

releasing factor, resulting in greater gonadotrophin release for a given amount of releasing factor¹⁵.

Simultaneous administration of DEC with a dose of 20 mg/kg body weight irrespective of route of administration, significantly blocked the precocious, puberty, vaginal opening and superovulating effects of EM in immature rats. It can be considered the DEC blocked the central feedback effect of estrogen on the release of gonadotrophins (anti-gonadotrophic). Since there was no ovulation in DEC-treated rats primed with estrogen, it can be surmised that DEC inhibits the production/or release of the ovulating hormone, LH or alters the sensitivity of the pituitary to endogenous hypothalamic gonadotrophin releasing factor.

In animals treated with EM and EM + DEC combination, there was a very significant increase ($p < 0.001$) in the uterine weight (vs control and DEC-treated animals) confirming the non-interference of DEC on the peripheral action of estrogen *i.e.*, uterus.

ACKNOWLEDGEMENT

The authors wish to thank Burroughs Wellcome & Co., Bombay for generous gift of pure diethyl carbamazine citrate.

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EFFECT OF MAGNESIUM ON THE TOXICITY OF CHROMIUM AND LEAD TOWARDS *ESCHERICHIA COLI* AND *AEROBACTER AEROGENES*

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ABSTRACT

The effect of magnesium on the toxicity of chromium, and lead towards *E. coli* and *A. aerogenes* was studied. In the absence of any heavy metal in the growth medium magnesium alone enhanced the growth of organisms to a great extent. Increasing concentration of magnesium increased the growth and diminished the toxicity effect of chromium both in *E. coli* and *A. aerogenes*. A similar result was obtained during the studies on the influence of increasing concentrations of magnesium on the toxicity of lead in *E. coli* as well as *A. aerogenes*.

INTRODUCTION

THERE have been some reports regarding the interaction of nontoxic metals with toxic metals diminishing or enhancing the toxicity of the latter by the former. The ion antagonism has been studied but not sufficiently. In *Lactobacillus arabinosus* overcoming of Zn toxicity by Mn was reported¹. Subsequently the improvement of the growth and the elimination of toxic effect of copper by EDTA were reported². The removal of the lethal effect of Cd and Zn by EDTA,

citrate, aspartate and pyruvate in *Pseudomonas* and *A. aerogenes* was also observed^{3,4}. Also the toxicity of Hg was reported to be decreased by Na₂S addition in mixed rumen bacteria⁵. However the inefficiency of ions such as MgCl₂, CaCl₂, MnCl₂ and NaCl in abolishing or diminishing toxic effect of Hg salts in *Staphylococcus aureus* was observed⁶ and the elimination of the killing effect of CdCl₂ with the addition of CaCl₂ was noted. Babich⁷ noted that the high concentration of NaCl unaffected, increased or decreased the toxicity of Zn in different organisms. In

the present communication effects of magnesium on the toxicity of chromium and lead in case of two organisms, *E. coli* and *A. aerogenes* have been described.

MATERIALS AND METHODS

The organisms *E. coli* and *A. aerogenes* isolated from the lake water of the University and deposited in the stock culture of this laboratory were used for these studies⁸. The medium used for culturing the organisms was the routine nutrient broth medium. The liquid medium was prepared by adding beef extract, (3 g) and peptone (5 g) in a litre of double distilled water and sterilized by autoclaving the medium for 15 min. The pH was adjusted to 7.6. The organisms usually grown at 37° C in a nutrient broth medium were maintained at 5° C. Subcultures from stock cultures stored at 5° C were incubated for 48 hr at 37° C. These were again subcultured and after incubation for 48 hr used as an inocula to inoculate experimental liquid medium. The experiments were carried out in Ehrlenmeyer flasks of 100 ml, capacity with side arm.

The metal salts (chromium as chromium trioxide, lead as lead nitrate and magnesium as magnesium sulphate) were of analytical grade. Stock solutions of each salt were made in sterilized double-distilled water. The individual flasks containing the growth medium were inoculated by 1 ml of the inoculum, prepared as above before incubation. The final volume in the flask was 50 ml. Flasks were then incubated for 48 hr at 37° C on a gyrotory shaker. Optical density was measured at 530 nm of Lumichem-20 (Scientific Instruments Service, India).

RESULTS AND DISCUSSION

It was observed that as little as 12 ppm of Cr in the growth medium almost completely stopped the growth of *E. coli* if little or no magnesium was present, and even after 48 hr incubation the optical density remained nearly the same. However, if much magnesium was applied (10 to 200 ppm), slight growth occurred. The effect of Mg in decreasing the toxicity of Cr was more pronounced at 2, 4, 8 and 10 ppm of Cr in the medium. With the increase in concentration of magnesium in the range of 10, 50, 100 and 200 ppm, the toxicity of heavy metal decreased and the growth rate increased (figure 1).

In *A. aerogenes*, 10 ppm was the concentration of Cr which almost completely inhibited the growth of this organism, and if Mg was not added to the medium, the initial density remained unchanged. However, if Mg was present in the range of 10 to 200 ppm, the initial density increased slightly showing some growth. The effect of Mg on the growth of *A. aerogenes* and toxicity of chromium was as pro-

nounced as in *E. coli* at 2, 4, 6 and 8 ppm of chromium in medium. With increasing concentration of Mg, the toxicity successively decreased and the growth rate subsequently increased (figure 2).

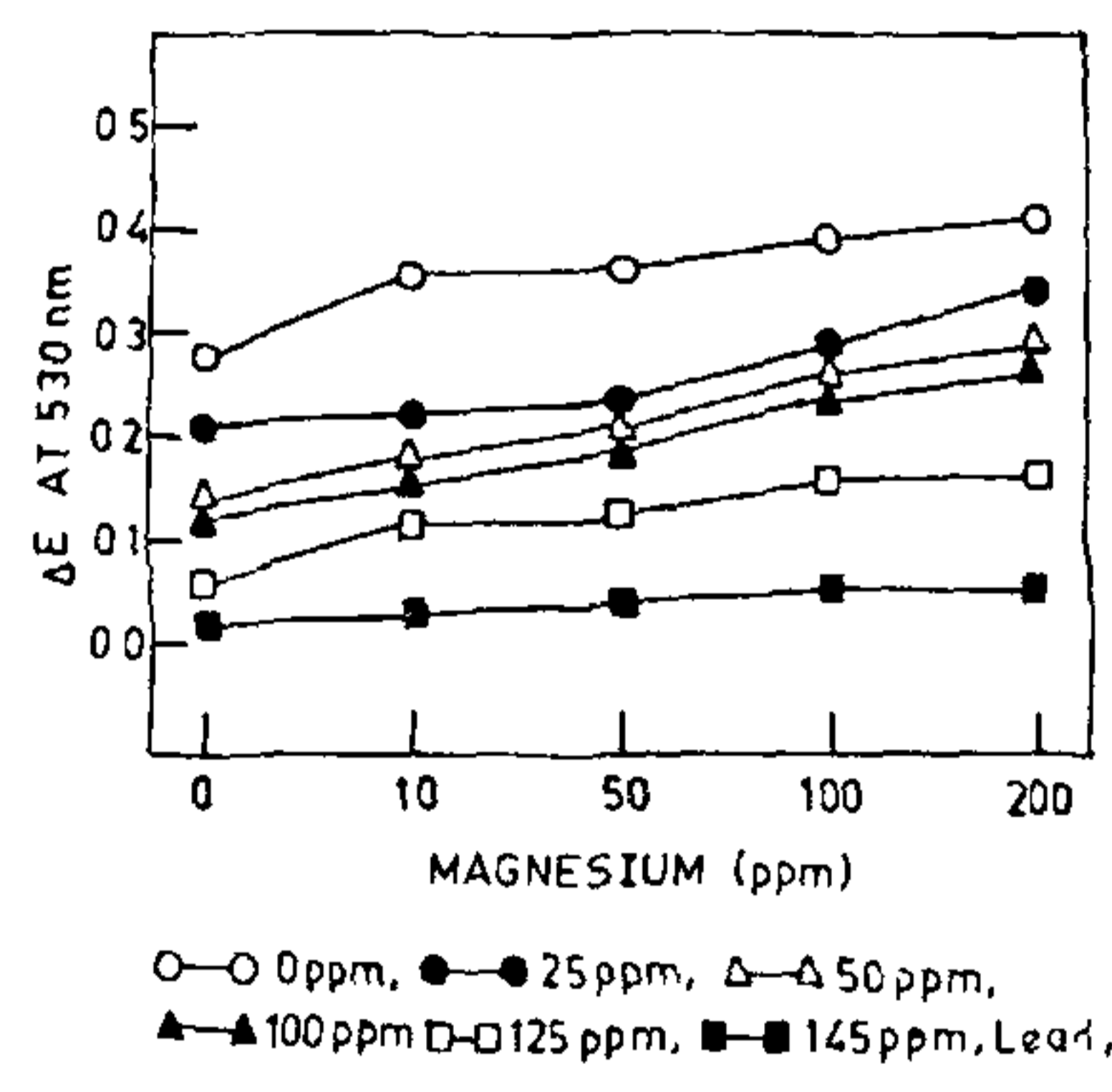
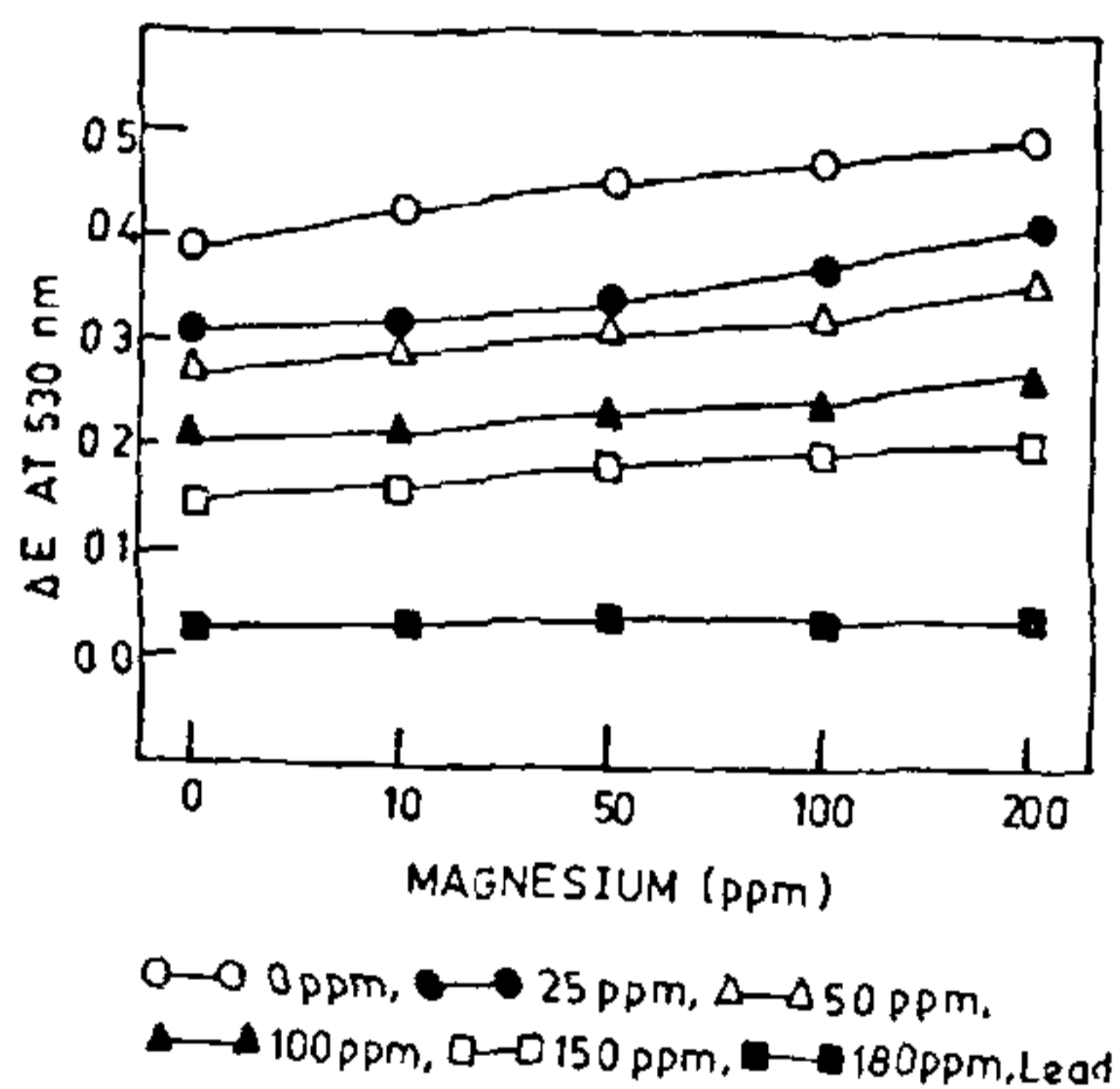
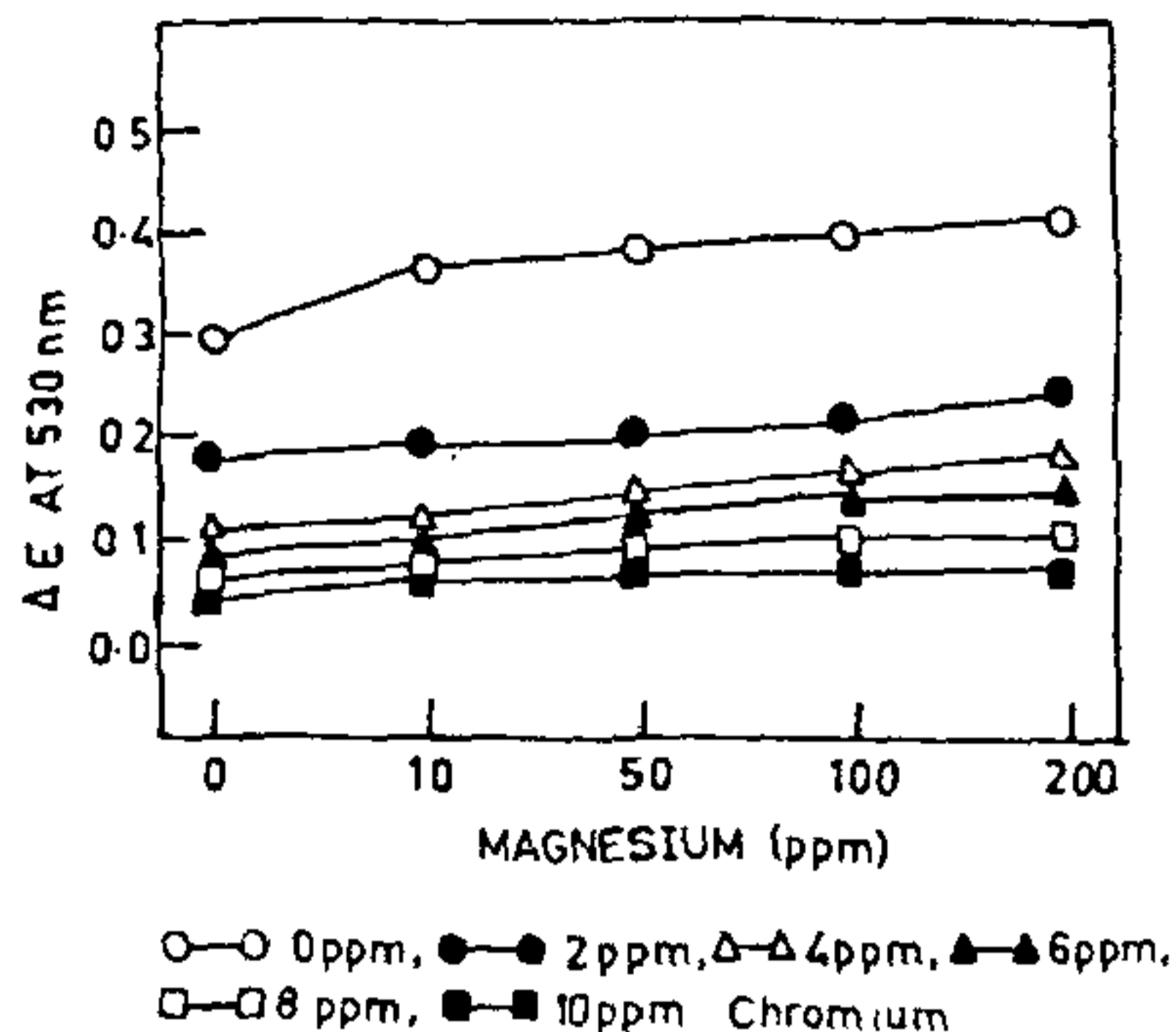
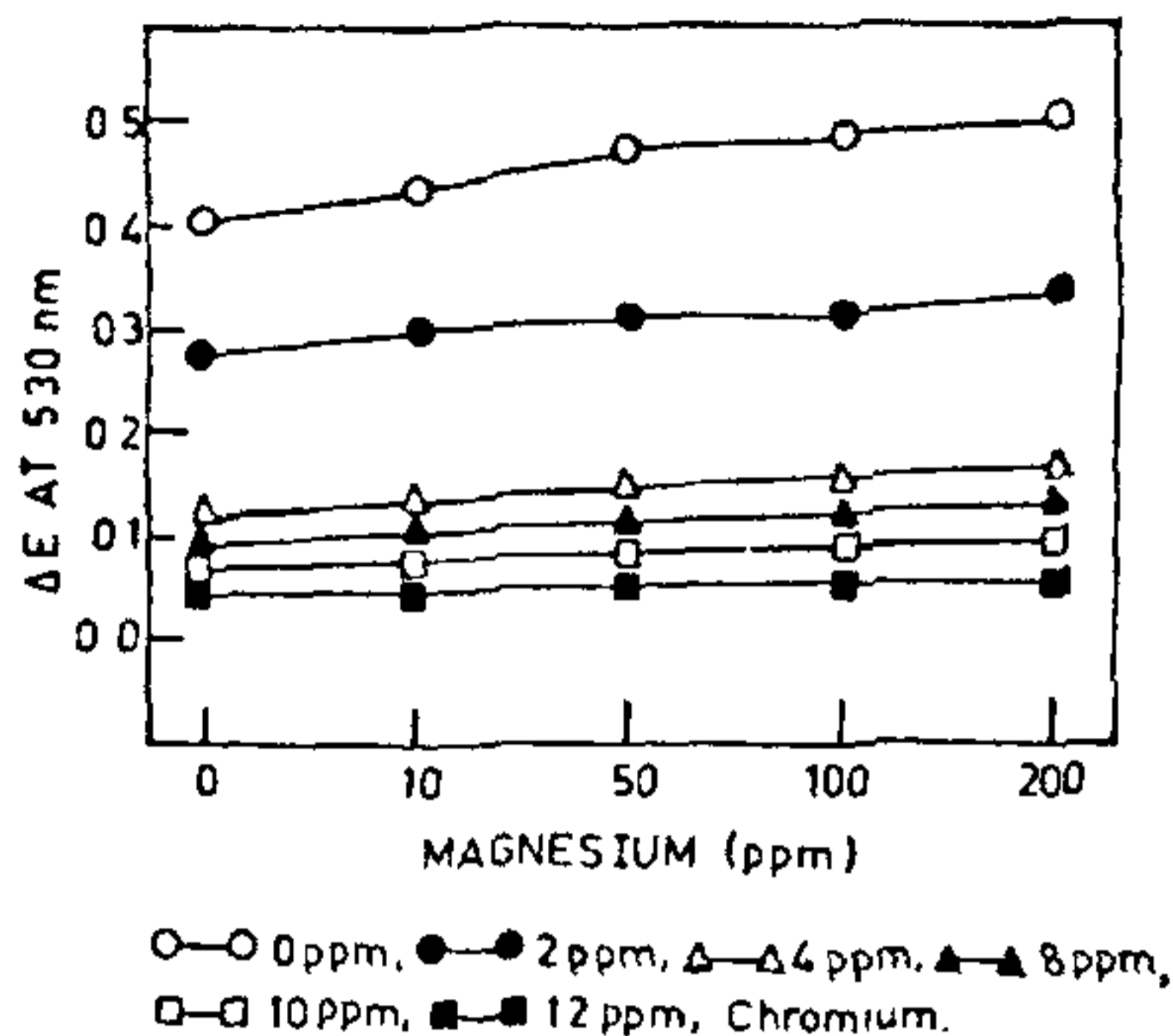
In figure 3, similar observations are shown involving lead and *E. coli*. Lead was less toxic than chromium and organisms could tolerate Pb to a great extent. The results were similar to those of chromium in the growth medium. Increasing magnesium concentration diminished the toxicity of Pb. High magnesium levels had a restraining influence both on the degree of toxicity and on the uptake of heavy metal. Toxic effect of Pb at low magnesium concentration was ameliorated by an increase in magnesium content in the medium.

Figure 4 presents the results with Pb and *A. aerogenes*. Inclusion of magnesium (10–200 ppm) could not arrest the inhibitory effect of lead on the growth when lead was added in the concentration of 145 ppm. However, if low concentrations of lead (25–100 ppm) were present in medium, increasing concentrations of magnesium increased the growth rate and removed the lethal effect of Pb to a great extent.

The results indicate that if inhibitory metal ion was absent in the growth medium, Mg alone promoted the growth of organisms to a great extent. With increasing concentrations of Mg, growth rate also increased (figures 1–4). This is in agreement with the findings of other workers^{2,9,10}, who in pure cultures of different organisms found that Mg stimulated the growth.

It was seen, that if a little magnesium was present, the toxicity effect of the inhibitor metal ion was unchanged. However, if higher doses of magnesium were applied, good growth of organisms occurred even at higher concentrations of inhibitor. With the increase in magnesium concentration, toxicity of inhibitor successfully decreased. The antitoxicity effect of Mg was more pronounced at lower concentrations of the inhibitor and higher concentrations of magnesium (figures 1–4). The findings agree with those of Abelson and Aldous⁹, but contradict those of Kondo, Ishikawa and Nakahara⁶ who reported inefficiency of Mg in abolishing or diminishing the toxicity effect of Hg in *Staphylococcus aureus*. The toxicity of the inhibitor ions at low concentration of Mg may be attributed to the interference in the normal metabolic role of magnesium, while lower toxicity of inhibitor at higher concentration of Mg may be due to the role of magnesium in diminishing the quantity of inhibitor bound by the cell⁹.

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Figures 1-4 . 1&2. Effect of magnesium on the toxicity of chromium towards 1. *E. coli*. 2. *A. aerogenes*. 3&4. Effect of magnesium on the toxicity of lead towards 3. *E. coli*. 4. *A. aerogenes*.

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