THE EFFECT OF N-NITROSODIETHYLAMINE AND N-NITROSOPYROLIDINE ON GAMMA GLUTAMYLTRANSPEPTIDASE (GGT) AND ALKALINE PHOSPHATASE (ALP) IN WHITE MICE LIVER

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Introduction

Small amounts of carcinogenic N-nitrosocompounds are found in food and may be formed in the acid contents of the stomach by the reaction between nitrite and amines in the food. Common carcinogenic volatile nitrosamines in the food are N-Nitrosopyrolidine (NPYR) and N-Nitrosodiethylamine (NDEA). These carcinogenic chemicals exert their cancer initiating effects only if it is converted by metabolic processes to chemically reactive species. The greatest capacity to metabolise these alkylating agents is found in the liver (Wan Ngah et al. 1997). In the management of cancer, the determination of the activities of GGT (EC 2.3.2.2) and ALP (EC 3.1.3.1) are regarded as indicators for liver damage. GGT is a useful marker in human melanoma, breast cancer and lung cancer and ALP activity is also a marker for cell differentiation. ALP may be seen in any disease process of the liver and its elevation is also a sensitive test for detection of liver disease). The effect of nitrosamines on key detoxification enzymes will be determined.

Materials and Methods

Male white mice (60, five weeks old) were divided into five groups for five different treatment time: 24, 36, 48, 60 and 72 hours (12 mice in each group). They were than subdivided to four groups and treated with 0.5, 2.5 and 5.0mg NDMA/kg of body weight respectively. Livers were excised after the different treatment periods and homogenised with Tris- HCL pH 7.4 and centrifuged at 10,000g for 45 min at 4°C. The supernatant was filtered through glass wool and stored at 4°C. Gamma glutamyl transpeptidase enzyme was assayed by the method of Jacobs (1971) and Alkaline phosphatase by the method of Bessey et al. (1946). Protein determination was carried out by the method of Biuret (Gornall et al. 1949).

Results and Discussion

The specific activity measured was compared between treated and control groups with different treatment time and doses.

Activity expressed as changes in percentage and control value was 100%. GGT specific activity increased about 50% compared to control after 36 hours subjected to NDEA by forced feeding with 30 and 40mg/kg of body weight. Activity subsequently decreased at 48 hours and 60 hours and increased again at 72 hours in treated samples. For ALP, its activity was elevated in the first 24 hours and the increment was about 60% after exposure to 20mg NDEA/kg of body weight. After 24 hours activity of ALP remained below control and yielded expression at 60 hours NDEA liver sample. For samples treated with NPYR, the induction of activities of GGT and ALP in liver were mainly seen for the 60 and 72 hours treatments. The changes in specific activity for ALP and GGTP ranged between 50% and 60% compared to control after treatment with 40mg NPYR/kg of body weight of mice. From the results, there was an induction of activity for both enzymes. Nitrosamines are supposedly distributed uniformly during a short period of exposure, but for NPYR, the expression of ALP and GGT specific activities predominantly takes place after 60 and 72 hours and this is because liver may be incapable of metabolising carcinogen in 'first pass' clearance, thus greater fraction of this carcinogen becomes available at later hours.

NDEA's effect on these enzymes varied from time to time and both were not concurrently expressed. Specific activity of these enzymes were dependant not only on its concentration in the sample, but also on the latency of the enzymes present because the stimuli used may cause changes in the enzyme micro-environment leading to increase activity and *vice versa*. Since the production of an enzyme follows general scheme of protein synthesis, different proteins may be synthesised in cancerous liver and chemical carcinogens could inhibit some protein synthesis and elevate certain other detoxification enzyme activities (Schlaeger, 1975).

Conclusions

These results suggested that feeding of NPYR and NDEA to mice increased both enzyme activities in liver and both enzymes can be used as tools or markers to further identify diseases in liver caused by other N-nitroso compounds before and after administration.

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