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EFFECTS OF COLLAR ROT AND RING-BARKING ON THE RHIZOSPHERE MICROFLORA AND CERTAIN CHEMICAL CONSTITUENTS OF SWORD BEAN PLANTS

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It is known that changes in the metabolism of plants will usually be reflected in the microbial population of the rhizosphere also. If the metabolism of the plant is altered by any means, it can bring about quantitative and qualitative changes in the root exudates which in turn will influence the microbial population of the rhizosphere.

Infection of plants by pathogens can also alter the host metabolism and thereby bring about changes in the rhizosphere microflora. Lakshmikumari (1960) observed that virus infection was able to bring about changes in the rhizosphere microflora of *Dolichos labial)* plants. However, the information available on this aspect is only very meagre. It is in this context that the present study was undertaken.

The collar rot of sword bean plants *(Canavalia gladiata* Jacq. DC.) caused by *Rhizoctonia* sp. was selected for this study mainly because the organism usually girdles the stem, renders the phloem tissue inactive and finally kills the plant. The blockage of the phloem tissue will prevent the downward translocation of nutrients.

Ring-barking will also prevent the downward translocation of nutrients which in turn can bring about the death of the plant eventhough this death is not strictly similar to that brought about by Rhizoctonia infection.

A comparative study was therefore made of the changes that are brought about in the microbial population of the rhizosphere of sword bean plants by Rhizoctonia infection and by ring-barking, An attempt was also made to assess the changes that are brought about in the carbohydrate and other nutrient contents of the root system as a result of the above treatments.

Materials and Methods

A local variety of sword bean *(Canavalia gladiata* Jacq. DC.) was used in the study. Single plants were raised in earthen pots of 30 cm diameter using a potting mixture consisting of farm yard manure, river sand and garden soil in the ratio 1:1:1.

Twentyfive days old plants were used for the experiment. They were divided into three lots, one lot was inoculated with *Rhizoctonia* sp., the second lot was ring* barked and the third lot was kept as control.

Inoculations were done at the region just above the cotyledons (approxi. mately 5 to 7 cm above the ground level) with five days old culture of *Rhizoctonia* sp. isolated from naturally infected plants. Culture bits were applied at this region and covered with moist cotton wool.

Ring-barking was done at the region just above the Cotyledons as in the case of inoculated plants using sterile razor blades. Two parallel cuts encircling the stem were made in the bark and the ring like piece of bark formed by the cut was removed from the stem. The width of the bark removed was about 1 cm. The control plants were not given any treatment. The assessment of rhizosphere microflora was done by the dilution plate method. The first root sample was drawn on the day of inoculation. Subsequent samples were drawn at three days interval. Flasks containing 100 ml of sterile distilled water into which the roots were added, were shaken for about half an hour on a mechanical shaker. Appropriate dilutions were prepared from this suspension. The final dilution used for plating was 1:1,000,000.

For making the counts of bacterial, actinomycetes and fungal colonies, one ml of the suspension was plated using 15 ml. of the appropriate medium per petridish.

Soil extract agar (Taylor and Lochbead, 1938), peptonedextrose agar with rose bengal and streptomycin, and Ken-knight's agar were used for the determination of the populations of bacteria, fungi and actmomycetes respectively. The populations are expressed in terms of millions per gram of dry soil.

For the determination of the changes in the chemical constituents in the root system, samples were collected from the same set of plants which were used for the rhizosphere studies. Altogether three samples were drawn, the first sample on the date of the treatment, and the second and third samples, 10 and 18 days after the treatments.

Results and discussion

First visible signs of infection were noted on the third day after inoculation when the felty growth of the fungus was formed at the inoculated region. By this time the leaves showed a number of oily or water soaked lesions. In about eight days after inoculation the leaves become dull grey in colour and started drooping with a characteristic inward rolling. Complete wilting and death of plants with shedding of leaves occurred by the 16th day.

the total carbohydrates fell from 31.23 per cent to 16.22 per cent only in the inoculated plants, it fell to 6,5 per cent in the ring-barked plants. This may be due to the fact that while in the inoculated plants the downward movement of food materials was stopped only gradually that in the ring-barked plants it occurred abruptly with the result that the reserve food materials present in the root system might have been depleted faster on account of the normal metabolic activities. Similar results have been obtained by Mason & Maskell (1928), Leach (1937) and Napper (1938) in other plants. They noted a faster depletion of starch and other nutrients in the root system of plants which were ring-barked.

Thus it is evident that collar rot as well as ring-barking can bring about marked changes in the rhizosphere microflora as well as in the chemical constituents of sword bean plants although the magnitude of these changes are not similar.

Summary

An attempt was made to assess the changes that were brought about in the microbial population in the rhizosphere of sword bean plants as a result of infection by *Rhizoctonia* sp. which causes the rotting of the collar region and the subsequent death of the plant. Healthy and ring-barked sword bean plants were used for comparative purposes. An assessment of the changes which were brought about in the carbohydrates and other nutrient contents of the root system of the plant was also made.

An increase in the microbial population in the rhizosphere of sword bean plants was noted as the plants became older. But this increase was more pronounced in plants which were inoculated with *Rhizoctonia* sp. and also in those which were ring-barked. The highest microbial population was, however, noted in the former. This increase in population was observed in all the three groups of organisms, namely bacteria, actinomycetes and fungi. The increase in the microbial population is considered due to the qualitative and quantitative changes which were brought about in the root exudates as a result of the treatments.

The wide difference in the microbial population in the plants which were inoculated with *Rhizoctonia* sp. and in those which were ring-barked is considered due to the difference in the nature of reaction of the root system of the plants to the two treatments. While the former brought about a diseased condition and the ultimate death of the plant, the latter brought about only the death of the plant as a result of the mechanical injury.

There was a marked reduction in the carbohydrate and nitrogen contents in the root system of ring-barked and inoculated plants. This reduction was very pronounced in the former. It is considered that while in the inoculated plants the

An increase in the percentage of total carbohydrates was noted in healthy plants as they became older. In the ring-barked and inoculated plants, on the other hand, the percentage of total carbohydrates decreased as a result of the treatment. This reduction was very highly pronounced in the ring-barked plants. While the percentage of total carbohydates in healthy plants increased to 38.58 per cent from 31.23 per cent in 18 days, it fell to 16.22 and 6.50 per cent respectively in the inoculated and ring-barked plants during the same period.

Although there was a fall in the percentage of total nitrogen as the plants became older, this fall was very pronounced in the ring-barked and inoculated plants While the percentage of nitrogen in healthy plants, fell from 2.56 to 2.06 in eighteen days it fell to 0.43 and 0·18 per cent in the inoculated and ring-barked plants respectively, during the same period.

There was an increase in the microbial population in the rhizosphere of sword bean plants as the plants became older. But this increase was more pronounced in plants which were inoculated with *Rhizoctonia* sp. and also in those which were ring-barked. The highest microbial population was noted in plants which received the former treatment. While the total microbial population in healthy plants rose from 73.21 million to 216.51 million in 18 days, it rose to 245.25 and 306.17 million in the ring-barked and inoculated plants respectively, during the same period The increase in the microbial population was evident in all the three groups of organisms, namely, bacteria, actinomycetes and fungi.

Ring-barking as well as inoculation with a pathogen can be expected to influence the metabolism of the plant, which in turn can bring about quantitative and qualitative changes in the root exudates. These changes will normally be reflected in the microbial population of the rhizosphere.

One of the reasons for the very high microbial population in the rhizosphere of inoculated plants as compared to that in the ring-barked plants may be that while in the former the stopnage of the downward translocation of nutrients was gradual, in the latter it was instantaneous. The changes in the metabolic activities of the root system in the inoculated and ring-barked plants also cannot be similar. While the former brings about a diseased condition, the latter brings about the death of the plant as a result of the mechanical iujury. In the diseased plant the interaction between the host and the pathogen as well as the toxic substances produced by the latter can seriously affect the metabolism of the plant. It is known from the work of Kerr (1957) and others that Rhizoctonia can produce toxic metabolic products.

A marked reduction in the carbohydrate and nitrogen contents of the root system was noted in the inoculated and ring-barked plants as compared to that in healthy plants. This reduction was much greater in the ring-barked plants. While

Treatment	Age of plant: 25 days (day of treatment)		28 days (3 days after treatment)		32 days (6 days after treatment),		36 days (11 days after treatment)		40 days (14 days after treatment)		43 days (18 days after treatment))	
	Popu- lation	R. S. ratio	Popu- lation	R. S. ratio	Popu- lation	R, S. ratio	Popu- lation	R. S. ratio	Popu- lation	R. S. ratio	Popu- lation	R. S. ratio
Healthy	26.91	5.25	29.97	4.14	33.35	4.65	42.43	6.26	65.27	9.26	80.42	12.17
Ring-barked	26.91	5.25	33.70	4.65	49.91	6.96	49.01	7.25	85.48	12.10	89.00	14.07
Inoculated	26.91	5.25	34.81	4.82	53.98	7.52	64.64	9.54	103.36	14.64	110.73	17.01
Colrol soil	5.12	..	7.24	..	7.17	..	6.77	..	7.06	..	6.33	..

Treatment	Age of plant: 25 days (day of treatment)		28 days (3 days after treatment)		32 days (6 days after treatment)		36 days (11 days after treatment)		40 days (14 days after treatment)		43 days (18 days after treatment)	
	Popu- lation	R. S. ratio	Popu- lation	R. S. ratio	Popu- lation	R. S. ratio	Popu- lation	R. S. ratio	Popu- lation	R. S. ratio	Popu- lation	R. S. ratio
Healthy	4.42	3.83	4.41	4.54	4.99	4.94	5.30	4.53	7.33	6.10	12.55	9.43
Ring-barked	4.42	3.83	4.42	4.54	5.76	5.70	6.42	5.40	10.i2	8.35	15.05	11.31
Inoculated	4.42	3.83	4.64	4.78	6.34	6.27	8.12	6.94	16.10	13.12	18.33	13.77
Control soil	1.15	..	0.97	..	1.01	..	1.17	..	1.20	..	1.33	..

The leaves of the ring-barked plants exhibited a general discolouration from the third day onwards and no lesions were formed. By the 9th day the lower leaves started rolling inwards and became distinctly yellow in colour. Complete wilting and death of plants occurred by the 13th day.

A steady increase in the rhizosphere microflora of healthy plants was noted as the plants became older. Ring-barking as well as inoculation with *Rhizoctonia* sp. exerted a stimulatory effect on the rhizosphere microflora (Table 1). This stimulatory effect was noticeable in all the three groups of organisms, namely, bacteria, actinomycetes and fungi, from the third day after treatment.

The total bacterial populations in the healthy, ring-barked and inoculated plants on the third day after treatment were 49.35, 70.19 and 65.85 million respectively. The corresponding populations on the 15 th day after treatment were 125.54, 141.20 and 177.11 million (Table 2).

While the actinomycetes population in the healthy plants was only 80.42 million that in the ring-barked and inoculated plants were 89.00 and 110.73 million respectively, eighteen days after the treatment (Table 3).

Similarly while the fungal population in healthy plants was only 12.55 million that in the ring-barked and inoculated plants were 15.05 and 18.33 million respecti vely, eighteen days after the treatment (Table 4).

Changes in the chemical constituents in the root system

A decrease in the percentage of moisture in the root system of healthy, ring barked and inoculated plants was noticed as the plants became older. The reduction in the moisture content was greater in the inoculated and ring-barked plants. While the moisture content in the root system of healthy plants fell from 82.80 to 70.72 percent within a period of 18 days that in the ring-barked and inoculated plants fell to 61.52 and 63.88 per cent respectively.

A decrease in the total sugars was noted as the plants became older. This reduction was very highly pronounced in the case of ring-barked plants. While the percentage of total sugars in the healthy and inoculated plants fell from 9.63 to 6.35 and 5.36 per cent respectively within 18 days that in the ring-barked plants fell to 2.20 per cent during the same period.

Although the percentage of crude fibre in healthy plants increased as the plants became older, that in the ring-barked and inoculated plants decreased considerably. While the percentage of crude fibire which was 21.60 on the day of treatment rose to 32.23 per cent in the healthy plants in 18 days, it fell to 4.30 per cent and 10.86 per cent in ring-barked and inoculated plants respectively during the same period.

Table 1

Total [illegible] populations in the rhizosphere of healthy, inoculated and ring barked sword bean plants (in millions)

Treatment	Age of plant: 25 days (day of treatment)	28 days (3 days after treatment)	32 days (6 days after treatment)	36 days (10 days after treatment)	40 days (14 days after treatment)	43 days (18 days after treatment)
Healthy	73.21	83.73	97.09	88.57	175.01	218.51
Ring-barked	73.21	108.31	130.71	88.50	216.87	245.25
Inoculated	73.21	104.30	155.62	88.14	283.25	316.17
Control soil	8.53	20.16	19.25	11	20.57	19.83

Table 2

Total bacterial populations in the rhizosphere of healthy, ring-barked and inoculated sword bean plants (in millions)

Treatment	25 days (day of treatment)		28 days (3 days after treatment)		Age of Plant: 32 days (6 days after treatment)		36 days (10 days after treatment)		40 days (14 days after treatment)		43 days (18 days after treatment)	
	Population	R. S. ratio	Population	R. S. ratio	Population	R. S. ratio	Population	R. S. ratio	Population	R. S. ratio	Population	R. S. ratio
Healthy	41.87	3.41	49.31	4.43	53.75	5.30	61.84	5.53	102.41	8.31	125.54	9.31
Ring barked	41.87	3.41	70.19	5.87	75.80	6.77	94.07	8.42	120.47	9.73	141.20	11.60
Inoculated	41.87	3.41	64.85	5.51	89.10	8.00	123.84	11.04	163.79	13.30	177.11	14.55
Control soil	12.26	..	11.85	..	11.07	..	11.21	..	12.31	..	12.17	..

സംഗ്രഹം

റൈസക്കോണിയ എന്ന കുമിൾ മൂലമുണ്ടാകുന്ന രോഗം ബാധിച്ച വാളരിങ്ങച്ചെടികളുടെ റൈസോസ്ഫിയറിൽ ബാക്ടീരിയ, ആക്ടിനോമൈസീറ്റ്സ്, ഫംഗസുകൾ എന്നീ മൂന്നുതരം സൂക്ഷ്മജീവികളുടെ സംഖ്യ വളരെയധികം വർദ്ധിക്കുന്നതായി കാണപ്പെട്ടു. തണ്ടിൽനിന്നും ഒരു സെന്റീമീറ്റർ ഭാഗത്തെ തൊലി ഇളക്കിമാറ്റിയ ചെടികളിലെ റൈസോസ്ഫിയറിലെ സൂക്ഷ്മജീവികളുടെ സംഖ്യ രോഗബാധയില്ലാത്ത ചെടികളിലേതിൽനിന്നും കൂടുതലായിരുന്നു. രോഗം ബാധിച്ച ചെടികളിലേയും തണ്ടിൽനിന്നും ഒരു സെന്റീമീറ്റർ ഭാഗത്തെ തൊലി ഇളക്കിമാറ്റിയ ചെടികളിലെയും വേരുപടലത്തിലെ കാർബോ ഹൈഡ്രേറ്റിന്റേയും നൈട്രജന്റേയും അളവ് ഗണ്യമായ തോതിൽ കുറയുന്നതായി കാണുകയുണ്ടായി.