Biliary Excretion of a New Semisynthetic Cephalosporin, Cephacetrile

J. M. BROGARD, P. HAEGELE, M. DORNER, AND J. LAVILLAUREIX

Centre Hospitalo-Universitaire de Strasbourg, 67 Strasbourg, France

Received for publication 31 July 1972

The biliary excretion of a new derivative of cephalosporin, cephacetrile (CIBA 36 278 Ba), was studied (i) in the isolated perfused rabbit liver, (ii) in humans with a duodenal tube, and (iii) in patients after cholecystectomy with a Kehr's drain in the common bile duct. The biliary excretion of the antibiotic was very low in the perfused liver, and no antibiotic activity was found in liver tissue at the end of the experiment. This observation, together with the finding of a rapid decline of the antibiotic concentration in the circulating blood serum, favors the assumption that a metabolic transformation of CIBA 36 278 Ba in liver tissue takes place. In humans, the antibiotic concentration was found to be low both in the duodenal juice and in the bile obtained by external drainage. The biliary concentrations found in these subjects seem to be inferior to those required for the inhibition of the common bacteria of biliary infections. In renal failure, however, the biliary excretion of CIBA 36 278 Ba increased considerably.

In earlier studies on the derivatives of cephalosporins, only a small part of the injected dose was found in the bile. However, the concentrations attained with some of these drugs were sufficient to raise an interest in their possible use for the therapy of certain biliary infections (1).

The objective of the present study was to evaluate the biliary excretion of the new semisynthetic cephalosporin, cephacetrile (CIBA 36 278 Ba; celospor) (4, 5). This study was performed on the isolated perfused rabbit liver. In addition, antibiotic concentrations were determined in the duodenal juice from normal subjects and from patients suffering from renal failure, and in samples collected by external biliary drainage after cholecystectomy.

MATERIALS AND METHODS

Perfusion of the isolated rabbit liver. The isolated rabbit liver was perfused by homologous, defibrinated blood, added with heparin. The blood was oxygenated and maintained at a temperature of 37 C. It reached the liver via the catheterized portal vein and poured out freely through the hepatic veins. The adequate function of such a liver was mainly assessed by satisfactory liver blood flow, by a constant bile flow, by the blood sugar level, and by a continuous formation of urea. The duration of the perfusion was 6 hr. The bile was collected throughout the experiment by a catheter introduced into the common bile duct. Five such perfusions were carried out. In each of them, 6 mg of CIBA 36 278 Ba was added to the 300 ml of circulating blood at the end of the first hour of the perfusion. At the end of the perfusion, a piece of liver tissue was obtained for the determination of the antibiotic.

Clinical study: selection of subjects. In the first group of 10 normal subjects (urea plasma N levels less than 40 mg/100 ml and creatinine clearance more than 90 ml/min), the duodenal juice was collected by duodenal drainage at hourly intervals during the 4 hr after an intramuscular injection of 1 g of CIBA 36 278 Ba. Blood samples were drawn hourly. The activity of the antibiotic was determined both in the bile and in serum samples.

The same investigations were performed in five patients with renal failure, exhibiting plasma N levels ranging from 151 to 247 mg/100 ml and creatinine clearance ranging from 5 to 20 ml/min.

In the third group of five patients choledochectomy performed 6 days previously, the serum and biliary concentrations of CIBA 36 278 Ba were followed in the same way during 8 hr after an intramuscular injection of 1 g of CIBA 36 278 Ba. The bile was collected with the use of a Kehr's drain.

Determination of the antibiotic activity. The antibiotic activity was determined by a gel-diffusion method. Strains of Bacillus subtilis (3 × 10⁴ cells per/ml of gel) were used as the test organism. The liver concentration of CIBA 36 278 Ba was measured after crushing and emulsifying the tissue fragments. Serum was used for a calibration curve. It was found that equal concentrations of the antibiotic gave similar inhibition zones in the presence of bile, serum, and various dilutions.
RESULTS

Kinetics of CIBA 36 278 Ba concentrations during the perfusion of the isolated rabbit liver. A rapid decrease in the activity of the antibiotic in circulating blood was observed in the five perfusions studied (Table 1). Whereas the initial theoretical value was about 15 μg/ml (taking into consideration 300 ml of circulating blood and 80 g of perfused hepatic tissue), the average serum concentration found 1 hr after adding 6 mg of CIBA 36 278 Ba was 7.3 ± 0.6 μg/ml. After 5 hr of perfusion, the average concentration was only 1.2 ± 0.6 μg/ml.

The biliary concentration was low, except for one experiment in which it was found to be 17.5 μg/ml. The average maximal concentration for the four other experiments was 2.1 μg/ml. The biliary peak was found at the end of the first hour in two experiments, at the end of the second hour in two experiments, and at the end of the third hour in one experiment.

In none of these experiments could the antibiotic be detected in the liver fragments taken at the end of the perfusion. It should be noted that after 5 hr of perfusion no antibiotic could be detected in the serum of two among the five perfusion experiments.

Kinetics of the CIBA 36 278 Ba concentration in duodenal juice of normal subjects. In confirmation of previous findings (3), we observed that, when 1 g of CIBA 36 278 Ba was administered intramuscularly to 10 normal subjects, the maximal serum activity of about 15 μg/ml occurred approximately 1 hr after the injection (Table 2). The serum half-life of the subsequent decline was 1.5 hr.

The concentration of CIBA 36 278 Ba found in the duodenal juice was low: 0.7 ± 0.2, 1.2 ± 0.3, 0.6 ± 0.2, and 0.1 ± 0.1 μg/ml at the first, second, third, and fourth hour, respectively. The maximal concentrations ranged from 1.0 to 3.1 μg/ml and were attained at the first or second hour. In three of these subjects, no drug was found in the duodenal juice.

Kinetics of CIBA 36 278 Ba concentration in duodenal juice of patients with renal failure. As shown elsewhere by others (2–6) and by us, altered renal function is accompanied by a drop in the urinary elimination of CIBA 36 278 Ba. Thus, in the five patients with renal failure, the serum concentrations of the antibiotic rose significantly (40.6 ± 8.1 μg/ml) 1 hr after injection, and their subsequent decline was slowed down (Table 3). Under these conditions, a considerable increase was noted in the serum half-life of the antibiotic, varying between 13 and 30 hr in the five patients studied.

In duodenal juice, the concentration of CIBA 36 278 Ba was found to be higher than in normal subjects; the peaks, ranging from 29 to 7 μg/ml, were attained at the first hour in one patient, at the second hour in three patients, and at the third hour in one patient. The biliary transport of the antibiotic was still important during the third hour, but decreased rapidly during the fourth hour.

CIBA 36 278 Ba concentration in hepatic bile obtained by Kehr’s drain in cholecystectomized subjects. The drug concentrations in serum observed after intramuscular administration of 1 g of CIBA 36 278 Ba to cholecystectomized patients (Table 4) were comparable to those recorded in normal subjects with a duodenal tube, except for the higher concentrations reached at the first hour in the present group (average, 19.4 μg/ml versus 14.7 μg/ml).

In the five cholecystectomized subjects, the biliary concentrations were found to be low. The maximal values were 1.0, 1.6, 3.0, 4.8, and 6.8 μg/ml, and were found between the first and fourth hour. Complete disappearance of the antibiotic occurred within 4 hr in three patients and within 5 hr in one patient; a weak activity was found at the eighth hour in one patient.

DISCUSSION

The biliary elimination of CIBA 36 278 Ba was low in the isolated liver perfusion studies, although this is the only way in which the antibiotic may be excreted. As the disappearance of this drug from the serum was rapid, its inactivation by the liver may be assumed.

Control experiments proved that the serum concentration of CIBA 36 278 Ba varied to an insignificant degree only in defibrinated and heparinized blood conserved during 6 hr at 37

---

**Table 1. Mean concentrations of CIBA 36 278 Ba in serum and bile after addition of 6 mg of the antibiotic in five isolated rabbit liver perfusions**

<table>
<thead>
<tr>
<th>Time (hr)</th>
<th>Mean concn ± SE (μg/ml)</th>
<th>Serum</th>
<th>Bile</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>1</td>
<td>7.3 ± 0.6</td>
<td>5.2 ± 3.0</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>5.9 ± 0.7</td>
<td>4.5 ± 2.1</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>4.7 ± 0.9</td>
<td>2.8 ± 0.6</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>2.4 ± 0.6</td>
<td>1.8 ± 0.2</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>1.2 ± 0.6</td>
<td>0.6 ± 0.4</td>
<td></td>
</tr>
</tbody>
</table>

* Biliary volumes varied from 1 to 14 ml/hr.
C. On the other hand, if defibrinated and hep-arinized rabbit blood to which CIBA 36 278 Ba had been added circulated for 6 hr in the perfusion apparatus, without perfused liver, the antibiotic serum concentration declined by only 23% between the first and the sixth hour of the experiment. Various mechanisms may be considered to explain this drop in serum antibiotic activity: fixation of the antibiotic on the walls of the polyethylene tubes or of the glass of the perfusion unit, its oxidation in the oxygenator due to a prolonged contact with the gas mixture (two parts of O₂ and one part of 7% CO₂) or an increase in the plasma protein binding due to the in vitro conditions.

It was also possible to show that activity of the antibiotic incubated at 37°C with serum and liver homogenates remained stable for 6 hr. It should, therefore, be assumed that, in contrast to the liver incubated in vitro, the perfused liver is able to inactivate CIBA 36 278 Ba. In contrast to other cephalosporins (cephaloridine, cephalaxin [1]) studied with the use of isolated liver perfusion, the liver did not contain any CIBA 36 278 Ba at the end of the procedure. The existence of a hepatic metabolism of the antibiotic could account for the low biliary transport of CIBA 36 278 Ba, at least in its active form, as detected by the estimation of its inhibitory potency on test strains.
The human findings are in keeping with those made in animals. The weak concentration of the antibiotic in the duodenal juice may be due to dilution by other gastrointestinal juices. Actually, the low concentrations found in patients with Kehr’s drainage favor the assumption that this cephalosporin is only weakly excreted by the liver. The antibiotic activity of the bile after the injection of 1 g of CIBA 36 278 Ba is just sufficient to inhibit during about 5 hr the growth of some gram-positive organisms from its antibacterial spectrum (4). Therefore, it does not seem that biliary infections are an indication for the exclusive application of this new derivative of cephalosporin, at least at the doses applied by us.

On the other hand, in renal failure, its biliary excretion increased greatly and its activity in the bile was of such a degree that it equaled or exceeded during 2 to 3 hr the minimal inhibitory concentration for most of the susceptible organisms.

ACKNOWLEDGMENTS

This investigation was supported by a grant from CIBA Laboratories. We are indebted to D. Babin and C. Renaud
for technical assistance and to R. Braun for the microbiological assays.

LITERATURE CITED


