K. Uchida, T. Matsubara

Effect of Flomoxef on Blood Coagulation and Alcohol Metabolism

Summary: The effect of flomoxef, a newly developed oxacephem antibiotic with an N-hydroxyethyltetrazolethiol (HTT) side chain, on blood coagulation and alcohol metabolism was compared with that of a series of cephalosporin antibiotics with N-methyltetrazolethiol (NMTT), thiadiazolethiol (TDT) or methylthiadiazolethiol (MTDT) side chains in position 3' of the cephalosporin nucleus known to cause hypoprothrombinemia and bleeding in patients who are malnourished, debilitated and/or of high age. A disulfiram-like effect caused by inhibition of aldehyde dehydrogenase was observed for NMTT-containing antibiotics. Studies were carried out on healthy volunteers and on rats. Eight-day treatment with 2 g flomoxef i. v. once or twice daily in five and six healthy male volunteers, respectively, did not cause any significant changes in prothrombin time (PT), coagulation factors II, VII, IX or X, in hepaplastin values or fibrinogen levels, activated partial thromboplastin time (APTT), platelet counts, bleeding time, or collagen- and ADP-induced platelet aggregation. Inhibition of vitamin K epoxide reductase was observed in rats treated with flomoxef, yet to a much lesser extent than observed for cephalosporins with NMTT, TDT or MTDT side chains. This defect was quickly normalized by vitamin K injection. There were no differences between oxacephem (1-O) and cephem (1-S) compounds with respect to effects on blood clotting and platelet aggregation. Flomoxef and its side chain HTT showed no influence on alcohol metbolism.

Zusammenfassung: *Wirkung von Flomoxef auf Blutgerinnung und Alkoholmetabolismus.* Die Wirkung von Flomoxef, einem neuen Oxacephem-Antibiotikum mit N-hydroxyethyl-tetrazol-thiol (HTT)-Seitenkette auf Blutgerinnung und Alkoholmetabolismus wurde mit den Wirkungen einer Reihe von Cephalosporinen verglichen, die N-methyl-tetrazol-thiol (NMTT)-, Thia-diazol-thiol (TDT)- oder Methyl-thiazol-thiol (MTDT)-Seitenketten in Position 3' des Cephalosporin-Kernes haben und bekanntlich bei Mangelernährung, schlechtem Allgemeinzustand und/oder hohem Alter Hypoprothrombinämie und Blutungen hervorrufen können. Antibiotika mit NMTT-Seitenkette können außerdem eine Disulfiram-Reaktion auslösen, indem sie die Aldehyddehydrogenase hemmen. Die Untersuchungen wurden bei gesunden freiwilligen Probanden und bei Ratten durchgeführt. Fünf beziehungsweise sechs männliche Probanden erhielten Flomoxef in einer Dosierung von einmal oder zweimal täglich 2 g intravenös appliziert. Unter der achttägigen Applikation ließen sich keinerlei signifikante Veränderungen der Prothrombinzeit (PT), der Gerinnungsfaktoren II, VII, IX oder X, der Hepaplastin-Werte, der Fibrinogen-Spiegel, der aktivierten partiellen Thromboplastinzeit (APTT), der Thrombozytenzahlen, der Blutungszeit oder der Kollagen- oder ADP-induzierten Plättchenaggregation erkennen. Bei Ratten wurde unter Behandlung mit Flomoxef eine Hemmung der Vitamin K-Epoxid-Reduktase festgestellt, die jedoch erheblich geringer ausgeprägt war als mit Cephalosporinen mit NMTT-, TDT- oder MTDT-Seitenketten. Die Störung wurde rasch durch Vitamin K-Injektionen ausgeglichen. Oxacephem (1-O)- und Cephem (1-S)-Verbindungen unterscheiden sich nicht im Einfluß auf die Blutgerinnung und Plättchenaggregation. Ein Einfluß von Flomoxef oder seiner Seitenkette HTT auf den Alkoholmetabolismus war nicht festzustellen.

Introduction

Cephalosporin antibiotics possessing an N-methyltetrazolethiol (NMTT), thiadiazolethiol (TDT) or methylthiadiazolethiol (MTDT) side chain as the 3'- position substituent of the cephalosporin nucleus are known to cause hypoprothrombinemia and bleeding in some malnutritional, debilitated or aged patients [1–3] and in patients with renal dysfunction [4–6]. In addition, NMTT-containing antibiotics show a disulfiram-like effect by inhibiting aldehyde dehydrogenase [7–10].

A newly developed oxacephem antibiotic, flomoxef, possesses an N- hydroxyethyltetrazolethiol (HTT) side chain which is different from NMTT but somewhat resembles it in the structure. Therefore, we examined the effects of flomoxef and HTT on blood coagulation activity as well as alcohol metabolism and compared them with those of NMTT-containing antibiotics.

The Effect of Flomoxef on Healthy Volunteers

2 or 4 g (bid) of flomoxef were injected intravenously (i. v.) into five and six healthy male volunteers daily for eight days, and blood clotting activities and plasma coagulation factor levels were examined before, during and after the treatment (Table 1). No significant change was found in prothrombin time (PT), Factors II, VII, IX and X, the hepaplastin value and fibrinogen levels. Figure 1 shows the

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Table 1: Effect of flomoxef on blood coagulation factors in healthy male volunteers.

		2 g/day (bid)			4 g/day (bid)		
		Before	During[a)]	After[b)]	Before	During[a)]	After[b)]
PT	(sec)	12.7[c)]	11.9	11.5	12.1	12.4	12.3
Factor II	(%)	96.4	94.8	84.4	85.7	89.0	98.8
Factor VII	(%)	81.6	92.2	102.6	109.2	100.8	108.5
Factor IX	(%)	110.2	112.4	115.6	111.7	100.8	128.0
Factor X	(%)	78.8	82.0	97.4	112.5	116.7	118.2
Hepaplastin	(%)	107.4	105.2	105.2	125.7	106.5	116.2
Factor I	(mg/dl)	190.0	216.4	216.9	262.2	257.0	246.7

a) On the fifth day of the treatment;
b) On the first day after treatment for eight days was stopped;
c) Mean values in 5–6 volunteers;
PT: Prothrombin time.

Table 2: Effect of β-lactam antibiotics on prothrombin time in male rats fed a vitamin K deficient diet.

	Dose (mg/kg/day)	Prothrombin time (sec)			
		Exp. 1	Exp. 2	Exp.3	Exp. 4
Control		16.0 ± 0.4[a)]	17.5 ± 0.4	22.9 ± 1.3	21.2 ± 2.4
Latamoxef	300	25.3 ± 0.9*	28.4 ± 3.6*	–	48.8 ± 7.9*
Ceftriaxone	300	16.0 ± 0.3	–	–	–
Cefotaxime	300	15.9 ± 0.2	18.8 ± 0.8	–	18.1 ± 1.4
Cefamandole	300	–	29.6 ± 3.0*	–	37.5 ± 3.2
Cefoperazone	300	–	33.8 ± 3.7*	–	–
Carbenicillin	300	–	19.0 ± 1.1	–	16.6 ± 1.4
Cefmetazole	300	–	–	> 53.0	–
Cefotetan	300	–	–	34.5 ± 3.1*	–
Cefotiam	300	–	–	20.4 ± 1.3	–
Cefoxitin	300	–	–	25.8 ± 1.4	–
Ceftezole	300	55.4 ± 15.5*	–	–	–
Cefazolin	300	–	48.5 ± 9.7*	–	–
Flomoxef	300	21.5 ± 1.8*	–	–	–

a) Mean ± S.E. in 5–6 rats;
b) The antibiotics were injected once a day (i.v.) for 8 days;
* Statistically significant ($P < 0.05$).

changes in PT, activated partial thromboplastin time (APTT), platelet counts and bleeding time, but no remarkable change was found. Figure 2 shows the changes in collagen- and ADP-induced platelet aggregation, but no inhibition was found.
These results suggest that flomoxef, 2 g or 4 g daily for eight days, causes no effect on either blood coagulation activities or platelet aggregation in healthy volunteers.

Effects of Cephalosporin Antibiotics and Flomoxef on Blood Coagulation in Rats

The effects of various cephalosporin antibiotics on PT were examined in male rats. When rats were kept on an ordinary diet which contained sufficient amounts of vitamin K_1 (ca. 500 ng/g diet), none of the cephalosporins produced hypoprothrombinemia. However, when the rats were kept on a vitamin K deficient diet (ca. 30 ng/g diet), some of the

Table 3: Effects of latamoxef and flomoxef on prothrombin time (PT), activated partial thromboplastin time (APTT), plasma prothrombin and PIVKA levels in male rats kept on a vitamin K deficient diet.

	PT (sec)	APTT (sec)	Prothrombin (U/ml)	PIVKA (U/ml)
Control	15.2 ± 1.3[b)]	25.2 ± 1.8	66.1 ± 11.8	4.6 ± 0.5
Latamoxef[a)]	24.5 ± 2.8*	38.4 ± 2.6*	26.6 ± 3.6*	6.9 ± 0.5*
Flomoxef	14.8 ± 0.8	24.5 ± 1.4	62.4 ± 9.8	4.9 ± 0.2

a)The antibiotics (300 mg/kg/day) were injected once a day (i.v.) for 3 days;
b)Mean ± S. E. of 4 rats;

*Statistically significant ($P < 0.05$);
PIVKA: Proteins induced by vitamin K absence.

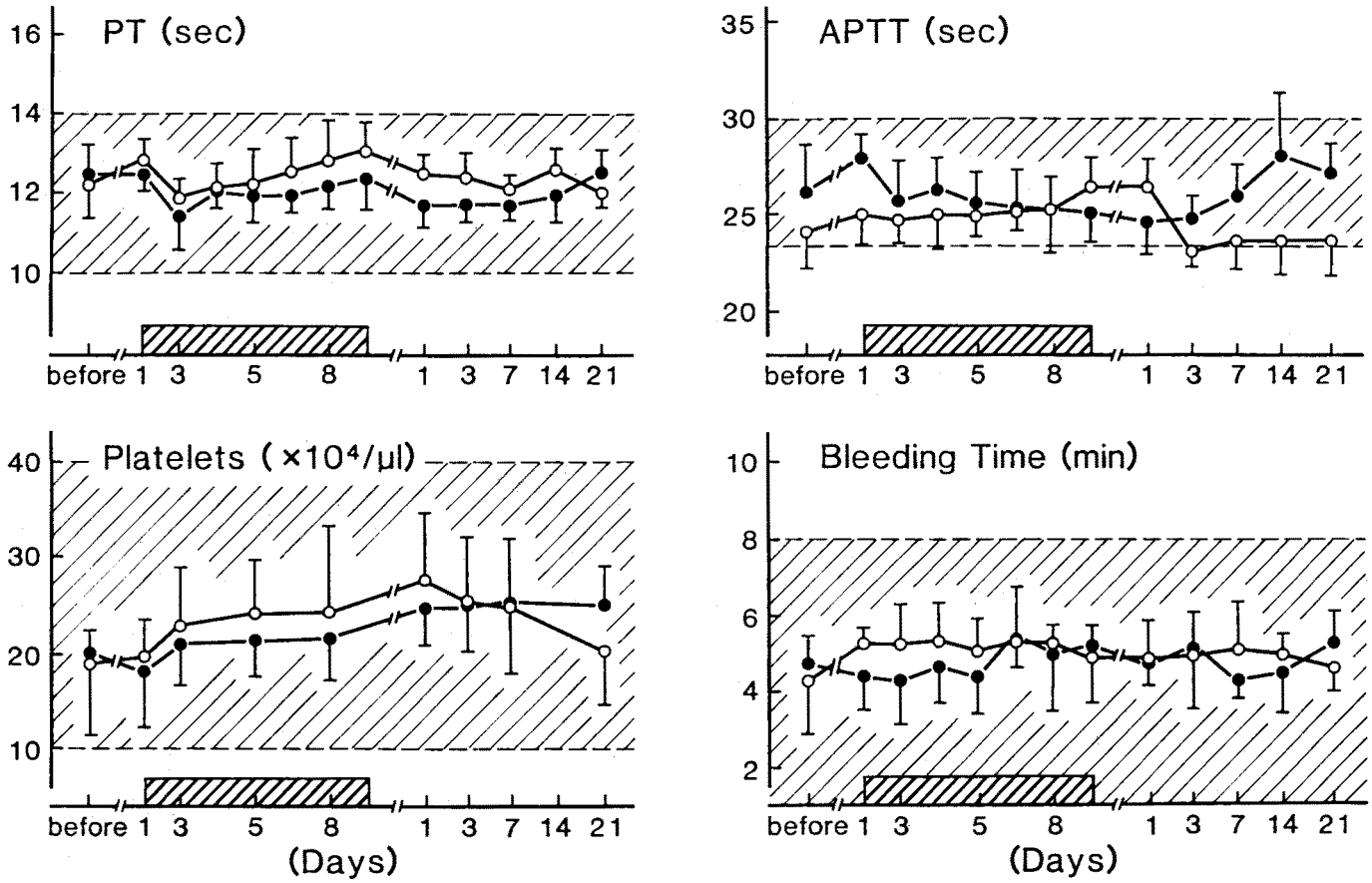


Figure 1: Effect of flomoxef on blood clotting parameters and platelet number. Healthy male volunteers were injected (i. v. d.) flomoxef for 8 days as shown in the figure by bars. Blood samples were obtained before, during and after the treatment with flomoxef, and their prothrombin time (PT), activated partial thromboplastin time (APTT), platelet number and bleeding time were determined. Closed and open circles represent daily 2 g (b. i. d.; no. = 5) and 4 g (b. i. d.; no. = 6) injections, respectively.

cephalosporins which contained an NMTT, TDT or MTDT side chain increased PT (Table 2). Flomoxef, although the side chain is different from NMTT, also increased PT in vitamin K deficient rats when the animals were treated with high doses of flomoxef for a long period. However, the treatment of vitamin K deficient rats with flomoxef for three days caused a slight or no effect on blood clotting parameters, even though latamoxef clearly showed hypoprothrombinemic action under the same conditions (Table 3).

Subsequently, we attempted to examine the mechanism for the hypoprothrombinemia. Since γ-glutamylcarboxylase is a key enzyme in the synthesis of vitamin K dependent clotting factors, we examined the effect of flomoxef on this enzyme activity compared with that of latamoxef [11]. As shown in Table 4, flomoxef caused no inhibition on γ-glutamylcarboxylase either *in vivo* or *in vitro*. Latamoxef slightly decreased the activity *in vitro*, but such a high concentration as 10 mM is not achievable *in vivo*. Of course, latamoxef showed no inhibition at lower concentrations such as 3 mM or less.

Figure 3 shows the effects of flomoxef and HTT on γ-glutamylcarboxylase activity *in vitro*. The enzyme activity in microsomes obtained from the ordinary and vitamin K deficient diet-fed rats was not modified by flomoxef and HTT, although a slight inhibition was found in normal rat liver microsomes in the presence of the highest concentration (10 mM) of the antibiotic or its side chain. These results suggest that flomoxef and its side chain, HTT, practically cause no inhibition of this enzyme.

Vitamin K reductase also plays an important role in the synethesis of vitamin K dependent clotting factors since vitamin K is usually supplied from diets as a biologically inactive quinone form which is reduced to an active hydroquinone form by vitamin K reductase. This enzyme was inhibited by disulfiram but not by the side chain of latamoxef, NMTT (Figure 4). The effects of various cephalosporins and their side chains on vitamin K reductase, including flomoxef and its side chain, are given in Figure 5 but no inhibition was found.

Vitamin K epoxide reductase, however, was inhibited by both NMTT and NMTT- containing antibiotics or flomoxef and its side chain as shown in Figure 6. The enzyme activity was determined by the method reported previously [12,

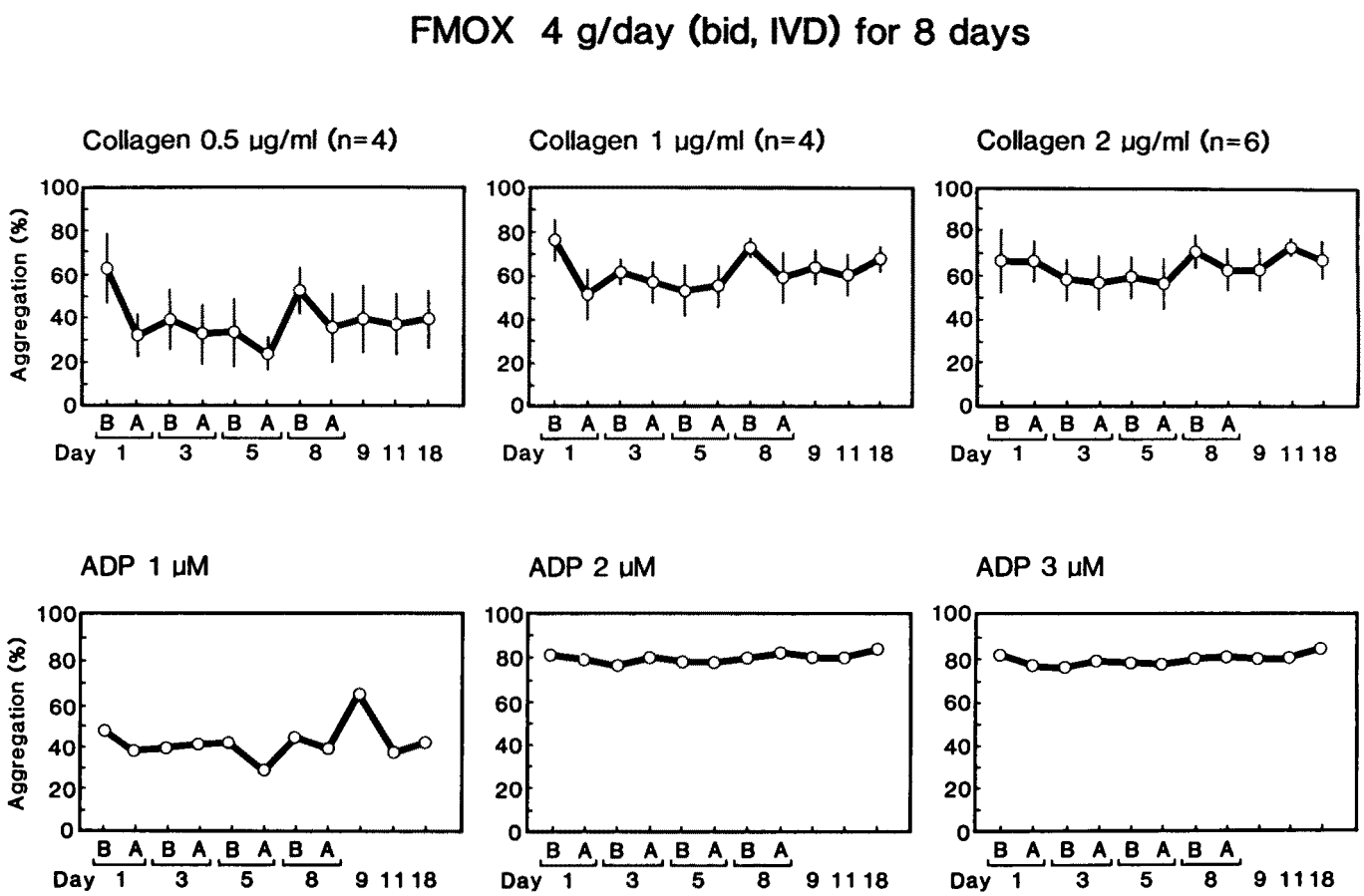


Figure 2: Effect of flomoxef on platelet functions. Healthy male volunteers were injected daily 4 g (b. i. d.) of flomoxef for 8 days (day 1 to day 8). Time required for flomoxef infusion was 1 hour, and blood samples during the administration period were obtained just before (B) or after (A) the infusion. Samples were also obtained 1 (day 9), 3 (day 11) and 10 (day 18) days after the last administration of flomoxef. Platelet aggregation was determined by adding various concentrations of collagen or ADP as the inducer.

13]. Then, we compared the effects of flomoxef and latamoxef on vitamin K epoxide reductase, where various doses of both compounds were injected intravenously for four days and the liver microsomes were prepared for the enzyme assay. As shown in Figure 7, both the antibiotics dose-dependently inhibited the enzyme activity, but the potency of flomoxef was about 1/3–1/10 that of latamoxef.

The results are summarized in Figure 8. Vitamin K is usually supplied from diets as a quinone form which is ineffective as a cofactor for γ-glutamylcarboxylase. This quinone vitamin K is reduced by vitamin K reductase to a hydroquinone form, the active form. In coupling with the reaction of γ-glutamylcarboxylation, which converts the glutamic acid (Glu) residue to γ-carboxyglutamic acid (Gla) residue, hydroquinone vitamin K is converted to vitamin K epoxide. This vitamin K epoxide is reduced to the original quinone vitamin K by vitamin K epoxide reductase and used again.

This cycle is called the "Vitamin K Cycle". When the supply of dietary vitamin K is reduced, vitamin K required for γ-glutamylcarboxylation is filled up by this vitamin K cycle. As shown in Figures 6 and 7, the NMTT side chain inhibits vitamin K epoxide reductase, in other words interrupts the vitamin K cycle. Therefore, NMTT-containing cephalosporins enhance vitamin K deficiency in rats fed the vitamin K deficient diet resulting in an increase of PT. This vitamin K deficiency is normalized by the administration of vitamin K, as shown in Figure 9, which demonstrates that the hypoprothrombinemia is quickly normalized after a vitamin K injection even though latamoxef treatment is continued because neither vitamin K reductase nor γ-glutamylcarboxylase is inhibited [11, 14]. This will be the mechanism for the hypoprothrombinemia produced by NMTT-containing antibiotics [15], and also the mechanism for flomoxef.

Recently, we could demonstrate that the side chains liberated from the cephalosporin antibiotics were responsible for the previously mentioned inhibitory effect, and the intact molecules of the antibiotics containing the side chains would not exert the inhibitory effect.

As a result of the inhibition of vitamin K epoxide reductase, plasma vitamin K epoxide levels increase. This evidence

Table 4: Effects of latamoxef and flomoxef on γ-glutamylcarboxylation.

	Control	Flomoxef	Latamoxef
	(x10³ dpm/mg protein/30 min)		
(1) *In vitro* (10 mM)			
Reduced vitamin K	15.12 ± 0.53	14.58 ± 0.08	12.34 ± 0.03*
Oxidized vitamin K	18.78 ± 0.40	18.20 ± 0.54	14.41 ± 0.64*
(2) *In vivo* (300 mg/day/rat for 8 days)			
Reduced vitamin K	11.41 ± 1.17	12.92 ± 0.79	12.11 ± 0.71
Oxidized vitamin K	15.12 ± 1.56	14.11 ± 1.31	13.99 ± 0.41

* Statistically significant ($P < 0.05$).

In the *in vitro* experiments, liver microsomes were prepared from intact rats and their carboxylation activity was measured as described previously (11) using reduced vitamin K or oxidized vitamin K (plus NADH) as a cofactor. Effects of flomoxef and latamoxef on the enzyme activity were determined by adding the antibiotics (10 mM) to the reaction system. *In vivo* effects of antibiotics were examined as follows: animals fed on a vitamin K deficient diet were given an i. v. injection of the antibiotics at 300 mg/kg once daily for 8 days, and their enzyme activities were determined as above.

Table 5: Effect of flomoxef on blood acetaldehyde and ethanol concentrations in male volunteers.

	Flomoxef treatment		
	Before	After	P
No. of volunteers	8	8	
Acetaldehyde (μg/ml)	0.17 ± 0.025	0.27 ± 0.129	NS
Ethanol (mg/ml)	0.36 ± 0.067	0.30 ± 0.078	NS

Flomoxef was injected (i. v. d.) once a day for 3 days (day 1, 0.5 g; day 2, 1.0 g; and day 3, 2.0 g). Each volunteer took liquor containing ca. 50 ml eq. of ethanol within a period of 20 minutes, 2 hours after the infusion of the antibiotic, and the blood acetaldehyde and ethanol levels were determined by a head-space gas-chromatographic method 40 minutes later.

has already been reported in patients treated with latamoxef by *Bechtold* et al. [16] and a similar result is obtained in rats. As shown in Figure 10, plasma vitamin K epoxide levels were positively correlated with the vitamin K levels in normal control rats, and the slope of the correlation line increased in the latamoxef treated rats. It should be emphasized that the plasma vitamin K epoxide levels (or the ratio of it to vitamin K) increase after treatment with these antibiotics, but it does not always indicate the deficiency of vitamin K. When sufficient amounts of vitamin K are present in the liver or plasma, synthesis of vitamin K dependent clotting factors normally proceeds even if plasma vitamin K epoxide levels are increased [14, 15].

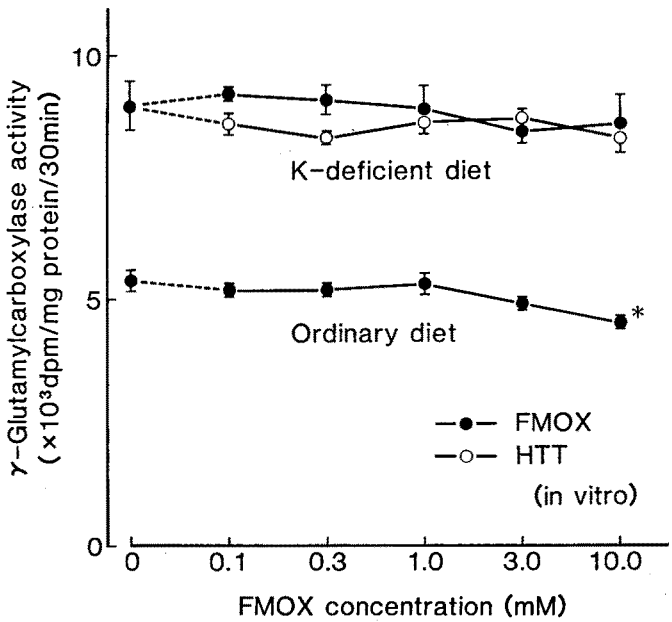


Figure 3: *In vitro* effect of flomoxef on γ-glutamylcarboxylase activity in liver microsomes. Liver microsomes were prepared from rats fed an ordinary diet or a vitamin K deficient diet, and the carboxylation activity was determined using a reduced vitamin K as a cofactor. Effects of flomoxef and its side chain, HTT, were examined by the addition of various concentrations of the compounds as shown in the figure.

Comparison of 1-Oxa and 1-Thia Cephalosporins in Regard to Their Effects on Blood Clotting Activities

Some people claim that the effect of oxygen replacement at the 1-position is not clear in regard to the safety of oxacephem antibiotics. Therefore, we synthesized 1-S congeners of latamoxef and flomoxef (Figure 11), and compared their effects on blood clotting activities and plasma clotting factor levels. The urinary excretion of flomoxef and the liberated side chain, HTT, was slightly higher than that of the 1-S congener (Figure 12). This corresponds

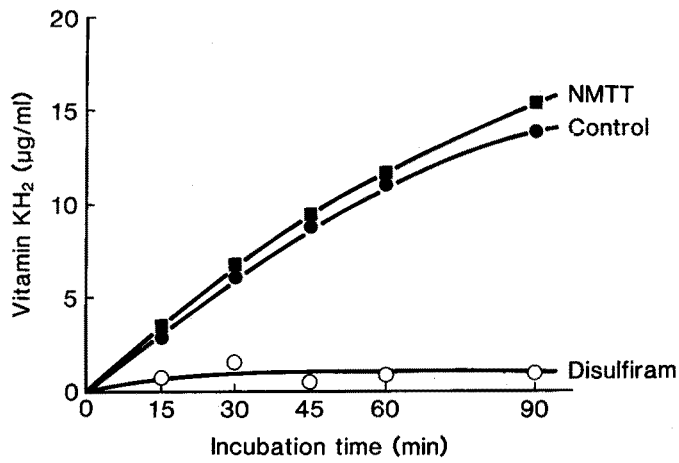


Figure 4: Time course changes in the formation of reduced vitamin K from the oxidized form by liver microsomal vitamin K reductase. Vitamin K reductase activity in rat liver microsomes was measured by detecting reduced vitamin K produced during the *in vitro* reaction [12]. Effects of NMTT and disulfiram *in vitro* were determined by adding 2 mM and 0.3 mM of the compounds, respectively.

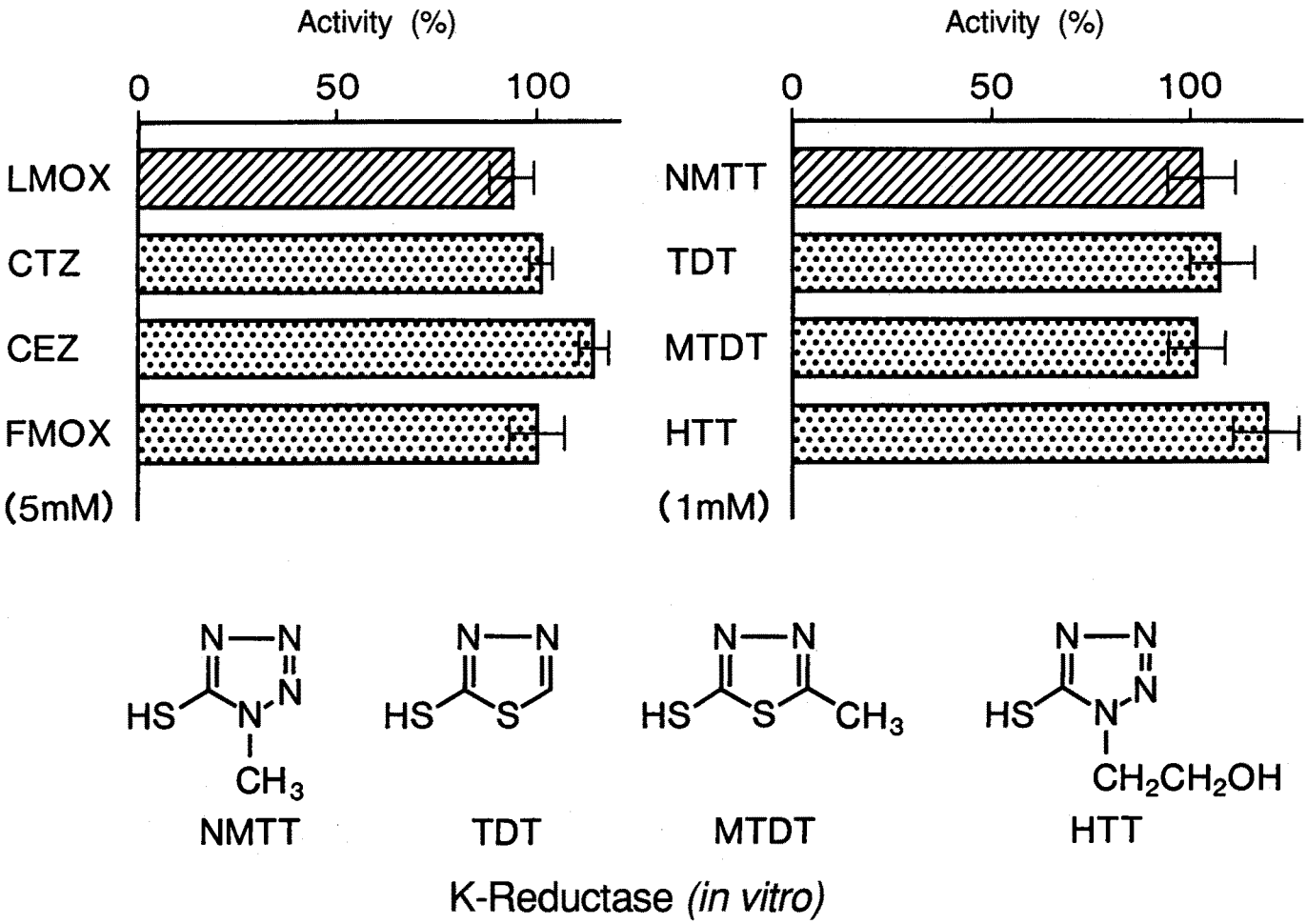


Figure 5: Effects of β-lactam antibiotics and their corresponding 3'-position side chain (heterocyclic thiol compounds) on liver microsomal vitamin K reductase activity. The vitamin K reductase activity was measured as described in Figure 4. Relative enzyme activities of microsomes in the presence of 5 mM antibiotics or 1 mM heterocyclic thiol compounds against the control are given in the figure. The values represent the mean ± S. E. of 5 determinations. LMOX: Latamoxef, CTZ: Ceftezole, CEZ: Cefazolin, FMOX: Flomoxef.

well to the fact that an oxacephem is more labile than the corresponding 1-S congener [17]. The effects of flomoxef and 1-S flomoxef on PT and plasma prothrombin levels are given in Figure 13. Actually, both the compounds dose-dependently increased PT in rats kept on a vitamin K deficient diet and decreased the plasma prothrombin levels, but no significant difference was found between the two compounds. Our previous data [18, 19], show that ADP- or collagen-induced platelet aggregation is inhibited by latamoxef and its 1-S congener at high concentrations *in vitro*, but no difference between the two compounds is found in the inhibitory effect.

As a conclusion, although the antibacterial activities of oxacephalosporins are much higher than the corresponding 1-S congeners [20, 21], their effects on plasma clotting factor levels or platelet aggregation are almost the same.

The Effect of Flomoxef on Alcohol Metabolism

The effect of flomoxef on alcohol metabolism in rats was examined in comparison with those of NMTT-containing antibiotics, latamoxef, cefmenoxime and disulfiram [22, 23]. As shown in Figure 14, latamoxef, cefmenoxime and disulfiram decreased the low Km aldehyde dehydrogenase (ALDH) activity and increased the blood acetaldehyde level, but flomoxef showed no effect. This data suggest that NMTT-containing cephalosporins show a disulfiram-like effect but flomoxef does not [24]. The low Km ALDH was inhibited by either disulfiram or NMTT, but the high Km ALDH was not (Figure 15).

Subsequently, we confirmed that neither alcohol incompatibility nor a disulfiram-like effect was observed in healthy male volunteers who had received flomoxef for three days (Table 5).

Conclusion

The following can be concluded: (I) The hypoprothrombinemia produced by NMTT, TDT or MTDT-containing cephalosporins is due to the inhibition of vitamin K epoxide

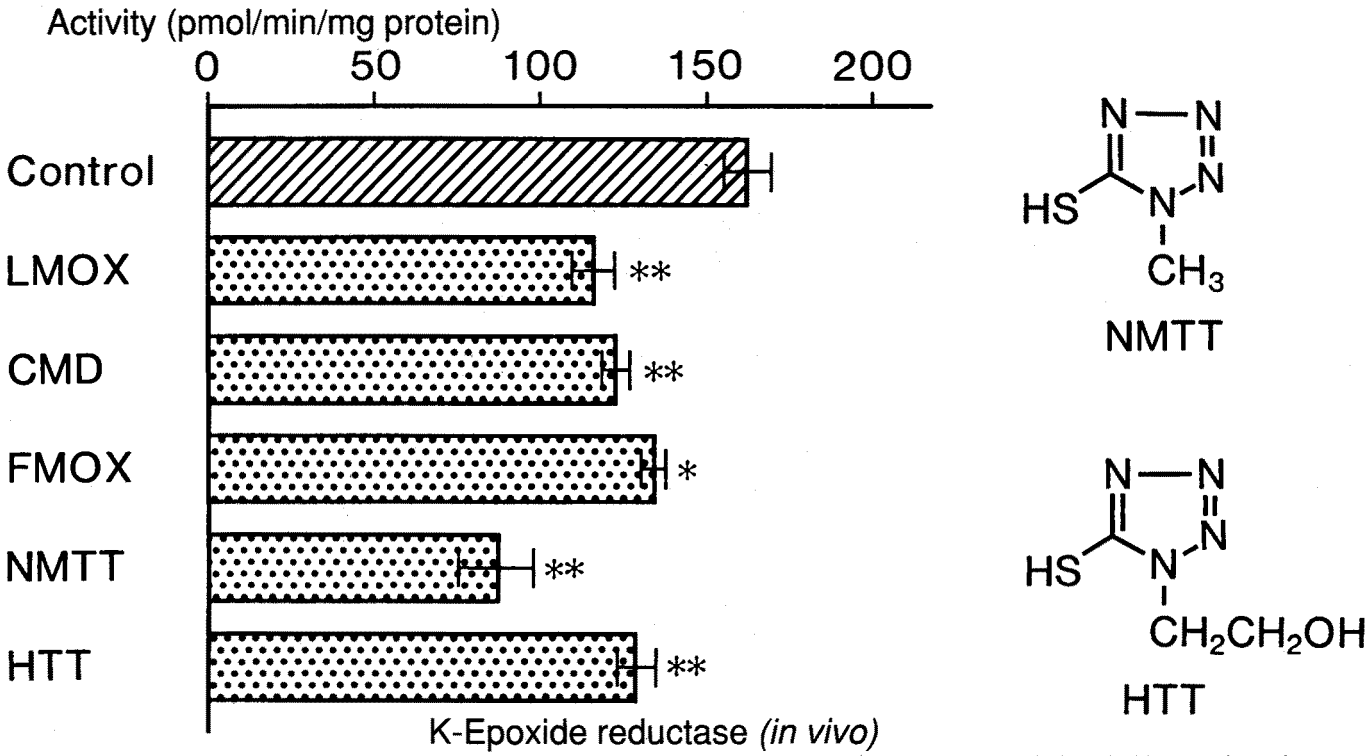


Figure 6: Effects of β-lactam antibiotics and heterocyclic thiol compounds on liver microsomal vitamin K epoxide reductase activity in rats. Animals were intravenously given the antibiotics (300 mg/kg) or heterocyclic thiol compounds (0.5 mmol/kg), once daily for 3 days. The vitamin K epoxide reductase activity was determined by detecting the conversion of vitamin K epoxide to the quinone form [13]. Specific enzyme activities in the control and the treated rats are given in the figure. The values in the figure represent the mean ± S. E. of 4 animals. **Statistically significant ($P < 0.01$) against the control. LMOX: Latamoxef, CMD: Cefamandole, FMOX: Flomoxef.

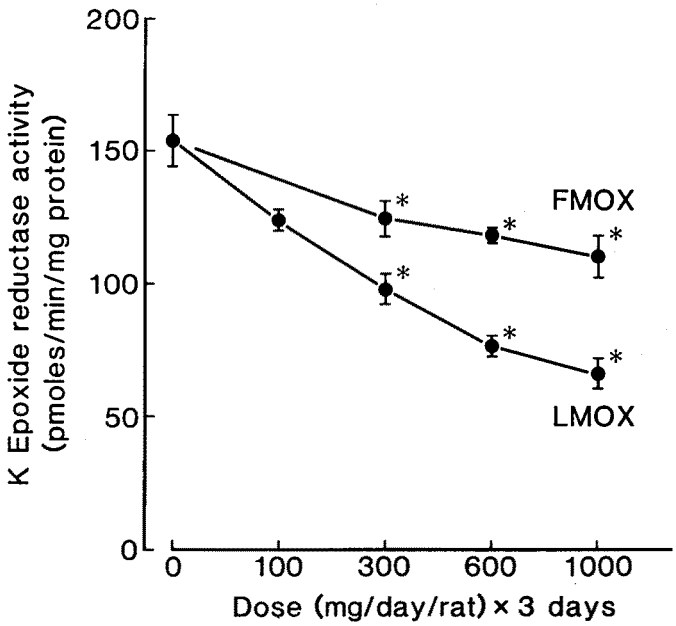


Figure 7: Effects of various doses of latamoxef and flomoxef on hepatic vitamin K epoxide reductase activity in rats. Animals were treated in the manner described in Figure 6 except that various doses of the antibiotics were injected as shown in the figure. The values in the figure represent the mean ± S. E. of 4 animals. * Statistically significant ($P < 0.05$) against the control. LMOX: Latamoxef, FMOX: Flomoxef.

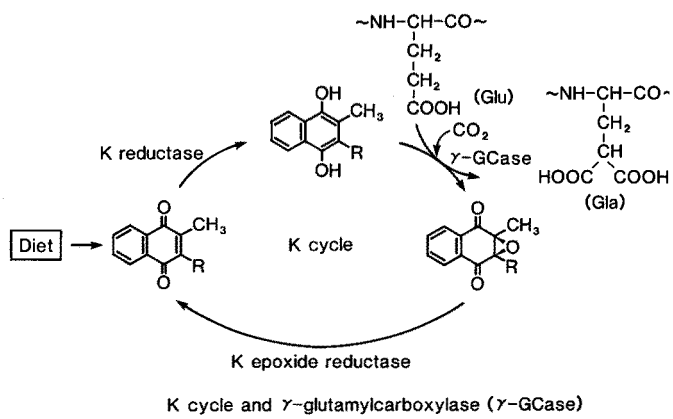


Figure 8: Vitamin K cycle and γ-glutamylcarboxylase.

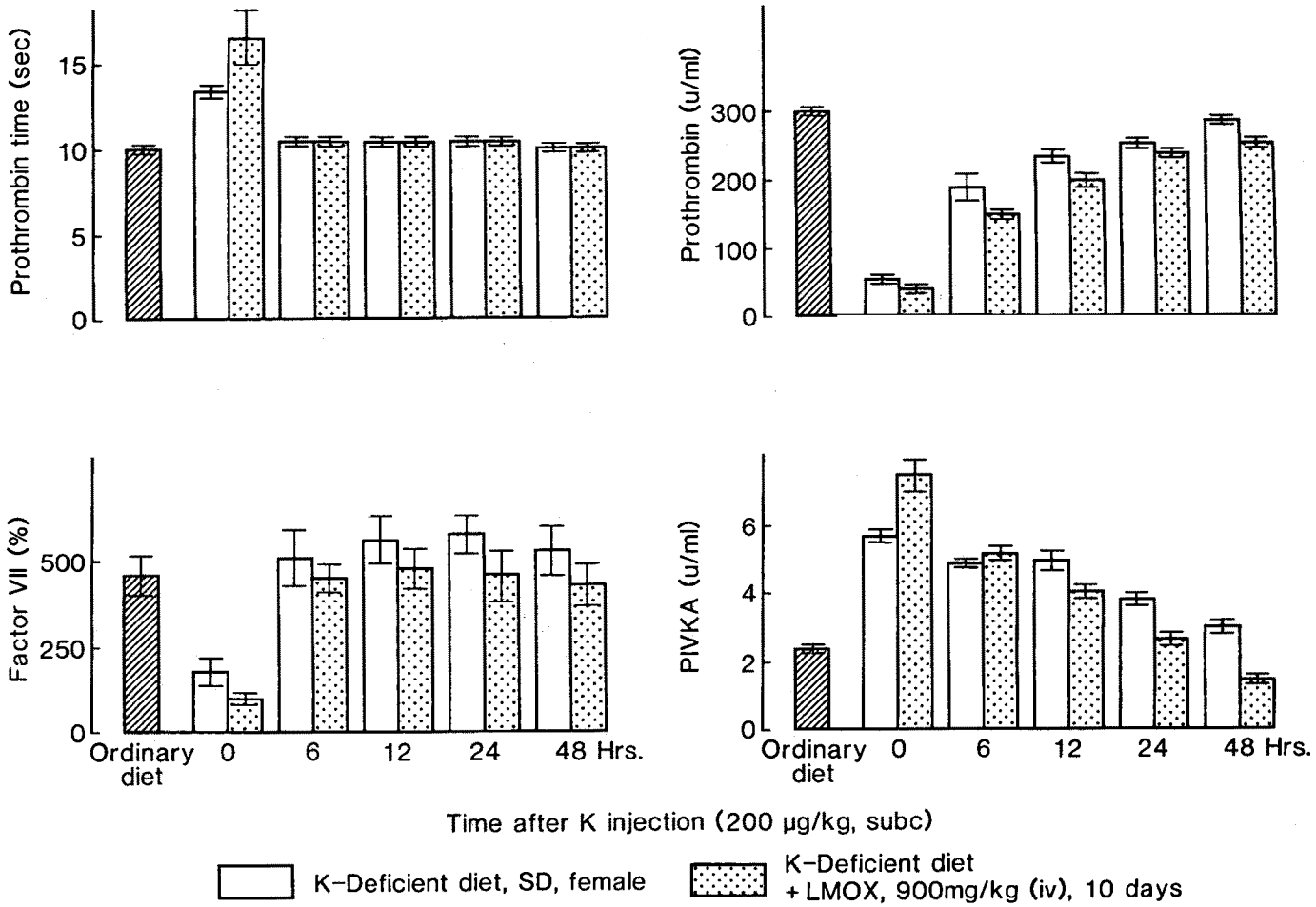


Figure 9: Recovery of the latamoxef-induced coagulopathy by a single injection of vitamin K. Rats were maintained on a vitamin K deficient diet and one half of them received the treatment of latamoxef (900 mg/kg/day, i. v.) for 10 days. Then, the animals were given a single injection of vitamin K (200 µg/kg, s. c.). The animals were further maintained on the vitamin K deficient diet with daily treatment of the antibiotic. The blood samples for the determination of blood coagulation parameters were obtained 6–48 hours after the vitamin K injection. The values represent the mean ± S. E. of 6 rats.

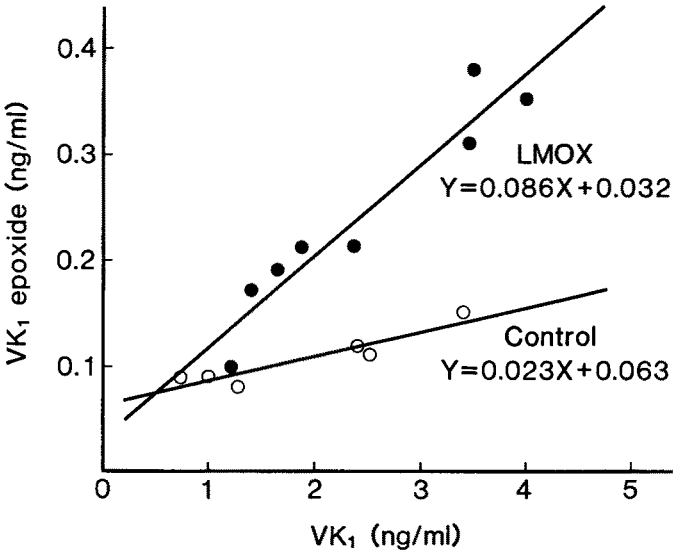


Figure 10: Correlation between the plasma levels of vitamin K_1 and vitamin K_1 epoxide. Rats maintained on an ordinary diet were divided into two groups, and the animals in one group were intravenously given latamoxef at 300 mg/kg/day for 7 days. Blood samples were obtained 3 hours after the last treatment, and the vitamin K_1 (VK_1) and vitamin K_1 epoxide concentrations in plasma were determined. The values in the individual animal are plotted as shown in the figure.

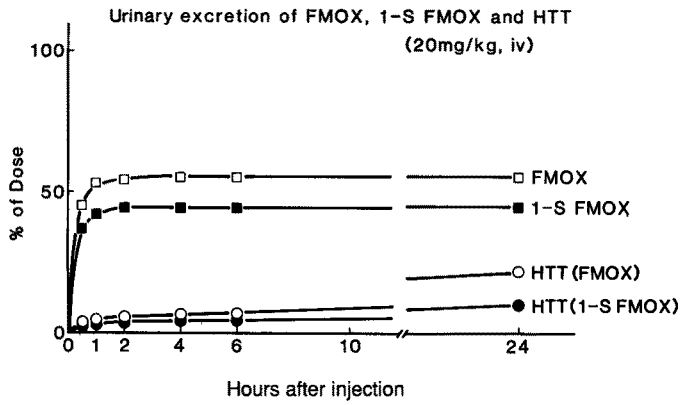


Figure 12: Urinary excretion of flomoxef (FMOX), its 1-thia congener (1-S FMOX) and the 3'-position substituent (HTT). Rats were given flomoxef and its 1-thia congener intravenously at 20 mg/kg, and the urine samples were collected periodically. Cumulative amounts of unchanged flomoxef and its 1- thia congener, and HTT liberated from the parent antibiotics in urine are shown in the figure.

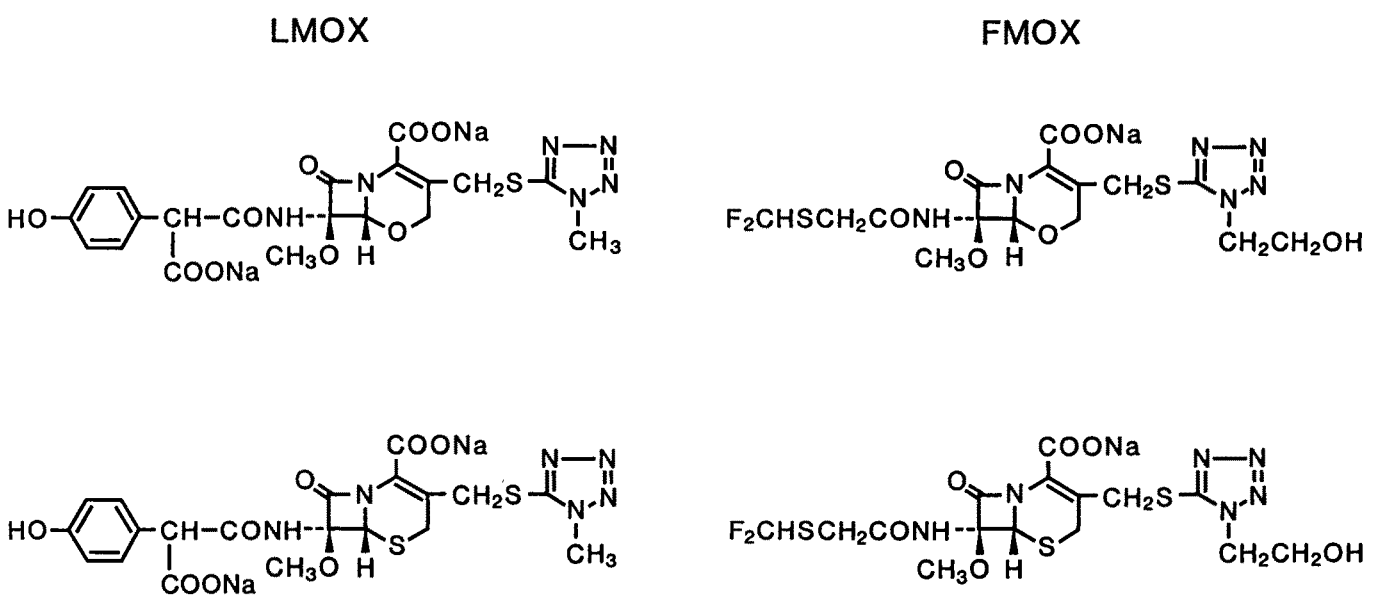


Figure 11: Chemical structures of latamoxef and flomoxef and their 1-thia congeners.

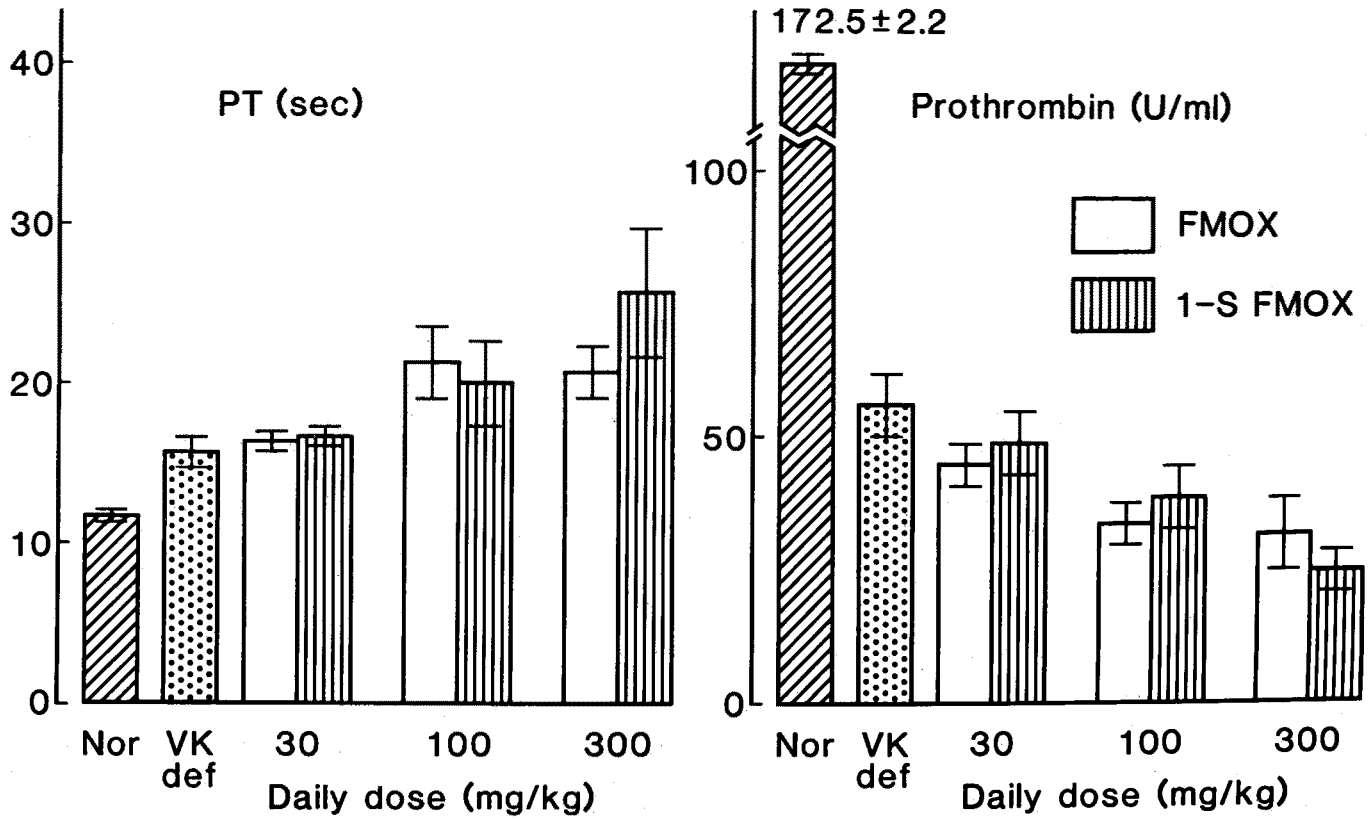
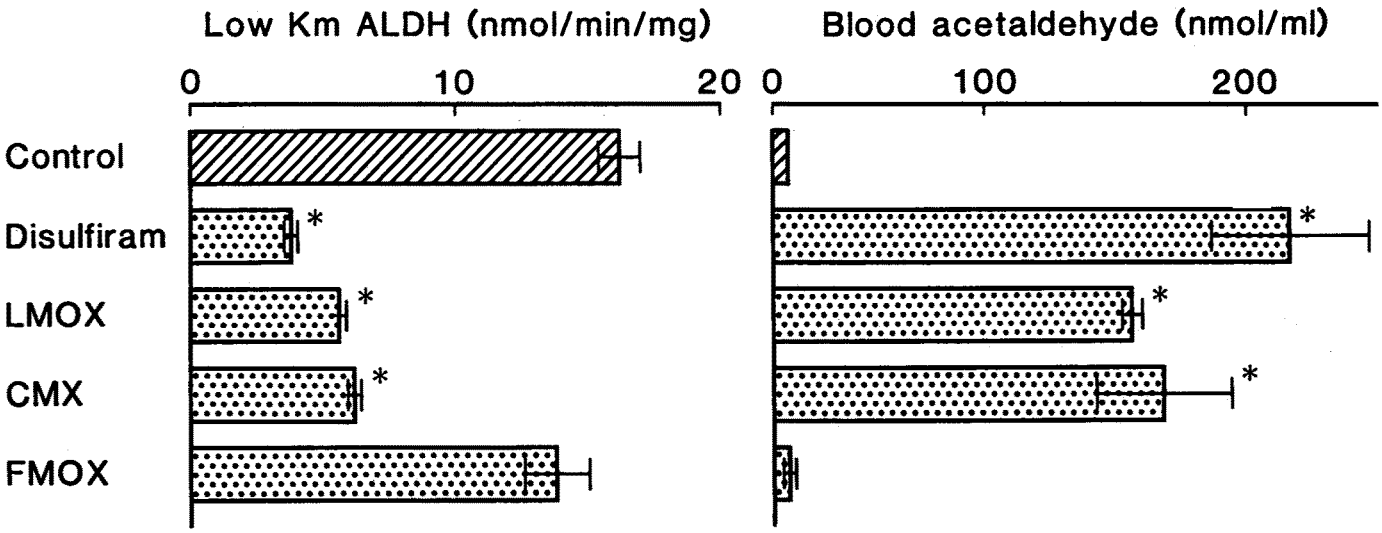


Figure 13: Comparison of flomoxef (FMOX) and its 1-thia congener (1-S FMOX) in the hypoprothrombinemic effects of the antibiotics on prothrombin time and plasma prothrombin level. Animals maintained on a vitamin K deficient diet were intravenously given flomoxef and its 1-S congener at 30, 100 and 300 mg/kg once daily for 7 days. The values in the figure represent the mean ± S. E. of 6 animals.



Disulfiram 500 mg/kg (po); Antibiotics 1000 mg/kg (subc.);
* Statistically significant ($P < 0.05$) Mean ± SE ($n = 5$).

Figure 14: Effects of disulfiram and β-lactam antibiotics on hepatic low Km aldehyde dehydrogenase activity and blood acetaldehyde level. Rats were given subcutaneous injection of the antibiotics (1,000 mg/kg) or oral administration of disulfiram (500 mg/kg), and their liver samples were obtained 24 hours later. The low Km aldehyde dehydrogenase (ALDH) activity was determined using isolated mitochondrial fraction. Control and the antibiotic- or disulfiram-pretreated rats were orally given ethanol (2,000 mg/kg) 24 hours later, and the blood acetaldehyde levels were determined 1 hour after the ethanol treatment. LMOX: Latamoxef, CMX: Cefmenoxime, FMOX: Flomoxef.

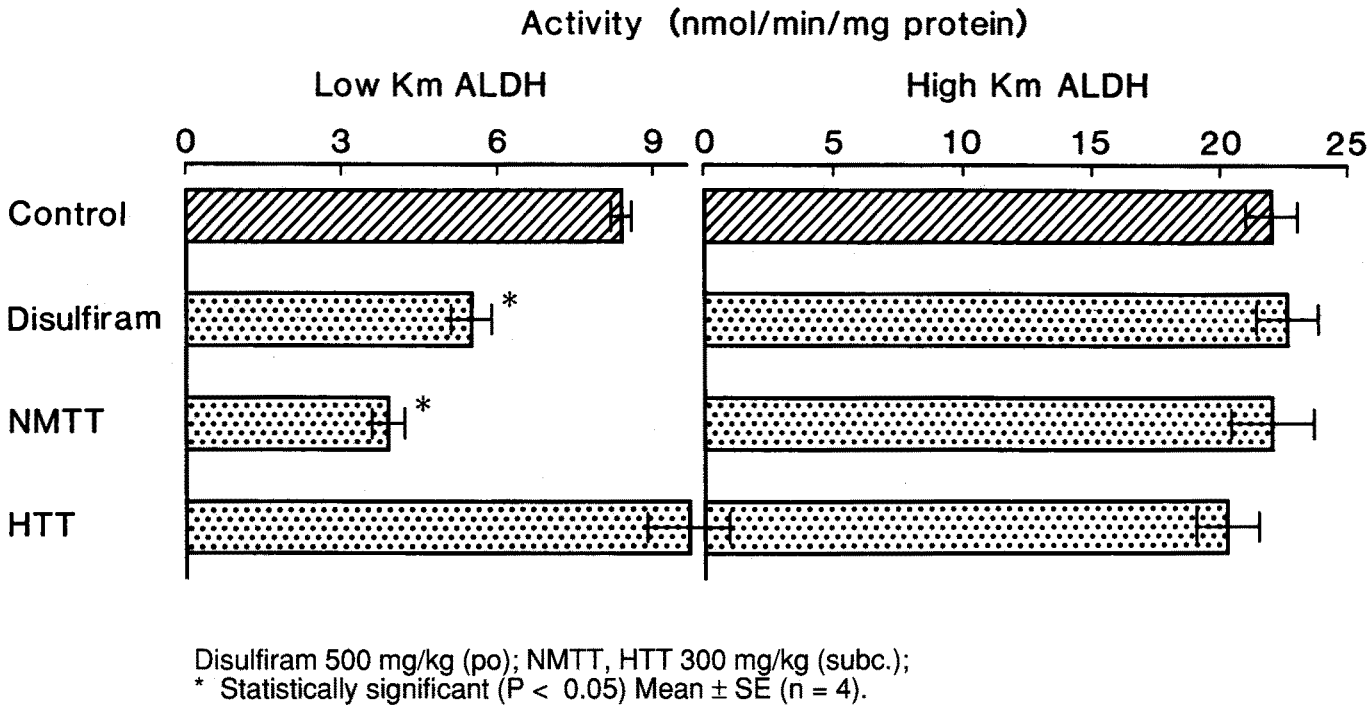


Figure 15: Effects of disulfiram and heterocyclic thiol compounds on liver aldehyde dehydrogenase activity in rats. Animals were given disulfiram or the heterocyclic thiol compounds shown in Figure 6. The liver samples were obtained 5 hours later and their low and high Km ALDH activities were determined using mitochondrial fraction.

reductase, and quickly normalized by vitamin K injection; (II) Flomoxef also inhibits vitamin K epoxide reductase but to a much lesser extent; (III) No difference is found between oxacephem (1-O) and cephem (1-S) compounds in the effects on blood clotting activities and platelet aggregation; (IV) Flomoxef and its side chain, HTT, show no influence on alcohol metabolism.

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