frac{(2)(169-13P)(-13)}{n_1} + \frac{39+13P)(+13)}{n_2} + \frac{(2)(35+13P)(13)}{n_3} + n_4$$

$$P = \frac{13\, n_2n_3n_4 - 3\, n_1n_3n_4 - 2.6923\, n_1n_2n_4 + n_1n_2n_3}{n_2n_3n_4 + n_1n_3n_4 +}$$

(iii) IGm B Pl

Gm A	Gm B	IGm B	Pl	IP1
gm a	**gm b**	igm **b**	pl	ipl

				IGmB Pl 2+P	IGmB pl 1-P	igmb Pl 1-P	igmb pl P
GmA	**GmB**	**Ipl**	27	GR	GR	GR	GR
GmA	**GmB**	**ipl**	9	PR	GR	PR	GR
GmA	**gmb**	**Ipl**	9	GR	GR	GR	GR
gma	**GmB**	**Ipl**	9	GS	GS	GR	GR
GmA	**gmb**	**ipl**	3	PR	GR	PR	GR
gma	**GmB**	**ipl**	3	PS	GS	PR	GR
gma	**gmb**	**Ipl**	3	GS	GS	GS	GS
gma	**gmb**	**ipl**	1	PS	GS	PS	GS

Then the frequencies of GR (Green Resistant), GS (Green Susceptible), PR (Purple Resistant) and PS (Purple Susceptible) classes will be :

GR = 165+3P, GS = 43-3P, PR = 39-3P and PS = 9+3P

$$0 = \frac{(165+3P)^2}{n_1} + \frac{(43-3P)^2}{n_2} + \frac{(39-3P)^2}{n_3} + \frac{(9+3P)^2}{n_4}$$

differentiating the equation (iii) we get,

$$0 = \frac{(2)(165+3P)(3)}{n_1} + \frac{43\text{-}3P)(\text{-}.lo93)}{} + \frac{39\text{-}3P)(\text{-}3)}{n_3} + \frac{9+3P)(3)}{n_4}$$

$$P = \frac{-55\, n_2n_3n_4 + 14.3334\, n_1n_3n_4 + 13\, n_1n_2n_4 - 3}{n_2n_3n_4 + n_1n_3n_4 + n_1n_2n_4 +}$$

(iv) Gm B ipl

Gm A IGm B Pl Gm B IP1

gm a igm b pl gm b ipl

				GmB IPl 2+P	GmB ipl 1-P	gmb IPl 1-P	gmb ipl P
GmA	**IGmB**	**Pl**	27	GR	PR	GR	PR
GmA	**IGmB**	**pl**	9	GR	GR	GR	GR
GmA	**igmb**	**Pl**	9	GR	PR	GR	PR
gma	**IGmB**	**Pl**	9	GS	PS	GS	PS
GmA	**igmb**	**pl**	3	GR	GR	GR	GR
gma	**IGmB**	**pl**	3	GS	GS	GS	GS
gma	**igmb**	**Pl**	3	GR	PR	GS	PS
gma	**igmb**	**pl**	1	GS	GR	GS	GS

Then the frequencies of GR (Green Resistant), GS (Green Susceptible), PR (Purple Resistant) and PS (Purple Susceptible) classes will be :

GR = 165+3P, GS = 43-3P, PR = 39-3P and PS = 9+3P

$$0 = \frac{(165+3P)^2}{n_1} + \frac{(43-3P)^2}{n_2} + \frac{(39-3P)^2}{n_3} + \frac{(9+3P)^2}{n_4} \quad \text{(iv)}$$

differentiating the equation (iv) we get,

$$0 = -\frac{(2)(165+3P)(3)}{} + \frac{(2)(43-3P)(-3)}{} + \frac{39-3P)(-3)}{n_3} + \frac{(2)(9+3P)(3)}{n_4}$$

$$P = \frac{-55\, n_2n_3n_4 + 14.3334\, n_1n_3n_4 + 13\, n_1n_2n_4 - 3}{+ n_1n_3n_4 + 111112114 +}$$

(v) Gm B ipl

Gm A Gm B Pl IGm **B** **IP1**

gm **a** gm **b** pl igm **b** **ipl**

				IGmB **IP1** 2+P	IGmB **ipl** 1-P	igmb **Ipl** 1-P	igmb **ipl** P
GmA	**GmB**	**PI**	27	GR	PR	GR	PR
GmA	**GmB**	**pl**	9	GR	GR	GR	GR
GmA	**gmb**	**Pl**	9	GR	PR	GR	PR
gma	**GmB**	**Pl**	9	GS	PS	GR	PR
GmA	**gmb**	**pl**	3	GR	GR	GR	GR
gma	**GmB**	**pl**	3	GS	GS	GR	GR
gma	**gmb**	**Pl**	3	GS	PS	GS	PS
gma	**gmb**	**pl**	1	GS	GS	GS	GS

Then the frequencies of GR (Green Resistant), GS (Green Susceptible), PR (Purple Resistant) and PS (Purple Susceptible) classes will be:

GR = 168-9P, GS = 40+ 9P, PR = 36+ 9P and PS = 12- 9P

$$O = \frac{(168-9P)^2}{n_1} + \frac{(40+9P)^2}{n_2} + \frac{(36+9P)^2}{n_3} + \frac{(12-9P)^2}{n_4} \quad \text{(v)}$$

differentiating the equation (v) we get,

$$0 = \frac{(2)(168-9P)(-9)}{n_1} + \frac{(2)(40+9P)(9)}{n_2} + \frac{(2)(36+9P)(9)}{n_3} + \frac{(2)(12-9P)(-9)}{n_4}$$

$$P = \frac{-18.6667\, n_2n_3n_4 + 4.4445\, n_1n_3n_4 + 4\ 111112114 - 1.3334}{n_2n_3n_4 + 111113114 + n_1n_2n_4 +}$$

(vi) m B Pl

Gm A	IGm B	IPl	Gm B	Pl
gm a	igm b	ipl	gm b	pl

				GmB Pl 2+P	GmB pl 1-P	gmb Pl 1-P	gmb pl P
GmA	**IGmB**	**IPl**	27	GR	GR	GR	GR
GmA	**IGmB**	**ipl**	9	PR	GR	PR	GR
GmA	**igmb**	**Ipl**	9	GR	GR	GR	GR
gma	**IGmB**	**Ipl**	9	GS	GS	GS	GS
GmA	**igmb**	**ipl**	3	PR	GR	PR	GR
gma	**IGmB**	**ipl**	3	PS	GS	PS	GS
gma	**igmb**	**Ipl**	3	GR	GR	GS	GS
gma	**igmb**	**ipl**	1	PR	GR	PS	GS

Then the frequencies of GR (Green Resistant), GS (Green Susceptible), PR (Purple Resistant) and PS (Purple Susceptible) classes will be :

$$GR = 166 - P, \quad GS = 42 + P, \quad PR = 38 + P \quad \text{and} \quad PS = 10 - P$$

$$O = \frac{(166-P)^2}{n_1} + \frac{(42+P)^2}{n_2} + \frac{(38+P)^2}{n_3} + \frac{(10-P)^2}{n_4} \quad \text{.....(vi)}$$

differentiating the equation (vi) we get,

$$O = \frac{(2)(166-P)(-1)}{n_1} + \frac{(2)(42+P)(1)}{n_2} + \frac{(2)(38+P)(1)}{n_3} + \frac{(2)(10-P)(-1)}{n_4}$$

$$\mathbf{P = \frac{-166\, n_2n_3n_4 + 42\, n_1n_3n_4 + 38\, n_1n_2n_4 - 10\, n_1n_2n_3}{n_2n_3n_4 + n_1n_3n_4 + n_1n_2n_4 + n_1n_2n_3}}$$