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Correlation Between Human Erythrocyte Aldehyde Dehydrogenase Activity and Sensitivity to Alcohol

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INOUE, K., M. FUKUNAGA AND K. YAMASAWA. *Correlation between human erythrocyte aldehyde dehydrogenase activity and sensitivity to alcohol.* PHARMAC. BIOCHEM. BEHAV. 13(2) 295–297, 1980.—Young healthy Japanese men were given 0.48 g ethanol/kg body weight orally. Those responding with a marked increase in heart rate after alcohol also exhibited facial flushing and had higher acetaldehyde levels than those not responding, in spite of similar blood alcohol levels. The activity of aldehyde dehydrogenase in erythrocytes was found to correlate significantly ($r=-0.73$, $p<0.01$) with the increase in heart rate after alcohol drinking. It is suggested that erythrocyte aldehyde dehydrogenase activity affects or reflects blood acetaldehyde levels and physiological response to alcohol, and may prove useful as a marker for alcohol sensitivity in Orientals.

Aldehyde dehydrogenase Alcohol sensitivity Human erythrocytes Blood acetaldehyde

SOME of the physiological effects on the cardiovascular and central nervous system after ingestion of ethanol have been suggested to result from the formation of acetaldehyde, the first metabolite of ethanol [8]. There is also some evidence that ethnic and individual differences in sensitivity to alcohol may be related to acetaldehyde levels in the body. Those who react strongly to alcohol ingestion with such symptoms as facial flushing, increase in heart rate and blood pressure changes, have higher blood and breath acetaldehyde levels than those who are less sensitive to alcohol [1, 7, 10].

Blood acetaldehyde levels are raised by inhibition of aldehyde dehydrogenase (ALDH) with, for example, disulfiram (Antabuse), and these higher levels are associated with the unpleasant physiological effects mentioned above. Thus, individual variability in sensitivity to alcohol may in part be caused by differences in the activity of ALDH. We have previously studied [3,4] the ALDH activity in human erythrocytes and this enzyme has kinetic correlation between individual differences in sensitivity to alcohol and ALDH activity in erythrocytes.

METHOD

Sixteen healthy Japanese male volunteers (aged 22–27) were randomly selected and blood samples were collected from the median cubital vein to assay ALDH activity. Each subject was then asked to drink 3 ml/kg body weight of Japanese rice wine (16% alcohol w/v). One hour later, heart rate of each subject was counted and blood samples were again collected to determine alcohol and acetaldehyde levels. Blood ethanol and acetaldehyde were measured by gas chromatography using the head-space procedure described in [4], except that deproteinization of the sample by perchloric acid was not carried out for the determination of ethanol. ALDH activity was measured spectrophotometrically in an assay system consisting of 100 mM sodium phosphate buffer (pH 7.4), 1 mM NAD and 20 mM propionaldehyde. Hemoglobin-free samples were prepared essentially as described previously [3]. 0.8 ml of stroma-free hemolysate was applied to a 1.2×10 cm column of CM-Sephadex C-50 equilibrated with 20 mM sodium phosphate, 1 mM EDTA and 0.1% 2-mercaptoethanol (pH 6.0) and eluted with the same buffer. Because enzyme activity was found in eluate from the void volume (2 ml) to 9 ml, the 1–11 ml fractions were pooled and assayed. The assay was performed within five hours of blood collection. Loss of enzyme activity was shown to be negligible within eight hours if intact erythrocytes or hemoglobin-free preparations in the presence of EDTA and 2-mercaptoethanol were stored at 0°C. The recovery of activity was calculated to be 90% by adding a known amount of purified enzyme to the stroma-free hemolysate. Hemoglobin content was determined by the cyanmethemoglobin technique.

RESULTS AND DISCUSSION

Out of 16 tested subjects, 10 responded with a noticeable flushing of the face after drinking the wine, and this same group showed a marked increase in heart rate. Between the alcohol-responding group and the non-responding group (6 subjects) there was no difference in blood alcohol levels (Table 1). On the other hand, blood acetaldehyde level of the alcohol-responding group was higher than that in the non-responding group, and these results parallel findings reported by other workers [1, 7, 10].

A strong negative correlation ($r=-0.73$, $p<0.01$) was

TABLE 1

ETHANOL AND ACETALDEHYDE CONCENTRATIONS IN THE BLOOD OF ALCOHOL-RESPONDING AND NON-RESPONDING GROUPS

	Alcohol-responding subjects	Non-responding subjects
Number of subjects	10	6
Ethanol (μmole/ml)	9.98 ± 1.95	9.77 ± 2.17
Acetaldehyde (nmol/ml)	13.96 ± 3.66	7.85 ± 2.12

Blood samples were collected 1 hour after ethanol intake (0.48 g/kg b. wt.).

found when erythrocyte ALDH activities were plotted against the increment in heart rate (Fig. 1). These results suggest that individual variability in the signs and symptoms of alcohol drinking that are ascribed to acetaldehyde is affectded by the differences in ALDH activity, i.e. those more sensitive to alcohol have a lower activity of ALDH. The activity of ALDH in erythrocytes is lower than that in the liver, which is the major site of acetaldehyde metabolism *in vivo*. This raises the question as to what extent the erythrocyte enzyme activity reflects that of the liver. The kinetic properties and the sensitivity to inhibitors of the erythrocyte enzyme is similar to the liver cytosolic enzyme, one of the two major isozymes in the liver [3]. It is possible, therefore that the activity of the erythrocyte enzyme can serve as an index of the activity of the liver cytosolic enzyme.

Wolff [9] has shown from clinical observation that Orientals are significantly more sensitive to alcohol than are Occidentals, and he hypothesized that the ethnic differences were determined genetically rather than by social environmental factors. This hypothesis was strongly supported by a recent investigation on the polymorphism of liver ALDH: Goedde *et al.* [2] reported that about 50% of Japanese lacked the ALDH isozyme with low Km for acetaldehyde. This enzyme is presumably identical to the mitochondrial enzyme. On the other hand, all the liver samples of German subjects had both high and low Km ALDH isozymes. Thus, in Japanese and other Orientals lacking the low Km ALDH any differences in the activity of cytosolic ALDH is probably more closely related to the sensitivity to alcohol.

Furthermore, a very recent report by Jenkins and Peters [5] suggests a possible role for liver cytosolic ALDH in alcoholic Occidentals. The activity of liver cytosolic ALDH was specifically lowered in non-cirrhotic alcoholics, suggesting that higher blood acetaldehyde in alcoholics [6] may be caused by a reduction in the activity of this enzyme. It is thus possible that in healthy Occidentals too differences in activity and/or properties of cytosolic ALDH is related to sensitivity to alcohol.

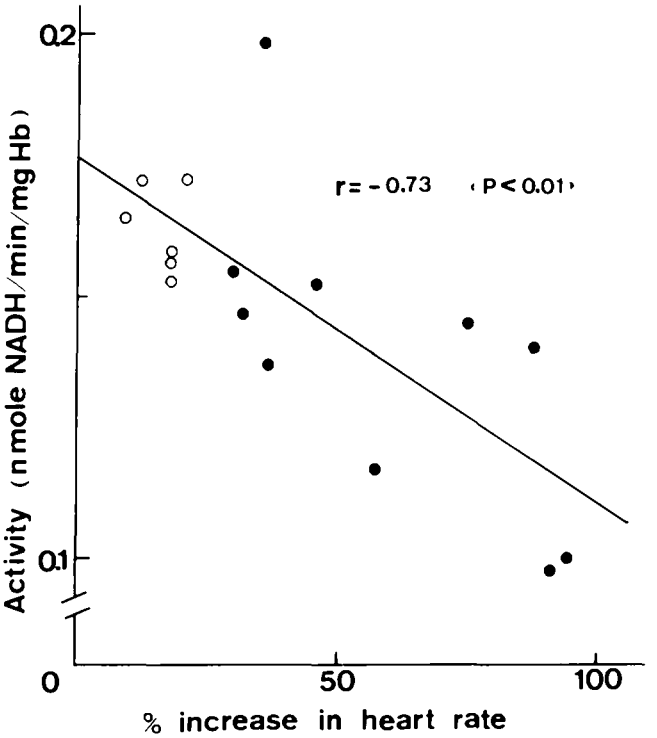

FIG. 1. Relationship between erythrocyte aldehyde dehydrogenase activity and increase in heart rate. Erythrocytes were analysed spectrophotometrically using a hemoglobin-free preparation. ●, Alcohol-responding subjects; ○, Non-responding subjects.

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