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Effect of *Azorhizobium caulinodans* on yield in plant growth regulator induced nodulated wheat

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ABSTRACT

In the laboratory seedling were induced with nodule-like outgrowths using different growth regulators 2,4-D,IBA and NAA in nitrogen free Hoagland solution. Induced seedlings were inoculated with *Azorhizobium caulinodans* (ORS 571) in wheat variety C-306. One set was also raised as control. The treated paranoulated wheat seedlings were transferred to pot culture. The data was collected on 30, 60 and 90 days after sowing, which reveals that as a *Azorhizobium caulinodans* with 2,4-D treated plants shows maximum photosynthetic rate followed by NAA and IBA combinations. The biomass production was maximum in *Azorhizobium caulinodans* treated with 2,4-D followed by IBA and NAA.

KEYWORDS

Plant Growth Regulators, Nodulation, *Azorhizobium caulinodans*

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Wheat is main food of North India. It is a tough task to meet out the increasing demand of food for burgeoning population. In this situation farmers use a huge amount of nitrogenous fertilizers to get maximum yield, which creates many health hazards as well as soil is also becoming barren day by day. Hence by using biofertilizer for nitrogen fixation, we can solve these problems unitely. *Azorhizobium caulinodans* plays a significant role to increase wheat yield, which used as biofertilizer for wheat crop. *Azorhizobium caulinodans* has been reported to fix nitrogen and also increase yield (Ridge *et al*., 1993, Christian Weninger and Vanderleyden, 1994). The use of Plant growth regulators with *Azorhizobium caulinodans* affects tillering, leaf area, photosynthetic rate and its biological yield was the object of the present study in the para nodulated and uninoculated control.

MATERIALS AND METHODS

Seeds of wheat variety C-306 were sterilized by dipping in $HgCl_2$ 5% for three minutes and rinsed thoroughly with water. These seeds were then sown in sand gravel. After seven days seedlings were transferred to test tubes covered with black paper, filled with Nitrogen free Hoagland solution with different treatments to induce para-nodules with 2, 4-D (0.5 ppm), IBA (8 ppm) and NAA (8 ppm). These para-nodulated seedlings were introduced to *Azorhizobium caulinodans* (ORS 571). After 7-8 days induced para-nodules can be seen with naked eyes. The plants were then transferred in cemented pots (50 x 50 x 50 cm^3) and 12 plants were maintained in each pot. The tillers were counted at different stage of growth and the leaf area was measured using the leaf area meter (LICOR 3000 model USA) and was expressed as cm^2/plant. The chlorophyll content in leaf was determined at different stages using the method described by Hiscox and Israelstam (1979). Photosynthetic rate, stomatal conductance and resistance were measured by IRGA (Infra Red Gas Analyzer) LICOR 6250. At harvest the total biomass production was measured including straw and grain yield in different treatments. The data were statistically analyzed.

RESULTS AND DISCUSSION

The addition of the synthetic auxin 2,4-D to wheat seedlings resulting in the formation of nodule like out growths that can best be described as modified lateral roots. We found that concentration (0.5 ppm) formed 100% of plants these out growths, addition of 2, 4- D also significantly reduce shoot and root length but dry weight was unaffected. Synthetic

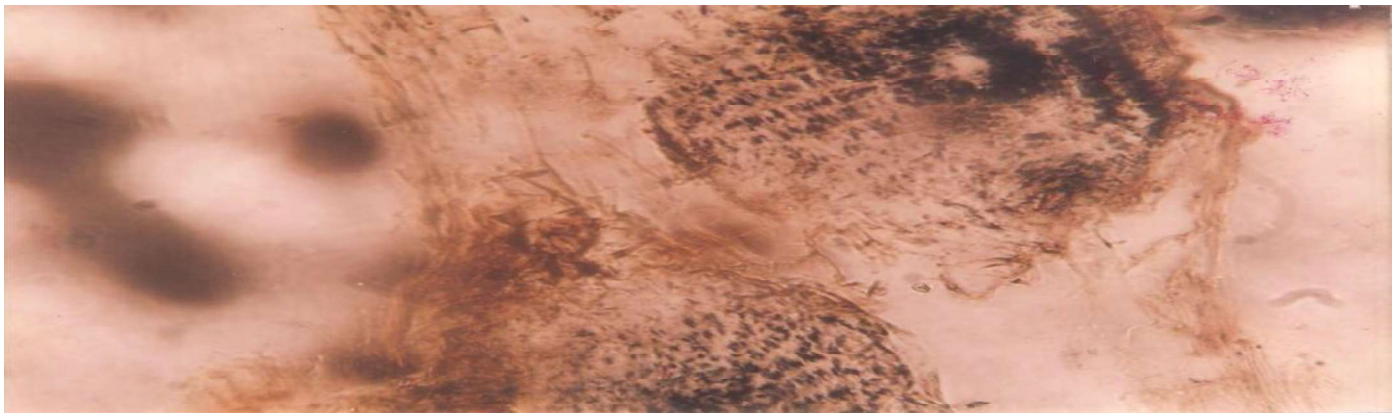

Fig. 1. Colonization of *Azorhizobium caulinodans* within para-nodules of wheat roots

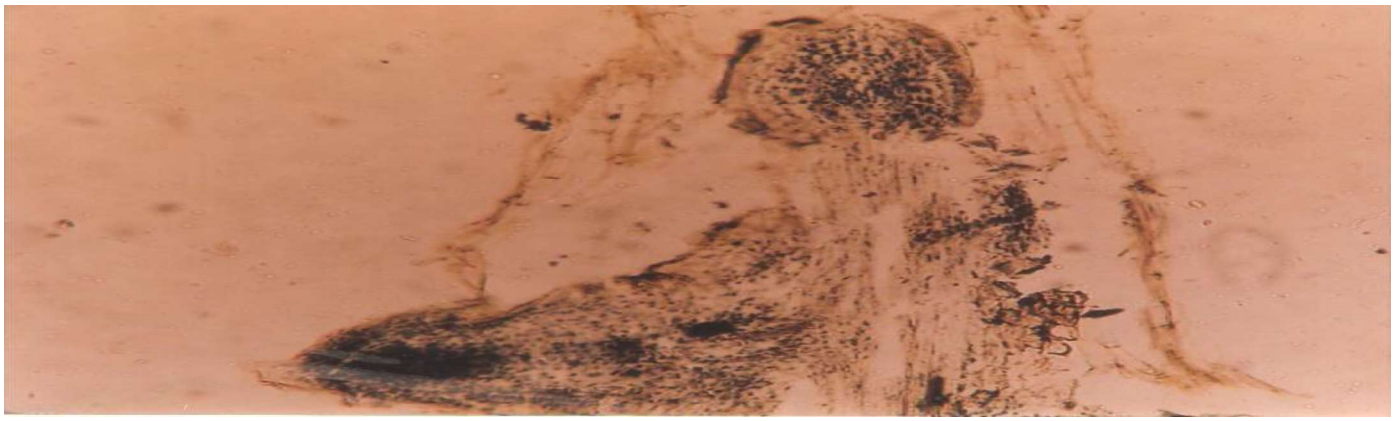

Fig. 2. Colonization of *Azorhizobium caulinodans* within para-nodules of wheat roots

Table 1. Showing the effect of plant growth regulators and *Azorhizobium caulinodans* on tillering, leaf area, chlorophyll content and photosynthetic rate at 60 DAS

Treatments	Tiller no. 60 DAS	Leaf Area 60 DAS	Chlorophyll content 60 DAS	Photosynthetic Rate 60 DAS	Biological Yield
Control	3.74	66.52	2.956	11.383	10.24
A. caulinodans	4.50	94.25	3.082	21.787	12.38
2,4-D	4.13	70.25	3.058	23.290	12.21
2,4-D + Azo.	5.64	114.52	3.538	31.930	13.56
IBA	4.26	75.25	2.954	13.480	14.29
IBA + Azo	5.75	118.62	3.615	17.040	15.42
NAA	4.03	67.52	2.931	14.940	12.08
NAA + Azo.	4.26	108.42	3.106	18.730	13.11
CD at 5%	0.296	5.64	0.177	1.238	0.912

auxin NAA (8 ppm) induces some root thickenings, but at lower concentration had no effect on root development. NAA does not play a very significant role in bio-mass, yield, root shoot length and on other factors. IBA (8 ppm) also shows some outgrowth on roots of wheat seedlings and cause proliferation, high leaf area, high tiller number, good bio-mass and better grain yield. Cytological investigations revealed that there were more *Azorhizobium caulinodans* present within the para-nodule than in the root system. This indicates that the major part of ethylene production come from the *Azorhizobium caulinodans* activities within the para-nodules. The total chlorophyll contents at 60 DAS revealed that it was more with *Azorhizobium caulinodans* which was better than control, however the plants inoculated with *Azorhizobium caulinodans* have more chlorophyll content than the plants grown with 2,4-D IBA and NAA alone. However the plants treated with IBA + *Azorhizobium caulinodans* had higher total chlorophyll content followed by 2,4-D < *Azorhizobium caulinodans* and the lowest chlorophyll content was noted in NAA treated either with *Azorhizobium caulinodans* or alone. The results obtained had been supported by the earlier work done by division of Microbiology I.A.R.I. (Kaushik *et al.,* 2008) where the wheat seedlings treated with *Azorhizobium caulinodans* had higher nitrogenase activity, enhanced higher biomass used as an index of nitrogen fixation.

The photosynthetic rates measured at 60 days after transplantation in the pots revealed that it was higher in *Azorhizobium caulinodans* treated plants. The plants treated with 2,4-D along with *Azorhizobium caulinodans* had shown the higher photosynthetic rates with better stomatal conductance and least stomatal resistance, whereas the plants treated with IBA or NAA either alone or with *Azorhizobium caulinodans* had poor photosynthetic rate consequent upon the relatively low stomatal conductance and higher stomatal resistance. Our results are in confirmatory with the earlier reports (Tchan and Kennedy 1991, Panwar 1993 in wheat and rice) and (Saikia *et al*., 2006 in maize). It is evident from Table 1 that IBA treated plants either alone or with *Azorhizobium caulinodans* have higher seed and straw yield (biological yield) followed by 2,4-D treated plants and NAA treated plants, however, it was better than untreated plants (control). IBA inoculated with *Azorhizobium caulinodans* had produced maximum biological yield followed by 2,4-D and NAA. The NPK content in seed and straw was relatively more with 2,4-D treated *Azorhizobium caulinodans* inoculated plants followed by IBA and NAA treated plants either alone or with different bio fertilizers used in this study. These results are in confirmatory with the earlier report by (Elanchezhian and Panwar, 1997) in wheat and (Naidu, 2001) in rice through non significant differences were found in potash content in seed and straw. The protein content was maximum in *Azorhizobium caulinodans* treated plants, either alone or with auxins.

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