RESULTS AND DISCUSSION

Almost all biochemical and metabolic processes of plants are determined by the age of the leaf (Atkinson et al, 1967; Patil and Hegde, 1983). There is decline in nucleic acids, proteins, clorophylls, sugars (Thimann et al, 1974, Tetley and Thimann 1974) and polyphenols (Judel, 1972) in aging leaves. It is therefore interesting to study the effect of leaf age on certain parameters.

A. Leaf Plastochron Index (LPI) :

Prior to harvesting the leaf material for the experimental purpose, the leaf plastochron index (LPI) which determines the age of the leaf (Erikson and Michelini 1957) was analysed in Soybean (Glycine max) variety JS-335 and MACS-13. The leaf plastochron index analysed is depicted in Table-1. It is vividly clear from the Table that the variety MACS-13 exhibit more LPI than JS-335. This clearly indicates MACS-13 has a capability to grow fast than that of JS-335.

B. Chlorophylls :

The chlorophyll play a major role in light reactions of photosynthesis and hence the chlorophyll content as well as the state of these pigments have direct bearing on the photosynthetic efficiency of the plant. Photosynthesis stops when the amount of chlorophyll in active complexes in vivo drops below the minimum level.

The changes in chlorophyll content in the leaves of two different varieties of soybean during different growth stages are given in Table-2. It can be seen from the Table that chlorophyll content goes on increasing upto flowering stage which is refered in the Table as medium mature stage with 20.41 and 47.17 LPI for JS-335 and MACS-

Table : 1 Leaf Plastochron index of Soybean (Glycine max) var. JS-335 and MACS-13

Leaf Stage	Leaf number from the base	Leaf Plastochron Index LPI
JS-335		
Young	14 - 18	13.66 ± 2.3
Medium Mature	5 - 13	20.41 ± 1.9
Mature	1 - 4	26.91 ± 1.5
MACS-13		
Young	26 - 30	34.64 ± 2.7
Medium Mature	6 - 25	47.14 ± 3.1
Mature	1 - 5	59.64 ± 2.25

Table : 2 Total chlorophylls and carotenoids content in the leaves of Soybean (Glycine max) var. JS-335 and MACS-13

Leaf Material	mg 100 $^{-1}$ fresh tissue	
	Total Chlorophylls	Carotenoids
JS-335		
Young	83.4 ± 5.1	37.53 ± 3.33
Medium Mature	266.8 ± 4.5	146.3 ± 5.4
Mature	100.2 ± 6.5	55.11 ± 5.0
MACS-13		
Young	155.8 ± 7.5	77.5 ± 4.6
Medium Mature	354.6 ± 5.0	185.3 ± 5.2
Mature	165.2 ± 5.0	110.5 ± 3.5

13 respectively and thereafter there is significant decrease at mature stage. The total chlorophyll content was appeared to be more in MACS-13 at all the stage of plant age or plastochron index, than that of JS-335.

Like that of chlorophyll, process of photosynthesis is also dependent upon carotenoid content in leaf tissue. In addition to light harvesting role, carotenoids protects chlorophylls by receiving the potentialy harmful quanta from chlorophyll triplets (Mayfield et al 1986) and in this way they are destroyed (Goodwin, 1976). The carotenoid also absorb radient energy as accessary pigments, protect chlorophyll molecules against lethal oxidation and photobleaching (Mayfield et al, 1986) and help in maintaining the confirmation of the pigment protein complexes (Heldix et al 1982). The carotenoid level studied in two different varieties of soybean viz. JS-335 and MACS-13 with respect to age depicted in Table-2, are indicative of the fact that their synthesis is more at medium mature stage than that of rest of the stages. Thus the data recorded in Table-2 for chlorophyll and carotenoids clearly suggest that both these pigments gradually increase in their level upto medium mature stage and then declines as the on set of maturity.

C. Nitrogen and proteins :

Nitrogen play an important role in crop plants for it is linked with the productivity. All the breeding programmes of the recent years are oriented to the nitrogen geared proctivity. The variation in nitrogen and protein content in the leaves of soybean varieties JS-335 and MACS-13 at different growth stages are given in Table-3. The leaves of medium mature stage exhibited high nitrogen and obviously protein content

Table : 3 Nitrogen and protein content in the leaves of Soybean (Glycine max) var. JS-335 and MACS-13

Leaf Material	g 100 $^{-1}$ g fresh tissue	
	Nitrogen	Protein
JS-335		
Young	0.75 ± 0.25	4.28
Medium Mature	1.85 ± 0.05	10.56
Mature	1.15 ± 0.05	6.57
MACS-13		
Young	0.9 ± 0.1	5.14
Medium Mature	2.48 ± 0.11	14.16
Mature	2.1 ± 0.15	12.0

than any other stage. Among the two varieties the higher nitrogen content was observed in MACS-13 and in both the varieties the nitrogen content decreased with increase in plant age. The reduction in nitrogen level after medium mature stage or beyond 20 LPI is quite natural in view of metabolic role of leaves because at seed filling stage one can notice a decline in nitrogen level in the leaves which can be attributed to the translocation of nitrogen to the developing seeds. The decrease in total nitrogen along with increase in leaf age and/or plant age is reported by several workers (Rao et al 1983; Insun et al 1987). According to Woolhouse (1967) senescence causes decline in the content of nitrogen, protein, nucleic acid and other macromolecules. The reduction in protein nitrogen content is probably due to the accelerated proteolytic activity which may result in compete breakdown and/or alteration of some proteins. Williams (1979) observed that the nitrogen accumulation in the leaves was maximum in 40 days old groundnut. Moreover, the nitrogen accumulation by most of the crop is dependent upon nitrogen supply and on other environmental factors and plant genotype (Tinker, 1979). In the present investigation the nitrogen accumulation gradually increases with the plant age upto flower initiation (20 LPI onwards) and thereafter declines which possibly be used for grain filling.

D. Polyphenols :

Secondary metabolites of plants do not exhibit a direct role in primary metabolism. However, secondary products of many chemical classes present in plant are believed to function during pathogenic attack or any type of stress (Harborne, 1986). Polyphenols represent a seconday product of plant metabolism, accumulate

in all infected plants (Srinivasan, 1983). Several reasons have been ascribed for accumulation of polyphenols in diseased plants, the major one is a defance mechanism adopted by the host plant, (Sasikumar et al, 1979). The polyphenol content studied in leaf, stem and root of soybean at different growth stages is recorded in Table-4 and represented histographically in Fig. 1 and 2. It is very clear from the Table and Figure that the maximum accumulation of polyphenol is found in root of both the cultivars JS-335 and MACS-13. The polyphenol level is found to be more at vegetative stage in MACS-13 whereas it is less in JS-335. In case of stem the level of polyphenol has no consistancy with respect to growth stages. The overall picture clearly indicate that there is large fluctuation in the polyphenol level in leaf, stem and root at all the stages of growth. The general trend observed in most of the plants reveals that the polyphenol level gradually increase from young to mature stage of growth and then decline (Hillis and Swain, 1959, Judel 1972). From the present data of polyphenol content, it can be surmised that the susceptibility of both the varieties to rust infection can be atributed to the fluctuating level of polyphenols in the plant at different growth stages. Hence it is very much essential to concentrate upon studying the threshold level of polyphnol and age of plant to device a time shedule for spraying better suited agrochemical to overcome the disease infection or biotic stress.

Looking to the polyphenol level in the leaf tissue it appears that the stage between vegetative and flower initiation in case of JS-335 and flower initiation stage of MACS-13 may be ideal for employing foliar applicaion of agrochemicals which will help the plant to develop disease resistance.

Table : 4 **Polyphenol content in leaf, stem and root of Soybean (Glycine max) var. JS-335 and MACS-13 at different growth stages.**

Soybean Growth Stages	POLYPHENOL CONTENT (g 100^{-1} g dry tissue)		
	Leaf	**Stem**	**Root**
JS-335			
Vegetative	0.04	0.36	1.42
Flower initiation	0.40	0.26	1.48
Pod formation	0.57	0.53	0.063
Mature	0.75	0.50	1.39
MACS-13			
Vegetative	0.87	0.48	2.40
Flower initiation	0.34	0.61	2.25
Pod formation	0.67	0.37	1.20
Mature	0.59	0.57	1.32

The values are mean of three determinations.

Figure 1
Histogram showing level of Polyphenol contents in Soybean (Glycine max) var. MACS-13 at different growth stages.

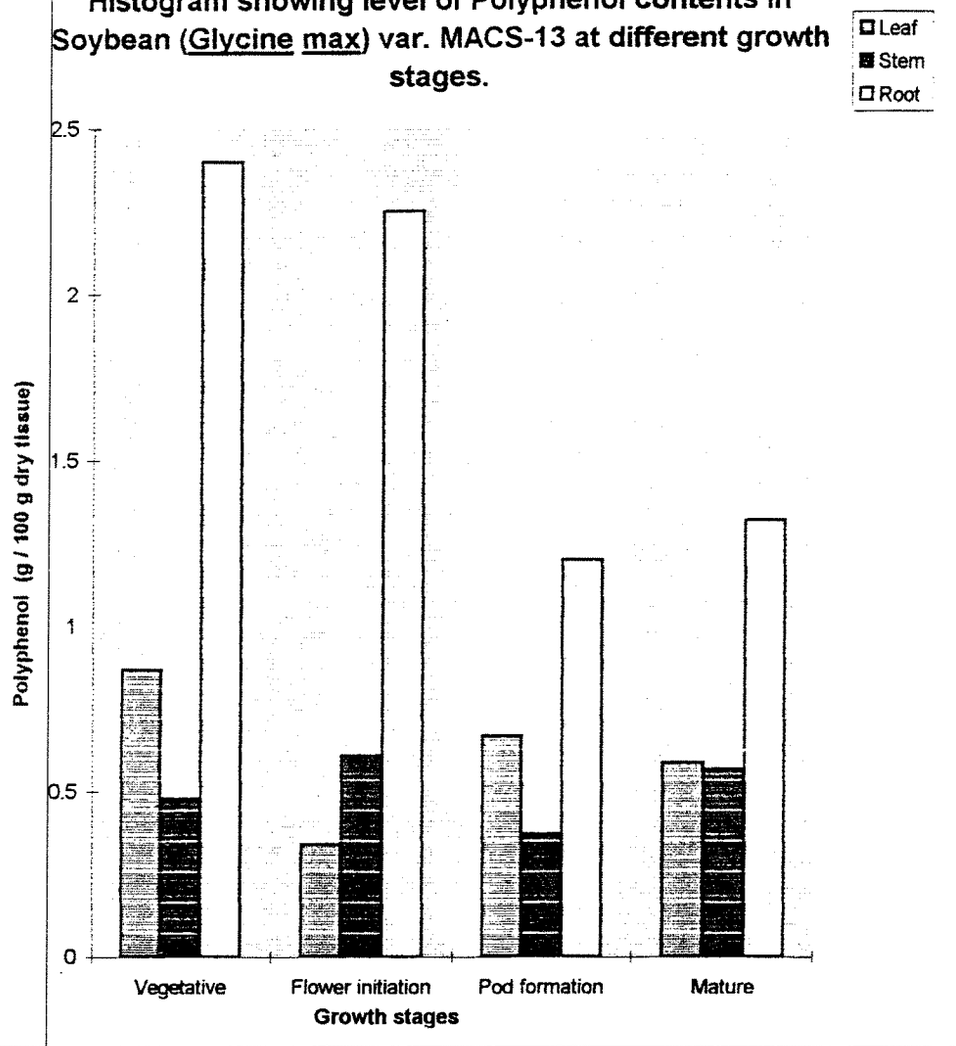
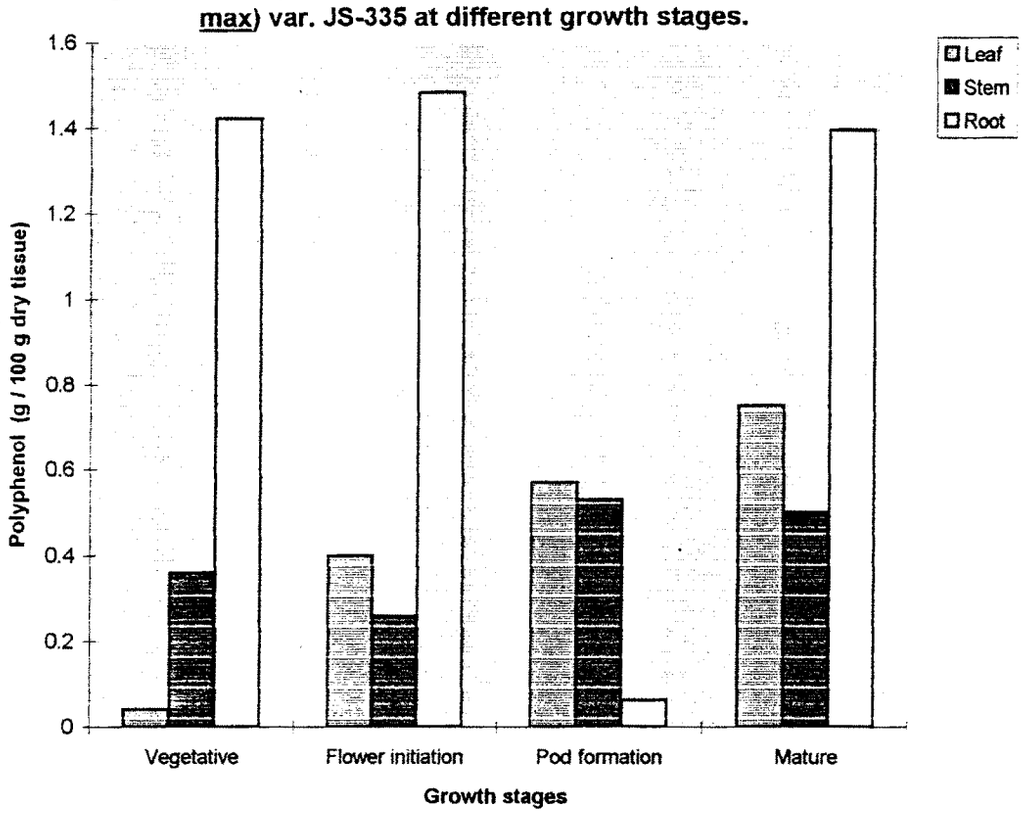


Figure 2

Histogram showing level of Polyphenol content in Soybean (Glycine max) var. JS-335 at different growth stages.



E. Polyphnol oxidase :

The phenol oxidase system play an important role in respiration by transfering electrons from respiratary substrates to other hydrogen or electron acceptors. Quinone is the oxidation product of phenol which may be reduced to their original phenol-form by respiratory carriers. The increase in polyphenol oxidase activity in diseased plant or under biotic stress is generally accompanied the increased concentration of phenolic substances. This enzyme has different substrate specificities which vary with plant species, plant organ and ontogenic state of plant (1970). The major role ascribed to polyphenol oxidase present in plant tissue is the elaboration of polymerised protective dark brown melanin product which could resist infection from an injured or cut surface of a plant tissue (Adamson and Abigor, 1980).

With this view in mind the activity of an enzyme polyphenol oxidase was scored in the leaves of soybean varieties JS-335 and MACS-13 with respect to plastochron age and or growth stages. The data is depicted in Table-5 and in Fig. 3. It is very clear from the Table and Figure that the enzyme activity in both the varities is slightly decreased at medium mature stage of growth (20, 41 LPI for JS-335 and 47.14 LPI for MACS-13). However, the said activity is at higher ebb at young and mature stage when expressed on chlorophyll basis, while on protein basis the activity went on increasing in MACS-13. This clearly indicates that the activity changes with ontogenic stage as stated earlier.

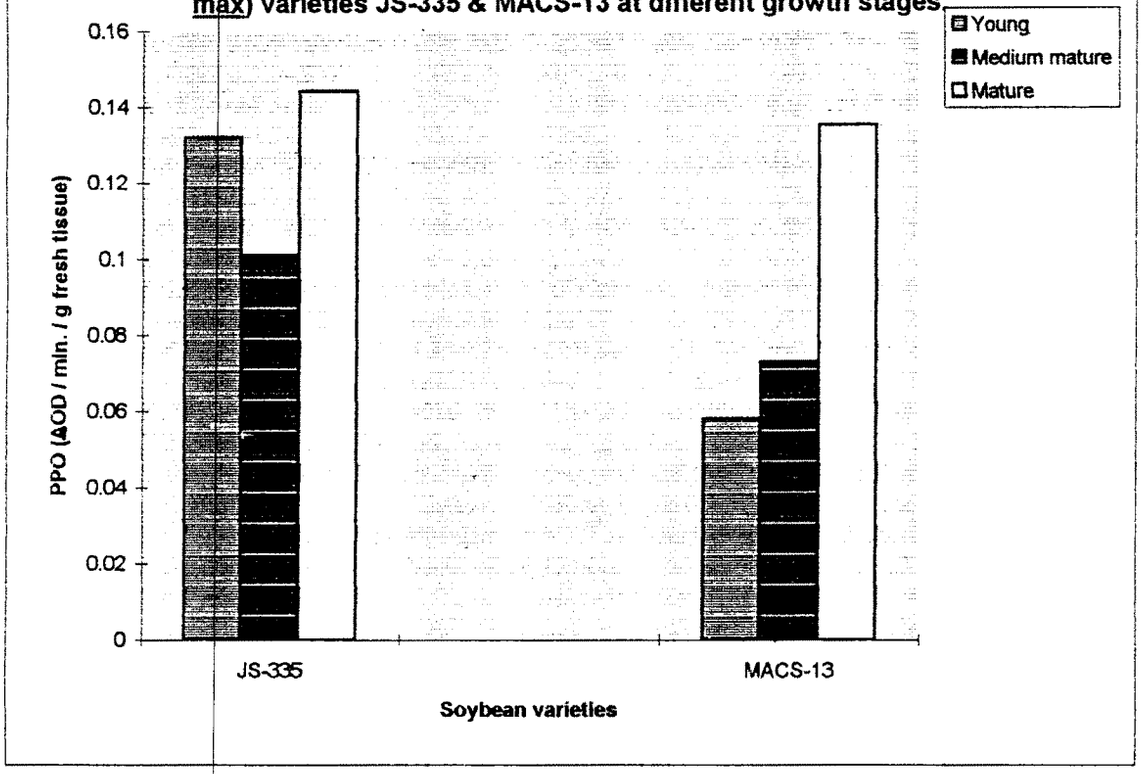
Table : 5 Polyphenol oxidase activity in the leaves of Soybean (Glycine max) var. JS-335 and MACS-13 at different growth stages.

Soybean Growth Stages	POLYPHENOL OXIDASE		
	Δ OD min^{-1} g^{-1} fresh tissue	Δ OD min^{-1} mg^{-1} chlorophyll	Δ OD min^{-1} mg^{-1} protens
JS-335			
Young	0.132	0.791	0.070
Medium mature	0.101	0.570	0.048
Mature	0.144	0.899	0.059
MACS-13			
Young	0.058	0.690	0.026
Medium mature	0.073	0.547	0.037
Mature	0.135	1.686	0.082

The values are mean of three determinations.

Figure 3

Histogram showing PPO activity in the leaves of Soybean (Glycine max) varieties JS-335 & MACS-13 at different growth stages.



E. Phenylalanine ammonia lyase (PAL) :

This enzyme catalyses the deamination of phenylalanine to trans-cinnamic acid. Thus formed cinnamic acid can then be metabolised to form variety of compounds such as flavonoids, coumarins and lignins. According to Iredale and Smith (1974) the phenylalanine ammonia lyase activity may have a regulatary role in phenol, coumarin and flavonoid biosynthesis. In disease resistance mechanism, this enzyme is increasingly reccognized to play an important role in the conversion of phenylalanine and tyrosine to coumaric acids. These provide the phenyl propane carbon skeleton for the synthesis of flavonoids, phenolic phenyl propanes and lignin which are important in disease resistance. Resistant varieties are apparently characteristic of rapid conversion of phenylalanine and tyrosine to coumaric acids. The significance of PAL in phytoalexin synthesis has also been stressed in recent reviews (Mahadevan, 1979).

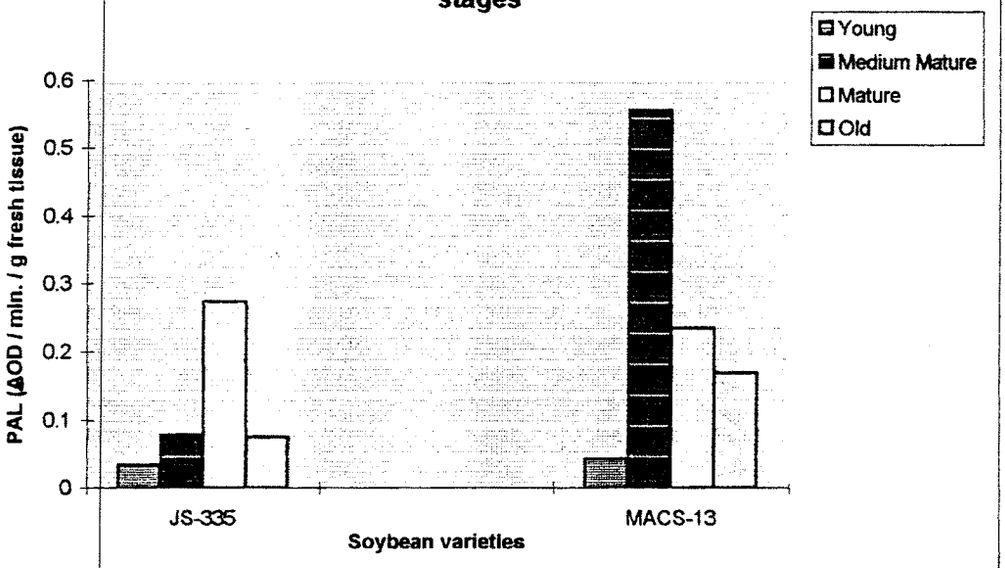
In order to know the fate of PAL which is mainly involved in polyphenol biosynthesis, its activity has been studied in soybean varieties JS-335 and MACS-13 with respect to growth stages or plastochron age. The data is represented in Table-6 and Fig. 4. It is vividly clear from the Table and Figure that the PAL activity is more at medium mature and mature growth stage as compared to young and old stage of growth. This led us to conclude that the plant age or the leaf plastochron largely contribute towards the enzyme activity. The increased activity of PAL may help in triggering flavonoid biosynthesis in soybean. The stimulation of this enzyme at young to medium mature growth stages can be acheived by the foliar application

Table : 6 Phenylalanine ammonia lyase activity in the leaves of Soybean (Glycine max) var. JS-335 and MACS-13 at different growth stages.

Soybean Growth Stages	Phenylalanine ammonia lyase		
	Δ OD min^{-1} g^{-1} fresh tissue	Δ OD min^{-1} mg^{-1} chlorophyll	Δ OD min^{-1} mg^{-1} protens
JS-335			
Young	0.0336	0.295	0.0205
Medium mature	0.0781	0.532	0.0236
Mature	0.273	2.338	0.118
Old	0.0754	1.107	0.0377
MACS-13			
Young	0.042	0.757	0.0087
Medium mature	0.554	5.395	0.0846
Mature	0.234	2.158	0.0405
Old	0.168	2.289	0.0291

The values are mean of three determinations.

Figure 4
Histogram showing PAL activity in the leaves of soybean (Glycine max) varieties GS-335 and MACS-13 at different growth stages



of suitable agrochemical which may possibly help in flavonoid biosynthesis and which will make the plant resistant to biotic stress even at early growth stage.

F. IAA oxidase :

The accumulation or destruction of auxin is frequently associated with IAA oxidase, an enzyme which destroys the auxin. Inhibition of IAA oxidase leads to the accumulation of IAA and vice-versa. Disease development is usually associated with alteration in oxidative enzymes, particularly peroxidase, phenol oxidase, ascorbic acid oxidase, however, changes in IAA oxidase during disease development and with plant age have not received much attention and hence it is thought worthwhile to study this enzyme in soybean at different growth stages.

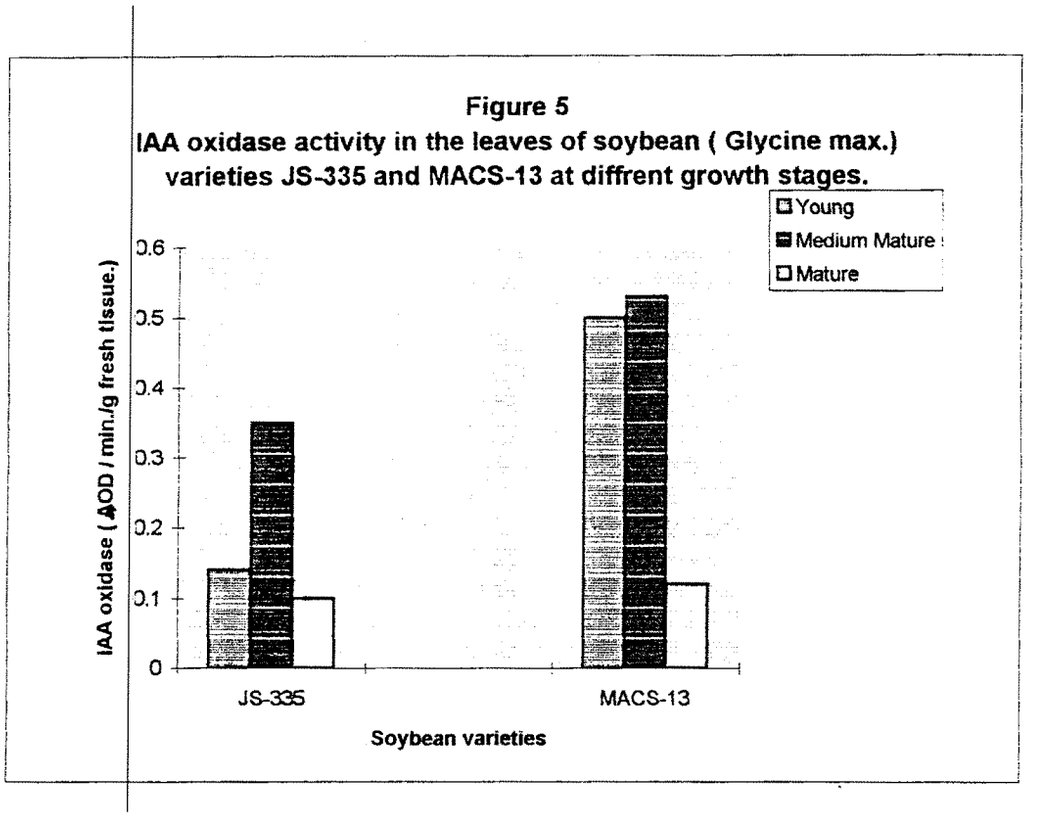
The activity of IAA oxidase studied in soybean at different growth stages show less activity at young and mature stage in case of JS-335 while in MACS-13 it is at mature stage (Table 7 and Fig. 5). Several reasons have been attributed to the lowered activity of IAA oxidase in diseased plants. These includes enhansed activity of polyphenol oxidase (Ramawat el al 1980; Lee et al) while Vidyasekaran and Durairaj, 1973) are of the opinion that decreased activity of catalase and low concetration of manganese may be responsible for low activity of IAA oxidase. On the other hand increased IAA-oxidase activity has been ascribed to low phenolic content and stimulation of catalase activity (Kosuge, 1969). Polyphenols like caffeic acid, chlorogenic acid (Mehrotra, 1980) also inhibit the activity of IAA oxidase. In the present investigation the low activity of IAA oxidase may be due to stimulated activity of Polyphenol oxidase and increase in total phenolic contents.

Table 7 : IAA oxidase activity in the leaves of Soybean (Glycine max) var. JS-335 and MACS-13 at different growth stages

Soybean Growth Stages	Δ OD min^{-1} g^{-1} fresh tissue
JS-335	
Young	0.14
Medium mature	0.35
Mature	0.10
MACS-13	
Young	0.50
Medium mature	0.53
Mature	0.12

The values are mean of three determinations.

Figure 5
IAA oxidase activity in the leaves of soybean (Glycine max.) varieties JS-335 and MACS-13 at diffrent growth stages.



G. Phenolic compounds :

The phenolic compounds from the whole plant powder (Leaf + stem + root), at young, medium mature and mature growth stage were detected by employing unidimenssional paper chromatography in sybean (Glycin max) var. JS-335 and MACS-13. The probable identification of different phenolic compounds was established by comparing the fluorescence under UV-light, UV + NH_3, mix ture of (1:1) 0.3% $FeCl_3$ + 0.3% K_3 Fe $(CN)_6$, and corresponding Rf values of authentic standards. The chromatographs of phenolic compounds detected on paper chromatography are very well seen in Plate 1 to 3 (JS-335) and Plate 4 to 6 (MACS-13). The probable identification of the compounds is given in Table 8 to 10 (JS-335) and Table 11 to 13 (MACS-13). Similarly the quantification of phenolic compounds is also given in Table 14.

It is very clear from the Table 14 that the concertration of phenolic compound goes on increasing from young to medium mature growth stage and decline at maturity. The phenolic compounds identified in JS-335 includes proanthocyanodins, Coumaric acid, Khellin, Catechin, D-catechin, Quercetin derivatives Tannic acid, Catechol + Quinic acid, Caffeic acid, Ferulic acid. Out of these phenolic compounds Quercetin derivatives Tannic acid, Catechol + Quinic acid, Caffeic acid and Ferulic acid are not seen at Medium mature stage in JS-335, but the other compounds such as Myricetin Benzoquinone, Kaempferol + Ellagic acid, 4-hydroxyl betaphenyl coumarin acetate are noticed. In MACS-13 the compounds detected at young stage are more or less the same at Medium mature and Mature stage of growth except few compounds

Table 8 : Detection of Polyphenol from whole plant powder in Soybean var. JS-335 at Young stage of growth

Spot No.	Colour under UV	Colour under UV + NH_3	0.3% $FeCl_3$ + 0.3% $K_3Fe(CN)_6$ 1:1 Mixture	Rf x 100 in BAW	Probable Identification
1.	Yellowish brown	Yellowish brown	Faint blue	4.24	Proanthocyanodins
2.	Yellow	Yellowish brown	Blue	13.33	Coumaric acid
3.	Faint violet	Deep violet	Blue	23.63	Khellin
4.	Faint violet	Deep violet	Blue	30.30	Catechin
5.	Deep violet	Bluish yellow	Blue	36.96	D-catechin
6.	Yellowish brown	Dark brown	Blue	46.66	Quercetin
7.	Dark yellow	Yellow brown	Dark Blue	57.57	Quercetin derivatives
8.	Deep violet	Violet	Dark Blue	70.90	Tannic acid
9.	Faint violet	Faint violet	Deep Blue	82.42	Catechol + Qunic acid
10.	Deep violet	Deep violet	Deep Blue	90.90	Caffeic acid
11.	Faint Yellow	Yellow brown	Deep Blue	96.96	Ferulic acid

BAW : n Butenol : Acetic Acid : Water (4:1:5)

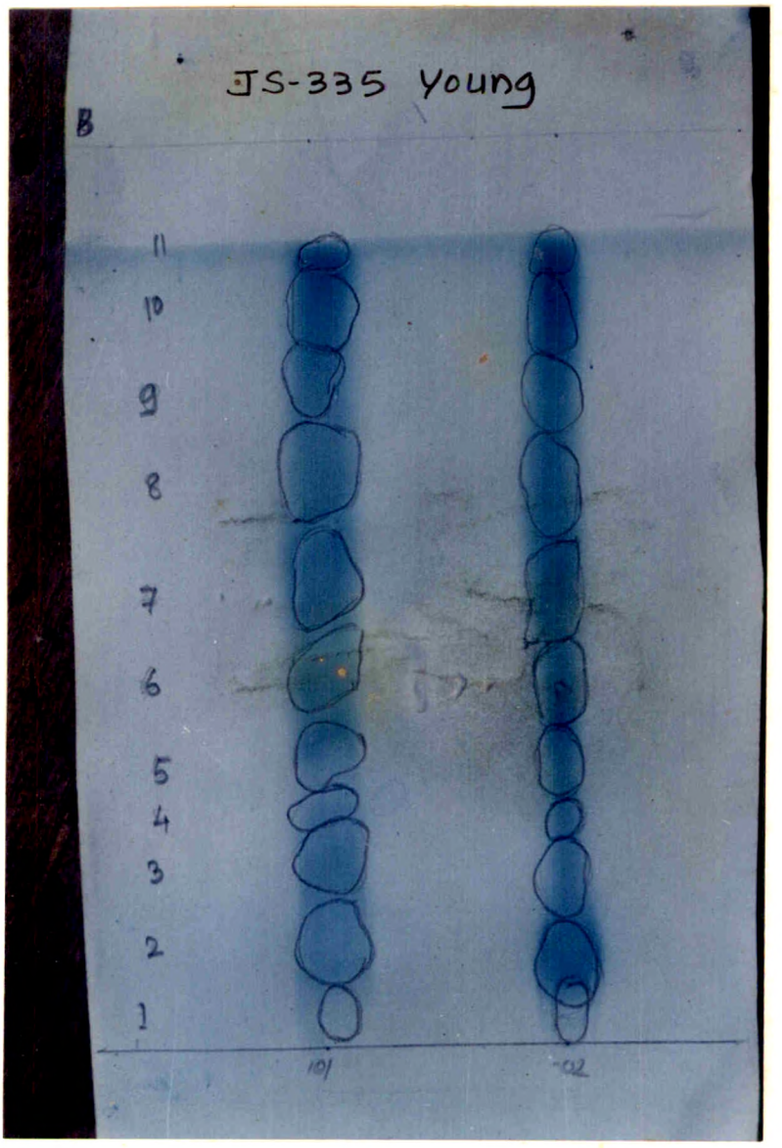


Plate-1

Table 9 : Detection of Polyphenol from whole plant powder in Soybean var. JS-335 at medium mature stage of growth

Spot No.	Colour under UV	Colour under UV + NH_3	0.3% $FeCl_3$ + 0.3% $K_3Fe(CN)_6$ 1:1 Mixture	Rf x 100 in BAW	Probable Identification
1.	Yellowish brown	Yellowish brown	Faint Blue	5.55	Proanthocyanodin
2.	Yellow	Yellowish brown	Faint Blue	13.33	Coumaric acid
3.	Faint violet	Deep violet	Blue	21.11	Khellin
4.	Faint violet	Deep violet	Blue	28.88	Catechin
5.	Deep violet	Bluish yellow	Dark Blue	36.66	D-catechin
6.	Yellowish brown	Dark brown	Dark Blue	46.11	Quercetin
7.	Dark yellow	Yellow brown	Blue	55.0	Myrecetin
8.	Deep violet	Violet	Dark Blue	65.55	4-hydroxy betaphenyl cumarin acetate
9.	Faint violet	Faint violet	Dark Blue	75.55	Flavanol
10.	Deep violet	Deep violet	Deep Blue	86.11	Benzoquinone
11.	Faint Yellow	Yellow brown	Deep Blue	94.44	Kaemphferol + Ellagic acid

BAW : n Butenol : Acetic Acid : Water (4:1:5)

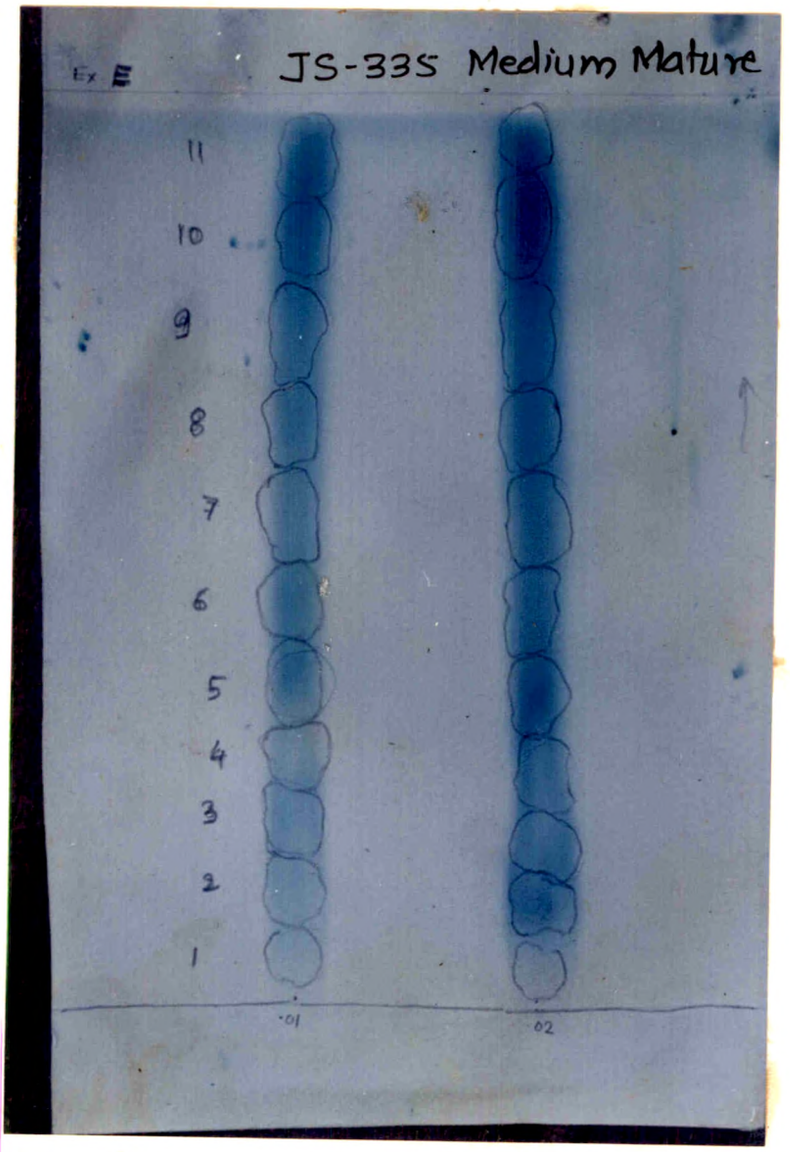


Plate-2

Table 10 : Detection of Polyphenols from whole plant powder in Soybean var. JS-335 at mature stage of growth

Spot No.	Colour under UV	Colour under UV + NH_3	0.3% $FeCl_3$ + 0.3% $K_3Fe(CN)_6$ 1:1 Mixture	Rf x 100 in BAW	Probable Identification
1.	Yellowish brown	Yellowish brown	Faint Blue	2.80	Unknown
2.	Yellow	Yellowish brown	Faint Blue	9.5	Proanthocyanodins
3.	Faint violet	Deep violet	Faint Blue	16.85	Coumaric acid
4.	Faint violet	Deep violet	Blue	24.71	Khellin
5.	Deep violet	Bluish yellow	Dark Blue	34.26	Flavonoid
6.	Yellowish brown	Dark brown	Dark Blue	40.44	Flavonoid
7.	Dark yellow	Yellow brown	Blue	51.68	Myrecetin
8.	Deep violet	Violet	Dark Blue	61.79	Quereetin derivatives
9.	Faint violet	Faint violet	Dark Blue	74.15	Epicatechin + Gallic acid
10.	Deep violet	Deep violet	Deep Blue	86.51	Benzoquinone
11.	Faint Yellow	Yellow brown	Deep Blue	95.50	Kaempferol + Ellagic acid

BAW : n Butenol : Acetic Acid : Water (4:1:5)

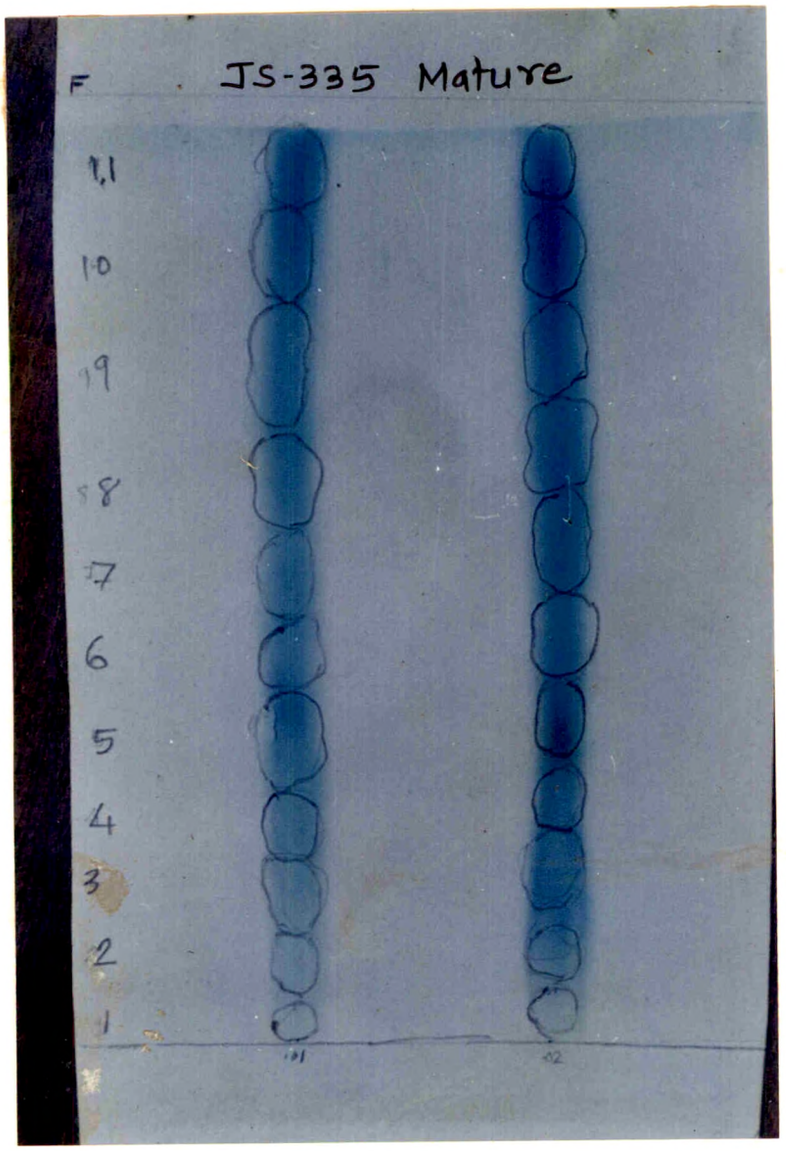


Plate - 3

Table 11 : Detection of Polyphenols from whole plant powder in Soybean var. MACS-13 at young stage of growth

Spot No.	Colour under UV	Colour under UV + NH_3	0.3% $FeCl_3$ + 0.3% $K_3Fe(CN)_6$ 1:1 Mixture	Rf x 100 in BAW	Probable Identification
1.	Yellowish brown	Yellowish brown	Faint Blue	3.63	Proanthocyanodins
2.	Yellow	Yellowish brown	Dark Blue	13.93	Coumaric acid
3.	Faint violet	Deep violet	Blue	22.42	Khellin
4.	Faint violet	Deep violet	Dark Blue	29.69	Catechin
5.	Deep violet	Bluish yellow	Dark Blue	46.16	Quercetin
6.	Yellowish brown	Dark brown	Blue	52.72	Myricetin
7.	Dark yellow	Yellow brown	Blue	62.42	Quercetin derivatives
8.	Deep violet	Violet	Dark Blue	72.72	Gallic acid
9.	Faint violet	Faint violet	Deep Blue	82.42	Catechol + Qunic acid
10.	Deep violet	Deep violet	Deep Blue	89.69	Caffeic acid
11.	Faint Yellow	Yellow brown	Deep Blue	96.36	Kaempferol + Ellagic acid

BAW : n Butenol : Acetic Acid : Water (4:1:5)

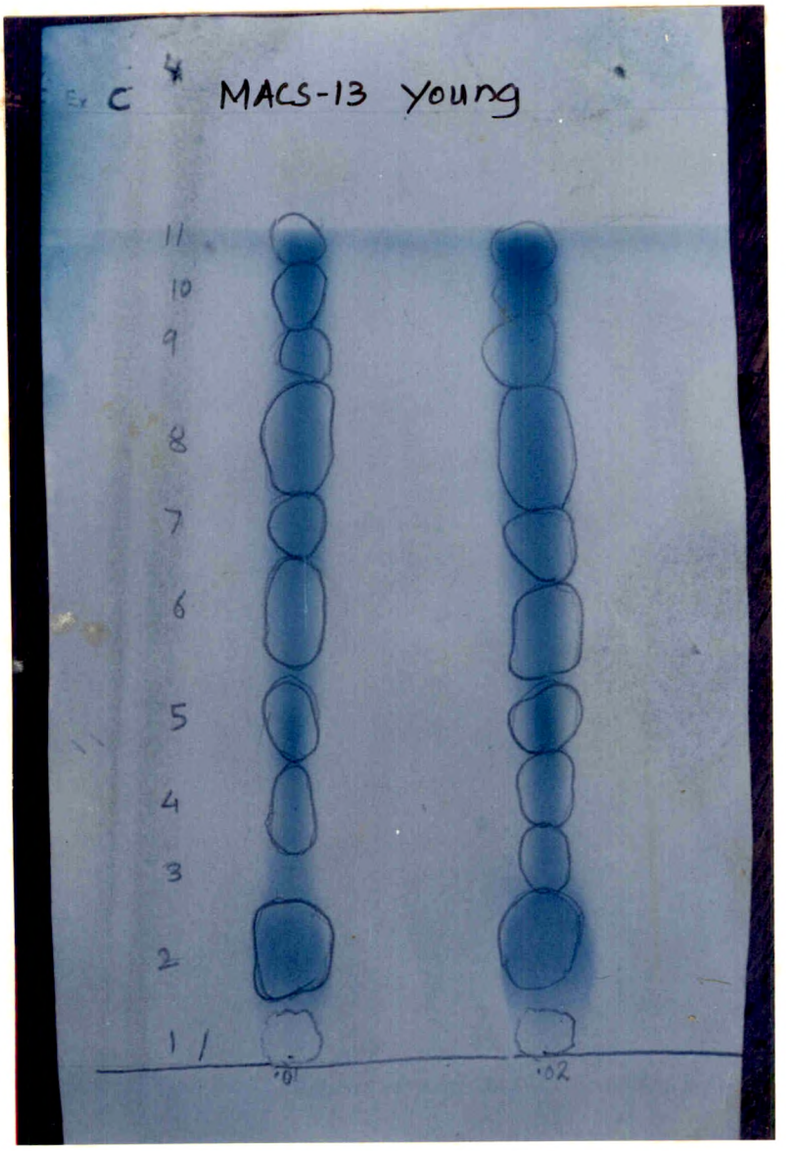


Plate- 4

Table 12 : Detection of Polyphenols from whole plant powder in Soybean var. MACS-13 medium mature stage of growth stage of growth

Spot No.	Colour under UV	Colour under UV + NH_3	0.3% $FeCl_3$ + 0.3% $K_3Fe(CN)_6$ 1:1 Mixture	Rf x 100 in BAW	Probable Identification
1.	Yellowish brown	Yellowish brown	Faint Blue	5.61	Proanthocyanodins
2.	Yellow	Yellowish brown	Faint Blue	14.04	Coumaric acid
3.	Faint violet	Deep violet	Faint Blue	21.91	Khellin
4.	Faint violet	Deep violet	Blue	31.46	Catechin
5.	Deep violet	Bluish yellow	Dark Blue	43.82	Quercetin
6.	Yellowish brown	Dark brown	Blue	55.05	Myricetin
7.	Dark yellow	Yellow brown	Dark Blue	67.41	Coumiryl acid
8.	Deep violet	Violet	Dark Blue	78.08	Chlorogenic acid
9.	Faint violet	Faint violet	Deep Blue	86.51	Catechol + Qunic acid
10.	Deep violet	Deep violet	Blue	93.25	Kaempferol
11.	Faint Yellow	Yellow brown	Faint Blue	96.96	Ferulic acid

BAW : n Butenol : Acetic Acid : Water (4:1:5)

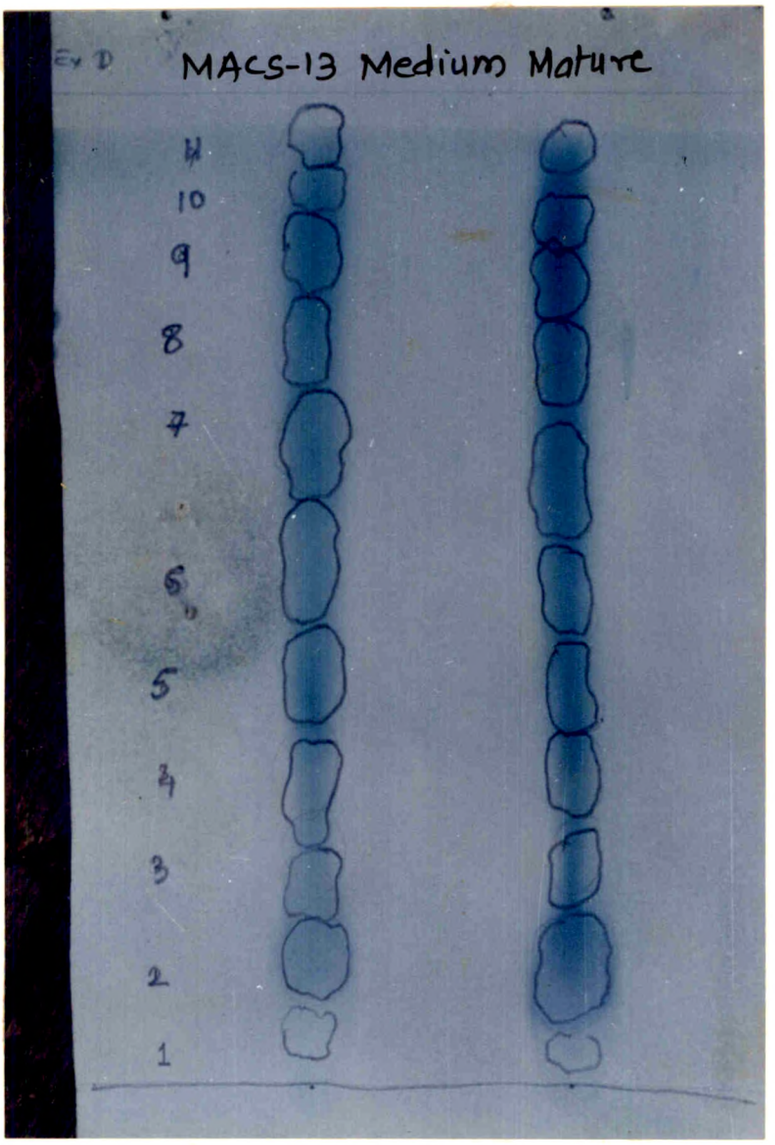


Plate - 5

Table 13 : Detection of Polyphenols from whole plant powder in Soybean var. MACS-13 at mature stage of growth

Spot No.	Colour under UV	Colour under UV + NH_3	0.3% $FeCl_3$ + 0.3% $K_3Fe(CN)_6$ 1:1 Mixture	Rf x 100 in BAW	Probable Identification
1.	Yellowish brown	Yellowish brown	Faint Blue	7.22	Proanthocyanodins
2.	Yellow	Yellowish brown	Faint Blue	17.77	Flavan
3.	Faint violet	Deep violet	Blue	28.33	Catechin
4.	Faint violet	Deep violet	Dark Blue	36.11	D-catechin
5.	Deep violet	Bluish yellow	Blue	46.11	Quercetin
6.	Yellowish brown	Dark brown	Blue	55.55	Myricetin
7.	Dark yellow	Yellow brown	Dark Blue	66.11	Phenyl coumarin acetate
8.	Deep violet	Violet	Dark Blue	75.0	Epicatechin + Gallic acid
9.	Faint violet	Faint violet	Deep Blue	83.88	Catechol + Qunic acid
10.	Deep violet	Deep violet	Deep Blue	92.22	Caffeic acid
11.	Faint Yellow	Yellow brown	Deep Blue	97.22	Ferulic acid

BAW : n Butenol : Acetic Acid : Water (4:1:5)

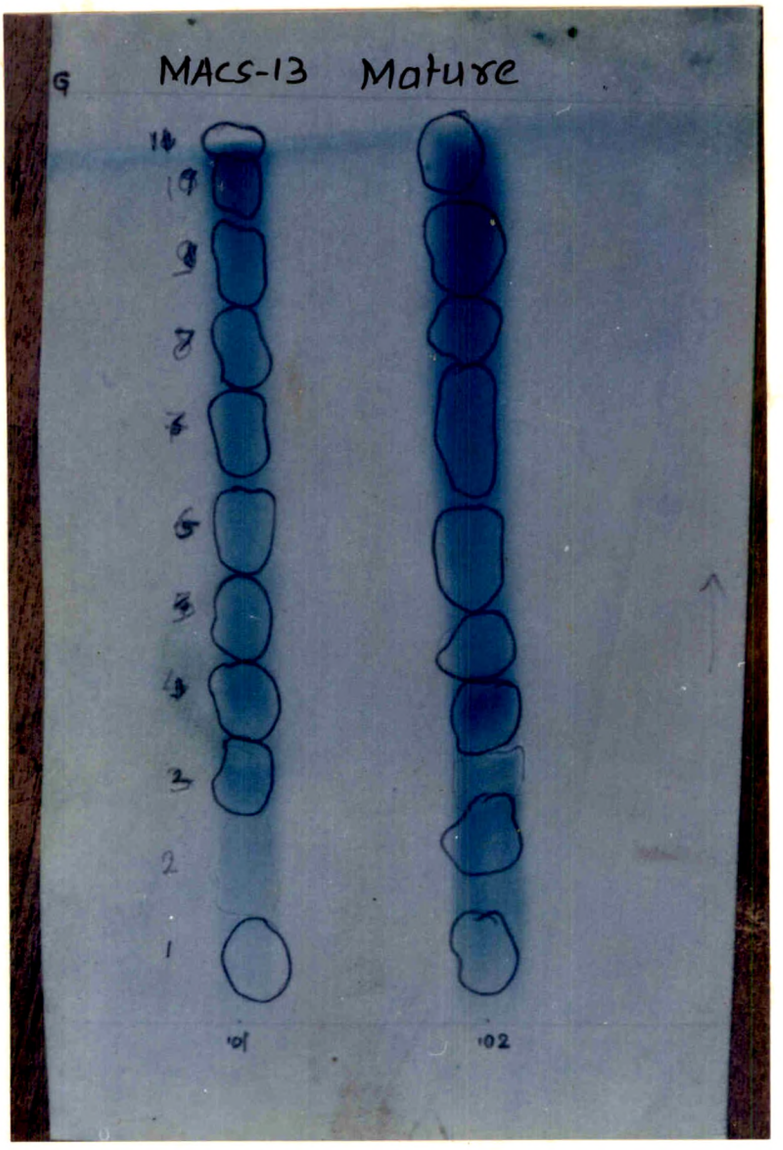


Plate- 6

Table 14 : Quantification of Phenolic compounds (ug/spot) detected in Soybean var. JS-335 and MACS-13 at various growith stages.

Compound	Quantification ug / spot					
	JS-335			MACS-13		
	Young	Medium Mature	Mature	Young	Midium Mature	Mature
Proanthocyanodin	5.85	7.30	5.85	3.9	4.8	3.09
Coumaric acid	13.8	55.2	43.80	9.2	36.8	--
Kellin	12.0	48.0	38.09	8.0	30.0	--
Catechin	17.0	68.0	--	11.33	45.5	25.36
D-catechin	21.3	85.2	--	--	--	11.2
Quercetin	18.5	74.0	--	12.15	56.8	11.2
Quercetin derivatives	24.8	--	58.73	75.0	--	--
Tannic acid	22.5	--	--	--	--	--
Catechal + Qunic acid	11.7	--	--	10.5	40.0	66.0
Caffeic acid	27.8	--	--	18.5	--	66.0
Ferulic acid	31.2	--	--	--	82.5	30.40
Flavonoid	--	--	16.90	--	--	--
Myrecetin	--	51.3	40.71	16.0	50.3	27.15

Contd.

Compound	Quantification µg / spot					
	JS-335			MACS-13		
	Young	Medium Mature	Mature	Young	Midium Mature	Mature
Epicatechin + Gallic acid	--	--	58.00	--	--	38.0
Benzoquinone	--	121.0	99.04	--	--	--
Kampferol + Ellagic acid	--	124.8	96.03	20.8	--	--
4-hydroxy betaphenyl coumarin acetate	55.0	--	--	--	--	--
Flavanol	--	60.0	--	--	--	--
Coumiryl quinic acid	--	--	--	--	34.2	--
Chlorogenic acid	--	--	--	--	35.5	--
Kaempferol	--	80.0	--	--	--	--
Gallic acid	--	--	--	7.60	--	--
Flavan	--	--	--	--	--	29.2
Phenyl coumarin acetate	--	--	--	--	--	40.0

such as coumaryl quinic acid and chlorogenic acid (Medium mature stage) Flavan and phenyl coumarin acetate (Mature stage).

The concentration of these compounds (µg/spot) is low at young, higher at medium mature and become less at mature growth stage (Table 14).

The role of individual compounds, their biosynthesis and metabolism with respect to plant age in soybean will certainly throw a light on defence mechanism against disease development in soybean, which is in progress. From the present data at this, juncture it can only be said that the susceptibility of both the varieties to rust infection observed in past few years in Maharashtra is possibly be due to lower concentration of phenolic compounds in soybean at early stage of growth. The further investigation on this line for the induction of polyphenol by triggering PAL activity is necessary to arrive at correct conclusion.