**Table 1** *Specific activities of key enzymes of carbohydrate metabolism in cell-free extracts of strains of Azotobacter and Azomonas*

| Strain | ED enzymes | 6PGD | PFK | FDA | GL3PD | ISDH |
|---|---|---|---|---|---|---|
| *Azotobacter chroococcum* $BI_1$ | 3 | 27 | 30 | 5 | 375 | 3143 |
| *Azotobacter chroococcum* $BI_2$ | 3 | 6 | 2 | 9 | 181 | 1319 |
| *Azotobacter vinelandii* 2821 | 4 | 354 | 32 | 5 | 699 | 3055 |
| *Azotobacter vinelandii* Ka | 5 | 21 | 96 | 8 | 429 | 295 |
| *Azomonas agilis* 2819 | 127 | 195 | 7 | 45 | 601 | 1179 |
| *Azomonas macrocytogenes* 2454 | 27 | 40 | 107 | 25 | 38 | 845 |

Specific activities expressed as nmol of product formed per minute per mg of protein.

strains studied suggests an operational TCA cycle in all the strains. Activity of NADP-6-phosphogluconate dehydrogenase, the key enzyme of the PP pathway, was also detected in all the strains.

On the basis of the activity of the ED enzymes and of FDA it is possible to distinguish between the strains of *Azotobacter* and *Azomonas*. This separation based on enzymatic determination is consistent with the classification of De Smedt *et al*[16], which is based on rRNA cistron homology, for these strains.

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## VISCOMETRIC STUDIES ON THE EFFECT OF NEAR-UV LIGHT IRRADIATION ON DNA IN PRESENCE OF CHLORPROMAZINE

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CHLORPROMAZINE (CPZ), a phenothiazine drug, is now widely used as an antidepressant tranquilizer. The drug is highly photosensitive. It has some effects on patients who use it continuously[1,2]. Dark incubation of human red blood cell (RBC) with this drug (concentration $3 \times 10^{-4}$ M) causes haemolysis of the RBC[3]. Near-UV light (365 nm) irradiation of RBC in the presence of relatively low concentration of the drug causes extensive haemolysis[3]. Photomutagenesis in presence of CPZ has been reported in Chinese hamster cells[4] and in *Salmonella typhimurium*[5]. Photoinduced inactivation of adenovirus[6] and bacteriophage[7] has also been observed. In this paper we report the results of viscometric studies on the effect of near-UV light (365 nm) irradiation on DNA in presence of CPZ.

CPZ, as CPZ hydrochloride (made in the USSR), was obtained from Sun Pharmaceutical Industries, Vapi, Gujarat, India. A stock solution of CPZ, 1 mg/ml in 0.01 M NaCl, was freshly prepared before each experiment. The absorption spectrum of CPZ solution (19 $\mu$g/ml) shows two absorption maxima, one at $\lambda = 255$ nm, with molar extinction coefficient

$\varepsilon = 30{,}680\ M^{-1}\ cm^{-1}$, and the other at $\lambda = 305$ nm, with $\varepsilon = 3900\ M^{-1}\ cm^{-1}$. The spectrum was recorded on a Carl Zeiss PMQ II spectrophotometer.

Highly polymerized calf thymus DNA (Sigma Chemical Co., USA) at a concentration of 1 mg/ml was allowed to disperse thoroughly in 0.01 M NaCl for 5 days at 4°C. This was used as stock solution. Stock DNA diluted to a concentration of 50 $\mu$g/ml gave an optical density (OD) of 1.0 at 260 nm. DNA–CPZ mixture was prepared 10–20 min before irradiation.

Near-UV light (365 nm) irradiation of DNA, CPZ or DNA–CPZ mixture was done using a Phillips HPW125 lamp. Sample (2–3 ml) to be irradiated was taken in a 5 cm diameter glass petri dish and kept at a distance of 25 cm from the lamp.

Viscometric measurements were made with a Canon Manning Semimicro Viscometer type 75 A500 (Canon Instrument Co., USA) in a constant temperature bath maintained at $35 \pm 0.1$°C. Viscosity was expressed as the ratio of the specific viscosity ($\eta'_{sp}$) of the treated or irradiated sample to that of the corresponding reference (untreated or unirradiated) sample ($\eta_{sp}$).

Incubation of DNA with different concentrations of CPZ (25 to 100 $\mu$g/ml) at 37°C for one hour in the dark did not cause changes in the viscosity of the DNA, compared to control untreated DNA. Irradiation of DNA with near-UV light for 45 min in absence of the drug also did not cause change in viscosity.

However, DNA when irradiated by near-UV light in the presence of different concentrations of CPZ showed a linear decrease in viscosity (figure 1). A similar fall in viscosity of the DNA was observed when DNA in the presence of a fixed concentration of CPZ was irradiated for different periods of time (figure 2). This fall in viscosity of DNA possibly indicates degradation and denaturation of DNA. The presence of CPZ in the DNA sample during irradiation seems to be obligatory for such photoinduced changes in DNA.

Visible-light induced photodegradation of DNA in the presence of acridine dye and acridine orange has also been observed[8]. Production of singlet oxygen during irradiation was thought to be one of the factors responsible[9]. We therefore tried to investigate whether singlet oxygen might cause the photoinduced degradation of DNA (50 $\mu$g/ml) in the presence of CPZ (100 $\mu$g/ml). DNA–CPZ mixture was irradiated with near-UV light for 45 min in the presence of $10^{-3}$ M sodium azide ($NaN_3$), which quenches singlet oxygen[10]. We observed no significant difference in the extent of fall in viscosity of DNA ($\eta'_{sp}/\eta_{sp} = 0.39$) compared to the sample irradiated in the absence of azide.

The results showed that singlet oxygen, if produced during irradiation, was not responsible for

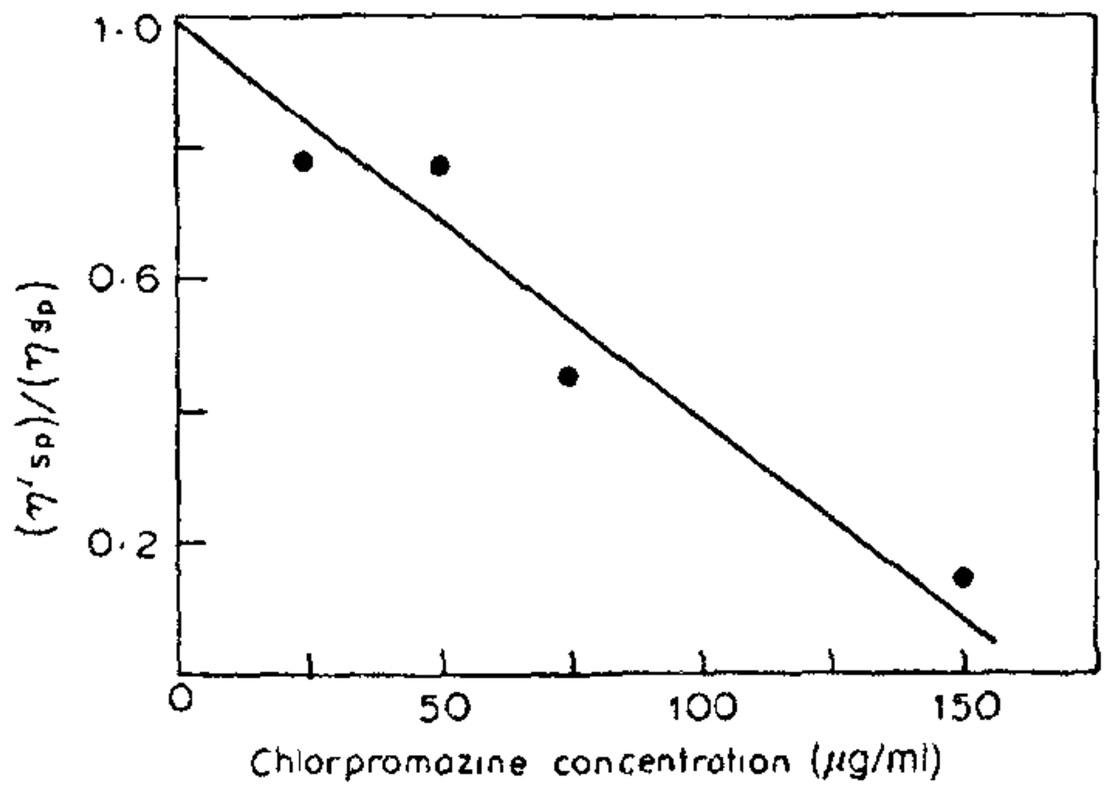



**Figure 1.** Decrease in viscosity of DNA (50 $\mu$g/ml) irradiated with near-UV light for 45 min in presence of different concentrations of CPZ.

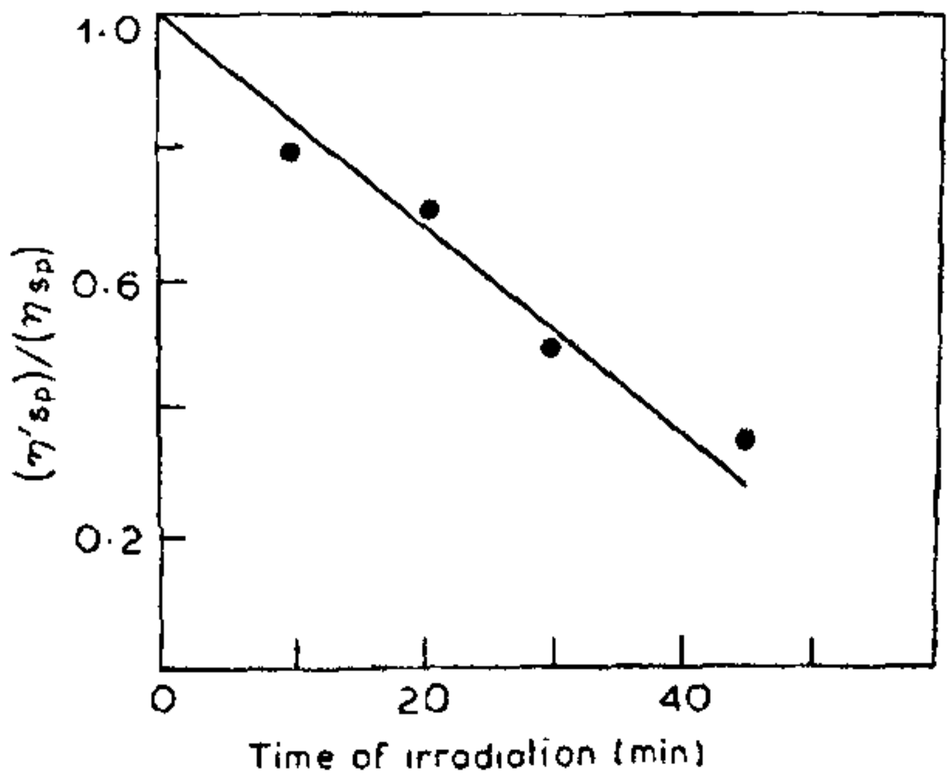
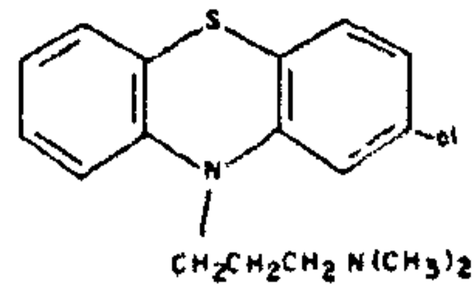



**Figure 2.** Decrease in viscosity of DNA (100 $\mu$g/ml) irradiated by near-UV light for different periods of time in presence of CPZ (100 $\mu$g/ml).




Structure of chlorpromazine

the near-UV-light induced degradation of DNA in presence of CPZ.

If, however, near-UV-light irradiated CPZ (45 min irradiation) was added to unirradiated DNA (final concentrations DNA 50 $\mu$g/ml, CPZ 100 $\mu$g/ml), a fall in viscosity of the DNA was observed up to 10 min after addition of the irradiated drug. After 10 min the rate of fall gradually slowed down; after 25 min $\eta'_{sp}/\eta_{sp}$ was about 0.70.

In the case of photoinduced haemolysis of RBC in presence of CPZ, Kochever and Lamola[3] observed that photoproducts of CPZ were able to haemolyse RBC possibly by membrane damage.

Our preliminary results also indicate that near-UV-light induced damage to DNA in presence of CPZ is primarily due to the products of CPZ produced during irradiation. However, the products are yet to be identified clearly.

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## *IN VITRO* STUDIES ON THE EFFECT OF CADMIUM ON GOAT EYE LENS

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ENVIRONMENTAL pollution by toxic metals is of global significance. Among the many toxic materials, cadmium, a non-essential element, has in recent times posed serious problems to both occupational health and community health[1]. Cadmium manifests its toxicity in humans and animals by accumulating in almost all organs. Kidney is the main target, where it is concentrated mainly in the cortex[2]. Cadmium is also toxic to the central nervous system, causes alteration of cellular functions in the lungs and affects both humoral and cell-mediated immune response in animals[3]. Whereas the toxic effects of cadmium have been related to specific chemical and physical properties of the ion[4], the molecular nature of defects in fundamental biochemical processes that are directly related to the toxicity of cadmium is yet to be understood. In this communication we report the effect of cadmium on ascorbic acid, glutathione (GSH) and proteins of goat eye lens.

Fresh goat eyes were obtained from a local butcher and were transported to the laboratory in an ice box. The lenses were dissected from the eyeballs by the posterior approach method. Each lens was weighed and incubated in a plastic petri dish containing 10 ml of tissue culture medium (pH 7.4) with or without cadmium chloride in the concentration range 20–500 $\mu$M for 10 h. The lenses were then taken out, wiped free of medium and weighed again. Lenses were individually and separately homogenized in 0.1 M Tris buffer (pH 7.4), and GSH[5], ascorbic acid[6], and total, soluble and insoluble protein[7] were determined.

Lenses incubated without cadmium chloride were clear and transparent whereas lenses incubated in medium containing cadmium showed a whitish appearance on the surface. Changes in biochemical properties are listed in table 1. The pH of the medium before and after incubation did not change and remained in the range 7.28–7.34. The average weight determined from almost similar-sized lenses was $1275.8 \pm 37.3$ mg. Average total protein was about 395 mg per lens and the ratio of soluble protein to total protein was about 0.5. Increasing the concentration of $Cd^{2+}$ in the medium did not affect