CHAPTER - III

RESULTS AND DISCUSSION

Orobanche aegyptiaca is a total root parasite of tobacco (*Nicotiana tabacum* L.) (Plate I,II) and brinjal (*Solanum melongena* L.) (PlateIV). The presence of *Orobanche* shoots in the infected fields of tobacco and brinjal are the most important indications. Parasitized plants do not show any external symptoms except the reduction in size. In heavily infested fields, there is considerable reduction in size as well as yellowing or even wilting of leaves. When *Orobanche* shoot is carefully taken out from the soil along with tobacco and brinjal, it shows a swollen clone connected to infected roots of these plants by means of a very fragile attachment through which it may draw its nurishment. The growth rate and yield of both the plants is greatly decreased due to *Orobanche* infection, which may absorb nutrients, water and minerals from the vascular system of roots of infected plants by means of haustoria.

Orobanche aegyptiaca, is a perennial plant without leaves. Plants erect 6-12 in. high. Stem more or less branched from the base, more or less round, pale brown, scales few, lanceolate, sparingly pubescent or glabrous. Flowers hermaphrodite, irregular in spikes. Flowers sessile or the lower shortly pedicellate, numerous in cylindric spikes; bracts lanceolate, pubescent bracteoles, narrowly linear, shorter than the calyx. Calyx pubescent, campanulate, 4-toothed, submembranous, teeth lanceolate, subulate from a broad base, corolla long pubescent outside, the upper part violet the lower part nearly white, the upper lip 2-lobed, the lobes obtuse or subacute, the lower lip equally 3-lobed, the lobes usually rounded, tube slightly curved near the top, constricted above the insertion of stamens about $\frac{1}{4}$ in. above the base. Filaments inserted low down in the tube, pubescent at the base, anthers obovate, wolly stigma, large with two semi-circular lobes, ovary superior, 1-celled, numerous ovules, fruit 2-volved, capsule containing many seeds. Seeds subglobose, or ovoid reticulate. An abundant amount of seed is produced by parasite those drop in the soil and germinate only in the presence of congenial host. Otherwise the seedsmay lie dormant and remain viable in the soil for as long as thirteen years.

1. Pathological Anatomy :

Tobacco (*Nicotiana tabacum* L.)

Anatomy of Healthy Root :

T.S. of root (Plate VA) shows normal dicot root structure. The epidermis, the outermost layer of the root shows parenchymatous cells which are compressed radially and some of them are disorganized. Below epidermis, there is 2-3 layered periblem made up of parenchymatous cells. The cells of which are radially compressed. The cells are compactly arranged.

There is no distinct endodermis. Inside the periblem there are parenchymatous cells scattered here and there containing some secretory substances. Phloem consists of radial rows of cells. Distinct cambium can not be recognised. The xylem consists of large, angular or circular vessels and tracheids.

Anatomy of Orobanche Infected Root :

The root of tobacco infected with *Orobanche aegyptiaca*, a total root parasite in T.S. (Plate VB) shows many changes in anatomical characters. At the point of contact of root of host and parasite a parenchymatous tissue is developed. From study of many serial sections it seems that there is no direct contact between vascular tissues of parasite and host. The parenchymatous tissue in between vascular cylinders of host and parasite acts as a conducting tissue. The host root shows some drastic changes in anatomical structures. Most of the tissues of cortex are found to be parenchymatous only. There is no distinct epidermis and periblem. The cells of phloem are reduced in size. The secretory cells which are found outside the phloem in healthy root are not seen in root infected with *Orobanche*. The vascular cylinder is reduced drastically. The vessels are few and smaller in size as compared to those in healthy root. The central portion of the root is mostly occupied by parenchymatous cells.

Brinjal (*Solanum melongena* L.)

Anatomy of Healthy Root :

T.S. of root (Plate VI A) shows ruptured epidermis, 2-3

(A) Healthy Tobacco (*Nicotiana tabacum* L.) plant.

(B) Tobacco field infected with *Orobanche aegyptiaca* Pers.

PLATE - I

Tobacco plant infected with Orobanche aegyptiaca Pers.

PLATE-II

(A) Healthy Brinjal (*Solanum melongena* L.) plant.

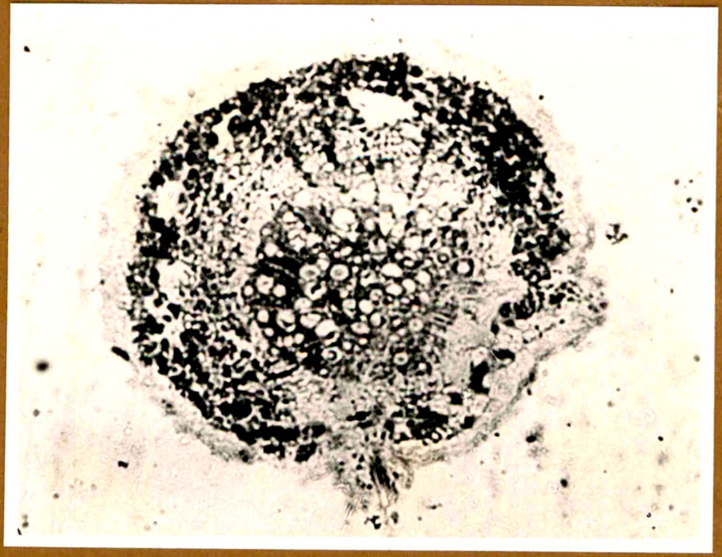
(B) Brinjal field infected with *Orobanche aegyptiaca* Pers.

PLATE-III

Brinjal plant infected with *Orobanche aegyptiaca* Pers.

PLATE-IV

(A) Microphotograph of root of healthy <u>Nicotiana</u> <u>tabacum</u> L. X40NS.



(B) Microphotograph of root of <u>Nicotiana</u> <u>tabacum</u> L. infected by <u>Orobanche</u> <u>aegyptiaca</u>. X40N.S.

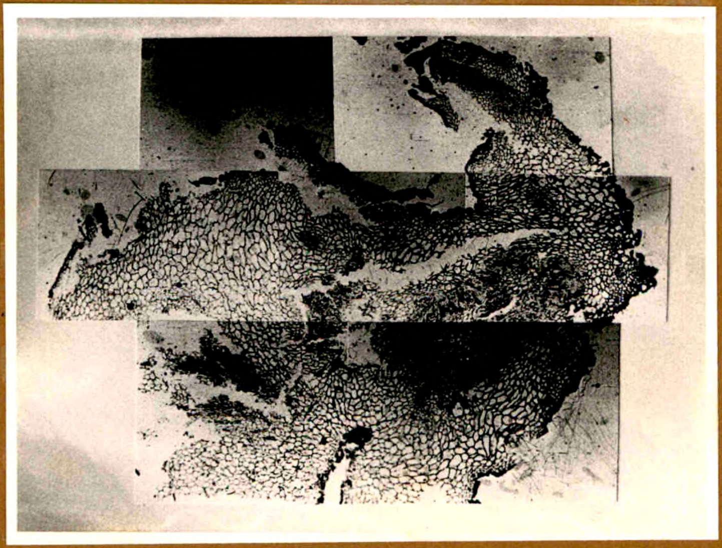
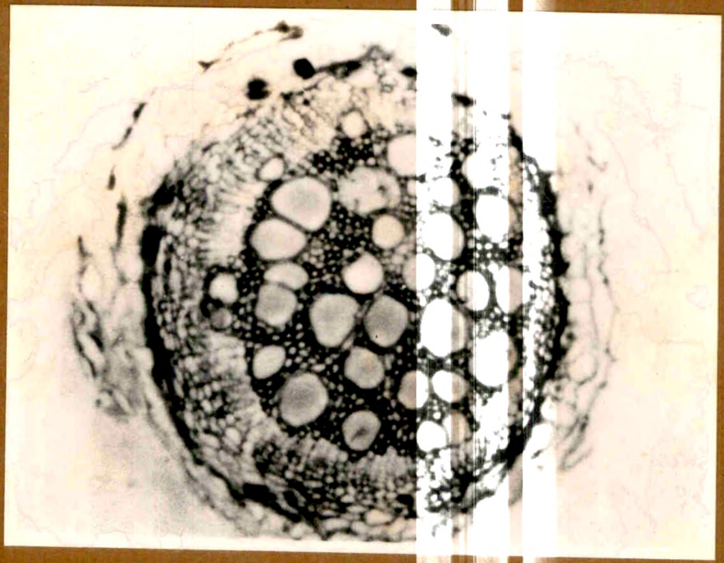


PLATE-V

(A) Microphotograph of root of healthy *Solanum melongena* L. X40N.S.



(B) Microphotograph of root of *Solanum melongena* L. infected by *Orobanche aegyptiaca*. X40N.S.

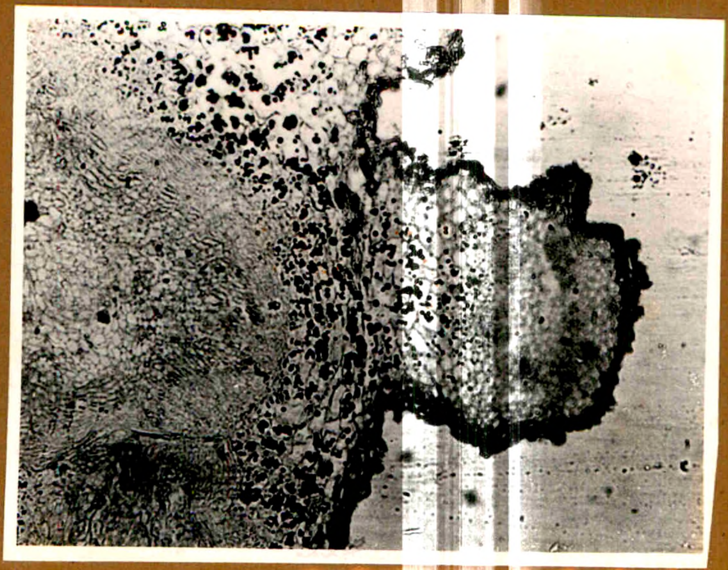
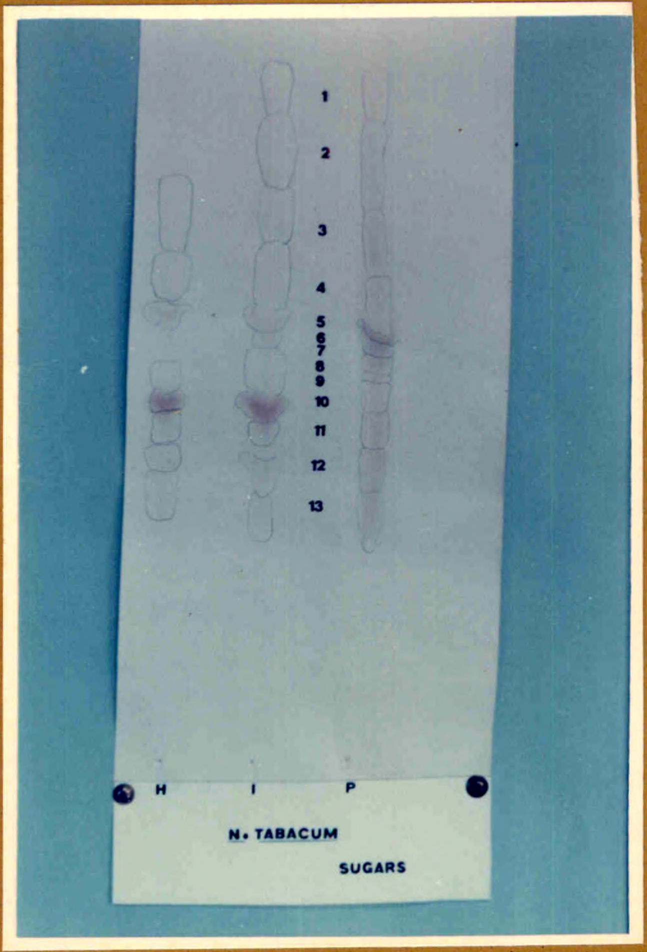


PLATE-VI

(A) Sugar composition of healthy and infected (by <u>Orobanche</u>) leaves of *Nicotiana tabacum* L. and parasite.



(B) Sugar composition of healthy and infected (by <u>Orobanche</u>) leaves of *Solanum melongena* L. and parasite.

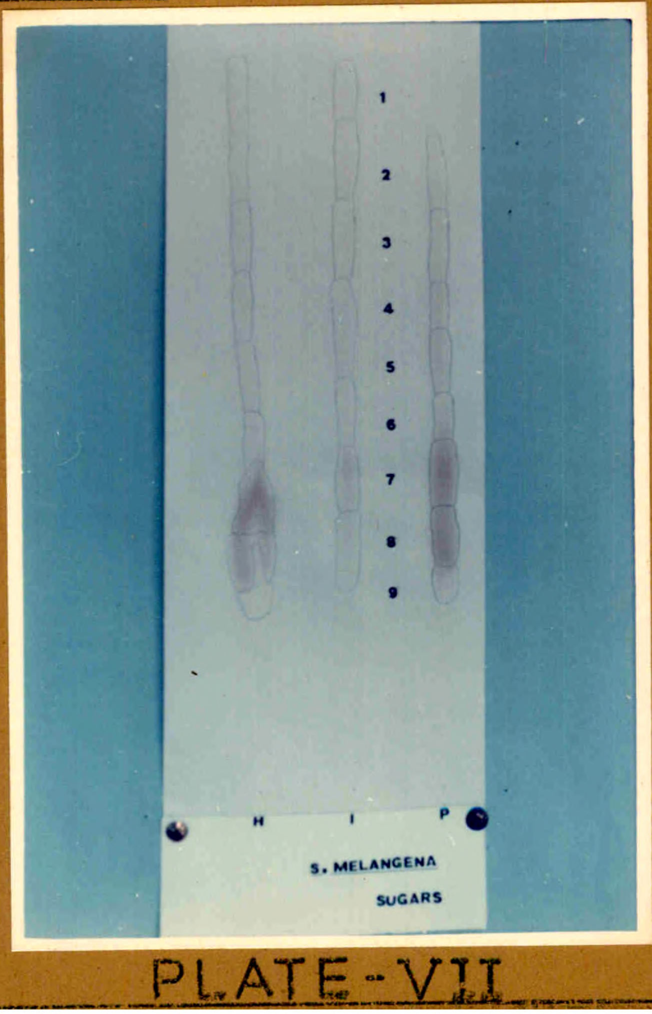
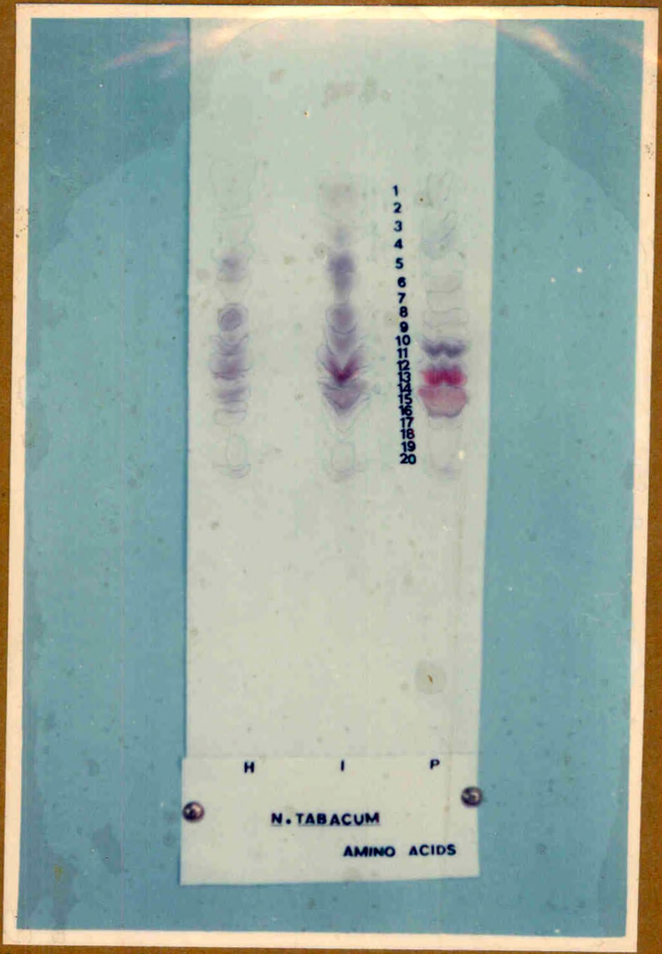


PLATE - VII

(A) Amino acid composition of healthy and infected (by *Orobanche*) leaves of *Nicotiana tabacum* L. and parasite.



(B) Amino acid composition of healthy and infected (by *Orobanche*) leaves of *Solanum melongena* L. and parasite.

PLATE - VIII

layered hypodermis, parenchymatous cortex and vascular tissues.

Epidermal cells are compressed, stretched and are found mostly in ruptured conditions. The hypodermis is made up of compactly arranged rectangular cells without air spaces. The cell walls are slightly lignified. The outer tangential cell walls are more thicker than inner walls. Below hypodermis, there is a cortex of 6-7 layers made up of comparatively large irregularly shaped parenchyma cells containing reserved food materials. Cortical cells contain intercellular spaces. Few sclereid cells are found scattered in cortical region.

Xylem cylinder is surrounded by secondary phloem. The phloem cells are arranged in radial rows. Secondary xylem consists of large vessels and fibres. Vessels are distributed irregularly. The vessels in T.S. are either circular or angular in shape. The vascular rays are usually uniseriate made up of parenchyma cells which contain reserve food. In the centre there are metaxylem and protoxylem elements. The secondary growth seems to be little excentric.

Anatomy of Orobanche Infected Root :

When the parasite, Orobanche comes in contact with host root it develops contact with root and epidermal cells are probably destroyed. / The cortical cells of Orobanche contain
(Plate VI B)

starch grains. Similarly parenchymatous cells containing starch grains grow in the cortical region of root. It forms a tissue which look like a cortex of root of host plant, however, the cortical cells of host tissue do not contain starch grains. The parasitic parenchymatous cells grow upto xylem elements of root of host and almost encircle the xylem cylinder of root of host.

The arrangements of these food absorbing and transferring cells is not definite. They look like cortical cells and are arranged like cortical cells. They are found in contact with xylem and phloem elements of host root and probably absorb the food material and nutrients and transfer to parasitic plant. No vascular connection is developed between the host and parasite. It seems that the absorbing parenchyma is responsible for absorption and transfer of food material and nutrients.

The anatomy of infected root in transverse section shows reduction in all types of tissues. Cortex is reduced. It is having lesser number of cell layers than the cortex in healthy root. Similarly the size of cortical cells of infected root is reduced. The vascular tissue is reduced. The xylem vessels are smaller in size and fewer in number. The xylem seems to be more or less disorganized.

The available reports on <u>Orobanche</u> indicated that <u>Orobanche</u> infection brought about the anatomical changes in roots of host plant. Dorr & Kollmann(1975) studied the structural features of parasitism of Orobanche, where they observed that differentiation of xylem connection of O.crenata with the host Vicia faba tissue by light microscopy. Upon contact with tracheary elements of the host these cells show usual differentiation before turning into a water conducting xylem element.

According to Eames and Laurence (1972) Orobanche makes only one host connection but penetratesdeeply into the root of the host. Saghir et al. (1973) reported that Orobanche infecting tomato seems able to modify host cortical cells into xylem elements, that unite immediately with the stele of the parasite which after ramifying the host tissue form the endophytic system.

Studies on tomato infected with Orobanche ramosa by Saghir and Dastgheib (1978)revealed that xylem elements and undifferentiated parenchyma of the host and parasite were continuous in the haustorium but no phloem continuity with the host has been observed. Anatomical changes resulting from the parasitism of tomato by Orobanche ramosa reported by Pennypacker et al. (1979) showed that the point of attachment of parasite to the host consisted of a mass of undifferentiated polymorphic Orobanche parenchyma cells extending from tomato epidermis to the xylem tissue. The polymorphic cells connected the xylem cells, occasionally penetrated them, and then undifferentiated into parasite-xylem vessel elements. These newly differentiated parasite xylem vessels connected the host xylem to the main vascular system of the parasite. Other polymorphic cells become tightly appressed to phloem sieve cells of the host. *Orobanche ramosa* established both a xylem and a phloem connection with its host.

Sadik *et al*. (1986) have studied the ultrastructure of contact areas between endophytic system of *Arceuthobium oxycedri* and the tissues of the host *Juniperus oxycedrus* which showed that crushed zones often occur around the large cortical strands and around the primary sinkers in the secondary phloem of the host. At the tip of sinkers, the cells of the parasite, in the course of their growth, damage the cells of the host and sometimes penetrate in the lumen of tracheids.

In the present study we have observed in *N.tabacum* root infected with *Orobanche aegyptiaca*, the vascular cylinder is reduced drastically. The vessels are smaller in size as compared to normal root. Cells of the phloem are reduced in size. In infected root of *S.melongena*, the parasitic parenchymatous cells grow upto the xylem elements of root of host and almost encircle the xylem cylinder of root of the host. No vascular connection is developed between the host and the parasite. It seems that the absorbing parenchyma is responsible for absorption and transfer of food material and nutrients. Although the cells connecting the phloem cells and the main body of the parasite was not identified under microscope Orobanche aegyptiaca may obtain needed metabolites from the host vascular system through the specialized parenchyma cells or by the destruction of the host tissue.

2. Physiological Studies :

A) Organic constituents :

i) Chlorophylls :

The influence of Orobanche aegyptiaca, a total root parasite infection on chlorophyll contents of the leaves of Nicotiana tabacum and Solanum melongena has been recorded in Table No.1 and depicted in Fig.1. It is evident that the chlorophyll content is increased in the leaves of infected N.tabacum and that decreased in the leaves of infected S.melongena. The percent increase in the leaves of infected N.tabacum in Chl. 'a' is about 12% and that in Chl. 'b' is about 13%. Thus the total chlorophyll content is increased by about 12%. However, there is a slight decrease in Chl.a/b ratio. On the other hand Chl.'a' in the leaves of infected S.melongena is found to be decreased by about 19%, while the Chl. 'b' level is increased by about 10%. Thus, the total amount of chlorophyll is decreased by about 10%, while the

Table No.1 : Influence of Orobanche aegyptiaca, a total root parasite, on chlorophyll contents* of leaves of Nicotiana tabacum L. and Solanum melongena L.

Plant		Chlorophylls**			
		'a'	'b'	'a + b'	a/b
N.tabacum L.	Healthy	84.77 (100.00)	37.55 (100.00)	128.57 (100.00)	2.25 (100.00)
	Infected	94.99 (112.05)	42.54 (113.28)	144.59 (112.46)	2.23 (99.11)
S.melongena L.	Healthy	83.26 (100.00)	35.92 (100.00)	125.23 (100.00)	2.31 (100.00)
	Infected	67.17 (80.68)	39.43 (109.79)	112.44 (89.78)	1.70 (73.59)

* Values are expressed in mg 100^{-1}g fresh tissue.

** Values in paranthesis are percent of healthy.

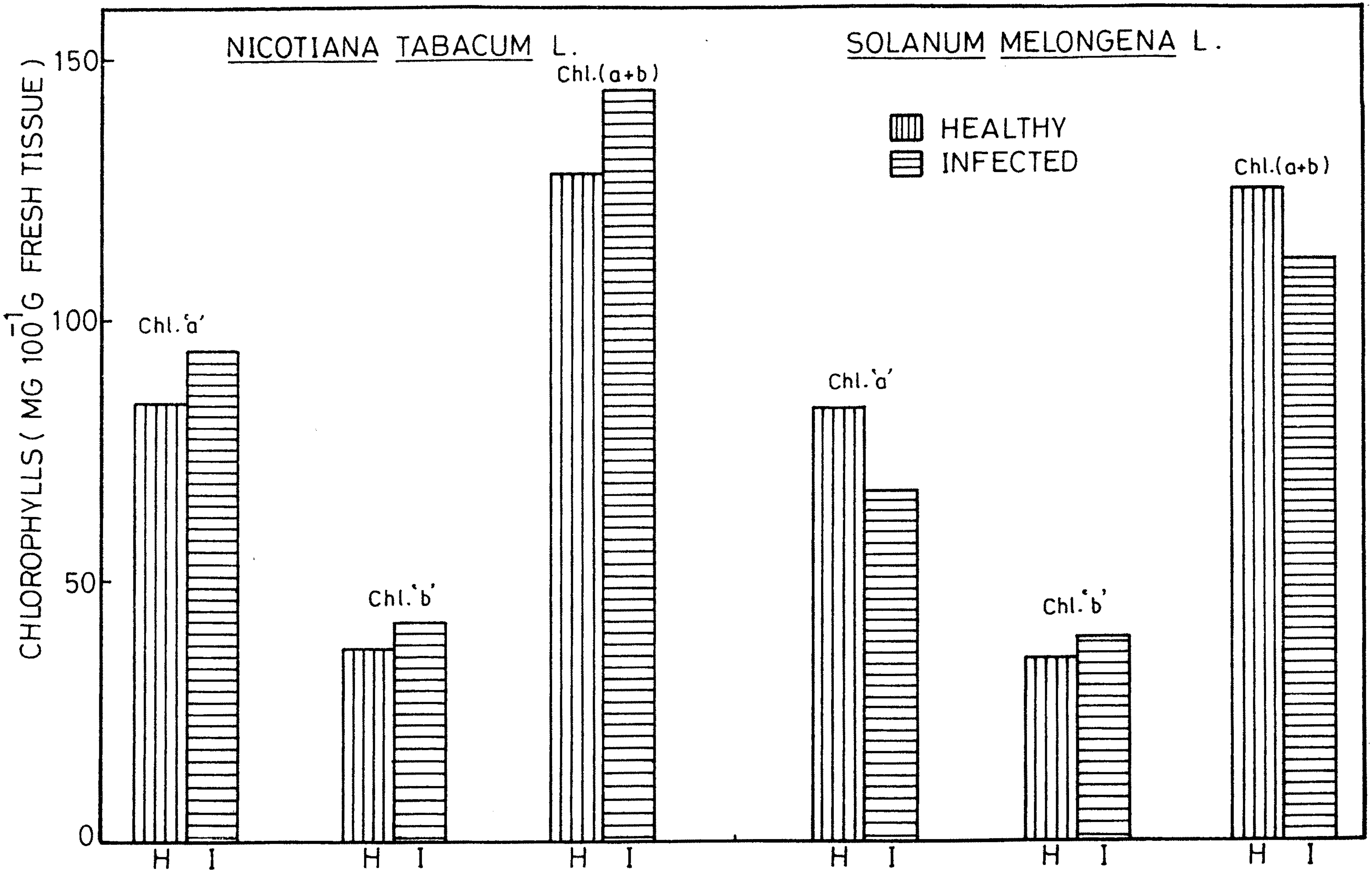


Fig. 1 — INFLUENCE OF <u>OROBANCHE AEGYPTIACA</u>, A TOTAL ROOT PARASITE ON CHLOROPHYLL CONTENTS OF LEAVES OF NICOTIANA TABACUM L. AND SOLANUM MELONGENA L.

Chl. a/b ratio is decreased by about 26% indicating more damage to chlorophyll 'b' due to infection.

Chlorophylls are the main energy trapping pigments, in the process of photosynthesis. According to Allen (1954) and Livne (1964) a reduction in the photosynthesis usually accompanies the chlorophyll loss in the late stages of infection. The chlorophyll content of the leaves is reduced by infection, so that the overall photosynthetic activity of leaves is probably suppressed. The available reports on the effect of *Orobanche* infection on chlorophyll content of host indicated similar trends.

Singh and Krishnan (1971) have reported a decrease in chlorophyll content of the leaves of *Petunia* infected by *Orobanche*. Saghir and Dastgheib (1978) on the other hand have reported that the photosynthesis and protein synthesis were not much affected by parasitism but remained unchanged in *Petunia hybrida* and *Nicotiana tabacum* infected with *Orobanche*. Baccarini and Melandri (1967) have reported a negligible amount of chlorophylls in *Orobanche hederae*, but carotenoids were found to be uniformly distributed throughout the plant. Ismail and Obeid (1976) have reported that *Cuscuta hyalina* and *Striga hermonthica* possess some chlorophyll but there was no trace of any chlorophyll found in *Orobanche ramosa*. Decreased chlorophyll content has also been reported in *Mangifera indica*

Not cited

leaves infected by Dendrophthoe falcata (Loranthaceae) (Kumar and Mukherjee, 1973).

Many reports on fungal infection describe considerable changes in the chlorophyll contents of the host tissue. Considerable decrease in total chlorophylls was reported by Magyarosy *et al.* (1976) in sugarbeet infected by powdery mildew, Siddaramaiah *et al.* (1979) in groundnut affected by rust, Vijayakumar and Rao (1980) in wheat infected by *Alternaria*, Sankpal and Nimbalkar (1980) in sugarcane infected by smut, Johri and Padhi (1981) in groundnut affected by *Cercospora* and Singh *et al.* (1982) in wheat infected by *Puccinia*. Roberts and Wood (1982) have recorded reduced chlorophylls in tobacco leaves infected by cucumber mosaic virus.

Swain and Prasad (1988) have reported a reduction in chlorophylls 'a' and 'b' fractions in rice infected with root-knot-nematode, *Meloidogyne graminicola*, which was consistantly higher in susceptible variety Annapurna, however, in the cultivar Udaya both the fractions showed an increase by 43.5 and 24.3% respectively, which could be due to the ability of plants to compensate for the ill effects of nematode injury to the roots. Increased chlorophyll a/b ratio have also been reported in brinjal infected by little leaf disease (Mitra and Sengupta, 1980).

Several reasons have been put forth to account for the low chlorophyll content. Among those the loss of structural integrity of the chloroplast (Chen and Chen, 1974; Garg and Mandahar, 1977 b; Tamlinson and Webb, 1978; Mitra and Sengupta, 1980), lowered rate of protein synthesis and the accelerated rate of its breakdown (Meyer *et al*., 1960; Tugnawat, 1977; Duggal *et al*., 1981), and the toxic metabolites released by pathogen (Pero and Main, 1970), are worth considering.

In the present investigation, the low chlorophyll content of diseased brinjal can be explained on the basis of above reports. According to Livne (1964) increased photosynthetic rate in many cases is correlated with need for increased catabolism as evidenced by respiratory increase. According to Bottrill and Potassingham (1969), deficiency of minerals is known to decrease the content of chlorophylls. The more likely explaination by Singh and Krishnan (1971) was that the effect of parasite has to decrease the amount of available nitrogen, which in turn affects both protein and chlorophyll synthesis. Smith *et al*. (1969) suggested that the effect of infection by angiosperm parasites resemble those caused by microbial cellular pathogens, a[illegible] decrease in rate of photosynthesis and also decreased chlorophyll content.

It can be suggested that the low content of chlorophylls in *Orobanche* infected *S.melongena* leaves may impair the rate of photosynthesis with consequent decrease in the yield of infected brinjal. Increased chlorophyll content in the leaves of infected *N.tabacum* may be a need for increased catabolism in the infected tissue.

ii) *Polyphenols* :

The polyphenol contents of the leaves of *Nicotiana tabacum* and *Solanum melongena* infected with *Orobanche aegyptiaca*, a total root parasite, and of the parasite has been recorded in Table No.2 and depicted in Fig.2. It is evident that the polyphenol content is increased slightly in the leaves of infected *N.tabacum* (2%) and to some extent in those of *S.melongena* (29%). The parasite on *S.melongena* has shown high level of polyphenols (237% of that in the host) and that on *N.tabacum* also has higher polyphenol content (115% of that in the host).

Khanna *et al*. (1968) have studied total phenolics in three parasitic angiosperms, *Cuscuta species*, *Orobanche aegyptiaca* and *Dendrophthoe falcata* and in their respective hosts. It was found that in *Medicago sativa* infected with *Cuscuta compestris* and in *Impatiens balsamina* infected with *C.compestris* the amount of total phenolics was increased. In *Petunia hybrida* infected with *C.reflexa* also was found to be increased. In *Aegle marmelos*, *Barringtonia acutangula*, *Grevillea robusta*, *Mangifera indica*, *Mimusops elengi* infected with *Dendrophthoe*

Table No.2 : Polyphenol contents* of **healthy** and infected (by _Orobanche aegyptiaca_) leaves of _Nicotiana tabacum_ L. and _Solanum melongena_ L. and parasite

Plant		Total polyphenols**
N.tabacum L.	Healthy	0.60 (100.00)
	Infected	0.61 (101.66)
Orobanche aegyptiaca	Parasite	0.69 (115.00)
S.melongena L.	Healthy	0.38 (100.00)
	Infected	0.49 (128.94)
Orobanche aegyptiaca	Parasite	0.90 (236.84)

* Values are expressed in g 100^{-1} g fresh tissue

** Values in paranthesis are percent of healthy

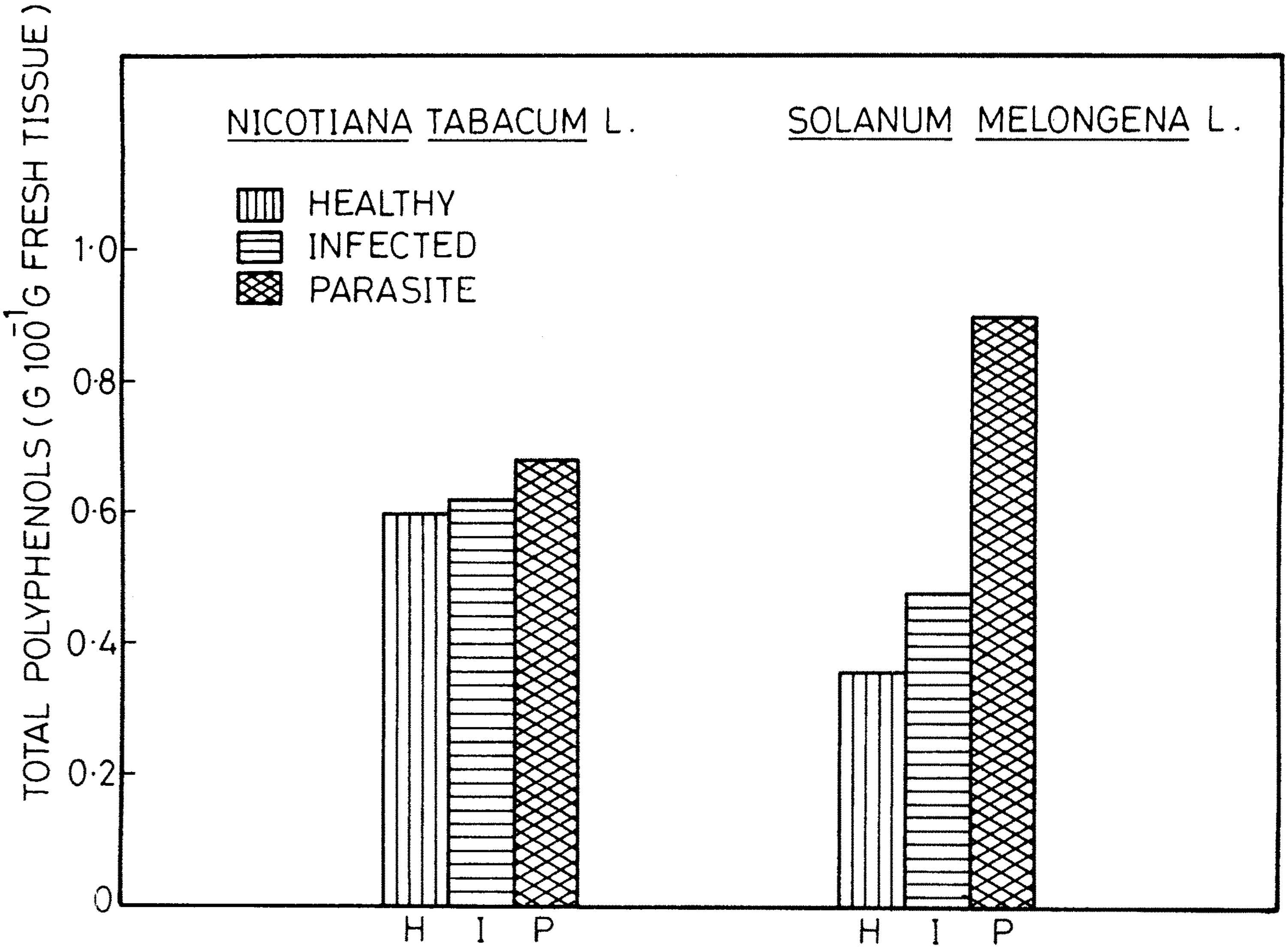


Fig. 2 - POLYPHENOL CONTENTS OF HEALTHY AND INFECTED (BY *OROBANCHE* SP.) LEAVES OF *NICOTIANA TABACUM* L. AND *SOLANUM MELONGENA* L. AND PARASITE.

falcata total phenolics were increased. The parasite contained lower amount of total phenolics than that in its different hosts. Infection by *Orobanche aegyptiaca* was accompanied by an increase in the phenolics in leaves, defoliated stem and roots of *Petunia hybrida*, but there was no demonstratable effect in *Nicotiana tabacum*, whose roots and leaf blades were analysed. However, the parasite tissue contained a higher amount of phenolics than the tissues of the hosts.

Conflicting reports on the effect of fungal diseases on polyphenol content are available. Sivaprakasam *et al*. (1975) in brinjal infected with *Verticillium*, Vijayakumar and Rao (1980) in wheat affected by *Alternaria*, Srinivasan (1968) and Sankpal and Nimbalkar (1980) in red rot and smutted sugarcane, Agarwal *et al*. (1982) in turmeric infected by *Taphrina*, Sharma *et al*. (1983) in maize affected by late blight and have reported a remarkable increase in polyphenol contents. On the other hand the work of Purohit *et al*. (1979), and Dhingra *et al*. (1982) has indicated a decline in the phenolic content of *Cyperus* affected by white rust and in *Brassica* infected with *Albugo* respectively.

Various reasons have been attributed to the accumulation of polyphenols in the diseased plants. High concentration of phenolics seems to be a general feature of parasitic angiosperms. The high amount of phenolics in *Cuscuta* species and in *Orobanche* is parallel in the accumulation of starch (Singh et al., 1968) and phosphate and phytic acid (unpublished data). When the phenolics were calculated in terms of starch free dry weight of tissue (Singh et al., 1968), the disparity between parasite and the host tissue was more prominent. Owing to the limited amount of chlorophyll in Cuscuta (MacLeod, 1962) and the lack of it in Orobanche (Baccarini and Melandri, 1967), the two parasites were to a marked extent dependant on the host for supply of intermediates for carbohydrate transformations. The metabolism of these compounds may be expected to result in accumulation of phenolics as an end product. It is generally agreed that phenolic compounds are involved in same manner in the mechanism of resistance of plants against infection by viral, bacterial and fungal pathogens (Farkas and Kiraly, 1962; Cruickshank and Perrin, 1964; Hare, 1966). However, the phenolics in the host of angiosperm parasites have not been studied so far, in order to explain the susceptibility or resistance to infection or for the possible changes following the infection by the parasites. It is not clear how an accumulation of phenolics protects the infected host against further growth of angiosperm parasite, since the tissues of Cuscuta and Orobanche have a higher content of phenolics as compared to that in the host. One of the functions of phenolics in the tissues may be to control metabolism, by inhibiting a sensitive enzyme (Pridham, 1965). It is likely that the increased amount of phenolics in Orobanche infected N.tabacum and S.melongena may also have similar effects.

iii) Carbohydrates :

The carbohydrate contents of the leaves of Nicotiana tabacum and Solanum melongena, infected with Orobanche aegyptiaca, a total root parasite and that of the parasite has been recorded in Table No.3 and depicted in Fig.3. It is evident that the leaves of infected N.tabacum contain increased amount of reducing sugars, total sugars and starch, but the amount of non-reducing sugars is decreased. The parasite on N.tabacum also shows an increased amount of reducing sugars. However, the amount of non-reducing sugars, total sugars and starch is decreased in the parasite. The leaves of infected S.melongena show increased level of both reducing as well as non-reducing sugars and hence total sugars but starch content is decreased. The parasite on S.melongena has decreased amount of non-reducing sugars but that of reducing sugars, total sugars and starch is increased.

Total carbohydrates :

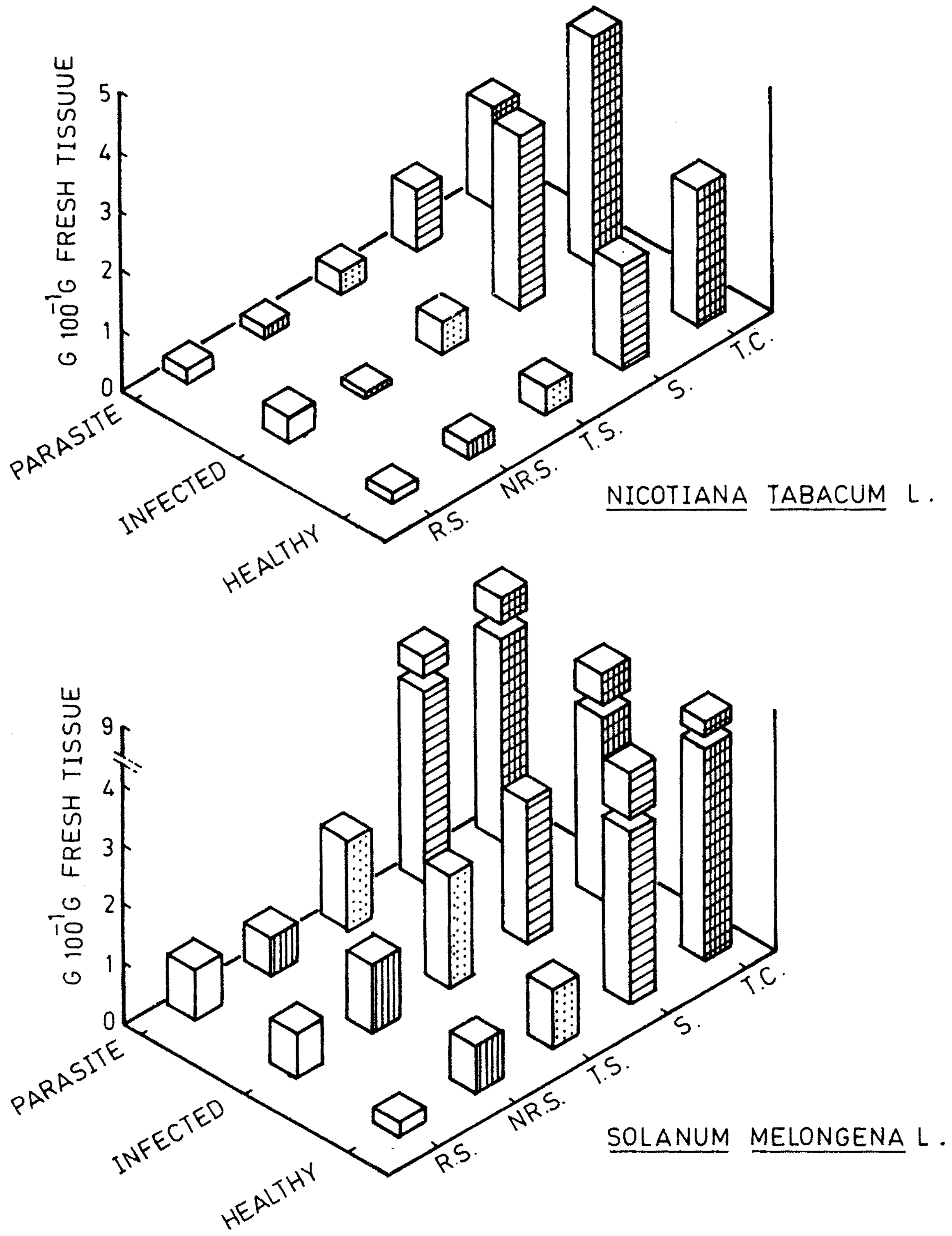
It is observed that the percent increase in total carbohydrates in the leaves of infected N.tabacum was about 68% of that in the leaves of healthy plants. On the other hand the total carbohydrate content in the leaves of infected S.melongena

Table No.3 : Carbohydrate contents* of healthy and infected (by Orobanche aegyptiaca) leaves of Nicotiana tabacum L. and Solanum melongena L. and parasite

Plant		Carbohydrates**				
		Reducing sugars	Non-Reducing sugars	Total sugars	Starch	Total carbohydrates
N.tabacum L.	Healthy	0.15 (100.00)	0.28 (100.00)	0.44 (100.00)	1.75 (100.00)	0.35 (100.00)
	Infected	0.43 (286.66)	0.10 (35.71)	0.54 (122.72)	2.97 (169.71)	3.95 (168.08)
Orobanche aegyptiaca	Parasite	0.25 (166.18)	0.14 (50.00)	0.40 (90.90)	1.06 (60.54)	1.72 (73.26)
S.melongena L.	Healthy	0.27 (100.00)	0.79 (100.00)	1.06 (100.00)	5.16 (100.00)	6.50 (100.00)
	Infected	0.75 (277.77)	1.21 (153.16)	1.96 (184.90)	2.43 (47.09)	5.16 (79.38)
Orobanche aegyptiaca	Parasite	0.88 (325.92)	0.66 (83.54)	1.54 (145.28)	6.69 (129.65)	9.13 (140.46)

* Values are expressed in g 100^{-1}g fresh tissue.

** Values in paranthesis are percent of healthy.



R.S. – REDUCING SUGARS, NR.S. – NON-REDUCING SUGARS, T.S. – TOTAL SUGARS, S – STARCH, T.C. – TOTAL CARBOHYDRATES.

Fig. 3 – CARBOHYDRATE CONTENTS OF HEALTHY AND INFECTED (BY *OROBANCHE* SP.) LEAVES OF *NICOTIANA TABACUM* L. AND *SOLANUM MELONGENA* L. AND PARASITE.

are decreased by about 21% due to parasite effect. On the other hand the parasite on *N.tabacum* has shown 27% decrease and that on *S.melongena* has exhibited about 40% increase in total carbohydrates.

Kumar and Mukherjee (1973) have reported a decrease in the amount of total carbohydrates in *Mangifera indica* infected with *Dendrophthoe falcata*. Singh *et al*. (1972) have also made similar observations in tomato (*Lycopersicon esculentum* Mill.) infected with *Orobanche cernua* and mustard (*Brassica campestris* L.) infected with *O.aegyptiaca*. Singh *et al*. (1979) have further reported that as a result of removal of host plants, the amount of acid hydrolysable carbohydrates decrease in *Orobanche cernua* and *O.aegyptiaca*.

Orobanche parasitism brings about a decrease in the level of total carbohydrates in *S.melongena* leaves, which may be due to hydrolysis of reserve carbohydrates to their simplerforms. Singh *et al*. (1968) have stated that the host plants like tobacco and brinjal respond to *Orobanche* infection with increased phosphorylase activity, whereas Ekaterina (1959) has reported increased hydrolytic activity of invertase in sunflower plants affected by *Orobanche cernua*. The results obtained in the present study fully support these findings that one of the effects of *Orobanche* parasitism is the reduction in the level of stored carbohydrates of the host. However, the parasite on

S.melongena maintains a higher concentration of total carbohydrates, as compared to that in the host plant. As *Orobanche* is devoid of chlorophyll, all sugars must be obtained from the host. Sugars translocated to *Orobanche* from the host are utilized in the elaboration of complex carbohydrates of the parasite. This suggests that *Orobanche* possesses the capacity to synthesize complex reserve carbohydrates from the simple sugars.

Starch :

The observations recorded in Table No.3 and depicted in Fig.3 indicate that *Orobanche* infection also affects the starch content of the leaves of *N.tabacum* and *S.melongena*. It is noted that the starch content is increased in *N.tabacum* (169.57%) and that decreased in *S.melongena* leaves (47.15%) due to infection. But in the parasite on *N.tabacum* this polysaccharide shows a remarkable decrease. However, that in the parasite on *S.melongena* increases by about 30% of healthy.

Singh *et al*. (1968) have reported that starch accumulation is a characteristic feature of angiosperm parasites. They observed the accumulation of starch in four species of *Cuscuta* (*Cuscuta compestris*, *C.indecora*, *C.planiflora* and *C.reflexa*), a leafy mistletoe (*Dendrophthoe falcata*) and chlorophyll lacking parasite, *Orobanche aegyptiaca*. The highest starch accumulation occurred in *O.aegyptiaca* than that in

Petunia hybrida and Solanum melongena. The starch content was decreased in P.hybrida infected with O.aegyptiaca. In Dendrophthoe falcata infected branches of Cassia siamea, Mangifera indica, Melia azedarch and Mimusops elengi, the starch content was found to be decreased. But, the parasite contained greater amount of starch than that in the host. In Cuscuta infected host plants, Lantana camera, Medicago sativa and Petunia hybrida, starch level was fallen down and the parasite contained the highest amount of this polysaccharide. Saghir and Dastgheib (1978) have also recorded significantly decreased starch content in Vicia faba infected with Orobanche crenata.

Fungal infection also alters the level of starch in host plant tissues. Accumulation of starch has been reported by MacDonald and Strobel (1969) in wheat affected by Stripe rust, Sankpal and Nimbalkar (1979) in sugarcane affected by smut, Hwang et al. (1983) in barley infected by Erysiphe and on the other hand Vidhyasekaran and Kandaswamy (1972) in bean affected by powdery mildew, Mogle and Mayee (1981) in pearl-millet infected by downy mildew and Goel et al. (1983) in Coriandrum affected by Protomyces have observed decreased amount of starch.

Several reasons have been attributed to the disturbed starch metabolism in the host. Alabi and Naqvi (1977) suggested that starch is normally not utilized for the growth of pathogen and hence it gets accumulated. Reduction in starch content of the host tissue infected by Orobanche indicates the parasite's dependance on the host intermediates for its starch synthesis and to increase the level of phosphorylase activity in the host, which provides an increased supply of readily diffusable carbohydrates for the parasite. In the present investigation parasitism by Orobanche also resulted in the changes in starch content, but these are not prominent. A decrease in starch content in the leaves of infected S.melongena, however, support the idea that the parasite utilizes starch from the host tissue for its growth.

Soluble sugars :

The observations recorded in Table No.3 indicatedthat the percent increase in the amount of total sugars in the leaves of infected N.tabacum is by about 23%, while that in the leaves of S.melongena is about 85%. The parasite on S.melongena also showed about 45% increase but nearly 9% decrease is observed in the parasite on N.tabacum.

Singh et al. (1972) have reported a decrease in the amount of total sugars in tomato (Lycopersicon esculentum Mill.) infected with Orobanche cernua and mustard (Brassica campestris L.) infected with O.aegyptiaca. Yorganci and Serin (1986) have also reported decrease in the amount of total sugars in tobacco leaves infected with tobacco mosaic virus. Chaudhari and Garg (1955) have made similar observations in brinjal infected with little leaf disease. Sivaprakasam _et al_. (1974) however, could observe accumulation of total sugars in brinjal leaves infected with _Verticillium_.

Fungal infections also cause either an increase or a decrease in the total sugar content of the host tissue. Vidhyasekaran _et al_. (1974) in _Setaria_ affected by rust, Mandokhot _et al_. (1979) in maize infected by _Drechslera_ and Hwang _et al_. (1983) in barley infected with _Erysiphe_ have reported an accumulation of total sugars. While, Alabi and Naqvi (1977), Kapur _et al_. (1978), Sankpal and Nimbalkar (1980), Agarwal _et al_. (1982), Goel _et al_. (1983) have recorded decreased amount of total sugars in groundnut infected by smut, turmeric infected by _Taphrina_, groundnut affected by _Cercospora_, _Coriandrum_ infected with _Protomyces_. Singh and Prasad (1988) have also made similar observations in tomato fruit during pathogenesis by _Corenbacterium michiganese_.

From the present investigation, it is evident that the accumulation of soluble sugars (total sugars) in the leaves of infected _N.tabacum_ and _S.melongena_ may be due to the disturbances in normal phloem transport or due to the release of amylases in the disorganized host cells as suggested by Okasha _et al_. (1968). The accumulation of sugars can be regarded as a nutrient supplying mechanism for the pathogen. Sugars are precursors of phenolics and the accumulation of sugars in the infected leaves would also result in accumulation of phenolics (Vidhyasekaran *et al*., 1971) which may indicate resistance to pathogen or parasite.

Reducing sugars :

In general, infection causes to increase the amount of reducing sugars, though in some cases it is decreased. The results obtained in the present investigation indicate that *Orobanche* affected *N.tabacum* and *S.melongena* leaves contain higher quantities of reducing sugars than that in the healthy ones (Table No.3).

It was noted that the percent increase in reducing sugar content in the leaves of infected *N.tabacum* and *S.melongena* is about 187 and 178% respectively. The parasite on *N.tabacum* has shown 66% increase in the amount of reducing sugars, while the parasite on *S.melongena* has recorded 226% increase in it.

Singh *et al*. (1967) have reported that in *Solanum melongena* infected with *Orobanche aegyptiaca* the amount of reducing sugars, fructose and glucose decreases in the roots. The parasite, however, contain increased amount of glucose. Similar observations have been made by them in *Petunia* infected with *O.aegyptiaca*. Singh *et al*. (1972) have reported higher concentrations of fructose and glucose in the host leaf and root tissues of tomato (*Lycopersicon esculentum* Mill.) and mustard (*Brassica campestris* L.) infected with *Orobanche cernua* and *O.aegyptiaca* respectively, which naturally lead to the higher level of total reducing sugars. Singh *et al*. (1967) have reported remarkable changes in the level of reducing sugars in *Lantana camera* and *Petunia hybrida* infected with *Cuscuta reflexa*. It was found that in *L.camera*, the amount of pentose sugar was increased. In *P.hybrida*, however, fructose was in greater amounts, while glucose level was found to be decreased. They also reported an increase in the level of glucose while a decrease in that of sucrose in *Medicago sativa* infected with *Cuscuta compestris*. Similar trend was observed in the same plant infected with *Cuscuta indecora*. In *Cassia siamea*, *Mangifera indica*, *Melia azedarach*, *Mimusops elengi* infected with *Dendrophthoe falcata*, the host tissues showed lower amounts of fructose and glucose than that in the parasite.

The effect of fungal infection on the reducing sugar contents of the host tissue are conflicting. An increase in reducing sugar content was also found by Garg and Mandahar (1976-a) in Abelmosclus affected by powdery mildew, Sankpal and Nimbalkar (1980) in sugarcane var. CO 740 infected by smut and

Goel et al. (1983) in Coriandrum infected by Protomyces. On the other hand the level of reducing sugars was found to be decreased (Garg and Mandahar, 1977 a) in barley infected by Pyrenophora. Chahal and Kang (1979) in borwn sarson infected by Alternaria, Saini and Suppal (1982) in chicory infected by Alternaria and have also found a decline in the level of reducing sugars.

Various reasons have been advanced for greater amount of reducing sugars in the diseased plants. Yarwood and Jacobson (1955) and Heitefuss and Williams (1976) stated that the diseased tissues and organs may become powerful metabolic sink due to increased metabolic activities at infection sites, resulting in accumulation of reducing sugars. An inverse relationship has been observed between reducing sugars and phenolic contents of the host and hence the tissues containing more reducing sugars will have less phenols, become more susceptible to the particular disease. Horsfall and Dimond (1957), Sohi and Rawal, Gupta and Chatrath (1979), Marimuthu and Kandaswamy (1981) have supported this view.

In the present investigation it was noted that the leaves of infected N.tabacum and S.melongena contained greater amount of reducing sugars. The study of individual sugars by Singh et al. (1967) in Orobanche aegyptiaca infected Petunia hybrida and Solanum melongena indicated that both glucose and fructose occured on O.aegyptiaca. Singh *et al*. (1972) stated that the important translocatory carbohydrate in most of the plant species is sucrose, however, other soluble carbohydrates such as fructose and glucose have also been reported to be translocated. Thus the observed increase in reducing sugar content of the parasite may be due to its translocation from host to parasite.

Non-reducing sugars :

The leaves of infected N.tabacum record nearly 64% decrease in the level of non-reducing sugars (probably sucrose, the major sugar) and also the value for non-reducing sugars in the parasite is about 50% lower than that in the leaves of healthy plant (Table No.3 Fig.3). However, non-reducing sugar content in the leaves of S.melongena infected by **Orobanche** is increased by about 53% over that in the healthy leaves. The parasite, in this case, however, shows nearly 16% decline in the level of non-reducing sugars (Table No.3).

Fungal pathogens also cause the accumulation or depletion of non-reducing sugars in the host plant tissue. Sahi and Rawal (1977) have reported that Septoria leaf spot disease of cowpea caused an increase in the level of non-reducing sugars, while, Gangopadhyay and Chattopadhyay (1977), Sankpal and Nimbalkar (1980), Agarwal *et al*. (1982), Goel *et al*. (1983)

have reported a decrease in the non-reducing sugar contents of rice affected by leaf spot, sugarcane affected by smut, turmeric affected by *Taphrina*, *Coriandrum* affected by *Protomyces* respectively.

Singh *et al*. (1979) have reported that the host shoot removal results in an increase in the amount of fructose and glucose in *Orobanche cermua* and *O.aegyptiaca* at 1st stage of their growth, but higher amount of fructose and glucose was also observed in *O.cernua* at half and full growth stages of *Orobanche* but a decrease was observed in these sugars in *O.aegyptiaca* after removal of the host plants.

The lowered photosynthetic rate in the leaves of *S.melongena* infected with *Orobanche* may be one of the reasons for the increase in the level of non-reducing sugars in this plant.

The chromatographic analysis (Plate VII & Table No.4) revealed that in the leaves of healthy *N.tabacum*, two sugars namely, Xylose and ribose are found to be present prominantly. In the leaves of infected *N.tabacum*, however, sucrose and ribose are detected which are also detected in the parasite on the same plant. The leaves of healthy *S.melongena* have shown the presence of glucose, ribose and xylose in remarkable amounts, however, in the leaves of infected *S.melongena*, only glucose

Table No.4 : Sugar composition of healthy and infected (by *Orobanche aegyptiaca*) leaves of *Nicotiana tabacum* L. and *Solanum melongena* L. and parasite.

Sugar	*N.tabacum*			*S.melongena*		
	Healthy	Infected	Parasite	Healthy	Infected	Parasite
Glucose	-	-	-	+	++	+++
Fructose	-	-	-	+	++	+++
Maltose	-	-	-	-	-	-
Arabinose	-	-	-	-	-	-
Ribose	+	++	+++	-	-	-
Lactose	-	-	-	-	-	-
Mannose	-	-	-	-	-	-
Xylose	+	-	-	+++	-	-
Sucrose	-	+	+	-	-	+

\+ - Present
++ - In appreciable amount
+++ - Prominant
\- - Absent

and ribose are found to be dominant. The parasite appears to be rich in sucrose and ribose.

Reports on the changes in the level of sugars in the host tissue due to *Orobanche* infection indicate both favourable as well as adverse effects on the sugar metabolism. A decrease in the level of fructose, glucose and sucrose in roots and an increase in that of sucrose in the leaves of *Solanum melongena* infected by *Orobanche aegyptiaca* has been reported by Singh *et al*. (1967). However, it was found that the parasite contained increased amount of fructose and glucose, while that of sucrose was decreased. In *Petunia* infected with *O.aegyptiaca* also the same trend was observed. Singh *et al*. (1967) have reported that the amount of pentose increases while that of sucrose decreases in the leaves of *Lantana camera* infected by *Cuscuta reflexa*. In *Petunia* infected with *C.reflexa* the level of glucose and sucrose was found to be decreased while that of pentose sugar was increased. In *Medicago sativa* infected with *C.compestris* there was accumulation of glucose while the level of sucrose was found to be declined. Similar trend of variation was also observed in *M.sativa* infected by *C.indecora*. In *Cassia siamea*, *Mangifera indica*, *Melia azedarach*, *Mimusops elengi* infected with *Dendrophthoe falcata* the host tissue showed lower amounts of fructose and glucose than that in parasite. The sucrose content was found to be increased in the infected *M.azedarach* and *M.indica* while it was decreased in infected *C.siamea* and *M.elengi*. In sugarcane there was three-fold decrease in the level of sucrose and a comparable increase in glucose and other reducing sugars when infected with *Aegintia* (*Orobanchaceae*) (Lee and Goseco, 1932). Whitney (1973) has reported that sucrose is the most common carbohydrate transported in the host plants, but it is glucose and fructose that accumulates in the tissue of parasites.

A reduced level of reducing as well as total sugars has been reported by Hopkins and Hampton (1969) in tobacco infected by tobacco mosaic virus. The adverse effect of fungal infection on total sugar content has been reported by Magyarosy *et al*. (1976), Upadhyay and Dwivedi (1979), Sankpal (1981) and Prasad (1983). Chaudhari and Prasad (1978) have reported fast deplition of glucose, fructose, maltose and raffinose in *Cajanus cajan* plants infected with *Fusarium*. A decrease in the level of sucrose and glucose and that increase in the level of fructose in chilli leaves infected with *Xanthomonas* have been reported by Thind *et al*. (1982).

In the present investigation only a little change in the level of sugars in the leaves of infected *S.melongena* is noted. Sucrose present in the parasite of both the plants indicate that most abundant sugar in the parasite is sucrose similar to the observations of Singh *et al*. (1967). Ribose sugar present in the parasite on both the plants which is also present in the host tissue indicates that the parasite probably derive both the sugars from the host.

iv) Nitrogen Metabolism :

The various fractions of nitrogen of the leaves of *Nicotiana tabacum* and *Solanum melongena* infected with *Orobanche aegyptiaca*, a total root parasite and that of the parasite have been recorded in Table No.5 and depicted in Fig.4 and 5. It is evident that in the leaves of infected *N.tabacum*, nitrate-nitrogen, nitrite-nitrogen and soluble protein-nitrogen contents are decreased and that of insoluble-nitrogen is increased. It is also evident that inspite of some changes in different forms of nitrogen, the total nitrogen content of the host tissue is increased. In the parasite on *N.tabacum* the level of nitrate-N and that of soluble protein-N is increased, while that of nitrite-N and insoluble-N is decreased. However, the total nitrogen content is increased considerably in *Orobanche* on *N.tabacum*. A decrease in the level of nitrate-N, soluble protein-N and total nitrogen and an increase in that of nitrite-N and insoluble-N is observed in the leaves of infected *S.melongena*. The parasite on *S.melongena* has shown a decrease in the level of nitrite-N, insoluble-N and also in that of total nitrogen but the amount of nitrate-N and soluble protein-N is found to be increased.

Table No.5 : Nitrogen fractions* of healthy and infected (by *Orobanche aegyptiaca*) leaves of *Nicotiana tabacum* L. and *Solanum melongena* L. and parasite

Plant		Nitrogen fractions (g 100^{-1}g dry tissue)				
		Nitrate-N	Nitrite-N	Sol-protein-N	Insoluble nitrogen	Total nitrogen
N.tabacum L.	Healthy	0.18 (100.00)	0.054(100.00)	0.098(100.00)	0.53 (100.00)	0.80 (100.00)
	Infected	0.12 (66.66)	0.024(44.44)	0.096 (97.95)	0.83 (156.66)	1.04 (130.00)
Orobanche aegyptiaca	Parasite	0.24 (133.33)	0.005 (9.25)	0.22(224.48)	0.43 (81.13)	0.89 (111.25)
S.melongena L.	Healthy	0.14 (100.00)	0.027(100.00)	0.20 (100.00)	0.83 (100.00)	1.17 (100.00)
	Infected	0.12 (85.71)	0.048(177.7)	0.17 (85.00)	0.86 (103.61)	1.15 (98.29)
Orobanche aegyptiaca	Parasite	0.21 (150.00)	0.009 (33.33)	0.40 (200.00)	0.20 (24.09)	0.81 (70.08)

* Nitrite-N values multiplied by 10^{-2}.

** Values in paranthesis are percent of healthy.

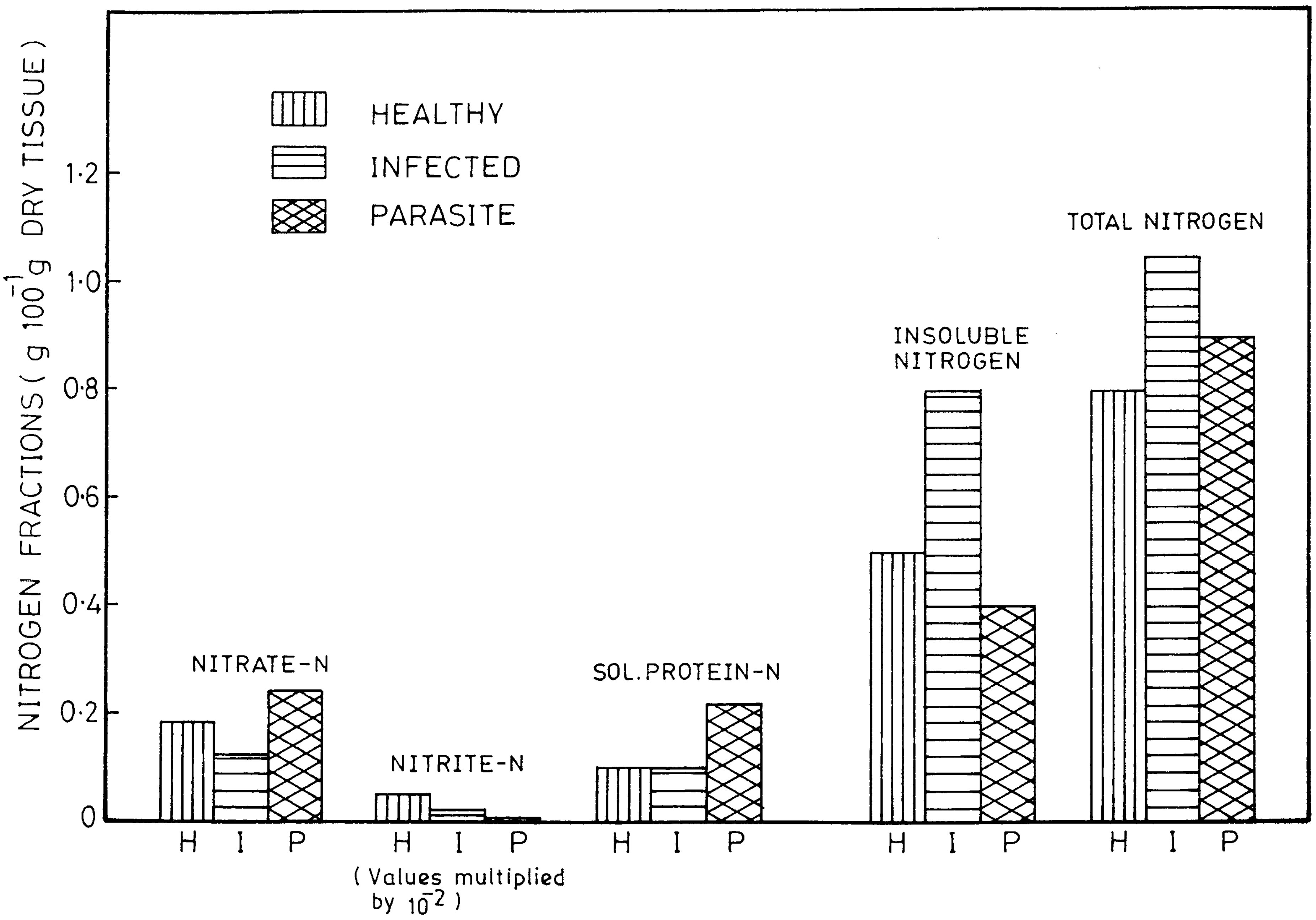


Fig.4 - NITROGEN FRACTIONS OF HEALTHY AND INFECTED (BY OROBANCHE SP.) LEAVES OF NICOTIANA TABACUM L. AND PARASITE.

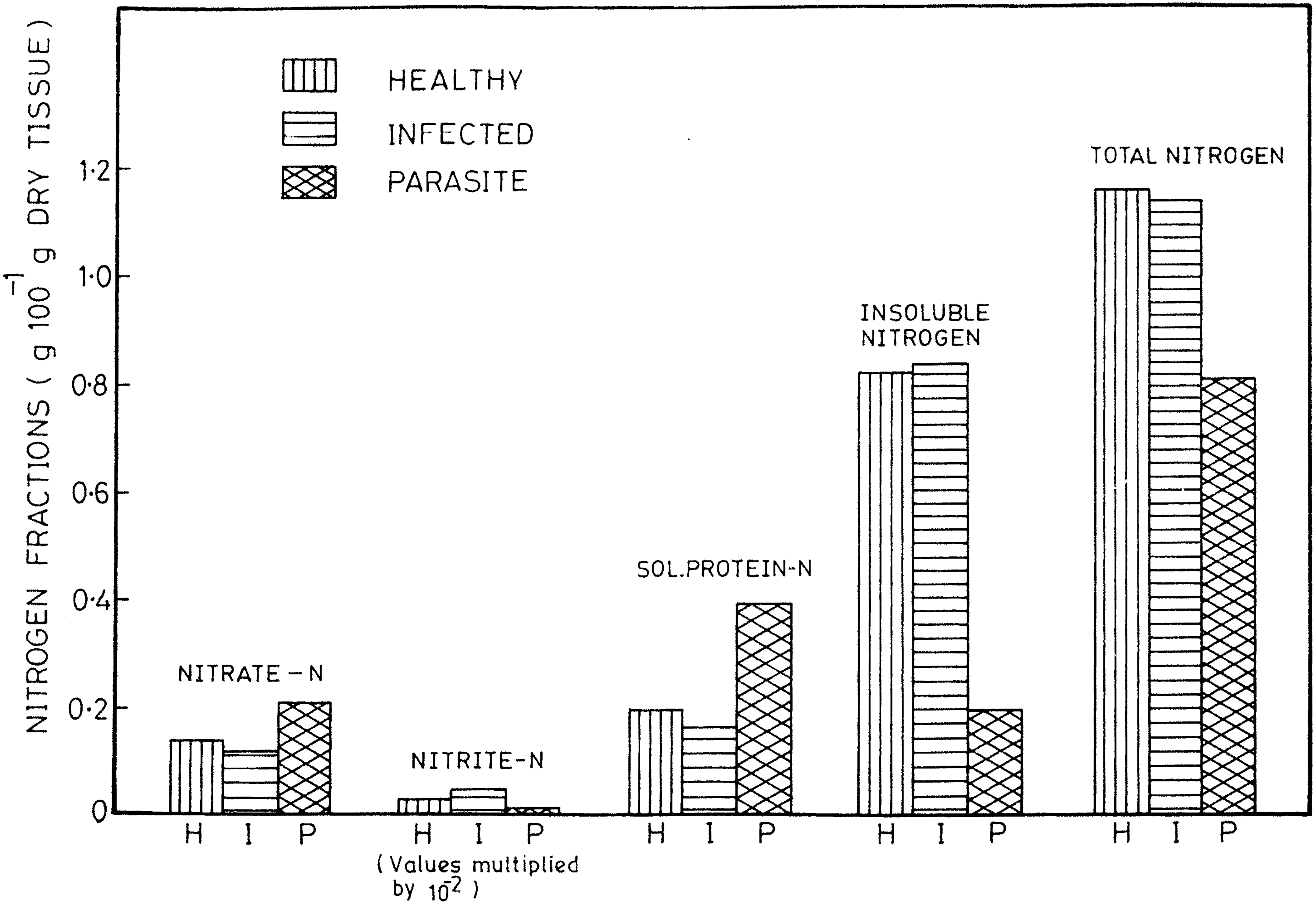


Fig. 5– NITROGEN FRACTIONS OF HEALTHY AND INFECTED (BY *OROBANCHE* SP.) LEAVES OF *SOLANUM MELONGENA* L. AND PARASITE .

Total Nitrogen :

The changes occured in total nitrogen contents of *N.tabacum* and *S.melongena* due to *Orobanche* infection have been recorded in Table No.5 and depicted in Fig.4 and 5. It is clear that the total nitrogen content of the leaves of infected *N.tabacum* is increased by about 30%, but in the leaves of infected *S.melongena* it is slightly decreased by about 1-2%. In the parasite on *N.tabacum* also it is increased by 11% while decreased in *S.melongena* by about 30%.

The available reports indicate that *Orobanche* infection causes remarkable changes in the total nitrogen content of the host plant. Singh *et al*. (1971) have reported that in tomato (*Lycopersicon esculentum* Mill.) infected with *Orobanche cernua* Loeffl. the total nitrogen content increases considerably, especially in the leaves and roots. Singh *et al*. (1972) have reported an increase in total nitrogen content in tomato (*Lycopersicon esculentum* Mill.) infected with *Orobanche cernua* and mustard (*Brassica campestris* L.) infected with *O.aegyptiaca*. While, Saghir and Dastgheib (1978) have recorded a decrease in it in beans infected with *Orobanche crenata*. Similarly, Agrawal *et al*. (1982) have also reported reduced total nitrogen content in *Cistanche tubulosa* (*Orobanchaceae*) infected *Aerva*, *Colliganum* and *Calotropis* roots. It was also found that the parasite contained higher nitrogen content than that in the host roots. Singh *et al*. (1979) observed that the concentration of total nitrogen decreases in *Orobanche cernua* and *O.aegyptiaca* after the removal of host roots. Kumar and Mukherjee (1973) have reported a decrease in total nitrogen content in *Mangifera indica* infected with *Dendrophthoe falcata*.

Generally, in fungus infected plants the total nitrogen and protein contents of the host-pathogen complex increases. An increase in total nitrogen content has been reported by Sindha Mathar and Vidhyasekaran (1978) in sunflower infected with rust and Sankpal and Nimbalkar (1980) in sugarcane infected with smut. On the other hand a decrease in total nitrogen content has been reported by Mujumdar and Raychaudhari (1978) in smutted date palm, Vijaykumar and Rao (1980) in resistant variety of wheat infected with *Alternaria* and Zuber *et al*. (1983) in rice infected with blight disease.

It is reported that MLO infection has both favourable as well as adverse effects on the nitrogen content of the host tissue. Chaudhari and Garg (1955) found a high level of total nitrogen in the leaves of brinjal affected by little leaf disease. Sivaprakasam *et al*. (1976) in brinjal affected by little leaf disease and Raychaudhari and Varma (1980) in sandle affected by spike disease have recorded low nitrogen content in the affected hosts.

An increase in total nitrogen content has been reported by Mukherjee et al. (1979) in tobacco (Nicotiana tabacum) infected with cucumber mosaic virus, Singh and Mall (1978) in pigeon pea fruits infected with arhar (pigeon pea) mosaic virus by Dubey and Joshi (1985) in chilli (Capsicum annum) infected with cucumber mosaic virus.

Nitrogen plays an important role in disease resistance. According to Vidhyasekaran and Kandaswamy (1971), Vidhyasekaran (1974) and Rao et al. (1978) increased nitrogen content results in an increase in protein content which adversely affects the concentration of phenols. Now it is well established that phenol concentration governs the extent of disease resistance.

In the present investigation the observed increase in total nitrogen content in the leaves of infected N.tabacum and parasite of the same plant, which also shows increased polyphenol content may result in an increase in disease resistance. Singh et al. (1972) noticed that nitrogen content of Orobanche was much lower as compared to that of the host. This was probably due to its low nitrogen requirement. The parasite on S.melongena shows a decrease in total nitrogen content which may be due to its low nitrogen requirement similar to the observations of Singh et al. (1972). An increase in total nitrogen content in the parasite of N.tabacum may be due to high rate of translocation of nitrogenous compounds from the host to the parasite.

Nitrate (NO_3^-) nitrogen :

Due to *Orobanche* infection there is about 33% decrease in the nitrate-nitrogen of the leaves of infected *N.tabacum*. In the leaves of infected *S.melongena* also the amount of nitrate-nitrogen is decreased by about 14%. Percent increase in the nitrate-N in the parasite on *N.tabacum* is about 33%, and that on *S.melongena* is about 50%.

Orobanche infection results in an increase in nitrate-N content in stem and roots of tomato (*Lycopersicon esculentum* Mill.) infected with *Orobanche cernua* and mustard (*Brassica campestris* L.) infected with *O.aegyptiaca* as reported by Singh *et al*. (1972). Singh *et al*. (1979) observed that after the removal of host plant, the nitrate-N level was increased in *Orobanche cernua* and *O.aegyptiaca*. But in *O.cernua* it was in negligible amounts during the second stage of its growth.

Parthasarathi *et al*. (1962) have reported an increase in nitrate-N content in sandle infected with spike disease. Singh *et al*. (1978) have also reported an increase in nitrate-N level in cowpea (*Vigna sinensis*) leaves infected with southern bean mosaic virus. Increased level of nitrate-N has also been reported by Tewari *et al*. (1983) in *Cucurbita maxima* fruits infected with three strains of watermelon mosaic virus.

Decrease in the nitrate-N content of the host plants, N.tabacum and S.melongena, while, its increase in the parasite tissue may be due to absorption of nitrate-N by the parasite directly from the soil and inhibition of its utilization in the parasite body due to availability of ready nitrogenous material from the host tissue.

Nitrite (NO_2^-) nitrogen :

It was noted that the percent increase in nitrite-nitrogen content of Orobanche infected S.melongena leaves is about 78%. In the leaves of infected N.tabacum its content is decreased by about 66%. In the parasite on N.tabacum the nitrite-N is decreased by about 91% and that on S.melongena by about 67%.

Singh et al. (1978) have found an increase in the level of nitrite-N in cowpea (Vigna sinensis cv. Laldana) infected with southern bean mosaic virus. Tewari et al. (1983) have also made similar observations in Cucurbita maxima fruits infected with strains of watermelon mosaic virus.

In the present investigation, it was found that the nitrite-N content is decreased in N.tabacum infected with Orobanche. The parasites on N.tabacum and S.melongena have shown a lower amount of nitrite-N which may be due to low nitrite-N requirement by the parasite.

Soluble protein nitrogen :

From the Table No.5 it is clear that in Orobanche infected N.tabacum leaves the soluble protein-nitrogen is slightly decreased by about 2%. However, in S.melongena infected with Orobanche, it is decreased by some more margin (about 15%). It is noted that percent increase in soluble protein-N in the parasite on N.tabacum was greater (124%) than that on S.melongena (100%).

Singh et al. (1978) have reported that in cowpea (Vigna sinensis cv. Lal dana) infected with southern bean mosaic virus the protein-N content was increased. Mukherjee et al. (1979) have also recorded an increase in soluble protein-N in tobacco (Nicotiana tabacum L.) leaves infected with cucumber mosaic virus. Fungal infection also causes remarkable changes in protein-N content of the host tissue. Gangopadhyay and Chattopadhyay (1977) have reported a decline in protein-nitrogen content in rice plants infected with brown spot disease.

In the present investigation, increased amount of soluble protein-nitrogen was observed in the parasite only of both the cases i.e. in N.tabacum and S.melongena which indicates that parasitic organisms direct the synthesis of their own constructive proteins interfering the host metabolism.

Insoluble nitrogen :

The changes in the contents of insoluble nitrogen due to *Orobanche* infection in *N.tabacum* and *S.melongena* leaves as recorded in Table No.5 and depicted in Fig.4 and 5 indicate that the percent increase in insoluble nitrogen in *N.tabacum* (57%) was greater than that in *S.melongena* (4%). It was noted that the percent decrease in insoluble nitrogen in the parasite on *N.tabacum* is by about 19% and that of *S.melongena* is by about 76%.

Singh *et al*. (1972) have reported that when tomato (*Lycopersicon esculentum* Mill.) and mustard (*Brassica campestris* L.) are infected with *Orobanche cernua* and *O.aegyptiaca* respectively, the insoluble nitrogen content of the host tissue (leaf) increases considerably, while in the roots it is increased to some extent. Krishnarao and Nayadu (1979) have found decreased amount of insoluble nitrogen in rice leaves infected with *Xanthomonas oryzae*.

From the present investigation, it appears that the increase in insoluble nitrogen content of *N.tabacum* and *S.melongena* leaves due to *Orobanche* infection and that decrease in parasite tissue may be due to its low requirement.

The chromatographic analysis (Plate VIII & Table No.6) has revealed that DL-alanine, DL-aspartic acid, DL-DOPA, DL-methionine,

Table No.6 : Amino acid composition of healthy and infected (by *Orobanche aegyptiaca*) leaves of *Nicotiana tabacum* L. and *Solanum melongena* L. and parasite

Amino acid	*N.tabacum*			*S.melongena*		
	Healthy	Infected	Parasite	Healthy	Infected	Parasite
1. DL-Alanine	+++	++	+++	++	++	-
2. DL-Aspartic acid	++	-	-	++	-	+++
3. DL-DOPA	+++	-	+++	-	-	+
4. L-Histidinemonohydrochloride	-	-	-	+	-	-
5. DL-nor-Leucine	-	-	-	++	++	-
6. DL-Methionine	++	+++	+	++	-	-
7. L-Proline	++	+++	+	-	++	-
8. DL-Tryptophan	-	-	-	-	-	-
9. DL-α Amino-n-butyric acid	-	-	-	-	+	-
10. L-Cysteinemonohydrochloride	-	-	-	-	-	-
11. Glutamic acid	++	+	+++	-	-	-
12. Hydroxyproline	++	+++	-	+++	-	-
13. L-leucine	-	-	-	-	-	-
14. L-Orinithinemonohydrochloride	+++	+++	-	-	-	++
15. DL-serine	-	++	-	-	++	-
16. L-Tyrosine	+	++	+++	-	+	+
17. L-Argininemonohydrochloride	-	+++	+++	-	-	+
18. L-Cystine	-	-	-	-	++	-
19. Glycine	-	-	+++	-	+++	-
20. DL-iso-Leucine	-	-	-	-	+	++
21. L-Lysinemonohydrochloride	-	-	-	++	-	+
22. DL-β-Phenylalanine	+++	+++	+++	+	+	-
23. DL-β-Threonine	-	+	-	-	-	-
24. DL-Valine	+	+	+	-	-	+

+	Present
++	In appreciable amount
+++	Prominent
-	Absent

DL-proline, glutamic acid, hydroxyproline, L-ornithinemonohydrochloride, L-tyrosine, DL-β-phenylalanine and DL-valine are found to be present in the leaves of healthy N.tabacum. However, serine, arginine and threonine in addition to the amino acids detected in the leaves of healthy plant, are present in the leaves of infected N.tabacum. The amino acid DL-DOPA is not detected in the leaves of infected N.tabacum. In the parasite DL-alanine, DL-methionine, L-proline, DL-DOPA, glutamic acid, L-tyrosine, L-argininemonohydrochloride, glycine, DL-β-phenylalanine and valine are detected. On the other hand in the leaves of healthy S.melongena DL-alanine, DL-aspartic acid, DL-histidinemonohydrochloride, DL-nor-leucine, DL-methionine, hydroxy proline, L-cystine, L-lysinemonohydrochloride and DL-β-phenylalanine are found to be present. The leaves of infected S.melongena have shown the presence of DL-alanine, DL-nor-lecuine, L-proline, DL-2-amino-n-butyric acid, DL-serine, L-tyrosine, L-cystine, glycine, DL-isoleucine and DL-β-phenylalanine. In the parasite, on this plant, DL-aspartic acid, DL-DOPA, L-ornithinemonohydrochloride, L-tyrosine, L-argininemonohydrochloride, DL-isoleucine, L-lysinemonohydrochloride and DL-valine are detected.

Chromatographic determination of the amino acid components by Misra et al. (1970) has revealed that the apical region of Cuscuta differs quantitatively from the haustoria, bearing reason in containing β-alanine, glycine and two unidentified amino acids. The interveining regions contained only β-alanine and glycine but not the other unidentified amino acids. The apical region contained lower amounts of alanine, aspartic acid, isoleucine, leucine, phenylalanine, serine and valine and the higher amount of cysteine, cystine, histidine, lysine, proline, tyrosine and another unidentified amino acid. Arginine, and glutamic acid were the most abundant amino acids in all the regions of the parasite vine.

Favourable as well as adverse effects of fungal infection on the concentration of amino acids in the host tissue have been reported. The work of Prasad and Chaudhari (1975) in the leaf 24 tissues of *Cajanus cajan* infected with *Fusarium* and Mogle and Mayee (1981) in pearlmillet affected by stem gall disease revealed that the decreased amino acid content in the host tissue is due to the pathogen attack. A decrease in the concentrations of lysine, histidine, serine, aspartic acid, valine tryptophan, β-alanine, and glycine in the seedlings of sesamum infected with *Aspergillus* has been reported by Misra *et al.* (1988). On the other hand an increased level of free amino acids and amides has been reported by Raggi (1974) in beans affected by rust, Thompson (1978) in the roots of potato infected with *Rhizopus*, Patil *et al.* (1979) in wheat infected by *Alternaria*, Sankpal (1981) in smutted sugarcane and by Goel *et al.* (1983) in Coriandrum affected by stem gall disease. The amino acids especially aspartate, alanine, glycine, serine, methionine,leucine and the amides like aspergine and arginine were found to be accumulated to a greater extent.

Barna and Janos (1981) suggest that the high concentration of amino acids creates unfavourable conditions for the growth of pathogen and thus induces disease resistance. They have further suggested that the amino acids (tryptophan, phenylalanine) participate in the phenol metabolism, which also adds to the inhibition of growth and multiplication of the pathogen.

Various reasons have been put forth to account for accumulation or reduction in amino acid contents in the diseased plants. This increase in the level of amino acids may be due to accelerated proteolytic activity or due to the impaired translocation of amino acids and their utilization. While, the fall in the amino acid level may be due to their rapid utilization by the pathogen towards increased protein synthesis.

The level of both bound and free amino acids in the three mistletoe genera, Amyema, Aroceuthobium and Phoradendron (Greenham and Leonard, 1965) and the presence of aspartate and glutamate transaminases in Cuscuta extracts (MacLeod, 1963) attest to a high level of protein synthesis. Since these enzymes can produce several amino acids from corresponding α-keto acids and glutamic acid and their presence suggest that the parasite does not need to obtain all its amino acids from the host (MacLeod, 1963). Generally the amino acids found in the parasite resemble to those in the host.

In the present investigation we have noted an increase in the level of amino acids in the leaves of N.tabacum and S.melongena which probably creates unfavourable conditions for the growth of parasite and thus may induce disease resistance, while the decrease in amino acids in the parasite may be due to the rapid utilization of amino acids by the parasite.

v) Nicotine in Tobacco :

The nicotine content of the leaves of Nicotiana tabacum infected with Orobanche aegyptiaca, a total root parasite and that of the parasite has been recorded in Table No.7 and depicted in Fig.6. It is evident that the leaves of healthy N.tabacum contain 0.93% nicotine (dry wt basis). In the leaves of infected N.tabacum there is about 196% increase in the level of this substance. However, the parasite tissue has shown a decrease in nicotine content by about 9%.

Orobanche infection brings about considerable changes in nicotine content of the host plant. As the synthesis of nicotine is known to take place in the root (Dawson, 1942) and as Orobanche is a total root parasite the nicotine content of the host tissue is found to be affected in the diseased tobacco

Table No.7 : Nicotine content* of healthy and infected (by *Orobanche aegyptiaca*) leaves of *Nicotiana tabacum* L. and parasite

Plant		Nicotine**
N.tabacum L.	Healthy	0.93 (100.00)
	Infected	2.75 (295.69)
Orobanche aegyptiaca	Parasite	0.85 (91.39)

* Values are expressed g 100^{-1}g dry tissue.

** Values in paranthesis are percent of healthy.

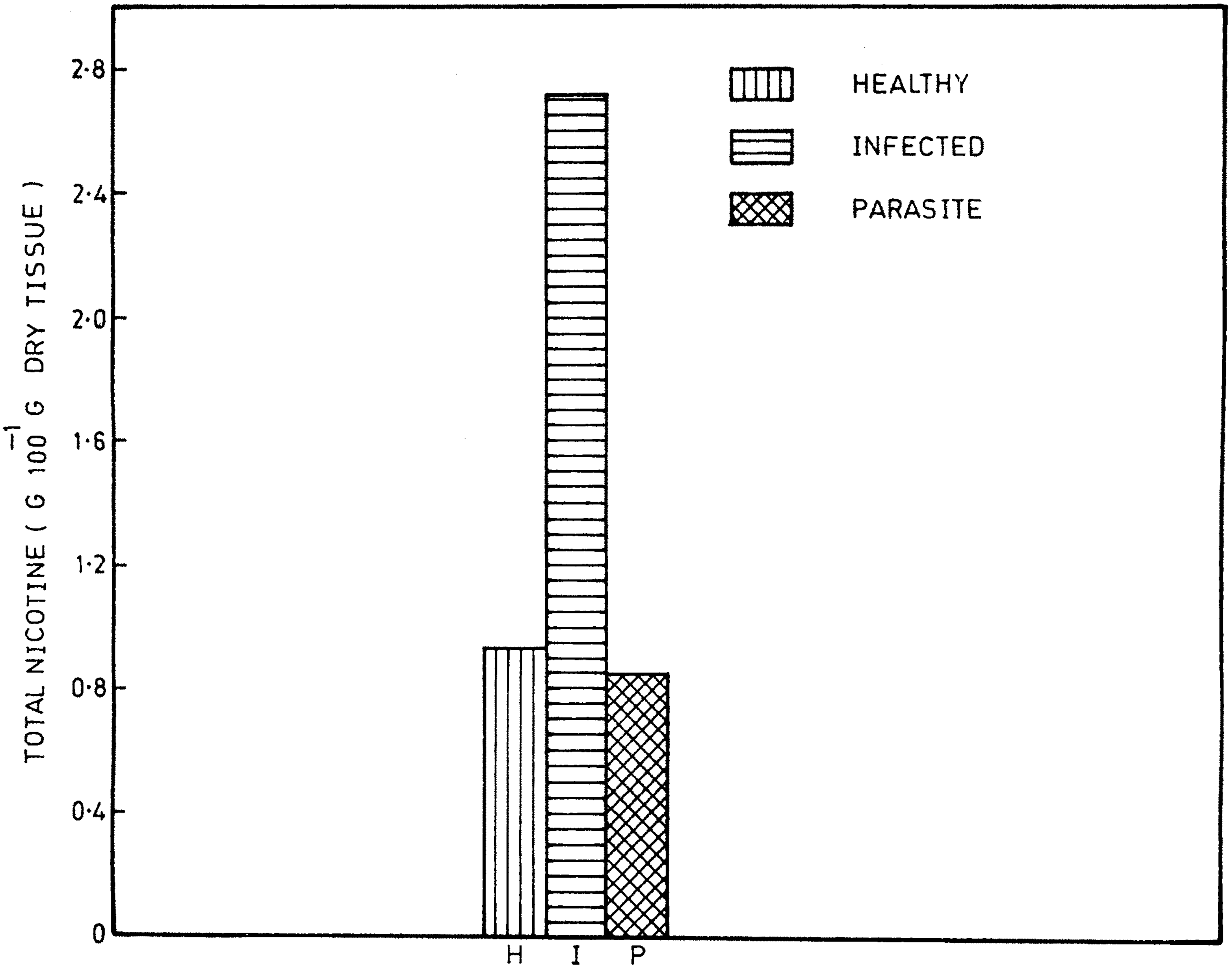


Fig.6 - NICOTINE CONTENT OF HEALTHY AND INFECTED (BY *OROBANCHE* SP.) LEAVES OF *NICOTIANA TABACUM* L. AND PARASITE.

plants. Iskender (1975) has reported that the nicotine content increases in mildew resistant Turkish tobacco but in mildewed tobacco the nicotine content was much lower than that in the healthy plants. Gujar and Hegde (1983) have reported that the nicotine level in the leaves of tobacco plants decreases by about 7% due to TMV infection when the disease incidence was about 70%. Davis and Rich (1987) have reported an increase in the level of nicotine content in tobacco roots infected with nematode, *Meloidogyne incognita* and this increased nicotine content was toxic to nematode infection indicating its relation to the disease resistance.

It is a well established fact that the nicotine metabolism is markedly influenced by environmental stress. Disease is also a type of stress. Here it is caused by *Orobanche*, a total root parasite which is likely to adversely affect the nicotine metabolism in tobacco. Silberschmidt (1930) has shown that the nicotine content is increased in TMV infected leaves at the expense of carbohydrates. In the present investigation we have also noted an increased amount of nicotine in the leaves of infected *N.tabacum*. Nicotine is an alkaloid and as alkaloids are nitrogen containing compounds, the availability of nitrogen is expected to play an important role in biosynthesis and accumulation of such alkaloids in the plant. According to Gujar and Hegde (1983) reduction in the level of nicotine content can be reconciled both with reduction in the photosynthetic leaf area as well as decreased rate of photosynthesis. We have noted a decrease in the level of nicotine in the parasite which may be due to the lack of chlorophyll in the parasite which may be one of the reasons for decreased nicotine content, similar to the findings of Gujar and Hegde (1983).

vi) Organic acids :

It is evident from Table No.8, that succinic, malic, oxalic and tartaric acids are found to be present in the leaves of healthy *N.tabacum*. In the leaves of infected *N.tabacum*, however, succinic, citric and ascorbic acids are detected. The parasite has also shown the presence of succinic, citric and tartaric acids. On the other hand in the leaves of healthy *S.melongena*, succinic, malic and citric acids, are found to be present. However, in the leaves of infected *S.melongena* succinic and fumaric acids are detected, while in the parasite the same organic acids have been found to be present predominantly.

The diseases caused by fungal pathogens also induce the changes in the level of organic acids of the host plant. Increase in the level of organic acids was reported by Sankpal (1981) in smutted sugarcane leaves and Reddy and Rama Gopal (1982) in rust infected groundnut which was as it was suggested due to the accelerated synthesis of malate, citrate and succinate, while, the work of Magyarosy *et al*. (1976) in sugarbeet

Table No.8 : Organic acid composition of healthy and infected (by *Orobanche aegyptiaca*) leaves of *Nicotiana tabacum* L. and *Solanum melongena* L. and parasite

Standard organic acid	*N.tabacum*			*S.melongena*		
	Healthy	Infected	Parasite	Healthy	Infected	Parasite
Succinic acid	+	++	+++	+	+	++
Malic acid	+	-	-	+	-	++
Oxalic acid	+	-	-	-	-	-
Citric acid	-	+	+	+	-	++
Ascorbic acid	-	+	-	-	-	-
Fumaric acid	-	-	-	-	+	-
Tartaric acid	+	-	++	-	-	-

+ - Present
++ - In appreciable amount
+++ - Prominant
- - Absent

infected by powdery mildew has indicated slow rate of synthesis of organic acids in affected plant tissue. A decrease in the level of citric and fumaric acids in leaf leachates of barley, infected with *Alteranaria* has been reported by Dixit and Gupta (1983).

In the present study we have observed that the level of organic acids in the leaves of infected *N.tabacum* and *S.melongena* is altered to some extent due to *Orobanche* infection. The presence of succinic, malic, oxalic and tartaric acids in the leaves of healthy *N.tabacum* and their absence except that of succinic acid in infected leaves may be due to deterioration or utilization by pathogen which shows the similarity with the findings of several workers like Farkas and Kiraly (1962), Kapoor and Tandon (1969). It seems likely that succinic, citric and tartaric acids in the parasite on *N.tabacum* may be derived from the host. Work of Vayonis (1954) on virus disease of tobacco indicated that increase in phosphorus content causes an increase in the rate of respiration. Dekock (1964) has reported that the disturbances in the normal working of TCA cycle may result in the accumulation of malate and citrate. The increase in the level of organic acids increases the susceptibility of the host as indicated by work of Schlegel (1957), Nour-Eldin (1955) and Taniguchi (1962).

B. Inorganic constituents :

i) Sodium (Na^+) :

The sodium content of the leaves of *Nicotiana tabacum* and *Solanum melongena*, infected by *Orobanche aegyptiaca*, a total root parasite and that of the parasite has been recorded in Table No.9 and depicted in Fig.7. It can be seen that the sodium content is increased in the leaves of infected *N.tabacum* by about 26%. However, it is decreased in those of infected *S.melongena* by about 16%. The parasites on *N.tabacum* and *S.melongena* have shown an increase in the level of sodium content by about 152 and 93% respectively. The parasites on both the plants have shown higher amounts of sodium than that in the leaves of infected plants.

Sodium is a functional element for all terrestrial plants (Nicholas, 1961). Sodium can replace potassium partly. It is an activator of transport ATP-ases in animals and possibly in plants. This monovalent nutrient is required for photosynthesis to increase the CO_2 assimilation. Brownell and Crossland (1972) demonstrated that C_4 plants require Na^+ as an essential nutrient. Though sodium is found in tobacco plant, it does not seem to have any active role in its life (Indian Tob. Mongr., 1961).

Very scanty information is available on the effect of

Table No.9 : Inorganic constituents* of healthy and infected (by *Orobanche aegyptiaca*) leaves of *Nicotiana tabacum* L. and *Solanum melongena* L. and parasite.

	N.tabacum			*S.melongena*		
	Healthy	Infected	Parasite	Healthy	Infected	Parasite
Sodium	4.13 (100)	5.21 (126.15)	10.43(252.54)	6.74 (100)	5.65(83.83)	13.04(193.47)
Potassium	20.26 (100)	23.52 (116.09)	58.88(290.62)	29.15 (100)	25.06(85.97)	43.98(150.87)
Calcium	263.47 (100)	335.32 (127.27)	123.75(46.97)	171.65 (100)	255.48(148.84)	195.60(113.95)
Magnesium	78.28 (100)	101.30 (129.41)	223.66(285.72)	473.64 (100)	63.15(13.33)	19.07(4.03)
Phosphorus	31.47 (100)	32.28 (102.57)	25.82(82.05)	61.33 (100)	40.35(65.79)	38.74(63.17)
Iron	2.22 (100)	2.78 (125.23)	3.41(153.60)	1.74 (100)	1.16(66.68)	4.13(237.36)
Zinc	0.11 (100)	0.12 (109.09)	0.07(63.64)	0.10 (100)	0.06(60.00)	0.07(70.00)
Copper	0.10 (100)	0.13 (130.00)	0.06(60.00)	0.06 (100)	0.05(83.33)	0.05(83.33)

* Value are expressed in Meq 100^{-1} g dry tissue.

** Value in paranthesis are percent of healthy.

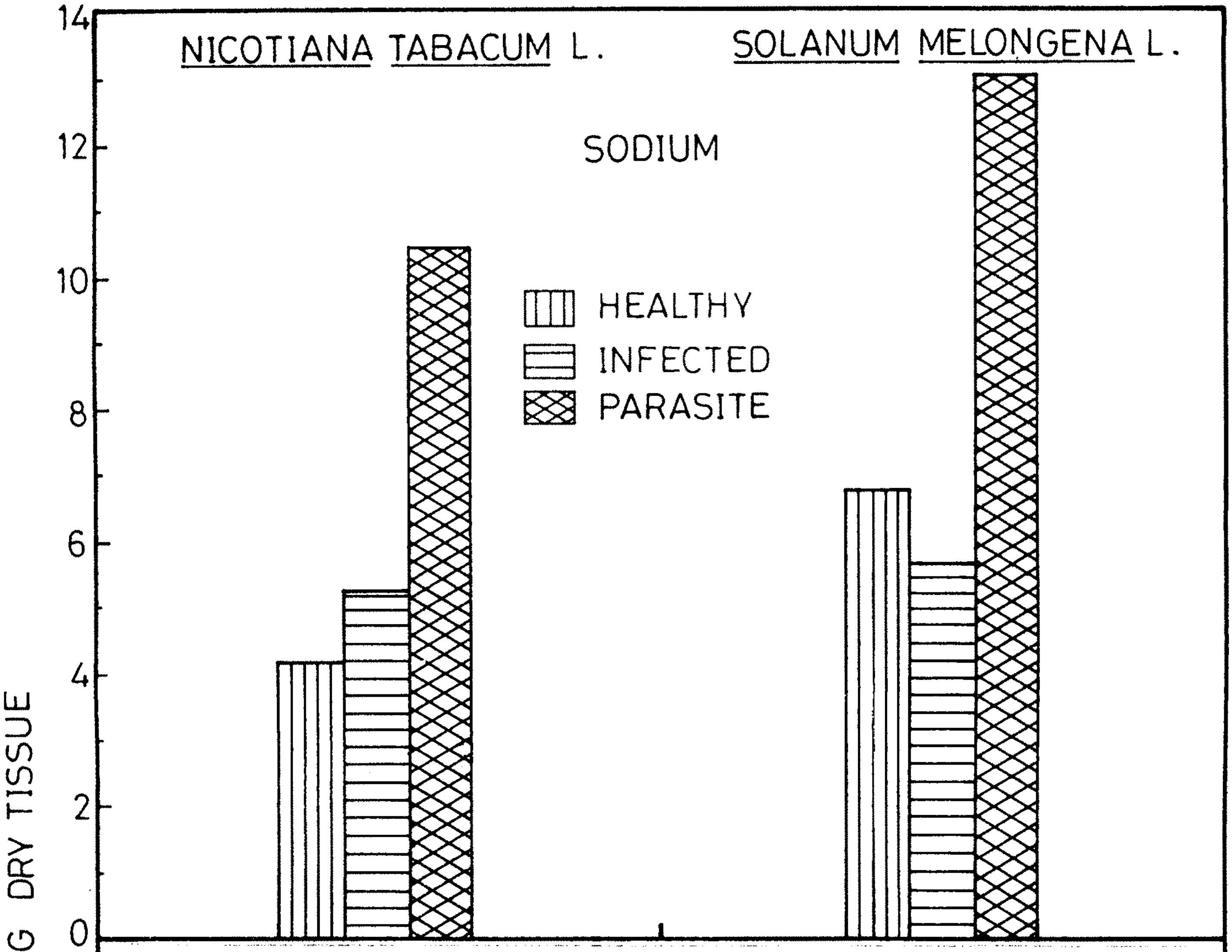


Fig.7 - SODIUM CONTENT OF HEALTHY AND INFECTED (BY *OROBANCHE* SP.) LEAVES OF *NICOTIANA TABACUM* L. AND *SOLANUM MELONGENA* L. AND PARASITE.

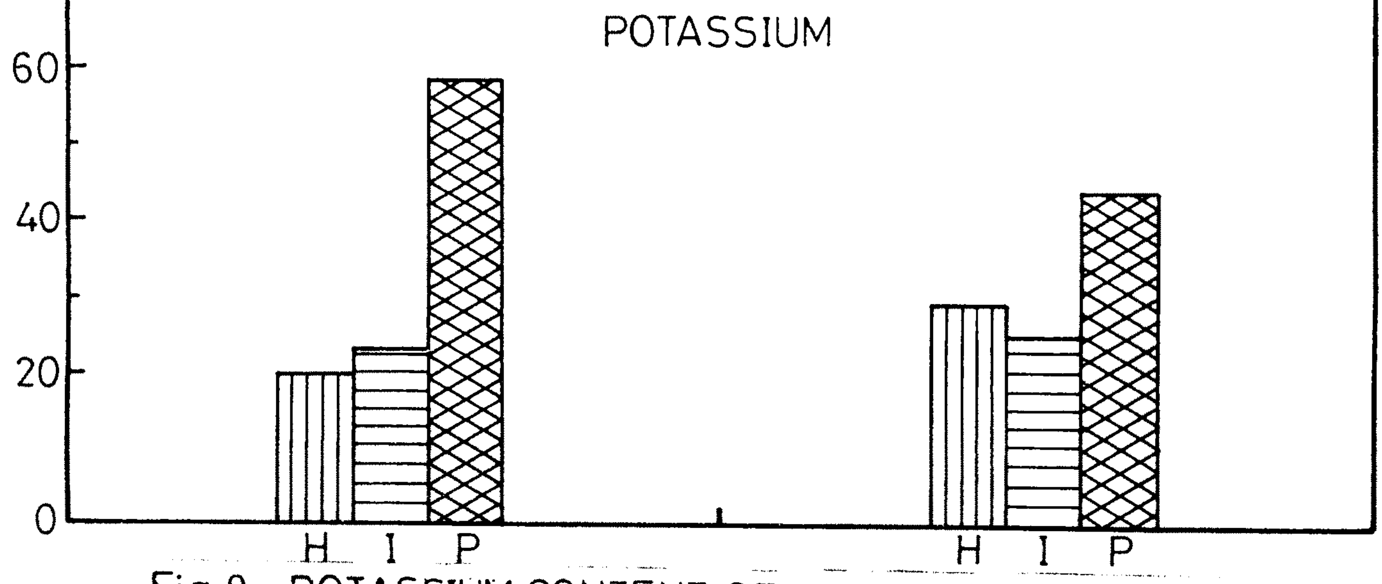


Fig.8 - POTASSIUM CONTENT OF HEALTHY AND INFECTED (BY *OROBANCHE* SP.) LEAVES OF *NICOTIANA TABACUM* L. AND *SOLANUM MELONGENA* L. AND PARASITE.

parasite on the sodium content of the host plant, which has shown an increase as well as decrease in it. Wallace et al. (1978) have reported that sodium is generally excluded from Cuscuta nevadensis regardless to its concentration in the host tissue and Cuscuta contained less sodium than that in the hosts. Agrawal (1982) has reported that sodium content was lower in Cistanche tubulosa (Orobanchaceae) infected host roots (Aerva, Colliganum, Calotropis) and the parasite body as compared to that of Ca^{2+} and Mg^{2+}.

Fungal infection also causes an increase or decrease in the level of sodium content of the host. The increased sodium content was reported by Balasubramanian (1975) in green ear disease affected jowar leaves. Increase in sodium content has also been reported by Gupta (1975) in Coriander infected by stem gall disease, Hegde and Karande (1978) in green ear disease affected bajara and Sankpal and Nimbalkar (1980) in sugarcane infected by smut. On the other hand Sivaprakasam et al. (1974) in brinjal leaves infected by Verticillium and Patil and Kulkarni (1977 b) in sunflower infected by Puccinia, have reported a decrease in sodium content in the infected plants.

According to Balasubramanian (1975) the increase in the level of sodium in the host tissue may be due to the changed metabolism of the host in the presence of pathogen. The increase and decrease in sodium content in Orobanche infected N.tabacum and S.melongena respectively may be due to destructive activities of pathogen or infection might have stimulated or inhibited the absorption of sodium.

ii) Potassium (K^+) :

The potassium content of the leaves of Nicotiana tabacum and Solanum melongena, infected by Orobanche aegyptiaca, a total root parasite and that of the parasite has been recorded in Table No.9 and depicted in Fig.8. It is found that the potassium content is increased in the leaves of infected N.tabacum by about 16%, while it is decreased in the leaves of infected S.melongena by about 14%. The parasites on N.tabacum and S.melongena have shown an increase in potassium content by about 191 and 51% respectively. The parasites of both the plants have shown higher level of potassium than that in the leaves of infected plants.

Potassium is the most important monovalent cation in the tissues with respect to its physiological and biochemical functions. Potassium plays significant role in stomatal opening and closing. It enhances the translocation of photosynthates. It may indirectly promote the synthesis of various organic compounds such as protein, sugars and polysaccharides. It activates several enzymes. Potassium has an important role as an osmotic regulator. It is highly important in raising disease resistance in many crop plants. Potash is important for normal, healthy growth of tobacco and it also produces drought resistance in the leaf.

Potassium content is markedly decreased in *Orobanche cernua* infected tomato (*Lycopersicon esculentum* Mill.), especially, in the leaves as reported by Singh *et al*. (1971). They further reported that *Orobanche* always maintains the potassium content usually higher than that in the hosts. Saghir and Dastgheib (1978) have reported reduced potassium content in beans infected by *Orobanche cernua* but the parasite contained higher level of potassium. Agrawal (1982) has reported that the potassium content is lower in *Cistanche tubulosa* (*Orobanchaceae*) infected host roots (*Aerva*, *Colligonum*, *Calotropis*). Accumulation of potassium was higher at the infection sites, to the levels substantially higher than that in the adjacent host tissue in *Orobanche*, *Cuscuta*, *Viscum* and *Loranthus* (Nicoloff, 1923; Gill and Hawksworth, 1961). Horak (1974) reported that potassium content of *Loranthus europaeus* and *Viscum laxum* leaves was higher than that in the leaves of corresponding host tree. Kumar and Mukherjee (1973) have reported decreased potassium content in the leaves of *Mangifera indica* infected by *Dendrophthoe falcata*, a leafy mistletoe (*Loranthaceae*), but in the parasite leaves there was remarkable accumulation of potassium.

Singh *et al*. (1979) have reported that the concentration of potassium is decreased in *Orobanche aegyptiaca* and *O.cernua* in the absence of host root or when the host root was removed. They further noticed that *Orobanche* spp. show a limited capacity to absorb minerals through their own root system.

Fungal pathogens also influence the potassium content of the host tissue. Sivaprakasam *et al*. (1974), Patil and Kulkarni (1977 b), and Sankpal and Nimbalkar (1980) have reported decreased potassium content in brinjal infected by *Verticillium*, in sunflower affected by *Puccinia*, in smutted sugarcane respectively. Sarkar and Joshi (1977) have also reported a decrease in potassium content in little leaf affected brinjal plant. On the other hand, increased potassium content in the infected tissues has been reported by Balasubramanaian (1975) in sorghum infected by *Sclerospora*, Sivaprakasam *et al*. (1976) & Srinivasan and Chelliah (1979) in *Solanum melongena* infected by little leaf disease, and Ahmad *et al*. (1982) in barley infected with brown rust.

In the present investigation it was noted that potassium content of the leaves of *N.tabacum* infected with *Orobanche* was increased. Sastry and Nariani (1962) and Mohan *et al*. (1978) have suggested that the increase in potassium content may develop greater resistance against the invading pathogen by reducing the rate of multiplication. They are also of the opinion that potassium may increase the resistance through the secondary metabolites like phenols. The parasite of both the plants has shown higher potassium content as compared to that in their host tissues. This indicates that probably there is maximum absorption of potassium by the parasite, *Orobanche*. It has also been stated that both potassium and phosphorus are readily and preferentially transported from healthy to infected tissues and get accumulated there (Yarwood and Jacobson, 1955; Roberts and Jensen, 1970). The present observations in *Orobanche* infected *N.tabacum* and in the parasite of both the plants are also on the similar lines.

iii) *Calcium* (Ca^{2+}) :

The calcium content of the leaves of *Nicotiana tabacum* and *Solanum melongena*, infected by *Orobanche aegyptiaca*, a total root parasite and that of the parasite has been recorded in Table No.9 and depicted in Fig.9. It is evident that the calcium content gets increased in the leaves of infected *N.tabacum* by 27% and *S.melongena* by 49%. In the parasite of *S.melongena* the calcium content is increased by 14%. However, the parasite on *N.tabacum* showed a decrease by 53% in calcium content.

Calcium is an important and essential cation required by the plants. Its optimum value for terrestrial plants is about

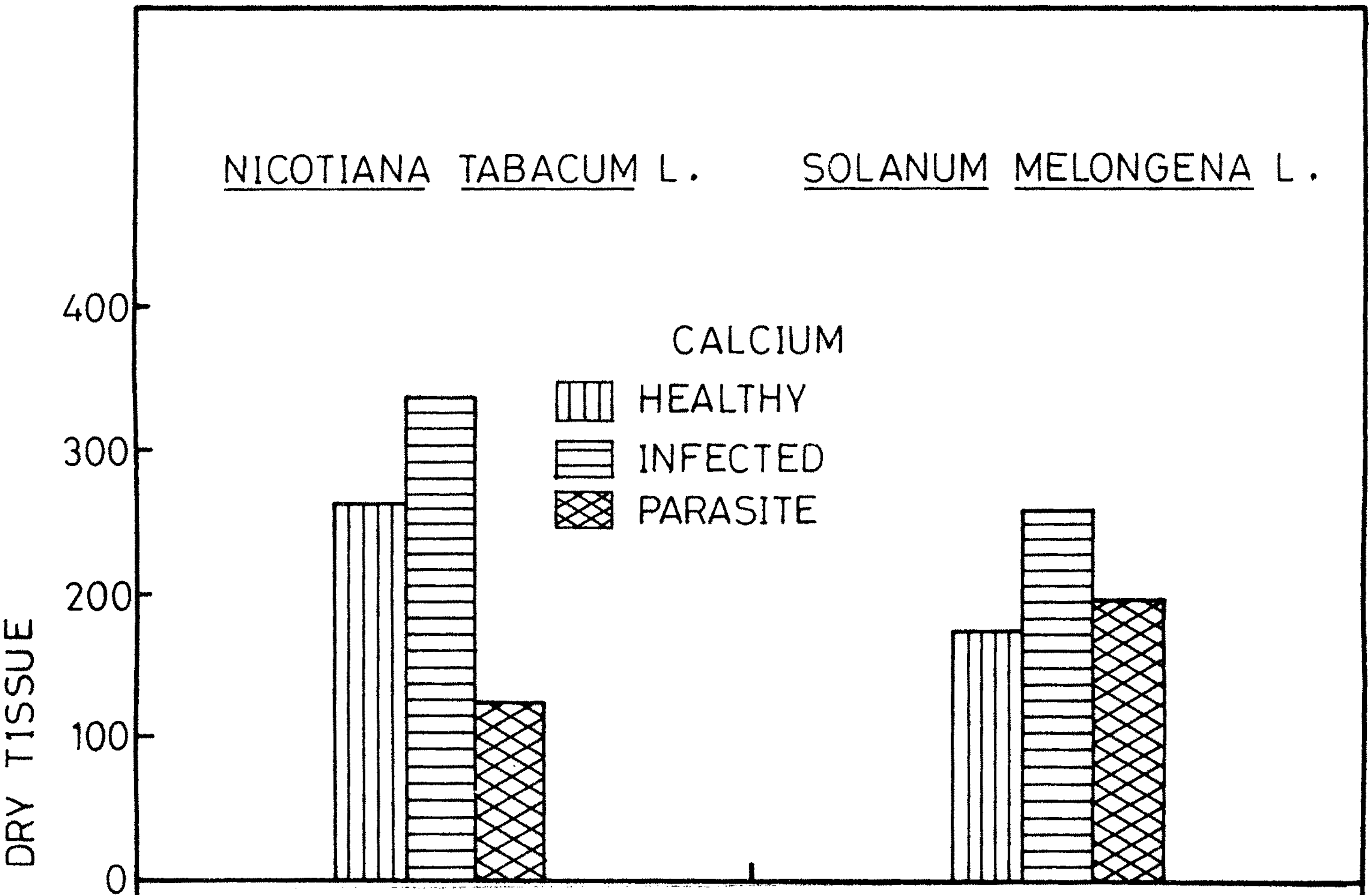


Fig.9 - CALCIUM CONTENT OF HEALTHY AND INFECTED (BY OROBANCHE SP.) LEAVES OF NICOTIANA TABACUM L. AND SOLANUM MELONGENA L. AND PARASITE.

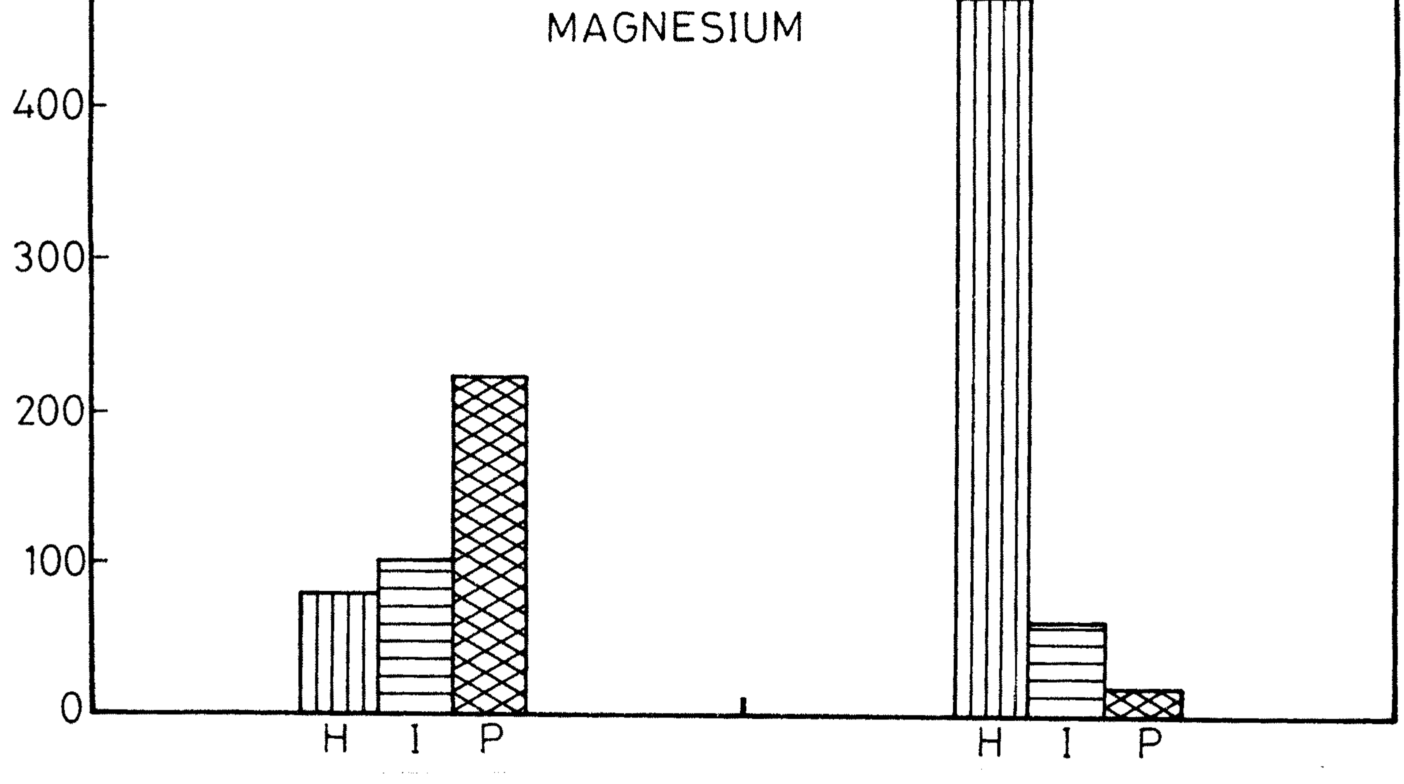


Fig.10 - MAGNESIUM CONTENT OF HEALTHY AND INFECTED (BY OROBANCHE SP.) LEAVES OF NICOTIANA TABACUM L. AND SOLANUM MELONGENA L. AND PARASITE

0.5% or 125 μmol g^{-1} of dry tissue (Epstein, 1972). Calcium is of fundamental importance for membrane permeability and maintenance of cell integrity. It also has a role to play in ion uptake. Calcium is required for cell elongation and cell division (Burstrom, 1968). It also plays a role in retarding senescence. Calcium may also activate some membrane-bround enzymes (Rensing and Corenelius, 1980).

It has been reported that due to *Orobanche* infection there is only a little change in the calcium content of the host plants. Singh *et al*. (1971) have reported increased calcium content in *Orobanche cernua* infected tomato (*Lycopersicon esculentum* Mill.) leaves. However, the trend of variation was not well defined in the stem and roots of infected plant. Saghir and Dastgheib (1978) have reported no change in calcium content in beans infected by *Orobanche cernua*, but the parasite exhibited lower calcium content. Agrawal (1982) has reported increased calcium content in *Aerva*, *Colliganum* and *Calotropis* roots infected with *Cistanche tubulosa* (*Orobanchaceae*). The parasite body also showed higher calcium content as compared to that of sodium. In *Vicia faba* infected with *Cuscuta europea* L., the host-tip records higher calcium content as compared to that of parasite tip (Malik and Komal, unpublished). Horak (1974) have reported that K : Ca ratio in *Loranthus europaeus* and *Viscum laxum* leaves was higher than that in the leaves of corresponding host trees. Singh *et al*. (1979) have reported that by removal of host roots, the concentration of calcium is decreased in *Orobanche aegyptiaca* and *O. cernua* indicating that by the removal of host roots *Orobanche* spp. showed limited capacity to absorb minerals through their own root system.

The diseases caused by fungi also either cause an increase or a decrease in calcium content of the affected plants. Higher concentrations of calcium have been reported by Sivaprakasam *et al*. (1974) in brinjal infected by *Verticillium*, Balasubramanian (1975) in sorghum affected by downy mildew Patil and Kulkarni (1977 b) in sunflower affected by *Puccinia* and Sankpal and Nimbalkar (1980) in smutted sugarcane. On the other hand, low calcium concentration has been reported by Kulkarni and Kulkarni (1978) in mango infected with Capnodium, and Misra and Padhi (1981) in pearlimillet infected with downy mildew. Sarkar and Joshi (1977)have also reported reduced calcium content in brinjal leaves infected by little leaf disease.

The possible involvement of calcium in disease resistance was indicated by Walter (1967), who stated that grey mould disease of tomato may be controlled with sufficient calcium. Kiraly and Gilly (1976) have confirmed the vital role of calcium in disease resistance against fungal infection. Singh and Verma (1974) and Ghorpade and Joshi (1981) have supported this view. In Orobanche infected N.tabacum and S.melongena leaves the increased calcium content may be involved in resistance to parasite, but due to the low rate of translocation of calcium from host to parasite, probably shows the values for calcium content lower than that in infected host leaf tissue.

iv) Magnesium (Mg^{2+}) :

The magnesium content of the leaves of Nicotiana tabacum and Solanum melongena, infected by Orobanche aegyptiaca, a total root parasite and that of the parasite has been recorded in Table No.9 and depicted in Fig.10. It can be seen that the level of magnesium is increased by about 29% in the leaves of infected N.tabacum. The leaves of infected S.melongena, however, have shown a remarkable decrease (87%) in the magnesium content. The parasite of N.tabacum shows an increase (186%) in magnesium content, however, that of S.melongena have shown a decrease (96%) in it.

Magnesium is generally taken up by the plants in lower quantities than those of either Ca^{2+} or K^{+}. The magnesium level in the plant tissue is usually of the order of 0.5% dry weight. It is an essential constituent of chlorophyll and is also associated with many of plant proteins. Magnesium is a cofactor in almost all enzymes activating phosphorylation process, and is therefore, of paramount importance in energy metabolism.

It is observed that the magnesium content increases in the leaves and roots of tomato (*Lycopersicon esculentum* Mill.) infected with *Orobanche cernua* as reported by Singh *et al.*, (1971). The parasite contained smaller amounts of magnesium than that in the host. Wolswinkel (1978) has reported that K^+ and Mg^{2+} were drained by *Cuscuta europea* L. from the host *Vicia faba*, whereas, such draining action by the parasite was not observed in case of calcium. Malik and Komal (unpublished) have found that the concentration of magnesium near the host-parasite contact area was almost doubled to that of *Cuscuta* stem tip. Agrawal (1982) has reported that the magnesium level was much higher in *Aerva*, *Colliganum* and *Calotropis* roots infected with *Cistanche tubulosa* (*Orobanchaceae*) and the parasite also contained higher amounts of magnesium. Singh *et al.* (1979) have reported that the concentration of magnesium decreases in *Orobanche aegyptiaca* and *O. cernua* due to the removal of host roots which indicated that *Orobanche* spp. showed limited capacity to absorb minerals through their own root system.

Fungal diseases also influence the magnesium metabolism of the infected plants. An increase in the Mg^{2+} content has been reported by Balasubramanian (1975) in the leaves of jowar, infected by downy mildew, Hegde and Karande (1978) in the leaves of bajara infected by downy mildew and Sankpal and Nimbalkar (1980) in the leaves of sugarcane infected by smut. While, it was found to be decreased in Ipomea leaves infected by white rust (Misra and Padhi, 1980). Sivaprakasam _et_ _al_., (1976) have also recorded a low magnesium content in the leaves of MLO infected brinjal plant.

In the present study an increase as well as a decrease in magnesium content have been observed in _Orobanche_ infected _N.tabacum_ and _S.melongena_. Goswami _et_ _al_. (1976) stated that the infection inhibits the uptake and translocation of magnesium which results in its low concentration. Singh _et_ _al_. (1971) have reported that _Orobanche_ maintains the calcium and magnesium concentrations lower while those of phosphorus and potassium are kept slightly higher which suggests that the withdrawal of minerals from the host tissue is selective.

According to Huber (1978), the pathogens associated with roots cause destruction of the roots and reduce the absorption of nitrogen, phosphorus, potassium, calcium, magnesium and sodium by reducing the amount of functional absorptive tissue. Based on these observations it is concluded that the decrease in magnesium content in _Orobanche_ infected _S.melongena_ may be due to reduced absorption of magnesium through the infected roots.

v) Phosphorus (P^{5+}) :

The phosphorus content of the leaves of *Nicotiana tabacum* and *Solanum melongena*, infected by *Orobanche aegyptiaca*, a total root parasite and that of the parasite has been recorded in Table No.9 and depicted in Fig.11. It can be seen that the phosphorus content of the leaves of infected *N.tabacum* is increased (2%), while that decreased (34%) in the leaves of infected *S.melongena*. The parasite of both the plants has shown a decrease (18 and 37% respectively in the parasites on *N.tabacum* and *S.melongena*) in the phosphorus content.

The phosphorus content of the parasite on both the plants is lower as compared to that in the host plants.

Phosphorus concentration of the terrestrial plants is 0.2% of dry weight (Epstein, 1972). It is one of the important plant nutrients, an integral of phospholipids in the cell wall and the other cell membranes, sugar phosphates and adenylate compounds like ATP, ADP and AMP. Phosphorus is also a constituent of a variety of organic compounds which are essential for the structure and metabolism of plants. Hall and Baker (1972) have shown that inorganic phosphorus plays a vital role in phloem transport. In tobacco plant, even though the phosphorus requirement is not as high as that of potassium or nitrogen, it is a major element which plays an important role in productivity and hence it is essential ingredient of N, P,

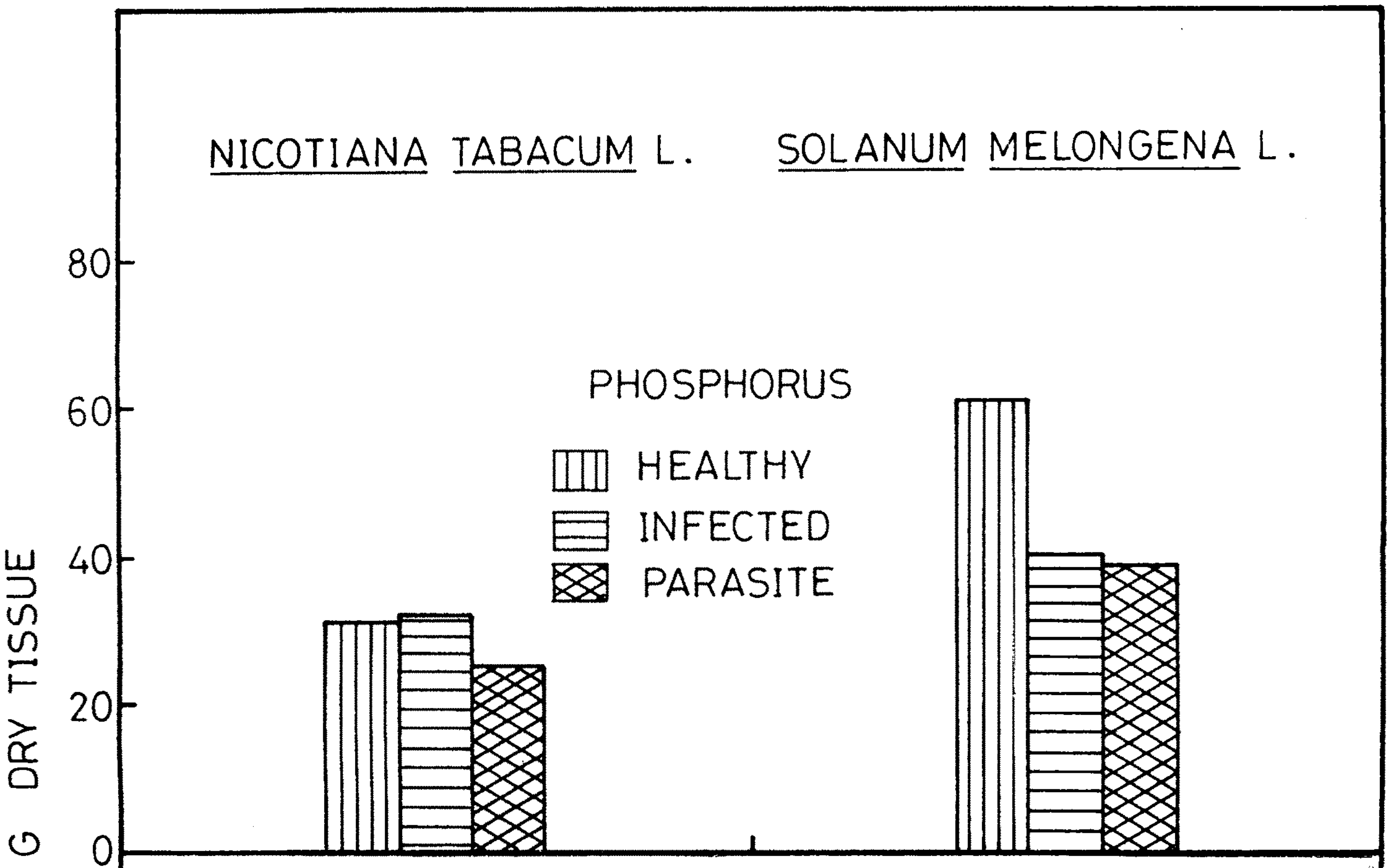


Fig.11- PHOSPHORUS CONTENT OF HEALTHY AND INFECTED (BY *OROBANCHE* SP.) LEAVES OF *NICOTIANA TABACUM* L. AND *SOLANUM MELONGENA* L. AND PARASITE.

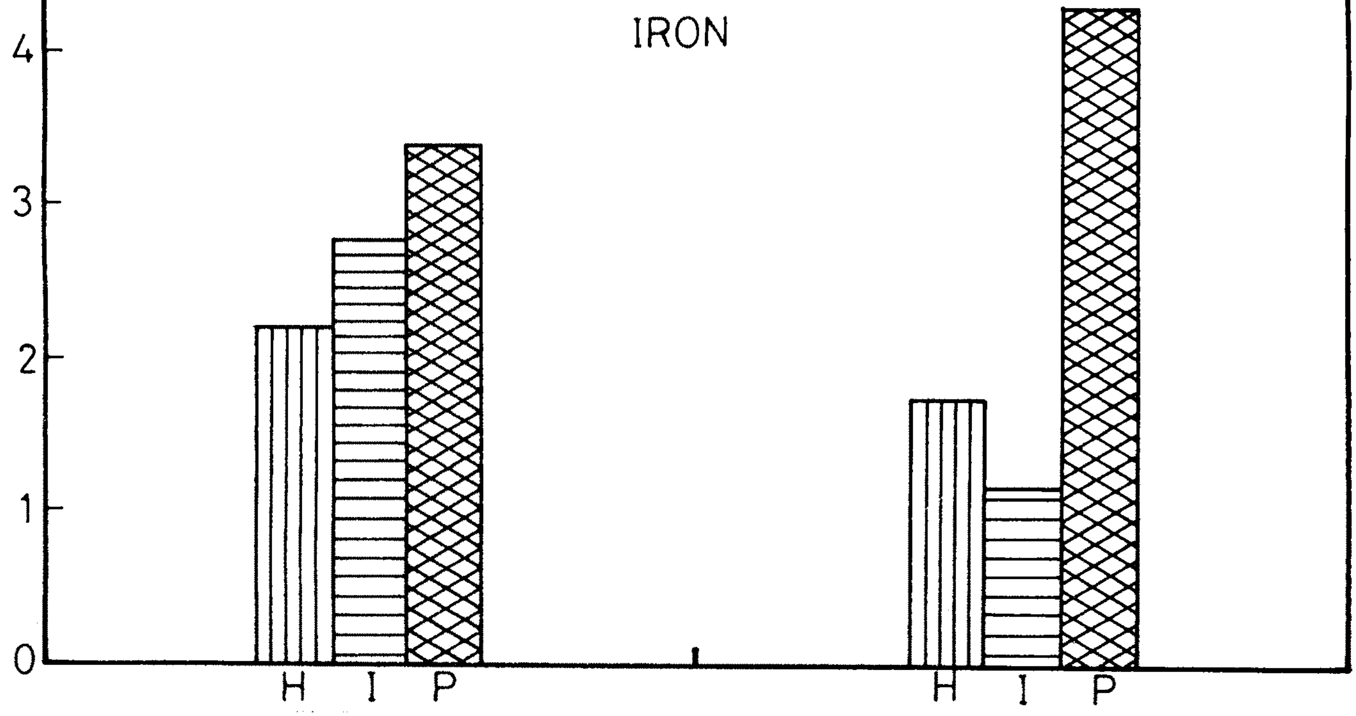


Fig.12- IRON CONTENT OF HEALTHY AND INFECTED (BY *OROBANCHE* SP.) LEAVES OF *NICOTIANA TABACUM* L. AND *SOLANUM MELONGENA* L. AND PARASITE.

K fertilizers. Singh and Singh (1977 a) observed that upto certain level, increasing phosphorus content in the medium enhances the plant growth but heavy accumulation of it results in stunted growth.

Orobanche infection leads to considerable changes in phosphorus content of the host plants. Singh *et al*. (1971) have reported a decrease in phosphorus content in tomato (*Lycopersicon esculentum* Mill.) leaves infected with *Orobanche cernua*. However, in the roots it was found to be increased under infected conditions. *Orobanche* contained greater amount of phosphorus substantiating the fact that the parasite has received more phosphorus. Saghir and Dastgheib (1978) have reported no change in phosphorus content in beans infected by *Orobanche crenata*. However, the comparison between shoots of the host and the parasite, in this case, revealed that the parasite contained higher amount of phosphorus as compared to that in its host. Nicoloff (1923) and Gill and Hawksworth (1961) have reported that inorganic phosphorus accumulates at the infection site to the levels substantially higher than that in the adjacent host tissue, in case of *Orobanche*, *Cuscuta*, *Viscum* and *Loranthus*. Malik and Komal (unpublished) have reported that the concentration of phosphorus was 35% more than that in contact host tip in the parasite tip area in *Vicia faba* infected with *Cuscuta europea*, while the middle portion of the parasite had this level nearly 40% lower than that in the control (Healthy plant). Singh *et al*. (1979) have reported that by the removal of host roots the concentration of phosphorus decreases in *Orobanche aegyptiaca* and *O.cernua* which indicates that *Orobanche* spp. show a limited capacity to absorb minerals through their own root system. Agrawal (1982) has reported that the phosphorus content of the roots of *Aerva*, *Colligamum* and *Calotropis* was much lower than that in the parasite, *Cistanche tubulosa*. Kumar and Mukherjee (1973) have recorded decreased phosphorus level in *Mangifera indica* infected by *Dendrophthoe falcata* (*Loranthaceae*).

Phosphorus content of the host plants also shows a disturbance due to diseases caused by even other pathogens. Sarkar and Joshi (1977)have reported a decrease in phosphorus content in brinjal infected by little leaf disease. On the other hand, increased level of phosphorus was reported in tobacco infected by tobacco mosaic virus (Mitra and Majumdar, 1980).

Similar observations have been reported in case of several fungal diseases. Sivaprakasam *et al*. (1974) in brinjal infected by *Verticillium*, Patil and Kulkarni (1977 b) in sunflower infected by rust and Sankpal and Nimbalkar (1980) in sugarcane infected by smut have reported decreased level of phosphorus due to infection. On the other hand Balasubramanian

(1981) in Sorghum infected by downy mildew and Ahmad et al. (1982) in barley infected by brown rust have reported an accumulation of phosphorus in the diseased plants.

Several reasons have been attributed to the increased level of phosphorus in the diseased plants. Yarwood and Jacobson (1955), Shaw (1963) and Hare (1966) have attributed this increase to the pathogen induced sinks in host tissue, which accumulates greater quantities of phosphorus during pathogenesis. According to Hare (1966) and Johri (1975), the reduced carbohydrate content and accelerated breakdown of starch also cause an increase in phosphorus content. However, in Orobanche infected tobacco leaves, the photosynthetic activities and starch breakdown are probably inhibited or prevented due to infection, but in Orobanche infected brinjal leaves, the low rate of photosynthesis only probably results in decreased phosphorus content.

vi) Iron (Fe^{3+}) :

Iron content of the leaves of Nicotiana tabacum and Solanum melongena infected with Orobanche aegyptiaca, a total root parasite and that of the parasite has been recorded in Table No.9 and depicted in Fig.12. It can be seen that the leaves of infected N.tabacum show an increase (25%) in the iron content. However, it is decreased (33%) in the leaves of infected *S.melongena*. The parasite on both the plants has also shown an increase (54% of healthy in case of parasite on *N.tabacum* while, that 137% in that of *S.melongena*).

Iron occurs in prosthetic group of certain proteins, notably the cytochromes, which function in electron transport and the enzymes, peroxidase and dehydrogenases. It has a specific role in chlorophyll synthesis. Ferredoxin is an iron--containing protein which acts as an electron carrier in photo--synthetic phosphorylation and in nitrogen fixation. Haemoglobin, another haemprotein, is found in nitrogen fixing root nodules. Iron functions both as a structural component and also as a cofactor for enzymatic reactions. Oxidation reduction reactions are most commonly associated with Fe- containing systems.

It can be seen that there is no marked change in the iron content of *Orobanche* infected host plants (Fig.12). The parasite has shown the maximum amount of iron. Malik and Komal (unpublished) have reported that in *Vicia faba* infected with *Cuscuta europea*, the host tip has maximum iron content. Wallace *et al*. (1978) have reported that *Cuscuta nevadensis* contains lower quantity of iron as compared to that in the leaves of host plants. Kumar and Mukherjee (1973) have also reported a decrease in iron content in *Mangifera indica* infected with *Dendrophthoe falcata*.

The increase in iron content was reported by Patil and Kulkarni (1977 a) in sunflower affected by *Puccinia*, and by Kapur *et al*. (1978) in date palm affected by smut which indicated the fungal pathogens also cause to increase the iron content of the tissue. On the other hand, Sankpal and Nimbalkar (1980) in sugarcane affected by smut and Balasubramnian (1981) in jowar infected with downy mildew, reported a decrease in iron content. Sarkar and Joshi (1977) have also made similar observations in little leaf infected brinjal.

The concentration of active iron plays an important role in determining the growth and concentration of pathogen in host tissue, which governs the degree of disease resistance (Sastry, 1964; Hegde, 1967). It was noted that the disease resistance decreases with increasing the iron content of the tissue. Hegde and Karande (1978) have supported this view. In the present investigation this has been observed only in *S.melongena* which may be due to the ability of host plant to resist the activity of parasite infection.

vii) *Zinc* (Zn^{2+}) :

The zinc content of the leaves of *Nicotiana tabacum* and *Solanum melongena* infected by *Orobanche aegyptiaca*, a total root parasite, and that of the parasite has been recorded in Table No.9 and depicted in Fig.13. It is clear that the zinc content is increased (9%) in the leaves of infected *N.tabacum*

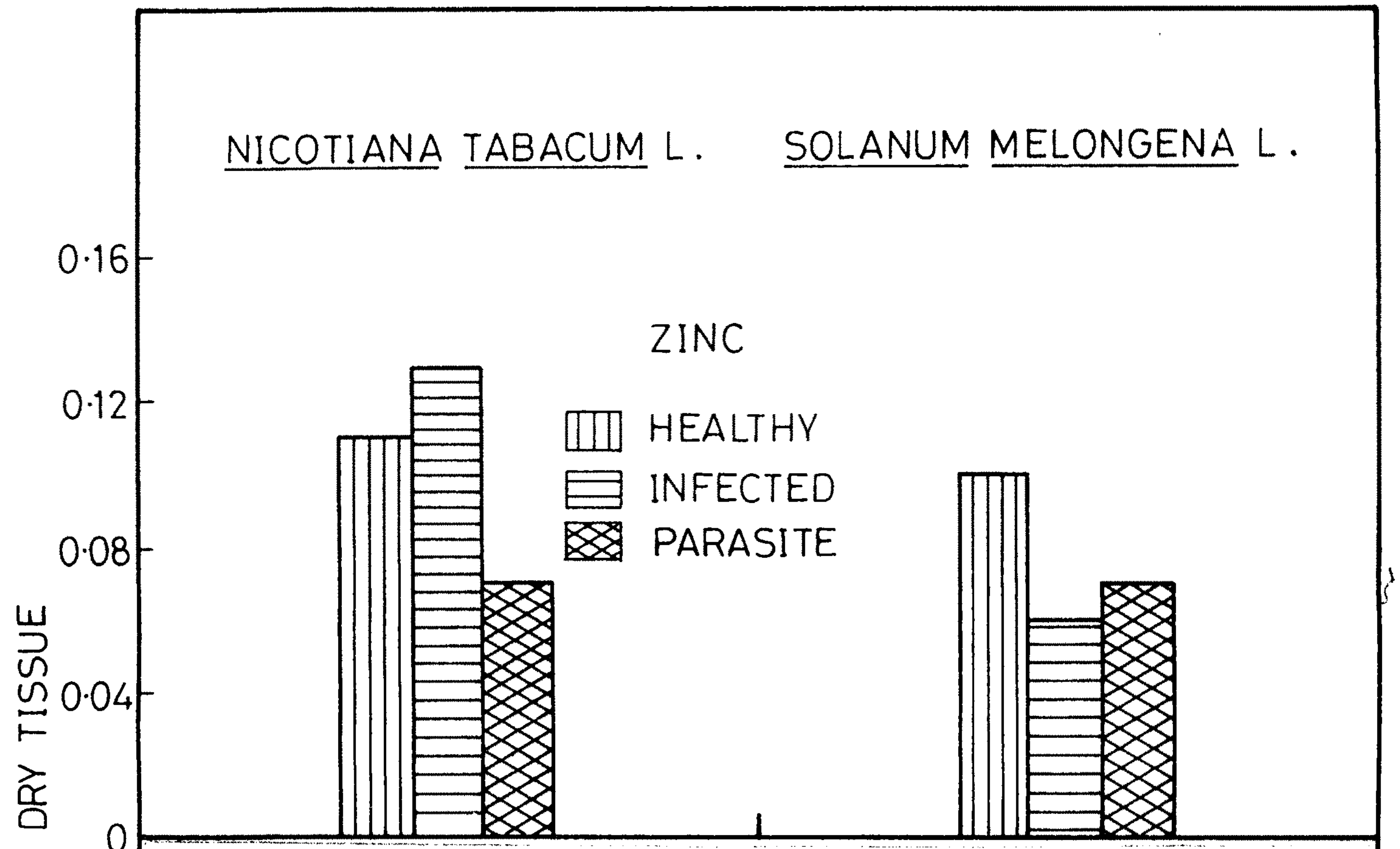


Fig.13 - ZINC CONTENT OF HEALTHY AND INFECTED (BY OROBANCHE SP.) LEAVES OF NICOTIANA TABACUM L. AND SOLANUM MELONGENA L. AND PARASITE.

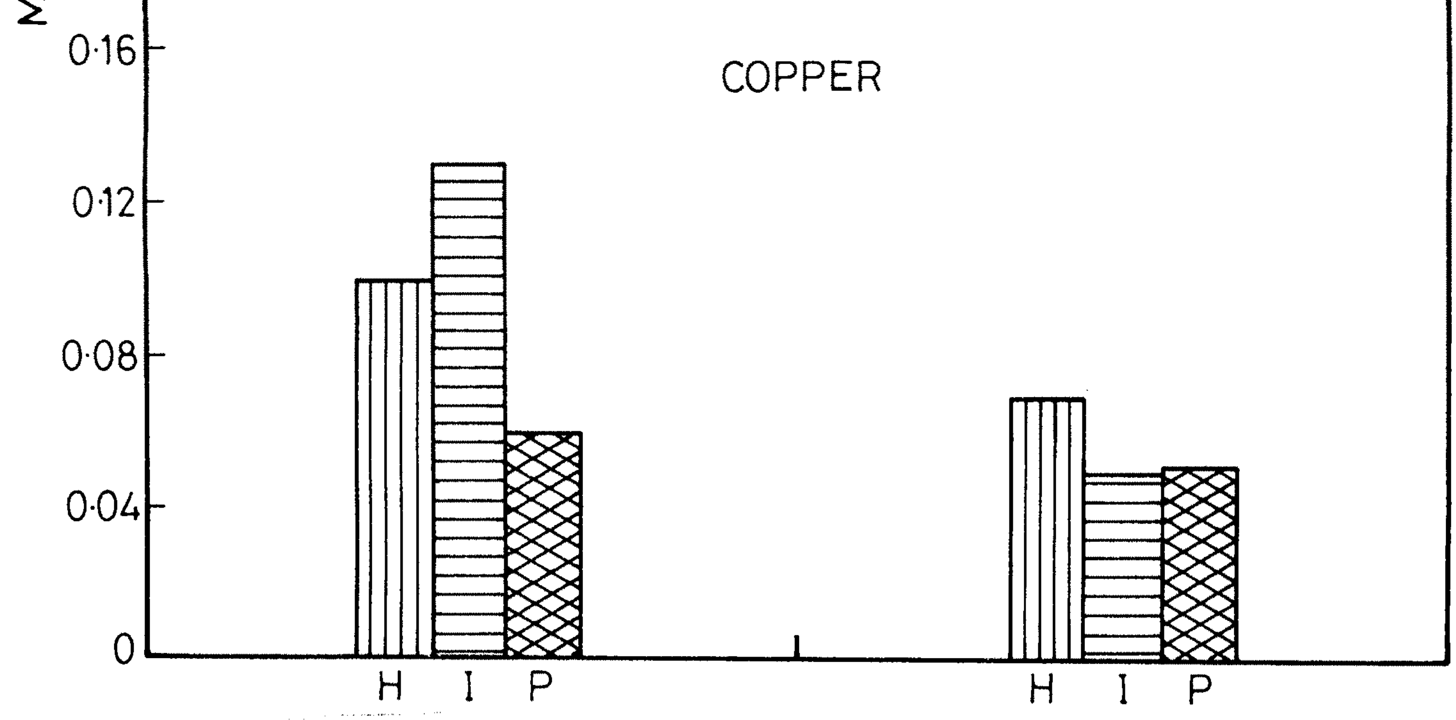


Fig.14 - COPPER CONTENT OF HEALTHY AND INFECTED (BY OROBANCHE SP.) LEAVES OF NICOTIANA TABACUM L. AND SOLANUM MELONGENA L. AND PARASITE.

and that decreased (40%) in the leaves of infected S.melongena. The parasite of both the plants has shown the values for zinc lower (36%, 30% respectively in parasites on N.tabacum and S.melongena) than those for the infected host plants.

Zinc is a micronutrient. Its optimum concentration for terrestrial plants is 0.03 μ mol g^{-1} or 20 ppm (dry wt basis, Epstein, 1972). It is needed for protein metabolism and appears to be involved some how in the production of chlorophylls. This micronutrient is associated most commonly with auxin, probably it prevents the oxidation of such growth hormones. Zinc appears to be necessary for synthesis of tryptophan, precursor of IAA (Tsui, 1948) and through this compound auxin levels are affected. Zinc plays a role in several enzyme systems such as dehydrogenases, phosphodiestarases, carbonic unhydrase etc. It also promotes the synthesis of cytochrome C and stabilizes the ribosomal fractions. This divalent cation is closely involved in nitrogen metabolism in plants. Zinc, along with Cu, has been shown to be the constituent of enzyme superoxide dismutase.

Malik and Komal (unpublished) have reported that the zinc content of Vicia faba infected with Cuscuta europea L. is nearly equal to that in the parasite tip while that parasite stem tip has the concentration of this nutrient nearly equal to that of host.

In sandle infected with spike disease, the zinc content was found to be decreased in the leaves, stem and roots of the host (Raychaudhari and Varma, 1980). Balasubramanian (1981) in chlorotic leaves of sorghum infected by downy mildew have reported an increase in the zinc content. Loneragan et al. (1979) and Orabi et al. (1981) have stated that in corn plants (Zea mays L.) the application of phosphorus causes an increase in zinc content. In the present investigation we have also noted that the phosphorus content of the leaves of N.tabacum infected by Orobanche is increased which may be one of the reasons for greater accumulation of zinc in the affected tobacco leaves. Wen (1983) has reported that when Cu, Zn and Cd were taken by the tobacco plant they get distributed throughout the plant, but in the roots the accumulation is more and the contents of total nitrogen, total alkaloids and the crude ashes in the leaves increase in proportion of Zn, Cu and Cd applied to the plant. But, the amount of phosphorus and that of reducing sugars was found to be increased.

Zinc plays an important role in disease resistance as reported by Singh et al. (1970), Deshmukh and Mayee (1978) and Misra et al. (1981). They have noted that the application of zinc causes to decrease the concentration of viral and fungal pathogens and to increase disease resistance in the host by lowering the disease incidence, while Wutscher and Hardesty (1979) and Shevchenka et al. (1980) are of the opinion that the trace element like zinc increases the disease resistance by increasing the phenolic contents which thereby checks the growth of invaded pathogen. However, the present observations indicated that the increased zinc content in the leaves does not produce such a significant effect.

viii) *Copper* (Cu^{2+})

The copper content of the leaves of *Nicotiana tabacum* and *Solanum melongena* infected by *Orobanche aegyptiaca*, a total root parasite, and that of the parasite has been recorded in Table No.9 and depicted in Fig.14. It is evident that the copper content of the leaves of infected *N.tabacum* is increased (30%) while that of the leaves of infected *S.melongena* is decreased (17%). The parasite on both the plants, has shown a decrease (40 and 17% respectively in *N.tabacum* and *S.melongena*) in the copper content. In other words, the parasite of both the plants has shown lower amount of copper as compared to that in the host plants.

Copper occurs in the soil almost exclusively in divalent form. The copper content of most of the plants is generally between 2-200 ppm dry wt. Copper is a constituent of chloroplast protein, plastocyanin, which forms the part of electron transport linking the two photochemical systems of photosynthesis, (Bishop, 1966; Boardman, 1975.). Copper appears to participate in both protein and carbohydrate metabolism. Copper is a cofactor in enzyme synthesis as well as DNA and RNA synthesis. Copper indirectly influences nodule formation also. In tobacco plant, as copper is required only in traces shortage does not usually occur. With a deficiency of the element, the younger leaves suddenly loose their turger and show permanent wilting (McMurtrey, 1941). On the other hand tobacco is adversely affected by the toxic action when too big a dose of copper is applied as a fungicide.

Malik and Komal (unpublished) have reported in Vicia faba infected by Cuscuta europea L. that the copper content of the host tip is increased, but that in the parasite tip was kept low.

Several reports indicate that the fungal diseases cause an increase in copper content of the host plant. An increase in copper content in the leaves of cobbage infected with Plasmodiophora and in the leaves of Sorghum infected with downy mildew was reported by Betz et al. (1980) and Balasubramanian (1981) respectively. According to Sastry (1963) increased copper content in the diseased plant favours the growth and development of pathogen. Increased copper content in the leaves of infected N.tabacum may favour the growth and development of Orobanche but in S.melongena the decreased copper content resulting in low degree of parasitism. The parasite contained low copper content which may be due to low draining of copper from host by the parasite.

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