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## RESPONSE OF *WESTIELLOPSIS PROLIFICA* TO SALT-STRESS II. UPTAKE OF $Na^+$ IN THE PRESENCE OF $K^+$ AS CHLORIDE, NITRATE AND PHOSPHATE

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Most of the higher plants react to salt-stress either through osmotic adjustments[1] by sequestering the inorganic ions[2], or secretion of excess polysaccharides[3]. Indications of increased nitrogen demand and accumulation of $K^+$ by certain algae under salt-stress are also available. Cyanobacterial response to salt-stress, is, however, little understood. We have therefore studied the physiological aspects of cyano-bacterial adaptation to salts[4]. In this report we examine the influence of $K^+$ as KCl, $KNO_3$ and $KH_2PO_4$ on the uptake of $Na^+$ and $K^+$ by cyanobacteria.

*Westiellopsis prolifica* ARM 366 was grown in BG-11 medium[5] devoid of combined nitrogen at $30 \pm 1$°C under continuous illumination (2400 lux). $Na^+$ uptake was examined at a fixed concentration of $Na^+$ as 400 mM NaCl which is equivalent to 23.4 mg NaCl/ml. $K^+$ at graded concentrations of 20, 50, 100, 150 and 200 $\mu$g/ml was added as KCl, $KNO_3$ and $KH_2PO_4$ and their influence was examined on growth, pigments (chlorophyll)[6], nitrogen fixation[7] and uptake of $Na^+$ and $K^+$. To estimate $Na^+$ and $K^+$, the pellet was dried, ashed in a muffle furnace for 15 min at 500°C and dissolved in 0.1 M $HNO_3$ and estimated in flame photometer[8], using a digital flamephotometer (Elico).

Supplementation of growth medium with KCl or $KH_2PO_4$ was not effective to overcome the salt-stress and the lowered chlorophyll level remained unaffected. A marginal improvement in chlorophyll content was, however, observed due to $NO_3^-$, suggesting that $NO_3^-$ although partially effective, was not sufficient to overcome the salinity stress. The biomass turnover was slightly higher (up to 12%) in the presence of KCl and $KNO_3$ but not in $KH_2PO_4$.

**Table 1** *Influence of $K^+$ as $Cl^-$, $NO_3^-$ and $PO_4^-$ on the growth and acetylene reduction activity of Westiellopsis prolifica in presence of 23.4 mg NaCl/ml*

| Treatment | Chlorophyll $\mu$g/ml | Dry weight mg/ml | $\mu$mol $C_2H_4$ mg/chl/h |
|---|---|---|---|
| BG-11 | 5.43 | 5.93 | 1.91 |
| NaCl | 0.58 | 0.51 | 2.85 |
| + KCl $\mu$g/ml | | | |
| 20 | 0.55 | 0.52 | 2.06 |
| 50 | 0.55 | 0.49 | 2.18 |
| 100 | 0.53 | 0.55 | 2.89 |
| 150 | 0.55 | 0.50 | 2.78 |
| 200 | 0.56 | 0.56 | 2.73 |
| + $KNO_3$ $\mu$g/ml | | | |
| 20 | 0.66 | 0.56 | 3.4 |
| 50 | 0.61 | 0.51 | 3.7 |
| 100 | 0.59 | 0.54 | 2.9 |
| 150 | 0.68 | 0.57 | 3.3 |
| 200 | 0.63 | 0.53 | 3.6 |
| + $KH_2PO_4$ $\mu$g/ml | | | |
| 20 | 0.56 | 0.44 | 3.09 |
| 50 | 0 60 | 0.48 | 3.70 |
| 100 | 0.54 | 0.45 | 3.20 |
| 150 | 0.60 | 0.50 | 3.70 |
| 200 | 0 58 | 0.48 | 3.90 |

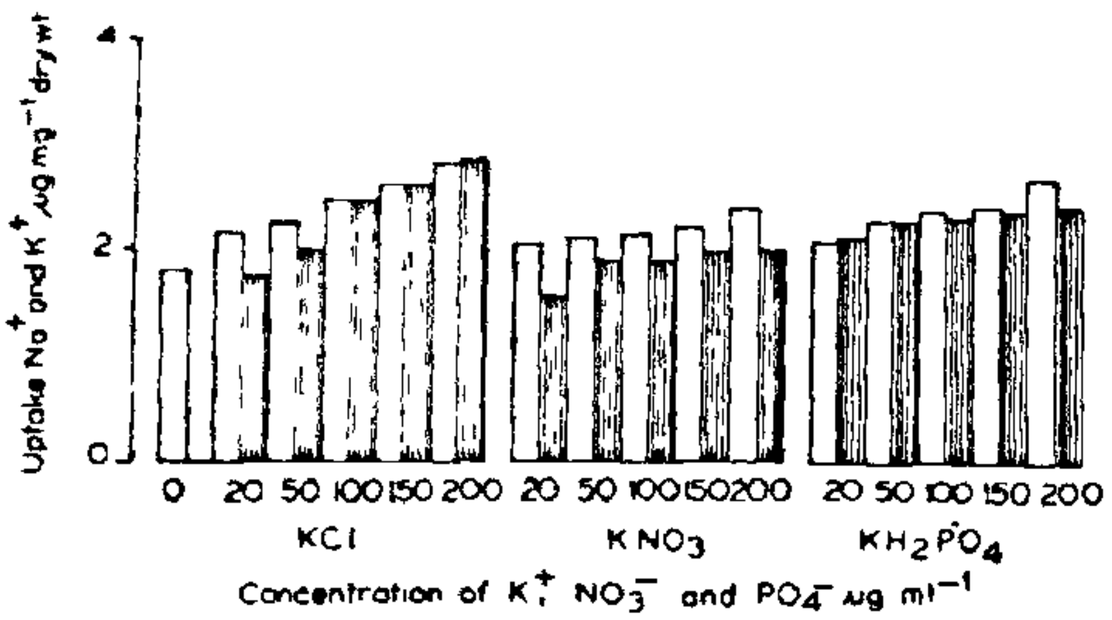


**Figure 1.** Uptake of $Na^+$ by *W. prolifica* in presence of fixed concentration of NaCl (23.4 mg NaCl/ml) and graded concentrations of K (20, 50, 100, 150, 200 $\mu$g/ml) as KCl, $KNO_3$ and $KH_2PO_4$.

Nitrogenase activity measured as acetylene reduction activity was higher in the presence of NaCl-stress. Supplementation of the growth medium with $K^+$ as salts of chloride, nitrate and phosphates showed that $K^+$ as KCl was either inhibitory or neutral while $NO_3^-$ and $PO_4^-$ were stimulatory. These results indicate that nutrients like nitrogen and phosphorus are limited under salt stress. Many cyanobacterial strains have been found to grow and tolerate high levels of salinity. These include halophilic forms such as *Microcoleus chthonoplastes* (20–25% NaCl)[9], *Spirulina subsala* (> 3 M NaCl)[10], *Calothrix scopulorum* (5% NaCl)[11], *Anabaena torulosa*[12] and euryhaline forms which could grow in freshwater and in varying degrees of salinity[13] and increased nitrogen fixation by salt adapted *Nostoc muscorum* has also been reported[14].

$Na^+$ uptake in the presence of fixed level of NaCl and varying concentrations of KCl, $KNO_3$ and $KH_2PO_4$ was a concentration-dependent phenomenon up to 200 $\mu$g/ml KCl and not in the presence of $KNO_3$ and $KH_2PO_4$ (figure 1). *W. prolifica* maintained a low internal $Na^+$ concentration (2.05 to 2.8 $\mu$g/ml dry weight of alga) and this uptake seems to be related partially with $K^+$. *Anabaena torulosa* showed an increased nitrogen demand under salt-stress and a similar response was also observed with *W. prolifica* where external supply of combined nitrogen could partially improve the level of chlorophyll (table 1). These results show that partial recovery in chlorophyll content is possible by supplying nitrogen as $KNO_3$ and $Na^+$ uptake is influenced by the presence of $K^+$ in the salty environment.

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## SOMATIC HYBRIDIZATION ATTEMPTS BETWEEN *SORGHUM BICOLOR* (L.) MOENCH AND *ORYZA SATIVA* L.

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PROTOPLAST fusion is widely used for transferring nuclear and cytoplasmic traits between sexually incompatible species. Somatic hybridization in the gramineae has proved difficult but varying degrees of success could be achieved ranging from the identification of heterokaryons in fusion between sorghum and maize[1] to that of successful plant regeneration from somatic hybrids of rice and