

growth/nutritional deficiency/rise in temperature/non availability of water and pest infestation. The increase in yield due to application of *Azolla* and N fertilizer in Kalinga-2 (Table II) was more in Rabi in comparison with CR-1005 in Kharif where there was a little increase. The response towards the application of 10-12 tons fresh *Azolla* ha containing about 20-25 kg N was similar to the application of 30 kg N/ha (ammonium sulphate) and increasing the amount of *Azolla* increased the yield (Tables II and III). The application of 30 and 50 kg N/ha through the ammonium sulphate and 10-12 tons of *Azolla* could increase the yield almost similar to the application of 50-60 and 70-80 kg N/ha through the ammonium sulphate. The increase in straw yield also followed the similar trend.

The shortage and high price of N fertilizers as well as the fear of pollution due to their excess use, initiated the research on alternative technology where utilization of synthetic nitrogen fertilizers could be minimized. The *Azolla* is one of them which will help in reducing the requirement of N fertilizers besides providing organic matter, due to its rapid multiplication (doubling time of 2-4 days under ideal conditions) by utilizing atmospheric nitrogen as nitrogen source. One of the reasons of its rapid multiplication is the occurrence of both photosynthesis and nitrogen fixation in the leaves. The multiplication of the fern was found to be rapid in soils of pH 5.8 to 7.5 to highly acidic or highly alkaline soils. The increase in the yield due to the utilization of *Azolla* is reported upto 40% in different observations<sup>9</sup>. The studies conducted at Central Rice Research Institute also showed significant increase in yield of different high yielding varieties of rice<sup>1-4</sup>.

The present study give rise to the following conclusions :

1. Response towards *Azolla* was comparatively better in short duration varieties (Supriya and Kalinga-2) than the medium duration varieties (like IR-8 and CR-1005). The medium duration varieties probably received more biologically fixed nitrogen due to longer submergence of fields.
2. The response towards *Azolla* and N fertilizer was comparatively better in Rabi than Kharif.
3. The effect of *Azolla* applied on N basis was comparable to the effect of the application of chemical N fertilizer.
4. *Azolla* and chemical N fertilizer could be used together for higher benefit.
5. The unincorporated *Azolla* also increased the yield significantly.

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#### OCCURRENCE OF A COMPLEX VIRAL INFECTION ON *ALTHAEA ROSEA* L.

SYMPTOMS produced by whitefly-transmitted agents have been categorized into mosaic, leaf curl and yellows types<sup>1</sup>. For each of the symptoms different agents are involved. Since, occasionally whiteflies are known to carry more than one virus at a time<sup>2</sup>, a disease transmitted by them could not be interpreted as to be caused by a single agent. Cotton leaf curl in Africa, described for Egyptian cotton (*Gossypium barbadense*), was reported to induce yellow mosaic symptoms when transmitted to *G. hirsutum* varieties<sup>3</sup>. Later work<sup>4</sup> indicated the presence of mixed infection with the two whitefly-transmitted diseases.

During the year 1975, many of the hollyhock plants grown at different places within National Botanic Gardens, Lucknow, showed severe vein yellowing symptoms accompanied with leaf curling and dark green vein thickening which rarely bursts into leafy enations. Deformities were easily observed on the underside of the leaves which included cupping of the leaves, thickening and zig-zag twisting of the veins. Severe vein yellowing could, however, be observed both on the upper and lower surfaces of the leaf.

An observation of the infected and healthy plants throughout the season, till flowering, showed that the plants, which developed only vein yellowing, grew to a smaller height than the normal with a slight reduction in leaf lamina and produced comparatively faded flowers. The plants showing cupping, leaf curling and

thickening of the veins were considerably reduced in size and produced none or only a few small flowers as compared to healthy plants. Severity of the disease in plants, showing both types of symptoms, was enhanced resulting in extreme dwarfing, cupping of the leaves and zig-zag twisting of the swollen veins. The plants neither grew to a size more than 10 inches in height nor produced any flowers. However, the severity of symptoms was completely dependant on the time of infection of each virus at different stages of the plant growth.

Since whiteflies are able to carry more than one virus at a time, whiteflies collected from field-grown *Nicotiana tabacum* plants were multiplied on a series of plant species consisting of *Nicotiana tabacum*, *Phaseolus vulgaris*, *Nicotiana rustica*, *Glycine max*, *Phaseolus aureus*, *Lycopersicon esculentum*, *Zinnia elegans*, *Althaea rosea*, and *Clitoria ternatea*. Transfer of generations of whiteflies to such a series of host plants made them completely virus-free. None of the tests done with whiteflies grown on *Clitoria ternatea* produced any symptoms on the aforesaid test plants. Non-viruliferous whiteflies thus multiplied were allowed to feed on diseased plants of hollyhock showing both leaf curl and vein yellowing symptoms. The handling of the whiteflies during transmission test was carried out using a method<sup>5</sup> described earlier. Viruliferous whiteflies were transferred at a rate of 20 per test plant. In each test, at least five plants of one species were used.

Data included in Table I showed that only 2 plant species showed vein yellowing symptoms while five

others produced only leaf curl accompanied by vein thickening. *Althaea rosea*, however, developed both vein yellowing and leaf curl symptoms. Back inoculation tests from infected plants to *Althaea rosea* produced either only vein yellowing or leaf curl and vein thickening type of symptoms (Fig. 1). It is, therefore, inferred that *Abelmoschus esculentus* can be used to separate vein yellowing agent while several species of Solanaceae mentioned in Table I can be a tool for separating leaf curl agent from the doubly infected plants.

TABLE I

Transmission of a complex infection of *Althaea rosea* to other hosts through whiteflies

Hosts	Symptoms	Incubation Period in host	Back Inoculation on <i>Althaea rosea</i>
<i>Ageratum conyzoides</i>	..	NA	..
<i>Abelmoschus esculentus</i>	VY	18-22 Days	VY
<i>A. moschatus</i>	VY	20-25 Days	VY
<i>Althaea rosea</i>	LC, VY, VT	25-30 Days	LC, VT, VY
<i>Capsicum annuum</i>	LC, VT	15-20 Days	LC, VT
<i>Carica papaya</i>	LC, VT	25-30 Days	—
<i>Cucumis sativus</i>	—	NA	—
<i>Dolichos lablab</i>	—	NA	—
<i>Lycopersicon esculentum</i>	LC, VT	15-20 Days	LC, VT
<i>Nicotiana rustica</i>	LC, VT	18-22 Days	LC, VT
<i>N. tabacum</i>	LC, VT	20-22 Days	LC, VT

— = No symptoms,  
LC = Leaf Curling,  
NA = Not applicable,  
VT = Vein thickening,  
VY = Vein yellowing.

Susceptibility of the different hosts tested, indicates the involvement of hibiscus vein yellowing<sup>6</sup> and tobacco leaf curl<sup>7</sup> viruses in complex infection of *Althaea rosea* plants. This is being reported for the first time along with the separation of individual virus from the mixture using hosts as filter.

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FIG. 1. View of leaf portions of *Althaea rosea*.  
Top left = Healthy  
Top right = Vein yellowing only.  
Bottom left = Vein yellowing + enations  
Bottom right = Swollen veins showing yellowing accompanied with leaf cupping.

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### FREE PROLINE ACCUMULATION AND REDUCTION IN RWC UNDER MOISTURE STRESS IN GENOTYPES OF SAFFLOWER

SCREENING of varieties for relative drought tolerance is of practical significance in that it helps in recommending the variety directly for cultivation under rainfed conditions. Several laboratory tests have been developed and used. A few studies show a correlation between performance in laboratory tests and actual field trial. Accumulation of free proline in leaves appears to be an adaptive mechanism for drought tolerance. A relationship between proline accumulation under stress and yield stability index has been shown in barley genotypes (Singh *et al.*<sup>1</sup>, 1972). Such a relationship has been established (Mehkri *et al.*<sup>2</sup>, 1976) in groundnut genotypes also. Decrease in relative water content (RWC) in the leaf under a given stress is another parameter (Barrs<sup>3</sup>, 1968). Using these two parameters, differences in safflower genotypes were studied as a measure of drought tolerance.

Six genotypes of safflower SF-11, 143-20, S-2, C-438, A-1 and 16-4-P<sub>2</sub>-2 were grown in a single pot

(18 × 12 × 12" size). One month old seedlings were subjected to stress by withholding water for 5 days. Determinations of proline and RWC were done on 4th and 5th day after withholding water and continued up to three days after rewatering. Rewatering was done after 5th day. Since the genotypes were raised in the same pot and subjected to stress, the attributes on proline accumulation and RWC are directly comparable between genotypes.

Free proline in the leaves was determined using the method of Bates *et al.*<sup>4</sup> (1973), and RWC by the method of Barrs and Weatherly<sup>5</sup> (1976).

The data in Table 1 indicated that free proline increased in the leaves of all genotypes under stress. But the magnitude of the increase differed among genotypes. The variety 143-20 showed a 35.9 fold increase in proline over the control followed by the variety S-2. The varieties A-1 and 16-4-P<sub>2</sub>-2 showed relatively lesser increase in proline under stress. However, the absolute amounts of proline when compared among varieties indicated that SF-11 had the highest under peak stress followed by 143-20. Varieties A-1 and 16-4-P<sub>2</sub>-2 were still low in the absolute amount also. On rewatering the decrease in proline was faster in SF-11 and 143-20 compared to other varieties (Fig. 1). This aspect could be a measure of the resumption of the metabolic activity following rewatering. Under conditions of intermittent moisture stress this point is of importance. Differences amongst genotypes in free proline accumulation have been noticed by other workers also in sorghum (Sinha and Rajagopal<sup>6</sup>, 1975).

The data on the reduction of RWC under stress bears a significant relationship with the ability to accumulate proline. The varieties 143-20 and S-2 which showed

TABLE I

Variety	Proline			RWC		
	Control	Stress 120 hr after	No. of times increase over Control	Control	Stress 120 hr after	% reduction on stress
SF-11	102	2620	25.7	86.5	42.7	50.6
143-20	68	2445	35.9	84.3	45.8	45.6
S-2	70	2260	32.0	85.0	43.1	49.3
C-438	82	2015	24.5	90.2	41.4	54.1
A-1	81	1645	20.3	87.4	42.0	51.9
16-4-P <sub>2</sub> -2	58	1307	22.5	88.0	42.7	51.5

Free proline (g/gm fresh wt.) and RWC in genotypes of safflower 120 hours after withholding water. All the genotypes were raised in the same pot. Control values represent data collected before withholding water.