Protection by Cerium Chloride on CCl_4-Induced Hepatotoxicity

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Summary. Pretreatment of rats with cerium chloride, an inhibitor of the DMES, protects against CCl_4 intoxication. The protection was obtained against the fatty infiltration in the liver, the decrease of triglyceride secretion from the liver into the plasma compartment, the polysomal disaggregation and the rise of serum transaminases.

Recently, Ärvela and Kärki[1] showed that cerium chloride inhibits in rats the activity of the drug-metabolizing enzyme system (DMES). The pathophysiological state of the DMES may condition the reactivity of the liver against many exogenous compounds[2]. Among toxicants, CCl_4 is metabolized within the DMES into toxic products[3,4], so that inhibition or induction of the DMES leads to a decreased or an increased toxicity, respectively[5,6].

In the present study, cerium chloride was given to rats before administration of CCl_4, to see if the lanthanon interfers with CCl_4-induced hepatotoxicity.

Materials and methods. Male Wistar rats, weighing 250–300 g, were used. The animals were maintained on a standard diet (Ditta Piccioni, Brescia, Italy), devoid of antioxidants.

CCl_4, in mineral oil, was administered by stomach tube, at the dose of 2.5 or 5.0 ml/kg b.wt., in animals fasted for 12–16 h. Control animals received an equal volume of mineral oil. Cerium chloride was injected i.v. at the dose of 4.0 mg/kg b.wt., 72 h before CCl_4. Triton WR 1339 (Rohm and Haas, Philadelphia, USA), as a 20% solution (w/v) in 0.9% NaCl, was injected into the saphenous vein in the amount of 500 mg/kg b.wt. Hexobarbital (Winthrop, Laboratories Inc., New York, USA) was i.p. injected in the amount of 120 mg/kg b.wt. Those rats whose blood was not withdrawn, were killed by decapitation. Blood samples were taken from the aorta under pentobarbital anesthesia.

Total liver and plasma lipids were extracted according to the method of Folch et al.[7]. Total glycerides were determined by the method of Van Handel and Zilversmit[8]. Glutamic-oxaloacetic (GOT) and glutamic-pyruvic (GPT) transaminases were determined by a test combination method (Sclavo, Siena, Italy). Liver polysomal patterns were obtained by the method previously described[9].

Results and discussion. We have administered cerium chloride after having tested the maximum inhibition of hexobarbital oxidation by determining sleeping times in cerium chloride-treated rats. At 24 and 72 h after treatment with cerium chloride, sleeping times (expressed in min) were: 41.5 ± 3.6 and 88.6 ± 5.9, respectively, as compared with control rats (29.6 ± 3.7).

In rats pretreated with cerium chloride 72 h before CCl_4, a 30% mortality was observed, while in the control rats receiving CCl_4 alone the mortality reached a percentage of 90. The animals were observed for 7 days after CCl_4 intoxication, given at the dose of 5.0 ml/kg b. wt.

Fatty infiltration induced by CCl_4 24 h after its administration, as evaluated by the determination of the triglyceride content in the liver, was partially protected by pretreatment of rats with cerium chloride (Table I). The Triton-induced hypertriglyceridemia in CCl_4-poisoned rats was lower than in control rats, while in cerium-pretreated rats then intoxicated with CCl_4 it was higher than in rats treated with CCl_4 alone (Table I). Hence, one may deduce that cerium pretreatment ameliorates the lipoprotein secretion from liver into the plasma compartment.

CCl_4 intoxication is known to cause a shift of heavier polysomes to the dimeric-monomeric ribosomes[10]. As shown in Table II, such polysomal disaggregation was prevented by pretreating the rats with cerium chloride.

CCl_4-induced liver necrosis was also prevented by pretreating the rats with cerium chloride. As shown in the Figure, serum transaminases levels were lower in cerium-pretreated animals than in those receiving CCl_4. These findings were also supported by histological examinations showing that, in liver of rats treated with

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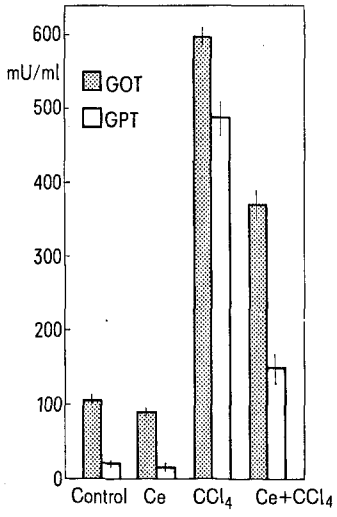
Table I. Effect of cerium chloride on the liver and plasma triglycerides in rats intoxicated with CCl_4

Treatment	Liver triglycerides[a] (mg/100 g b.w.)	Plasma triglycerides[b] (mg/100 ml)
a) Control	17.79 ± 2.22 (7)	462.57 ± 87.90 (5)
b) Cerium chloride	15.72 ± 0.54 (8)	442.71 ± 49.21 (6)
c) CCl_4	151.00 ± 7.22 (8)	229.80 ± 12.53 (7)
d) Cerium chloride + CCl_4	86.55 ± 7.88 (8)	342.33 ± 13.89 (5)

Mean ± SE. Number of animals is given in parentheses. Cerium chloride was administered at the dose of 2.5 ml/kg b.wt.; Triton WR 1339 was administered i.v. at the dose of 500 mg/kg b.wt. 90 min before sacrifice. The animals were killed 24 h after CCl_4 administration. Statistical significance by *t*-test was: [a] a–c, a–d and c–d: $p < 0.001$; [b] a–c and c–d: $p < 0.001$.

CCl_4 alone, the centrolobular necrosis was more severe than in liver of rats pretreated with cerium chloride and intoxicated with CCl_4.

The toxicity of CCl_4 is assumed to be due to reactive metabolites, such as $\cdot CCl_3$ free radical, which are produced in the DMES[3]. Since cerium chloride depresses the DMES[1], it may be implied that protection by cerium chloride in CCl_4-intoxication may be due to a diminished biotransformation of the haloalkane in the liver. A protective effect has been shown by us[11] and CARLSON[12] with lead nitrate and methylmercury respectively. Also in these cases, protection is due to inhibition of the DMES induced by the metals, and consequently to reduction of CCl_4 metabolism into toxic products.



Serum transaminases glutamic oxaloacetic (GOT) and glutamic pyruvic (GPT) under the following experimental conditions: Cerium chloride was given i.v. at the dose of 4.0 mg/kg b.wt., 72 h before intoxication; CCl_4 was given p.o. at the dose of 2.5 ml/kg b.wt. The rats were sacrificed 24 h after intoxication. Vertical bars represent the SEM from at least 6 rats.

Table II. Protection by cerium chloride on the liver polysomal damage induced by CCl_4

Treatment	Polysomes[a] / Total ribosomes
a) Control	0.59 ± 0.02
b) Cerium chloride	0.59 ± 0.08
c) CCl_4	0.39 ± 0.05
d) Cerium chloride + CCl_4	0.53 ± 0.05

[a] Calculated by the areas of the polysomal patterns. Mean $\pm$ SE. 5 animals were used for each group. CCl_4 was given p.o. at the dose of 2.5 ml/kg b.wt., 1 h before sacrifice. Statistical significance by t-test was: a–c; c–d: $p < 0.001$.

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Depression of Neurones in the Rat Cerebral Cortex by Leptazol

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Summary. The convulsant drug leptazol was applied by microiontophoresis to 116 neurones in the cerebral cortex of rats. The firing of 101 cells was reduced. Only 6 cells were excited.

Pentamethylenetetrazol (leptazol) is a convulsant drug whose mode of action is unknown. Several other convulsants act by interfering with the actions or release of inhibitory neurotransmitters in the central nervous system. Strychnine blocks the postsynaptic actions of glycine[1] and both picrotoxin and bicuculline antagonise γ-aminobutyric acid[2]. Leptazol does not antagonize central inhibitory transmitters in this way[3,4] and it has been suggested that leptazol could have a direct excitatory action on some cells[5–8]. The present experiments were undertaken to examine this possibility.

Materials and methods. Male hooded Listar rats weighing 250–300 g were anaesthetized with urethane, 1.25 g/kg i.p. Experiments were performed in the somatosensory cerebral cortex on cells which were either randomly encountered at various depths or identified as pyramidal tract cells by antidromic stimulation of the medullary pyramid[9].

Details of the preparation of animals, and of the microiontophoretic techniques used have been given previously[9]. Five-barrelled micropipettes with overall

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