INTERACTION OF LECTINS WITH DIFFERENT PLANT PROTOPLASIS

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PLANT protoplasts have been isolated from various sources and their agglutination reactions with different newly purified phytolectins have been studied to understand the nature of the cell membrane.

The plant protoplasts have now been used to study the nature of the cell membrane of plants. By the influence of lectins protoplasts may be aggregated to facilitate the cell fusion. Concanavalin A induces the agglutination of protoplasts from cultured carrot cells^{2,3}. Lectin may be helpful in detecting a specific type of protoplasts from a mixture of different sub-populations⁴. No further work on this aspect has yet been reported.

The materials used for isolation of protoplasts were as follows:

Epicotyls of Oryza sativa, Triticum aestivum, Butea monosperma and Trichosanthes anguina; leaves of Momordica charantia var. muricata, Cephalendra indica, and Nicotiana tabecum; the callus tissues of Daucus carota, Canavalia ensiformis and C. gladiata. The epicotyls were mostly seven days old and the leaves were expanded and young. The callus tissues were grown in MS media⁵.

The epicotyls were dissected into small pieces and incubated in enzyme solution containing 0.5% macerozyme R-10, 5.5% cellulase Onozuka R-10, 0.1 M CaCl₂ in 0.7 M mannitol⁶⁻⁸ (pH 5.8). The leaf pieces were also treated for the yield of protoplasts. The callus tissues were incubated in the enzyme solution^{2,9} containing 2% macerozyme R-10 and 5.5% cellulase Onozuka R-10 in 0.6 M Mannitol and 0.1 M CaCl₂ (pH 5.6) for 3-6 hr at 37° C with occasional shaking. Protoplasts were then separated^{2,4} from the enzyme solution. The protoplasts $(2 \times 10^{5-6}/\text{ml})$ were incubated with the lectins $(40 \text{ to } 50 \,\mu\text{g/ml})$ in a ratio of 1:1 (v/v).

The lectins were purified from seeds of the plants Momordica charantia var. muricata (MCm A)¹⁰, Trichosanthes anguina (TAA)¹¹. Butea monosperma (BMA)¹². The concanavalin A (Con A) was supplied by V. P. Chest Institute. These lectins interacted with the above plant protoplasts. The agglutination reactions were found positive by the lectins MCmA, TAA, BMA and Con A with the protoplasts from O. sativa, B. monosperma, T. aestivum and D. carota. MCm A agglutinated M. charantia var. muricata leaf

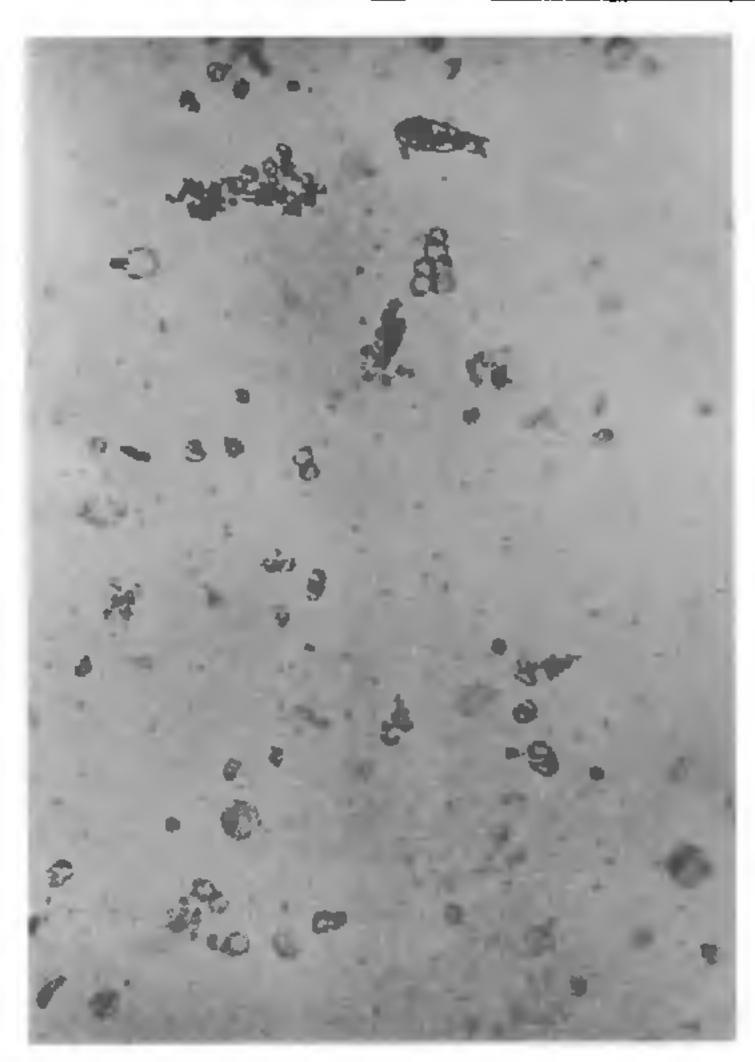


Fig. 1. Agglutination of Momordica charantia varamuricata leaf protoplasts influenced by MCmA lectin.

protoplasts (Fig. 1) but the agglutination was negative in the cases of N. tabecum, C. ensiformis and C. gladiata protoplasts. The agglutination could not be ascertained in the protoplasts of T. anguina and C. indica. Similarly TAA induced the agglutination reaction in the protoplasts of T. anguina and M. charantia var. muricata. No clear confirmation was obtained by TAA in the agglutination of the protoplasts from C. indica, N. tubecum and species of Canavalia. The agglutination reaction was highly influenced by BMA lectin in the protoplasts of T. anguina, N. tabecum, C. ensiformis and C. gladiata. No clear indication of positive agglutination of the protoplasts of M. charantia var. muricata and C. indica was found by BMA. Con A agglutinated the proplasts of N. tabecum, C. ensiformis and C. gladiata. No agglutination was found in cases of the protoplasts from M. charantia var. muricata, C. indica and could not be ascertained in the case of T. anguina. D-galactose was shown to inhibit the agglutination reaction mediated by MCm A, TAA and BMA, but in the case of Con A, D-glucose was effective.

It is possible that non-agglutinating lectins are also capable of binding the protoplast membrane receptors but the interprotoplast due to the excessive distance the attachment between receptors of adjacent cells is not fruitful. The lectin MCm A, TAA, BMA and Con A agglutinated the protoplasts nonspecifically from the same plant tissues as well as others. It appears that plant lectins failed to distinguish between

protoplasts of various sources on the basis of agglutinating nature.

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THE PRODUCTIVITY IN INDUCED MUTANTS OF MOONG BEAN

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In recent years, induced mutants have been directly released as improved varieties in a wide group of crop plants. The 93 registered mutant crop varieties released so far, indicate the increasing popularity of adopting mutation breeding technique for crop improvement. The present study deals with the performance of some of the important mutants in moong bean.

Two inbreds of moong bean (Vigna radiata (L.) Wilczek), viz., Pusa Baisakhi and S-8, obtained from Genetics Division, I.A.R.I., New Delhi, were treated with ethyl methane sulphonate (0.1%, 0.2%, 0.3%), nitroso methyl urea (0.01%, 0.02%, 0.03%), gammarays (20, 40, 60 kR) and their combinations (20 kR gamma-rays +0.1% EMS and 20 kR gamma-rays +0.01% NMU in the variety Pusa Baisakhi only) and directly sown in the field. Seeds from each of the M_I plant were collected on the individual plant

basis and sown in the field in randomized block-single row design to raise the M_2 generation. Similarly M_3 and M_4 generations were raised. Some true breeding mutants, isolated in M_2 generation, were carried over to M_4 generation to study the stability in the superiority of their productivity. The description of these mutants has already been presented². The experiment was designed in replicates of two. The protein content in the grain was determined by modified Kjeldahl's method³. Two essential amino acids namely methionine and tryptophan were analysed by colorimetric methods^{4,5}.

The data, on total grain yield, protein, methionine and tryptophan contents studied in induced mutants are summarised in Table I.

Grain yield per plant

All the mutants were high yielding and the yield per plant ranged from 5.4 (g) to 10.1 (g) in the variety Pusa Baisakhi and 5.7 (g) to 14.0 (g) in variety S-8. This increase in grain yield ranged from 55.5% to 191.4% in the variety Pusa Baisakhi and 0.6% to 146.9% in the variety S-8 as against their respective controls.

Protein Contents

In Pusa Baisakhi, all the mutants except two, showed an increase in protein content ranging from 0.2% to 23.7% whereas in S-8, the increase ranged from 8.5% to 27.3% over their respective controls. It is interesting to find that most of the mutants in both the varieties have shown simultaneous increase in grain yield and protein content as well.

Methionine Content

Out of the 20 mutants, 18 showed an increase in methionine content ranging from 4.5% to 93.0% in Pusa Baisakhi and 48.3% to 135.5% in S-8 compared to their respective controls.

Tryptophan Contents

Only 13 mutants out of the 20 were found with increased tryptophan contents as compared to their respective controls. This increase in the tryptophan contents ranged from 3.9% to 108.4% in Pusa Baisakhi and 9.7% to 22.7% in S-8.

It was interesting to note that some of the mutants, viz., bigger grain size in Pusa Baisakhi, large number of pods per plant in Pusa Baisakhi and S-8 (isolated from 40 kR gamma-ray treatment) and long pod mutants in both the varieties have shown simultaneous increase in grain yield, protein, methionine and typtophan contents. The studies are in progress to establish, at least, a few top ranking mutant strains as varieties in this important pulse ctop.

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