Communications to the Editor

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ALTERATIONS IN THE SYNTHESES OF RNA AND DNA BY CHROMIUM(III) IN REGENERATING RAT LIVER

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The administration of trivalent chromium (Cr(III)) caused a significant enhancement of RNA synthesis in rat liver after partial hepatectomy. Cr was accumulated in the liver cell nuclei and concentrated in the nucleoli. Of RNAs synthesized in the nuclei, nucleolar RNA synthesis was most enhanced by the administration of Cr(III). Furthermore, DNA synthesis in the Cr(III)-administered rat liver after partial hepatectomy was initiated a little earlier and was less synchronized than in control rats.

KEYWORDS———chromium(III); regenerating rat liver; partial hepatectomy; nucleus; nucleolus; RNA synthesis; DNA synthesis

The biological action of trivalent chromium (Cr(III)) has been understood from two points of views; one is that it acts as an essential trace element for maintenance of normal glucose tolerance,[1] and the other possibility is that it acts as the ultimate carcinogen or mutagen produced intracellularly from hexavalent chromium (Cr(VI)).[2] Presumably related to the latter action is the fact that Cr(III) binds to DNA and enhances *in vitro* RNA synthesis[3] and also that the administration of Cr(III) in mice causes a significant enhancement of hepatic RNA synthesis,[4] whereas Cr(VI) does not enhance RNA synthesis either *in vitro* or *in vivo*.[3,4]

On the other hand, it has been reported that with partial hepatectomy of rats endogenous Cr became concentrated in the liver nuclei.[5] This, together with the fact that RNA synthesis is activated after the partial hepatectomy, suggests that regenerating liver may be good material for assaying the effect of Cr on RNA synthesis. The effect on hepatic DNA synthesis can also be assayed, because a synchronized DNA synthesis is initiated approximately 18 h after the hepatectomy.[6] By using partially hepatectomized rats under these considerations, the present study indicates that Cr(III) administration causes a significant enhancement of nucleolar RNA synthesis and also induces a less synchronized DNA synthesis in the regenerating liver.

Male Wistar rats (110-120 g) were intraperitoneally administered an aqueous solution (0.5 ml/rat) of $CrCl_3$ (5 mg Cr/kg body weight) 24 h before the partial hepatectomy which was performed with removal of the main lobes (*ca.* 68% of the liver) in the usual manner.[7] To determine the accumulation of Cr in the regenerating liver, some rats received $^{51}CrCl_3$ (30 μCi/rat; NEN) in the $CrCl_3$ solution. Control rats received 0.5 ml of distilled water in the same manner. At 0 to 12 h after the hepatectomy, RNA synthesis in the regenerating liver was assayed by the incorporation of [6-^{14}C]orotic acid (0.5 μCi/rat; RCC Amersham, 59 mCi/mmol), intra-

venously injected 15 min before the assay time, into the whole cellular RNA and nucleolar RNA; the latter was fractionated from the nucleoli obtained by sonicating the nuclear preparation of the liver.[8] The DNA synthesis in regenerating liver was determined 0 to 48 h after the partial hepatectomy by the incorporation into liver DNA of [6-^{3}H]thymidine (5 μCi/rat; NEN, 21.5 Ci/mmol) injected intravenously 60 min before the assay time.

As shown in Fig. 1a, RNA synthesis in the regenerating rat liver 0 to 12 h after partial hepatectomy was significantly enhanced by the pre-administration of $CrCl_3$ (5 mg Cr/kg body weight). Fig. 1a also shows that in the course of regeneration the accumulation of Cr in the liver proceeds with time. In particular, the nuclear Cr increased from ca. 20% of cellular Cr at 0 h to ca. 40% at 6-12 h after the hepatectomy. Since in regenerating liver at the early stage the nucleolar RNA is predominantly synthesized,[9] its synthesis was assayed as indicated in Fig. 1b. Cr accumulation in the nucleoli increased with time, and throughout the experimental period nucleolar RNA synthesis was significantly enhanced in the regenerating liver of $CrCl_3$-dosed rats. In regenerating liver the rate of nucleoplasmic RNA synthesis is known to be relatively low and $CrCl_3$ administration did not enhance it (data not shown). Therefore, it may be said that the administration of $CrCl_3$ specifically enhanced the nucleolar RNA synthesis in this system. A rather rapid accumulation of Cr in the nucleoli after the hepatectomy, shown in Fig. 1b, may suggest that the accumulated Cr directly enhanced the RNA synthesis, as was found in an in vitro system,[3] although this is not certain. The fate of newly synthesized nucleolar RNA in $CrCl_3$-dosed rats is now being investigated in our laboratory.

Nucleolar RNA synthesis has been known to be linked to subsequent initiation of DNA synthesis.[10] As indicated in the $CrCl_3$-administered rats in Fig. 2,

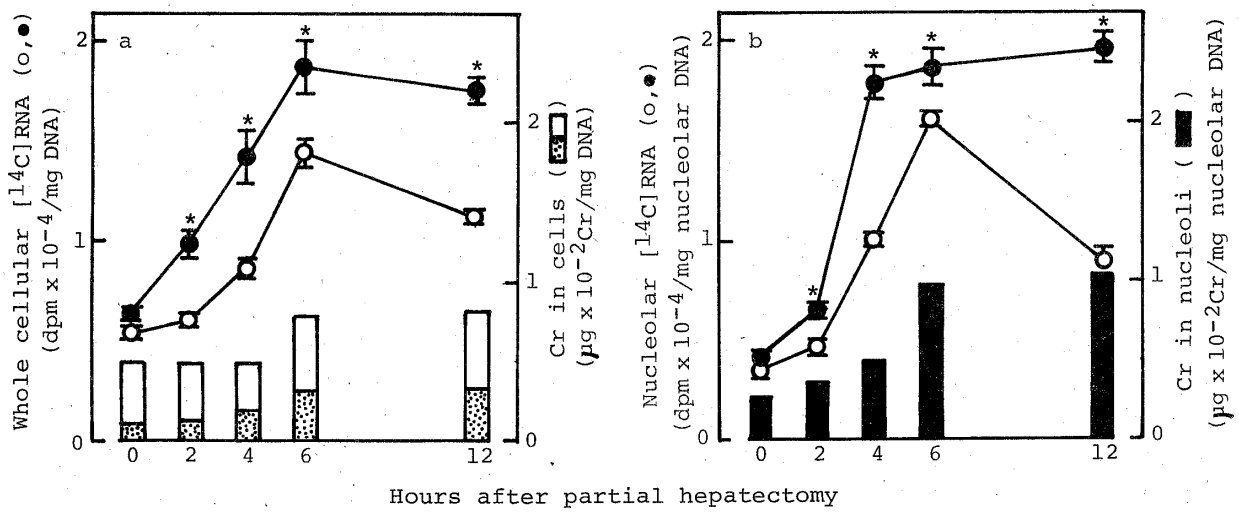

Fig. 1 Cr Accumulation and RNA Synthesis in the Whole Cells (a) and Nucleoli (b) of Regenerating Rat Liver

Cr accumulation: ▨ in whole cells, ▨ in nuclei, ■ in nucleoli.
● $CrCl_3$-dosed (5 mg Cr/kg), o control.
Each point and vertical line indicates mean ± SE for 3 rats.
* Significantly different from each control ($p < 0.05$).

DNA synthesis was initiated a little earlier and continued longer, i. e., was less synchronized than in control rats. Although it is uncertain whether this alteration in DNA synthesis resulted from the enhanced RNA synthesis in the nucleoli or from direct action of Cr accumulated in the nuclei or else, the alteration must be of biological interest. Cr has been found to be concentrated in the nuclei of a variety of cells,[4,5,11] suggesting that this element may have a role in or effect on nuclear functions. The present study may support this idea, although it remains to be determined whether it is a physiological or toxicological action.

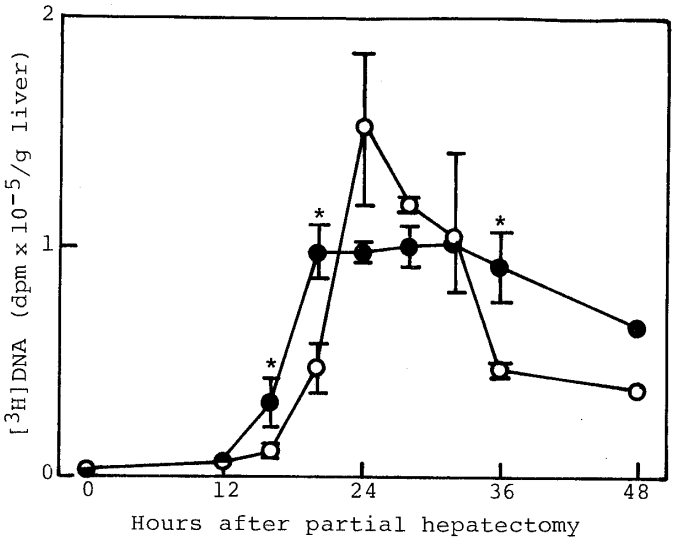

Fig. 2 DNA Synthesis in Regenerating Rat Liver after $CrCl_3$ Administration

● $CrCl_3$-dosed (5 mg Cr/kg), o control. Each point and vertical line indicates mean ± SE of 3-4 rats except experiments at 0, 12 and 48 h (one rat for each group).
* Significantly different from each control ($p < 0.05$).

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