Growth and yield response of canola to bacterial inoculants: a three-year field assessment

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ABSTRACT

Nine rhizobacteria able to produce auxins (IAA-like substances) in *vitro* were assessed for the ability to promote growth of canola *(Brussica napus* L., cv. Legend) in a growth chamber. Strains that exhibited plant growth promotion activity in this screening were then tested as seed inoculants for canola at multiple field sites (n=3 or 4; 16 reps.) in Saskatchewan over a three-year period. Plant growth and yield response varied from site to site and year to year. For example, in the 1993 study *Bacillus* strain HPR3 increased ($P<0.05$) seed yield by 104 to 158 $kg \cdot ha^{-1}$ (8-18%) at two of the four sites, but had no effect at the other sites. In the 1994 study, *Xunthomonas* strain HPR2, strain HPR3 and *Arthrobucter* strain HPR9 increased ($P<0.05$) canola seed yield by 90 to 109 $kg \cdot ha^{-1}$ (7%) at the Melfort site only. In the 1995 study, strain HPR9 increased (non-significant) seed yield by 140 $kg \cdot ha^{-1}$ (12%) at the Melfort site. Non-significant seed yield increases (45-90 $kg \cdot ha^{-1}$; 5-8%) also were noted for other strains at other sites. None of the inoculants affected seed oil (%) content, seed emergence or plant height. Sometimes inoculants decreased yields at some sites. Our results demonstrate that some rhizobacteria may benefit canola growth, but edaphic and climatic conditions can influence inoculant efficacy. Furthermore, site-to-site field variability suggests that extensive field testing will be required to demonstrate the potential usefulness of rhizobacteria as biofertilizers.

INTRODUCTION

Plant growth-promoting rhizobacteria (PGPR) colonize the rhizosphere and enhance the growth of many plant species. However, responses of field crops to inoculation with PGPR strains are often highly variable from one year to another, and from one field location to another. Variability in soil and climatic conditions, both spatial and temporal, can be a barrier to interpretation of field response data (De Freitas & Germida, 1992; Kloepper et al., 1988). Previously, we isolated and identified several PGPR strains from crop rhizospheres which enhanced the growth of lettuce, cabbage, onion and canola in growth chamber studies (De Freitas & Germida, 1995). We hypothesized that these bacteria could be used as seed-applied inoculants to enhance canola yield on a wide range of soil quality and climatic conditions in Saskatchewan. A collaborative research project conducted with the Saskatchewan Wheat Pool Product Development Branch allowed us to test this hypothesis. The objective of this work was to assess the in-field effectiveness of plant growth-promoting rhizobacteria (PGPR) as inoculants for canola.

MATERIALS & METHODS

Field plots were established by the Saskatchewan Wheat Pool Product Development Branch in 1993, 1994, and 1995 at 3 or 4 locations in Saskatchewan. Plots (8.5 m^2) were seeded (6.0 kg $seeds \cdot ha^{-1}$) and fertilized at seeding with N and P according to field testing recommendations. Replicates (N-16) were harvested at maturity and seed yield determined.

Eight bacterial strains including HPRL (Unknown), HPR2 *(Xanthomonas* sp), HPR3 *(Bacillus* sp), HPR5 *(Pseudomonas* sp), R85 *(Xanthomonas* sp 1), HPR9 *(Arthrobacter*

sp), R92 ***(Bacillus*** sp), and 2W7M4 ***(Xanthomonas*** sp 2) were assessed during the 3-year study. Rhizobacteria were grown in Luria Bertani medium (Bric, et al., 1991), centrifuged, concentrated and inoculated to Vitavax RS coated canola ***(Brassica naps*** L., var. Legend) seeds ***(ca.*** 10^6 - 10^7 cfu per seed). Talc was added to the mix to soak up moisture and form a coat around the seed.

RESULTS

During the 1993 study inoculation with HPR1 or HPR3 increased ($P<0.05$) grain yield of canola at Beatty and Lake Lenore, respectively. These increases (up to 18%) were 207 and 158 kg·ha^{-1} in both sites, respectively (Table 1).

Table 1. Grain yield of canola grown at Manitou, Beatty, Lake Lenore, and Yorkton 1993 field plots. Average of 16 reps.

Treatment	Manitou		Beatty		Lake Lenore		Yorkton	
	kg·ha^{-1}	%[a]	kg·ha^{-1}	%	kg·ha^{-1}	%	kg-ha-^{1}	%
Control	1388		1172	-	836	0	683	
HPR1	1253	-10.0	1379*	17.6	836	-3.7	769	12.6
HPR2	1306	-0.05	1271	8.4	805	[illegible]	723	5.6
HPR3	1431	3.1	1276	8.8	994*	18.8	772	13.0
HPR5	1355	-0.02	1260	7.5	833	-0.3	737	7.9
R85	1430	3.0	1136					-6.1
R92	1382	-0.004	1261	-3.7 6.0	794 853	-5.0 2.0	641 834	22.1
2W7M4	1364	-0.01	1211	3.3	806	-3.6	779	14.0

a Percentage yield relative to the control; * Different from the uninoculated control at $P<0.05$.

During the 1994 study, strains HPR2, HPR3 and HPR9 increased ($P<0.05$) grain yield from 6.7% to 8.1% at the Melfort site. In addition, strain HPR2 increased grain yield by 7.5% at the Yorkton site, but this was not significant due to variability between replicates (Table 2).

Table 2. Grain yield of canola grown at Yorkton, Watrous and Melfort field 1994 plots. Average of 16 reps.

Treatment	Melfort		Yorkton		Watrous	
	kg-ha-^{1}	%[a]	kg-ha-1	%	kg-ha-^{1}	%
Control	1338		1358	-	1003	
HPR2	1428*	6.7	1460	7.5	971	-3.1
HPR3	1447*	8.1	1213	-10.7	971	-3.1
HPR9	1430*	6.9	1247	-8.1	905	-9.8

[a] Percentage yield relative to the control; * bifferent from the uninoculated control at $P<0.05$.

During the 1995 study, there were some non-significant seed yield responses to the inoculants at three of the four field locations (Table 3). For example, at the Melfort site,

inoculation with the three isolates resulted in non-significant increases in grain yield of canola by 6.2 to 20% (73-140 kg-ha-t). Four of the 16 replications were lost due to poor growing conditions during the 94/95 field season. Nevertheless, HPR9 yielded 140 kg·ha-1 more than control plots. Other isolates e.g., HPR2 and HPR3 increased grain yield by 46 and 93 kg·ha-1 (5 - 8.1%) at the Yorkton and Watrous sites, respectively.

Although the yield increases observed for 1995 were not significant, it is important to note that similar yield increases ($P<0.05$) of 90 - 109 kg·ha-1 were obtained for HPR inoculated canola at the 1994 Melfort site (Table 2). Despite these positive responses, it should be noted that some inoculants also resulted in a yield decline at the Rosetown (116 - 136 kg-ha-t), Yorkton (72 kg·ha-1) and Watrous (104 kg·ha-1) sites (Table 3). There were no differences with respect to seed oil (%) content, emergence or plant height at any site.

Table 3. Grain yield of canola grown at Melfort, Rosetown, Yorkton and Watrous 1995 field plots. Average of 16 reps.

Treatment	Melfort[a]		Rosetown		Yorkton		Watrous	
	kg·ha-1	%[b]	kg-ha-1	%	kg·ha-1	%	kg-ha-1	%
Control	1160	6.4	1528		915	-	114	-
HPR 2	1234	6.3	1395		961	5.0	1045	-9.0
HPR3	1233	[illegible]	1392	-8.9	879	-3.9	1242	8.1
HPR9	1300	12.0	1412	-7.6	873	-4.6	1156	0.6

[a] 12 replications; [b] Percentage yield relative to the control.

CONCLUSIONS

- It is apparent that inoculation of canola with PGPR that produce IAA-like substances can significantly enhance the growth and yield of canola. However, crop response to inoculation may vary depending on field conditions;
- In our study, yield differences were limited to a specific site. The reason for this is unknown, but edaphic and climatic conditions can influence inoculant efficacy;
- Our results indicate that extensive multi year field tests will be required to demonstrate the potential usefulness of rhizobacteria as biofertilizers.

REFERENCES

Bric, J.M., Bostock, R.M. & Silverstone, SE. 1991. Rapid ***in situ*** assay for indoleacetic acid production by bacteria immobilized on a nitrocellulose membrane. Appl. Environ. Microbiol. 57:535-538.

De Freitas, J.R. & Germida, J.J. 1992. Growth promotion of winter wheat by fluorescent pseudomonads under field conditions. Soil Biol. & Biochem. 24: 117-l 146.

De Freitas, J.R. & Germida, J.J. 1995. Identification of IAA-producing rhizobacteria as biofertilizers for vegetable and field crops. In: Proceedings of 87th Annual Meeting of the American Society of Agronomy - ASA, CSSA, SSSA, Saint Louis, MI. Oct. 29 - Nov. 3, 1995.

Kloepper, J.W., Lifshitz, R. & Schroth, M.N. 1988. ***Pseudomonas*** inoculants to benefit plant production. IS1 Atlas of Science: Animal and Plant Sciences, pp. 60-64.

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