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Increase of Sialyltransferase Activity in the Small Intestine Following Thermal Injury in Rats

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The acute phase protein response following inflammation is associated with an increased total protein-bound carbohydrate content in plasma in the form of glycoproteins. Glycosyltransferases in liver may serve as a regulator of this increased glycosylation activity in the plasma and may also serve as a marker for the acute phase response. Sialyltransferase is an example of a glycosyltransferases in which sialic acid is transferred to oligosaccarides of glycopeptides in the Golgi prior to glycopeptide secretion. In this study, sialyltransferase activities were determined in plasma, liver, and intestinal mucosa following a standardized 25% body surface area thermal injury in the rat. A statistically significant increase in sialyltransferase activity was found in liver and small intestine which were maximal at 24 hours after the injury. These increased sialyltransferase activities were accompanied by a statistically significant 2 to 4 fold elevation in plasma sialyltransferase activity at 24 hours. The plasma and liver elevations in these activities were similar to elevations seen in other models of acute inflammation using turpentine injections and bacterial infections. The increased sialyltransferase activity within the rat intestine was comparable to increases in intestinal sialyltransferase activity following colchicine treatment and may represent a similar mechanism(s).

Plasma activity of sialyltransferase, which is an enzyme which transfers sialic acid to the oligosaccarides of glycoproteins, increases in rats with bacterial infections (1), following turpentine injections (2,3), and following thermal injury (4) and is considered to be an acute phase marker in rats.(1)

Previous studies have shown that total protein-bound carbohydrate in the form of glycoproteins increases in response to injury and that most of these

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serum glycoproteins are synthesized in the liver prior to secretion.(4) In models of acute inflammation, elevated plasma sialyltransferase activity appears to result from an increased biosynthesis and release of this enzyme by the liver.(3) Inflammation may regulate the activity of glycosyltransferases in rat liver and result in increased glycosylation.

In the acute inflammatory reaction model using turpentine, elevation of sialyltransferase activity has been identified in liver homogenates.(3) However, tissue levels of sialyltransferase activity in organs other than liver have not been measured. In this study, we have determined the sialyltransferase activity in plasma, liver, and small intestine following a 25% body surface area thermal injury. The tissue sialyltransferase activity was not only elevated in plasma, but also in liver and small intestine of thermally-injured rats. These results suggest that elevated tissue activities in the intestine, liver, and plasma may be components of the acute inflammatory reaction following thermal injury in the rat and that the intestine may actively participate in that reaction.

METHODS

Animal experiments. Adult Lewis male rats weighing 175-200 g (Charles River Laboratories, Inc., Wilmington, MA) underwent a standardized thermal injury according to previously described methods.(5-7)

Animals were sacrificed by decapitation immediately after injury, 24, and 48 hours after thermal injury; the livers and small intestine were harvested. At $4^{0}C$, the small intestine was rinsed with ice-cold 0.9% NaCl and small intestinal mucosa was scraped. Liver and intestinal mucosa were homogenized in 9 volumes of 0.1 M Tris-HCL buffer pH 7.4. The homogenate was centrifuged at low speed (1,000 x g) for 10 min to remove nuclei and cell debris. The supernatant was then centrifuged at 105,000 x g for 1 h in a Beckman L5-65 ultracentrifuge with a 50.3 Ti rotor. The resulting pellets were resuspended in the same buffer by homogenization and immediately used for enzyme assay.

Plasma sialyltransferase activities were determined in blood taken from the abdominal aorta.

Using asialofetuin (Sigma Chemical Co., St. Louis, MO) as an exogenous protein acceptor, the sialyltransferase activity was measured within the linear portion of product formation with respect to incubation time and protein concentration as previously described.(8)

Using standard assays (Sigma), plasma alanine aminotransferase (SGPT) and aspartate aminotransferase (SGOT) were determined in blood samples which were taken from the tail vein without anesthesia.(7) Animals showing elevated aminotransferase activities at 24 and 48 h were used in this study. Glycosidic-bound sialic acid in plasma was determined by the periodate-resorcinol method.(9) Protein was determined by the method of Lowry using bovine serum albumin as a standard.(10)

RESULTS AND DISCUSSION

The present study was undertaken to determine the influence of thermal injury upon sialyltransferase activities in plasma, liver, and small intestinal mucosa. In other studies, elevation of the plasma activity of this enzyme has been found to be a component of the acute inflammatory reaction in rats induced by turpentine injection (3), bacterial infection (1), and thermal injury (4). In addition to increased plasma enzyme activity, elevated activity of this enzyme has also been identified in liver homogenates following turpentine injection.(3)

Consistent with the acute phase response following thermal injury, the sialic acid content of the plasma (glycosidic-bound sialic acid) significantly increased 1.5 to 1.8 fold by 24 and 48 hours respectively ($p < 0.05$ for both) as shown in Table 1. This increase in sialic acid content occurred even though plasma protein concentration significantly decreased to 67% and 60% of initial values at 24 and 48 hours respectively ($p < 0.05$).

Plasma sialyltransferase activity was significantly increased 3.8 fold and 2.7 fold at 24 and 48 h as compared to initial values (ANOVA, $p < 0.05$) as shown in Table 1. This elevation in plasma enzyme activity was similar to

Table 1. Plasma Protein, Glycosidic-bound Sialic Acid Content, and Sialyltransferase Activity Following Thermal Injury

Time	Protein Concentration	Glycosidic-bound Sialic Acid Content	Plasma Sialyltransferase Activity
h	mg/ml	nmol/ml	nmol/h/ml
0	58.9 ± 1.9	2.01 ± 0.12	17.7 ± 1.02
24	39.3 ± 2.9*	2.97 ± 0.09*	63.8 ± 2.03*
48	35.0 ± 1.2*	3.60 ± 0.21*	47.0 ± 1.99*

Values are Means ± SE of 3 plasma samples.

* Significantly ($p < 0.05$) different from 0-h control.

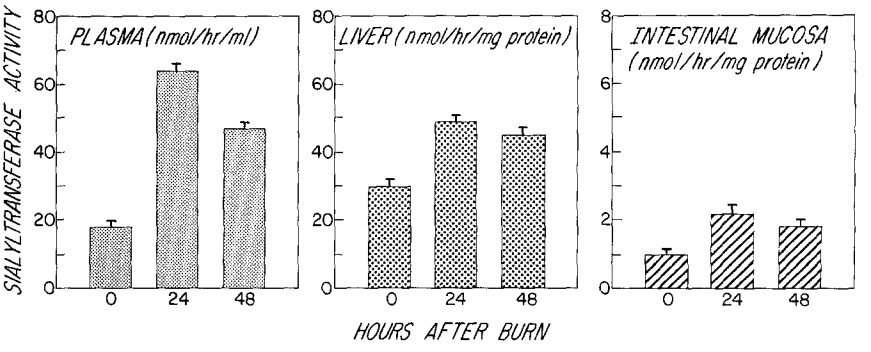

Figure 1. Sialyltransferase activity in plasma, liver, and small intestinal mucosa following thermal injury. Sialyltransferase activity was determined in plasma and in both liver and intestinal mucosa immediately after burn injury, 24, and 48 hours after injury.(n = 4, Mean $\pm$ SE)

elevations in plasma activity which have been previously reported by others.(4)

In addition to the elevation of plasma sialyltransferase activity, significant elevations in sialyltransferase activity were found in liver and small intestine following burn injury by ANOVA ($p < 0.05$) as shown in Figure 1. The activity of hepatic sialyltransferase enzyme was increased 1.6 and 1.5 fold over initial values; intestinal sialyltransferase enzyme activity was increased to 2.1 and 1.8 fold.

Though the mechanism(s) for enhanced glycosyltransferase activities in the intestine are not clear, such enzyme activities suggest that the small intestine may also actively participate in the inflammation either as a key component or in the general response to inflammatory mediators.

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