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COMMENTARY

ETHANOL METABOLISM AND ETHANOL–DRUG INTERACTIONS

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Ethanol is one of the most commonly ingested drugs in man, and its effects on the body are numerous and varied. Some of the effects of ethanol ingestion are due to a direct action of ethanol or its metabolites, while others result from modification of the metabolism of endogenous substances or drugs which share a common metabolic pathway with ethanol.

Some of the well known characteristics of ethanol are that it is highly soluble in water, rapidly absorbed by the gastrointestinal tract, not bound to plasma proteins, distributed in body water and oxidized to acetaldehyde by more than one enzyme system. This Commentary deals with methodology in the determination of rates of ethanol metabolism *in vivo*, with the controversy regarding the existence and relative importance of various pathways of ethanol oxidation, and with the mechanisms and significance of various drug–ethanol interactions.

Metabolism of alcohol

Ethanol, following its distribution in body water, is eliminated from the body in a linear fashion down to low ethanol concentrations [1]. Some early studies suggested that rates of ethanol elimination were dependent on the concentration of ethanol [2, 3]; however, more recent studies have not corroborated these observations [4, 5].

The rates of elimination of ethanol from the blood are very variable among normal individuals even when they are of the same age and have not ingested any alcohol or drugs for a period of 1 month or longer. The range of variation in rates of ethanol disappearance found in most studies has been between 12 and 20 mg/100 ml of blood/hr, which calculates to about 70–150 mg/kg body weight/hr. On the other hand, the rate of ethanol elimination determined on different days in the same individual varies little. Therefore, studies of the effect of drugs on ethanol metabolism are best designed by using each individual as his own control rather than by comparison of two groups of subjects. The best means to administer the ethanol load for the determination of the rate of elimination is by the intravenous route. Oral and intraperitoneal administration leads very often to non-reproducible results because of an erratic and delayed absorption of the ethanol. Claims that the rates of ethanol elimination from the blood were similar whether ethanol was given by the oral or intravenous route when blood samples were taken starting 1 hr after its administration [6] could not be reproduced in our laboratory because of delayed absorption resulting in many cases in peak blood levels of ethanol later than 1 hr after the ingestion of the loading dose.

Measurement of pulmonary excretion of $^{14}CO_2$ after intravenous injection of ^{14}C-ethanol is an unreliable way to estimate rates of ethanol metabolism, since it deviates and lags behind the rate of decrease of ethanol from the blood or body, and in certain experimental conditions, CO_2 production may depend on total body metabolism [7]. In addition, in studies where labeled ethanol was given after administration of unlabeled ethanol, a lag time in mixing may have accounted for initial higher hepatic concentration and metabolism of the labeled ethanol [8].

Acute administration of large doses of ethanol (1·5 g/kg and more) results in hypothermia [9]. This effect is, curiously, not prominent when the acute ethanol dose is given to animals previously treated chronically with ethanol. Since hypothermia decreases cardiac output, arterial pressure, liver blood flow [10] and the rate of ethanol metabolism [11], it is imperative that body temperature be maintained during comparative studies of rates of ethanol metabolism *in vivo* after acute ethanol loads.

Oxidation of ethanol

Ethanol is eliminated from the body mainly by metabolism in the liver, and only minimally by urinary excretion and pulmonary alveolar air [7, 12]. Three enzyme systems have been shown to oxidize ethanol to acetaldehyde *in vitro*: alcohol dehydrogenase, catalase and a microsomal ethanol-oxidizing system (Fig. 1). The existence of these separate enzyme systems and their roles in the metabolism of ethanol *in vivo* have been a matter of controversy.

Alcohol dehydrogenase

All evidence indicates that hepatic alcohol dehydrogenase is the principal enzyme responsible for ethanol elimination *in vivo*. Alcohol dehydrogenase is present

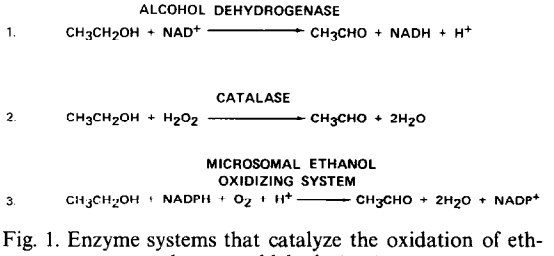

Fig. 1. Enzyme systems that catalyze the oxidation of ethanol to acetaldehyde *in vitro*.

in the soluble fraction of liver cell homogenates [13]. It is also found in other tissues in the body such as gastric and intestinal mucosa, the kidney and brain [14, 15]. However, the calculated total extrahepatic alcohol dehydrogenase activity is less than one-tenth the total activity of alcohol dehydrogenase found in the liver. Nevertheless, the ability of extrahepatic tissues to metabolize ethanol may contribute to a number of biochemical and pharmacological effects produced by ethanol ingestion. The evidence that alcohol dehydrogenase is the principal enzyme responsible for ethanol oxidation *in vivo* as follows:

(1) Its ethanol-oxidizing activity *in vitro* is higher than that of the other ethanol-oxidizing systems in both animals and man.

(2) Its K_m value *in vitro* for ethanol (1 mM) is lower than that of the other ethanol-oxidizing systems and in agreement with an apparent K_m value *in vivo* obtained from the non-linear portion of ethanol disappearance curves at low blood ethanol concentrations in animals and in man [1, 16]. Possible errors in deriving ethanol kinetics *in vivo* include consideration of the body as a single homogeneous compartment and dependence on blood flow of ethanol extraction by the liver [17].

(3) Pyrazole, a very effective competitive inhibitor of liver alcohol dehydrogenase *in vitro* [18], also markedly inhibits ethanol metabolism *in vivo* [19, 20]. High doses of pyrazole administered to rats (7·0 m-moles/kg) results in 89 per cent inhibition of the rate of ethanol elimination *in vivo* [19]. Pyrazole (2 mM) binds to hepatic microsomes, producing a typical type 2 spectrum, and inhibits pentobarbital and aniline hydroxylation [21]; however, pyrazole *in vitro* has no [22] or only a slight inhibitory effect [23] on the microsomal ethanol-oxidizing system, and no effect on catalase [24]. Pyrazole *in vivo* was shown to have no effect on the microsomal ethanol-oxidizing system at 23 hr after its administration in one study [25], while it resulted in moderate inhibition in another study [26]. It has a delayed inhibitory effect on catalase which reaches a maximum 28 hr after administration and is probably caused by an as yet unidentified metabolite of pyrazole [24]. Such delayed inhibition would be expected to contribute little, if at all, to the inhibition of ethanol elimination *in vivo* by pyrazole. Further studies of the enzyme activities at various time intervals after pyrazole administration may provide more definitive information about the use of this compound in the study of ethanol metabolism.

(4) The demonstrated decreases in hepatic NAD^+/NADH ratios after ethanol administration in the perfused liver [27] and *in vivo* [28, 29] are compatible with ethanol oxidation by alcohol dehydrogenase, although oxidation of acetaldehyde by aldehyde dehydrogenase could also account for these changes. The $NADP^+$/NADPH ratio also falls but returns to normal earlier than the NAD^+/NADH ratio [30]. It is tempting to speculate that the rapid return of the low $NADP^+$/NADPH ratio to normal may be due to NADPH-mediated microsomal ethanol oxidation. However, the concentration of ethanol measured in the liver at the time of the observed return of the ratio to normal was below the K_m for ethanol of the microsomal ethanol-oxidizing system so that optional oxidation of ethanol by this system could not have been achieved.

The principal role of alcohol dehydrogenase in ethanol oxidation *in vivo* has been challenged on the basis of a pH optimum for the enzyme *in vitro* of 10·6, and too low an activity at physiologic intracellular pH to account for known rates of ethanol elimination *in vivo* in animals and in man [25]. These arguments have not been validated by this investigator after measurements of alcohol dehydrogenase activity. In man, the mean activity of the liver enzyme was 1·5 μmoles/mg of protein/hr at pH 7·0 and 37°. Since the mean concentration of protein in the liver is about 100 mg/g of wet liver weight and the total liver weight in an adult man is estimated to be 1·2 kg, the calculated average total liver alcohol dehydrogenase activity is 180 m-moles (8·3 g)/hr. This corresponds to 118 mg/kg body weight/hr in a 70-kg man, which is the range of known rates of ethanol metabolism *in vivo*. Similar determinations in a 200-g rat reveal a hepatic alcohol dehydrogenase activity of 0·55 μmole/mg of protein/hr at pH 7·0 and 37°. The total alcohol dehydrogenase activity calculated from a protein concentration of 100 mg/g of liver wet weight and a liver weight of 8·0 g is 440 μmoles (20·2 mg)/200-g rat/hr or 110 mg/kg body weight/hr. The total maximum activity of alcohol dehydrogenase *in vitro* in the rat admittedly accounts for about only 50 per cent of the known rate of ethanol elimination *in vivo*, and leaves room for ethanol oxidation by other enzyme systems.

Catalase

The ability of catalase to oxidize ethanol *in vitro* in the presence of a hydrogen peroxide-generating system was first demonstrated by Keilin and Hartree [31]. Catalase, however, has not been considered to be of importance in ethanol metabolism *in vivo*, since administration of 3-amino-1,2,4-triazole, a potent inhibitor of catalase, has no effect on ethanol metabolism *in vivo* or by tissue slices *in vitro* [32]. These observations contrast with the effect of 3-amino-1,2,4-triazole in inhibiting metabolism of methanol *in vitro* [33] and *in vivo* in a number of animals [34].

Microsomal ethanol-oxidizing system

The enzymatic nature and the role of ethanol oxidation *in vivo* by microsomes has been a matter of extensive study and controversy since its initial description by Orme-Johnson and Ziegler [35]. It was originally suggested that ethanol oxidation by the microsomes was mediated through the mixed function oxidase system which metabolizes a number of drugs, since it required NADPH and oxygen and was inhibited by carbon monoxide [25, 36]. In further support of this concept was the observation that ethanol could bind to cytochrome P-450 producing a modified type 2 spectrum [21]. Furthermore, sodium cyanide, sodium azide and the administration of 3-amino-1,2,4-triazole (all inhibitors of catalase) and pyrazole (an inhibitor of alcohol dehydrogenase) produced only slight inhibition of the NADPH-dependent ethanol-oxidizing activity by the microsomes, suggesting that neither of these contaminating enzymes was responsible for the observed ethanol-oxidizing acti-

vity [25]. Other investigators, however, observed that substitution of NADPH by a system generating H_2O_2 resulted in comparable oxidation of ethanol by the microsomes [37, 38]. In view of this and the finding of a more significant inhibition by catalase inhibitors than found by Lieber and DeCarli [25, 36], it was postulated that ethanol was oxidized by catalase in the presence of H_2O_2 generated from NADPH and oxygen by NADPH oxidase. Subsequently, Thurman *et al.* [26] provided further support for this mechanism by demonstrating sufficient H_2O_2 formation from NADPH by microsomes, and the same Michaelis–Menten constant for ethanol oxidation with both the NADPH- and H_2O_2-generating systems, as well as similar inhibition by catalase inhibitors, by a peroxidating substrate of catalase, and by a H_2O_2-utilizing process in the presence of both generating systems. Oshino *et al.* [39] pointed out that variabilities in the results by various investigators regarding the effect of catalase inhibitors on ethanol oxidation by microsomes was related to both the amount of H_2O_2 generated and the amount of contaminating catalase, and more specifically to the ratio between the two. A low ratio between the H_2O_2 generated and catalase concentration favors the peroxidatic reaction, while a high ratio favors a catalatic reaction. The catalase inhibitor would be expected to be less effective at a low ratio when the generation of H_2O_2 is low and rate limiting, as in the case of endogenous H_2O_2 generation from NADPH and oxygen by NADPH oxidase, than at a high ratio when an exogenous H_2O_2-generating system is used and catalase concentration is rate limiting. Therefore, catalase inhibitors cannot be used in determining the relative contributions of catalase or other enzyme systems to ethanol oxidation by microsomes unless the ratio of the H_2O_2 production to catalase is defined in each experiment.

A report [40] that hepatic microsomes from mutant mice, whose catalase had been rendered inactive by heat, failed to oxidize ethanol appeared to support the concept that catalase contamination by the microsomes was responsible for ethanol oxidation by microsomes. However, these findings were shown to be due to the use of an insensitive method of determining ethanol oxidation (measurement of ethanol disappearance in the reaction mixture), since subsequent experiments in which acetaldehyde production was measured revealed that microsomes virtually devoid of catalase were indeed able to oxidize ethanol at rates similar to those found with microsomes of normal mice [41]. While a number of the studies described provided new evidence for the ability of catalase present in the microsomes to participate in the oxidation of ethanol, they did not resolve the question as to whether or not catalase was the principal or only mediator of the NADPH-dependent ethanol oxidation by microsomes. Indeed, more recent studies show that ethanol oxidation is catalyzed by a microsomal component rich in cytochrome P-450 and separate from contaminating alcohol dehydrogenase and containing only trace amount [23] or no catalase [42]. The cytochrome P-450 fraction contained small amounts of cytochrome *c* reductase and lipid. Addition of extra small amounts of cytochrome *c* reductase enhanced the ethanol-oxidizing activity, while addition of large amounts of cytochrome *c* reductase or of lipid resulted in inhibition of the ethanol-oxidizing activity [23]. A more purified fraction of cytochrome P-450 prepared by Barakat *et al.* [43], free of cytochrome *c* reductase, had only a trace of ethanol-oxidizing activity. These later experiments suggested that cytochrome P-450 by itself was inactive in oxidizing ethanol; however, the results obtained by mixing the cytochrome P-450 and cytochrome *c* reductase fractions cannot be interpreted, since neither the amounts of cytochrome *c* reductase added nor the amounts of lipid present in the fractions were defined, and as pointed out above, an excess of either results in inhibition of the ethanol-oxidizing activity. Addition of excess lipid to fractions of cytochrome P-450 and cytochrome *c* reductase have also been shown to inhibit ω-hydroxylation of fatty acids by these components [44]. A possible mechanism for cytochrome P-450-mediated ethanol oxidation is that cytochrome P-450 acts like a peroxidase, as has been demonstrated for the hydroxylation of benzphetamine and ethylmorphine in the presence of a superoxide-generating system [45]. In support of this mechanism is our observation of ethanol oxidation by the cytochrome P-450 microsomal fraction in the presence of either a H_2O_2- or an NADPH-generating system [23]. In the case of the NADPH-generating system, hydrogen peroxide would be produced from NADPH and oxygen, by NADPH oxidase. However, recently Teschke *et al.* [22] have observed ethanol oxidation by their microsomal fraction with the NADPH-, but not with the H_2O_2-generating system. Two observations are at variance with ethanol oxidation by mixed function oxidase: (1), the concomitant inhibition of ethylmorphine demethylation and cytochrome *c* reductase, but not of ethanol oxidation, by adding increasing amounts of antibody to cytochrome *c* reductase [46], and (2) the lack of inhibition of ethanol oxidation by SKF-525A [47, 48]. These differences suggest that there may exist another enzyme system in the microsomes which does not require cytochrome P-450, as was shown to be the case for the mixed function amine oxidase [49]. Further purification of the different microsomal fractions followed by reconstitution experiments and possibly the use of antibodies against the various components will be required to resolve the existing controversy of the enzymatic nature of ethanol oxidation by the microsomes.

The possible role of the microsomal ethanol-oxidizing system *in vivo* is also a matter of controversy that remains unresolved. Lieber and DeCarli [50] from data *in vitro* estimated that the microsomal ethanol-oxidizing system could account for 20–25 per cent of ethanol oxidation in rats. Studies with pyrazole in which alcohol dehydrogenase activity *in vitro* is fully inhibited demonstrate a maximum of 90 per cent inhibition of ethanol elimination *in vivo*, suggesting some oxidation of ethanol by another enzyme [19]. Lieber and DeCarli [50] extended the above observation further by demonstrating that the K_m and V_{max} values *in vivo* obtained from the non-linear portion of the ethanol disappearance curve after pyrazole administration approximated the values for K_m of 8·6 mM and V_{max} of 29·2 μmoles/kg body weight/min for the microsomal ethanol-oxidizing system.

Studies showing a lack of effect of the administration of SKF-525A on rates of ethanol metabolism *in vivo* in animals [47, 48] have been presented as evidence against a role of microsomal ethanol oxidation *in vivo*. However, studies *in vitro* have shown that ethanol oxidation [25], as well as the oxidation of some other lipid-soluble compounds [51], is not inhibited by SKF-525A. The role of the microsomal ethanol-oxidizing system has been postulated to be greater at higher ethanol concentrations because its K_m for ethanol is higher than that of alcohol dehydrogenase. In support of this hypothesis are the findings of Thieden [52] of higher rates of ethanol oxidation by liver slices when the concentration of ethanol was raised from 4 to 80 mM. Furthermore, pyrazole appeared to be a more effective inhibitor of ethanol oxidation at low than at high ethanol concentrations. In addition, administration of ethanol *in vivo* at high and low doses with pyrazole revealed an increased malate/pyruvate ratio with the high as compared with the low dose of ethanol, which is compatible with NADPH-dependent ethanol oxidation at the high ethanol dose [53]. The $NADP^+$ formed may be reduced by malic enzyme with conversion of malate to pyruvate, which in turn is carboxylated and reduced back to malate. The end result is an increase in the malate/pyruvate ratio, since malic enzyme is the enzyme with the lowest activity in this cycle. Also, in support for a function *in vivo* of the microsomal ethanol-oxidizing system in man was the recent observation that blood acetaldehyde levels, which were constant at very high blood ethanol levels (33–54 mM), declined sharply as ethanol concentration fell to 24 mM (110 mg/100 ml) [54]. At this latter ethanol concentration, the microsomal ethanol-oxidizing system, but not alcohol dehydrogenase, would become unsaturated, resulting in decreased acetaldehyde production from ethanol.

Induction of the microsomal ethanol-oxidizing system has been postulated to be the mechanism for increases in the rates of ethanol metabolism in chronic alcoholics after recent heavy ethanol ingestion [6, 55] and in rats after ethanol administration [56, 57], since the increases in the rates of ethanol elimination were associated with increases in the NADPH-dependent ethanol-oxidizing activity but no changes in alcohol dehydrogenase. However, after withdrawal of alcohol in man [55] and rats [58] the rates of ethanol disappearance from the blood fell rapidly to normal, while the NADPH-dependent ethanol-oxidizing activity remained elevated for a prolonged time. The lack of parallelism between the fall in the rate of ethanol elimination and the activity of the microsomal ethanol-oxidizing system suggests that factors other than the increase in enzyme activity cause the changes in the rate of ethanol oxidation after ethanol administration. Both hepatic blood flow and the availability of coenzymes may regulate the rates of ethanol degradation. Studies of the effect of acute administration of ethanol on hepatic blood flow have resulted in contradictory results [59–61], while the effect of chronic administration of ethanol on hepatic blood flow remains unknown. The availability of coenzymes is probably more important. In the case of ethanol oxidation by alcohol dehydrogenase, studies *in vitro* show that the dissociation of the alcohol dehydrogenase–NADH complex is rate limiting [62]. Also, mitochondrial uncoupling agents, such as dinitrophenol, which increase the ability of mitochondria to oxidize NADH, have been shown to increase the rate of ethanol metabolism by rat liver slices [63]. Furthermore, studies *in vitro* demonstrate that stimuli to increased reoxidation of NADH, such as elevated pyruvate concentrations in children with type I glycogen storage disease [64], or infusion of fructose in dogs [65] and man [66], enhance the rate of ethanol metabolism.

Effects of ethanol on drug metabolism

The administration of ethanol can result in inhibition or acceleration of drug metabolism depending on the dosage and time interval between the administration of the ethanol and the drug. Inhibition occurs from direct interference of alcohol with the metabolism of the drug, while acceleration of drug metabolism is due to induction of drug-metabolizing enzymes by ethanol, and is observed after discontinuation of prolonged ethanol administration.

Ethanol, as mentioned before, binds to cytochrome P-450, producing a modified type 2 spectrum [21], and inhibits the reduction of cytochrome P-450 by NADPH which is a characteristic of type 2 binding substances [67]. It has also been shown to inhibit the binding to cytochrome P-450 of aniline, a type 2 binder, but not of hexobarbital, a type 1 binder [68]. As would be expected from these observations, ethanol is a stronger inhibitor of metabolism by microsomes of type 2 than of type 1 binding drugs [69]. Ethanol *in vitro* is a competitive inhibitor of aniline and pentobarbital hydroxylases, and a mixed inhibitor of the demethylation of aminopyrine and ethylmorphine [70], and alters the metabolism of certain drugs such as 4-hydroxyphenazon and 4-aminoantipyrine from the major oxidative to the minor conjugative pathway [71]. Ethanol has also been demonstrated to inhibit drug metabolism *in vivo*. It inhibits the rates of disappearance of phenobarbital and meprobamate from the blood of humans [70] and the rate of phenobarbital disappearance from the blood in animals [71]. Ethanol also decreases the urinary excretion of *p*-hydroxyphenobarbital and increases the amounts of phenobarbital in tissues after phenobarbital administration in animals [72, 73]. The concomitant administration of ethanol and drugs such as the barbiturates and chloral hydrate results in an enhanced depressant effect of these drugs on the central nervous system. The effect of ethanol and chloral hydrate given in combination is greater than the simple summation of the effects obtained when either drug is given alone. This potentiation of effect appears to be due to the mutual inhibition of the metabolism of these drugs [74].

Ethanol is an inducer, not only of its own metabolism but also of the metabolism of a number of drugs. In animals, its administration results in increases of the smooth endoplasmic reticulum, cytochrome P-450, cytochrome *c* reductase, and of the activity of a number of microsomal enzymes [57, 75, 76]. In humans, ethanol administration was shown to induce pentobarbital hydroxylase, but not benzpyrene hydroxylase [77].

Ethanol differs from other inducers, such as phenobarbital and polycyclic hydrocarbons, in that it fails to stimulate liver growth and in that it is a relatively weak inducer [57]. The increases in enzyme activity after ethanol administration are of the order of 1-fold and often only significant when expressed per g of wet liver weight and not when expressed per mg of microsomal protein. This contrasts with 3- to 10-fold increases in enzyme activities, expressed per mg of microsomal protein, induced by other compounds. Increases in microsomal enzyme activity in rat occur as early as 48 hr and are maximal after 2–3 weeks of ethanol administration. However, this author has observed that, after more prolonged administration of ethanol (6 weeks), increases in the activities of the microsomal enzymes are no longer apparent. This lack of demonstrable induction coincides with the development of increasing hepatic fatty infiltration and hepatocellular injury due to prolonged ethanol administration, which is not a feature of prolonged administration of other drugs such as phenobarbital.

The increases in metabolism *in vitro* of drugs after ethanol administration have been demonstrated to be accompanied by increases in the metabolism *in vivo* of certain drugs such as meprobamate in aminals [78] and of meprobamate and pentobarbital [78] and antipyrine [79] in man. Also, increases in the rates of metabolism of commonly used drugs such as tolbutamide [80, 81], warfarin and diphenylhydantoin [81] were shown in chronic alcoholic patients after heavy alcohol intake. Rats which become tolerant to alcohol have cross tolerance to some drugs such as pentobarbital and hexobarbital. However, in one study, the increased tolerance to the above drugs was not associated with an increase in their metabolism, suggesting that decreased central nervous system sensitivity to drugs may be another mechanism, in addition to increased metabolism, for the increased tolerance to drugs after ethanol ingestion [82].

Effect of drugs on ethanol metabolism

In contrast to the ready inhibition of drug metabolism by ethanol, the inhibition of ethanol metabolism by drugs is uncommon. The reason for this is that most drugs are metabolized by microsomal enzymes while ethanol, as mentioned before, is principally oxidized by alcohol dehydrogenase. Therefore, inhibition of ethanol metabolism usually occurs only when the drug or a metabolite of the drug administered is a substrate and or an inhibitor of alcohol dehydrogenase. Chlorpromazine is an example of a drug which is an inhibitor of alcohol dehydrogenase and whose administration results in inhibition of ethanol metabolism [83]. Chloral hydrate is another well known inhibitor of ethanol metabolism. In this latter case, the inhibition is due to its metabolite trichloroethanol, which is a substrate and a competitive inhibitor of ethanol oxidation by alcohol dehydrogenase [84]. In one study, phenobarbital, a drug metabolized by microsomal enzymes, was observed to decrease 1-^{14}C-ethanol metabolism to $^{14}CO_2$ by liver slices and to inhibit alcohol dehydrogenase activity [85]. However, more recent studies have not demonstrated any changes in the rates of ethanol disappearance from the blood 24 hr after phenobarbital administration, when phenobarbital was still detectable in the blood, nor any inhibition of alcohol dehydrogenase *in vitro* by phenobarbital concentrations ranging from 0·1 to 1·0 mM [50, 72].

A number of drugs have been shown to enhance the rates of ethanol metabolism. The principal mechanisms postulated for their action have been: (1) increase in the activity of the microsomal ethanol-oxidizing system; (2) increases in hepatic blood flow and liver weight; and (3) increase in the reoxidation of reducing equivalents.

(1) Phenobarbital was shown to increase the rates of ethanol disappearance from the blood and the activity of the microsomal ethanol-oxidizing system in animals, if sufficient time is allowed between the administration of the last dose of phenobarbital and the determinations for the drug to clear the blood [50]. The time interval for clearance of a dose of 100 mg/kg body weight was 48 hr. Earlier determinations by this author [86] and other investigators [87] showing no increase in the rates of ethanol disappearance from the blood or in the enzyme activity were done 24 hr after the last dose of phenobarbital. However, in one recent animal study in which a 48-hr interval was used, the increase in the activity of the microsomal ethanol-oxidizing system was not accompanied by an increase in the rate of ethanol disappearance from the blood [88]. The discrepancy in results between the studies may be related to the dosage and duration of phenobarbital administration or to the sex of the animals used. In the study showing increases in the rates of ethanol disappearance from the blood, the phenobarbital was given in a dose of 80 mg/kg by the intraperitoneal route for 4 days to female rats [50], while in the study showing no increase in the rate, the phenobarbital was given in a dose of 100 mg/kg orally for 1 week to male rats [88]. The surprising finding is that the increase in the activity of the microsomal ethanol-oxidizing system was greater in the study failing to show an increase in the rate of ethanol disappearance from the blood than in the one showing an increase in the rate. No levels of phenobarbital were detectable at the time of the determinations in either study. In man, phenobarbital administration resulted in increases in the rates of ethanol disappearance from the blood but in no changes in the activities of either alcohol dehydrogenase or the NADPH-dependent ethanol-oxidizing activity in liver homogenates [89]. The discrepancies between the changes in the rates of ethanol disappearance and the induction of the microsomal ethanol-oxidizing activity after the administration of phenobarbital suggest that a mechanism other than the increase in the microsomal activity is responsible for the increases in the rates of ethanol metabolism. Phenobarbital may increase ethanol metabolism by its effects of increasing liver weight [90], hepatic blood flow [91] and biliary flow [92].

(2) Increases in the rates of ethanol disappearance after clofibrate administration were found to be directly proportional to increases in liver size produced by the drug [93]. No increases in alcohol dehydrogenase activity were found when the enzyme was expressed per g of liver and, unfortunately, the activity of the microsomal ethanol-oxidizing system was not determined. Prior studies showing the ability of

clofibrate to increase the capacity of the liver to utilize reducing equivalents [94] were not confirmed.

(3) Fructose has been the most effective and consistent compound to increase the rates of ethanol disappearance from the blood in animals [65, 95] and man [96–98]. The amounts of fructose that have to be administered to obtain an increase in ethanol metabolism are large, ranging from 1 to 2 g/kg body weight. In studies showing no effect of fructose on ethanol metabolism, smaller doses were given [99]. The postulated mechanism accounting for the acceleration of rates of ethanol elimination is dissociation of the alcohol dehydrogenase–NADH complex by glyceraldehyde formed from fructose. In support of this mechanism are studies showing increases in ethanol oxidation by liver slices on the addition of D-glyceraldehyde, increases in the formation of glycerol and α-glycerophosphate from fructose in the presence of ethanol due to reduction of glyceraldehyde by the alcohol dehydrogenase–NADH complex [100], and abolition of the fructose effect by pyrazole [101].

Conclusions

Ethanol is principally oxidized to acetaldehyde by alcohol dehydrogenase and to a lesser extent by the microsomal ethanol-oxidizing system. The contribution of the microsomal system to ethanol oxidation is greater at high ethanol concentrations, but whether or not it is a factor for the observed increased metabolism and tolerance to ethanol after ethanol ingestion is still a matter of controversy. A very significant aspect of ethanol oxidation by the microsomes is that it influences the microsomal metabolism of a number of drugs. Inhibition of drug metabolism results from direct interference by ethanol, while acceleration of drug metabolism is due to induction of microsomal enzymes by ethanol and becomes apparent after discontinuation of its ingestion. Since alcoholic patients are frequently given tranquilizer drugs for treatment of their alcoholism, and in addition, receive drugs for the treatment of other diseases, the effects of ethanol on drug metabolism have obvious implications. Some drugs, in turn, have been shown to inhibit or accelerate ethanol metabolism. Inhibition of ethanol metabolism caused by drugs is usually due to their inhibition of alcohol dehydrogenase activity. On the other hand, the possible mechanisms for the acceleration of ethanol metabolism by drugs are varied and less well defined and include increases in the microsomal ethanol-oxidizing activity, in hepatic blood flow and liver weight and in the reoxidation of reducing equivalents. Further research on the effects of drugs on ethanol metabolism is bound to provide further knowledge of the pathogenesis of alcohol-associated metabolic changes and alcoholism and aid in the discovery of new therapeutic approaches.

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