

reduction in germination for 1 and 2 h was 40 and 50% in untreated-exposed while in treated-exposed it was 30 and 40%. After 48 h there was a slight increase in seed germination of untreated-exposed and the reduction was 40 and 48% whereas in treated-exposed sets it was 25 and 38%. The protease enzyme activity which hydrolyses storage protein and helps in seedling growth, was less in untreated-exposed sets as compared to treated-exposed sets. The above findings show that ascorbic acid mitigates the adverse effects of SO₂ on seed germination and on protease activity. The protein content in cotyledons of exposed sets was higher, showing that hydrolysis was reduced due to SO₂. As the exposure period increased the negative effects of the SO₂ increased (table 1). The ascorbic acid content was greater in treated sets indicating that it has been taken directly by the seedling and therefore has an important role in plant metabolic pathways and acts as a reductant⁷. Sulphur dioxide accumulates in plants as SO₃²⁻ and ferredoxin reductase system is required for the photoreduction to sulphide¹⁴. The excess of sulphur accumulated in plant as sulphate and sulphite which causes injury is volatilized from the leaves as H₂S and this reaction is light-dependent. Light is needed for the supply of reductant¹⁵. Ascorbic acid acts as a powerful reductant and seems to mediate the reduction of sulphite to H₂S thereby reducing the toxicity of SO₂⁸. In the present study the sulphite accumulated in the treated-exposed seeds might have converted into H₂S using ascorbic acid as a reductant thereby reducing the toxicity of SO₂. The lower increase in the seedling sulphur content of treated-exposed sets supports this assumption. Further investigation is necessary to understand the resistance of plants against air pollution.

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CHARACTERIZATION OF A STREPTOMYCIN-RESISTANT MUTANT OF *SCENEDESMUS DIMORPHUS* (TURP.) KUETZ

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STREPTOMYCIN has been used in algae mainly to study its effect on chlorophyll formation and in isolation of apochlorotic races of *Euglena*^{1,2}. Ebringer^{3,4} attributed inhibition of chlorophyll synthesis to the chemical structure of streptomycin in which the carbohydrate molecule contains glycolate linkages and a large number of hydrophilic groups (amino or guanidine groups)⁵.

A few reports⁶⁻¹⁰ are on record, pertaining to the effect of streptomycin on pigment production in algae. The present study incorporates biochemical analysis of chlorophyll pigments quantitatively and the qualitative analysis of free and protein bound amino acids of streptomycin-resistant mutant of *S. dimorphus*.

A wide range of concentrations 0.05 to 2 mg of streptomycin per 100 ml of the medium were used but 1 mg/100 ml proved lethal¹¹. Cultures from sub-lethal concentration (0.5 mg/100 ml) were sub-cultured several times in fresh nutritive medium and used for analysis. Chlorophyll pigments were

extracted from fresh sample in 80% acetone and their quantity was calculated with the help of Parsons and Stricklands's formulae¹².

The free and bound aminoacids were analysed by unidirectional ascending paper chromatography.

Absorption spectra of chlorophyll pigments of control as well as treated samples showed two major peaks at 420 and 660 nm (figure 1). The first peak is said to be due to chlorophyll *a*¹³. But according to Kumar⁶, it represents chlorophyll *a* and carotenoids both. While the second peak was essentially due to chlorophyll *b*^{6,13,14}. In the treated samples, the optical density values were markedly lowered at both these wavelengths. An additional shoulder peak was seen at 600 nm in the streptomycin-treated sample, perhaps due to another form of chlorophyll *b*¹³. This peak has also been recorded in *Chlorococcales*⁸ and *Desmids*¹⁰ treated with streptomycin. But it still awaits its identification. Work on this line is in progress in our laboratory.

This antibiotic had more pronounced effect on chlorophyll *a* than on chlorophyll *b*; even the total chlorophyll content was also very much reduced. In normal cultures, chlorophyll *a* and chlorophyll *b* contents were estimated to be 8.721 and 3.703 mg/500 mg fresh weight. While in treated samples the values of chlorophyll *a* and chlorophyll *b* were 2.1095 and 1.7950 mg/500 mg fresh weight respectively. With this, chlorophyll *a* and chlorophyll *b*

contents were lowered by 75.81% and 51.52% respectively in the treated culture samples and the total chlorophyll contents were reduced by 68.57% when compared with the normal ones. This observation further gets its support from previous reports⁸⁻¹⁰. The loss of pigment colour might be due to the conversion of some precursor of either the pigment or the plastid by the drug². While according to Ebringer^{3,4} this antibiotic contains aminosugar moieties in their molecule which interfere with the metabolism of ribose and deoxyribose, contained in the nucleic acids of the chloroplast and this resulted in the bleaching of the cells.

Qualitative analysis of free and protein-bound amino-acids of controlled and streptomycin-treated samples yielded quite interesting variations. In normal samples, the fifteen amino-acids registered were alanine, L-arginine, aspartic acid, DL-2-amino-*n*-butyric acid, L-cysteine, cystine, glutamic acid, isoleucine, lysine, DL-methionine, phenylalanine, valine and three unidentified ones whose *R_f*

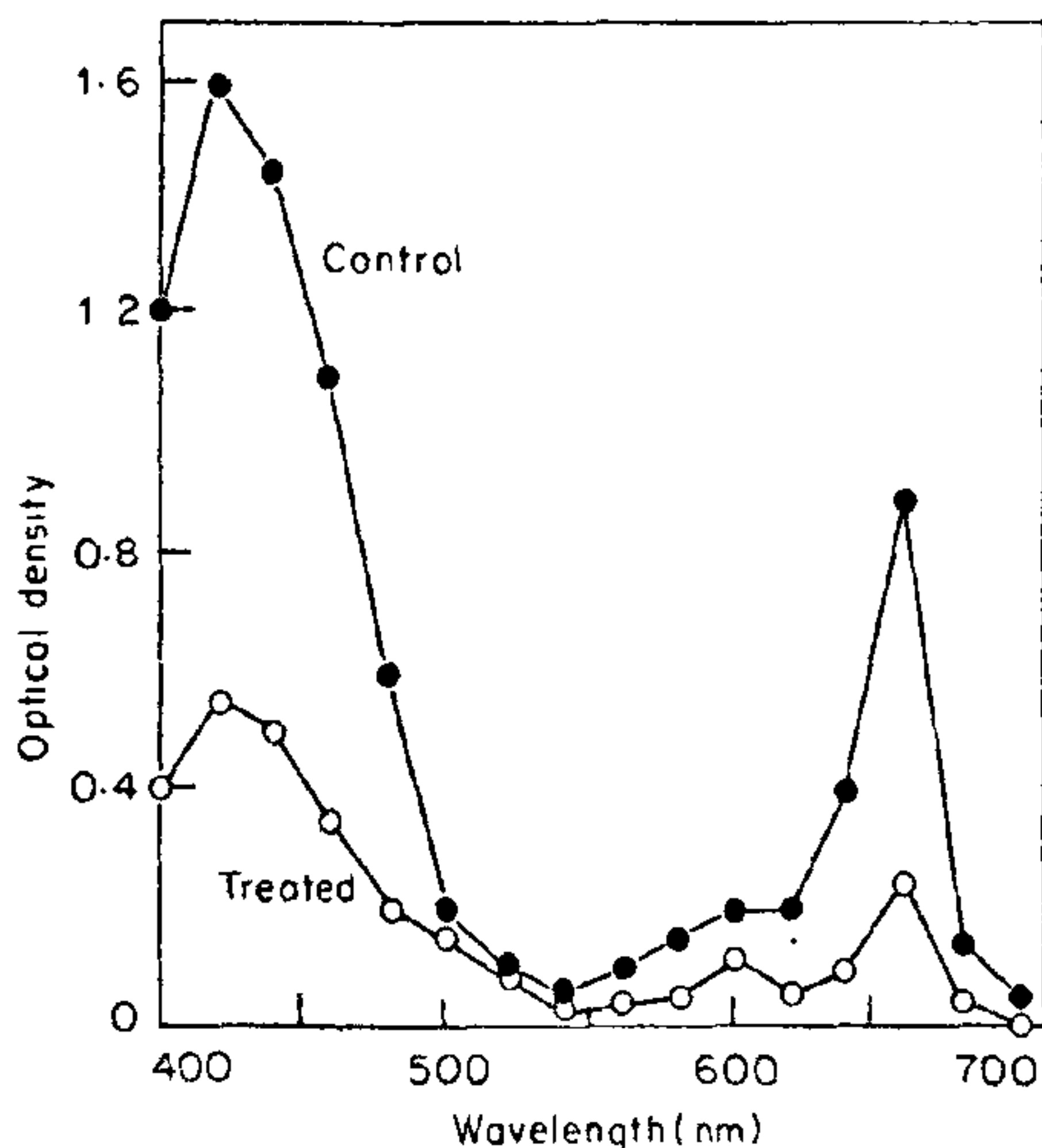


Figure 1. Absorption spectra of chlorophyll pigments of *S. dimorphus* after streptomycin treat-

Table 1 Presence of free and bound amino acids in streptomycin-resistant mutant of *S. dimorphus*

Name of the amino acid	Control		Treated	
	Free	Bound	Free	Bound
Alanine	+	-	-	+
L-Arginine	-	+	+	+
Aspartic acid	+	-	-	+
DL-2-amino- <i>n</i> -butyric acid	-	+	-	-
Citrulline	-	-	-	-
L-Cysteine	-	+	+	+
Cystine	-	+	+	-
Glutamic acid	-	+	+	-
Glycine	-	-	-	+
Histidine	-	-	-	+
Isoleucine	-	+	-	+
L-leucine	-	-	-	-
Lysine	+	-	+	-
DL-Methionine	+	-	+	-
DL-ornithine	-	-	-	-
Phenylalanine	-	+	-	-
Serine	-	-	-	+
Threonine	-	-	-	-
Tryptophan	-	-	-	-
Tyrosine	-	-	+	-
Valine	-	+	-	+
Proline	-	-	+	+
Unidentified	-	+	-	-
Unidentified	-	+	-	-
Unidentified	-	+	-	-
Unidentified	-	-	-	+

values were very high (table 1). While in antibiotic-treated samples, the number of amino-acids was enhanced by one and the newly expressed amino-acids were glycine, histidine, serine, tyrosine, proline and an unidentified one which was different from those obtained in the untreated sample due to its varied R_f value. Amino-acids which got completely suppressed by streptomycin were DL-2-amino-*n*-butyric acid, phenylalanine and three unidentified ones.

These observations revealed that streptomycin had a promoting effect as far as the number of amino-acids was concerned. However, certain amino-acids were very sensitive to the drug. It may be inferred that streptomycin induced changes in both free and bound amino-acids of the algal samples under study, which may be attributed to the effect *per se* as well as enzymatic breakdown of algal proteins into amino-acids.

Streptomycin might have brought these biochemical variations by affecting the sample at genetic level because these changes persisted even after several subculturing in the fresh nutritive medium.

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EFFECT OF ALTERNARIA BLIGHT ON OIL CONTENT OF RAPE-SEED AND MUSTARD

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ALTERNARIA blight caused by *Alternaria brassicae* (Berk.) Sacc. is a serious disease of rape-seed and mustard. All aerial parts of the plants including pods are attacked and the yield is greatly reduced¹. Nijhawan and Hussain² analysed the seeds of rape-seed from healthy plants and plants infected with *A. brassicae* for their chemical constituents, and demonstrated that oil content of seeds from diseased plants was reduced. Since then many new cultivars have been introduced and recommended to growers and are grown on commercial scale. Of late, the disease has assumed greater significance in India in view of the importance of oilseed production in the national economy. The present study was undertaken to examine the possible effect of the disease on the oil content of seeds of a number of recommended cultivars of rape-seed and mustard which are grown on commercial scale for oil extraction.

Samples of seeds of 8 recommended cultivars of rape-seed (*Brassica campestris* L.) and 9 of mustard [*B. juncea* (L.) Czern and Coss.] were collected from plants heavily infected with *A. brassicae* as well as from disease-free plants separately for each cultivar and analysed for oil content. The oil content was estimated by the cold percolation method³.

The results indicate that the disease caused reduction in the oil content of the seeds to varying extents in all the cultivars of rape-seed and mustard included in the study. Variations to this effect were observed between the cultivars of rape-seed and mustard. Greatest loss of oil content was recorded in YST-151, T-42 and K-88, the rape-seed cultivars. The loss in oil content of the seeds from diseased plants of rape-seed cultivars over the seeds from healthy plants ranged between 14.58 and 35.97%. The highest loss (35.97%) was observed in T-42. The range of loss in oil content of seeds from diseased plants of mustard cultivars over the seeds