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# LOW PLASMA TESTOSTERONE VALUES IN MEN DURING HANGOVER

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SUMMARY

Plasma testosterone, estradiol, estrone and luteinizing hormone concentrations were measured by radioimmunoassay in healthy volunteers who after fasting for 10 h consumed 1·5 g ethanol/kg body wt. Of this group five suffered from severe hangover while another five had essentially no hangover. Ten to twenty hours after drinking, the testosterone concentrations were significantly decreased in all subjects, but in the 5 subjects with severe hangover the decrease was more pronounced. Estradiol values decreased during the hangover period, but were normal at the time of acute intoxication, whereas estrone values, in the few cases determined, showed a tendency to increase during acute intoxication. A compensatory increase in the plasma concentration of luteinizing hormone was found in all subjects.

## INTRODUCTION

Chronic alcohol intake is known to cause gynecomastia and testicular atrophy in some male subjects [1, 2]. These phenomena are suggested to be due at least partly to altered steroid metabolism in the cirrhotic liver [1–3]. In addition, it has recently been shown that acute alcohol intake can also induce remarkable changes in the metabolism of steroid sulfates [4, 5]. These changes seem to be attributable to the reduction of the hepatic redox state during alcohol oxidation [5]. The biological activity of neutral steroid sulfates as such has not been evaluated with certainty, but it is probably low or completely absent and thus the physiological significance of the alcohol induced changes in their metabolism is not clear. Testosterone is the main androgenic hormone in man and it is one of the immediate precursors of estradiol in the testes [6]. Therefore, we decided to study the effects and after-effects of a single relatively large dose of alcohol on plasma concentrations of testosterone, estradiol, estrone and luteinizing hormone (LH).

## MATERIAL AND METHODS

In all 19 healthy male medical students (aged 19–25) participated in our larger hangover study [7]. All experiments were begun at 6 p.m. Before the session every volunteer fasted for 10 h. Each experimental session involved 6 subjects—3 receiving ethanol 1·5 g/kg body wt. as a 20% solution, which they drank at a constant rate during the 3 h from 6 to 9 p.m. This dose of ethanol was found suitable for inducing hangover but was not large enough to cause too severe and dangerous intoxication. The other three volunteers at each session consumed an equal volume of water. Two weeks later the roles were reversed so that each subject served as his own control.

The intensity of intoxication and hangover were estimated by rating the subjective feeling and objective signs using special test forms [8]. The subjective symptoms were transformed into length units so that each subject marked his general feeling of the degree of intoxication or hangover on a scale of 100 mm. The distance of the mark from the zero point was taken as a measure of subjective feeling. Some simple tests were performed to evaluate the physical signs of intoxication. The results of these were rated from 0 to 4 and the sum was used as a rough score of disturbance during intoxication. In addition to the scale method, a rating of the intensity of fatigue, headache, dizziness, nausea, thirst and tension from 0 to 4 was used to express the subjective feeling of hangover. A rating of the intensity of paleness, tremor and perspiration from 0 to 2 was used to score the objective signs of hangover.

For testosterone determinations we selected from the group of 19 the five who according to the test program had most severe hangover and five who had

essentially no hangover, to investigate whether the intensity of hangover has any correlation with plasma testosterone concentration. According to the hangover scale the subjective feeling of hangover was most intensive 15 h after the start of the experiment (Fig. 1). The scores of target symptoms and physical signs of hangover correlated well with the hangover scale value, and all these measures of the intensity of hangover were significantly different in the mild and severe hangover groups.

Blood samples were drawn into heparinized tubes 0, 4, 8, 12, 15 and 20 h after the start of the experiment. Plasma was separated and stored at −80°C for 1–4 months before the determinations were made.

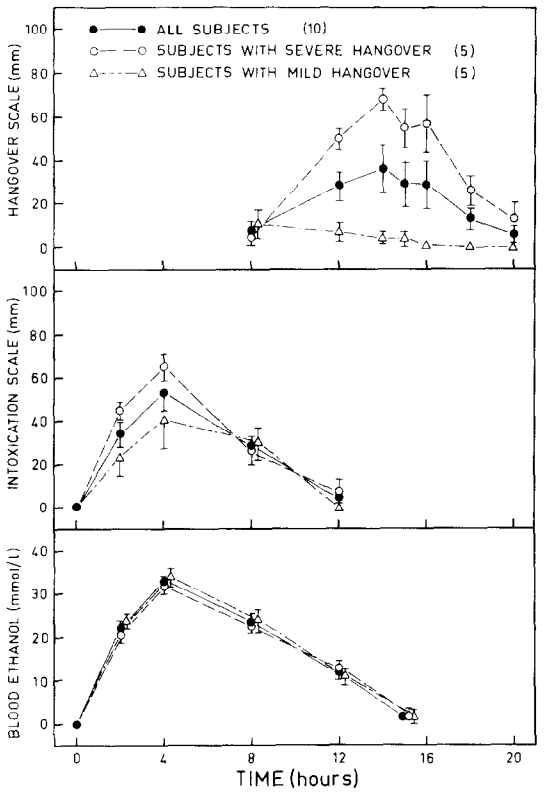


Fig. 1. Blood ethanol concentration and the intensity of the subjective feeling of alcohol intoxication and hangover. All subjects fasted for 10 h after which they drank 1·5 g ethanol/kg body wt. from 0 to 3 h as a 20% aqueous solution. In all, there were 10 subjects. The subjective feeling of alcohol intoxication and hangover were converted to length units so that every volunteer could mark his general feeling on a scale of 100 mm (intoxication scale and hangover scale). The distance of the mark from the starting point of the scale was used as a measure of the feeling of intoxication or hangover. Five subjects, who at any time had a hangover scale value of more than 50 mm, constitute the severe hangover group. The other five subjects always rated their hangover less than 20 mm and they constitute the mild hangover group. Each point represents mean ± SEM from 5 or 10 experiments.

Testosterone was determined principally according to the method of Ismail *et al.* [9]. One ml of serum was precipitated in the cold with 50% ammonium sulfate. This procedure was repeated and then the testosterone bound to sex hormone binding globulin in the ammonium sulfate-precipitate was extracted from these fractions with benzene–petroleum ether [2:5]. Radioimmunoassay was performed using 1,2,6,7-$^{3}$H-testosterone (The Radiochemical Centre, Amersham, England) and antiserum (raised in rabbits against testosterone-3-bovine serum albumin, produced by Searle Diagnostic, High Wycombe, Bucks, England) in 0·133 M borate buffer pH 8·0 containing 0·02% gelatine.

The LH radioimmunoassay method was principally that of Den Hollander *et al.* [10, 11], with the following modifications: Purified human LH (National Pituitary Agency, National Institute of Arthritis, Metabolism and Digestive Diseases, Bethesda, U.S.A.) was radioiodinated using $^{125}$I according to a modified Greenwood–Hunter method[12]. Human Pituitary Luteinizing Hormone (68/70) (National Institute for Medical Research, London, England) was used for the construction of the standard curve. The concentrations used ranged from 1–900 mIU human LH/ml. Antigen (unknown or standard), labelled antigen and anti-human LH (National Pituitary Agency, National Institute of Arthritis, Metabolism and Digestive Diseases, Bethesda, U.S.A.) were incubated for a period of 48 hours in a volume of 550 μl. Insolubilized second antibody (immunosorbent) was then added (500 μl) and the tubes were rotated at room temperature for 6 h before centrifugation after which the supernatant was discarded and the solid phase washed and counted using an LKB-Wallac gamma counter.

Plasma estradiol-17β and estrone were determined according to the methods of Onikki and Adlercreutz[13, 14]. This method is based on the same principle as the testosterone assay. The radioimmunoassay of estradiol-17β was performed both before the reduction and after all the estrone present was reduced to estradiol. The antibody used, was raised in a rabbit against 11α-hydroxyestradiol-11-succinyl-bovine serum albumin. From the difference in these two determinations the plasma estrone value was calculated.

Blood ethanol was determined by gas chromatography as described earlier [8]. Statistical significances were tested by linear regression analysis [15] and by the *t*-test according to de Jonge[16].

## RESULTS

Blood ethanol concentration was at its maximum 4 h after the start of drinking (Fig. 1). At this time the

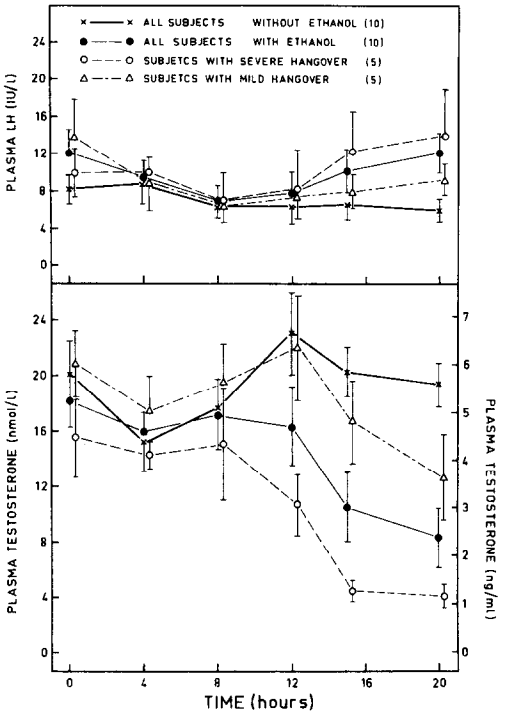


Fig. 2. Plasma concentrations of testosterone and luteinizing hormone (LH) during alcohol intoxication and hangover. Experimental conditions as in Fig. 1. The groups are also as in Fig. 1, except that the group "all subjects without ethanol" represents values obtained from the control sessions during which the same 10 subjects drank water instead of ethanol. Each point represents mean ± SEM of 10 or 5 experiments.

subjective feeling of alcohol intoxication was most severe (Fig. 1). The score for objective signs of intoxication also showed a maximum simultaneous with the latter parameters. Those who subsequently had severe hangover seemed to be more intoxicated than those whose hangover was mild, although blood ethanol concentrations were similar in both groups (Fig. 1).

Plasma testosterone concentrations did not change during the acute intoxication period (Fig. 2) and the values were similar in the severe and mild hangover groups. In the control group the concentrations were lowest in the late evening (10 p.m.) and rose from then until morning (6 p.m.) as has been found when the diurnal variation in plasma testosterone concentration has been studied (*e.g.* 17). Twelve hours after the start of the experiment, when the subjects began to suffer from hangover, the concentration of testosterone decreased rapidly and significantly especially in the severe hangover group (Fig. 2). There was also some decrease in the mild hangover group but this was not nearly as great as in the severe hangover group, in which the lowest values were only 19% of the respective values during the control period. In the correlation analysis a significant inverse correlation between the intensity of hangover and the plasma testosterone concentration was found ($r = 0{\cdot}76$ at 15 h). During the control period there were no differences in testosterone concentration between the mild and severe hangover groups. During hangover estradiol concentration decreased to about 63–80% (15 h:51 ± 10 ng/l) of the control level (81 ± 2 ng/l). These values are somewhat higher than most authors have reported but are consistent with values obtained previously with the same method [13]. In contrast, the plasma concentration of estrone, where measured, showed a tendency to increase during the intoxication period.

To elucidate the possible role of gonadotrophins in mediating the observed changes, we measured the plasma LH concentration in these samples. When compared to the control values no decrease in its concentration was found during the experimental period (Fig. 2). On the contrary, there was a significant, obviously compensatory increase in its concentration during hangover.

## DISCUSSION

Ethanol seems to have no acute effects on plasma testosterone and LH concentrations. Similar observations have also been made by Toro *et al.*[18]. The increased plasma concentration of estrone during the intoxication phase is difficult to assess because of the small number of samples analyzed and because both the testes and adrenals contribute to the circulating level [6].

During and after halothane anaesthesia [19] and after surgical procedures [20] testosterone concentration in human plasma has also been shown to decrease considerably. The reason for the testosterone decrease in these situations and during hangover is not known. It must certainly be due to decreased production and/or an increase in catabolism or excretion. The decrease in production could be due to insufficient LH-stimulation or impairement of biosynthesis.

The simultaneous decrease in estradiol and testosterone levels during the hangover period might suggest that there is an impairement of LH-production. However, no decrease in plasma LH was found during the experimental period. LH is generally accepted as the pituitary gonadotrophin which controls testosterone secretion [21]. It has not, however, been found to be raised in association with the testosterone increase observed *e.g.* in physical exercise [22]. Similarly a lack of association between androgens and LH has been observed in some other situations [23, 24]. Therefore,

the possibility of ethanol affecting testosterone secretion *via* the hypothalamus or higher centres remains open. On the basis of this study we cannot reach any conclusion concerning a possible direct effect of ethanol on the testis, but high LH values simultaneous with the lowest testosterone level support the existence of such a mechanism.

The pathway of testosterone catabolism includes many reductive steps, most of which occur in the liver [25]. Ethanol oxidation profoundly changes the redox state of the liver by increasing its content of reducing equivalents [26]. This change may affect the catabolic pathways of testosterone metabolism by favouring reductive reactions and inhibiting oxidative steps. However, the effect of ethanol on the hepatic redox state is maximal during ethanol oxidation, and the testosterone concentrations are lowest many hours after the completion of ethanol elimination (Figs. 1 and 2). In addition, the rate of ethanol oxidation and thus the reduction of the hepatic redox state are similar in those who suffered from severe hangover and in those whose hangover was mild [7]. Thus, it seems improbable that the difference in plasma testosterone between the severe and mild hangover groups is a result of changes in the hepatic redox state.

In chronic alcoholics Fabre *et al.*[27] found increased urinary excretion of testosterone glucuronide. They also found that alcohol intake increased the excretion of this conjugate in those subjects [27]. Thus, in short term experiments, alcohol may also enhance the excretion of testosterone and so decrease its plasma concentration.

It can be concluded that after one large dose of pure ethanol the concentration of testosterone in plasma is strikingly decreased and that this decrease is related to the intensity of the hangover. However, the mechanism of this phenomenon and its physiological significance are obscure. The results suggest that direct effects of ethanol may cause endocrinological disturbances in male alcoholics by lowering plasma testosterone concentration independent of the status of liver function. This effect is therefore of general interest in regard to the normal functioning of the reproductive glands.

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