

EFFECTS OF METALAXYL ON AGRONOMIC TRAITS OF ZERO TANNIN LENTIL

A. Matus¹ and A. Slinkard²,

¹ Department of Crop Science and Plant Ecology and ²Crop Development Centre, University of Saskatchewan, Saskatoon, SK. S7N 0W0.

ABSTRACT

The seed coat of lentil (*Lens culinaris* Medikus) contains tannin precursors which oxidize during storage, discolouring seed from the normal olive green to deep brown. Seed coats of zero tannin (ZT) lentil do not discolour during storage due to the absence of these tannin precursors. ZT lentil has a thinner, more delicate seed coat than standard lentils. Thus, ZT lentil seed is highly susceptible to mechanical damage and germinating seeds have a high frequency of seed rot. The effect of fungicide on seedling emergence and other agronomic traits of ZT lentil was investigated using a split-plot design replicated three times for two years. The main effect was lentil lines and the split effect was untreated seed vs. seed treated with the fungicide metalaxyl (6g a.i./100 kg seed). Seedling emergence of ZT lentil was greatly improved following fungicidal seed treatment with metalaxyl. Fungicidal seed treatment had no effect on agronomic traits of ZT lentil, other than seedling emergence and seed yield. Commercial production of ZT lentil must involve fungicidal seed treatment and special precautions during seeding, harvesting and cleaning to minimize mechanical damage to the seeds. These ZT lines did not yield competitively with Eston or Laird lentil. A second cycle of hybridization of the better ZT lentil lines to adapted lines followed by selection for yield should increase the frequency of high yielding ZT lentil lines.

INTRODUCTION

The seed coat of lentil (*Lens culinaris* Medikus) contains phenolic compounds which can discolour seed from the normal olive green to deep brown on prolonged storage due to an oxidative polymerization of low molecular weight soluble tannin to high molecular weight condensed tannin (Nozolillo and de Bezeda 1984) and possibly other compounds such as aurones (Vaillancourt 1984). Discoloured lentil seeds are unacceptable to the consumer and accordingly are discounted and downgraded.

The Crop Development Centre of the University of Saskatchewan discovered a single recessive gene that prevented the formation of tannins in the seed coat of lentil. Subsequently some zero tannin (ZT) lentil lines were developed and tested. Seed increase of these lines was a problem, probably because of poor seed quality. The seed coat is thinner than normal seed coats, the seeds are vulnerable to mechanical damage (Vaillancourt 1984, Matus 1991) and a high frequency of seed rot occur when the seeds are planted. Metalaxyl is a fungicidal seed treatment highly effective in controlling seed rot caused by Phycomycetes such as *Pythium ultimum*.

Therefore, the objective of this study was to determine the effects of metalaxyl seed treatment on seedling emergence, seed yield, days to first flower, days to maturity, plant height and 1000-seed weight of ZT lentil.

LITERATURE REVIEW

Pythium ultimum is the most common cause of seed rot and seedling blight in chickpea (Cicer arietinum), and reductions in seedling emergence of more than 90% have been reported (Kraft et al. 1988). According to Kaiser and Hannan (1983), seed treatment with metalaxyl reduced seed rot and increased seed yield of chickpea. de Bezeda (1980) reported that the ZT lentil line PI 345635 was more susceptible to seed rot than tannin containing lines. Germinating seeds of lentil exude water soluble phenolic compounds that inhibit germination of fungal spores (Agrawal and Khare 1973). Furthermore, Parashar and Sindhan (1986) found that pea (Pisum sativum L.) cultivars resistant to powdery mildew have high levels of phenolic compounds in their stems and leaves. Black-seeded cultivars of bean (Phaseolus vulgaris) were resistant to Rhizoctonia solani infection, while white-seeded cultivars were susceptible to seed infection and pre-emergence damping-off (Prasad and Weigle 1976). Poor emergence of white-seeded kabuli chickpea was also associated with Pythium infection (Kaiser and Hannan 1981), whereas brown- or black-seeded desi types were more resistant to infection (Kaiser and Hannan 1983).

MATERIALS AND METHODS

The ZT lines were F₂-derived F₆ lines from crosses between the ZT line P.I. 345635 and Laird or Eston. In 1989 Laird, Eston and Rose lentil plus 13 ZT lentil lines with or without fungicidal seed treatment were grown at the University of Saskatchewan Preston Plots. In 1990 Laird and Eston lentil plus 14 ZT lentil lines with or without fungicidal seed treatment were grown on the University of Saskatchewan Sutherland Farm. The fungicide was Apron 69 LSD which contains metalaxyl [N-(2,6 dimethylphenyl)-N-(methoxyacetyl)-alanine methyl ester] (Thompson 1985) and was applied to the seed as a powder at slightly above 6 g a.i. per 100 kg seed.

Individual plots consisted of four rows, 4 m long and 0.3 m apart. The experimental design was a split plot with three replications. The main plot consisted of a single lentil line and the split plot consisted of the fungicidal seed treatment and untreated seed. The seed for each plot was inoculated with Rhizobium leguminosarum "C" at time of seeding by using dead wheat as the carrier and planting it in the row with the lentil seed. The seeding rate was about 40 seeds per linear meter of row. The seeding dates for the two experiments were 4 May 1989 and 19 May 1990.

Data were collected in both years on seedling emergence, days to flower, days to maturity, plant height, seed yield and 1000-seed weight as follows: seedling emergence was determined as the number of plants in a meter of one of the inner rows four weeks after planting. Number of days to first flower was recorded when 50% of the plants in a plot were in flower. Number of days to maturity and plant height (cm) were recorded at maturity. To minimize mechanical damage to the seed

coat, the lentil plots were cut by hand, bagged and dried using hot air for 48 hours. The lentil plants were gently threshed in a belt thresher, and cleaned with a Clipper cleaner. The clean seed from each plot was weighed and yield converted to kg per hectare. A random sample of 200 seeds from each plot was counted, weighed and the weight multiplied by 5 to obtain 1000-seed weight.

RESULTS AND DISCUSSION

Effect of Metalaxyl Seed Treatment on Seedling Emergence

Table 1. Seedling emergence of tannin-containing lentils and zero tannin lentils untreated or treated with metalaxyl fungicide and grown on the Preston Plots in 1989 and the Sutherland Farm in 1990

Lines	Preston 1989			Sutherland 1990		
	Fungicide	Control	Mean	Fungicide	Control	Mean
	Plants/m row [†]					
Laird	36.0	34.4	35.2	40.0	40.0	40.0
Eston	35.0	35.4	35.2	40.0	37.6	38.8
Rose	33.0	33.0	33.0	----	----	----
Tannin mean	34.7	34.3	34.5	40.0	38.0	39.0
ZT-22	----	----	----	37.6	23.6	34.3
ZT-23	23.6	18.0	20.8	32.6	24.4	28.5
ZT-24	24.0	19.0	21.5	34.0	25.0	29.5
ZT-28	24.4	18.4	21.4	37.6	26.0	31.8
ZT-31	23.0	19.0	21.0	39.4	28.6	34.0
ZT-34L	23.6	17.4	20.5	36.4	21.4	28.9
ZT-34S	24.4	22.4	23.4	34.4	23.6	29.0
ZT-35	19.6	15.4	17.5	37.0	20.4	28.7
ZT-36	23.0	14.6	18.8	41.0	24.6	33.0
ZT-41	20.0	15.0	17.5	33.6	24.6	29.1
ZT-42L	22.4	18.4	20.4	31.4	26.4	28.9
ZT-42S	20.0	15.6	17.8	37.4	23.4	30.4
ZT-46	25.6	19.0	22.3	35.6	20.6	28.1
ZT-47	28.4	14.6	21.5	34.0	23.6	28.8
ZT mean	23.2	17.4	20.3	35.8	24.0	30.0
ST. error (fung*line)	1.1	1.1	0.8	2.4	2.4	1.7
C.V (%)	---	---	8.6	---	---	13.1

[†] A stand of 35 plants per meter of row is more than adequate.

Metalaxyl fungicidal seed treatment improved average seedling emergence of ZT lentil lines (33% in 1989 and 50% in 1990) (Table 1). The tannin containing lines (Eston, Laird and Rose) had a significantly higher seedling emergence than the mean of the ZT lines in both experiments. Differences in seedling emergence among ZT lines and among tannin-containing lines were not significant. The fungicide by line interaction was significant in both experiments due to the lack of response of the tannin containing lines to fungicidal seed treatment relative to the marked response of the ZT lines (Table 1). In addition, the fungicide by ZT interaction was significant for the Preston Plots, indicating that the fungicide disproportionately affected seedling emergence of at least one of the ZT lines (ZT-47, Table 1).

The seed coat of Eston, Laird and Rose lentil is high in tannin and acts as a protective barrier against fungal infection. Lentil seeds without tannin in the seed coat are more susceptible to seed rot than lentil seeds with tannin in their seed coat. The thin seed coat of ZT lentil cracks easily (Vaillancourt 1984, Matus 1991) and causes reduced germination and emergence in the field, due to the high incidence of seed rot (Prasad and Weigle 1976) and/or imbibitional chilling injury (Tully et al. 1981). ZT lentil seeds have a thin seed coat, imbibition occurs more rapidly and the amount of nutrients that extrude and/or leak out is probably greater than in tannin containing lentils. These nutrients provide a rich medium for growth of seed rotting pathogens (Spaeth 1989).

Different mechanisms have been proposed to explain how tannin protects plants against microorganisms. According to Swain (1979), tannins act through their ability to combine with protein, reducing protein digestibility. Others have suggested that tannins may act as enzyme inhibitors (Bell 1981; Allen and Friend 1983).

Effect of Fungicidal Seed Treatment on Seed Yield

Table 2. Seed yield of tannin-containing lentils and zero tannin lentils untreated or treated with metalaxyl fungicide and grown on the Preston Plots in 1989 and Sutherland Farm in 1990.

Lines	Preston 1989			Sutherland 1990		
	Fungicide	Control	Mean	Fungicide	Control	Mean
	kg/ha					
Laird	1508	1378	1443	2125	2230	2177
Eston	1603	1474	1539	1916	2230	2072
Rose	1232	1197	1215	----	----	----
Tannin mean	1448	1350	1399	2021	2230	2125
ZT-22	----	----	----	1464	1368	1416
ZT-23	106	50	79	1344	1302	1323
ZT-24	70	48	60	1447	1660	1704
ZT-28	799	492	646	1826	1575	1701
ZT-31	68	54	61	988	920	954
ZT-34L	135	83	109	1560	1429	1495
ZT-34S	194	130	162	1426	1220	1341
ZT-35	217	105	161	1189	1111	1150
ZT-36	85	58	72	1017	1111	1064
ZT-41	115	78	96	1441	1378	1410
ZT-42L	211	171	191	1260	929	1094
ZT-42S	168	103	136	1187	1086	1137
ZT-46	142	105	124	1115	1082	1199
ZT-48	66	61	63	1228	903	1066
ZT mean	183	118	151	1324	1220	1290
St. error (fung*lin)	33	33	23	83	83	59
C.V. (%)	---	---	14.6	---	---	10.5

Tannin-containing lentil outyielded the ZT lentils in both year (Table 3). Differences in seed yield were highly significant among ZT lentil lines but not among tannin-containing lines. Metalaxyl fungicidal seed treatment improved seed yield of almost every ZT line, but no consistent fungicidal effect was detected for the tannin-containing lentil lines Eston, Laird and Rose (Table 2). Differences in seed yield due to fungicide for tannin vs. ZT lentil lines were highly significant on the Sutherland Farm in 1990, due to a slight yield decrease from fungicide treatment of tannin containing lines and a large yield increase from fungicide treatment of the ZT lines (Table 2). Differences in seed yield due to metalaxyl fungicide among the ZT lentil lines were highly significant on the Preston Plots, but not on the Sutherland Farm in 1990, probably due to severe bird damage on the Preston Plots rather than a fungicide effect. Metalaxyl fungicidal seed treatment had no effect on number of

days to first flower, days to maturity, plant height, and 1000-seed weight (data not presented).

CONCLUSIONS

Two conclusions can be made from these studies; 1) seedling emergence and seed yield of ZT lentil lines were greatly improved following metalaxyl fungicidal seed treatment, and 2) metalaxyl fungicidal seed treatment had no effect on other agronomic traits of ZT lentil.

ACKNOWLEDGMENTS

The authors wish to acknowledge the assistance provided by Dr. R. J. Baker in the statistical analysis, Mr. K. Thai in conduct of the experiments, and Ms. Maria A. Matus in the collection of field data.

LITERATURE CITED

Agrawal, S. C., and M. N. Khare. 1974. An antifungal chemical exuded by germinating seeds of lentil. *Sci. and Cult.* 40:464-465.

Allen, F. H. E., and J. Friend. 1983. Resistance of potato tubers to infection by Phytophthora infestans: a structural study of haustorial encasement. *Physiol. Plant Pathol.* 22:285-292.

Bell, A. A. 1981. Biochemical mechanisms of disease resistance. *Ann. Rev. Plant Physiol.* 32:21-81.

de Bezeda, M. V. 1980. Effect of environmental conditions on lentil seeds. M.Sc. Thesis, University of Ottawa, Ottawa, ON.

Kaiser, W. J., and R. M. Hannan. 1981. Preemergence damping-off of chickpea in the Palouse region of Eastern Washington caused by Pythium ultimum. *Phytopathology* 71:230. (Abstr.).

Kaiser, W. J., and R. M. Hannan. 1983. Etiology and control of seed decay and preemergence damping-off of chickpea by Pythium ultimum. *Plant Disease* 67:77-81.

Kraft, J. M., M. P. Haware, and M. M. Hussein. 1988. Root rot and wilt diseases of food legumes. Pages 565-575. In R. J. Summerfield (ed.). *World crops: Cool season food legumes*. Kluwer Academic Publisher. Dordrecht, The Netherlands.

Matus, A. 1991. Agronomic evaluation of zero tannin lentil. M.Sc. Thesis, University of Saskatchewan, Saskatoon, SK.

Nozolillo, C., and G. M. de Bezeda. 1984. Browning of lentil seeds, concomitant loss of viability, and the possible role of soluble tannins in both phenomena. *Can. J. Plant Sci.* 64:815-824.

Parashar, R. D., and G. S. Sindhan. 1986. Biochemical changes in resistant and susceptible varieties of pea in relation to powdery mildew disease. *Prog. Hort.* 18:135-137.

Prasad, K., and J. L. Weigle. 1976. Association of seed coat factors with resistance to *Rhizoctonia solani* in *Phaseolus vulgaris*. *Phytopathology* 66:342-345.

Spaeth, S. C. 1989. Extrusion of protoplasm and protein bodies through pores in cell wall of pea, bean, and faba bean cotyledons during imbibition. *Crop Sci.* 29:452-459.

Swain, T. 1979. Tannins and lignins. Pages 657-682. In G. A. Rosenthal and D. J. Jansen (ed.). *Hervibores: Their interaction with secondary plant metabolites*. Academic Press. New York.

Thompson, W. T. 1985. Agricultural chemicals. Book IV-fungicides. Pages 139-140. Thompson Publications. Fresno, CA.

Tully, R. E., M. E. Musgrave, and A. C. Leopold. 1981. The seed coat as a control of imbibitional chilling injury. *Crop Sci.* 21:312-317.

Vaillancourt, R. 1984. Seed coat darkening and the inheritance of tannin content in lentil. M.Sc. Thesis, University of Saskatchewan, Saskatoon, SK.