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Effect of Sodium Lauryl Sulphate on Rat Liver and Kidney Enzymes: An *In Vitro* Study

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The pronounced effect of surfactants on human through household commodities and cosmetic preparations (Schwuger and Bartnik, 1980; Nater and deGroot, 1983; Agarwal et al. 1986) are of special interest. Interaction of surfactants with proteins and lipids following absorption has already been established (Blackmore et al. 1979; Lay et al. 1983). Cooperative binding of anionic surfactants with proteins leading to enzymic inhibition or inactivation has also been reported (Schwuger and Bartnik 1980; Rogers and Yusko 1972; Blickhorn and Jones 1973; Jones et al.1973; Tanaka et al. 1975). However, very little is known about the *in vitro* effect of sodium lauryl sulphate (SLS), a commonly used anionic surfactant, on intact liver and kidney enzymes. Our previous *in vivo* studies on commercial synthetic detergent products containing anionic surfactant as major constituent, have shown significant variations in various liver and kidney enzymes (Gupta and Mathur 1984; Gupta et al. 1986). The present study deals with the *in vitro* effect of SLS on various enzymes of rat liver and kidney.

MATERIALS AND METHODS

Male albino rats (150 ± 10 g) were procured from Industrial Toxicology Research Centre animal breeding facility and maintained on standard pellet diet and water *ad libitum*. All the chemicals used were analytical grade purchased from Sigma, BDH and E. Merck. Rats were sacrificed by decapitation and liver and kidney removed washed and weighed. Liver and kidney homogenates (10% w/v) were prepared in cold 0.26 M sucrose using Potter-Elvehjem type homogenizer. A portion of homogenate was centrifuged at 900 g for 15 min and supernatant used as the source of enzyme. For succinic

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dehydrogenase and Mg-ATPase mitochondrial fractions were used. Mitochondrial fractions were obtained according to the procedure of Schneider and Hogeboom (1950), Mg-ATPase was determined according to the method of Emmelot and Bos (1965) and SDH by the method of Slater and Bonner (1952). Acid and alkaline phosphatases, GOT and GPT were determined in 900 g supernatant fractions according to the method described by Wootton (1964). Before adding the substrates, the reaction mixtures were pretreated with different concentrations of sodium lauryl sulphate for 15 min. at 37°C.

In separate set of experiments liver and kidney homogenates were first treated with SLS for 15 min. at 37°C and then immediately fractionated as above to achieve subcellular enzyme preparations for the assays of various enzymes. The protein content was determined according to the method of Lowry et al. (1951) using bovine serum albumin as standard.

Enzyme activities were expressed in terms of specific activity. The results were expressed as mean $\pm$ SE. Comparisons were made with appropriate controls employing Student's 't' test. Differences were considered significant at $P<0.05$.

RESULTS AND DISCUSSION

The *in vitro* effect of SLS on various liver and kidney enzymes are shown in Fig.1. All the enzymes studied showed significant inhibition at different levels of SLS. The inhibition pattern was more or less concentration dependent. Inhibition constant (Ki) as determined from Lineweaver-Burk plot revealed almost similar values for liver and kidney enzymes (Table 1). However, liver Mg-ATPase Ki value was five times higher than kidney. To obtain indepth knowledge regarding the type and nature of inhibition, more detailed kinetic

Table 1. Inhibitor constant (Ki) data on sodium lauryl sulphate interaction to rat liver and kidney enzymes

Enzyme	Ki (μM)	
	Liver	Kidney
ATPase	50.0	10.0
Succinic dehydrogenase	9.8	7.1
Acid phosphatase	12.5	14.3
Alkaline phosphatase	15.1	10.2
Glutamic oxaloacetic transaminase	20.0	16.2
Glutamic pyruvic transaminase	22.2	14.7

Ki values are determined from Lineweaver-Burk plot.

Table 2. *In vitro* effect of sodium lauryl sulphate (SLS) on liver and kidney enzymes

	Specific activity	
	Liver	Kidney
ATPase (n moles Pi/mg protein/min)		
0.0	172.89±12.71	133.39±2.85
10 µM	59.31±1.64*	41.56±1.26*
20 µM	36.78±1.49	15.17±1.02*
SDH (n moles $K_3Fe(CN)_6$ reduced/mg protein/min)		
0.0	42.56±3.88	67.78±2.20
10 µM	30.55±1.05**	43.89±3.22*
20 µM	6.42±0.69*	23.49±3.51*
Acid phosphatase (n moles/mg protein/min)		
0.0	13.23±1.03	33.64±0.64
10 µM	10.02±0.78	16.18±0.99*
20 µM	4.39±0.59*	12.84±1.10*
Alkaline phosphatase (n moles/mg protein/min)		
0.0	21.12±0.87	607.88±15.78
10 µM	10.34±0.99**	285.52±5.03*
20 µM	8.01±0.85*	224.68±12.61*
GOT (n moles hydrazones/mg protein/min)		
0.0	18.58±0.64	33.21±2.21
10 µM	12.24±1.54**	26.82±1.07
20 µM	11.15±2.60**	19.34±0.71*
GPT (n moles hydrazones/mg protein/min)		
0.0	33.21±2.23	24.12±1.60
10 µM	33.49±3.19	14.49±1.08**
20 µM	36.06±0.47	13.01±2.08**

Liver and kidney homogenates were treated with SLS for 15 min.
Other details are as given in the text.
*P<0.001; **P<0.01; Values are mean ± SD from 4-5 separate experiments.

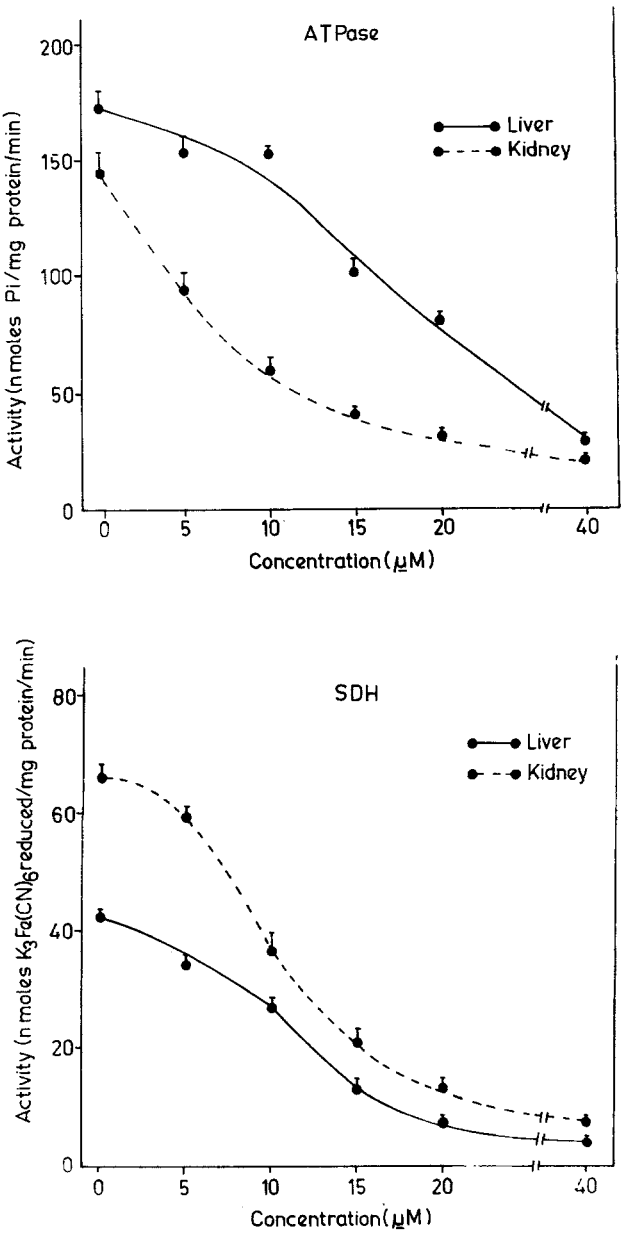
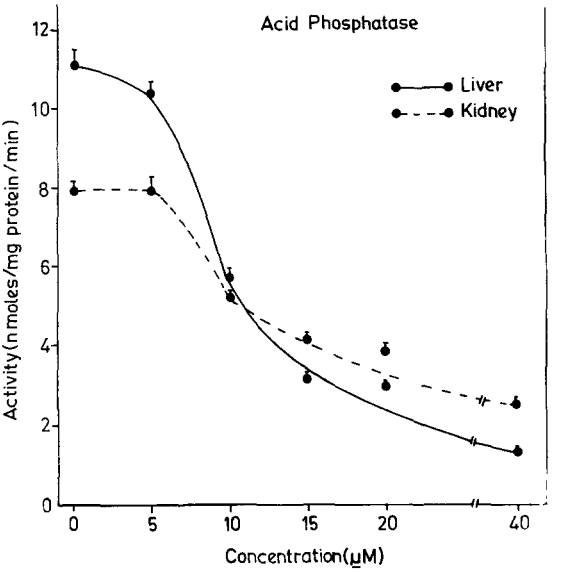
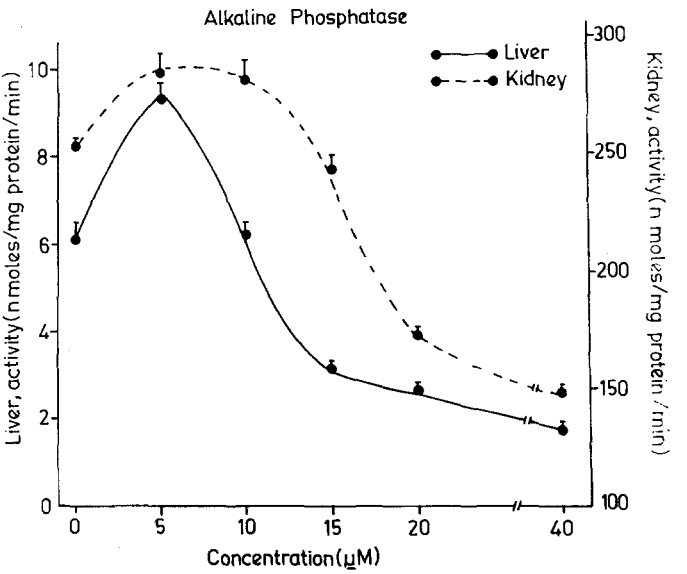


Figure 1. *In vitro* effect of sodium lauryl sulphate on various liver and kidney enzymes.





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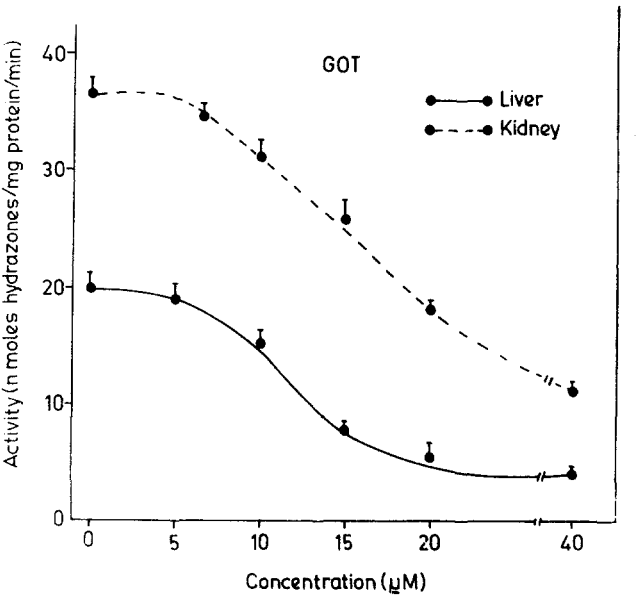

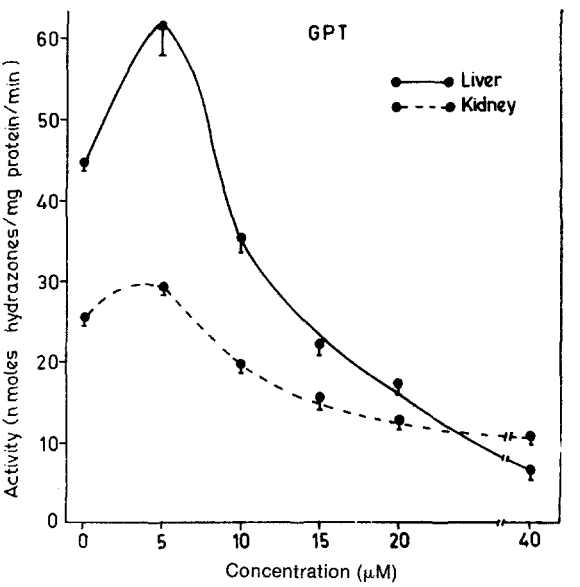

Figure 1 continued

studies are needed for each enzyme with purified preparations. Another set of experiments where the tissue homogenates were treated with SLS also revealed significant concentration dependent inhibition of all the liver and kidney enzymes (Table 2).

The toxicity of surfactants arises from its action on biological systems. Effects of surfactants are generally attributed to its ability to react directly with proteins leading to enzyme inhibition. The inhibition of membrane bound enzymes following *in vitro* treatment of SLS in the present study indicates that probably even the lower concentration of surfactant causes the cellular damage and also inhibits the enzyme activities as well. *In vitro* inhibition of Mg-ATPase by SLS has been reported in various tissues of fish (Verma et al. 1978). The inhibition of succinic dehydrogenase could be the cause of interaction of surfactant with sulfhydryl groups leading to impairment of energy metabolism (Verma et al. 1979a). Acid phosphatase, a lysosomal enzyme, hydrolyses the ester linkage of phosphate esters and helps in autolysis of the degenerated cells (De Duve et al. 1955). Inhibition of acid phosphatase, therefore, could be attributed to the action of surfactant with lysosomes (Gupta and Dhillon 1983). Alkaline phosphatase is another membrane enzyme which splits phosphorus esters at alkaline pH and mediates membrane transport. Besides, it has also been credited with a variety of other functions such as an intimate association with protein synthesis (Pilo et al. 1972), synthesis of certain enzymes (Summer 1965), secretory activity (Ibrahim et al. 1974) and glycogen metabolism (Gupta and Rao 1974). Thus the activation or inhibition of alkaline phosphatase activity in the present study could be indicative of a disturbance in these processes. Similar observations in *in vitro* studies have also been reported (Verma et al. 1979). Concomitantly GOT and GPT are known to act as an important link between carbohydrate and protein metabolism providing source of keto acids for Kreb's cycle and gluconeogenesis.

Earlier we have reported the inhibition of some of these enzymes in rat tissues following oral administration of common synthetic detergent (Gupta et al. 1986). The inhibition of various metabolic enzymes in the present *in vitro* study further strengthen the view that SLS could cause disturbance of normal metabolism and thereby create alterations in the physiology. However, further detailed *in vivo* and *in vitro* studies are needed to elucidate the mechanism of surfactant action.

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