Carnoy's fluid, they were washed gently with distilled water. The styles were dissected and placed on a clean slide having 2 to 3 drops of safranin 0 fast green stain (Safranin 0-150 mg, fast green 250 mg, 45% hot acetic acid 25 ml). The styles were left in the stain for 20 hr and thereafter the excess stain was blotted and cleared in 45% acetic acid for 15 min. The styles were then placed on a drop of glycerine, covered with a cover slip and gently pressed before examination.

The bluish green pollen and pollen tubes could easily be traced in the light red-coloured stigmatic and stylar tissues of greengram (figure 1) whereas in the reciprocal with blackgram as ovule parent, there was no pollen germination and pollen tube growth (figure 2).

The stigmatic exudate provides a portion of the carbohydrate needed for wall biosynthesis by the developing pollen tube[6]. According to Martin[7] pollen germination failure after distant cross-pollinations is probably related to differences in the stigmatic environments such as the size, degree of branching and type of papillae and the composition of stigmatic exudate. Since there is no difference in the stigmatic and stylar morphology between blackgram and greengram plants, the failure of pollen germination may be due to the composition of the stigmatic exudate of the blackgram plant which reacts with substances from the pollen of greengram resulting in the failure of stimulation for germination. The chemicals present in the stigmatic exudate of greengram probably had inhibitory action on the germination of the blackgram pollen. Thus the barrier to successful hybridization between blackgram and greengram appears to exist at the pre-fertilization stage itself.

2 February 1983; Revised 25 April 1983.

1. Buchholz, J., *Stain Technol.*, 1931, **6**, 13.
2. Chandler, C., *Stain Technol.*, 1931, **6**, 13.
3. Dionne, L. A., *Am. Potato J.*, 1958, **35**, 422.
4. Nair, M. K. and Narasimhan, R., *Stain Technol.*, 1963, **38**, 341.
5. Lewis, D. *Sexual incompatibility in plants*, Edward Arnold, London, 1959, 57.
6. Kroch, M., Labarca C. and Loewus. F., In *Pollen development and physiology*, (ed. J. Heslop Harrison), Butterworths, London, 1977, 273.
7. Martin, P. W., In *Pollen development and physiology*, (ed. J. Heslop Harrison), Butterworths, London, 1977, 262.

## NATURE OF SALT INJURY AT GERMINATION STAGE IN PADDY

K. S. GILL* AND S. K. DUTT
*Cental Soil Salinity Research Institute, Karnal 132 201, India.

*Central Soil Salinity Research Institute, Research Station, P.O. Canning Town, Distt. 24 Parganas, West Bengal

SEED germination in paddy under salt stress is limited due to the osmotic stress of the medium[1-5]. However, the mechanism of injury in paddy under salinity at seed germination stage has not been clearly studied so far. Hence the sequential changes were studied to establish the basis of salt injury at the initial stages of germination and early seedling growth

Seeds of two rice varieties *i.e.*, Basmati-370 and NC 1281 (fine and coarse grain respectively) were germinated in varying concentration of salt solution. Three levels of electrical conductivity (EC) *i.e.*, 1.3, 9.2 and 14.3 m mhos/cm were prepared using different dilutions of seawater from Mala river. Experiment was replicated sixteen times. Periodic observations on water absorption, amylase activity[6,7] total soluble protein[8], growth characters and sodium and potassium content in various parts of seed and seedlings from 6 hr to 8 days were recorded.

Water absorption rate was maximum upto 6 hr; there was then a sharp decrease at 24 hr at all salinity levels and at 48–72 hr there was an increase again but the increase was less at higher salinity levels in paddy seed. Delayed induction of seed germination, seed coat rupture and radicle emergence in paddy under salt stress have been reported[1]. This delayed germination was perhaps due to decreased water absorption under salt stress. Total soluble proteins increased with time at all salinity levels but salt and varietal differences were meagre at this stage. $\alpha$-amylase activity increased with time but decreased with increase in salinity (table 1). Interaction effect was non-significant upto 72 hr but effects were significant at 96 hr and thereafter. At 120 hr Basmati-370 had higher enzymatic activity upto EC 9.2 but at EC 14.9, coarse variety NC 1281 had higher activity.

It was interesting to note that at germination stage, much of the sodium of the medium is retained in the seed coat as compared to rice grain (endosperm) at 2 days after sowing. At later stages upto 8 days, also husk contained more sodium while root had higher sodium than shoot and NC 1281 had lower sodium content than Bas-370. Potassium was more in the endosperm than in husk and shoot had more potassium than root and variety NC 1281 had higher potassium than Bas-370. At 8 days of sowing, the root and

TABLE 1

*Effect of salinity on α-amylase activity in rice grain (unit change/mg protein/min) sodium, potassium (Conc. $10^3$ ppm) and dry weight accumulation at 8 days germination stage.*

| EC (m mhos/cm) | Variety | α-amylase activity | | Dry weight | | Sodium | | | | Potassium | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | 96 hr | 120 hr | (mg)/20 seedlings | | | | | | | | | |
| | | | | Root | Shoot | Husk | Rice | Root | Shoot | Husk | Rice | Root | Shoot |
| 1.3 | Bas. 370 | 1.34 | 2.99 | 39 | 48 | 2.73 | 1.70 | 10.8 | 7.41 | 0.50 | 0.74 | 5.3 | 11.8 |
| | NC. 1281 | 1.81 | 2.27 | 60 | 74 | 2.88 | 1.68 | 11.0 | 8.45 | 0.92 | 1.26 | 7.0 | 13.3 |
| 9.2 | Bas.. 370 | 1.04 | 2.27 | 19 | 36 | 6.29 | 2.76 | 14.6 | 8.75 | 0.68 | 0.79 | 7.8 | 26.6 |
| | NC 1281 | 0.72 | 1.88 | 22 | 58 | 4.99 | 2.93 | 15.1 | 8.40 | 0.56 | 1.37 | 10.2 | 28.3 |
| 14.9 | Bas 370 | 0.62 | 1.09 | 11 | 20 | 6.64 | 3.22 | 18.2 | 13.23 | 1.18 | 1.76 | 14.4 | 19.6 |
| | NC 1281 | 0.60 | 1.24 | 22 | 32 | 6.54 | 3.47 | 16.8 | 12.22 | 0.83 | 1.51 | 16.6 | 29.7 |
| CD at 5% | Variety | 0.01 | 0.19 | NS | 13 | NS | NS | NS | NS | NS | 0.85 | 1.15 | NS |
| | Salinity | 0.23 | 0.23 | 56 | 17 | 0.915 | 0.714 | 1.25 | 0.93 | 0.174 | 0.109 | 2.03 | 1.78 |
| | Variety Salinity | 0.03 | 0.03 | NS | NS | NS | NS | NS | NS | NS | 0.239 | 0.174 | NS |

shoot growth as well as their dry weight decreased under salinity in paddy. Variety NC 1281 had higher root and shoot weight than Bas-370. Thus, it is evident that the nature of salt injury in paddy at initial stages may be due to the reduced water absorption into the seed followed by decreased enzyme activity and higher absorption of sodium than potassium into the seed. Decrease in the growth at the initial stages may be due to the higher sodium than potassium in the root than in the shoot, and thus consequent decreased germination and seedling growth under saline conditions.

3 November 1982; Revised 28 February 1983

1. Gill, K. S. and Dutt, S. K., *Indian J. Agric. Sci.*, 1979, **49**, 374.
2. Gill, K. S., Ph.D. Thesis 1981., *Punjab Agric. Univ.*, Ludhiana.
3. Sinha, T. S. and Dutt, S. K., *Curr. Sci.*, 1974, **43**, 518.
4. Patolia, J. S. and Iyengar, E. E. R., *Oryaza*, 1979, **16**, 66.
5. Kaddah, M. T., *Soil Sci.*, 1963, **96**, 105.
6. Chrispels, N. J. and Varner, J. E., *Plant Physiol.*, 1967, **42**, 398.
7. Shuster, L. and Gifford, R H., *Arach. Biochem. Biophys.*, 1962, **96**, 534.
8. Lowery, H O., Roser Brough, N. J., Parrand, A. L. and Randall, R. J., *J. Biol. Chem.*, 1951, **193**, 265.

## INDUCTION OF RESISTANCE AGAINST TOBACCO MOSAIC VIRUS IN DETACHED LEAVES OF *NICOTIANA TABACUM* VAR SAMSUN NN BY LEAF EXTRACT OF *HELIANTHUS ANNUUS*

A. K. JOHARI, R. K. RAIZADA, K. M. SRIVASTAVA AND B. P. SINGH
Plant Virus Laboratory, National Botanical Research Institute, Lucknow 226 001, India.

RECENTLY a few reports dealing with induction of systemic resistance in plants against viruses have appeared[1-4]. Such resistance, however, develops after 24 hr of treatment and does not last for more than a couple of days. The present investigation deals with the effect of antiviral principle present in the leaf extract of *Helianthus annuus* on number of local lesions induced by tobacco mosaic virus (TMV) in detached leaves of *Nicotiana tabacum* var Samsun NN TMV culture was maintained on *N. tabacum* var White Burley and the inoculum was prepared in conventional way using phosphate buffer (pH 7, 0 1 M) as diluent. Extract from *H. annuus* was prepared by macerating the leaves in double-distilled water in a ratio of 1.2(W/V) followed by centrifugation at 3,000 rpm for 5 min. The supernatant, thus obtained, was used for testing its activity against TMV.

To study the effect of leaf extract of *H. annuus* on TMV *in vitro*, the extract was mixed with virus inoculum in equal amounts and the mixture, after an incubation of 30 min, was tested for its activity on *N. tabacum* var Samsun NN. Pre-inoculation treatments were made 15 min, 24 hr and 48 hr before virus-challenge on one half of the detached leaves. Untreated halves of the same leaves were inoculated with virus and served as control. Post-inoculation treatments were similarly made after 15 min and 24 hr of inoculation. In another experiment, the time required to move the active antiviral principle from treated to untreated half of the same leaves was judged by challenge inoculation of TMV at 15 min, 24 hr and 48 hr after treatment. Detached leaves of equal vigour from the same plant were treated with TMV and maintained as control. Detached leaves of all treatments were kept in petridishes containing glass-wool soaked with distilled water Lesions were counted after three days of virus inoculation and the percent inhibition was calculated by the following formula:

$$(C-T \times 100)/C$$

Where $C$ = lesion number in control samples and $T$ = lesion number in treated samples.

*H. annuus* leaf extract caused negligible reduction (only 8%) in a number of local lesions induced by TMV when it was mixed *in vitro* with clarified sap containing TMV. However, as apparent from the data in table 1 the inhibition of TMV in the treated half of the detached leaves enhanced with increase in lapse of time between treatment and challenge inoculation. While negligible inhibition was noted when the time between application of leaf extract and inoculation was 15 min, a high percentage of inhibition (85%) was recorded when there was a gap of 48 hr between treatment and challenge of the virus. Leaf extract of *H. annuus* when rubbed on detached leaves after 15 min and 24 hr of virus inoculation, however, could not appreciably effect the number of local lesions induced by TMV. The inhibition, if any, obtained in such a case was always below 6%.

Data compiled in table 2 reveal that the antiviral principle present in leaf extract of *H. annuus* could move from treated to untreated half of the same leaf. However, the time required for movement sufficient to