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RELATIONSHIP BETWEEN CYTOCHROME P450 2E1 AND ACETONE CATABOLISM IN RATS AS STUDIED WITH DIALLYL SULFIDE AS AN INHIBITOR

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Abstract—Previous studies have demonstrated that cytochrome P450 2E1 (P450 2E1) catalyzes the oxidation of acetone *in vitro*. The present study was designed to determine the importance of P450 2E1 in the catabolism of acetone in rats using diallyl sulfide (DAS) as an inhibitor of this enzyme. After a single intragastric dose of DAS, blood samples were collected from rats at different time points, and blood acetone concentrations were measured by gas chromatography. In a low DAS dose (50 mg/kg body weight) group, the maximum acetone level of 6-fold higher than the normal level was reached at 6 hr; the acetone level returned to normal at 48 hr. In a high dose (200 mg/kg) group, the maximum acetone level of 9-fold higher than the normal level was reached at 12 hr; the acetone level returned to normal at 60 hr. The turnover time and fractional turnover rate of elevated acetone were 15.8 ± 0.5 hr and 0.054 ± 0.001 hr^{-1}, respectively, for the low dose, and 19.2 ± 0.6 hr and 0.046 ± 0.005 hr^{-1}, respectively, for the high dose. In a chronic experiment, DAS (50 and 200 mg/kg, i.g.) was given to rats daily for 29 days, and elevated blood acetone levels were observed during the entire experimental period: 2.0 to 2.8 μg/mL for the low dose and 3.4 to 3.9 μg/mL for the high dose at 24 hr after the 1st, 7th, 14th and 28th doses versus 0.8 to 0.9 μg/mL for the control. The increase of blood acetone level was closely related to the decreases of *N*-nitrosodimethylamine (NDMA) demethylase activity and P450 2E1 content in liver microsomes. Consistent with the lack of cumulative effect from the multiple doses of DAS on acetone level, rather stable levels of the DAS metabolites, diallyl sulfoxide (45.0 μg/mL, range: 33.8 to 58.6 μg/mL) and diallyl sulfone (11.7 μg/mL, range: 6.9 to 15.6 μg/mL), were observed at 24 hr after the 1st, 7th, 21st and 28th doses with DAS (200 mg/kg) in the chronic experiment. It is likely that the inactivation and inhibition of P450 2E1 by DAS and its metabolites block the oxidation of acetone and cause its elevation in blood. The results strongly suggest an important role of P450 2E1 in acetone catabolism under physiological conditions.

Key words: cytochrome P450 2E1; acetone metabolism; diallyl sulfide; diallyl sulfone; suicide inhibition; *N*-nitrosodimethylamine demethylase

P450 2E1† is well recognized for its role in the bioactivation of many environmental chemicals to toxic and carcinogenic agents [1–3]. Acetone, one of the ketone bodies produced under ketogenic conditions such as fasting or diabetes, is both an inducer and a substrate of P450 2E1 [1, 3]. The existence of a gluconeogenic pathway for acetone was demonstrated by the incorporation of [^{14}C]-labeled acetone into glucose and amino acids during prolonged fasting and diabetic ketoacidosis [4–6]. The role of P450 2E1 in catalyzing the conversion of acetone to acetol and then to methylglyoxal, both intermediates in the gluconeogenic pathway, has been demonstrated *in vitro* [7–9]. Under normal physiological conditions, however, the importance of P450 2E1 in the biotransformation of acetone was still unclear.

If P450 2E1 plays a vital role in catalyzing the oxidative metabolism of acetone, inactivation of this enzyme would be expected to elevate the level of acetone in the body. DAS, a flavor compound derived from garlic, is a potent inactivator and competitive inhibitor of P450 2E1 [10, 11]. Studies on DAS metabolism in rat liver microsomes and *in vivo* indicate that DAS is first converted to DASO and then to $DASO_2$ [11]. All three compounds are competitive inhibitors of P450 2E1, and $DASO_2$ is a suicide inhibitor of this enzyme [11]. Treatment of rats with one dose of DAS results in a marked decrease of hepatic NDMA demethylase activity as well as of the P450 2E1 level [10]. The effects of chronic treatments, however, are not known.

In the present study, DAS was employed as an inhibitor of P450 2E1 to study its role in acetone catabolism in nonfasting rats. The effects of a single dose and repeated doses of DAS on the blood acetone level in rats were examined. The effects of chronic DAS treatments on P450 2E1-mediated NDMA demethylase activities in microsomes of liver, kidney, and lung as well as on the hepatic P450 2E1 level were examined. The blood levels of the DAS metabolites, DASO and $DASO_2$, were also analyzed in these studies.

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† Abbreviations P450 2E1, cytochrome P450 2E1; DAS, diallyl sulfide; DASO, diallyl sulfoxide; $DASO_2$, diallyl sulfone; NDMA, *N*-nitrosodimethylamine; PB, phenobarbital; DSF, disulfiram.

MATERIALS AND METHODS

Animals and sample collection. Male Sprague–Dawley rats (body weight approximately 250 g) were obtained from Taconic Farms (Germantown, NY) and maintained in air-conditioned quarters with 12-hr light–dark cycles. They were given a rat chow (Ralston Purina Co., St. Louis, MO) and water *ad lib*. Treatment of rats started after a week's acclimation. For a single dose treatment with DAS, rats were randomly divided into two groups and DAS was given intragastrically at a dose of 50 or 200 mg in 4 mL corn oil per kg body weight. Blood was taken into heparinized tubes from the orbital sinus [12] before dosage and 6, 12, 24, 48 and 60 hr after DAS administration. For chronic treatment with DAS, rats were randomly divided into 3 groups with 5 or 6 rats in each group. DAS was given intragastrically to the treatment groups at a daily dose of 50 or 200 mg/kg for 8 days. Rats in the control groups were given an equal volume of corn oil (4 mL/kg). In a second experiment, the rats were treated similarly for 29 days. Blood samples were taken at 24 hr after the 1st, 7th, 14th, 21st and 28th doses of DAS for measuring the concentrations of acetone and the metabolites of DAS. Rats were decapitated at 24 hr after the 8th or 29th dose of DAS. Immediately after the rats were killed, liver, lung and kidney were removed, frozen on dry ice, and stored at −70° until used.

Analysis of acetone level. Acetone in blood was analyzed using head-space gas chromatography as described by Eriksson [13] and DeMaster and Stevens [14] with some modifications. In brief, duplicate samples were prepared for analysis by the addition of 0.2 mL of blood to 0.6 mL of cold 0.72 N $HClO_4$. After mixing, the protein precipitate formed was removed by centrifugation at 16,000 *g* for 10 min at 4°. Two hundred microliters of the protein-free supernatant fraction was neutralized with 50 μL of 4 N NaOH, sealed in the septum vials, and stored at 4° until analyzed. Acetone was measured by GC on a Tenax 60/80 mesh column ($\frac{1}{8}$ in. × 6 ft, Alltech Inc., Deerfield, IL). The GC system consisted of an HS-101 head-space autosampler and an 8500 gas chromatograph (Perkin-Elmer Co., Norwalk, CT). Operating conditions for the head-space autosampler were: sample temperature, 90°; needle temperature, 120°; transfer temperature, 120°. Operating conditions for gas chromatography were: carrier gas (helium), 1 mL/min; air, 240 mL/min; H_2, 20 mL/min; oven, 110°. Linearity and recovery (100 ± 3%) were established.

An area under the concentration versus time curve (AUC) and an area under the moment versus time curve (AUMC) for increased acetone level between 0 and 60 hr after DAS treatment were calculated using a trapezoidal rule. The turnover time (t_t) was obtained by dividing $AUC_{0\text{-}60}$ with $AUMC_{0\text{-}60}$. Fractional turnover rate (k_t) was calculated using the plot of the logarithm of the elimination concentration versus time [15].

Measurement of DAS metabolites in the plasma. The plasma sample (200 μL) was precipitated by the addition of 20 μL of 25% (w/v) $ZnSO_4$ and then 20 μL of saturated $Ba(OH)_2$. The mixture was centrifuged at 16,000 *g* for 5 min, and 20 μL of the supernatant was analyzed by HPLC on a Supelcosil LC-ABZ column (4.6 × 250 mm, 5 μm, Supelco Inc., Bellefonte, PA) for DASO and $DASO_2$. A Waters 840 HPLC system was used. It consisted of a model 510 pump, a model 712 WISP autoinjector, a model 490E programmable multi-wavelength detector set at 232 nm for DASO and at 205 nm for $DASO_2$, and a model 380 digital professional data system. The mobile phase consisted of 900 μL of tetrahydrofuran in 1000 mL of methanol and water (8:92, v/v). The flow rate was 1.5 mL/min. Under these conditions, the retention times were 5.0 min for DASO and 6.5 min for $DASO_2$. The limits of detection for DASO and $DASO_2$ were estimated to be 1 μg/mL.

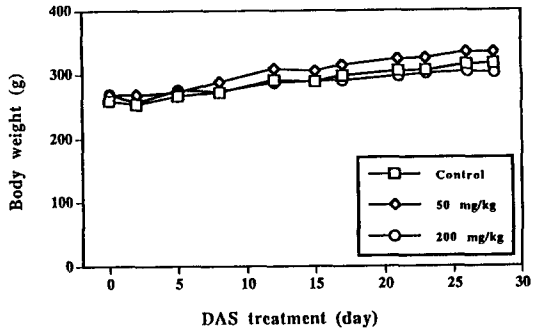

Fig. 1. Effect of chronic DAS treatment on the body weights of rats. DAS was given to rats intragastrically at a daily dose of 50 or 200 mg in 4 mL corn oil/kg body weight for 29 days. Rats in the control group were given an equal volume of vehicle. Each point represents the mean of 5 or 6 animals.

Other methods. Liver, lung and kidney microsomes were prepared as previously described [16]. NDMA demethylase activity was determined as previously described [10], with a substrate concentration of 0.1 mM NDMA. The P450 2E1 content in liver microsomes was determined by immunoblotting [17] with some modifications. In brief, microsomal protein (3.0 μg/well) was separated by polyacrylamide gel electrophoresis in the presence of sodium dodecyl sulfate. Proteins were then transferred to a nitrocellulose sheet according to the method of Towbin *et al.* [18]. Antibody raised against rat P450 2E1 was used as the first antibody, and the alkaline phosphatase-conjugated goat anti-rabbit antibodies were used as the second antibody in the staining reaction. The intensity of the immunoblot bands was quantified using a Shimadzu dual-wavelength CS-930 TLC scanner (Shimadzu Scientific Instruments, Inc., Columbia, MD).

All results are expressed as means ± SD. Statistical analysis was performed by the Newman–Keuls range test. P values of <0.05 were considered significant.

RESULTS

General observations. In the acute experiment, the rats did not show any obvious inactivity or signs

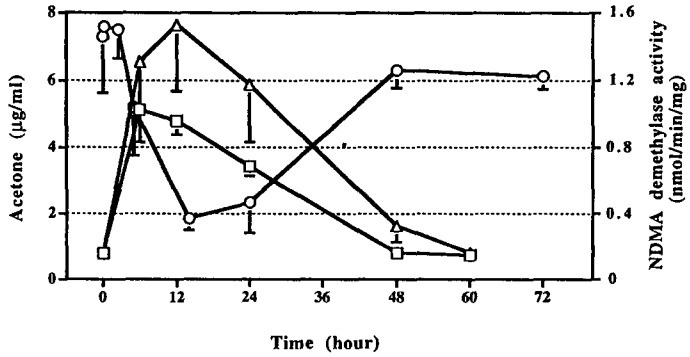

Fig. 2. Blood acetone levels and microsomal NDMA demethylase activity after a single oral dose of DAS. DAS (50 mg/kg, □—□; 200 mg/kg, △—△) was administrated to rats intragastrically. Blood was taken at 0, 6, 12, 24, 48 and 60 hr, and the acetone level was measured by head-space gas chromatography. Each data point represents the mean ± SD of 5 individual animals. The data for NDMA demethylase activity in rat liver microsomes are from a previous study with a dose of DAS of 200 mg/kg (○—○) under similar experimental conditions [10].

Table 1. Kinetic parameters of acetone accumulation and elimination after a single dose of DAS to rats

Dose (mg/kg)	C_0 (μg/mL)	C_6 (μg/mL)	C_{12} (μg/mL)	AUC_{0-60} [(μg·hr)/mL]	t_t (hr)	k_t (hr^{-1})	Recovery time (hr)
50	0.80	5.13	4.80	109.47	15.84	0.0538	48
	(0.09)	(0.98)	(0.41)	(11.34)	(0.47)	(0.0010)	
200	0.78	6.55	7.06*	199.77*	19.19*	0.0463*	60
	(0.05)	(2.39)	(2.12)	(73.01)	(0.60)	(0.0051)	

DAS (50 or 200 mg/kg) was administered to rats intragastrically in corn oil. Blood was taken at 0, 6, 12, 48 and 60 hr, and acetone levels were measured by head-space gas chromatography. C_0, C_6 and C_{12} are the concentrations of blood acetone at 0, 6 and 12 hr after DAS administration, respectively; AUC_{0-60} is the area under the curve for acetone concentration from 0 to 60 hr; t_t and k_t are turnover time and fractional turnover rate, respectively. Data are means (with SD in parentheses) of 5 individual animals.
* $P < 0.05$, compared with the 50 mg/kg group.

of toxicity after one dose of DAS (50 or 200 mg/kg, i.g.). In the chronic experiments, we could smell DAS in the rats treated with DAS. The rats in the treatment groups maintained normal drinking, eating and other behaviors. No signs of toxicity were observed. The growth rates of rats in the treated groups were the same as that in the control group (Fig. 1).

Elevation of acetone level by a single dose of DAS. Blood acetone levels were measured in rats prior to treatment and at 6, 12, 24, 48 and 60 hr following a single oral low (50 mg/kg) or high (200 mg/kg) dose of DAS. Both DAS-treated groups showed a time-dependent increase in the level of circulating acetone (Fig. 2). At 12 hr, the acetone level was elevated 6- and 9-fold over pretreatment levels for the low and high dose treatments, respectively (Table 1). The area under the curve for the increased concentration of blood acetone (AUC_{0-60}) of the high dose groups was about 2 times that of the low dose group. Turnover time (t_t), the time required to renew the amount in the pool of elevated acetone, was longer in the high dose group (19.2 ± 0.6 hr) than in the low dose group (15.8 ± 0.5 hr). The fractional turnover rate (k_t) in the high dose group was estimated to be 0.046 ± 0.005 hr^{-1} which was significantly lower than the 0.054 ± 0.001 hr^{-1} observed in the low dose group. Consistent with the estimations of the kinetic parameters of AUC_{0-60}, t_t and k_t, the recovery time for returning to the normal level of acetone in the high dose group (60 hr) was 12 hr longer than that in the low dose group (48 hr) (Table 1).

Elevation of acetone level by chronic treatments with DAS. DAS was given daily to rats at low doses (50 mg/kg daily, i.g.) or high doses (200 mg/kg daily, i.g.) for 29 days. The blood acetone level was measured at 24 hr following the 1st, 7th, 14th and 28th doses. In comparison to the vehicle control group, the low and high dose groups had significantly increased blood acetone levels: about 2.8- and 4.1-fold, respectively (Fig. 3). No apparent cumulative effect due to the multiple treatments of DAS was observed. The blood acetone levels were 2.4 μg/mL

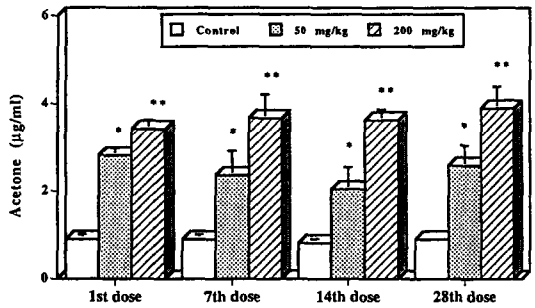

Fig. 3. Effects of chronic DAS treatments on blood acetone levels. DAS was given to rats intragastrically at a daily dose of 50 or 200 mg/kg for 29 days. Rats in the control group were given corn oil alone. Blood was taken at 24 hr after the 1st, 7th, 14th and 28th doses. Each bar represents the mean ± SD of 5 or 6 individual animals. Key: (*) significantly different from control ($P < 0.05$), and (**) significantly different from control and 50 mg/kg ($P < 0.05$).

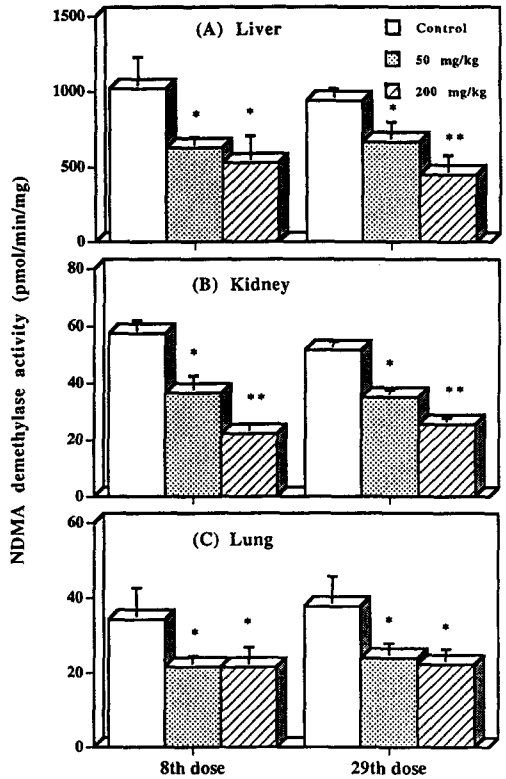

Fig. 4. Effects of chronic DAS treatments on microsomal NDMA demethylase activities in the liver (A), kidney (B) and lung (C). DAS was given to rats intragastrically at a daily dose of 50 or 200 mg/kg for 8 or 29 days, and control rats received the vehicle only. Animals were killed at 24 hr after the 8th or 29th dose of DAS. NDMA demethylase activity in liver microsomes was assayed with 0.3 mg microsomal protein and 0.1 mM NDMA in 0.5 mL with an incubation period of 20 min. Each bar represents the mean ± SD of 5 or 6 liver samples. With kidney and lung microsomes, 0.5 mg microsomal protein and a 30 min incubation were used. Kidney and lung microsomes were pooled samples, and each bar represents the mean ± SD of three determinations. The final concentration of NDMA was 0.1 mM. Key: (*) significantly different from control ($P < 0.05$), and (**) significantly different from control and 50 mg/kg ($P < 0.05$).

(range: 2.0 to 2.8 μg/mL) for the low dose and 3.6 μg/mL (range: 3.4 to 3.9 μg/mL) for the high dose in comparison to 0.9 μg/mL (range: 0.8 to 0.9 μg/mL) for the control.

Effects on NDMA demethylase activity and P450 2E1 content. To study the relationship between the levels of blood acetone and P450 2E1, P450 2E1-mediated NDMA demethylase activity and P450 2E1 content were studied. As shown in Fig. 2, the increase of acetone level corresponded with the decrease of NDMA demethylase activity after a single dose of DAS. For the chronic experiments, significant decreases in microsomal NDMA demethylase activity were observed in the liver, kidney, and lung after the 8th and 29th doses of DAS (Fig. 4). P450 2E1 content measured by immunoblotting was also decreased after the 8th and 29th doses with DAS (Fig. 5). Figure 6 presents the results of quantitation of immunoblot bands by a densitometer. Close correlations between acetone level (*in vivo*) and P450 2E1 level (*in vitro*) were found either among 1-week treated rats ($r = 0.906$, $N = 12$, $P < 0.001$) or among 4-week treated rats ($r = 0.886$, $N = 12$, $P < 0.001$). The results are consistent with the hypothesis that the elevation of blood acetone level *in vivo* after DAS treatment is caused by the decrease of P450 2E1 activity.

Effect of chronic treatment of DAS on the plasma concentrations of DASO and $DASO_2$. Our previous results indicate that DAS inhibits the activity of P450 2E1 by a competitive inhibition mechanism and by suicide inactivation of P450 2E1 via its metabolite $DASO_2$ [11]. In a preliminary study on the fate of DAS in rats, DAS was not detected, but DASO and $DASO_2$ were detected in the plasma 10 min after intragastric administration of DAS (200 mg/kg). In chronic treatments, the concentrations of DASO and $DASO_2$ were determined by HPLC at 24 hr after the 1st, 7th, 21st and 28th doses. In the high dose group, the concentrations of DASO and $DASO_2$ were 45.0 μg/mL (range: 33.8 to 58.6 μg/mL) and 11.7 μg/mL (range: 6.9 to 15.6 μg/mL), respectively, reflecting a lack of accumulation of the metabolites during the long-term treatment with DAS (Fig. 7). In the low DAS group of the chronic study, the metabolites were below the level of detection of the method employed.

Effect of phenobarbital treatment on blood acetone level. Treatment of rats with DAS is known also to induce P450 2B enzymes by 7-fold [10]. To examine whether P450 2B induction would affect blood acetone level, PB was used to induce P450 2B enzymes. Prior to treatment with PB and at 24 hr after the 1st, 2nd and 3rd doses of PB, the blood acetone levels were 1.13 ± 0.19, 1.17 ± 0.20, 1.40 ± 0.69 and 1.43 ± 0.75 μg/mL, respectively. The results showed that induction of P450 2B did not cause a decrease in blood acetone level,

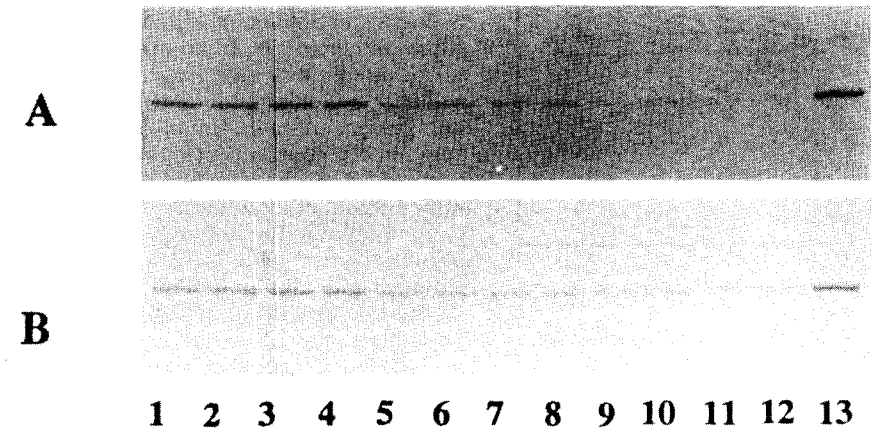

Fig. 5. Western blot of rat liver microsomal samples. Microsomes from the same experiment shown in Fig. 4A were subjected to SDS–PAGE followed by immunoblot analysis with anti-P450 2E1 IgG. (A) group of 8 doses; (B) group of 29 doses. Lanes 1–4, control liver; lanes 5–8, 50 mg/kg DAS-treated liver; lanes 9–12, 200 mg/kg DAS-treated liver; lane 13, acetone-induced liver. Lanes contained 3 μg of microsomal protein.

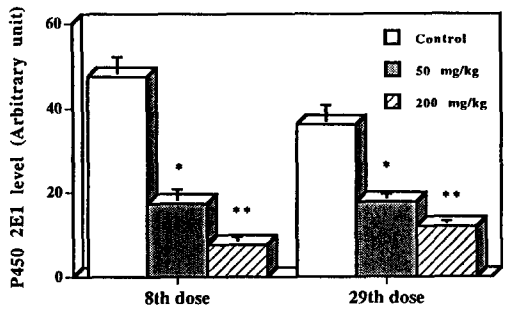

Fig. 6. Effects of chronic DAS treatments on the content of P450 2E1 in rat liver microsomes. Quantitation of immunoblot bands of Fig. 5 was conducted with a densitometer. Each bar represents the mean ± SD of 4 separate liver samples. Key: (*) significantly different from control ($P < 0.05$), and (**) significantly different from control and 50 mg/kg ($P < 0.05$).

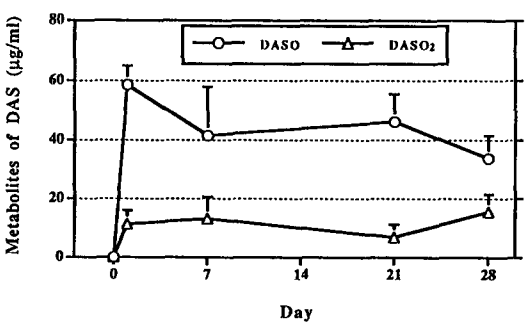

Fig. 7. Effects of chronic DAS treatments on the plasma concentrations of DASO and $DASO_2$. DAS was given to rats intragastrically at a daily dose of 200 mg/kg for 29 days. Blood samples were taken at 24 hr after the 1st, 7th, 21st and 28th doses. DASO and $DASO_2$ were analyzed by HPLC. Each data point is the mean ± SD of 5 or 6 individual animals.

suggesting that P450 2B is not involved in acetone metabolism. We previously observed that PB treatment decreased rat liver P450 2E1-mediated NDMA demethylase activity by 40% [19]. Such an effect may be related to the presently observed slight increase of blood acetone level (by 27%) after the 3rd dose of PB. Because of the large individual variations, however, the increase was not statistically significant.

Effects of DAS and $DASO_2$ on activity of aldehyde dehydrogenase. Aldehyde dehydrogenase is an enzyme responsible for the conversion of 1,2-propenediol to lactaldehyde in the acetone gluconeogenic pathway. To determine if DAS and its metabolite $DASO_2$ inhibit this enzyme, they were added to incubations with rat liver microsomes or cytosol (Fig. 8). No inhibitory effect on aldehyde dehydrogenase activity was observed. However, DSF, a compound also known to cause acetonaemia [14, 21, 22] strongly inhibited the cytosolic enzyme activity with an IC_{50} of 25 μM (Fig. 8).

DISCUSSION

The acetone level in an animal is affected by two factors. One is by its production under ketogenic conditions through the nonenzymatic decarboxylation of acetoacetate [23]. The other is the elimination of acetone by enzymatic oxidation and exhalation. The present results indicate that the elevation of the blood acetone level by DAS treatment is due mainly to the inhibition of acetone elimination. The time required to reach maximum concentration of acetone was shorter in rats treated with the low dose than with the high dose of DAS (Fig. 2). Similarly, the recovery time to return to normal blood acetone level was prolonged in the high dose group (Table 1). The AUC_{0-60} in the high dose group was significantly higher than in the low

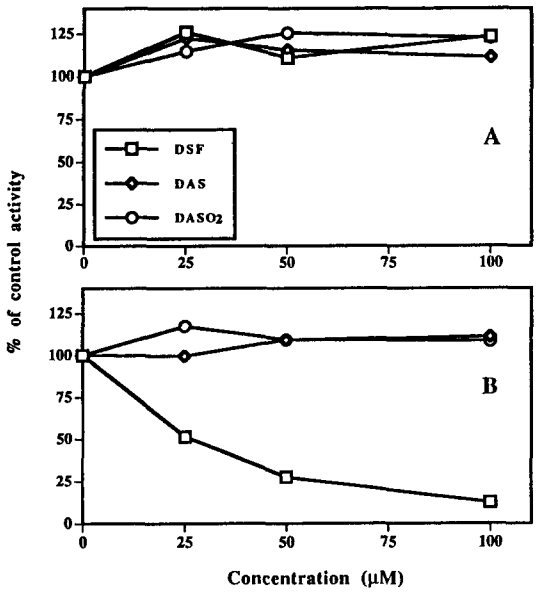

Fig. 8. Effects of DAS, $DASO_2$ and DSF on aldehyde dehydrogenase activity in rat liver microsomes (A) and cytosol (B). The activity of the enzyme was measured according to the procedure of Tottmar *et al.* [20]. The final concentration of substrate (propionaldehyde) was 5 mM. The control activity (100%) was 8.17 and 6.05 nmol/min/mg, respectively, for the microsomes and cytosol. Each data point is the mean of two determinations.

dose group, indicating a larger acetone pool in rats in the high dose group. Furthermore, a lower fractional turnover rate and longer turnover time were found in the high dose group, also suggesting that an inhibition of acetone elimination is responsible for the elevation [15]. Exhalation can be an important route of acetone elimination, especially in fasting and diabetic conditions [4, 5]. The relative importance of this route of elimination depends on the plasma acetone level. For example, it accounts for about 70% of the elimination when the plasma acetone level is at 8 mM and 20% when the plasma acetone level is at 2 mM [5]. The blood concentrations of the rats in our study was in the range of 0.017 to 0.130 mM, and the contribution of the exhalation in total elimination is expected to be much lower than 20%. In addition, the rats in the DAS treatment group behaved normally. Thus, a decrease of acetone exhalation due to DAS treatment is not believed to have taken place. Therefore, the elevation of the blood level of acetone in DAS-treated rats is attributed to the decrease of acetone catabolism.

In both humans and rats, administration of DSF was reported to cause a significant elevation of blood acetone level [14, 21, 22]. Differently from the situation in fasting, in which all of the ketone bodies are elevated [4, 24], DSF increased the blood level of acetone but not of acetoacetate or β-hydroxybutyrate. Since DSF is known to be a strong inhibitor of P450 2E1 [2, 25], we believe that the increase of acetone level following treatment with DSF, similar to the situation with DAS treatment, is due to the inhibition of the P450 2E1-mediated oxidative elimination of acetone.

Consistent with the suggestion that acetone oxidation is catalyzed by P450 2E1 *in vitro* [7, 9], our present results demonstrate the correlation between the elevation of blood acetone level and decreased P450 2E1 activity in rats. In the experiment with a single high dose of DAS, the increase of acetone level was correlated with the decrease of NDMA demethylase activity (Fig. 2), which is predominantly due to P450 2E1 [26–28]. Similarly, the increase of acetone level observed in the chronic experiment (Fig. 3) was accompanied by a decrease of NDMA demethylase activities (Fig. 4) and diminution of P450 2E1 content in liver microsomes (Fig. 5). These results suggest that the increase of acetone level *in vivo* is due to the inactivation and inhibition of P450 2E1 by DAS. The elevated levels of acetone, 4–9 times higher than normal, is comparable to those attained in fasting [24]. Such levels of acetone are believed to be only one of the factors contributing to the induction of P450 2E1 during fasting [24].

Our previous results indicate that DAS, DASO and $DASO_2$ are competitive inhibitors and that $DASO_2$ is a suicide inhibitor of P450 2E1 [11]. The lack of accumulation of DASO and $DASO_2$ in plasma during chronic treatment with the high dose of DAS (Fig. 7) suggests that DASO and $DASO_2$ are rather effectively metabolized and eliminated. During the chronic treatment, the P450 2E1 level was not progressively decreased, suggesting that the rate of suicide inactivation of this enzyme is at about the same rate as its biosynthesis. This result is consistent with the observation that the blood acetone level did not progressively increase as the DAS treatment prolonged. This is different from the case where animals were treated by repeat doses of DSF, which causes a continued increase of blood acetone level [21, 22], suggesting an accumulation of DSF metabolites. In addition, DSF may affect acetone metabolism at more than one step, because it also inhibits aldehyde dehydrogenase [29, 30], which catalyzes the metabolism of lactaldehyde converted from 1,2-propenediol, a metabolite of acetone. On the other hand, no inhibition of aldehyde dehydrogenase by DAS or its metabolite $DASO_2$ was found in our experiments (Fig. 8).

In conclusion, the present studies demonstrate that the inhibition of P450 2E1 results in the elevation of the blood acetone level in rats. The results strongly suggest that P450 2E1 is an enzyme responsible for acetone catabolism under physiological conditions.

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