EFFECT OF SEED TREATMENT WITH α-AMYLASE ON THE BIOMETRY OF THE SESAME PLANT AND ON BIOCHEMICAL COMPOSITION OF SEEDS

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ABSTRACT

Seeds of sesamum cv. C-50 and type-13 were soaked in α-amylase solutions (0.1 % to 3 %) for 18 hours before sowing. Presoaking upto 0.3 % of the enzyme solution increased the plant height, number of capsules on the main branch, fruiting length, seeds per capsules and the oil content of the harvested seed, but decreased the days to maturity and protein and soluble carbohydrate content of seeds. The grain yield also increased significantly in cv. C-50. Treatment with higher concentrations of the enzyme solution had the opposite effect on most of these parameters. Soaking with 0.2 % had given the maximum beneficial effect and lowered the maturity period by 15 days.

INTRODUCTION

MOBILIZATION of endosperm reserves during the germination of cereal grains is accompanied by the action of several hydrolytic enzymes, which become operative[1]. α-amylase, the enzyme responsible for starch hydrolysis in association with β-amylase and endo-β-glucanase, is synthesized (induced) during germination[2] and is under the direct control of gibberellic acid[3–6]. In oil seeds, direct involvement of amylases has not been shown. However, Yamada[7] reported that in castor, lipids from different parts of the seed, especially cotyledons and endosperm, disappear during germination followed by gradual accumulation of carbohydrate upto 4 days. In oilseeds, fats get converted to sugars[8,9] which are transferred to embryo. Thus, sugars are essential for seed germination and the seedling growth of oil yielding plants, and α-amylase can play a catalytic role by increasing the availability of sugars. This has been amply demonstrated, though indirectly, by the effect of gibberellic acid in inducing normal growth in certain dwarf varieties of garden pea and some single gene dwarf mutants of maize, and enhancing elongation in other plants[10]. The exogenous supply of α-amylase should, therefore, behave partly in a manner similar to the effect of gibberellic acid *viz* the increase in rate of plant growth. The only limitations being the permeability of the imbibing seed to this enzyme and the damage done to other tissues by the enzyme. The present study describes the effect of pretreatment of seeds by α-amylase on the biometry of the sesame plant and the biochemical composition of the harvested seeds.

MATERIALS AND METHODS

The seeds of sesamum varieties C-50 and Type-13 were obtained from the All India Coordinated Research Project on oil seeds (sesamum), University of Udaipur. Field experiments were done in plot size 4 m × 4 m with a plant-to-plant distance of 15 cm and row-to-row distance of 30 cm. The seeds were soaked in various concentrations of α-amylase solutions (0.1–3 %) for 18 hr at 20°C. The control was soaked in distilled water only. The treatment seeds were sown by hand dibbling. A basal dose of fertilizer N:P:K (30:20:20 kg/ha) was used. Developing seeds were harvested at varying time intervals after flowering. Data for two consecutive years with three replications for each treatment were collected.

Analytical procedures:

Oil content was determined by the method described in AOAC[11]. Crude protein, fatty acids, total free amino acids and water soluble carbohydrates were estimated according to Snell and Snell[12] and free fatty acid by the method of Radha Pant *et al.*[13]

RESULTS

The effect of presoaking of seeds in α-amylase solution on the biometry of the plant is given in tables 1 and 2 and on the chemical composition of the harvested seed in tables 3 and 4 for varieties C-50 and type-13 respectively.

Table 1 *Effect of pretreatment of seeds with α-amylase on the biometry of Sesamum variety C-50.*

Pretreatment with α-amylase (%)	Plant height (cm)	Seed moisture at 40 DAF (%)	Capsule on main branch (No.)	Fruiting length (cm)	Capsule length (cm)	Capsule breadth (cm)	Seeds per capsule (No.)	Plant maturity (Days)	Grain yield kg/plot**
Control	102	46.8	31	67.5	5.9	4.1	101*	110	0.61*
0.1	128	41.0	47	75.0	6.2	3.9	110	95	0.63
0.2	129	40.1	54	83.6	6.2	4.1	110	94*	0.81
0.3	117	39.7*	51	76.3	6.2	3.7	110	96	–
0.4	117	–	44	67.7	5.2	3.5	110	96	–
0.5	117	–	40	65.5	5.2	3.7	108	99	–
1.0	113	–	36	59.0*	4.8	3.8	106	97	–
2.0	98*	–	30	60.2	4.7	3.3	104	100	–
3.0	98*	–	29*	60.2	4.4*	3.2*	103	101	–
S. Em. ±	2.87		2.47	2.43	0.24	0.15	2.98	0.30	–
C. D. at 5%	8.21		7.05	6.93	0.70	0.42	8.52	0.85	–
C. D. at 1%	10.99		9.43	9.28	0.93	0.57	11.39	1.14	–

Underlined figures indicate maximum values. *minimum values. 40 DAF is the period for physiological maturity of the grain. **Plot size was 12 sqm.

Table 2 *Effect of pretreatment of seeds with α-amylase on the biometry of Sesamum Type-13.*

Pretreatment with α-amylase (%)	Plant height (cm)	Seed moisture at 40 DAF (%)	Capsule on main branch (No.)	Fruiting length (cm)	Capsule length (cm)	Capsule breadth (cm)	Seeds per capsule (No.)	Plant maturity (Days)	Grain yield kg/plot**
Control	88*	43.2	26	54*	4.4	3.0*	95*	106	1.10
0.1	131	34.9	27	72	5.6	3.1	110	92*	0.98
0.2	125	36.5	30	70	5.1	3.2	109	92*	0.90
0.3	115	35.7*	26	60	5.0	3.2	105	94	0.88*
0.4	116	–	24	60	4.6	3.1	105	98	–
0.5	102	–	22	57	4.6	3.1	104	103	–
1.0	102	–	21*	57	4.5	3.1	103	101	–
2.0	93	–	22	54*	4.4	3.2	100	105	–
3.0	88*	–	21*	54*	4.3*	3.0*	100	102	–
S. Em. ±	2.21	–	0.46	1.81	0.17	0.15	3.04	0.35	–
C. D. at 5%	8.32	–	3.93	5.17	0.49	0.43	8.69	1.00	–
C. D. at 1%	8.46	–	5.27	6.92	0.66	0.57	11.62	1.42	–

Underlined figures indicate maximum values. *minimum values. 40 DAF is the period for physiological maturity of the grain. **Plot size was 12 sqm.

Table 3 *Effect of pretreatment of seeds with α-amylase on the chemical composition of harvested seeds of sesame var. C-50*

Pretreatment α-amylase (%)	Moisture (%)	Protein (%)	Oil (%)	Soluble carbo-hydrate (%)	Free amino acids (%)	Free fatty acids (%)
Control	4.1	26.85	47.05*	12.41	2.37*	1.16
0.1	4.5	25.78	49.85	12.34	2.95	0.96
0.2	4.3	24.45*	51.30	12.33*	3.15	0.94*
0.3	4.4	25.14	49.23	13.48	2.70	0.97
0.4	4.3	25.65	47.66	13.54	2.61	1.19
0.5	4.2	26.06	47.85	13.56	2.57	1.43
1.0	4.0	26.78	47.37*	15.84	2.50	1.23
2.0	4.3	26.89	47.37*	15.79	2.49	1.20
3.0	4.3	26.99	47.37*	15.85	2.45*	1.07
S. Em.±	0.11	0.44	0.38	0.10	0.08	0.03
C. D. 5%	0.32	1.24	1.07	0.29	0.23	0.07
C. D. 1%	0.43	1.67	1.43	0.39	0.31	0.10

Underlined figures indicate maximum value. *Minimum values.

Table 4 *Effect of pretreatment of seeds with α-amylase on the chemical composition of harvested seeds of sesame var. Type-13.*

Pretreatment α-amylase (%)	Moisture (%)	Protein (%)	Oil (%)	Soluble carbo-hydrate (%)	Free amino acids (%)	Free fatty acids (%)
Control	3.8	25.97	48.10	11.35*	2.23	1.21
0.1	3.9	25.85	49.20	12.68	1.86*	0.97
0.2	4.1	25.40*	50.68	12.38	1.89	0.75*
0.3	4.0	26.11	47.90	13.20	2.22	1.21
0.4	3.9	26.03	47.76	13.93	2.96	1.21
0.5	4.0	26.10	46.56	13.92	3.21	1.24
1.0	4.3	26.40	46.32	14.05	2.91	1.45
2.0	4.0	26.40	46.26	14.20	2.54	1.44
3.0	3.8	26.86	46.25*	14.24	2.59	1.43
S. Em ±	0.10	0.12	0.27	0.21	0.06	0.04
C. D. 5%	0.28	0.33	0.78	0.60	0.17	0.10
C. D. 1%	0.38	0.44	1.04	0.80	0.22	0.14

Underlined figures indicate maximum values. *Minimum values.

Pre-soaking in 0.4% and lower concentrations of amylase solution:

The treatment of C-50 cv seeds with 0.2% solution increased plant height from 102 cm to 129 cm, the number of capsules on main branch from 31 to 54, the fruiting length from 67.5 cm to 83.6 cm, the number of seeds per capsule from 101 to 110 and the grain yield per plot from 0.6 kg to 0.8 kg. Days to maturity decreased, however, from 110 to 94. The effect was maximum at 0.1% level in the case of type-13, which showed an increase in plant height from 88 cm to 131 cm, fruit length from 54 cm to 72 cm, capsule length 4.4 cm to 5.6 cm, seeds per capsule from 95 to 110. The days to maturity also dropped from 106 to 92. However, the yield showed a decline. The physiological maturity of the developing seed of sesame is reported to be attained in 40 days after flowering, and is indicated by a decrease in seed moisture[14]. Seed moisture in both the varieties decreased at 0.3% treatment implying an enhancement of maturity. Oil content increased from 47.05% to 51.30% in C-50 seeds and from 48.10% to 50.68% in type-13 at 0.2% level, whereas protein content decreased from 25.97%

to 25.40% in type-13, and from 26.85% to 24.45% in C-50. Free fatty acids also showed a downward trend with the minimum at 0.2% level.

Pre-soaking in 0.5% and higher concentrations of amylase solution:

With higher concentrations of amylase solution used, the plant height, the number of capsules on main branch, fruiting length, length of capsules, breadth of capsule (only in C-50), and the number of seeds per capsule showed decreasing trend in both the varieties reaching values less than the control at 3% level. Protein and oil contents reverted to control values.

DISCUSSION

Pre-soaking of seeds of sesame var C-50 and type-13 with various concentrations of α-amylase solution showed a definite change in the biometry of the plants and in the chemical composition of the harvested seeds. Varying the concentrations of enzymes had varying effects and in fact, these effects can be grouped into two enzyme levels, namely, lower upto 0.3% and higher levels greater than 0.3%. In general, changes by treatment using these levels have been opposite. Seed treatment with low levels of α-amylase produced beneficial changes in the biometry and in the chemical composition. The oil content increased by 9% and the total free fatty acids decreased by 20% in C-50 and by 5.3% and 38% in type-13, respectively. Lowering of free fatty acids would improve the quality of the oil[15]. Similarly, the treatment with low levels of α-amylase also showed a beneficial trend in the plant in terms of an increased number of capsules on main branch, the fruiting length and the number of seeds per capsule. The lowering of moisture content of seed at 40 DAF showed that the effect is towards enhancement of the maturity, and consequently, a reduction in the harvest period by as much as fifteen days in both the varieties, which is an important feature of this study. Although grain yield has shown improvement only in the C-50 variety, the total beneficial effect is evident. Since these data represent a mean of two consecutive years of experiments done in mini-plots, it can be concluded that treatment with α-amylase would benefit the crop.

25 January 1985; Revised on 23 September 1985

1. Mayer, A. B. and Poljakoff-Mayber, A., *Metabolism of germinating seeds. The germination of seeds.* Pergamon Press, Frankfurt-am-Main, 1975, p. 106.
2. Bewley, J. D. and Black, M., *Physiology and biochemistry of seeds in relation to germination.* Vol 1. *Development, germination and growth.* Springer-Verlag, Berlin, Heidelberg, New York, 1978, p. 177.
3. Momotani, Y. and Kato, J., *Plant Physiol.*, 1966, **41,** 1395.
4. Tanaka, Y. and Akazawa, T., *Plant Physiol.*, 1970, **46,** 586.
5. Jacobsen, J. V., Scandalios, J. G. and Varner, J. E., *Plant Physiol.*, 1970, **45,** 367.
6. Tanaka, Y., Ito, T. and Akazawa, T., *Plant Physiol.*, 1970, **46,** 650.
7. Yamada, M., *Sci. Pap. Coll. Gen. Educ. Univ. Tokyo*, 1955, **5,** 161.
8. Bradbeer, C. and Stumpf, P. K., *J. Biol. Chem.*, 1959, **234,** 498.
9. Canvin, D. T. and Beavers, H., *J. Biol. Chem.*, 1961, **236,** 988.
10. Phinney, B. O., *Proc. Natl. Acad. Sci. USA*, 1956, **42,** 185.
11. Official methods of analysis. Association of Official Agricultural Chemists. Washington D. C. 10th edn., 1965.
12. Snell, F. D. and Snell, C. T., *Colorimetric methods of analysis.* Vol. III. D. Van Nostrand Co. Inc., Princeton, N. J. and New York 1953.
13. Radha Pant, Nautiyal, G. C. and Singh, J. B., *Indian J. Biochem. Biophys.*, 1973, **18,** 116.
14. Nanawati, G. C., *Lipid metabolism in the developing seeds* of *Sesamum indicum* L., Ph.D. Thesis, Udaipur University, Udaipur 1982.
15. Dhawan, S., Singhvi, S. C. and Simlot, M. M., *J. Food Sci. Tech.*, 1972, **9,** 23.