

## BINDING OF NAPROXEN TO SERUM PROTEINS IN PATIENTS WITH LIVER CIRRHOSIS AND HEALTHY INDIVIDUALS

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### ABSTRACT

A three-fold decrease of the *in vitro* ability of serum proteins to bind naproxen was shown in patients with cirrhosis of the liver compared to healthy individuals. This decrease was caused by quantitative changes in serum proteins in the sera of patients with liver damage. Lower doses of naproxen are therefore suggested in the treatment of patients with liver dysfunction.

**Keywords:** Naproxen; Liver, cirrhosis; Serum proteins.

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### INTRODUCTION

Naproxen is a non-steroidal, anti-inflammatory drug with high antiphlogistic and analgesic activity.<sup>1</sup> Previous studies have shown that the drug's hepatotoxicity is relatively low.<sup>2-5</sup> The present study attempted to measure binding of this drug to serum proteins in healthy human beings as compared to patients with cirrhosis of the liver. Hepatic cirrhosis is accompanied by significant plasma protein changes, and is a terminal state of chronic liver insufficiency. Although there is no clinical indication for naproxen treatment in patients with cirrhosis, investigations were carried out in order to determine the effect of plasma protein alterations induced by liver diseases upon the binding of naproxen to serum proteins.

On the other hand, chronic joint diseases are frequently accompanied by hepatic dysfunction, therefore changes in plasma protein patterns should be taken into consideration in dose estimations of the drug in clinical practice.

### MATERIALS AND METHODS

Naproxen was obtained commercially from Krka Nove Mesto, Yugoslavia. Sera were obtained from 15 patients of both sexes (ages 37-52 years) with liver cirrhosis. Sera were

obtained after 10 hours of fasting and prior to treatment. Patients were hospitalized in the First Ward of Internal Medicine of the 2nd Municipal Hospital in Sosnowiec. Diagnosis of the disease was based on the clinical picture and laboratory data. Control samples were obtained from 15 healthy blood donors (ages 25-48 years). Serum protein content was measured by the method of Lowry et al.<sup>6</sup> Human albumin was used as a standard. The capacity of each serum sample to bind naproxen under standard conditions was determined by equilibrium dialysis. Dialysis cells were made of collodium. Three separate measurements were made for each sample. Prior to binding measurement, sera were diluted with 0.1M Tris-HCl buffer (pH= 7.4) to a final concentration of 40g of protein per 1 liter of serum. This dilution was used in order to exclude the effect of total protein changes and the Donnan effect on the serum binding ability. The dilution was included in calculation of the results. 0.5 ml of the diluted serum was mixed with 1 ml of naproxen solution in the same buffer to a final drug concentration of 0.447 mg/ml. In preliminary investigations this concentration was shown to be sufficient to saturate the binding sites of the serum. The mixture was incubated at 37°C for 1 hour, and then transferred to dialysis cells and dialysed against 0.1M Tris-HCl buffer (pH= 7.4). The dialysis lasted 24 hrs at a controlled temperature of 20°C.

Control cells were also run, containing buffer in place of

## Naproxen Binding to Serum Proteins

**Table I.** Binding of naproxen to serum proteins in patients with cirrhosis of the liver and healthy individuals.

	mg of naproxen bound by 1 ml of serum	mg of naproxen bound by 1g of serum proteins	Percentage of naproxen bound by serum proteins
Healthy human beings	0.653 ± 0.061	10.05 ± 1.334	89.87 ± 5.13
Liver cirrhosis	0.189 ± 0.079*	3.437 ± 0.995*	30.70 ± 6.41*

\*P < 0.001

**Table II.** Hypothetical values of binding of naproxen to serum albumin in the sera of patients and in solutions of albumin.

	mg of naproxen bound by 1g of serum albumin	Percentage of albumin in total protein
Healthy human beings	19.06 ± 1.33	55 ± 4
Liver cirrhosis	8.11 ± 0.99*	40 ± 7
40% solution of human albumin	9.45 ± 0.92*	100 ± 0

\*P < 0.0001

sera, and showed that equilibrium was achieved within 24 hours. When the dialysis was over, the naproxen concentration was measured spectrophotometrically.<sup>7</sup>

The percentage of bound drug was calculated as follows;

$$\text{Percentage bound} = \frac{(C_T - C_F) \times 100}{C_T}$$

where  $C_T$  = total drug in cell, and  $C_F$  = free drug in both compartments of the cell.

The binding of the drug to serum albumin was measured in human albumin solution (40g/L) instead of serum. The routine electrophoretic method for protein separation was used to measure albumin content in the sera.

Statistical analysis of the differences was made with Student's t-test.

## RESULTS AND DISCUSSION

A significantly lower ability of serum proteins to bind naproxen was found in patients with liver cirrhosis. A

decrease in total protein level in patients with cirrhosis had no effect upon the obtained results. These results are summarized in Table I.

It is generally accepted that serum albumin is a protein fraction with a high binding ability. The results were also calculated as the amount of drug bound to the albumin fraction. Results of hypothetical values of binding of the drug to 1g albumin, assuming that albumin binds the drug exclusively, are shown in Table II. These calculations indicate that the drug is also bound by protein fractions other than albumin. Standard solutions of albumin, comparable to normal serum albumin levels, bind only about 90 percent of total drug by proteins.

A decrease in binding ability of the sera of patients with liver cirrhosis is probably caused by several factors. Cirrhotic liver damage is accompanied by quantitative changes in serum proteins. Hepatic failure diminishes metabolic activity of the liver, and plasma proteins are overloaded with some non-metabolized compounds. Changes in acid-base balance and ion concentrations may also influence binding of the drug to proteins.<sup>8,9</sup> All these phenomena are suggested as simultaneous causes of the decrease in binding ability of serum proteins.

It is possible that similar mechanisms exist in patients with liver disorders other than cirrhosis, and binding of naproxen to plasma proteins may be decreased under these conditions as well. The results obtained suggest that decreased doses of naproxen should be applied in the treatment of patients with liver dysfunctions.

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