HISTOLOGICAL AND BIOCHEMICAL BASIS OF RESISTANCE TO WOLLY APHID IN SUGARCANE (*Saccharum officinarum* L.)

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I. INTRODUCTION

Sugarcane (*Saccharum officinarum* L.) is the leading cash crop of India, the only source of sugar in the country and also source of food, fuel fodder and fibre, a sweet reed which has attracted the attention of many from monks to monarchs. Among sugar producing plants, sugarcane is responsible for about 60 per cent of world's sugar production, the remaining 40 per cent comes from sugar beet, a temperate zone crop. Sugarcane is cultivated mainly in the tropics, though it is grown in sub tropical areas in India.

This crop sustains the second largest organized agro-based industry in the country with an area of 4.36 million hectares, cane production of 281.5 million tonnes, productivity of 64.6 t/ha and sugar recovery of 10.36 per cent (Anon., 2004). Karnataka state ranks third with an area 0.385 million hectare, cane production of 32.4 million tonnes and productivity of 84.4 tonnes per ha. Sugarcane is adaptable to a wide range of agricultural situations, but its productivity is generally limited due to biotic and abiotic stresses. Northern Karnataka has major stake in the state in respect of area (74%) and production (69%), though this part of the state is top in the country for sugar recovery as it is favoured with cold and dry winter coinciding cane ripening/crushing season, but the cane productivity levels are lower, compared to southern Karnataka and Tamil Nadu. The lower cane productivity in this high potential region is primarily because of monoculture of sugarcane varieties either CoC 671 or Co 8011 over large and adverse agro-ecological zones.

Besides various abiotic and biotic stresses sugar cane woolly aphid (SWA) *Ceratovacuna lanigera* Zehenter has recently become a serious biotic constraint, threatening sugarcane cultivation itself, including sugar mills and foreign exchange, sugarcane woolly aphid has become a serious pest in peninsular India, causing significant losses in cane yield and sugar recovery, so far woolly aphid on sugarcane was considered as a minor or negligible pest of sugarcane, it was first reported in sugarcane in 1897 from Java (Zehenter, 1897). In India during 1958, the occurrence of *C. lanigera* was first time reported from Cocchebahar in West Bengal (Basu and Banerjee, 1958). Later, in 1963 the pest incidence was reported from Tripura (Behura, 1963), in 1967 from Assam (Saxena, 1967), in 1971 from Uttar Pradesh (Chakrabarti *et al.,* 1971).

In Indonesia loss due to the heavy incidence of *C. lanigera* has been reported up to 26 per cent in cane yield and 24 per cent in sugar content (Farina, 1994). In Maharashtra 7 to 39 per cent reduction in cane yield and 1.2 to 3.43 unit reduction in sugar recovery has been recorded. A total loss of about Rs. 874 crores has been estimated in India due to damage caused by SWA alone (Patil *et al.,* 2004).

Though synthetic insecticides are effective against the sugarcane woolly aphid, they do not find place in sugarcane ecosystem for reasons like operational hazards during application of insecticides, improper coverage of crop canopy after seven months. So, considering the fast multiplication, quick spread on the large area, existence of the pest in high humid places, very negligible population of predators during March to June, to save the reduction in the cane yield and sugar recovery loss and to avoid the further the further spread of woolly aphid, it is necessary to identify SWA resistant clones which would become important component of IPM.

Host plant resistance (HPR) is one of the important component of IPM, which is environmentally safe and ecologically stable. Anatomical characters are know to play a major role in attributing resistance against insects, characters like leaf thickness, vein toughness, compactness of the tissue are some of the important traits against aphid resistance. The localization of histochemical substances such as total polysaccharides, protein and total RNA also play important role in resistant mechanism it is possible to identify the susceptible and resistant clones by employing histochemical techniques.

Hydroxamic acids (Hx) are secondary plant chemicals present in Graminae (Niemeyer, 1991 and Baria Copaja and Niemeyer, 1992) showing significant deleterious effects on organisms such as fungi, bacteria and insects (Niemeyer, 1988 and Philogene and Lambert, 1990). Information from experiments in which aphids are reared on artificial diets containing DIMBOA the main Hx in wheat (Niemeyer *et al.,* 1988 and Niemeyer *et al.,* 1989)

and from electronic monitoring of aphid feeding (Argandona *et al.,* 1983) suggests DIMBOA exerts both toxic and antifeedent effects which results in inhibition of feeding (Argandona *et al.,* 1983).

In sugarcane, the cane and sugar yields are considered to be the complex characters. The information on the phenotypic and genotypic interrelationship of cane yield and CCS yield with their component characters and among yield and quality components *inter-se* would be of immense help to the sugarcane breeder. But the inter dependence of these component characters among themselves often influence the direct relationship with yield (both cane and sugar yield), as a result the information based on the correlation coefficients becomes not dependable. Path coefficient analysis, on the other hand provides direct and indirect effects of component traits which helps to understand true relationship of the characters.

So an attempt was made to study the possible resistant mechanisms and basis against woolly aphid in sugarcane with the following objectives.

1. Study of histological and histochemical basis of resistance to woolly aphid in sugarcane
2. Study of biochemical basis of resistance to woolly aphid in sugarcane
3. Association studies in SWA resistant clones for cane yield and its component traits

II. REVIEW OF LITERATURE

I. STUDIES ON SUGARCANE WOOLLY APHID

Cane yield is influenced by several factors like soil fertility, climate, variety, cultural practices and prevalence of pests and diseases (David and Easwaramurthy, 1986). Among these insect pests cause considerable losses in cane yield as well as sugar recovery. David and Nandagopal (1986) reported over 214 insect species which attack sugarcane crop from the first day, when the cane sets are planted in the soil and their damage in one or the other form continued until the crop is harvested. Out of these about a dozen are serious and are of regular in occurrence. These include termites, white grubs, borers, pyrilla, armyworms and grass hoppers which attack the crop at various stages of growth. Among sucking pests, pyrilla, whiteflies, bugs scale insects, mealy bugs and sugarcane woolly aphid are common.

Sugarcane woolly aphid, *Ceratovacuna lanigera* Zehnter was first reported on sugarcane in 1897 from Java (Zehntner, 1897) and it is a serious pest of sugarcane in Asia (Matsumura 1910 Coplland, 1917, Ishid, 1928 and Waterhouse, 1993).

In India it has been reported as a pest of sugarcane from West Bengal, Assam, Nagland, Sikkim, Tirpura and Uttar Pradesh (Saxena, 1967, Ghosh, 1974, Phukan *et al.,* 1988, Tripathi, 1992 and Gupta and Goswamy, 1995).

Since 2002, it has attained a serious status in Maharashtra and Karnataka (Anon., 2002a, and Patil, 2003 and it has also spread to Andhra Pradesh, Tamil Nadu, Goa, Kerala, Bihar and Uttaranchal.

Nature of damage

The insect prefers sugarcane as its primary host (Hill, 1993) Bamboo, *Miscanthus sinesis* and cynodon dactylon are reported as secondary hosts (Aoki *et al.,* 1984 and Anon., 2001). Both nymphs and adults desap the leaves ventrally by piercing their stylet through the stomata leading to development of whitish patches, which coalesce and turn yellow. Later affected leaves dry from tip down words along the margin before complete drying. Due to heavy secretion of honey dew which falls on lower leaves leading to development of sooty mould which reduce photosynthetic area affecting cane yield and quality, continuous infestation leads to reduction in the length, girth, height and sugar content of the stalks in susceptible varieties in vietnam (Anon., 1963) and loss in tonnage as well as sugar recovery was also reported from India (Tripathi, 1995). Gupta and Goswami (1995) assessed the effect of 25 and 100 per cent aphid infected leaves on yield and quality parameters of sugarcane and reported that 100 per cent infestation had detrimental effects on length (11.6% reduction), girth (3.5% reduction) height (16.6% reduction), inter nodal length (18.4% reduction) and further juice quality parameters also exhibited considerable reduction. The per cent reduction in sucrose, brix per cent, glucose, purity and CCS was 53.3, 32.3, 25.3, 31.7 and 64.0 respectively.

A study on loss estimation was carried at Agricultural Research Station Sankeshwar, involving different commercial varieties *viz.,* Co 92020, CoC 671, Co 86032, Co 8021 and Co 8011, there was significant reduction (30.35%) in both cane yield and sugarcane yield parameters in severely infested 10 month crop and further infestation at early stage (during germination to tillering phase) leads to complete drying causing 100 per cent loss (Anon., 2002b). In the infected areas even jaggery industry has suffered in terms of jaggery quality and recovery, seed germination loss to the extent of 40 per cent and considerable fodder (cane tops) quality deterioration has also been reported due to sugarcane woolly aphid.

Pest control strategies

To manage this pest, several management practices *viz.,* chemical, biochemical, cultural and host plant resistance have been suggested (Anon., 2002b, Patil, 2003, Nerkar, 2003, Lingappa *et al.,* 2003 and Joshi and Viraktmath, 2004).

Attempts to identify SWA resistant sugarcane germplasm have been made in Taiwan, Philippines and Indonesia. In Taiwan varietal differences for aphid incidence and its biology

were studied and indicated the variety ROC 1 to offer resistance to some extent as it was associated with larger nymphal period and fewer progenies per adult (Pan *et al.,* 1984). In a similar study at Philippines, varietal difference in relation to aphid biology were reported (Rueda and Calilung, 1974). In Indonesia a resistance breeding programme was initiated for SWA (Mirzwan *et al.,* 1995). There are no reports available on the level of resistance for SWA in India. However, earlier studies reported relative susceptibility of genotypes (Anon., 2002a) and varieties with lower nitrogen content, total soluble solids and higher silicon were reported less susceptible (Phukan, 1978).

Patil *et al.* (2005) out of ten identified SWA resistant clones, three clones *viz.,* SNK 44, SNK 61 and SNK 754 were promising for both cane and sugar yields with acceptable cane features. On artificial inoculation the aphids failed to survive and perpetuate on these resistant clones instead they wondered throughout the length of leaf after release leading to cent per cent mortality after 48 to 72 hrs of release. Whereas, in susceptible genotypes including commercial varieties, aphids immediately colonize within 24 hrs after release.

II. HOST PLANT RESISTANCE

A plant is neither susceptible to all the phytophagous insects nor any insect species is the pest of all the species of plants. Plant species which are fed upon by an insect are called as host plants which are not fed at all are non host plants. Host plant resistance is the result of interactions between two biological entries, the plant and the insect under the influence of various environmental factors (Dhaliwal *et al.,* 1993).

The earliest documentation of an insect resistant variety was a wheat variety under hill which was found to resist the attack of the Hussian fly *Mayetiola destructor* in New York, 1782. However the scientific investigations into the mechanisms of host plant resistance date back to approximately 100 years and the first significant contribution of host plant resistance in agriculture was made in 1980 when European grape vines were successfully grafted on to resistant root stock to save French vine industry.

Concept of host plant resistance

Snelling (1941) defined plant resistance as those characters that enable a plant to avoid, tolerate or recover from attacks of insects.

Painter (1951) described plant resistance as the relative amount of heritable quantities that influence the ultimate degree of damage done by insect.

Kogan (1982) defined resistance to insects as the inherent property that enables a plant to inhibit growth of insect populations.

Mechanisms of resistance

Painter (1951) grouped mechanisms of resistance into three main categories, *viz.,* non preference antibiosis and tolerance.

Kogan and Ortman (1978) proposed the term antixenosis instead of non preference.

A. Antixenosis

Antixenosis refers to the resistance mechanism by host plant to deter or reduce colonization by insects, the plant may deter the insects from feeding, sucking, oviposition or taking shelter and insects are unable to colonize (Kogan, 1982).

B. Antibiosis

Antibiosis refers to the adverse effect of the host plant on the biology (growth, development or reproduction) of the insects and their progeny infesting, it has adverse effect on physiology of the insect including consumption, assimilation, utilization and allocation for reproduction (Ananthkrishnan *et al.,* 1994).

Characteristic features of antibiosis (Painter, 1951)

The death of insects on resistant plants frequently takes place during the first instars, this death of the very young nymphs or larvae has been observed on resistant plants in several species of aphids. The killing of young larvae and eggs by proliferation of tissue in the case of the cotton boll weevil.

A consistent restlessness of the insect is some times observed when insects feed on resistant varieties. This behaviour on resistant varieties is often true for aphids.

A reduction in number of young ones is a characteristic feature of the aphid when placed on resistant cultivars.

Possible physiological explanations

1. Presence of toxin in resistant plant
2. The imbalance in available of nutrients in plants
3. The presence of growth or reproduction deterrent in plants

C. Non-preference

Insects does not prefer the plant as a good source of food.

III. BASIS OF RESISTANCE

A number of plant characteristics are known to render the cultivars less suitable or unsuitable for feeding, ovisposition and development of insect pests. Broadly these characteristics can be classified into two categories, *i.e.,* biophysical and biochemical.

A. Biophysical basis

The plant resistance is controlled by several morphological factors like remote factors. Eg., internal anatomy, colour, shape, size, etc and close range or contact factors *etc.*, thickening of cell walls and rapid proliferation of plant tissues, solidness and other plant and stem characteristics.

B. Biochemical basis

A wide array of chemical substances including inorganic chemicals, primary and intermediary and secondary substances are known to impart resistance to a wide variety of insect pests. Broadly, the chemicals imparting resistance to insects can be classified into two main categories.

1. Nutrition

The host plant may be deficient in certain nutritional elements required by the insects and hence prove resistant.

2. Allelochemicals

Allelochemicals are non-nutritional chemicals produced by organism of one species that affect the growth, health, behaviour or population biology of individuals of another species (Whittakar, 1970). The allelochemicals have been broadly classified into two categories, *viz.,* (a) *Allomones* tending to confer an adaptive advantage to the producing organism, *i.e.,* the host plant and (b) kairomones-tending to give an adaptive advantage to the receiving organism *i.e.,* the phytophagous insect. The involvement of allelochemicals in various types of insect plant relationships can determine the status of a plant either as a host (presence of kairomone) and non-host (absence of kairomone) or as resistant (presence of allomone) and susceptible (absence of allomone). Allomones are considered to be a major factor of insect resistance in plants and these have been exploited to increase levels of resistance in several agricultural crops.

As sugarcane woolly aphid (SWA) is a new pest of sugarcane and literature pertaining to host plant resistance of sugarcane is scanty, so an attempt is made to review literature pertaining to sucking insect pests of sugarcane and other crop species.

2.1 HISTOLOGICAL AND HISTOCHEMICAL BASIS OF RESISTANCE TO SUGARCANE WOOLLY APHID

2.1.1 Pyrilla

Pyrilla is one of the major sucking pest of sugarcane leaf, which causes considerable damage by reducing the vigour of plant.

Khanna *et al.* (1950) working on resistant varieties of sugarcane to pyrilla reported that resistant varieties differed significantly in their leaf anatomical structures, the phloem tissues of the resistant varieties was narrowed by a protecting covering formed by fusion of vascular sheath and sclerenchymatus tissue (rib) around the phloem. However such covering (protection) was not observed in susceptible varieties of sugarcane.

Kumarsinghe *et al.* (2001) while working on basis of resistance of sugarcane to pyrilla insect reported that sugarcane cultivars resistant to pyrilla had a thick layer of phloem fibre around the vascular bundle in leaf compared to susceptible ones, which had thin layer of covering around the vascular bundles.

2.1.2 Leaf hopper

Leaf hoppers are the important sucking pest of leaf and stem, which suck the sap by scraping the measophyll tissue of leaf and stem.

Harris (1930) in a study including pubescent and glabrous varieties of soybean, reported that pubescent varieties of soybean are known to be highly resistant to potato leaf hopper *Empoasca fabae*, where in leaf hoppers were unable to establish on plants whose epidermis is covered with a thick layer of long cellulose hairs.

Pinnemeyer and Tingey (1987) working on different types of trichomes in providing efficient defense against insects, reported that hooked trichomes are of more efficient in providing plant defense against insects than other types of trichomes, however in another experiment Arthur *et al.* (1987) concluded that tricome morphology and density did not play a major role in imparting resistance to leaf hopper in common bean.

Elden and Elgin (1992) conducted an experiment to know the mechanisms of host plant resistance to the potato leaf hopper in selected alfalfa lines and concluded that primary mechanism of resistant differed among the alfalfa populations studied, B16 population was associated with non preference due to dense pubescence however in Arc and ISC population expressed feeding or oviposition non-preference or both.

2.1.3 Aphids

Aphids are one of the most important sucking pests of crop plants, which feed on phloem sap of leaf and stem tissue of plants.

Annand (1946) while working on reaction of glabrous and hairy types of cotton for aphid resistance in the form of differential attraction of aphids concluded that differential attraction of the aphids could be due to the difference in the light intensities reflected from the two sorts of leaves, leading to fewer aphids in glabrous cotton leaves than on hairy types.

Campbell *et al.* (1982) studied aphid feeding behaviour on resistant and susceptible cultivars using electronic method of monitoring aphid probing, concluded that biotype (C) had significantly lower feeding time from the phloem of resistant cultivars compared to significantly more time in susceptible cultivars.

Khan and Agarwal (1990) in an experiment to study the different anatomical characters which impart resistance to aphid *Aphis gossypii* in cotton reported that non-preference type of resistance in cotton was due to anatomical characters such as less distance among hair basis, less thickness of parenchyma and more distance of palisade tissue from lower leaf epidermis.

Melaku *et al.* (1992) working in feeding behaviour of Russian wheat aphid on host plants such as wheat, oat, barley and non host sorghum using a computerized electronic insect feeding monitory, reported that Russian wheat aphid required four times longer time to locate the phloem and achieve committed phloem ingestion and further concluded that aphid salivated more and ingested less while feeding on sorghum than on wheat, oat and barley cultivars.

Webster *et al.* (1994) conducted an experiment on yellow sugarcane aphid and green bug in sugarcane to know the possible mechanism of resistance and concluded that leaf surface pubescence is of questionable under green bug plant resistance, however it was an effective resistance mechanism against the yellow sugarcane aphid.

Dhaliwal and Arora (1998) working on tolerant and susceptible cultivars of cotton to *Aphis gossypii* reported that aphid was unable to pierce the stylet in tolerant cultivars mainly due to stem hardness compared to susceptible cultivars.

2.2 BIOCHEMICAL BASIS OF RESISTANCE TO SUGARCANE WOOLLY APHID

Roach (1937) working on host plant resistance of woolly apple aphid *Erisoma lanigerum* Hausm involving resistant and susceptible genotypes, reported that when woolly apple aphid reared on resistant cultivar of northern spy, the number of young ones produced were smaller and their life cycle was shorter, however on susceptible there was normal life cycle.

Victor *et al.* (1968) working on biochemical basis of resistance to corn leaf aphids reported that DIMBOA, a hydroxamic acid isolated from corn leaf extracts was deleterious to aphids when fed on artificial diets and concluded that hydroxamic acid content in wheat and other graminae species correlated with resistance to the aphid *Metopolophium diethodum*, however he further proposed that hydroxamic acid acts as naturally occurring protective factors against aphids.

Experiments were conducted to determine the relationship between hydroxanmic acid total phenol and orthodihydroxy phenol concentrations in corn (*Zea mays* L.) tissues and resistance to the corn leaf aphids (*Rhopalosiphum meidis* (Fitch)) eleven corn in breeds were inoculated for resistance. A highly significant negative correlation (r= -0.06) was obtained between aphid infestation and hydroxanmic acid concentration, poor correlations were found between aphid infestation and both total phenol and orthodihydroxy concentrates (Beek *et al.,* 1982).

In order to find out the effect of DIMBOA supplemented in maize leaf diet Argandona *et al.* (1983) reported DIMBOA, the main hydroxamic acid isolated from maize leaf extract increased the mortality of *Schizaphis graminnum* and concluded that, the deleterious effects of DIMBOA on aphids was due to feeding deterrence and toxicity, the 2-□-0-glucoside of DIMBOA, the form in which DIMBOA naturally occurs in graminae had lethal effect on aphid and acted as feeding deterrent.

Thakary *et al.* (1988) carried out an experiment to know the effect of DIMBOA on aphid survival and reproduction rate in wheat, reported that DIMBOA the main benzoxazine isolated from wheat leaf extracts reduced survival and reproduction rates when added to artificial diets. Hydrolysis of glucoside to aglucon upon aphid infestation may be the reason for resistance of cereals to aphids.

Niemeyer *et al.* (1989) working on behaviour of aphids on resistant wheat genotypes in relation to hydroxamic acid DIMBOA content, reported that inverse relationship exists between DIMBOA levels and extent of aphid feeding on wheat plants and concluded that hydroxamic acid greatly affect the probing and setting behaviour of the aphids on resistant wheat plants.

Givovich *et al.* (1992) carried out an experiment to know the feeding behaviour pattern and post feeding behaviour of aphids on resistant wheat cultivars with high Hx levels. They reported that when aphids feed on phloem sap containing higher levels of Hx

concentration feed less and concluded that lesser feeding was indicated by the decreased honey dew secretion by aphids.

Givovich *et al.* (1994a) reported that although mean Hx content of whole leaf extracts differed significantly between resistant wheat cultivars to aphids but DIMBOA – glucoside concentrations in the phloem sap of these genotypes did not exhibit significant difference among themselves, however they further concluded that DIMBOA-glucoside was the only Hx related chemical present in phloem sap.

Givovich *et al.* (1994b) reported that there was more time required by aphid to reach the phloem, when the concentration of Hx was more in mesophyll and concluded that on wheat plants with high Hx levels aphids spend more time in searching for a suitable phloem vessel with increased frequency of probing and periods of ingestation and further concluded that aphids not only feed less but also lowered nutrients ingestation.

Niemeyer and Givovich (2000) experimented the effect of hydroxamic acid on aphid using electrical penetration graphs and phloem sap analysis recorded no significant difference of Hx in phloem sap but significant difference of Hx in whole leaf extracts. They also reported feeding deterrency by DIMBOA in the plant's mesophyll.

Nikus Feanette (2003) working on a proposed defense system in the poaceae consisting of glucosylated enzyme and cyclic hydroxamic acid DIMBOA and DIMBOA reported that □-glucosidase enzyme was located in plastids, cell walls and glucoside in cytoplasm and cells associated with vascular bundle.

2.3 ASSOCIATION STUDIES

2.3.1 Character association

Estimation of phenotypic, genotypic and environmental correlation between yield and yield attributes and among yield attributes themselves would provide information for the breeding programme when selection to be based on two or more characters simultaneously.

2.3.1.1 Yield and yield components

In sugarcane, cane yield is largely determined by stalk number, stalk length, stalk diameter and stalk weight (Singh and Singh, 1954).

Gangopadhyaya and Sarkar (1963) observed a significant correlation of cane yield with stalk number and height but stalk diameter, according to them was not important. However, Ethirajan (1965) noticed positive and highly significant correlation between cane yield and stalk diameter, apart from number of canes, stalk height and single cane weight.

Strong positive association between cane yield and stalk weight was reported by Herbert (1965), Brown *et al.* (1969) and James and Miller (1971). Rao and Krishnamoorthy (1968) observed negative association between stalk number and stalk weight. Legendre (1970) reported a high positive correlation between cane yield and its components *viz.,* stalk number, stalk weight and also the number of internodes per stalk.

Mariotti (1971) based on phenotypic correlations reported that cane yield was related with stalks per plot, stalk weight and stalk length. On the basis of correlation, stalk number per plot followed by stalk weight had high values of association with cane yield. He was emphatic that number of stalks per plot and stalk weight were most important characters in determining cane yield. High positive correlation between number of millable canes, stalk height and cane yield and negative association between diameter and number of stalks were reported by Batcha and Sahi (1972).

Positive and significant interrelationship of stalk number and stalk diameter with cane yield was noticed by Jams and Miller (1975) while Sahi and Patel (1975) reported a strong positive association of cane yield with stalk weight and stalk length. However, positive association of only stalk number with cane yield was reported by Miller and James (1974), Ramana Rao and Rao (1977), Khairwal *et al.* (1978) and Mariotti (1978).

Hooda *et al.* (1979) observed positive association between stalk weight as well as stalk length with cane yield. While, Tai *et al.* (1980) reported high positive association of stalk number and stalk weight with cane yield.

Sahi (1981) found a strong positive association of single cane weight and stalk diameter with cane weight at 10th month. Kang *et al.* (1983) observed strong association of cane yield with plant height, stalk diameter, stalk number and stalk weight; According to Punia *et al.* (1983) cane yield was influenced by the number of tillers per clump, the number of millable canes per clump, cane thickness and cane weight.

Reddy and Reddi (1985) while studying the association pattern in 12 varieties of sugarcane revealed the significant interrelationship of clump weight with stalk weight, stalk length, stalk diameter and stalk volume. The same authors in the year 1986b, reported highly significant phenotypic and high genotypic association of cane yield with stalk number per plot, stalk weight and stalk length.

Significantly high phenotypic and genotypic association of cane yield with number of millable canes per plot and length of millable cane was reported by Ramana Reddy (1988).

Sekhar (1986) observed close association of number of internodes, cane length and single cane weight with cane yield. On the other hand Saikar *et al.* (1986) found close association of cane yield with plant weight.

In a similar interrelationship study of cane yield with its components in the hybrid progenies of twelve intervarietal direct and reciprocal crosses at the first clonal generation, Reddy and Reddi (1987 b) observed highly significant association of cane yield with stalk number in all the 12 crosses, with stalk length in seven crosses and stalk diameter in five crosses. They suggested that the stalk number per plot followed by stalk weight were the major components influencing cane yield per plot.

Poor association of germination and early tillering with cane yield was reported by Nair and Sreenivasan (1990). They also noticed closer correlation between shoot height recorded on 90th day after planting with cane yield. Significant and positive correlation of cane yield with millable cane, millable height of cane, number of internodes and leaf area index was noticed by Gajera *et al.* (1991). While Pillai and Ethirajan (1993) reported significant positive correlation of cane yield with stalk number, stalk length and stalk weight and non-significant association with quality traits. Reddy and Somarajan (1994) did not notice any significant positive association of cane yield with any of the yield attributes like single cane weight, cane length, cane thickness and number of internodes except number of millable canes.

Positive correlation of cane yield with number of millable stalks, stalk weight, stalk length, diameter and internode number was reported by Sarvjeet Singh and Khan (1995). While Singh *et al*. (1995) observed significant positive association of stalk yield with number of stalks per clump and stalk weight. Das *et al* (1996) reported significant association of cane yield with stalk weight, stalk diameter and number of internodes per cane.

Significant positive correlation of cane productivity with number of millable canes, stalk height, stalk weight and internode length were observed by Kundu and Gupta (1997). While Rishipal *et al.,* (1998) noticed close association of cane yield with number of millable canes, single stalk weight and stalk height.

2.3.1.2 Association among yield components

Significant positive association between stalk weight and stalk diameter was reported by Herbert (1965), Miller and James (1974), Sahi and Patel (1975), Bathla (1978), Singh *et al.* (1981) and Ramana Reddy (1984). On the contrary, and Kang *et al.* (1983) observed a negative association between these two traits.

A negative correlation between stalk diameter and stalk number was reported by several research workers (Herbert, 1965, James and Flagout, 1969, Smith and James, 1969, James, 1971, Mariotti, 1971, Batcha, 1975, Sahi and Patel 1975, Balasundaram and Bhagyalakshmi, 1978a, and 1978 and Nair and Somaraja, 1984b).

A negative association between stalk weight and stalk number was reported by Sahi and Patel (1975), Ramana Rao and Rao (1977), Tai *et al.* (1980), Nair and Somarajan (1984 a and b) and Ramana Reddy (1988). While Singh and Jain (1968) noticed a negative correlation between stalk number and Nagarajan (1983) observed a negative association between stalk number and stalk length.

Kang *et al.* (1983) reported a significant positive association of plant height with stalk diameter and stalk weight. Highly significant negative association of stalk diameter with stalk number and stalk number with stalk weight was observed.

Ramana Reddy (1985) observed highly significant positive association between cane weight and cane length, While Reddy and Reddi (1985) in a study including twelve varieties, found highly significant positive association of stalk number with stalk length and average length of internode and significant negative association with stalk diameter, stalk volume and stalk weight. The association between stalk length and average length and internode, stalk diameter and stalk volume was found positive and highly significant.

A highly significant positive association of plant height with number of internodes and cane thickness was reported by Saikh *et al.* (1986). While Tehlan *et al*. (1986) reported significant negative association between plant height and stalk diameter and number of canes and number of internodes. Sekhar (1986) observed highly significant positive association between character pairs, cane length and number of internodes; number of internodes and single cane weight; single cane weight and cane length; stalk diameter and single cane weight.

Significant positive association between number of internodes and cane thickness was indicated by Rekhi and Gill (1987). Ramana Reddy (1988) reported significant positive association of stalk weight with stalk diameter and stalk length. Highly significant positive association of number of millable canes per clump with number of internodes per cane and stalk weight was noticed by Verma *et al.* (1988). He also observed highly significant positive association of stalk weight with number of internodes per stalk girth.

Positive association of stalk weight with diameter and stalk length and also between stalk length and number of internodes was reported by Madhavi *et al.* (1991). Patel *et al.* (1993) observed significant negative association of millable canes/ha with weight and stalk diameter and significant positive association of stalk weight with stalk diameter, Das *et al*. (1996) reported significant negative association of number of millable canes per ha with stalk weight and height of millable cane.

Positive correlation between cane weight and its thickness and negative correlation of NMC with individual cane weight and its length were reported by Das *et al.* (1997a).

In a crop like sugarcane, quality plays an important role as such it is imperative to understand the relationship of quality parameters with cane yield and its components and also with commercial sugar yield.

Number of sugarcane workers Mariotti (1968), Brown *et al.* (1969), Batcha and Sahi (1972), Richard (1975), Khairwal *et al.* (1978), Nair and Somarajan (1984a), Ramana Reddy (1984), Chauhan *et al.* (1987) and Premachandran (1995) observed negative association between sucrose per cent and cane yield.

A negative association between stalk diameter and sucrose per cent was reported by Herbert and Henderson (1959), Herbert (1965), Luna (1965), Richard (1975), Ramana Rao and Rao (1977) and Nageswara Rao *et al.* (1983). Positive association between stalk diameter and brix was reported by Herbert (1965) and Jagathesan *et al.* (1967). Highly negative significant association of sucrose with number of canes per row, cane length and cane yield was reported by Balasundaram and Bhagyalakshmi (1978a).

Positive and significant association between stalk number and brix was noticed by Smith and James (1969). On the contrary, James and Falgout (1969) revealed negative correlation of brix with diameter of stalks.

Strong positive association between brix and sucrose was reported by Herbert (1965), Richard (1975) and Nageswara Rao (1982). Kang *et al.* (1983) revealed highly

significant negative association of stalk diameter with brix, sucrose and purity. Brix had highly significant positive association with sucrose and purity. The association between sucrose and purity was also found highly significant.

Nageswara Rao *et al.* (1983) reported high negative association of brix with stalk diameter at both genotypic and phenotypic levels, while the clump weight and millable stalks per clump had low positive correlation with brix. While studying the association of quality attributes in 41 genotypes Punia and Paroda (1984) found that, CCS per cent had highly significant negative association with length of internode and cane yield per clump but high positive association among brix per cent, sucrose per cent, purity per cent and commercial cane sugar per cent were also observed.

A non-significant positive association of brix with number of stalks, single cane weight, length of millable canes and stalk diameter was reported by Ramana Reddy (1984). In another study in 1985, he reported a highly significant negative association of sucrose per cent with cane yield and its components *viz.,* stalk diameter, stalk weight and stalk length.

Tehlan *et al.* (1986) reported the highly significant and positive association of sucrose with plant height and brix. Brix had highly significant positive association with plant height and number of internodes. Highly significant positive association of brix with sucrose and CCS per cent was observed by Sekhar (1986).

Rekhi and Gill (1987) reported the significant positive inter relationship between sucrose and purity. Verma *et al.* (1988) reported highly significant negative association of sucrose per cent in juice with number of millable canes and number of internodes per cane. Purity coefficient had highly significant positive association with stalk girth and sucrose per cent in juice.

High positive association among brix per cent, sucrose per cent, purity per cent and CCS per cent was noticed by Hooda *et al.* (1989). Sreekumar *et al.* (1994) observed significant and positive correlations both at phenotypic as well as genotypic levels for brix with pol, purity, CCS per cent and CCS/ha, but negative association with millable cane count and yield/ha. Singh *et al.* (1995) observed significant correlation of number of green levels and stalk diameter with brix quality. Das *et al.* (1996) reported significant negative association of number of millable canes per hectare with sucrose per cent in juice and CCS percentage.

2.3.1.3 Association among yield components with CCS yield

Low significant correlation between cane yield per acre and yield of sugar per tonne of cane was noticed by Herbert (1965). Significant positive association of CCS yield with stalk number and stalk weight were reported by Stevenson (1965), Batcha (1975) and Tai *et al.* (1980). Legendre (1970) found a strong positive genotypic association between yield of sugar per acre and its two components, yield of cane per acre and sucrose. Batcha (1975), Sahi and Patel (1975), Balasundaram and Bhagyalakshmi (1978a), Dasado *et al.* (1980), Nair and Somarajan (1984a), Nagarajan (1983) and Sekhar (1986) reported highly significant association between sugar yield and cane yield and also its components.

Balasundaram and Bhagyalakshmi (1978a) reported non-significant negative association between sugar yield and sucrose. Hogarth *et al.* (1981) concluded that progress in breeding for greater yield of sugar per hectare is more likely with selection for greater yields of cane than for higher sugar content.

Reddy and Reddi (1986b) revealed that commercial cane sugar (CCS) yield was largely dependent on cane yield rather than on sucrose per cent in juice. Highly significant and positive association of CCS yield with stalk number per plot and stalk weight and stalk diameter was also observed.

The strong positive and significant association of CCS yield with number of millable canes per clump, number of internodes per cane, stalk weight, stalk girth, sucrose per cent in juice and purity coefficient were reported by Verma *et al.* (1988). Nair and Sreenivasan (1990) reported close association between height at 90^{th} day after planting and sugar yield.

Significant association of sugar yield with cane yield, sucrose per cent in juice, CCS per cent and number of internodes were observed by Patel *et al.* (1993).

Singh *et al.* (1994) observed significant positive correlation of CCS (t/ha) with most of cane yield components including number of leaves per cane. Premachandran (1995) reported either negative correlation or no correlation of sucrose percentage in the juice with commercial cane sugar yield. Das *et al.* (1996) observed significant association of CCS (t/ha) with cane yield, cane height and stalk weight, among the cane yield components and sucrose per cent in juice and CCS percentage among quality parameters. In a similar study, Das *et al.* (1997) also reported significant positive association of sugar yield with cane height at maturity apart from other cane yield components. Kundu and Gupta (1997) observed positive correlation of sugar yield with sucrose, brix and CCS percentage. While Rishipal *et al.* (1998) reported a close association of sugar yield with number of millable canes single stalk weight and stalk height.

In an investigation to study the mean performance of selected sugarcane genotypes for twenty cane yield and juice quality traits in northern dry zone of Karnataka (Tippeswamy *et al.,* 2002) reported genotypes Co 90008, Co 87025 and check variety Co 671 were superior out of sixty genotypes for cane and sugar yield.

Sanjeev Kumar *et al.* (2001) reported the close association of cane yield with number of tillers, number of millable canes per plot, germination percentage, length of inter nodes and single cane weight under moisture deficient conditions, while Kamat and Singh (2002) germinations per cent, number of shoots, LAI, single came weight sucrose per cent may taken into consideration while selecting rainfed tolerant varieties of sugarcane combining high yield and better juice quality based on correlation studies made under rainfed conditions.

Tippeswamy *et al.* (2003a) carried out an experiment to study character association and path coefficient analysis involving sixty sugarcane genotypes for twenty characters, out of these single cane weight, number of millable canes per plot, leaf area and germination per cent had positive and significant association with cane yield and CCS yield.

A field investigation was carried out by Tippeswamy *et. al.* (2003b) to know the character association for morpho-physiological traits in 60 sugarcane genotypes, among 11 characters studied single cane weight, number of millable canes per plot, germination per cent and leaf area were the major contributors to cane yield per plot.

2.3.1.4 Path coefficient analysis

As the number of independent variables influencing a particular dependent variable is increased, there is bound to be certain amount of independence. Due to their mutual association, the development of the dependent variable and their indirect effect exerted through pattern. Under such complex situation, the total correlation would be insufficient to explain the association and for effective manipulation of the characters. Path analysis furnishes, a method of partitioning the correlation coefficient in to direct and indirect effects and measures the relative importance of the factors involved.

Path coefficient analysis, which is simply a standardized partial regression analysis was developed by Wright (1921, 1923 and 1934) and later modified by Dewey and Lu (1959). Path coefficient analysis permits the partitioning of correlation coefficients into direct and indirect effects and gives a more realistic relationship of the character and helps in identifying the effective components. The available literature on path analysis both on cane and sugar yield with their component characters was briefly reviewed.

Bhide (1969) stated that cane yield was directly influenced by number of stalks, stalk height and diameter. However, James (1971) indicated stalk number, stalk diameter and stalk length in that order to be the most important components of cane yield. James and Miller (1971) reported the importance of stalk population in determining cane yield.

Stalk number, stalk length and stalk diameter were reported by Maroitti (1973a) to be the major components of cane yield based on the phenotypic path. But when the genotypic path was considered stalk number only appeared to be the strongly contributing trait to cane yield. Diameter ranked second, while length and density showed negative contribution.

Mariotti (1973b) observed that in one locality, number of stalks had maximum direct effect on commercial cane sugar yield while sucrose, stalk diameter and stalk length appeared to be poor contributors to the cane yield. However in another locality stalk number, diameter and length were proved to be very important contributors to cane yield.

Miller and James (1974) identified stalk population, diameter and length as the primary components of cane yield. The importance of number of millable canes followed by stalk weight on cane yield and sugar yield was stressed by Batcha (1975). Khairwal and Babu (1975) and Khairwal *et al.* (1977) observed negative direct effect of cane height as well as sucrose on cane yield.

The importance of stalk number and stalk weight on cane yield was reported by Balasundaram and Bhagyalakshmi (1978b). They also noticed stalk thickness as a negative component of sugar yield. Khariwal *et al.* (1978) found strong direct effect of stalk number on cane yield followed by stalk thickness and stalk weight, while Hooda *et al.* (1979) indicated maximum direct effect of stalk weight on cane yield.

The positive contribution of stalk weight, number of canes and available sugar ration to the sugar was observed by Nagatomi *et al.* (1982). Kang *et al.* (1983) in their study on genotypic path coefficient analysis indicated plant height to be less important than stalk diameter and stalk number as a component of cane yield, but at the phenotypic level all the three characters were of equal importance. Sucrose per cent had a large direct positive effect on sugar per tonne of cane, whereas brix had a small negative influence. Sangwan (1983) emphasized the maximum weightage to the brix.

The study of Nagarajan (1983) highlighted the significance, consistency and equal contributions of stalk weight and stalk number to sugar yield. Nair and Somarajan (1984b) reported that the millable stalks per plot and single stalk weight were the major components of sugar yield.

High phenotypic and genotypic direct effects of stalk number per plot and stalk weight on cane yield were observed by Reddy and Reddi (1986b). They also indicated that the direct effect of cane yield both at genotypic and phenotypic level was high and positive on CCS yield. The yield components *viz.,* stalk number, stalk length and stalk diameter had low direct effect on sugar yield. Sucrose content on the other hand had low direct effect on CCS yield. Low and negative direct effect of sucrose content on cane yield was also observed.

Tehlan *et al.* (1986) revealed the high direct effect of number of canes followed by stalk height on cane yield. Number of internodes was found to be the most important character contributing directly towards higher sucrose percentage. Purity percentage contributed to higher sucrose percentage indirectly through number of internodes.

Chauhan *et al.* (1987) reported that the number of millable canes per clump had high positive direct effect towards cane yield even via sucrose per cent in juice. The sucrose per cent in juice had highest direct effect on yield, medium effects via millable canes and low via tillers per clump.

Highest positive effect of purity of juice on sucrose content was reported Rekhi and Gill (1987). Indirect effects of sucrose via purity of juice were higher as compared to the indirect effects via other characters.

Based on phenotypic and genotypic path analysis study Ramana Reddy (1988) indicated that stalk diameter and stalk density were equally important for stalk weight.

Highest direct positive effect of CCS (t/ha) on cane yield followed by NMC per hectare, stalk weight and stalk diameter was reported by Das *et al.* (1996). Indirect components *i.e.,* individual cane weight and cane diameter, CCS and sucrose percentage influenced sugar yield (Das *et al.,* 1997). Maximum direct effect of stalk weight on cane yield, followed by number of millable stalks were observed by Das *et al.* (1997). However Bakshi Ram (1994) and Rishipal *et al.* (1998) reported high positive direct effects of NMC and single cane weight on both cane yield and sugar yield.

Tippeswamy *et al.* (2003a) carried out an experiment to study path coefficient analysis involving sixty sugarcane genotypes reported that, CCS per plot at harvest along with dry matter and number of internodes were the major contributor to cane yield per plot.

A field investigation was carried out by Tippeswamy *et al.* (2003b) to study the path coefficient analysis of cane yield per plot involving 11 characters and reported that single cane weight and number of millable canes per plot had high positive direct effect on cane yield per plot.

III. MATERIAL AND METHODS

The present investigation was carried out in the Department of Genetics and Plant Breeding, University of Agricultural Science, Dharwad during 2004-05. The field experiment was conducted at Agricultural Research Station Sankeshwar.

3.1 EXPERIMENT MATERIAL

3.1.1 Histological and histochemical study

For histological and histochemical study two SWA resistant and two susceptible commercial checks were selected.

I) Resistant

1. SNK 192

2. SNK 754

II) Susceptible

1. CoC 671

2. Co 92020

3.1.2 Fixation

Both resistant and susceptible clones leaf sample were collected separately and fixed in formalene, acetic acid and 70 per cent alcohol (FAA) in the ratio of 1:1:18, the material was allowed to remain in the solution for 24 hours.

3.1.3 Dehydration

Fixed material was thoroughly washed in 70 per cent alcohol and further dehydrated by passing through 80 per cent, 90 per cent and absolute alcohol. The dehydration was carried out at least for two hours interval. Further, dehydration was continued using alcohol and paraffin solvent butanol series.

Details of the alcohol, butanol grades are given below

SL. No.	Water concentration (%)	Ethanol (%)	n-butanol (%)
1	30	70	-
2	20	80	-
3	10	90	-
4	-	100	-
5	-	75	25
6	-	50	50
7	-	25	75
8	-	-	100

3.1.4 Paraffin infiltration

For the process of paraffin infiltration paraffin with a melting point of 58 to 68^{0}C was selected. Small quantity of paraffin was added to specimen tubes containing dehydrated plant material for cold infiltration to get at room temperature for 24 hours. Further, specimen tubes were kept in hot air oven maintained at 50^{0}C above melting point, fresh molten paraffin was added at an interval of four hour till last traces of butanol was removed.

3.1.5 Embedding

After the removal of n-butanol from the dehydrated material it was embedded in paraffin wax by adopting paper boat technique. The paper boats of appropriate size were prepared and inner surface of paper boat was smeared with glycerin. The dehydrated plant material with molten wax was poured into the boats immediately followed by pre-boiled molten wax. For the easy cutting of blocks the material was arranged in proper way in linear rows.

3.1.6 Microtoming

From the paraffin blocks containing plant leaf, thin sections of 9 □m thickness were obtained using rotary microtome.

3.1.7 Affixing the sections

Gelatin (one per cent) was used as an adhesive to fix the sections to slides, which was prepared in warm, distilled water. To this small quantity of potassium dichromate crystals were added and later it was filtered and used to fix the sections to slides.

Small amount of gelatin was smeared on the down slides and ribbons of convenient size were spread carefully, these slides were warmed on hot plate maintained at 45^{0}C to further stretch the sections. The sliders were tilted to remove excess gelatin later the sections were dried at room temperature for 72 hours and stored in a clean slide box.

3.1.8 Staining procedure for anatomical studies

The sections were stained by using a combination of stains for getting anatomical observations and photographs of tissue sections.

3.1.9 Deparaffinizing and hydrating the sections

Sections were deparaffinized by using xylene and were later hydrated using the alcohol series. As per the requirement, hydration of the sections were carried out following the steps listed below.

Chemical	Duration of the treatment
Xylene	5 minutes
Xylene + alcohol	5 minutes
Absolute alcohol	5 minutes
90 per cent alcohol	5 minutes
70 per cent alcohol	5 minutes
50 per cent alcohol	5 minutes
Water	5 minutes

3.1.10 Preparation of Safranin stain

A stock solution of safranin was made by dissolving 1 g of safranin in 100 ml of 50 per cent alcohol

3.1.11 Preparation of fast green stain

Fast green stain was prepared by adding 0.5 g of fast green in a mixture of 100 ml of 95 per cent alcohol.

3.1.12 Preparation of clove oil mixtures

Clove oil mixture was prepared by mixing of 50 per cent clove oil with 50 per cent xylene.

3.1.13 Staining schedule for histological Studies (Safranin and Fast green method)

Procedure

1. The sections were deparaffinized (5 minutes) as describe earlier
2. Stained in safranin for 24 hours
3. Excess stain was washed in running water
4. Gradually dehydrated by passing rapidly in a series of alcohol like 50, 70, 90 per cent and absolute alcohol
5. Counter stained with fast green for a short period of 3 minutes
6. Then passed in 90 per cent alcohol, and absolute alcohol
7. Excess stain was cleared by clove oil mixture
8. Passed in xylene and mounted in DPX
9. Various anatomical observations were recorded (10x) and group mean of resistance and susceptible clones were analyzed using 't' test

3.1.14 Histochemical studies

3.1.14.1 Test for total insoluble polysaccharides

Periodic Acid-Scgiffs (PAS) method was adopted for localization of the total insoluble polysaccharides.

Mild treatment with an oxidative agent was given to the tissue selections with periodic acid before staining with Schiff's reagent. During oxidation, carbon chains of polysaccharides containing one and two glycol groups are broken and are oxidized to aldehyde groups. Aldehyde groups react with basic fushin (n-sulfuric acid) to develop magenta colour complexes.

3.1.14.1.1 Preparation of schiffs reagent

One gram each of basic fuschin and potassium meta bisulphate were dissolved in 100 ml of 0.15 N HCL of 0.2 N HCL. Mixture was shaken for 2-3 hours until dye is concerted to fuschin sulphuric acid and then 300 mg of fresh activated charcoal was added and shaken for at least 30 minutes and filtered through hard filter paper. Care was taken to see that filtrate was clear and colourless and the collected Schiff's reagent was stored in refrigerator.

3.1.14.1.2 Staining schedule

a) The slides were deparafinized and hydrated as earlier described

b) Slides were treated with an oxiditative reagent (0.5 g of periodic acid dissolved in a solution of 100 ml of distilled water)

c) Stained in Schiff's reagent for 20 minutes at room temperature

d) The sections were rinsed in water and placed them in two per cent sodium bisulphate for 1-2 minutes.

e) Washed thoroughly in running water for ten minutes, later

f) Dehydrated by passing through alcohol and butanol grades and

g) Finally cleared in xylene and mounted in DPX mount

The assessment of polysaccharide constituents in the test was based on the appearance of magenta pink colour and its intensity in tissues.

3.1.14.2 Test for protein

Mercuric bromophenol blue method was adopted to asses the total protein (Maizia *et al.,*1953).

The principle behind this procedure is that basic proteins bind to the bromophenol blue even in the absence of mercury. Therefore excellent correlation exists between the amount of protein and the amount of dye bound.

3.1.14.2.1 Preparation of mercuric bromophenol stain

Ten gram of mercuric chloride and 100 g of bromophenol blue powder were dissolved in 100 ml of absolute alcohol.

3.1.14.2.2 Staining schedule

a) The sections were deparaffinized

b) Brought to absolute alcohol

c) Stained with mercuric bromophenol blue for 20 minutes later

d) Rinsed well in tap water till final blue colour is obtained at the sites of protein

e) Dehydrated in tertiary butyl alcohol (at 2 changes given for 10 minutes) and

f) Cleared in xylene and mounted on DPX mount

The assessment of protein constituents in this test was based on the appearance of blue + bluish green color in the tissues.

3.1.14.3 Test for total RNA

Azure B method was adopted to asses the total RNA in the leaf tissues, the principle behind is when Azure B is used the nucleic acids stain meta chromatic.

3.1.14.3.1 Preparation of stain

0.025 per cent solution of Azure B to be prepared in citrate buffer maintained at pH 4.0 by dissolving 25 mg of Azure B in 100 ml of citrate buffer. To prepare stock solution of citrate buffer 65 ml of 0.2 M citric acid (4.29 g citric acid dissolved in 100 ml of water) was mixed with 35 ml of 0.2 M sodium citrate (5.9 g sodium citrate dissolved in 100 ml of water).

3.1.14.3.2 Staining schedule

a) The sections were deparaffinized

b) Hydrated the sections in a customary way, then

c) Placed the sections in 0.025 per cent solution of Azure B prepared in citrate buffer at pH 4 for 12 hours later

d) Washed in water and dehydrated the sections by using tertiary buytl alcohol (TBA) for 30 minutes at least two changes of TBA given and

e) Passed through xylene and mounted on DPX

The assessment of DNA and RNA constituents in this test is based on appearance of blue colour for DNA and purple for RNA.

3.1.15 Histochemical assessment

Based on the degree of cytochemical reaction with specific reagents for various cellular chemical compounds as observed under microscope and rankings were given as mentioned below

a) Very rich (+++)

b) Rich (++)

c) Low (+)

d) Nil (-)

3.2 BIOCHEMICAL STUDY

For biochemical study nine susceptible including commercial checks and seven SWA resistant clones were used as indicated below.

Sugarcane clones used for biochemical analysis

Sl. No.	Clones	
	Susceptible	Resistant
1.	Co 740	SNK 044
2	CoC 671	SNK 049
3	CoC 7704	SNK 061
4	Co 85019	SNK 158
5	Co 88025	SNK 192
6	Co 88028	SNK 256
7	Co 94012	SNK 754
8	MS 6847	
9	SNK 822	

3.2.1 Collection of samples

The top second leaf was taken with tip discarded and only middle part was taken for biochemical analysis.

3.2.2 Determination of hydroxamic acid

3.2.2.1 Preparation of extracts

Two grams of plant tissue was macerated in nine ml H_2O, filtered through cheesecloth and left 15 minutes at room temperature. The extractant was adjusted to pH_3 with one M Hcl and centrifuged at 1000 RPM for 10 minutes. The supernatant was extracted into ethile ether (2 nol x 3) twice the volume in to 3 times and the organic phases were evaporated to dryness. These extracts were used for quantification of hydroxamic acids.

The data was analyzed using statistical package given by Panse and Sukatme (1967).

3.2.2.2 Quantification of hydroxamic acids

Hydroxamic acid forms with ferric chloride reagent (50 g of $FeCl_3$ 6 H_2O, 500 ml 95% ethanol and 5 ml of 1.5 M HCL) a blue coloured complex whose absorbance was measured at 590 nm. The concentrations of hydroxamic acid were compared for resistance and susceptibility.

3.3 ASSOCIATION ANALYSIS AMONG RESISTANCE CLONES

Association analysis was carried out using ten SWA resistant clones and field observations were recorded at different stages of crop growth.

3.3.1 Experimental detail

The experiment was conducted in medium deep black soil having a depth of 60 cm at Agricultural Research Station, Sankeshwar. Experiment was laid out is a homogenous blocks following Randomized Block Design with three replications, observations were recorded at various stages of crop growth and at harvest.

3.3.2 Observation recorded

3.3.2.1 Germination per cent

The germination per cent was recorded from the gross plot on 45^{th} day after planting and expressed as percentage of the buds planted.

3.3.2.2 Tiller number per plot

Tiller number per plant were recorded at 90 DAP for the SWA resistant clones under study.

3.3.2.3 Leaf area (cm^2)

Leaf area was estimated by measuring length and width of leaves and multiplied with a factor 0.75. From this data leaf area per plant was calculated and expressed in cm^2.

3.3.2.4 Millable cane height (cm)

Millable cane height was recorded at harvest of the crop.

3.3.2.5 Cane girth (cm)

The diameter of millable canes was recorded on the middle internode of the cane by using a vernier caliper at the time of harvest and expressed in centimeters.

3.3.2.6 Number of internodes

The number of internodes present on the stalk up to the last fully opened leaf were recorded in five canes and in each genotype at the time of harvest and expressed as number of internodes per stalk.

3.3.2.7 Single cane weight

The weight of millable canes was recorded at harvest and the average weight was worked out and expressed as weight of millable canes in kilograms.

3.3.2.8 Number of millable canes per plot (NMC/plot)

All the canes from each genotype (net plot) were cut, dressed counted and recorded as number of millable canes per plot.

3.3.1.8 Cane yield per plot (kg)

All the canes in the net plot were cut close to the ground level. The tops and trash were removed, cane yield per plot was recorded expressed as cane yield per plot in kilograms.

3.3.2.9 Commercial cane sugar yield per plot (kg) (CCS yield/plot)

Sugar yield was calculated from commercial cane sugar per cent as

$$\text{CCS per plot (kg)} = \frac{\text{CCS per cent x Cane yield per plot (kg)}}{100}$$

Where,

CCS per cent = [Sucrose per cent – (brix per cent- Sucrose per cent) x 0.40] x 0.75

3.3.3 Quality parameters

3.3.3.1 Brix per cent

Brix reading was made by using the brix hydrometer at 12th month after planting and at the time of harvest. The corrected brix reading was worked out using Bur-standards.

3.3.3.2 Sucrose per cent

It was estimated by Horne's dry lead acetate clarification method using polariscope at 12th month after planting and at the time of harvest (Iswaran, 1981). Hundred ml of the filtered juice was transferred to 250 ml conical flask to which one gram of basic lead acetate was added, stirred well and allowed to stand for about an hour until clear supernatant is obtained. This supernatant is filtered through Whatman No. 40 filterpaper and the clarified juice was filled into a 20 mm polariscope tube and sucrose reading was recorded. The corrected sucrose readings were obtained by comparing the sucrose reading measured with the corresponding corrected brix reading referring to Schmitz table.

3.3.4 Statistical methods

3.3.4.1 Analysis of varience

The mean values of the genotypes in each replication were used for analysis of variance .Replication wise mean values were subjected to RBD analysis (Snedecor and Cochran, 1967). The significance of differences among all the genotypes was tested by 'F' test.

The structure of ANOVA as follows

Source of variation	DF	MSS	Expectations	Variance ratio
Replication (r)	r-1	RMSS	-	
Genotypes (v)	v-1	VMSS	$\sigma^2e + r\sigma^2g$	VMSS/EMSS
Error	(r-1)(v-1)	EMSS	σ^2e	
Total	(rv-1)			

Where,

r = Number of replications

v = Number of genotypes

and

$$SE\ m = \sqrt{\frac{EMSS}{r}}$$

After testing for significance of difference among the means of different genotypes, further computations were done as detailed below.

3.3.4.2 Estimation of correlations

The analysis of variance (ANOVA) and analysis of covariance (ANCOVA) was done by following method described by Cochran and Cox (1957) for all characters.

Sl. No.	Source of variation	DF	MSS	Expected MSS
1.	Replication (r)	r-1	RMSP	-
2.	Genotypes (v)	v-1	VMSP	$\sigma^2e+\sigma^2g$
3.	Error	(r-1)(v-1)	EMSP	σ^2e
Total		Rv-1		

$$COV_{XY}\ (g) = \frac{VMSP - EMSP}{r}$$

$$COV_{XY}\ (e) = EMSP$$

$$COV_{XY}\ (p) = COV_{XY}\ (g) + COV_{XY}\ (e)$$

Phenotypic correlation

$$r_{xy}\ (p) = \frac{COV_{XY}\ (p)}{\sqrt{V_X\ (p) \times V_Y\ (p)}}$$

Genotypic correlation

$$r_{xy}\ (g) = \frac{COV_{XY}\ (g)}{\sqrt{V_X\ (g) \times V_Y\ (g)}}$$

Where,

COV_{xy} (g) = Genotypic covariance between characters X and Y

COV_{xy} (p) = Phenotypic covariance between characters X and Y

COV_{xy} (e) = Error covariance between characters X and Y

V_x (p) = Phenotypic variance of character X

V_y (p) = Phenotypic variance of character Y

V_x (g) = Genotypic variance of character X

V_y (g) = Genotypic variance of character Y

r_{xy} (p) = Phenotypic correlation coefficient among characters X and Y

r_{XY} (g) = Genotypic correlation coefficient among characters X and Y

The significance of correlation coefficients were tested by comparing 'r' values at (n-2) degrees of freedom where, 'n' is number of genotypes tested.

3.3.4.3 Estimation of path-coefficients

The correlation coefficients were further partitioned into direct and indirect effects with the help of path coefficient analysis as suggested by Wright (1921) and Dewey and Lu (1959).

Cane yield per plot was assumed to be dependent variable (effect) which is influenced by other characters, the independent variables (causes) directly as well as indirectly through other characters. The variation in cane yield unexplained by other causes was presumed to have been contributed by residual factor 'R' which is uncorrelated with other factors.

Path coefficients were obtained by solving the simultaneous equations which express the basic relationship between correlations and path coefficients. The equations were as follows.

$r_{1,y} = P_{1,y} + r_{1,2} P_{2,y} + r_{1,3} P_{3,y} + \ldots\ldots\ldots\ldots\ldots\ldots + r_{1,1} P_{1,y}$

$r_{2,y} = P_{2,y} + r_{2,1} P_{1,y} + r_{2,3} P_{3,y} + \ldots\ldots\ldots\ldots\ldots\ldots + r_{2,1} P_{1,y}$

$r_{3,y} = P_{3,y} + r_{3,1} P_{1,y} + r_{3,2} P_{2,y} + \ldots\ldots\ldots\ldots\ldots\ldots + r_{3,1} P_{1,y}$

„

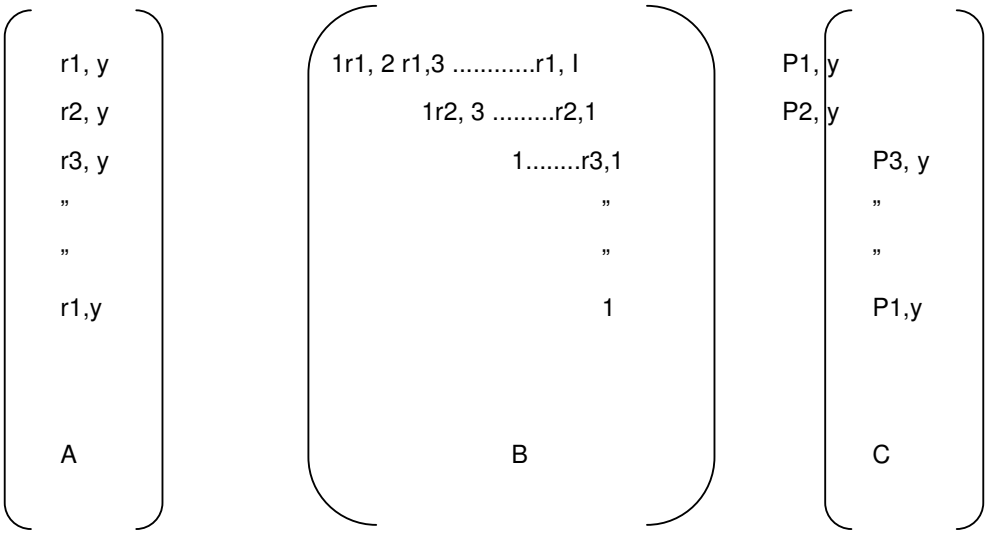
„

„

$r_{I,y} = P_{I,y} + r_{I,1} P_{I,y} + r_{1,2} P_{2,y} + \ldots\ldots\ldots\ldots\ldots\ldots + r_{I,18} P_{I,y}$

where, r,y to $r_{I,y}$ denote the correlation coefficients between independent characters 1 to I and dependent character 'y', $r_{1,2}$ to $r_{1,y}$ denote the correlation coefficients between all possible combinations of independent characters and $P_{1,y}$ to $P_{I,y}$ denote the direct effects of characters 1 to I on character y.

The above equation can also be written in matrix form as shown below



‘B’ matrix was inverted and inverted ‘B’ matrix was multiplied by ‘A’ matrix to obtain path coefficients. Residual factor which measures the contribution of rest of the characters of the usual scheme was obtained as follows.

$$P_{XY} = \sqrt{V_X(g) \times V_Y(g)}$$

where,

R^2 = --- $P^2_{i,\ y+2i<jPj,yrji}$

Which is the square of the multiple correlation coefficient (R) and is known as coefficient of determination.

IV. EXPERIMENTAL RESULTS

4.1 HISTOLOGICAL AND HISTOCHEMICAL STUDY

Sugarcane woolly aphid is one of the major sucking pests and it sucks the sap piercing through stomata of the lower leaf surface, so it is important to study the leaf morphology and anatomy to tackle the pests.

Leaf constitute one of the three basic organs of the plant, the other two being stem and roots, leaves are vital source of photosynthesis makes them abundant and are conspicuous features of our biotic community.

A large proportion of monocots have sessile leaves with a relatively broad, more or less sheathing base. The outer most layer of cells which extend all over the surface of the leaf is called as epidermis, the interior of leaf between upper and lower epidermis is called as ground tissue (mesophyll) in which the vascular bundles are embedded. The foliage leaves vary greatly in their internal structure and the differences are resulted in evolutionary adoptions and insect resistance, the typical leaf consist of following structures.

4.1.1 Epidermis

The epidermis, which is usually a single cell layer in thickness, protects the delicate mesophyll tissues against mechanical injury, loss of water, infection and other harmful influences and cells are filled with transparent cell sap and this outer walls are often marked by thickened and some what convex, epidermis consists of four important components *viz*.

a) Ordinary epidermal cells

b) Guard cells and stomata

c) Trichomes

d) Unusual epidermal cells

a) Ordinary epidermal cells

Ordinary epidermal cells lie between the more specialized cells of the epidermis and they are typically the most numerous and cover the greatest proportion of the plant body, they can have almost any shape, but they are most often tubular, they are elongated and parallel to the organ. The epidermal cells that lie over the veins of the leaves are usually elongated and parallel to the vein.

An almost universal feature of the epidermal cell is that they firmly attach with each other, intra cellular space do not occur between ordinary epidermal cells and only place present are the stomatal pores.

b) Guard cells and stoma

For successful activity of leaf particularly in the process of food manufacture occurs through interchange of gasses between the inner tissue and the atmosphere, this is possible if the epidermis is unbroken layer but accomplished by the presence of minute opening (Stoma) and the opening should have well guarded guard cells. Guard cells play a vital role in regulating stomata, guard cells are broadly divided into ellipictic and graminaceous. In the graminaceous type of stomata guard cells are characteristically dumbel shaped.

Stoma is the site of entry for various atmospheric gases and pests (insects and pathogens). The insects which are unable to probe through epidermis take the help of stoma to get into entry of the leaf tissue for gathering food.

c) Trichomes

Trichomes are the most beautiful and fascinating structure of plant epidermis. The term trichomes is applied to an artificial grouping of all cells that project out of the plane of

Resistant Clones

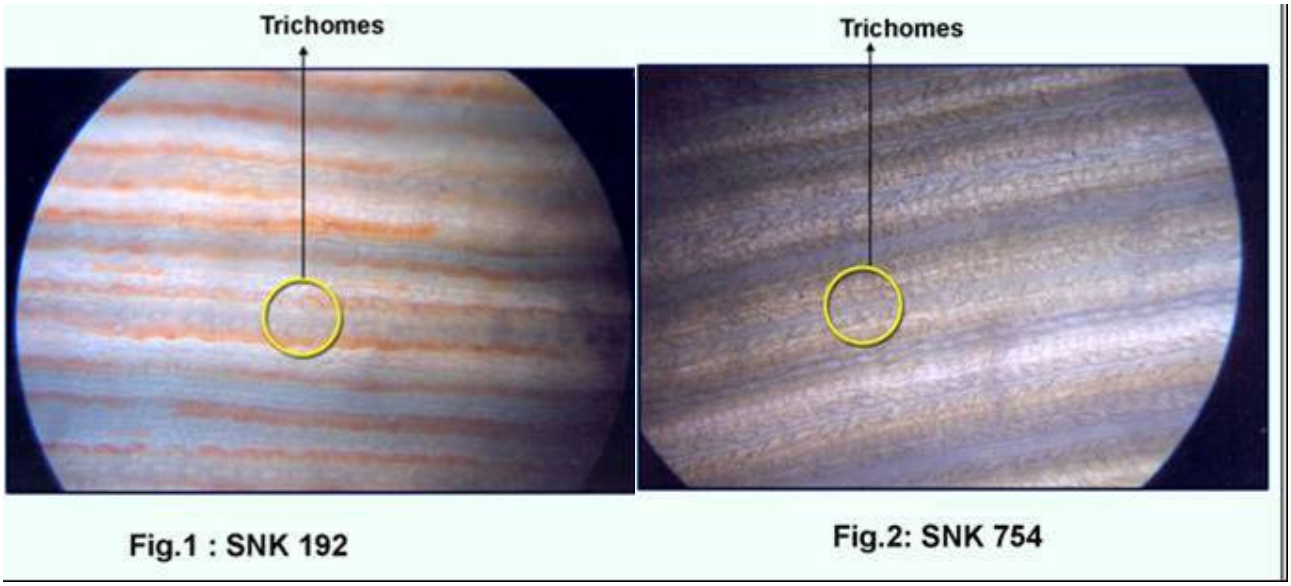


Fig.1 : SNK 192

Fig.2: SNK 754

Susceptible Clones

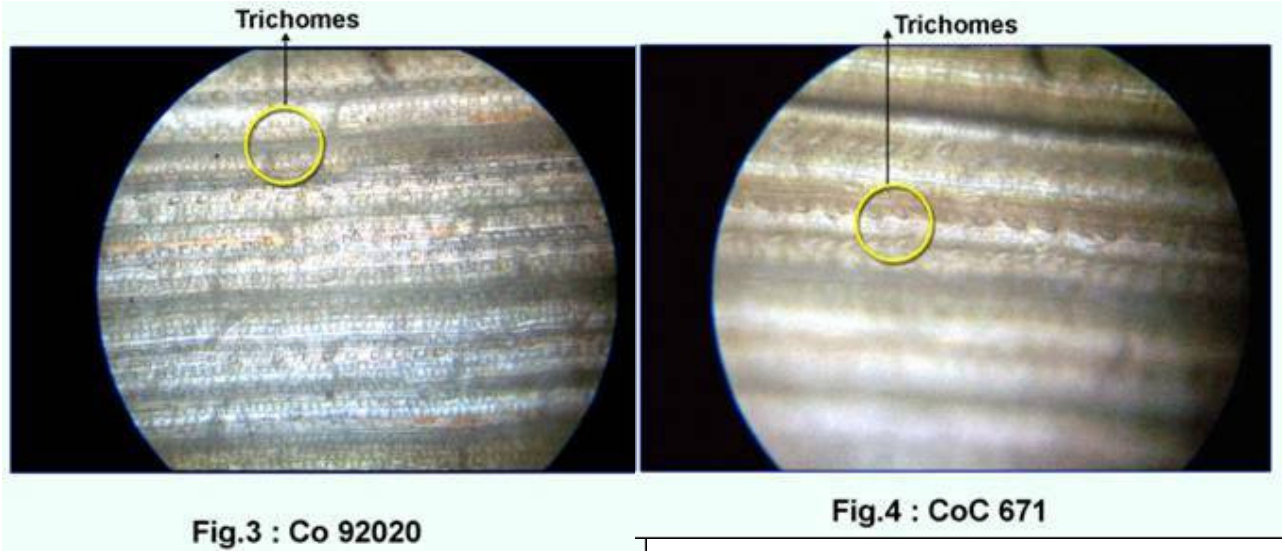


Fig.3 : Co 92020

Fig.4 : CoC 671

Plate 1. Comparative arrangement of trichomes in lower leaf epidermis of resistant and susceptible clones

epidermis. Trichomes furnish a rich field for morphogenetic investigations because of their great diversity and superficial position and relatively simple nature.

Trichomes assist the epidermis in its protective capacity, the functions of the trichomes are wonderfully diverse, the glandular trichomes can secrete water, salt, nector, terpine, adhesives and digestive enzymes. The non-glandular trichomes along with waxes protect against excessive sun light. As they die and dehydrate, their walls become more refractile and scatter light, such a structure is also deterrent against insects because of the hairs can tangle the insect to feed, the trichomes are of little nutritive value to insects.

d) Unusual epidermal cells

(i) Silica cells

Epidermis of grass species contain small and long cells usually grouped together as pairs, some times long cells may also be present, short cells may be either modified as silica or cork cells.

The silica cells contain silica bodies with various shapes (round, elliptical, dumbel and radial shape). These are extremely useful in taxonomy and mostly occur in monocot families.

(ii) Bulliform cells

The epidermis of many monocots, specially the grass and sedges may contain a specialized type of cells are called as bulliform cells. They are very large thin old epidermal cells arranged in long band parallel to the length of the leaf, they are turgid and swollen when leaf is open and flat, when they loose water the leaf folds minimizing its exposed area, during drought situations, but the primary role of bulliform cells is to unroll the young leaves when it is in the bud.

The epidermis of sugarcane exhibits typical layer of cell which are quite different in upper and lower epidermis.

In the upper epidermis ; Three types of zone are present.

a) A central zone consisting of long cells alternating with short groups, often with one or more of the central row composed only of short cells.

b) Stomatal zone, flanking the central zone

c) Marginal zones, bordering the stomatal zone

A band made up of one central zone, two adjutant stomata zones and two marginal zones constitutes one wide unit.

The pattern of the lower epridermis of the leaf bands is made up of two kinds of bands, the vein zones or costal zones overlaying the fibro vascular bundles. De Grott (1941) has studied the structure of the lower epidermis in order to estimates its value for the description of varieties. The intercostal bands are mainly made up of stomatae, short cells (cork cells and sometimes silica cells) and cells of intermediate length.

The costal bands are made up of long cells and cells of intermediate length, together with spine cells and silica cells the pattern of the costal bands exhibit greater variation than that of inter costal bands and they may be of some importance in the description of varieties. De Grott (1941). The great diversity in the pattern of the costal bands can be traced back to three basic types.

Type – I : Characterized by lack of spines, occurs very rarely *i.e.,* galgah nobilization.

Type – II : Characterized by the presence of central row of spines

Type – III : The costal bands are flanked at both sides by a row of spines.

Lower epidermis of resistance and susceptible clones (Plate 1) shows the epidermis of resistant clone covered with spines and stomata, in the form of zones (costal bands) on which spines are present and inter costal bands alternate with each other, in SNK 192 (Fig. 1) type II epidermis is present were spines are relatively small and not flanked on the stomatea

and SNK 754 clones (Fig. 2) type III epidermis is present *i.e.,* the costal bands are flanked at both sides by row of spines.

While, susceptible clones spines *viz.,* Co 92020 (Fig. 3) and Co 92020 had type II epidermis where spines are relatively very small compared to all clones under study and are not flanked on the stomatae in CoC 671 type III epidermis is present where spines are flanked on the stomatae.

4.1.2 Ground tissue

Ground tissue is very broad term were the vascular bundles are embedded and mostly made up of mesophyll tissue (parenchyma cells).

The term parenchyma refers to a tissue composed of living cells variable in their morphology and physiology but are generally thin walled and polyhedral in shape. It is the foundation of the plant body in the sense that parenchyma appears as a ground substance in which other tissues are embedded.

This tissue is the principle seat of essential activities of the plant such as photosynthesis, assimilation, respiration and secretion. Parenchyma cells are highly complex in the physiology as they possess living protoplasts.

4.1.2.1 Structure and content of parenchyma cells

Most of the parenchyma cells eg. that contain chloroplast are called as chlorenchyma and those act as storage cells usually have thin primary walls. The photosynthetic parenchyma contain usually single or numerous vacuoles, sugars, other carbohydrate products and tannins are present in vacuoles.

4.1.3 Vascular bundles

The arrangement of the vascular bundles *i.e.,* the venation imparts a characteristic appearance to leaves, monocots have parallel venation where bundles of relatively uniform size are arranged longitudinally, these veins are laterally interconnected by a small bundles throughout the leaf blade.

In monocots the longitudinal bundles may be of almost equal thickness, vary in size, the larger veins alternating within the bundles of various sizes showing quantitative and qualitative differences. The largest bundle contains xylem and phloem in comparable proportion, in C_4 plants the special significant feature of vascular system is close association with mesophyll and presence of bundle sheath. The vascular bundle consists of xylem, phloem and phloem fibre.

Xylem

Xylem is a complex tissue that has many functions in plants *i.e.,* conduction of water, mechanical support and storage of water

Phloem

The phloem is the principle food conducting tissue of the vascular plants, the phloem is composed of several kinds of cells concerned with different functions there fore it exemplifies a morphologically and physiologically complex tissue.

Phloem sap contains between 50 and 300 mg dry matter per mm and 80 to 90 per cent of this is sugar, amino acids which occur at concentrations of 20 to 80 mg/ml and other components were organic phosphates, phloem consists of sieve tubes and companion cells, sieve tubes are involved in translocation of food material.

Phloem is surrounded by phloem fibre a specialized sclerenchymatus tissue present around the phloem and above the bundle sheath, phloem fibre is made up of tissues (sclerenchyma cells). The terms sclerenchyma refers to thick walled cells often lignified,

Table 1: Mean microscopic measurements (10x) on leaf anatomical features of sugarcane woolly Aphid resistant and susceptible clones

Sl. No.	Character	Range (mm)	Group mean (mm)		Calculated 't' value
			Resistant	Susceptible	
1	Thickness of leaf	0.651 - 0.871	0.757	0.720	0.946
2	Distance between lower epidermis and phloem (sucking distance)	0.110 – 0.117	0.116	0.114	1.349
3	Distance between large and small vascular bundles	0.045 - 0.078	0.055	0.059	2.244**
4	Distance between large and medium vascular bundle	0.157 - 0.239	0.187	0.230	5.859**
5	Distance between medium and small vascular bundles	0.096 - 0.184	0.118	0.124	14.944**
6	Width of large vascular bundle	0.241 - 0.296	0.281	0.272	1.319
7	Width of phloem	0.081 - 0.119	0.118	0.084	28.485**
8	Width of parenchyma cells	0.010 - 0.034	0.025	0.011	8.690**
9	Thickness of phloem fibre	0.017 - 0.024	0.021	0.017	18.311**

* Significant at 5%
** Significant at 1%

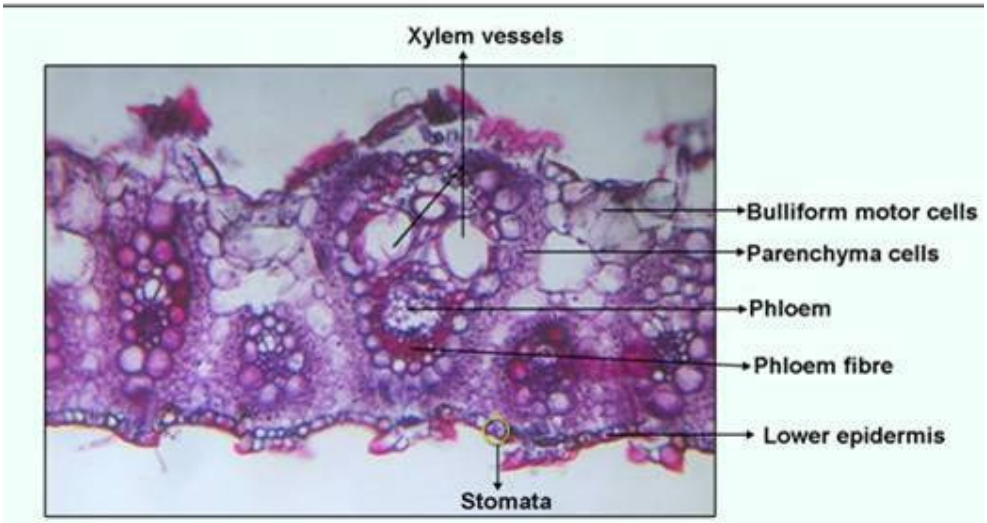


Fig.1

Fig .1

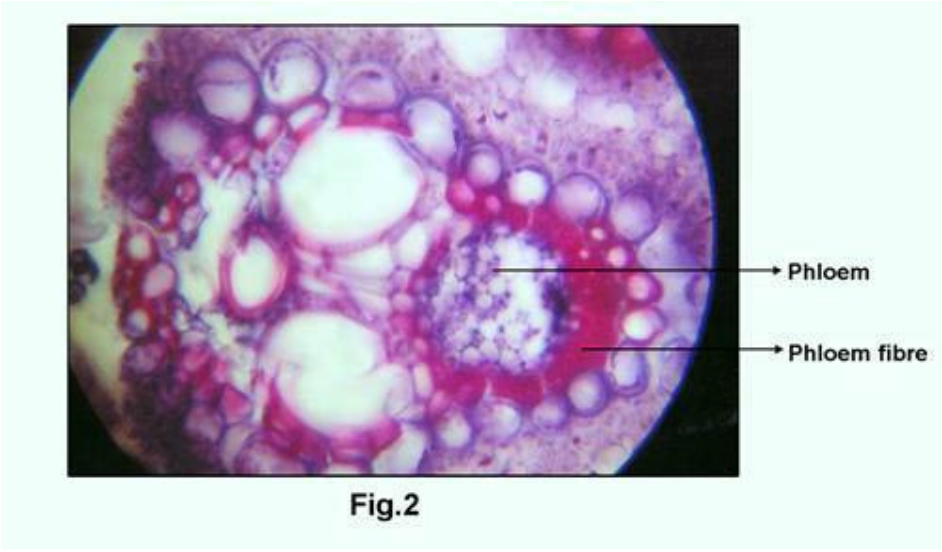


Fig.2

Fig .2

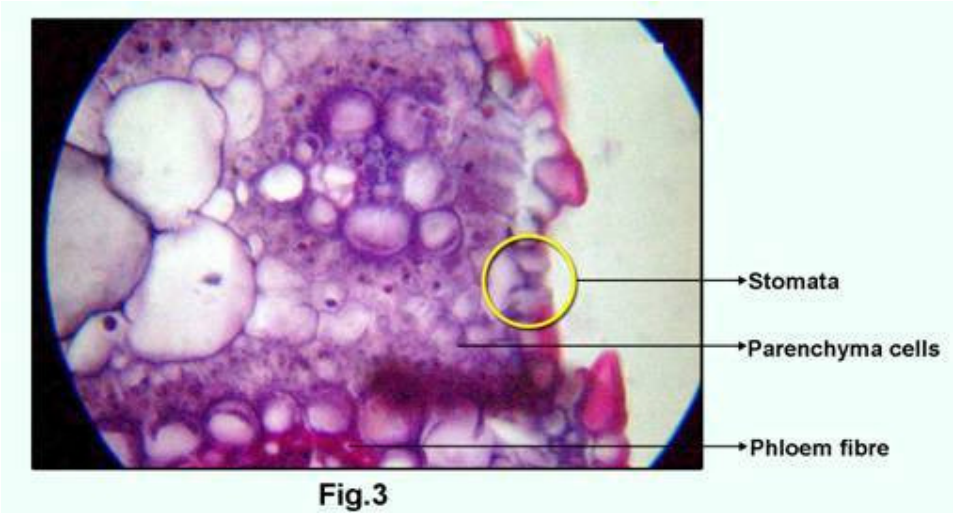


Fig .3

Plate 2. Fig 1: Transverse section of leaf blade of resistant clone SNK 192 with large vascular bundle flanked by two, small and medium ones (10x)

Fig 2: Thick phloem fibre around phloem (45x)

Fig 3: Distance between stomata and phloem separated by more number of parenchyma cells (Sucking distance (45x))

which principle function is to provide mechanical support and protect against invaders to the phloem, to avoid damage to phloem. Sclerenchyma cells exhibit elastic properties unlike collenchyma which exhibits plastic properties. Phloem fibres may be prominent in monocots where they form sheaths (wreath) enclosing the phloem and xylem tissue.

The comparative internal arrangement of leaf tissue between resistance and susceptible clones was studied by a microscopic examination of very thin slices or cross sections cut at right angle to the broad surface of the leaf, indicated increase of several well defined regions in the leaf blade, which can be studied by histology of cross sections of leaf blade under microscope.

4.1.3.1 Leaf anatomical observations

Anatomical observations were made and are tabulated in Table 1 for two resistant and two susceptible clones used in the present investigation.

4.1.3.1.1 Leaf thickness

Thickness of leaf measured as distance between upper and lower epidermis varied among genotypes. The mean distance between upper and lower epidermis varied from 0.651 to 0.871 mm, but was statistically on par with each other irrespective of resistant or susceptible clones.

The group mean distance of resistant clones was 0.757 mm compared to 0.720 mm of susceptible clones and these values were on par with each other.

4.1.3.1.2 Distance between lower epidermis and phloem (sucking distance)

The mean distance varied from 0.110 to 0.117 mm indicating less variation for the distance between lower epidermis and phloem bundle vessels indicating resistant and susceptible clones were on par with each other for this trait under study.

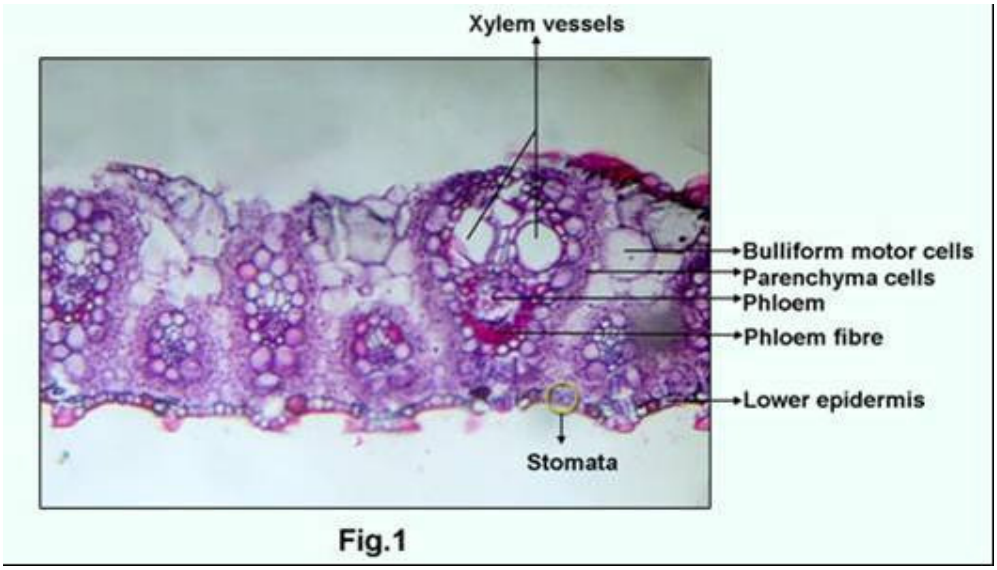


Fig.1

Fig .1

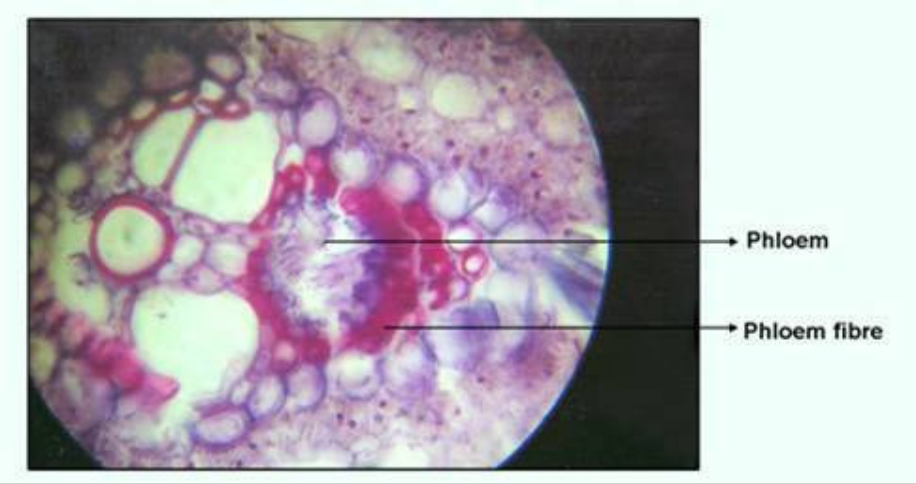


Fig .2

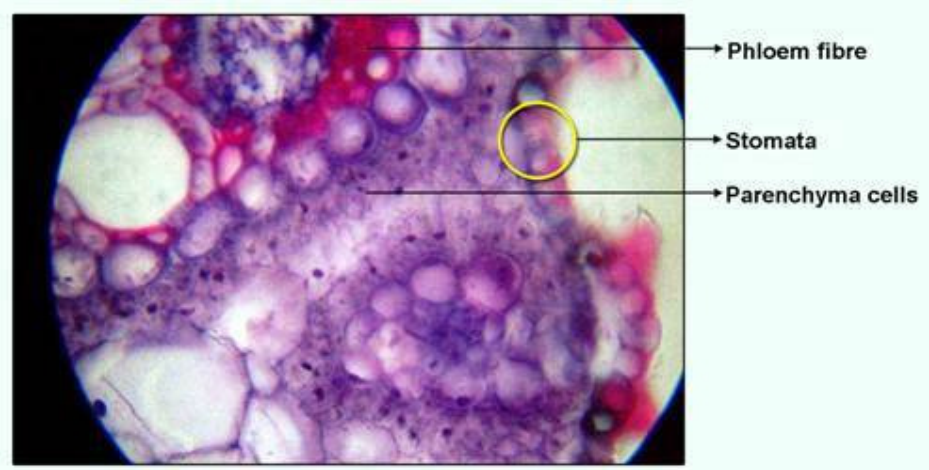


Fig .3

Plate 3. Fig 1: Transverse section of leaf blade of resistant clone SNK 754 with large vascular bundle flanked by two, small and medium ones (10x)

Fig 2: Thick phloem fibre around phloem (45x)

Fig 3: Distance between stomata and phloem separated by more number of parenchyma cells (Sucking distance (45x))

The group mean distance of resistant clone was 0.116 mm compared to 0.114 mm of susceptible clones.

4.1.3.1.3 Distance between large and small vascular bundles

This distance indicates the free space present between large and small vascular bundles. This is site where stylet of aphid enters the internal tissue.

The distance varied from 0.045 to 0.078 mm indicating presence of variation for the space between large and small vascular bundles.

Significant difference between group mean value of susceptible and resistant clones was observed. The group mean of resistant clones was 0.055 mm compared to 0.059 mm for susceptible clones.

4.1.3.1.4 Distance between large and medium vascular bundles

This distance indicates the free space available between large and medium vascular bundles. Space between large and medium bundles differed between resistant and susceptible clones significantly, the mean distance varied from 0.157 to 0.239 mm.

The group mean distance between large and medium vascular bundles of resistant clones was 0.187 mm compared to 0.230 mm of susceptible clones.

4.1.3.1.5 Distance between medium and small vascular bundles

The mean distance varied from 0.096 to 0.194 mm indicating less variation for the distance between small and medium bundle vessels however, group mean differed significantly.

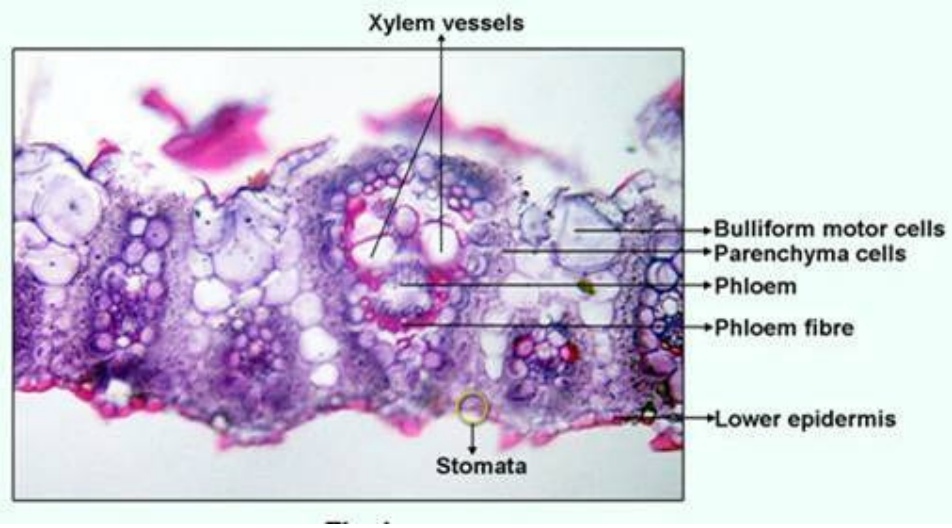

Xylem vessels
Bulliform motor cells
Parenchyma cells
Phloem
Phloem fibre
Lower epidermis
Stomata

Fig .1

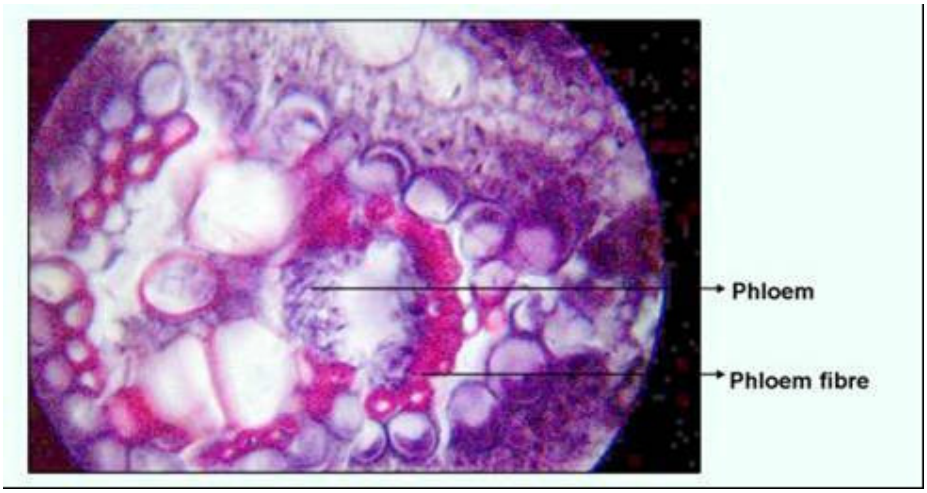

Phloem
Phloem fibre

Fig .2

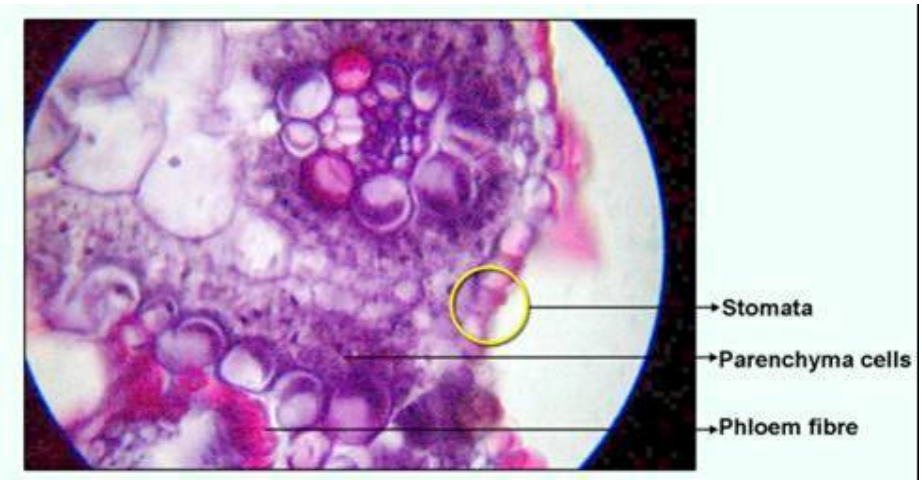


Fig .3

Plate 4. Fig 1: Transverse section of leaf blade of susceptible clone Co 92020 with large vascular bundle flanked by two, small and medium ones (10x)

Fig 2: Thick phloem fibre around phloem (45x)

Fig 3: Distance between stomata and phloem separated by more number of parenchyma cells (Sucking distance (45x))

The group mean distance of resistant clones was 0.118 mm compared to 0.124 mm of susceptible clones.

4.1.3.1.6 Width of large vascular bundles

This is the area where in the xylem and phloem tissues are embedded. There was no significant difference for width of large vascular bundles among the clones. However, mean width varied from 0.241 to 0.296 mm.

The group mean width of large vascular in resistant clone was 0.281 mm compared to 0.272 mm in susceptible clone.

4.1.3.1.7 Width of the phloem

The mean width of the phloem varied from 0.081 to 0.119 mm indicating more variation for the width of the phloem for the resistant and susceptible clones under study, thus the differed significantly.

The group mean width of phloem in resistant clones was 0.118 mm compared to 0.084 mm of susceptible clones.

4.1.3.1.8 Width of parenchyma cells

These are the cells involved in active photosynthesis and are encircling the bundle sheath, which is characteristic feature of C_4 plants.

Width of parenchyma cells around the large vascular bundle revealed presence of significant differences among group mean width of parenchyma cells for resistant and susceptible clones under study. The mean width varied from 0.010 to 0.034 mm indicating presence of variation. The group mean of resistance clones was 0.025 mm compared to 0.011 mm among susceptible clones.

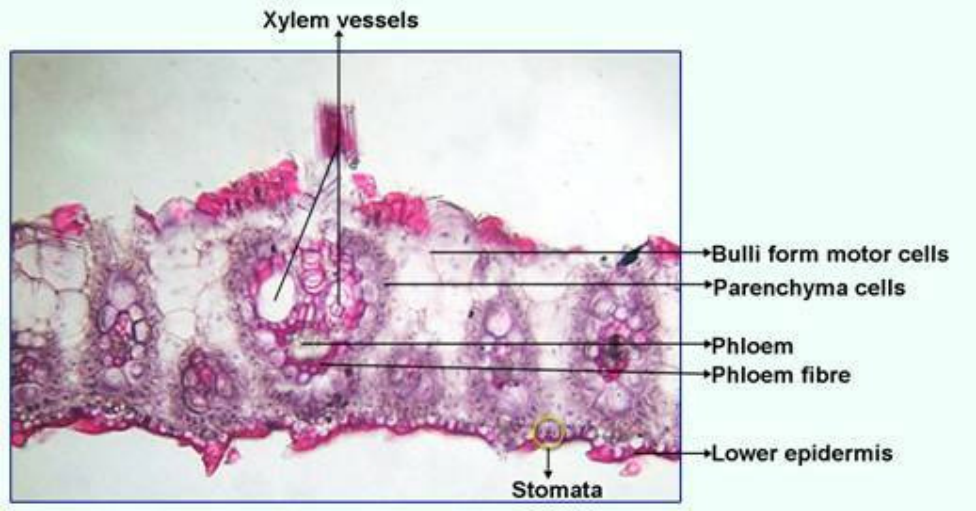

Xylem vessels
Bulli form motor cells
Parenchyma cells
Phloem
Phloem fibre
Lower epidermis
Stomata

Fig .1

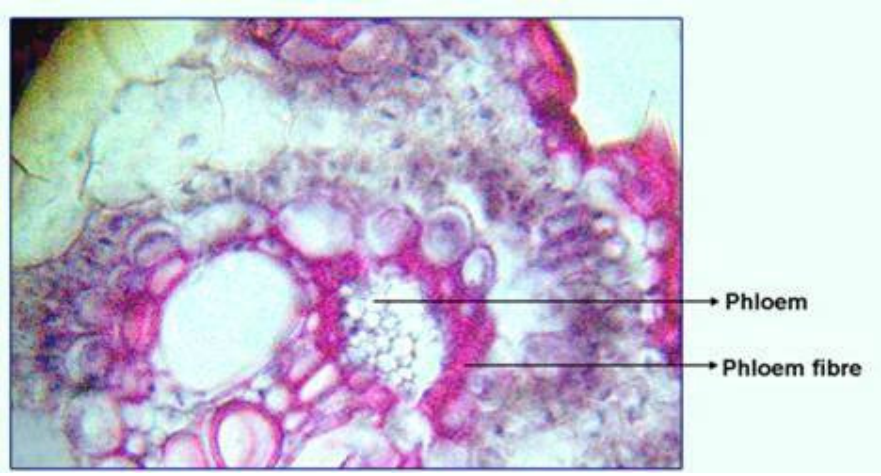

Phloem
Phloem fibre

Fig .2

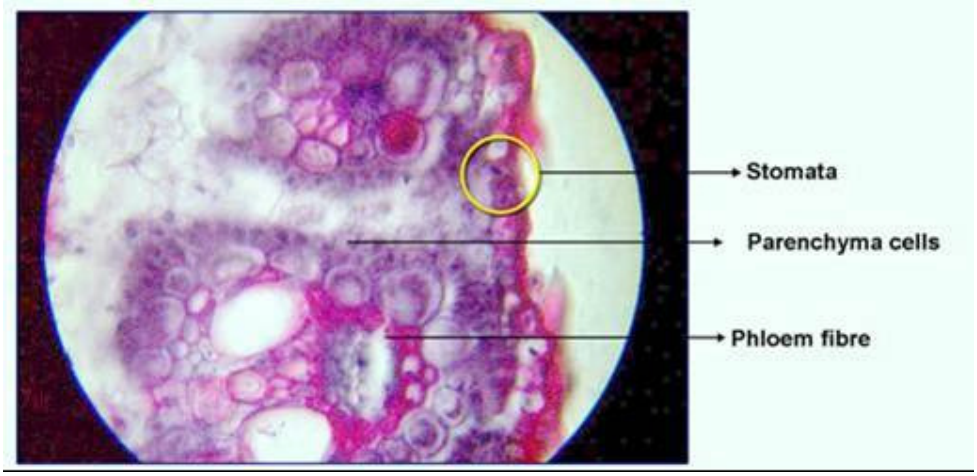


Fig .3

Plate 5. Fig 1: Transverse section of leaf blade of susceptible clone CoC 671 with large vascular bundle flanked by two, small and medium ones (10x)

Fig 2: Thick phloem fibre around phloem (45x)

Fig 3: Distance between stomata and phloem separated by more number of parenchyma cells (Sucking distance (45x))

4.1.3.1.9 Thickness of phloem fibre

Considerable variation for the thickness of phloem fibre surrounding the phloem was observed with mean values ranging from 0.017 to 0.024 mm.

The group mean values of resistance clone was 0.021 mm compared to 0.017 mm of susceptible clones, indicating greater thickness of phloem fibre around the phloem tissues in large vascular bundles.

4.1.4 Histochemistry

Histochemistry is a branch of practical botany wherein the biological molecules like polysaccharides, proteins and nucleic acids are localized in the tissue with the help of specific dyes.

The thin sections obtained after microtoming are exposed to specific dyes, which upon reaction localizes biological molecules in the tissue.

The histochemical results of the sugarcane leaf for various biological molecules are tabulated in Table 2 with plates 6 to 8 and results are explained below.

4.1.4.1 Histochemical study

4.1.4.1.1 Test for total polysaccharides

In general Co 92020 stained very rich (+++) and SNK 192 had low intensity of stain (+), total polysaccharides differed among the resistant and susceptible clones under study (Plate 6). In resistant clones SNK 192 (Fig. 1) had low staining (+) compared to SNK 754 rich (++) clone.

Among susceptible clones Co 92020 and CoC 671 (Fig. 3 and 4) stained very rich (+++). The parenchyma cells around the bundle sheath showed positive for the polysaccharide test (magenta pink) while other cells of the leaf tissue showed negative.

4.1.4.1.2 Test for protein

Total protein differed among the clones under study, protein differed among SWA resistant and susceptible clones. In resistant and susceptible clones protein was localized around the bundle sheath cells *i.e.,* parenchyma cells while phloem stained very low (light green). However, in resistant clones SNK 754 stained very rich (+++) compared to SNK 192 which stained very low (+) whereas among both susceptible clones Co 92020 and CoC 671 stained rich (++).

Table 2: Histochemical studies in the leaf tissue of woolly aphid resistant and susceptible clones of sugarcane

Sl No	Test for	Resistant clone		Susceptible clone	
		SNK 192	SNK 754	Co 92020	CoC 671
1	Total Polysaccharide	+	+ +	+ + +	+ + +
2	Protein	+	+ + +	+ +	+ +
3	Total RNA	+ +	+ +	+ + +	+ + +

Histochemical grade:
Very rich: + + +
Rich: + +
Low: +

4.1.4.1.3 Total RNA

Total RNA differed between resistant and susceptible clones under study and intensity varied from very rich to low.

Resistant clones SNK 754 and SNK 192 stained rich (++) compared to Co 92020 and CoC 671 stained very rich (+++).

All the parenchyma cells around the bundle sheath stained dark blue, while bundle sheath cells stained light blue, the intensity of stain was more towards lower epidermis in clones Co 92020 and CoC 671.

Resistant Clones

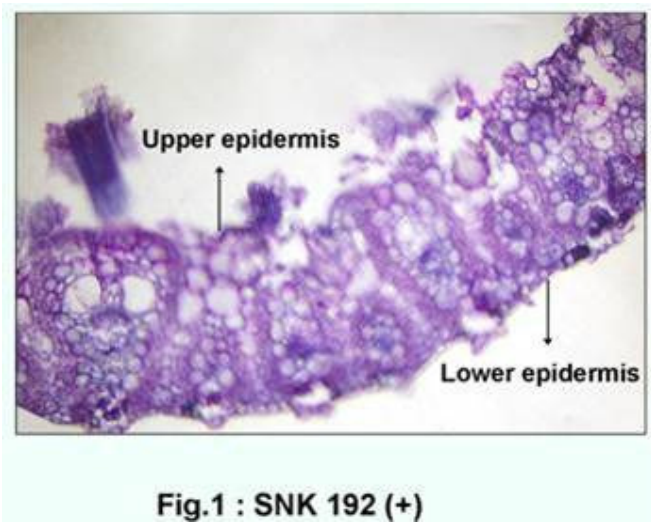


Fig.1 : SNK 192 (+)

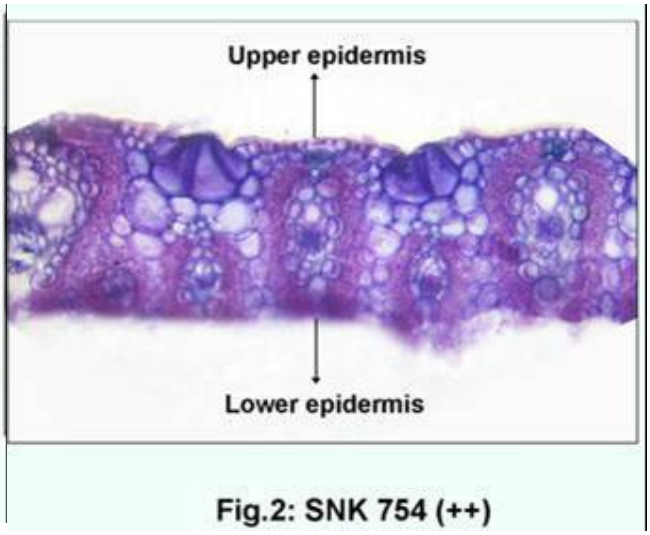


Fig.2: SNK 754 (++)

Fig .1: SNK 192 (+)

Fig .2: SNK 754 (++)

Susceptible Clones

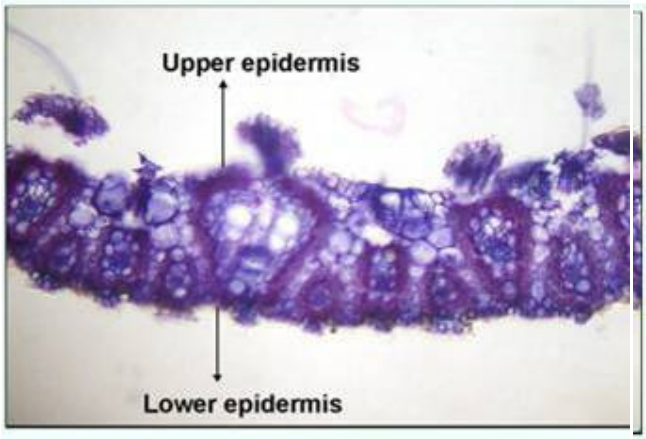


Fig.3 : Co 92020 (+++)

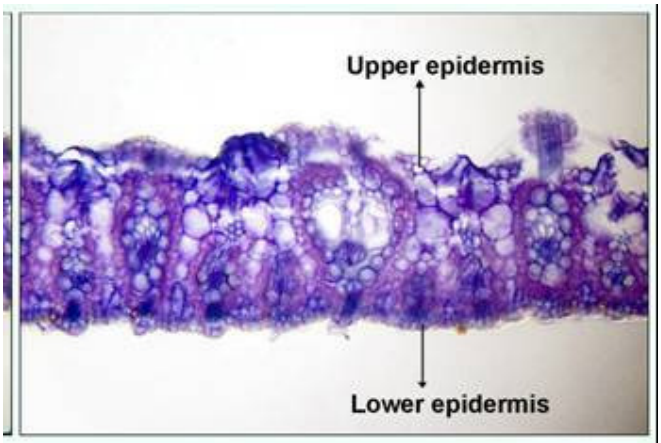


Fig.4 : CoC 671 (+++)

Fig .3: Co 92020 (+++)

Fig .4: CoC 671 (+++)

Plate 6: Histochemical differences for total polysaccharides in transverse sections of leaf blade (10x)

Resistant Clones

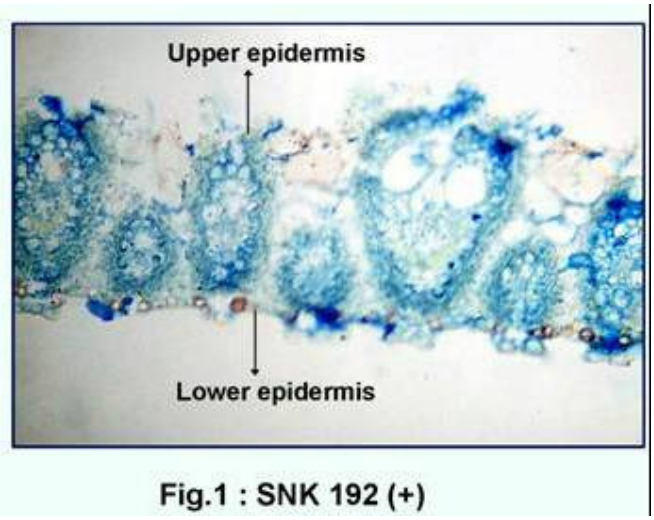


Fig.1 : SNK 192 (+)

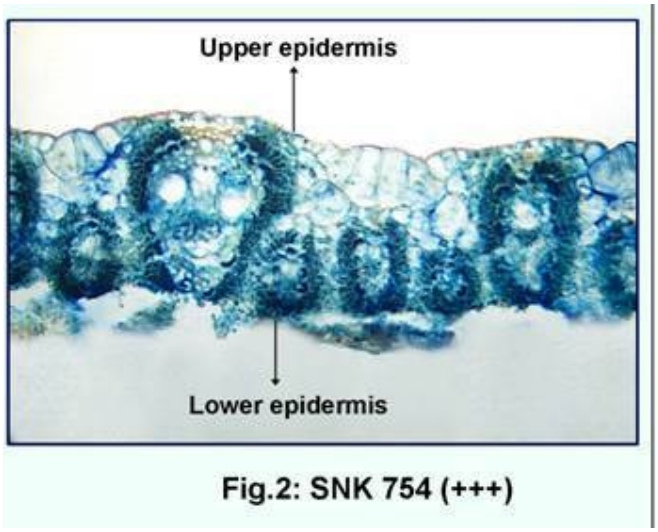


Fig.2: SNK 754 (+++)

Fig .1: SNK 192 (+)

Fig .2: SNK 754 (+++)

Susceptible Clones

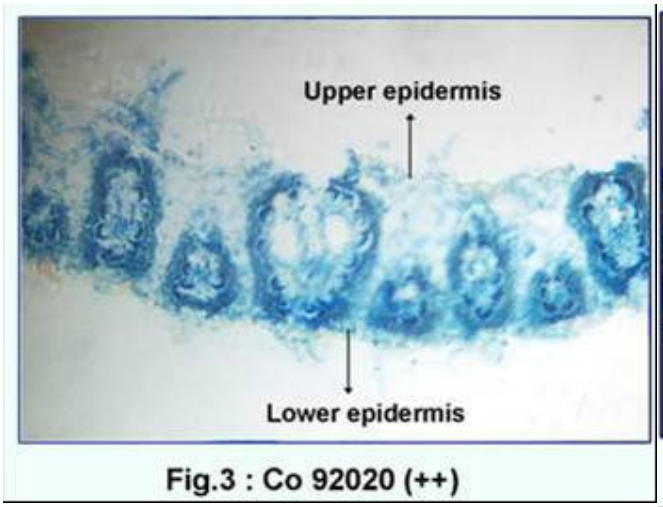


Fig.3 : Co 92020 (++)

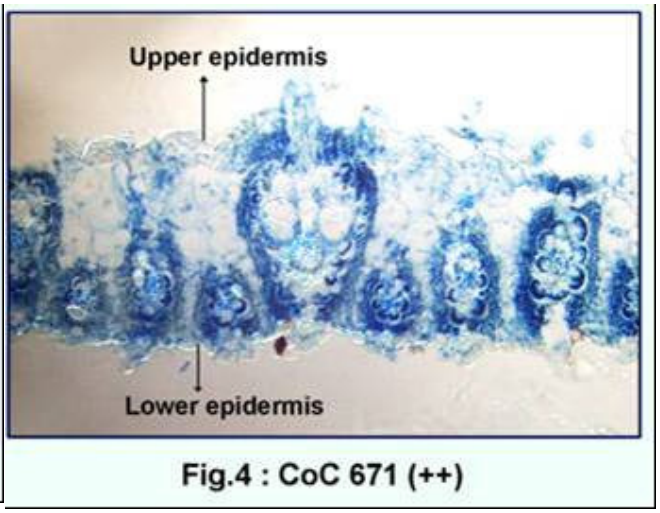


Fig.4 : CoC 671 (++)

Fig .3: Co 92020 (++)

Fig .4: CoC 671 (++)

Plate 7. Histochemical differences for protein in transverse sections of leaf blade (10x)

Resistant Clones

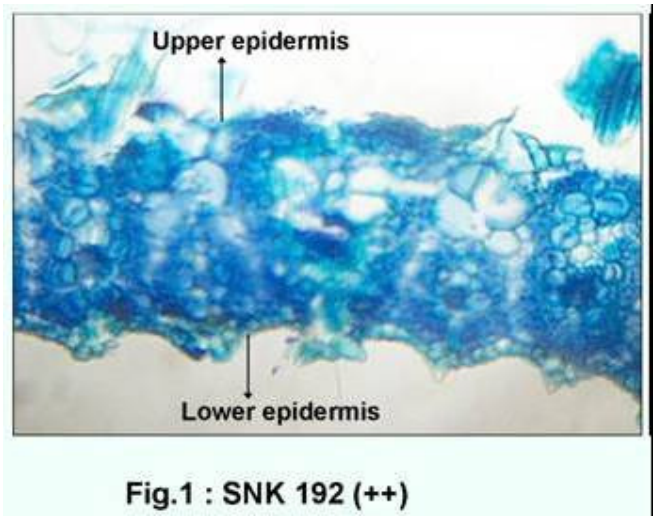


Fig 1: SNK 192 (++)

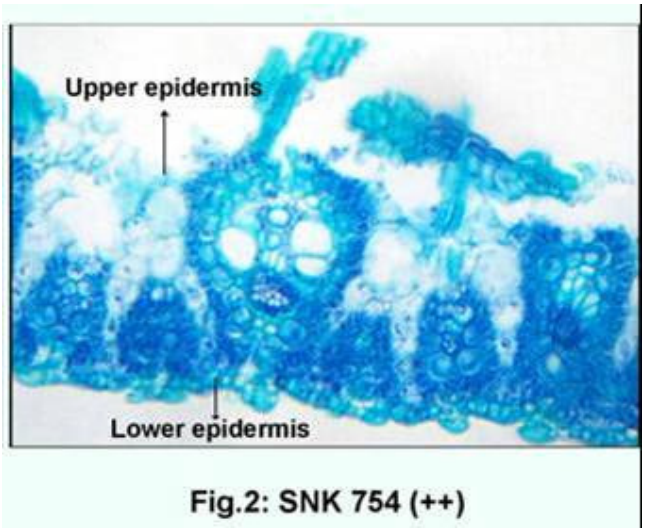


Fig 2: SNK 754 (++)

Susceptible Clones

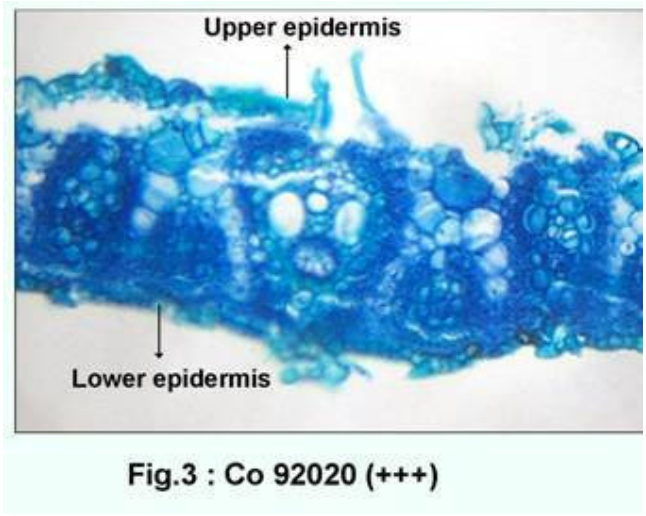


Fig 3: Co 92020 (+++)

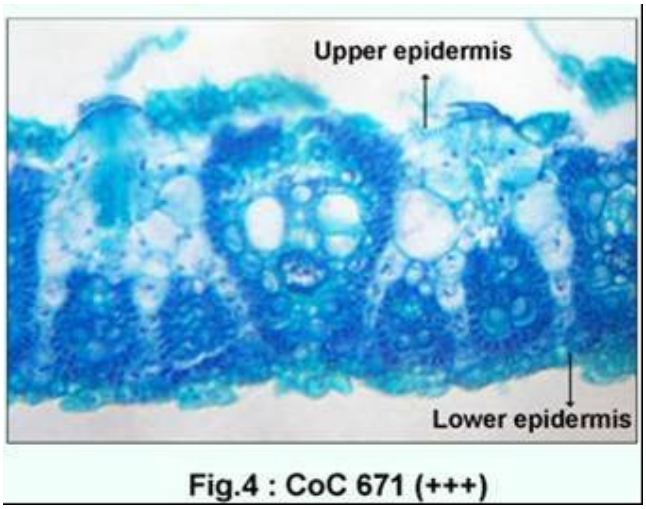


Fig 4: CoC 671 (+++)

Plate 8. Histochemical differences for total RNA in transverse sections of leaf blade (10x)

4.2 BIOCHEMICAL BASIS OF RESISTANCE TO SUGARCANE WOOLLY APHID

Among the most effective weapons developed by plants against phytophagous insects, phytochemicals are noxious, which adversely affect the survival, growth, development and behaviour of insects.

These include a wide variety of chemical molecules ranging from complex molecules such as DIMBOA in wheat, maize and most of the graminae species (Nicol *et al*., 1991), gossypol in cotton, ammonium nitrate in sweet clover and benzyl alcohol in barley (Dhaliwal and Bhathal, 1994).

Hydroxamic acids (Hx) are the group of secondary metabolites present in plant body as glucosides (glucoside a derivative of pyranose sugars in which hydroxyl aldehyde carbon is substituted with another group such as methyl group or phenolic compounds and mostly stored in vacuole) upon hydrolysis these glucosides release aglucons (DIMBOA a form of aglucon) which act as feeding deterrent or toxic effects to insects.

In the present study indirect estimation was made to find out the intensity of hydroxamic acid in the newly developed sugarcane clones due to non-availability of facilities involved in the actual quantification of hydroxamic acids. An attempt was made to quantify the hydroxamic acid based on relative aborbance values of resistant and susceptible clones and is presented in Table 3.

The overall group mean absence value for resistant clones was 0.477 per cent compared to susceptible clones with a group mean of 0.303 per cent. Highest per cent of absorbance values for hydroxamic acid was recorded by SNK 192 (0.588%) followed by SNK 256 (0.512%) among resistant clones compared to Co 740 (0.327%) followed by Co 88025 (0.322), however lowest absorbance value was recorded in SNK 158 (0.396%) among resistant clones compared to susceptible clone CoC 671 (0.270%). All the resistant clones under study recorded significantly higher absorbance values for hydroxamic acid compared to Co 740 which had highest absorbance values among susceptible clones.

4.3 ASSOCIATION STUDIES

The experimental results of present investigation in sugarcane woolly aphid resistant and susceptible clones on their yield and quality parameters are presented under the following sub headings.

4.3.1 Variability

4.3.2 Character association

4.3.3 Path coefficient analysis

The mean values obtained for various characters included in respect of all the clones are provided in appendix I. Analysis of variance for 13 characters included in the present study are presented in Table 4. As evident from table, mean sum of squares for most of the characters in the present investigation were highly significant for most the characters under study, except for millable cane height and juice extract per cent.

4.3.1.1 Germination per cent

The overall mean value of resistant clones for germination per cent was 78.53 per cent and varied from 47.50 to 89.17 per cent showing presence of variation in resistant clones for germination per cent. The clone SNK 44 (89.17%) had the highest germination per cent followed by SNK 256 (88.33%) whereas, SNK 02 (47.50%) showed lower germination per cent followed by SNK 192 (68.75%) and SNK 124 (75.42%). In general most of the resistant clones recorded lower germination per cent compared to commercial checks.

4.3.1.2 Tiller number per plot

Tiller number per plot at 90 DAP exhibited wide range of variation among the resistant clones studied, the mean value ranged from 72.67 to 344.33 per plot with overall mean of 186.70. The clone SNK 256 (344.33%) had the highest tiller number per plot followed

Table 3: Absorbance values of SWA resistant and susceptible clones for hydroxamic acid

Sl. No.	Clones	Mean absorbance (%)
I	**Resistant**	
1	SNK 044	0.410
2	SNK 049	0.475
3	SNK 061	0.484
4	SNK 158	0.396
5	SNK 192	0.588
6	SNK 256	0.512
7	SNK 754	0.478
Group mean		**0.477****
II	**Susceptible**	
1	SNK 822	0.320
2	Co 740	0.327
3	MS 6847	0.298
4	CoC 7704	0.282
5	CoC 671	0.270
6	Co 88028	0.317
7	Co 88025	0.322
8	Co 94012	0.321
9	Co 85019	0.277
Group mean		**0.303**
SE m $\pm$		0.014
CD at 5%		0.044
CD at 1%		0.054

by SNK 61 (241.33%). Whereas, clone SNK 02 (72.67%) recorded lower tillers/plot followed by SNK 192 (89.00%).

Clones SNK 256 (344.33%), SNK 61 (241.33%) and SNK 124 (228.67%) recorded higher tiller number per plot compared to that of best check Co 86032 (204.66%).

4.3.1.3 Leaf area 10th month (cm^2)

The range of mean value of resistant clones for leaf area was 234.88 to 685.15 cm^2 indicating wide variation with an overall mean of 417.80 cm^2. The clone SNK 754 (685.15 cm^2) had higher leaf area followed by SNK 57 (494.96 cm^2), whereas SNK 02 (234.88 cm^2) followed by SNK 44 (302.57 cm^2) recorded lower leaf area.

However, resistant clones SNK 754 (685.15 cm^2) and SNK 57 (494.96 cm^2) recorded higher leaf area than that of commercial checks.

3.3.1.4 Millable cane height (cm)

The overall mean value of resistant clones for millable cane height was 232.00 cm and varied from 163.33 to 283.33 cm. The clone SNK 49 and SNK 256 (283.33 cm) recorded highest millable cane height followed by SNK 124 (270.00 cm) where as the lowest millable cane height was recorded by SNK 192 (163.33 cm) followed by SNK 44 (180.00 cm).

However, SNK 124 (270.00 cm) recorded high millable cane height than superior commercial check CoC 671 (242.66 cm).

4.3.1.5 Cane girth (cm)

Cane girth recorded an overall mean value of 2.44 cm and varied from 1.80 to 2.90 cm. The highest cane girth among resistant clones was recorded by SNK 49 (2.90 cm) followed by SNK 57 (2.87 cm), whereas clones SNK 02 (1.80 cm) and SNK 256 (1.83 cm) recorded lower cane girth. None of the resistant clones recorded higher cane girth than best commercial check CoC 671 (3.00 cm).

4.3.1.6 Number of internodes

Number of internodes exhibited moderate range of variation among resistant clones under study, the mean values ranged from 15.33 to 20.67 with an overall mean of 18.00. Clone SNK 49 (20.67) recorded higher number of internodes followed by SNK 61 (20.00) whereas clone SNK 192 (15.33) followed by SNK 02 (17.00) recorded the lower number of internodes.

None of the resistant clones recorded higher number of internodes than best commercial check.

4.3.1.7 Single cane weight (SCW) (kg)

None of the resistant clones recorded higher SCW compared to best check CoC 671, with a overall mean value of 1.63 kg ranging from 0.53 to 1.75 kg. The highest SCW was recorded by SNK 49 (1.75 kg) followed by SNK 57 (1.58 kg) and the lowest cane weight was recorded by SNK 02 (0.53 kg).

4.3.1.8 Number of millable canes per plot (NMC/plot)

The overall mean value among resistant clones for number of millable canes per plot was 189.60 and varied from 105.00 to 376.67 millable canes per plot.

Clone SNK 256 (376.67 canes/plot) recorded the highest NMC per plot followed by SNK 754 (239.00 canes/plot) and clone SNK 02 (105.00 canes/plot) recorded the lowest NMC per plot followed by SNK 158 (131.67 canes/plot).

All the resistant clones except SNK 02 (105.00 canes/plot) recorded higher NMC per plot then commercial checks.

Table 4: Analysis of variance for different yield and quality characters in SWA resistant sugarcane clones

Source	Df	MSS for the characters												
		Germination per cent	Tillers per plot	Leaf area (cm^2)	Millable cane height (cm)	Cane girth (cm)	Number of internodes	Single cane weight (kg)	Number of millable canes per plot	Juice extract per cent	Brix per cent	Sucrose per cent	Cane yield (kg/ plot)	CCS yield (kg/plot)
Replication	2	240.69	30.69	83.08	2147.18	0.04	0.85	0.08	187.35	100.92	3.44	4.551	1369.813	0.118
Genotype	12	440.20*	13824**	35466.67*	4290.11	0.42*	16.12**	0.479**	21105.10**	51.12	4.01**	2.43*	14933**	197.61*
Error	24	162.40	1428	1176.45	4759.79	0.10	2.67	0.055	3963.80	60.27	1.14	1.00	507.29	12.92
SE		7.35	21.81	19.80	39.83	0.07	0.94	0.135	36.34	4.48	0.61	0.57	13.00	2.07
CD at 5%		21.47	63.68	57.79	115.696	0.20	2.75	0.395	106.10	12.408	1.80	1.69	37.95	6.05
CD at 1%		28.914	86.29	78.30	156.695	0.27	3.73	0.530	143.75	16.750	2.45	2.169	51.43	8.21

* = Significant at 5% level
** = Significant at 1% level

4.3.1.9 Juice extract per cent

The overall mean value of resistant clones for juice extract per cent was 59.07 per cent and varied from 54.33 to 67.00 per cent. The highest juice extract per cent was recorded in SNK 02 (67.00%) followed by SNK 19 (62.00%). Whereas the lowest juice extract per cent was recorded by SNK 124 (54.33%) followed by SNK 158 (55.67%).

However, the best commercial check for juice extract per cent was recorded by CoM 88121 (60.66%) which was lower than resistant clones SNK02 (67.00%), SNK 49 (62.00%) and SNK 61 (61.00%).

4.3.1.10 Brix per cent

The range of mean values for brix per cent was 16.73 to 21.27 per cent, with an overall mean of 19.09 per cent. The highest brix per cent was recorded by SNK 754 (21.27%) followed by SNK 44 (20.83%) and the lowest brix per cent was recorded by SNK 256 (16.73%) followed by SNK 158 (17.90%).

However, none of the commercial checks had higher brix per cent than most of the resistant clones except for SNK 192 (18.10%), SNK 158 (17.90%) and SNK 256 (16.73%)

4.3.1.11 Sucrose per cent

Sucrose per cent recorded moderate variation among resistant clones under study with a mean sucrose per cent of 16.53 and varied from 14.57 to 17.70 per cent. Clone SNK 754 (17.70%) recorded the highest. Sucrose per cent followed by SNK 44 (17.67%), whereas the lowest sucrose per cent as exhibited by clone SNK 256 (14.57%).

However, none of the commercial checks recorded higher sucrose per cent than SNK 754 (17.70%), SNK 44 (17.67%) and SNK 61 (17.63%).

4.3.1.12 Cane yield (kg/plot)

Cane yield at harvest recorded high range of variation among resistant clones under study with overall mean of 223.93 kg/plot and varied from 73.33 to 345.33 kg/plot.

The highest cane yield was recorded by SNK 49 (345.33 kg/plot) followed by SNK 256 (295.67 kg/plot) and the lowest cane yield was recorded by SNK 02 (73.33 kg/plot) however SNK 49 (345.33 kg/plot), SNK 256 (295.67 kg/plot) and SNK 61 (273.00 kg/plot) out beated superior commercial checks under study *viz.,* Co 86032 (207.66 kg/plot), CoM 88121 (206.00 kg/plot) and CoC 671 (191.00 kg/plot).

4.3.1.13 CCS yield (kg/plot)

CCS yield recorded higher variation with overall mean of 25.13 kg/plot and varied from 7.99 to 38.73 kg/plot. Clone SNK 49 (38.73 kg/plot) recorded the highest CCS per plot followed by SNK 61 (33.59 kg/plot) whereas clone SNK 02 (7.99 kg/plot) recoded the lowest CCS followed by SNK 192 (16.07 kg/plot). However, none of the commercial checks recorded higher CCS than top clones mentioned above.

4.3.2 CHARACTER ASSOCIATION

The genotypic and phenotypic correlation coefficients were determined to know the nature of relationship existing between cane yield, sugar yield and their component characters as well as their association among component characters themselves.

The degree of association of different quantitative characters with cane yield and also among themselves at genotypic and phenotypic levels are given in Tables 5 and 6 and results are explained below.

4.3.2.1 Germination per cent

Germination per cent had significant positive association with cane yield (0.985) CCS yield (0.972), NMC per plot (0.883) tiller number (0.793), single cane weight (0.735), number of internodes (0.665) and cane girth (0.638) at genotypic level only.

4.3.2.2 Tiller number

Strong significant positive association of tiller number was recorded with NMC per plot (0.883), germination per cent (0.793), millable cane height (0.785), cane yield (0.676) and CCS yield (0.617) at genotypic level, however millable cane height (0.682) and NMC per plot (0.616) recorded significant association with tiller number at phenotypic level.

4.3.2.3 Leaf area

None of the characters recorded strong significant positive association with leaf area at 10 month DAP at genotypic and phenotypic level, however juice extract per cent recorded strong negative association (-0.681) at genotypic level with leaf area.

4.3.2.4 Millable cane height

Millable cane height did recorded significant association for tiller number per plot (0.785, 0.682) at genotypic and phenotypic level respectively.

4.3.2.5 Cane girth

Cane girth recorded significant positive association with single cane weight (0.937), germination per cent (0.638) and number of internodes (0.610) at genotypic level. However, at phenotypic level single cane weight (0.856) only recorded positive significant association with cane girth none of the characters under study exhibited significant negative association with cane girth at genotypic and phenotypic level.

4.3.2.6 Number of inter nodes

Number of inter nodes recorded strong significant positive association with CCS yield (0.972), cane yield (0.924) single cane weight (0.854), sucrose per cent (0.835), brix per cent (0.793), germination per cent (0.665) and cane girth (0.610) at genotypic level, however at phenotypic level only CCS yield per plot (0.638) recorded positive significant association with number of inter nodes.

4.3.2.7 Single cane weight SCW (kg)

SCW had significant positive association with cane girth (0.937) number of inter nodes (0.854), CCS yield (0.747), germination per cent (0.735) and cane yield (0.668) at genotypic level, whereas at phenotypic level cane girth (0.856) and CCS kg/plot (0.637) recorded positive association with SCW.

4.3.2.8 NMC yield/plot

NMC yield in the present investigation associated (significant positive) significant and positively with tiller number (0.883) and germination per cent (0.812) at genotypic level whereas at phenotypic level tiller number (0.616) recorded significant positive association with NMC yield/plot.

4.3.2.9 Juice extract per cent

Juice extract per cent exhibited significant positive association with brix per cent (0.770) and tiller number (0.714) at genotypic level only, however germination per cent (-0.932) and leaf area (-0.681) recorded significant negative association with juice extract per cent at genotypic level.

4.3.2.10 Brix per cent

Correlation coefficient analysis revealed strong positive significant association of brix per cent with sucrose per cent (0.964), juice extract per cent (0.770) and tiller number of internodes (0.714) at genotypic level, similarity at phenotypic level only sucrose per cent (0.779) recorded significant positive association with brix per cent.

Table 5: Genotypic correlation among yield and yield components in SWA resistant sugar cane clones

Characters	Germination percent	Tillers per plot	Leaf area (cm^2)	Millable cane height (cm)	Cane girth (cm)	Number of internodes	Single cane weight (kg)	No. of millable canes per plot	Juice extract per cent	Brix per cent	Sucrose per cent	CCS yield (kg/ plot)	Cane yield (kg/ plot)
Germination percent	1.000	0.793**	0.499	0.198	0.638*	0.665*	0.735*	0.812**	-0.932**	0.020	0.146	0.972**	0.985**
Tillers per plot		1.000	-0.004	0.785**	-0.209	0.373	0.085	0.883**	-0.714*	-0.461	-0.358	6.17*	0.676
Leaf area (cm^2)			1.000	-0.206	0.528	0.355	0.338	0.100	-0.681*	0.348	0.306	0.349	0.316
Millable cane height (cm)				1.000	-0.214	0.595	0.137	0.499	-0.171	-0.458	-0.324	0.468	0.545
Cane girth (cm)					1.000	0.610*	0.937**	-0.264	-0.315	0.519	0.597	0.590	0.513
Number of internodes						1.000	0.854**	0.091	0.065	0.793**	0.835**	0.972**	0.929**
Single cane weight (kg)							1.000	-0.172	-0.391	0.380	0.559	0.754	0.668
No. of millable canes								1.000	-0.576	-0.372	-0.501	0.478	0.595
Juice extract per cent									1.000	0.770**	0.542	-0.273	-0.341
Brix per cent										1.000	0.964**	0.270	0.113
Sucrose per cent											1.000	0.313	0.166
CCS yield (kg/ plot)												1.000	0.987**
Cane yield (kg/ plot)													1.000

* = Significant at 5% level
** = Significant at 1% level

Table 6: Phenotypic correlation among yield and yield components in SWA resistant sugar cane clones

Characters	Germination percent	Tillers per plot	Leaf area (cm^2)	Millable cane height (cm)	Cane girth (cm)	Number of internodes	Single cane weight (kg)	No. of millable canes per plot	Juice extract per cent	Brix per cent	Sucrose per cent	CCS yield (kg/ plot)	Cane yield (kg/ plot)
Germination percent	1.000	0.537	0.282	0.151	0.352	0.178	0.436	0.211	-0.517	0.027	0.056	0.422	0.469
Tillers per plot		1.000	-0.004	0.0682*	-0.153	0.274	0.120	0.616*	-0.256	-0.285	-0.249	0.472	0.563
Leaf area (cm^2)			1.000	-0.145	0.507	0.208	0.311	0.090	-0.254	0.265	0.217	0.322	0.305
Millable cane height (cm)				1.000	-0.138	0.369	0.254	0.203	0.053	-0.324	-0.201	0.357	0.401
Cane girth (cm)					1.000	0.364	0.856**	-0.268	-0.212	0.380	0.452	0.539	0.475
Number of internodes						1.000	0.480	0.113	0.384	0.199	0.323	0.631*	0.586
Single cane weight (kg)							1.000	-0.245	-0.146	0.234	0.382	0.637*	0.574
No. of millable canes								1.000	-0.036	-0.279	-0.300	0.468	0.590
Juice extract per cent									1.000	0.048	0.011	0.004	-0.014
Brix per cent										1.000	0.779**	0.248	0.081
Sucrose per cent											1.000	0.368	0.138
CCS yield (kg/ plot)												1.000	0.960**
Cane yield (kg/ plot)													1.000

* = Significant at 5% level
** = Significant at 1% level

4.3.2.11 Sucrose (pol) per cent

Sucrose per cent recorded significant positive association with brix per cent (0.964) and number of inter nodes (0.835) at genotypic level. however, at phenotypic level only brix per cent (0.779) recorded strong significant positive association with sucrose per cent.

4.3.2.12 CCS yield/plot

CCS yield associated significantly and positively with cane yield (0.987), number of internodes (0.972), germination per cent (0.972), single came weight (0.754) and tiller number (0.617) at genotypic level, however, at phenotypic level it associated significant and positively with cane yield (0.960), single cane weight (0.637) and number of inter nodes (0.631).

4.3.2.13 Cane yield kg/plot

Cane yield recorded significant positive association with CCS kg per plot (0.987), germination per cent (0.985), number of internodes (0.924), tiller number (0.676) and single cane weight (0.668) at genotypic level recorded significant positive association with cane yield.

4.3.3 PATH COEFFICIENT ANALYSIS

The path coefficient analysis at genotypic and phenotypic levels was worked out for cane yield (kg/plot) and presented in tables 7 and 8 by taking components of cane yield into consideration. Results are presented below with respect to genotypic and phenotypic path coefficient analysis.

4.3.3.1 Direct effects

Out of twelve, five and three traits showed high direct positive effects to cane yield at genotypic and phenotypic level respectively.

The characters which recorded higher positive direct effects to cane yield are CCS yield per plot (0.565), NMC per plot (0.389), cane girth (0.363), millable cane height (0.275) and brix per cent (0.172) at genotypic level. Similarly at phenotypic level CCS yield per plot (0.724), NMC per plot (0.253) and cane girth (0.226) recorded high positive direct effect to cane yield. Whereas, very low positive direct effect was exhibited by number of internodes (0.041), germination per cent (0.009) and juice extract per cent (0.003) with cane yield at genotypic level. However, at phenotypic level low positive direct effect was exhibited by millable cane height (0.096), germination per cent (0.085) and juice extract per cent (0.084) to cane yield.

High negative direct effect was recorded by sucrose per cent (-0.189) and tiller number per plot (-0.163) to cane yield at genotypic level. However, at phenotypic level high negative direct effect to cane yield was exhibited by sucrose per cent (-0.201), whereas low negative direct effect to cane yield was recorded by leaf area (-0.030) and tiller number (-0.020).

4.3.3.2 Indirect effects

4.3.3.2.1 Germination per cent

High positive indirect contribution of germination per cent to cane yield per plot was through CCS yield (0.595), NMC per plot (0.316) and cane girth (0.232) at genotypic level, whereas CCS yield (0.305) recorded high indirect effect on cane yield at phenotypic level.

However, indirect negative contribution was through tiller number (-0.129), single cane weight (-0.022), leaf area (-0.032) and sucrose per cent (-0.028) at genotypic level. Whereas, at phenotypic level germination per cent exerted its indirect negative effect was through juice extract per cent (-0.043) on cane yield.

4.3.3.2.2 Tiller number per plot

Tiller number per plot had its positive effect expressed through CCS yield (0.348), NMC (0.344) and millable cane height (0.216) on cane yield at genotypic level. Whereas, at

Table 7: Genotypic path analysis of cane yield components on cane yield in sugarcane woolly aphid resistant clones

Characters	Germination percent	Tillers per plot	Leaf area (cm^2)	Millable cane height (cm)	Cane girth (cm)	Number of internodes	Single cane weight (kg)	No. of millable canes per plot	Juice extract per cent	Brix per cent	Sucrose per cent	CCS yield (kg/ plot)	rg
Germination percent	**0.009**	-0.129	-0.032	0.055	0.232	0.027	-0.022	0.316	-0.003	0.003	-0.028	0.595	0.985**
Tillers per plot	0.007	**-0.163**	0.000	0.216	-0.076	0.0015	-0.003	0.344	-0.002	-0.079	0.068	0.348	0.676*
Leaf area (cm^2)	0.004	0.001	**-0.065**	-0.057	0.192	0.015	-0.010	0.039	-0.002	0.060	-0.058	0.197	0.316
Millable cane height (cm)	0.002	-0.128	0.013	**-0.275**	-0.078	0.024	-0.004	0.194	-0.001	-0.079	0.061	0.264	0.545
Cane girth (cm)	0.006	0.034	-0.034	-0.059	**0.363**	0.025	-0.028	-0.103	-0.001	0.089	-0.113	0.333	0.513
Number of internodes	0.006	-0.061	-0.023	0.164	0.222	**0.041**	-0.025	0.036	0.000	0.137	-0.158	0.586	0.924
Single cane weight (kg)	0.007	-0.014	-0.022	0.038	0.340	0.035	**-0.030**	-0.067	-0.001	0.065	-0.106	0.422	0.668*
No. of millable canes	0.007	-0.144	-0.006	0.137	-0.096	0.004	0.005	**0.389**	-0.002	-0.064	0.095	0.270	0.595
Juice extract per cent	-0.008	0.116	0.044	-0.047	-0.114	0.003	0.012	-0.224	**0.003**	0.133	-0.103	-0.154	-0.341
Brix per cent	0.000	0.075	-0.023	-0.126	0.188	0.032	-0.011	-0.145	0.002	**0.172**	-0.205	0.152	0.113
Sucrose per cent	0.001	0.058	-0.020	-0.089	0.217	0.034	-0.017	-0.195	0.002	0.187	**-0.189**	0.117	0.166
CCS yield (kg/ plot)	0.009	-0.100	-0.023	0.129	0.214	0.042	-0.022	0.186	-0.001	0.047	-0.059	**0.565**	0.987**

Residue = 0.0100

* = Significant at 5% level
** = Significant at 1% level

Diagonal values – Direct effects

Table 8: Phenotypic path analysis of cane yield components on cane yield in sugarcane woolly aphid resistant clones

Characters	Germination percent	Tillers per plot	Leaf area (cm^2)	Millable cane height (cm)	Cane girth (cm)	Number of internodes	Single cane weight (kg)	No. of millable canes per plot	Juice extract per cent	Brix per cent	Sucrose per cent	CCS yield (kg/ plot)	rp
Germination percent	**0.085**	-0.011	-0.001	0.015	0.080	-0.001	0.003	0.053	-0.043	0.002	-0.011	0.305	0.469
Tillers per plot	0.046	**-0.020**	0.000	0.066	-0.035	-0.001	0.001	0.156	-0.021	-0.020	0.050	0.341	0.563
Leaf area (cm^2)	0.024	0.000	**-0.030**	-0.014	0.115	-0.001	0.002	0.023	-0.018	0.018	-0.044	0.233	0.305
Millable cane height (cm)	0.013	-0.013	0.004	**0.096**	-0.031	-0.002	0.002	0.051	0.004	-0.023	0.041	0.258	0.401
Cane girth (cm)	0.030	0.003	-0.015	-0.013	**0.226**	-0.002	0.006	-0.068	-0.018	0.026	-0.091	0.390	0.475
Number of internodes	0.015	0.005	-0.006	0.035	0.082	**-0.004**	0.003	0.028	0.032	0.014	-0.065	0.547	0.586
Single cane weight (kg)	0.037	-0.002	-0.009	0.024	0.193	-0.002	**0.007**	-0.062	-0.012	0.016	-0.077	0.461	0.574
No. of millable canes	0.018	-0.012	-0.003	0.020	-0.061	0.000	-0.002	**0.253**	-0.003	-0.019	0.061	0.339	0.590
Juice extract per cent	-0.044	0.005	0.008	0.005	-0.048	0.002	-0.001	-0.009	**0.084**	0.003	-0.018	0.003	-0.014
Brix per cent	0.002	0.006	-0.008	-0.031	0.086	-0.001	0.002	-0.070	0.004	**0.070**	-0.157	0.180	0.081
Sucrose per cent	0.005	0.005	-0.007	-0.019	0.102	-0.001	0.003	-0.076	0.008	0.054	**-0.201**	0.266	0.138
CCS yield (kg/ plot)	0.036	-0.009	-0.010	0.034	0.122	-0.003	0.005	0.118	0.000	0.017	-0.074	**0.724**	0.960**

Residue = 0.0120

* = Significant at 5% level

** = Significant at 1% level

Diagonal values – Direct

phenotypic level CCS yield (0.341) and NMC per plot (0.156) contributed positive indirect effect.

Negative effect was exerted through brix per cent (-0.079) and cane girth (-0.076) on cane yield at genotypic level however at phenotypic level tiller number excreted through cane girth (-0.035), juice extract per cent (-0.021) and brix per cent (-0.020).

4.3.3.2.3 Leaf area at 10^{th} month (cm^2)

The major indirect positive contribution of leaf area to cane yield was through CCS yield (0.0197) and cane girth (0.197) at genotypic level, similarly at phenotypic level it was also through CCS yield (0.233) and cane girth (0.115).

Negative indirect effect was through sucrose per cent (-0.058) and millable cane height (-0.057) at genotypic level, whereas at phenotypic level it was contributed through sucrose per cent (-0.044) and juice extract per cent (-0.021).

4.3.3.2.4 Millable cane height (cm)

High positive indirect contribution on cane yield was through CCS yield (0.264) and NMC pre plot (0.194), while it was low through sucrose per cent (0.061) and number of internodes (0.024) at genotypic level, whereas at phenotypic level it was recorded through CCS yield (0.258) and NMC (0.051).

Negative indirect effect on cane yield was observed through tiller number (-0.128), brix per cent (-0.079) and cane girth (-0.078) at genotypic level, however at phenotypic level it was through cane girth (-0.031) and brix per cent (-0.023).

4.3.3.2.5 Cane girth (cm)

The major indirect positive effect of cane girth to cane yield was contributed by CCS yield (0.333) and brix per cent (0.089) at genotypic level, similarly at phenotypic level it came through CCS yield (0.390) and brix per cent (0.026).

Negative indirect effect of cane girth on cane yield was exerted through sucrose per cent (-0.113) and NMC (-0.103) at genotypic level, whereas, at phenotypic level it recorded through sucrose per cent (-0.091) and NMC per plot (-0.068).

4.3.3.2.6 Number of internodes

Path analysis revealed that number of internodes exerted major positive indirect effect through CCS yield (0.586), cane girth (0.222), millable cane height (0.164) and brix per cent (0.137) to cane yield at genotypic level whereas at phenotypic level it was through CCS per plot (0.457) and cane girth (0.082).

Negative indirect effect of internodes to cane yield was through sucrose per cent (-0.158) and tiller number (-0.061) at genotypic level, however at phenotypic level it exerted only through sucrose per cent (-0.065).

4.3.3.2.7 Single cane weight (SCW) (kg)

Single cane weight had its major impact on cane yield through CCS yield (0.422) and cane girth (0.340) at genotypic level. Whereas, CCS yield (0.461) and cane girth (0.193) recorded positive indirect effect to cane yield at phenotypic level through SCW.

Negative indirect effect on cane yield contributed through sucrose per cent (-0.106) and NMC per plot (-0.067) at genotypic level, similarly at phenotypic level it was exerted through sucrose per cent (-0.077) and NMC per plot (-0.062) on cane yield.

4.3.3.2.8 Number of millable canes per plot (NMC/plot)

Positive indirect contribution of NMC to cane yield was through CCS yield (0.270), millable cane height (0.137) and sucrose per cent (0.095) at genotypic level, whereas at

phenotypic level it was exerted through CCS yield (0.339), sucrose per cent (0.061) and millable cane height (0.020).

However, negative indirect effect of NMC to cane yield was through tiller number (-0.144), cane girth (-0.096) and brix per cent (-0.064) at genotypic level and cane girth (-0.061) and brix per cent (-0.019) at phenotypic level.

4.3.3.2.9 Juice extract per cent

A high positive indirect contribution of juice extract per cent to cane yield was expressed through brix per cent (0.133) and tiller number (0.116) at genotypic level, whereas, at phenotypic level very low contribution came through tiller number (0.005) and millable cane height (0.005).

However, high negative indirect contribution came through NMC per plot (-0.224), CCS yield (-0.154) and cane girth (-0.114) at genotypic level, whereas at phenotypic level it was through cane girth (-0.048) and germination per cent (-0.44).

4.3.3.2.10 Brix per cent

Brix per cent had its indirect effects expressed through CCS yield (0.152) and cane girth (0.188) to cane yield at genotypic level similarly at phenotypic level also it recorded through CCS yield (0.180) and cane girth (0.082).

Negative indirect effect of brix per cent to cane yield was expressed through sucrose per cent (-0.205) NMC (-0.145) and millable cane height (-0.126) at genotypic level. Whereas, at phenotypic level it was through sucrose per cent (-0.157) and NMC per plot (-0.070).

4.3.3.2.11 Sucrose per cent

Positive indirect contribution of sucrose per cent to cane yield was through cane girth (0.217), brix per cent (0.187) and CCS yield (0.177) at genotypic level, whereas CCS yield (0.266) and cane girth (0.102) expressed at phenotypic level.

Negative indirect effect of sucrose per cent to cane yield was NMC (-0.195) and millable cane height (-0.089) at genotypic level, however, at phenotypic level its effect expressed through NMC (-0.076) only.

4.3.3.2.12 CCS yield (kg/plot)

The indirect contribution of CCS yield to cane yield was exerted through cane girth (0.214), NMC per plot (0.186) and millable cane height (0.129) at genotypic level, whereas at phenotypic level it exerted through cane girth (0.122) and NMC per plot (0.118).

Negative indirect effect of CCS yield to cane yield was through tiller number (-0.100) and sucrose per cent (-0.059) at genotypic level. However, at phenotypic level it was through sucrose per cent (-0.074) only.

V. DISCUSSION

Sugarcane is one of the important commercial crops in the world, it is main source of energy for man for his food as well as industrial needs. The increased sugar production today is mainly attributed to hybridization programme between cultivated varieties and wild *Spontaneum* types and subsequent nobilization in the past. The hybrid canes of Coiambatore became world famous and are being cultivated all over the world.

Today with the exploding population, particularly in Asia, the need for higher levels of sugar production has became very important. But the production has reached a plateau and efforts are being made to break it, to improve the potential of sugarcane all over the world.

In Karnataka cultivation of sugarcane is spread in four irrigation command areas besides farmers controlled irrigation system over an area of 3.13 lakh ha with a production of 249.18 lakh tonnes of cane. Productivity in the state is higher (79.56 tonnes/ha) than national average and has a potential of 125 tonnes/ha. Cane yield is markedly influenced by many factors like soil fertility, climate, variety, moisture stress, cultivation practices, weeds, insect pest and diseases.

So far, woolly aphid *Ceratovacuna lanigera* Zehntner on sugarcane was considered as a minor or negligible pest of the crop. Zehntner (1897) reported sugarcane woolly aphid, *C. lanigera* from Java for the first time. Its incidence was reported in sugarcane by Department of Agriculture in Athani (Belgaum) during September 2002 in Karnataka for the first time. Now the pest has assumed a serious status on sugarcane crop grown in the Krishna and Ghataprabha river basin. The sugarcane woolly aphid *C. lanigera* belongs to the family pemphigidae and order Homoptera.

It is very difficult to conclude exactly what resistance mechanism and basis operating against sucking pests in crop plants (Victor *et al*., 1994a). However in the present investigation identified SWA resistant and susceptible commercial checks were investigated for the possible basis of resistance to woolly aphid and evaluated for their performance for their cane yield, sugar yield and their components at different stages of crop growth.

The data collected on Histological, histochemical, biochemical, association among characters and their direct as well as indirect effects on cane yield and sugar yields and mean performance were analyzed. The results obtained are discussed under the following sub headings.

1. Histological and histochemical basis of resistance
2. Biochemical basis of resistance
3. Association studies

Plants continue to exist in spite of later evolution of organisms that depend upon plants for their own existence – the herbivores. It is not only the general principle of natural balance but also the evolutionary characteristics of plants that enable them to avoid, tolerance or recover from the injury by insects. This ability of plants to withstand the attack of insect enemies is derived from certain morphological and or biochemical characteristics which exert deleterious effects on the insects (Ghosh, 1995).

Feeding behaviour and food quality of aphid

Most of not all species of aphids feed on the phloem sap of plants, which they obtain by tapping the phloem elements with their stylets. Phloem elements are living cells and are located at some depth within a plant. The contents of these elements (phloem sap) are rich in sugars and relatively poor in amino acids, especially those that are essential for growth and development of aphid (Dixon, 1998).

Aphids after landing on the host scan the surface of a plant accepted as a potential host with the tip of their proboscis. The tactile receptors on the tip of proboscis detect the counters of veins, their preferred feeding site (Tjallingii, 1978). Then they probe into the plant with their mandibular and maxillary stylets, which together form hallow needle like structure.

The sap that passes into the gut of the aphids consists mainly of a concentrated solution of simple sugars and weak solution of amino acids. The low concentration of amino acids would also appears to put a severe constraint on the rate of growth they can achieve (Dixon, 1998).

Aphids can substantially reduce the osmotic concentration of the ingested fluid, by converting mono and disaccharides into trisaccharides like melezitose (Michel, 1942 and Bacon and Dickinson, 1957) and oligosaccharides. Aphids secrete excess sugars in the form of honey dew as they are solely dependent on amino acids and little amount of sugars (Walter and Mullin, 1988, Fisher *et al.,* 1994 and Rhodes *et al.,* 1996). In order to fuel their very high rate of growth therefore, aphids need to process large quantities of food and use the nitrogen it contains effectively for growth and development (Mittler, 1958).

5.1 HISTOLOGICAL BASIS OF RESISTANCE

An attempt was made to study the comparative anatomy of sugarcane woolly aphid resistant and susceptible clones, to know the possible basis of resistance of sugarcane against woolly aphid.

Trichomes play a very vital role in deterring the insects in their normal activities on plant, they become obstacle while moving, sucking egg and young ones laying (Pinnemeyer and Tingey 1987). Among resistant clones SNK 192 had type II epidermis wherein costal bands are characterized by the presence of central row spines. Whereas, in SNK 754 type III epidermis, where stomatae are flanked at both sides by row of spines, however, in susceptible clone Co 92020 type II epidermis is present. Whereas, in CoC 671 type III epidermis is present as type II and type III epidermis are observed both in resistant and susceptible clones and are not confined to any category (resistant and susceptible) of clone, so there may be no role of trichomes in deterring aphids against feeding and various activities , which are contradicting to the observations made by Pinnemeyer and Tingey(1987),but in accordance with the results of Arthur *et al* .,(1987) where they found trichome morphology and density did play any mojor role in imparting resistance against leaf hopper in Common Bean.

In the present study leaf thickness was measured as a distance between lower and upper leaf epidermis which revealed non significant difference among resistance and susceptible group of clones, however group mean thickness of resistant clones was higher, compared to susceptible clones, this indicates varying amount of different tissues making up the internal leaf anatomy. In susceptible clones leaf thickness was less indicating lesser amount of tissue in turn lower number of cells making up the internal leaf anatomy. Interestingly the group mean distance of resistant clones for distance between stomatal and phloem was more compared to susceptible clones even though they did not differ significantly. So it is possible to say that large number of cells and greater distance between stomata of lower epidermis to phloem in resistant clones increase the distance and energy required by the aphid to reach the phloem for gathering food, similar observations was also made by Chandrashekar (1994) wherein he reported that more number of cells in leaves of resistant cotton genotypes compared to susceptible ones for sucking pest (insects).

Distance between large and small, large and medium and medium and small vascular bundles varied significantly among the resistant and susceptible clones. This distance indicates the free space between vascular bundles, in susceptible group mean distance was more higher indicating presence of more free space occupied by spongy tissue compared to resistant clones which had lower row space with compactness internal leaf anatomy. Thus as discussed earlier greater number of cells with low free space may add toughness to internal leaf anatomy of resistant clones compared to susceptible clones increased free space adds succulence to internal leaf anatomy as noticed in case of susceptible clones, width of phloem in resistant clones was more compared to susceptible clones indicating availability of more amount of food to aphid for sucking.

Parenchyma cells are important cells of leaf where metabolic activities takes place. Significant difference was observed for the width of parenchyma cells around the bundle sheath between resistant and susceptible clones. For successful tapping of food, aphid has to pass its stylet invariably through intracellular or intercellular to reach the phloem. Klingauey

(1987) has reported that most of aphid species penetrate inter cellularly supported by the action of hydrolytic enzymes in their saliva particularly pectinase which dissolves middle lamella of cell wall, thus presence of thick layer of parenchyma cells may acts as barrier for aphid stylet penetration but during this process aphid may invariably damage more number of parenchyma cells which may lead to change in chemical nature of leaf by way of triggering host defense mechanism.

Phloem fibre is one of the important part of vascular bundle and its main function is to provide mechanical strength against collapse and avoid phloem damage by invaders. In the present study resistant clones had significantly higher thickness of phloem fibre compared to susceptible clones as phloem fibre is made up of sclerenchymatus tissue which is dead and lignified, presence of extra thickness of phloem fiber and its gritty nature in resistant clones may act as strong mechanical barrier for stylet penetration to suck the sap from phloem. Similar observations were also made by earlier workers Khanna *et al.* (1950) and Kumarsinghe *et al.* (2001), where they noticed thickened pholem fibre around phloem tissue and vascular bundles respectively.

In general presence of large amount of internal leaf tissue with wider distance between stomata of lower epidermis and phloem and more thickness of parenchyma cells around vascular bundles with greater thickness of phloem fiber around the phloem tissue may inhibit piercing and also phloem sap ingestion in resistant clones. Whereas, absence or presence of above features in lower amounts may motivate the insect for high rate of piercing and ingestion of phloem sap from phloem in susceptible clones.

5.1.1 Histochemistry

The sections obtained from the leaves of resistant and susceptible clones were subjected to histochemical studies. Three major parameters considered for histochemical studies, were total polysaccharides, protein and total RNA in the tissue through staining procedures.

The histochemical studies on total polysaccharides protein and total RNA of the resistant clones are presented in plate 6, 7 and 8. For total polysaccharides resistant clone SNK 192 and SNK 754 stained rich (++) compared to susceptible clones which stained very rich (+++) indicating more amount of total polysaccharides in susceptible clones which may attract woolly aphid for high rate of sucking which are in accordance with results of (Chandrashekar, 1994) where he observed higher amount of polysaccharide in susceptible clones compared to resistance clones .

There was presence of protein difference in leaf tissue between resistant and susceptible clones, where resistant clones SNK 192 stained low (+) and SNK 754 stained very rich (+++) compared to susceptible clone where, Co 92020 and CoC 671 stained rich (++), These results are contradicting to results obtained by Chandrashekar (1994), where he observed low staining intensity for protein in resistant clones compared to susceptible cotton genotypes.

As aphids are dependent mainly on amino acids and little on sugars for their growth and development and they secrete excess sugars as honey dew, so presence of more polysaccharides in susceptible clones and less in resistant clones many not be the reason for imparting resistance or susceptibility to sugarcane which are in accordance with views expressed by Walter and Mullin (1988), Fisher *et al.* (1994) and Rhodes *et al.* (1996).

Total RNA among resistant clones SNK 192 and SNK 754 stained rich (++) whereas in susceptible clone Co 92020 and CoC 671 stained very rich (+++).The role of total RNA in resistance need to be established.

5.2 BIOCHEMICAL BASIS OF RESISTANCE

In the present investigation Hx absorbance per cent values (measured as intensity of blue colour produced after addition of ferric chloride reagent to plant leaf extracts) varied significantly among the resistant and susceptible clones, resistant clones recorded significantly higher mean absorbance values than susceptible clones for Hx absorbance in leaf tissue extracts. These results are in accordance with the results of Victor *et al.* (1983),

Nicol *et al.* (1992), Givovich (1994a), Givovich (1994b) and Niemeyer *et al.* (2000), where in they also reported higher amount of Hx in leaf tissues of wheat cultivars resistant to aphids.

Patil *et al.* (2005) in a field study on behaviour of released apterous nymphs involving resistant and susceptible clones had reported that released apterous nymphs could not colonize on resistant clones leading to cent per cent mortality after 48-72 hours of release compared to susceptible clone where released apterous nymphs colonized immediately after 24 hours of release. Further in resistant clones there was no honeydew secretion by the aphids indicating the aphid ingested less or did not ingest any sap from phloem, death of aphids might be due to toxic effect of sap of resistant clones or feeding deterrence leading to starvation death.

Based on absorbance values related to Hx content of resistant clones and field observations made by Patil *et al.* (2005), it is appropriate to conclude that resistant mechanism operating may be antibiosis or antixenosis (feeding deterrence or toxic effect) and basis of resistance may be biochemical as Hx content higher in resistant clones compared to susceptible clones indicated from mean absorbance per cent value.

If the resistant mechanism operating is antibiosis then there should be decreased growth rate and lower fecundity rate of released apterous nymphs, but such observations where not made by Patil *et al.* (2005). Instead, they noticed cent per cent mortality of released apterous nymphs on resistant clones, without any further growth of released apterous nymphs. The sudden death of released apterous nymphs within 48-72 hours after release may be due to toxic sap of resistant clone leading to starvation in the form of feeding deterrence, which is supported by no honey dew exudates on resistant clones indicating possibility of no ingestion of phloem sap or may be very less ingestion because of toxic nature of sap. These views are supported by the findings of earlier workers Victor *et al.* (1983), Givovich (1994a) and Niemeyer (2000).

Where they observed higher amount of Hx in wheat cultivars which resistance to aphid and attributed feeding deterrence due to toxic effects of phloem sap, however, Niemeyer (2000) in his findings reported that feeding deterrence was present at mesophyll tissue level and not in phloem. The higher amount of Hx in resistant clones could be due to inherent capacity to produce high Hx or more number of parenchyma cells damage.

Interestingly histological observations in the presence study also supports views explained above in the form of more number of parenchyma cells around vascular bundle sheath when aphid attempts to gather food from phloem, the stylet has to penetrate through mesophyll cells to reach phloem, it is most likely that more number of cells may get injured during this process in resistant clones triggering the activity of □-glucosidase enzyme present in cell wall of mesophyll tissue(Nikus fennette 2003), the enzyme □-glucosidase cleaves glucoside to aglucons which is a form of Hx acid which acts as a toxic compound or feeding deterrent to aphid probing. These views were also expressed by Halfman and Hafmanova (1969), Nicol *et al.* (1992) and Givovich *et al.* (1994b).

It is also in accordance to the hypothesis put forth by Edward and Wratten (1987) where wound related induction of chemical changes in plant tissues have a defensive role. Any damage to leaf, however small it is, causes local change in leaf chemistry and insect responds to the induced change by moving away from the vicinity of damage.

5.3 ASSOCIATION STUDIES

5.3.1 Character association

Selection for a specific character is known to result in correlated response in certain other character (Falconer, 1964). Generally plant breeders make selection for one or two attributes at a time, then it becomes important to know the effect on other characters. Simple phenotypic correlations indicate broadly the type of association that exists between various attributes. But simple phenotypic correlations by themselves do not provide any reliable basis for selection. Hence the genotypic correlations which are based on the heritable part of the observed variation enable the assessment of the pattern of inherent relationship that exists between various traits.

The genotypic and phenotypic associations involving ten resistant clones were attempted to identify the trait (s) contributing to cane yield by utilizing the overall data of the genotypes. In the present investigation for majority of the character pairs, the genotypic correlation coefficients were higher than the corresponding phenotypic correlation coefficients and further the sign of the genotypic correlation coefficients matched that of the phenotypic correlation coefficients.

Similar results were also indicated by Kang *et al.* (1983) and Reddy and Reddi (1986a and 1986b) in sugarcane. These results suggest that reliance can be placed on the correlation coefficients both at genotypic as well as phenotypic level.

The lower phenotypic correlation coefficient values obtained in the present study could be due to modifying effect of environment on the character associations at the genotypic level. Many of the earlier sugarcane workers (Anand and Torrie, 1963 and Bhatt *et al.,* 1968) also reported the role of environment in modifying the nature of association in relation to their corresponding phenotypic values, which were taken for interpreting the results.

Among 13 characters studied germination per cent, tiller number, single cane weight, number of internodes and CCS per cent had positive and significant genotypic association with cane yield at genotypic level. Whereas, at phenotypic level, only CCS per plot had positive and highly significant positive association with cane yield per plot.

In the present investigation germination per cent recorded positive and significant association with cane yield, CCS yield, NMC per plot, tiller number per plot, single cane weight, number of internodes, and cane girth at genotypic level. Whereas, at phenotypic level only CCS per plot had positive significant association with cane yield. literature regarding association of germination per cent with cane yield components are very scanty, however, Nair and Sreenivasan (1990) reported poor association of germination per cent with cane yield. Whereas, Tippeswamy *et al.* (2003a) reported positive significant association of germination per cent with NMC per plot and cane yield.

Tiller number recorded positive significant association NMC per plot, germination per cent, millable cane height, cane yield and CCS yield at genotypic level, whereas at phenotypic level tiller number associated positively and significantly with millable cane height and NMC per plot. There are no reports available in the literature regarding association of tiller number with millable cane height, juice extract per cent, NMC per plot and CCS per plot. Where as Punia (1983) reported positive association of tiller per clump to cane yield, Nair and Sreenivasan reported poor association of early tillering with cane yield. However, present results are in agreement with the results of Sekhar *et al.* (1986) for cane girth and Stevensson (1965), Batcha (1965) and Jai *et al.* (1980) for CCS yield and Pillai and Ethirajan (1993) and Singh and Khan (1995) for cane yield.

Leaf area is a important physiological trait determining the total biomass production and inturn determines productivity. Leaf area in present investigation recorded non-significant association with cane yield which are contradicting to the results obtained by Gajera *et al.* (1991) and Tippeswamy *et al.* (2003a) where they reported significant positive association with cane yield. The present non-significant association with cane yield might be attributed due to recording of leaf area at ten month stage of crop.

Millable cane height and single cane weight are considered as main yield components as evident by literature by different workers. However, millable cane height recorded non-significant association with cane yield whereas, single cane weight recorded significant positive association with cane yield.where as, Gangopadhyaya and Sarkar (1965), Marriotti (1971), Kundu and Gupta (1997) recorded significant association of millable cane height, whereas Herbert (1965), Sai and Patel (1975), Bathla (1978), Yadav and Sharma (1978), Ramanareddy (1994) and Tippeswamy (2003a) recorded positive association of single cane weight with cane yield. The non-significant association of millable cane height with cane yield could be due to major contribution by single cane weight which nullified the effect of cane height.

Cane girth recorded significant association with single cane weight, germination per cent and number of internodes at genotypic level, however at phenotypic level it recorded

positive significant association with single cane weight. These results are not in accordance with the results obtained by Balasundaram and Baghyalakshmi (1978a), Kang *et al.* (1981), Reddy and Reddy (1997b) and Tippeswamy *et al.* (2003a) this non-significant association could be attributed mainly due to less number of genotypes included in the present investigation.

Number of internodes recorded strong positive and significant association with CCS yield, cane yield, single cane weight, sucrose per cent, brix per cent, germination per cent and cane girth at genotypic level. However, at phenotypic level it associated significantly only with CCS yield, which are in accordance with the observations of Verma *et al.* (1996) for sucrose per cent and CCS yield per plant, Legendre *et al.* (1970), Sekhar (1986), Singh and Khan (1995) and Das *et al.* (1996) and Tippeswamy (2003a) for cane yield, Tehlan (1986) for brix per cent.

NMC per plot recorded significant positive association with germination per cent and tiller number at genotypic and only tiller number at phenotypic level, whereas negative association but not significantly for SCW and cane girth at genotypic and phenotypic level however Tippeswamy *et al.* (2003a) reported significant association of NMC per plot to cane yield per plot this non significant association could be due to major contribution by SCW and number of internodes.

Juice extract per cent recorded positive association with tillers number and brix per cent. However, literature regarding association of juice extract per cent with tiller number and brix per cent are not available.

The ultimate objective in sugarcane breeding is to improve the sugar productivity per unit area.

Brix per cent and sucrose per cent are the major constituents responsible for the quality of cane juice. Hence the improvements in these traits would help to improve the sugar productivity.

Brix per cent recorded positive significant association with sucrose per cent and juice extract per cent at genotypic level, whereas at phenotypic level it recorded significant positive association only with sucrose per cent. These results are in agreement with the results of Herbert (1965), Lesnic and Vencorsky (1974), Richard (1975) and Nageswar Rao (1982) for sucrose per cent, so improvement for any of the character *i.e.*, brix or sucrose per cent will improve both characters as they are associated positively, which was observed in the present study.

CCS is one of the important parameters of sugarcane breeding where CCS is given prime importance during selection in the present study CCS recorded significant positive association. Cane yield and the results are in accordance with the results of Reddy and Reddi (1986b), Singh *et al.* (1994) Das *et al.* (1996) and Tippeswamy *et al.* (2003a) for association of CCS yield per plot with cane yield at harvest.

In general in the present investigation among 12 characters under study, revealed germination per cent, tiller number, single cane weight, number of internodes, cane girth, CCS yield had positive and significant association with cane yield at genotypic level. Whereas, at phenotypic level only CCS yield had positive and significant association with cane yield, so these characters can be considered as important yield (cane and sugar yield) determining factors and selection can be practiced based on these characters. However, special importance should be given for SCW, number of internodes and cane girth which are principal cane yield components and final selection should be made based on CCS yield, so improvement in any of the principal cane yield components traits in general and CCS yield in particular would certainly bring improvement in cane yield.

Among quality parameters sucrose per cent and brix per cent exhibited positive association among themselves, so improvement in any of the characters in general and sucrose in particular would certainly bring improvement in sugar yield.

5.3.2 Path coefficient analysis

In sugarcane, the cane and sugar yields are the complex characters and they are influenced by number of interrelated component traits. The interdependent of the component characters among themselves often influence the direct relationship with cane yield and as a result the information based on correlation coefficients becomes undependable. Since path coefficient analysis gives a more realistic relationship of characters and an attempt has been made to identify the effective components of cane and sugar yield.

5.3.2.1 Path coefficient analysis for cane yield per plot

Path coefficient analysis was used in working out the direct and indirect effects of 12 characters on cane yield per plot in the present study. The genotypic path coefficient analysis accounted for all the variation in cane yield as indicated by low residual effects.

The results of present investigation indicated some interesting facts. Those characters with high positive correlation have not always shown direct effects. Many a times they were associated with low direct effects and negative direct effects. In the present investigation characters which had high positive association to cane yield *viz.,* germination per cent and number of internodes had low positive direct effect. Whereas, tillers number and single cane weight had negative direct effects on cane yield. Similar trend was also observed by Tippeswamy *et al.* (2003b) for germination per cent single cane weight, cane formed shoots and stem diameter.

From the breeders point of view, those characters would be useful as selection criteria which are not only having high positive association but also exert high direct effects. In this context CCS per plot had emerged as a single character which had high positive significant association with cane yield per plot but also exerted, high direct effect on cane yield per plot. Similar trend was also observed by Tippeswamy *et al.* (2003b). Where, they reported CCS per plot had significant positive association and high direct effect on cane yield.

Based on these results it is evident that CCS per plot appears to be a best criteria for the improvement of cane yield, the same view was also expressed by Patel *et al.* (1993). However, among the traits which had significant positive association on cane yield *viz.,* germination per cent and number of internodes had low direct effects and tiller number and single cane weight had negative direct effects influencing the cane yield indirectly and positively through CCS per plot. Negative direct effects of positively correlated characters were also reported by Sharma and Singh (1984), Patel *et al.* (1993) and Tippeswamy *et al.* (2003).

Among the yield components millable cane height, cane girth and NMC per plot had positive but not significant association with cane yield but recorded high positive direct effects on cane yield, hence these traits can also be considered as selection criteria for improving cane yield.

Among juice quality parameters brix per cent had direct positive effect on cane yield where as sucrose per cent recorded high (maximum) negative direct effects on yield at the time of harvest. Khairwal and Babu (1975), Khairwal *et al.* (1977), Reddy and Reddy (1977) and Tippeswamy *et al.* (2003) also reported negative direct effect of sucrose per cent on cane yield.

Hence, from genotypic and phenotypic path analysis it can be said that CCS per plot had maximum direct effect on cane yield and it also contributed indirectly via germination per cent, tiller number, single cane weight, millable cane height, cane girth, number of internodes, NMC and brix per cent. Hence, for improvement of cane yield in sugarcane emphasis must be placed on CCS yield per plot at harvest, however yield components *viz.,* millable cane height, cane girth and NMC per plot may also be considered as selection criteria for improvement in cane yield.

5.3.3 Mean performance of genotypes

The analysis of variance revealed the presence of highly significant differences among woolly aphid resistant and susceptible clones for most of the character under present study.

In the present investigation some of the woolly aphid resistant clones differed significantly for tiller number, millable cane height, leaf area, brix per cent, sucrose per cent, CCS per plot, NMC per plot and cane yield (kg/plot) compared to commercial checks CoC 671, Co86032 and CoM 88121.

For germination per cent, none of the resistant clones had significantly higher mean values than commercial checks, however clones SNK 256 and SNK 44 recorded numerically higher mean value than one of the commercial check CoM 88121. Although the clones did not differ significantly for germination per cent, but at the age of 90 DAP only one resistant clone SNK 256 recorded significantly higher mean than best check Co86032, however clones SNK 61, SNK 124 had the potentiality of producing numerically higher mean values than superior check Co86032, clones for leaf area clone SNK 754 and SNK 57 recorded significantly higher mean values than the best check CoM88121.

Among the ten resistant clones under study for millable cane height all the resistant and susceptible (checks) were on par with each other, however resistant clones SNK 256, SNK 124 and SNK 61 and SNK 49 had the capability of providing numerically higher mean values than commercial checks, resistant clones differed significantly for cane girth, number of internodes and single cane weight compared to commercial checks but none of the resistant clones recorded significantly higher mean values than checks. Resistant clones differed significantly for NMC compared to commercial checks and clone SNK 256 recorded significantly higher mean value than superior check Co 86032.

Sugar yield and quality are two important parameters of concern to breeder and sugar industry, so it is important to known the performance of newly identified SWA resistant clones for sugar yield and quality characters. Juice extract per cent did not differ significantly among resistant and susceptible clones, however recorded significantly higher mean values than commercial checks.

Sucrose pre cent in juice is most important character contributing to juice quality thus resistant clones differed significantly for sucrose per cent compared to checks. However, none of the resistant clone recorded significantly higher mean than best check CoC 671. For brix per cent resistant clones differed significantly compared to checks and clones *viz.,* SNK 754 and SNK 44 recorded significantly higher mean values then best check Co 86032.

Resistant clones differed significantly for CCS per plot and cane yield per plot, however clones SNK 49, SNK 754, SNK 44 and SNK 61 recorded significantly higher mean values for CCS per plot and for cane yield clones *viz.,* SNK 49, SNK 256, SNK 061, SNK 44 and SNK 754 recorded significantly higher mean than best check Co 86032. The higher contribution to cane yield was mainly due to higher contribution made by NMC per plot.

In general, based on mean performance resistant clones SNK 49, SNK 256, SNK 44 and SNK 754 were identified as best clones for cane yield, where as for sucrose content three clones *viz.,* SNK 754, SNK 44 and SNK 61 were identified as best clones. However, clones SNK 754, SNK 44 and SNK 61 proved their superiority over commercial checks for both cane and sugar yield (superiority in terms of higher mean value) which are ultimate end products of sugarcane.

In this situation were there is threat for sugarcane cultivation itself due to woolly aphid menace, it would be advisable to grow these newly identified SWA clones and save the farmers and sugar industry from severe economic loss.

Table 9: Top five superior performing SWA resistant clones selected for cane yield and its component traits

Sl. No.	Germination per cent	Tillers per plot	Leaf area (cm^2)	Cane girth (cm)	Number of internodes	Single cane weight (kg)	Number of millable canes per plot	Brix per cent	Sucrose per cent	Cane yield (kg/plot)	CCS yield (kg/ plot)
1	SNK 044 (89.17)	SNK 286 (334.33)**	SNK 754 (685.15)**	SNK 057 (2.79)	SNK 061 (21.66)	SNK 049 (1.71)	SNK 256 (437.33)**	SNK 754 (21.27)**	SNK 754 (17.70)	SNK 049 (345.33)**	SNK 49 (38.73)**
2	SNK 256 (88.33)	SNK 061 (241.33)	SNK 57 (496.96)*	SNK 044 (2.74)	SNK 049 (20.66)	SNK 061 (1.56)	SNK 754 (241.00)**	SNK 44 (20.83)*	SNK 44 (17.67)	SNK 256 (296.00)*	SNK 61 (33.59)**
3	SNK 754 (85.83)	SNK 124 (228.67)	SNK 196 (486.28)	SNK 158 (2.53)	SNK 057 (18.66)	SNK 057 (1.55)	SNK 044 (178.00)	SNK 671 (19.43)	SNK 61 (17.63)	SNK 061 (273.00)**	SNK 44 (31.13)*
4	SNK 057 (85.42)	SNK 049 (194.67)	SNK 49 (466.00)	SNK 061 (2.47)	SNK 754 (18.52)	SNK 044 (1.48)	SNK 061 (176.04)	SNK 457 (18.44)	SNK 57 (16.67)	SNK 044 (266.66)*	SNK 754 (29.88)*
5	SNK 758 (84.58)	SNK 754 (191.33)	SNK 158 (447.05)	SNK 754 (2.45)	SNK 044 (18.50)	SNK 158 (1.30)	SNK 192 (174.00)	SNK 124 (18.83)	SNK 49 (16.60)	SNK 754 (252.00)	SNK 256 (29.16)
Checks											
1	Co 86032 (92.00)	Co 86032 (204.67)	CoC 671 (3.04)	CoM88121 (43.00)	CoC 671 (24.61)	CoC 671 (1.88)	CoM88121 (163.35)	CoM88121 (18.6)	CoC 671 (16.21)	Co 86032 (207.66)	Co 86032 (23.53)
2	CoC 671 (90.92)	CoM88121 (187.33)	Co 86032 (2.79)	Co 86032 (415.25)	CoM88121 (21.66)	Co 86032 (1.63)	Co 86032 (162.00)	Co 86032 (18.39)	Co 86032 (16.01)	CoM88121(206.00)	CoM88121 (22.29)
3	CoM88121 (86.67)	CoC 671 (156.00)	CoM88121 (2.73)	CoC 671 (380.00)	Co 86032 (20.33)	CoM88121 (1.35)	CoC 671 (101.56)	CoC671 (17.89)	COM88121 (15.94)	CoC 671 (191.00)	CoC 671 (21.78)
CD at 5%	21.47	63.68	57.79	0.20	2.75	0.395	49.01	1.80	1.69	37.95	6.05
CD at 1%	NS	86.29	79.00	0.27	3.73	0.130	66.41	2.45	NS	51.43	8.21

* = Significant at 5% level

** = Significant at 1% level

FUTURE LINE OF WORK

1. The greater role of histological studies involving specific fluorescent dyes to locate glucosidase and glucoside in leaf tissue needs to be undertaken.
2. There is need to locate the feeding deterrence against aphid in leaf tissue using electronic feeding monitory system
3. Actual quantification of hydroxamic acids and their threshold levels in resistant and susceptible clones need to be under taken using HPLC
4. Identified superior woolly aphid resistance clones *viz.,* SNK 754, SNK 44 and SNK 61 needs to be tested over location in large scale trials to confirm their superiority

VI. SUMMARY

The present investigation was carried out to understand the mechanism and possible basis of resistance against woolly aphid involving recently identified resistant clones through histological and biochemical studies. Resistant clones along with commercial checks were evaluated to get information on mean performance nature of association and path analysis for cane yield and its component traits.

The experimental material was planted in a randomized block design with three replication at Agricultural Research Station, Sankeshwar. Leaf samples for histological and histochemical study were collected at 10 months and for biochemical work at six months crop. Field observations were recorded for cane and sugar yield traits at various stages *viz.,* germination per cent tiller number, leaf area, single cane weight, millable cane height, cane girth, number of internodes, juice extract per cent, brix per cent, sucrose per cent, number of millable canes, CCS yield per plot and cane yield per plot.

The salient findings of present investigations are summarized below

1. Comparative leaf anatomical observations were made through histological studies for SWA resistant and susceptible clones for internal arrangement of various tissues that make up the leaf. Significant anatomical differences were observed between resistant and susceptible clones *viz.,*

    i) Distance between large and small, large and medium and medium and small vascular bundles was more in susceptible clones

    ii) Thickness of parenchyma cells around bundle sheath and width of the phloem was more in resistant clones

    iii) Greater thickness of phloem fibre around phloem tissue in resistant clones,

2. Histochemical studies involving resistance and susceptible clones

    i) Total polysaccharides in resistant clone SNK 192 stained low (+) whereas SNK 754 stained rich (++) and in susceptible clones Co 92020 and CoC 671 stained very rich (+++)

    ii) Protein content in resistant clone SNK 192 stained low (+) and SNK 754 stained very rich (+++) and for susceptible clones Co 92020 and CoC 671 stained rich (++).

    iii) Total RNA content in resistant clone SNK 192 and SNK 754 stained rich (++) however, Co 92020 and CoC 671 stained very rich (+++).

3. Mean absorbance values for leaf hydroxamic acid concentration differed significantly between resistant and susceptible clones.

4. Association study revealed significant association of germination per cent, tiller number, single cane weight, number of internodes, cane girth and CCS yield with at cane yield at genotypic level, whereas at phenotypic level only CCS yield per plot had positive and significant association with cane yield per plot. Among quality parameters sucrose per cent and brix per cent exhibited strong association among themselves.

5. Path coefficient analysis of cane yield revealed that CCS yield at harvest had large contribution to cane yield, followed by NMC per plot, cane girth, millable cane height, brix per cent and can be used as selection criteria for improvement in cane yield as indicated by high direct effects.

6. Based on the mean performance, the SWA resistant clones *viz.,* SNK 754, SNK 044 and SNK 061 were identified as superior over commercial checks for both cane and sugar yield sucrose per cent. However, these clones need to be further tested in large scale trials to confirm their superiority in yield and quality attributes.

VII. REFERENCES

AKARAKAI, N., 1989, Flight periodicity and the effect of weather conditions on the take off of *Ceratovacuna lanigera* Zehntner (Homoptera : Aphididae). *Applied Entomology and Zoology*, 24 : 264-272.

ANAND, 1946, Aphid reaction to glabrous and hairy type of cotton. *Annals of Applied Biology*, 33 : 23-25.

ANAND, S. C. AND TOTTIE, J. H., 1963, Heritability of yield and other traits and interrelationships among traits in F_3 and F_4 generations of three soybean crosses. *Crop Science*, 3 : 508-511.

ANANTHKRISHNAN, J. N., VENKATESHAN, S. AND SRIDHAR, S., 1994, Energetics and resource allocation in the Lepidopteran complex infesting the cotton plant interms of cultivar density and plant chemical quality in : T. N. Ananthkrishnan (eds). *Functional dynamics of Phytophagous Insects*, Oxford and IBH publishing Co. Pvt. Ltd., New Delhi, India, pp. 95-114.

ANONYMOUS, 1963, Annual work progress report on crop improvement program of rice, sugarcane, vegetable and field crops. Directorate of Rural Affairs, Vietnam, p. 266.

ANONYMOUS, 2002a, Report on woolly aphid *Ceratovacuna lanigera* Zehntner (Homoptera : Pemipigidae) on sugarcane in Northern Karnataka, University of Agricultural Sciences, Dharwad.

ANONYMOUS, 2002b, Status of sugarcane woolly aphid in northern Karnataka and its management, University of Agricultural Sciences, Dharwad, pp. 1-14.

ANONYMUS, 2004 Statistical outline of India, *Indian sugar,* 12 :50-64

AOKI, S., KUROSU, U. AND USUBA, S., 1984, First instar larvae of the sugarcane woolly aphid, *Ceratovacuna lanigera* (Homoptera : Pemphigidae), attack its predators. *Kontyu*, 52 : 458-460.

ARGANDONA, V. H., CORCUERA, L. J., NIEMEYER, H. M. AND CAMPBELL, B. C., 1983, Toxicity and feeding deterrency of hydroxamic acids from Gramineae in synthetic diets against the green bug. *Schizaphis graminum*. *Entomoligia Experimentalis et Applicata*, 34 : 134-138.

ARGANDONA, V. H., NIEMEYER, H. M. AND CORCUERA, L. J., 1981, Effect of content and distribution of hydroxamic acids in wheat on infestation by the aphid *Schizaphis graminum*. *Phytochemistry*, 20 : 673-676.

ARTHUR, W., SCHAAFRMA, CESAT CORDONA, JULIAX, JORNEGAY, ASISON, M. WYLDE AND THOMAS, E. M., 1987, Resistance of common bean lines to the potato leaf hoppers. *Journal of Economic Entomology*, 91 (4) : 981-986.

AVASTHY, P. N., 1977, Integrated control of sugarcane pests and diseases. *Sugar News*, 9 (6 & 7) : 72-74.

BACON, T. S. AND DICKINSON, B., 1957, The origin of melezitose a biochemical relationship between the lime tree (*Tillia* spp) and an aphid (*Eucallipterus tiliae* L.). *Biochemical Journal*, 66 : 289-299.

BAJIT SINGH, SINGH, S. AND RISIHPAL, 1994a, Direct and indirect effects of characters affecting cane yield of five sugarcane crosses. *Crop Research*, 8 (2) : 302-304.

BAKSHIRAM AND HEMAPRABHA, G., 1995, Interspecific hydbirdization of *Saccharum* species and its consequences on yield and quality. In *Genetic Research and Education*, Current Trends and the Next Fifty years (Eds. Sharma *et al.*). *Indian Society of Genetics and Plant Breeding*, New Delhi, 1082-1086.

BALASUNDARAM, N. AND BHAGYALAKSHMI, K. V., 1978, Path analysis in sugarcane. *Indian Journal of Agricultural Research*, 12 (4) : 215-218.

BARRIA, B. N., COPAJA, S. V. AND NIEMEYER, H. M., 1992, Occurrence of DIMBOA in wild *Hordeum* species and its relation to aphid resistance. *Phytochemistry*, 31 : 89-91.

BASU, A. N. AND BANERJEE, S. N., 1958, Aphids of economic plants of West Bengal. *Indian Agriculture, 2* : 89 – 112.

BATCHA, M. B. G. R., 1975, A study on the association of yield and yield contributing characters in sugarcane under North Bihar conditions. *Ph. D. Thesis*, Bihar University, Ranchi.

BATCHA, M. B. G. R. AND SAHI, B. K., 1972, Studies on the association of certain agronomic traits in sugarcane under subtropical condition. *Paper Presented at the Diamond Jubilee Symposium of Sugarcane Breeding Institute*, Coimbatore.

BATHLA, H. V. L., 1978, Study of correlation between cane yield and different attributing characters. *Indian Sugar Crops Journal*, 3 (3) : 45-46.

BATHLA, H. V. L., 1981, Effects of different yield attributing characters on cane yield. *Indian Sugar Crops Journal*, 8 (1) : 19-20.

BEHURA, B. K., 1963, Prakruti. *Journal of Urkal University of Sciences,* 3 : 40- 65.

BEEK, A. L., NIEMEYER, H. M. AND VICTOR, 1982, Role of various biochemicals in resistance to wheat aphid. *Phytochemistry*, 16 : 50-56.

BHATT, G. M., SANDHU, P. S. AND SINGH, A., 1968, A study of genetic variability and correlation in soybean. *Journal of Research,* Punjab Agricultural University 5 : 1-5.

BHIDE, V. S., 1969, Biometrical studies in the aspects of quality and yield and its components in sugarcane (*Saccharum officinarum* L.). *Ph. D. Thesis*, Agra University, Agra,

BROWN, A. H. D., 1965, Correlation between brix in juice and fibre in commercial hybrid sugarcane populations. *Proceedings of International Society of Sugarcane Technologists 12th Congress*, 38 : 361-369.

BROWN, A. H. D., DANIELS, J. AND LATTER, B. D. H., 1969, Quantitative genetics of sugarcane II. Correlation analysis of continuous variation of characters in relation to hybrid sugarcane breeding. *Theoretical and Applied Genetics*, 39 : 1-10.

CAMPBELL, B. C., McLEAN, D. L., KINSEY, M. G. JONES, K. E. AND DREYER, D. L., 1982, Probing behaviour of the green bug (*Schizaphis graminum* biotype C) on resistant and susceptible varieties of sorghum. *Entomologia Experimentalis et Applicata,* 31 : 140-146.

CHAKRABARTI, S., GHOSH, A. K. AND RAYCHAUDHURI, D. N., 1971, New records of aphids (Insecta : Homoptera) from Uttar Pradesh, India. *Science and Culture,* 37 : 247 – 248.

CHANDRASHEKAR, 1994, Histological and biochemical basis of resistance to insect pests in cotton. *M. Sc. (Agri.) Thesis*, University of Agricultural Sciences, Dharwad.

CHAUHAN, R. V. S., SINGH, S. B. AND SINGH, H. N., 1987, Correlation and path analysis in sugarcane. *Indian Sugar*, 37 (1) : 39-41.

COCHRAN, W. G. AND COX, G. M., 1957, *Experimental* Designs, Wiley, New York, pp. 611.

COPLAND, E. B., 1917, Disease and pests of sugarcane in the Philippines. *Philippines Agriculture and Forestry*, 5 : 343-346.

DAS, P. K., JENA, B. C. AND NAYAK, N., 1997a, Character association and path analysis of sugar yield in sugarcane. *Indian Sugar*, 46 (10) : 805-808.

DAS, P. K., JENA, B. C., NAYAK, N. AND PARIDA, A. K., 1996, Correlation and path analysis of cane yield in sugarcane. *Co-operative Sugar*, 27 (7) : 509-512.

DAS, P. K., PARIDA, A. K., NAYAK, N., MAHAPATRA, S. S. AND JENA, B. C., 1997, Path coefficient, regression and discriminate function in sugarcane. *Indian Sugar*, 47 (1) : 31-34.

DASADO, V. G., TAPAY, R.E. AND MIAYO, H. M., 1980, Selection indices for high yield in sugarcane. *Proceedings of International Society of Sugarcane Technologists*, *17th Congress* : 1104-1110.

DAVID, H. AND EASWARAMOORTHY, S., 1986, Biological control. In *Sugarcane Entomology in India* (Eds David *et al.,*) Sugarcane Breeding Institute, (ICAR), Coimbatore, pp. 383-422.

DAVID, H. AND NANDAGOPAL, V., 1986, Pests of sugarcane distribution symptomatology of attack and identification. In sugarcane Entomology in India, (Ed. David *et al.*) S.B.I. (ICAR) Coimbatore publication, pp. 1-29.

DE GROOT, 1941, Beschri jving der soortn van het svikerriet varietciten verhand, praefst, Java Suikerind, pp. 1697-1716.

DEWEY, D. R. AND LU, K. H., 1959, A correlation and path coefficient analysis of components of crested wheat grass seed production. *Agronomy Journal*, 51 : 515-518.

DHALIWAL, G. S., DILWARI, V. K. AND SAINI, R. S., 1993, Host plant resistance to insects : Basic concepts. In G. S. Daliwal and Ramesh Arore (eds) Trends in Agricultural Insect Pest Man Commonwealth Publishers, New Delhi, India pp. 469-511.

DIXON, 1998, Phloem sap analysis and their effect on aphid growth and development. *Journal of Experimental Biology*, 199 : 61-68.

DIXON, A. F. G., 1998, Aphid ecology, Publication, Chapman and Hall, London.

DOSADO, V. G., TAPAY, R. E. AND MIAYO, H. M., 1978, Characters association with yield of sixteen sugarcane varieties. *26th Philippines Sugar Technology*, Annual Convention, Manila.

EDWARD, P. J. AND WRATTEN, S. D., 1987, Ecological significance of wounded induce change in plant chemistry : In : Insect plants. Labeyric V. Fabers S. G., Lachaise D. (Eds) Dr. W. Junk publishers. Dardrecht Nether Land, pp. 213-218.

ELDEN, T. C. AND ELGIN, J. H., 1992, Mechanisms of resistance to the potato leaf hopper in selected alfalfa clones. *Journal of Economic Entomology*, 85 (2) : 576-582.

ETHIRAJAN, A. A., 1965, evaluation of plant attributes concerned with yield in sugarcane and assessing their behaviour in succeeding clonal population. *Ph. D. Thesis*, Madras University, Madras.

FALCONER, D. S., 1964, *Introduction to Quantitative Genetics*, Oliver and Boyd, London, p. 365.

FARINA, S. R., 1994, Problems on WSCA and strategies in its control at Tarapur sp. Report of the Mmomagen of Tarapur sp. Trakar Sugar Plantation Unpublished Report, pp. 9.

FISHER, D. V., WRIGHT, J. P. AND MITTLER, T., 1984, Osmo regulation by the aphid *Myzus persicae* : A physiological role for honeydew oligosaccharide. *Journal of Insect Physiology*, 30 : 387.

GAJERA, G. M., PATEL, H. S., PATEL, M. P., NAIR, P. L. AND MEHTA, N. J., 1991, Correlation studies in sugarcane. *Indian Sugar*, 15 (12) : 875-876.

GANGOPADHYAYA, M. AND SARKAR, R. P., 1963, A correlation study on crop weather relationship in sugarcane. *Indian Journal of Sugar Research and Development*, 8 : 1-12.

GHOSH, AK. K., 1974, Aphids (Homoptera : Insecta) of economic importance in India. *Indian Agriculture*, 18 : 135.

GIVOVICH, A. J., MORESE, S., CERDA, H., NIEMEYER, H. M., WRITTEN, S. D. AND EDWARD, P. J., 1992, Hydroxamic acid glucosides in the honeydew of aphids feeding on wheat. *Journal of Chemical Ecology*, 18 : 841-846.

GIVOVICH, A., SANDSTROM, J., NIMEYER, H. M. AND PETTERSSON, J., 1994, Presence of a hydroxamic acid glucoside in wheat phloem sap and its consequences for performance of *Rhopalosiphum padi* L. (Homoptera : Aphididae). *Journal of Chemical Ecology*, 20 (8) : 1923-1930.

GUPTA, M. M. AND GOSWAMY, P. K., 1995, Incidence of sugarcane woolly aphid and its effect on yield attributes and juice quality. *Indian Sugar*, 44 : 883-885.

HALFMAN AND HOFMANOVA, O., 1969, 1, 4-benzoxazine derivatives in plants, Sephadeoxa fractionation and identification of new glucoside. *European Journal of Biochemistry*, 8 : 109-112.

HARRIS, 1930, Studies on pubescent and glabrous types of soybean in relation to insect resistance. *Journal of Economic Entomology*, 29 : 31-33.

HERBERT, L. P., 1965, Association between yield components and yield of sugarcane varieties in Louisiana. *Proceedings of International Society of Sugarcane Technologist 12th Congress* : 760-763.

HERBERT, L. P. AND HENDERSON, M. T., 1959, Breeding behaviour of certain agronomic characters in progenies of sugarcane crosses. USDA Technology Bulletin, 1144-1154.

HILL, D. S., 1993, Major tropical pests (descriptions, biology and control). *In Agricultural Insect Pests of Tropics and their Control* (2nd, edn.), Coimbatore University Press, Coimbatore, p. 209.

HOGARTH, D. M., WUU, K. K. AND HEINZ, D. J., 1981, Estimating genetic variance in sugarcane using a factorial crosses design. *Crop Science*, 21 : 21-25.

HOODA, M. S., SINGH, S. AND CHAUDHARY, B. S., 1989, Interrelationships between cane yield and quality attributes in sugarcane.*Bharatiya Sugar*, 14 (3) : 67-68.

HOODA, R. S., BABU, C. N. AND KHAIRWAL, I. S., 1979, Association and oath analysis of nine characters in the progenies crosses at seedling stage. *Indian Journal of Agricultural Sciences*, 49 : 931-933.

ISHDA, M., 1928, The sugarcane woolly aphid and its control in Fomosa. *Journal of Formosan Sugar Planters Association*, 6 : 1-28.

ISHWARAN, V., 1981, *A Laboratory Hand Book for Agricultural Analysis Today and Tomorrow*, Published, pp. 300-301.

JAGATHESAN, D., VENKATARAMAN, M. R. AND SHAH, S. S., 1967, Introgression in *Saccharum. Current Science*, 36 : 217-218.

JAMES, N. I., 1971, Yield components in random and selected sugarcane populations. *Crop Science*, 11 : 906-908.

JAMES, N. I. AND FALGOUT, R. N., 1969, Association of five characters in progenies of four sugarcane crosses. *Crop Science*, 9 : 88-91.

JAMES, N. I. AND MILLER, J. D., 1971, Selection in row seedling crops of four sugarcane progenies. *Crop Science*, 11 : 245-248.

JAMES, N. I. AND MILLER, J. D., 1975, Selection in crops of sugarcane. II : Efficiency and optimum selection intensities. *Crop Science*, 15 : 37-40.

JOSHI, S. AND VIRKTAMATH, C. A., 2004, The sugarcane woolly aphid *Ceratovacuna lanigera* Zehntner)Hemiptera : Aphididae) its biology, pest status and control. *Current Science*, 87 : 307-316.

KANG, M. S., MILLER, J. D. AND TAI, P. Y. P., 1983, Genetic and phenotypic path analyses and heritability in sugarcane. *Crop Science*, 23 : 643-647.

KHAIRWAL, I. S. AND BABU, C. N., 1975, path coefficient analysis of cane yield in sugarcane. *International Society of Sugarcane Technologists and Sugarcane Breeders Newsletter*, 36 : 58-61.

KHAIRWAL, I. S., BABU, C. N. AND HOODA, R. S., 1978, The biometrical approach to an ideotype in sugarcane. *International Society of Sugarcane Technologists and Sugarcane Breeders Newsletter*, 41 : 51-54.

KHAIRWAL, I. S., BABU, C. N., ARORA, N. D. AND HOODA, R. S., 1977, Path analysis of sucrose content in sugarcane II. *International Society of Sugarcane Technologists and Sugarcane Breeders Newsletter*, 39 : 34-36.

KHAN AND AGARWAL, 1990, Resistance of cotton genotypes to sucking insects. *Journal of Economic Entomology*, 25 : 16-20.

KHANNA, K. L., SHARMA, S. L. AND HUSSAIN, M. Z., 1950, On pyrilla infestation and varietal resistance in sugarcane. *Current Science*, 19 : 251-252.

KLINGAEY, F. A., 1987, Host plant finding and acceptance in aphids, their biology natural enemies and control (Ed.) Minks A. K and Harrecuijin publisher. Elsevier Science Publishing Company, New York, pp. 197-208.

KOGAN, M. AND ORTMAN, E. F., 1978, Analysis – A new term proposed to clyne painters non prycence mortality of resistance. *Bulletin Ent. Sco. Ann*, 24 : 125-126.

KOGAN, M., 1982, Plant resistance in pest management. In : R. L. Mutcoft and W. H. Luckmann (eds). Introduction to insect pest management John Miley and Sons, New York, USA, pp. 93-134.

KUNDU, S. AND GUPTA, M. L., 1997, Variability and character association studies in *Saccharum officinarum* clones. *Indian sugar,* 46 (11) : 889-893.

LEGENDRE, B. L., 1970, Association involving yield of sugar cane acre and its components in sugarcane. *M. Sc. (Agri.) Thesis*, Louisiana State University, Louisiana.

LINGAPPA, S., KULKARNI, K. A., PATIL, R. K., TIPPANNAVAR, P. S. AND SHEKHARAPPA, 2003, Status of woolly aphid, *Ceratovacuna lanigera* Zehntner on sugarcane in Karnataka. *Brain Storming Session on Sugarcane Woolly Aphid*, MPKV, Rahiru, Maharashtra June 10th, 2003.

LINGAPPA, S., PATIL, R. K., TIPPANNAVAR, P. S., BALIKAI. R, A. AND PRASADKUMAR, 2004, Development of strategies for the management of woolly aphid, *Ceratovacuna lanigera* Zehntner (Homoptera : Aphididae) on sugarcane. *Project Report* submitted to Department of Agriculture, Government of Karnataka, April, 2004.

LUNA, A. L., 1965, Study on some characters of the progeny in four crosses of sugarcane. *Proceedings of International Society of Sugarcane Technology*, *12th Congress* : 764-772.

MADHAVI, D., REDDY, C. R., REDDY, P. M., REDDY, G. L. K., REDDY, K. R. AND REDDY, K. H. P., 1991, Correlation studies in sugarcane. *Co-operative Sugar*, 22 (6) : 379-381.

MARIOTTI, J. A., 1968, Estudia Estadistica elans problacionas derivados decincocruzamientos en cane de azueat. *International Sugar Journal*, 70 (3) : 95-151.

MARIOTTI, J. A., 1971, Phenotypic associations among characters at the single stool selection stage in sugarcane. *International Sugar Journal*, 73 (8) : 327-340.

MARIOTTI, J. A., 1972, On the Effectiveness of some genetic parameters used in the selection of sugarcane populations. *International Society of Sugarcane Technologists and Sugarcane Breeders Newsletter*, 29 : 8-15.

MARIOTTI, J. A., 1973a, Path analysis of yield components in sugarcane. *International Society of Sugarcane Technologists and Sugarcane Breeders Newsletter*, 32 : 19-22.

MARIOTTI, J. A., 1973b, The influence of environment on the relationships between yield and its components in sugarcane. *International Society of Sugarcane Technologists and Sugarcane Breeders Newsletter*, 32 : 19-22.

MARIOTTI, J. A., 1978, Investigations on clonal selection of sugarcane in the Argentina Republic. *International Sugar Journal*, 80 (3) : 27-43.

MELAKU GIRMA, GERALD, E., WILDE AND JHNO, REESE, 1992, Russian wheat aphid feeding behaviour on host and non-host plant. *Journal of Economic Entomology*, 85 (2) : 395-401.

MELARU GIRMA, GERALD, E. M. AND JOHN C. R., 1992, Russian wheat aphid feeding behavioural on host and non-host plants. *Journal of Economic Entomology* 85 (2) : 395-401.

MICHEL, E., 1942, Beitrage zur kenntnis von lachmus (petro chlorus) roboris L., einer wichtigen, Honigat averguerin an der eiche, *zeitchrift angewndte entomologie*, 29 : 284-285.

MILLER, J. D. AND JAMES, N. I., 1974, The influence of stalk density on cane yield. *Proceedings of international Society of Sugarcane Technologists 15th Congress*, 177-183.

MILLER, J. D. AND JAMES, N. I., 1975, Selection in six crops of sugarcane 1. Repeatability of three characters. *Crop Science*, 15 : 23-25.

MIRZAWAN, P. D. N. AND IRWAN, L. S., 1995, Problems of pests and diseases for *germplasm conservation and exchange* (Eds. Croft, *et al.,*). *Report of and International Workshop Brisbane, Queensland, Australia*, pp. 34-41.

MITTLER, T. E., 1958a, Studies on feeding and nutrition of *Tuberolachmus salignus* (Gmelin) (Homoptera : Aphididae). Ii the uptake of phloem sap. *Journal of Experimental biology*, 34 : 334-341.

MOTE, U. N. AND PURI, S. N., 2003, Present status of sugarcane woolly aphid, *Ceratovacuna lanigera* Zehntner, a new pest problem on sugarcane in Maharashtra. *Paper presented at brain storming session on sugarcane woolly aphid*, Maharashtra, India, 10th June, 2003.

NAGARAJAN, R., 1983, Studies on genotype environment interaction in selected sugar cultivars of yield and quality attributes. *Ph. D. Thesis*, Tamil Nadu Agriculture University, Coimbatore.

NAGATOMI, S., MAEDA, M. AND LO, C. C., 1982, Studies on selection methods for sugarcane breeding XVI analysis of yield components. *Japanese Journal of Tropical Agriculture*, 26 : 229-238.

NAGESWARA RAO, P., 1982, Pattern of genetic variation in the progenies of crosses of high and low sugar cultivars of sugarcane (*Saccharum*). *Ph. D. Thesis*, Andhra Pradesh Agriculture University, Hydrabad.

NAGESWARA RAO, P., RAHMAN, M. A. AND PANDURANGA RAO, C., 1983, Genetic variability and character association in sugarcane progenies. *Indian Journal of Agricultural Sciences*, 53 : 621-623.

NAIR, N. V. AND SOMARAJAN, K. G., 1984a, Genetic correlations and selection responses in a hybrid sugarcane population selected for adaptation to water logging. *Indian Journal of Agricultural Sciences*, 54 : 180-182.

NAIR, N. V. AND SOMARAJAN, K. G., 1984b, Genetic variability and character association in sugarcane. *Sugarcane*, 5 : 8-10.

NAIR, N. V. AND SREENIVASAN, T. V., 1990, Studies on some early attributes of sugarcane in relation to yield and yield components. *Indian Journal of Genetics and Plant Breeding*, 50 (4) : 354-358.

NERKAR, Y. S., 2003, Sugarcane varietal planning in Maharashtra and its effects on improving sugar recovery and production in the state. *Proceedings of All India Seminar on Sugarcane Variety, Cane and its Effects on Sugar Industry, STAI, Lonavla, Khandala*, pp. 7-16.

NICOL, D., COPAJA, S. V., WRATTEN, S. D. AND NIEMEYER, H. M., 1992, A screen of worldwide wheat cultivars for hydroxamic acid levels and aphid antixenosis. *Annals of Applied Biology*, 121 : 11-18.

NIEMEYER, H. M., 1988, Hydroxamic acids (4-hydroxy-1, 4-benzoxazin-3-ones), defense chemicals in the Graminaeae. *Phytochemistry*, 27 : 3349-3358.

NIEMEYER, H. M., 1991, Hydroxamic acid content of *Triticum* species. *Euphytica,* 37 : 289-293.

NIEMEYER, H. M. AND ARTURO GIVOVICH., 2000, Use of electrical penetration graphs and phloem sap chemical analysis in studies of the effects of hydroxamic acids in cereals on aphid (Homoptera : Aphididae) feeding behaviour. *Proceedings of the Entomological Society of America, Lanham*, MDUSA, pp. 237-244.

NIEMEYER, H. M., PESEL, E., COPAJA, S. C., BRAVO, H. R., FRANKE, S. AND FRANKE, W., 1988, Change in hydroxamic acid levels of wheat plants induced by aphid feeding. *Phytochemistry*, 28 : 447-450.

NIEMEYER, H. M., PESEL, E., FRANKE, S. AND FRANCKE, W., 1989, Ingestion of the benzoxazinone DIMBOA from wheat plants by aphids. *Phytochemistry,* 28 : 2307-2310.

NIKUS FEANETTE, 2003, Beta-glucosidase and hydroxamic acid glucosides. *Agearia*, 387.

PAINTER, R. H., 1951, Insect resistance in crop plants The McMillan Company, New York, USA.

PAN, Y. S., YANG, S. L. AND SHIEH, H. L., 1984, Occurrence of sugarcane woolly aphid *Ceratovacuna lanigera* Zehntner, in relation to sugarcane varieties. *Taiwan Sugar*, 31 : 6-13.

PANSE, V. G., 1957, Genetics of quantitative characters in relation to Plant Breeding. *Indian Journal of Genetics and Plant Breeding*, 36 : 58-61.

PANSE V. G. AND SUKATME, P. V., 1967, Statistical methods for agricultural work ICAR, New Delhi.

PATEL, M. M., PATEL, H. S., PATEL, A. D. AND PATEL, M. P., 1993, Correlation and path analysis in sugarcane. *Indian Sugar*, 43 (6) : 365-368.

PATIL, A. S., SHINDE, V. D., MAGAR, S. B., YADAV, R. G. AND WERKAR, Y. S., 2004, Sugarcane woolly aphid (*Ceratovacuna lanigera* Zehntner) its history and control measures. *Proceedings of the 66th Annual Convention of the Sugar Technologists Association of India*, pp. 133-155.

PATIL, S. B., TIPPANNAVAR, P. S., PATIL, S. A., BALASUNDARAM, N., LINGAPPA, S., GOWDA, M. V. C., KHADI, B. M., SALIMATH, P. M., NAYAKAR, N. Y., BHAT, B. N. AND KAMBAR, N. S., 2005, Identification of clones resistant to the sugarcane woolly aphid (*Ceratovacuna lanigera* Zehntner). *Current Science*, 88 : 284-288.

PHUKAN, E., 1978, Comparative response of certain sugarcane varieties to *Ceratovacuna lanigera* Zehntner (Hemiptera : Aphididae: Pemphigidae) and effect of meteorological factors on the natural population build up of the pest. *M. Sc. Thesis* Assam, Agricultural University of Jorhat, Assam.

PHUKAN, E., DATTA, S. K. AND SHARIFULLAH, M. D., 1988, Population buildup of sugarcane aphid *Ceratovacuna lanigera* Zehntner. *Cooperative Sugar*, 19 : 311-312.

PILLAI, S. V. AND ETHIRAJAN, A. S., 1993, Correlation between yield and yield components at three stages of selection in sugarcane. *Sugarcane*, 4 : 6-10.

PILLEMER, E. A. AND JINGEY, W. M., 1978, Hooked trichomes and resistance of *Phaseolus vulgaris* to *Emposca faba*. *Entomologia Experimentalis et Applicanta*, 24 : 83-94.

PREMACHANDRA, M. N., 1995, Character association and selection of clones in sugarcane under water logging. *Cooperative Sugar*, 27 (3) : 189-192.

PUNIA, M. S., 1981, Path analysis for yield and quality attributes in sugarcane. *M. Sc. (Agri.) Thesis*, Haryana Agricultural University, Hissar.

PUNIA, M. S. AND PARODA, R. S., 1984, Study on association and path coefficient analysis for quality attributes in sugarcane. *Indian Sugar*, 34 (3) : 289-292.

PUNIA, M. S., PARODA, R. S. AND HOODA, R. S., 1983, Correlation and path analysis of cane yield in sugarcane. *Indian Journal of Genetics and Plant Breeding*, 43 : 109-112.

RAMANA RAO, T. C. AND RAO, J. T., 1977, Analysis of quantitative variation in sugarcane hybrid populations involving *Saccharum robustum*. *Proceedings of International Society of Sugarcane Technologists 16th Congress*, pp. 239-243.

RAMANA REDDY, C., 1984, Genetic evaluation of sugarcane genotypes for quality characters. *Maharashtra Sugar*, 9 (2) : 57-59.

RAMANA REDDY, C., 1988, Correlation and path analysis in four intervarietal crosses of sugarcane. *Indian Sugar,* 38(3) : 735-737.

RAO, J. T. AND KRISHNAMOORTHY, T. N., 1968, Accelerating genetic improvement in sugarcane. *Indian Journal of Genetics and Plant Breeding*, 28 : 88-96.

REDDY, C. R. AND REDDI, M. V., 1978a, Studies on reciprocal crosses difference for economic characters in sugarcane (*Saccharum* spp. Hybrid). *Journal of Research*, Andhra Pradesh Agricultural University*,* 15 (2) : 89-96.

REDDY, C. R. AND REDDI, M. V., 1985, Studies on character Associations and degree of genetic determination in sugarcane (*Saccharum* spp.). *Journal of Research,* Andhra Pradesh Agricultural University, 13 (2) : 134-136.,

REDDY, C. R. AND REDDI, M. V., 1986a, Multiple regression analysis on cane yield in twelve intervarietal crosses of sugarcane. *Indian Journal of genetics and Plant Breeding*, 46 (2) : 315-318.

REDDY, C. R. AND REDDI, M. V., 1986b, Degree of genetic determination, correlation and genotypic and phenotypic path analysis of cane and sugar yield in sugarcane. *Indian Journal of genetics and Plant Breeding*, 46 (3) : 550-557.

REDDY, C. R. AND REDDI, M. V., 1987a, Effect of cane yield on the sugar yield of sugarcane. *Indian Journal of Agricultural Sciences*, 57 (10) : 698-700.

REDDY, C. R. AND REDDI, M. V., 1987b, Interrelationship of cane yield with its components. *Sugarcane*, 4 (1) : 14-16.

REDY, O. U. K. AND SOMARAJAN, K. G., 1994, Genetic varability and character association in interspecific hybrids of sugarcane. *Indian Journal of Genetics and Plant Breeding*, 54 (1) : 32-34.

REKHI, H. S. AND GILL, S. S., 1987, Correlation and path coefficient analysis of sucrose per cent in sugarcane. *Bharatiya Sugar*, 12 (5) : 27-30.

RHODES, J. D., CROGNA, P. R., DIXON, A. F. G., 1986, Uptake excretion and respiration of sucrose and amino acid in the pea aphid *Acyryjosiphum pisum. Journal Experimental Biology*, 199 : 1-51.

RICHARD, C. A., 1975, Genetic behaviour of sucrose content in sugarcane. *Ph. D. Thesis*, Louisiana State University, Louisiana.

RISHIPAL, B. S., CHAUDHARY, A. S., MEHTA AND KADIAN, S. P., 1998, Studies on character interrelationship in clonal generation of sugarcane (*Saccharum* spp. Hybrid). *Indian Sugar*, 47 (11) : 907-911.

ROACH, W. A., 1937, Studies on possible cause of immunity. *Annals of Applied Biology*, 24 : 206-210.

RUEDA, L. M. AND CALILUNG, V. J., 1974, Biological studies on the sugarcane woolly aphid, *Ceratovacuna lanigera* Zehntner (Hemiptera : Aphididae : Pemphigidae) of five varieties of sugarcane. *Philippines Entomology*, 3 : 129-147.

SAHI, B. K. AND PATEL, K. A., 1975, Correlation study of quality and yield contributing characters in early maturing sugarcane cultivars. *Indian Sugar*, 25 : 621-624.

SAHI, B. K., 1981, Further studies on heritability of some economic characters of sugarcane. *Ph. D. Thesis*, University of Bombay, Mumbai.

SAIKH, A. R., MUNTAJABUDDIN, K., BORUKAR, D. N. AND UPADHYAY, U. C., 1986, Correlation and path coefficient analysis of yield in sugarcane. *The Indian Sugar Crops Journal*, 12 (2) : 8-9.

SANGWAN, R. S., 1983, Correlation and path coefficient analysis in early maturing genotypes in sugarcane. *Maharashtra Sugar*, 8 : 31-32.

SAXENA, A. P., 1967, Biology of *Campyloneurus mutator* (Fabricius) (Hymenoptera : Braconidae). *Technical Bulletin*, *CIBC*, 9 : 61-72.

SEKHAR, A., 1986, Evaluation of promising types of sugarcane varieties for peninsular zones with particular reference to Tamil Nadu. *M. Sc. (Agri.) Thesis*, Sugarcane Breding Institute, Coimbatore.

SHARMA, M. L. AND KATIYAR, R. B., 1986, Studies on genetic variability, correlation and path analysis in sugarcane germplasm. *Indian Sugar*, 35 (12) : 639-647.

SHARMA, M. L. AND SINGH, H. N., 1984, Genetic variability, correlation and path coefficient analysis in hybrid populations of sugarcane. *Indian Journal of Agricultural Sciences*, 54(2) : 102-109.

SINGH, B. V. AND JAIN, R. P., 1968, Correlation studies in sugarcane. *Madras Agricultural Journal*, 55 : 287-288.

SINGH, H. N., SINGH, S. B., CHAUHAN, R. V. S. AND VISHWAKARAM, R. S., 1983, Variability for yield and quality in sugarcane. *Indian Journal of Agricultural Sciences*, 53 : 786-789.

SINGH, H. N., SINGH, S. B., VERMA, P. S. AND SINGH, R. V., 1981, Correlation and path analysis in sugarcane. *Indian Journal of Agricultural Research*, 15 : 126-130.

SINGH, M. M. AND SINGH, H., 1954, Certain correlation studies in sugarcane. *Proceedings of Conference on Sugarcane Research Development Workers,* Jalandhar, 2 : 70-81.

SINGH, R. K. AND SUDAMA SINGH, 1994, Early evaluation crosses for varietal improvement in sugarcane. *Sugarcane*, 3 : 17-21.

SINGH, R. K., SINGH, D. N., SINGH, S. K. AND SINGH, H. N., 1994, Genetic variability and correlation studies foreign commercial hybrids of sugarcane. *Agricultural Sciences Digest*, 14 (2) : 103-107.

SINGH, R. K., SINGH, S. AND SINGH, G. P., 1995, Genetic variability and yield relationship in adult sugarcane seedlings. *Sugarcane*, 1 : 18-21.

SMITH, G. A. AND JAMES, N. I., 1969, Association in characters within and repeatability between years in progenies of four sugarcane crosses. *Crop Science*, 9 : 819-821.

SNEDECOR, G. W. AND COCHRAN, W. G., 1967, *Statistical Methods,* 6th edition. Edited by Mohan Pramila, Oxford and IBH Publishing Co., New Delhi.

SNELLING, P. O., 1941, Resistance of plants to insect attack. *Botanical Review*, 7 : 543-586.

SREEKUMAR, K., JESSY, KURIAMOSE, M., THOMAS, MATHEW, ALEXANDER, D. AND SANTHAKUMARI, S., 1994, Variability, heritability and correlation on studies on the yield and quality characters of sugarcane. *Indian Sugar*, 44 (4) : 243-249.

SREEKUMAR, S. G., GOPINATHAN NAIR, V., SARASWATHY, P. AND SREEKUMARAN NAIR, S., 1989, Path analysis for cane yield, CCS percentage and sugar yield in the first ratoon crop of sugarcane. *Indian Sugar*, 38 (10) : 809-812.

STEVENSON, G. C., 1965, *Genetics and Breeding of Sugarcane*. Longmans, Green and Co., Ltd., London, p. 201.

TAI, P. Y. P., MILLER, J. D. GILL, B. S. AND CHEW, V., 1980, Correlations among characters of sugarcane in two intermediate selection stages. *Proceedings of International Society of Sugarcane Technologists 17th Congress*, 1119-1128.

TEHLAN, R. S., SINGH, R. K. AND SINGH, F., 1986, Studies on some morphological and quality parameters in relation to cane and sugar yield in sugarcane. *Indian Journal of Sugarcane Technology*, 3 : 38-42.

THAKERY, D. J., MORSE, S. AND LEECH, C., 1998, The role of hydroxamic acids in wheat and maize in conferring resistance to aphids. *Aspects of Applied Biology*, 17 : 225-227.

THRIPATHI, G. M., 1992, Biology of *Conobathra aphidivora* Meyr. Pyralid predator of sugarcane woolly aphid in Nagaland. *Journal of Applied Zoological Research*, 3 : 99-100.

THRIPATHI, G. M., 1995, Record of parasite and predator complex of sugarcane woolly aphid, *Ceratovacuna lanigera* Zehntner in Nagaland. *Indian Sugar*, 44 : 883-885.

TIPPESWAMY, S., KAJJIDONI, S. T., SALIMATH, P. M. AND GOUD, J. V., 2003a, Correlation and path analysis for cane yield, juice quality and their component traits in sugarcane. *Sugar Tech*, 5 (1 & 2) : 65-72.

TIPPESWAMY, S., KAJJIDONI, S. T., SALIMATH, P. M. AND GOUD, J. V., 2003b, Genetic enhancement of cane yield through morpho physiological parameters in sugarcane. *Sugar Journal*, 28 : 34-41.

TIPPESWAMY, S., KAJJIDONI, S. T., SALIMATH, P. M. AND SHOBHARANI, T., 2002, Mean performance of promising sugarcane genotypes for cane yield and juice quality traits in northern dry zone of Karnataka. *Indian Sugar* : 729-733.

TJALLINGI, W. F., 1978, Electronic recording of penetration behaviour by aphids. *Entomologia Experimentalis et Applicata*, 24 : 521-530.

VERMA, P. S., DHAKA, R. P. S. AND SINGH, H. N., 1988, Genetic variability and correlation studies in sugarcane. *Indian Journal of Genetics and Plant Breeding*, 48 : 213-217.

VICTOR, H. A., 1968, Hydroxamic acid content in maize leaf extract and resistance to aphid. *Phytochemistry*, 16 : 35-39.

VICTOR, H. A., LUIS, J. C., HERMANN, M. N. AND BRUCE, C. C., 1983, Toxicity and feeding deterrency of hydroxamic acids from graminae in synthetic diets against the green bug, *Schizaphis graminum*. *Entomologia. Experimentalis et Applicata*, 34 : 134-138.

WALTER, F. S. AND MULLIN, C. A., 1988, Sucrose dependent increased in oligosaccharide production associated glycosidase activities in the potato aphid *Macrosiphum euphorbiae* (Thomas). *Archives of Insect Biochemistry and Physiology*, 9 : 35-46.

WATERHOUSE, D. F., 1993, *The major Arthropod pests and weeds of Agriculture in Southeast Asia*, p. 141.

WEBSTER, J. A., RINYATULLAH, C., HANISSON, M. AND MIRPES, K. A., 1994, Leaf pubescence effects in wheat on yellow sugarcane aphids and green bugs (Homoptera : Aphididae). *Journal of Economic Entomology*, 87 : 231-240.

WHITTAKER, R. H., 1970, The biochemical ecology of higher plants. In E. Sondheimer and J. B. Simeone (eds). *Chemical Ecology*. Academic Press, New York, USA, pp. 43-70.

WRIGHT, S., 1921, Correlation and causation. *Journal of Agricultural Research*, 20 : 557-585.

WRIGHT, S., 1923, Theory of path coefficients. *Genetics*, 8 : 239-255.

WRIGHT, S., 1934, The method of path coefficients. *Annals of Mathematics and Statistics*, 5 : 161-215.

ZEHNTNER, L., 1897, Mededeelingen van het proe of station voor snikerriet in West Java Te Kagok Teal No. 49 De Planteluzien van het Suikerriet of Java X. *Ceratovacuna lanigera* Zehntner. *Archief voor de Java suker industrie, Jerarg* 8 Aft. pp. 13-24.

Appendix I: Mean performance of SWA resistant sugarcane clones for different yield and yield component traits

Clones	Germination per cent	Tiller number per plot	Leaf area (cm^2)	Millable cane height (cm)	Cane girth (cm)	Number of inter nodes	Single cane weight (kg)	No. of millable canes per plot	Juice extract per cent	Brix per cent	Sucrose per cent	Cane yield kg per plot	CCS yield kg/plot
SNK 02	47.50	72.67	234.88	215.00	1.80	17.00	0.53	105.00	67.00	19.47	16.50	73.33	7.99
SNK 44	89.17	153.00	302.57	180.00	2.83	18.33	1.55	174.33	59.67	20.83	17.67	260.67	31.13
SNK 49	76.67	194.67	485.13	283.33	2.90	20.67	1.75	197.00	62.00	19.33	16.60	345.33	38.73
SNK 57	85.42	161.00	494.96	233.33	2.87	18.00	1.58	155.67	57.33	18.97	16.67	244.33	25.72
SNK 61	83.75	241.33	384.60	268.33	2.47	20.00	1.52	176.33	61.00	19.43	17.63	273.00	33.59
SNK 124	75.42	228.67	361.40	270.00	2.27	17.00	1.18	168.67	54.33	18.83	16.50	178.67	20.46
SNK 158	84.44	191.00	447.40	226.67	2.45	17.33	1.30	131.67	55.67	17.90	15.63	166.33	18.55
SNK 192	68.75	89.00	466.28	163.33	2.47	15.33	0.97	171.67	57.67	18.10	15.80	150.00	16.07
SNK 256	88.33	344.33	315.68	283.33	1.83	17.33	0.75	376.67	58.00	16.73	14.57	295.67	29.16
SNK 754	85.83	191.33	685.15	196.67	2.53	19.00	1.09	239.00	58.00	21.27	17.70	252.00	29.88
CoM88121	86.6	187.33	430.00	232.83	2.7	20.00	1.35	163.33	60.66	18.6	15.94	206.00	22.29
CoC671	92.91	156.00	380.0	242.66	30	24.66	1.88	101.66	57.00	17.89	16.26	191.00	21.78
Co8632	95.00	204.66	415.25	228.16	2.79	20.33	1.63	130.00	59.66	18.39	16.01	207.66	23.53
CD at 5%	21.47	63.68	57.79	NS	0.20	2.75	0.395	106.10	NS	1.80	1.69	37.95	6.05
CD at 1%	NS	86.29	79.30	NS	0.27	3.73	0.530	143.75	NS	2.45	NS	51.43	8.21

HISTOLOGICAL AND BIOCHEMICAL BASIS OF RESISTANCE TO WOOLLY APHID IN SUGARCANE (*Saccharum officinarum* L.)

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ABSTRACT

Sugarcane is the leading cash crop of India, the only source of sugar in the country. The occurrence of sugarcane woolly aphid *Ceratovacuna lanigera* (Zehntner) has caused considerable damage threatening sugarcane cultivation itself. Therefore, the present work was carried out to know the possible mechanism and basis of resistance in newly identified resistant clones. Comparative leaf anatomical observations recorded through histological study between SWA resistant and susceptible clones indicated significant anatomical differences for distance between large and small, large and medium and medium and small vascular bundles and the distance was more in susceptible clones and further thickness of parenchyma cells around bundle sheath and thickness of phloem fibre around phloem was more in resistant clones compared to susceptible clones indicating more succulence in susceptible clones encouraging stylet insertion by aphid to suck sap. Histochemical studies for total polysaccharides revealed that resistant clone SNK 192 had low stain and SNK 754 stained rich. Whereas, susceptible clones Co 92020 and CoC 671 stained very rich, however protein and total RNA stained at varying intensity irrespective of resistant and susceptible clones. In biochemical basis of resistance mean observance values for leaf hydroxamic acid (DIMBOA and DIBOA) concentration recorded more in resistant clones and differed significantly between resistant and susceptible clones. Association studies involving ten resistant clones revealed positive significant association of germination per cent, tiller number, number of internodes, cane girth and CCS yield with cane yield at genotypic level. Whereas, at phenotypic level only CCS yield had positive significant association. Path coefficient analysis of cane yield revealed that CCS yield at harvest had large contribution followed by NMC and millable cane height to cane yield. Three resistant clones *viz.,* SNK 754. SNK 044 and SNK 061 were superior over commercial checks for cane yield and sucrose per cent.