Tohoku J. Exp. Med., 1989, **157**, 31-37

Effect of Blood Exchange Transfusion as an Initial Treatment of Acute Hemorrhagic Pancreatitis

KAZUNORI TAKEDA, TOSHIHIKO SUZUKI, MAKOTO SUNAMURA, SHUJI MATSUBARA, MASAO KOBARI and SEIKI MATSUNO

The First Department of Surgery, Tohoku University School of Medicine, Sendai 980

TAKEDA, K., SUZUKI, T., SUNAMURA, M., MATSUBARA, S., KOBARI, M. and MATSUNO, S. *Effect of Blood Exchange Transfusion as an Initial Treatment of Acute Hemorrhagic Pancreatitis*. Tohoku J. Exp. Med., 1989, **157** (1), 31-37 —— The effect of blood exchange transfusion on experimental acute pancreatitis was investigated. Acute hemorrhagic pancreatitis was induced in rats by the injection of sodium taurocholate into the pancreatic duct. One hour after the induction of pancreatitis, blood exchange transfusion was carried out in these rats. The survival rates by 24 hr significantly increased in the exchange transfusion group as compared to those in the non-treated control group. Blood exchange transfusion, though had not significant effect on the macroscopic findings at autopsy, reduced pulmonary edema. These results suggested that blood purification by exchange transfusion may be beneficial for patients with acute pancreatitis, especially at an early stage. ———— acute pancreatitis; blood exchange; blood purification; water content of the lung

Clinical feature of the severe acute pancreatitis is characterized by systemic organ failure including cardiovascular, pulmonary and renal insufficiency at an early stage. These complications have been attributed to the activated pancreatic enzymes themselves and secondarily to the toxic substances produced in the inflamed pancreas. To purify these noxious factors, we applied plasma exchange for acute pancreatitis as a blood purification on acute pancreatitis (Yamauchi et al. 1985). To objectively verify the effectiveness of the blood purification, we conducted the blood exchange transfusion to rats with acute experimental pancreatitis in view of the changes in the survival rate and the organ impairments.

MATERIALS AND METHODS

Wistar-strain rats weighing 250-300 g were used. After 12 to 18 hr fasting, the animals were laparotomized under ether anesthesia. A polyethylene tube, 1 mm diameter, was inserted into the femoral vein, so that the tip was indwelt in the inferior vena cava as a blood access for the exchange transfusion.

Received June 28, 1988; revision accepted for publication November 29, 1988.

Induction of acute pancreatitis

The common bile duct was temporarily clamped at its end, and 0.5 ml of 5% sodium taurocholate was infused retrogradely into the pancreatic duct via bile duct with a water pressure of 100 cm. The common bile duct was ligated at the upper and the lower portion of the infused region to prevent the leakage of the bile (Fig. 1).

The experimental animals were divided into two groups as follows: Group I: rats with acute hemorrhagic pancreatitis (non-treated control), and Group II: rats with acute hemorrhagic pancreatitis treated with blood exchange transfusion 1 hr after the induction of pancreatitis.

Blood exchange transfusion was performed in the following method using 12 ml of fresh blood exanguinated from healthy rats, six times of exchange transfusion (2 ml per each session) was made within 30 min with polyethylene tube previously indwelt in the inferior vena cava. After the exchange transfusion, animals were brought back to the general cage, and were given food and water freely.

Determination of the biochemical parameters and tissue water content of the lung

Serum amylase levels, serum glutamic pyruvic transaminase (GPT) levels, serum creatinine levels and serum total bilirubin levels were analyzed in these animals. Amylase was measured with Caraway's method (Caraway 1959). GPT, total bilirubin and creatinine levels were measured with autoanalyzer using Uni-Kit (Chugai Pharmaceutical Co., Tokyo).

Tissue water content of the lung was determined in both group I and group II. The rat of each group were sacrificed 12 hr after the induction of pancreatitis. The wet wight of lung and the dry weight incubated for 72 hr at 80°C were mesured. Tissue water content of the lung was determined with (wet weight-dry weight)/body weight.

Histological examinations of the pancreas, liver, kidneys and lungs

Ten rats in each group were sacrificed 12 hr after the induction of pancreatitis. The pancreas liver, kidneys and lungs were extirpated and fixed with formalin. The microscopic examinations were made by hematoxylin-eosin staining.

Survival rates in both experimental groups

The survival rates in both groups were calculated 12, 24, 36 and 48 hr after the

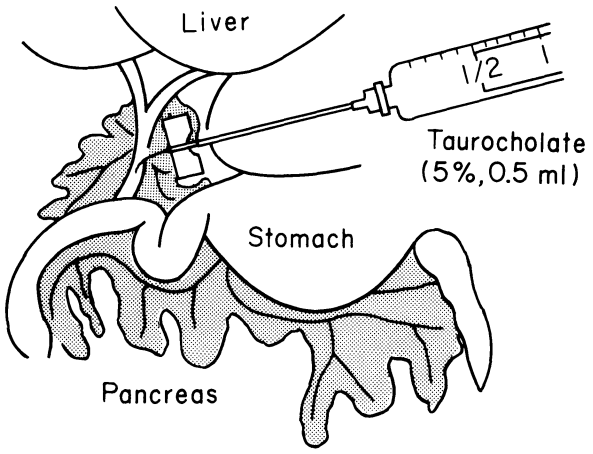

Fig. 1. Experimental model of acute pancreatitis in the rats.

induction of pancreatitis.

Statistical analysis

Statistical significance was evaluated with the chi-square and the unpaired Student's t-test, and $p<0.01$ was assesed to be significant.

Results

Histological changes in the pancreas

Macroscopic findings of the pancreas in group I (non-treated control) showed hemorrhagic pancreatitis with sporadic fat necrosis on the surface and 3 to 5 ml of hemorrhagic exudate accumulating in the abdominal cavity. Histological findings also revealed hemmorrhagic-necrotizing pancreatitis accompanying bleeding and necrotic changes in the parenchyma and inflammatory cellular infiltration (Fig. 2). These findings were not significantly different in group II.

Survival rates in both groups

In group I (non-treated control, $n=30$), the survival rates were 43.3, 13.3, 10 and 0% at 12, 24, 36 and 48 hr, respectively. On the other hand, the survival rates in group II ($n=20$) were 95% after 12 hr, 78% after 24 hr, 45% after 36 hr, and 10% after 48 hr. The survival rates of group II at 12 and 24 hr were significantly high ($p<0.01$) as compared to those of group I (Fig. 3).

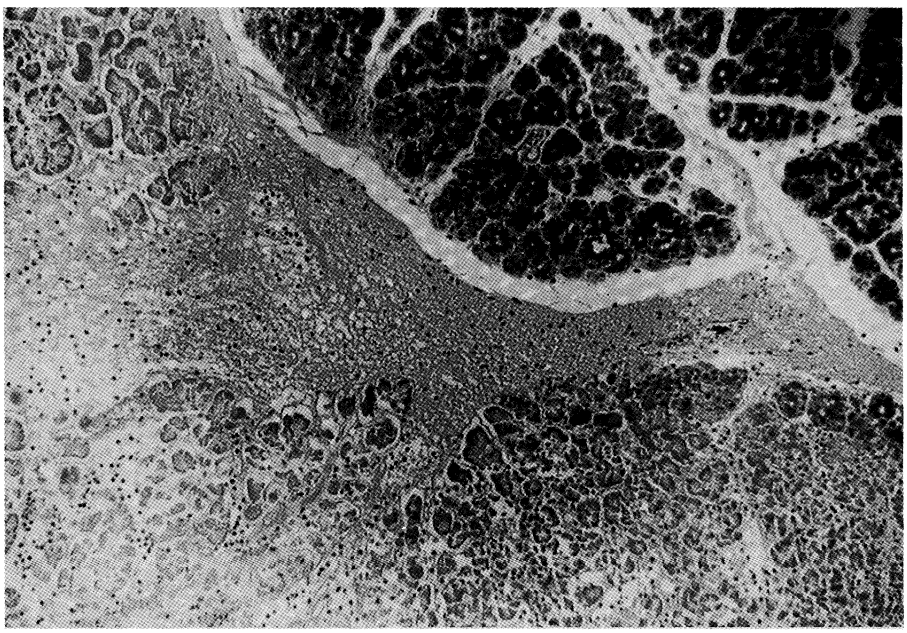

Fig. 2. Histological changes in taurocholate-induced pancreatitis in a rat after 6 hr.

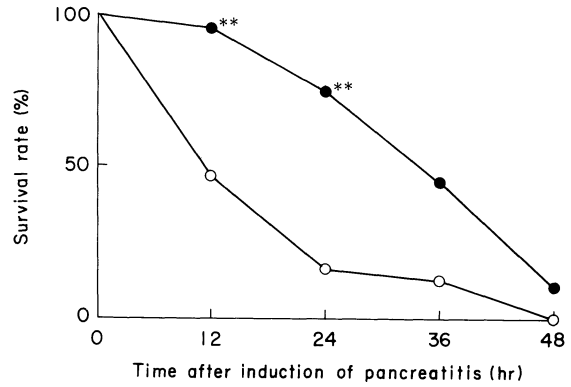

Fig. 3. Survival rates in both group I (○—○) and group II (●—●). $^{**}p<0.01$ compared to group I at each 12 and 24 hr after the induction of pancreatitis.

Changes in the serum amylase levels

In group I, the serum amylase levels elevated to 12,000±1,500 U/100 ml 6 hr after the induction of pancreatitis and 11,000±1,000 U/100 ml after 12 hr. In group II, they were 9,400±1,900 U/100 ml after 6 hr and 9,300±1,800 U/100 ml after 12 hr, respectively. There was no significant difference between group I and group II (Fig. 4).

Changes in the serum bilirubin, GPT and creatinine levels

Because of the ligation of the common bile duct, the sequential elevation of the serum bilirubin level was observed. The prevalue was 0.3±0.2 mg/100 ml in

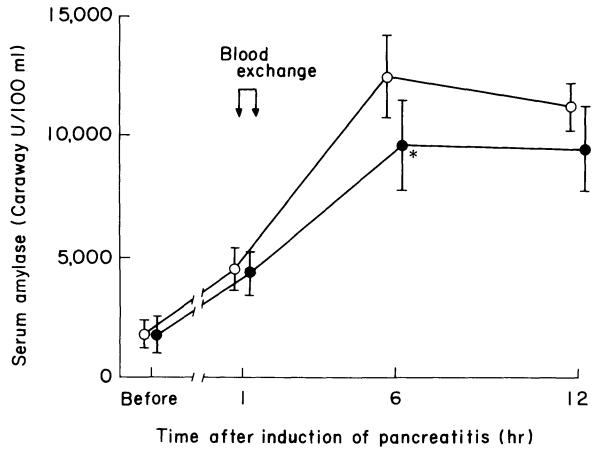

Fig. 4. Time course of serum amylase levels in both group I (○—○) and group II (●—●). $^{*}p<0.05$ compared to serum amylase levels in group I 6 hr after the induction of pancreatitis.

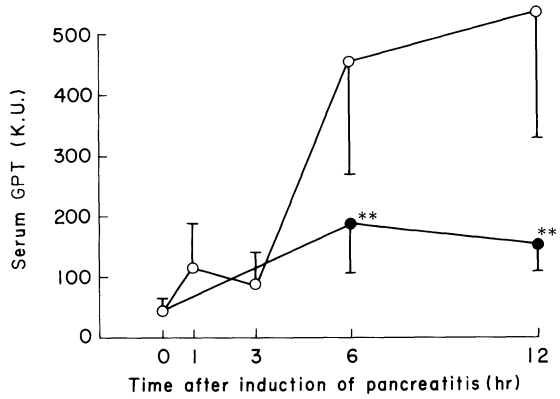

Fig. 5. Time course of serum GPT levels in both group I (○—○) and group II (●—●). $^{**}p<0.01$ compared to serum GPT levels in group I at each 6 and 12 hr after the induction of pancreatitis.

group I and 0.5 ± 0.2 mg/100 ml in group II. At 12 hr the serum bilirubin levels were 4.2 ± 2.1 mg/100 ml in group I and 3.8 ± 2.1 mg/100 ml in group II. There is no significant difference between group I and II. However, GPT levels in group I were 460 ± 180 Karmen Units (K.U.) after 6 hr and 530 ± 150 K.U. after 12 hr, whereas those in group II were 200 ± 40 K.U. after 6 hr and 180 ± 37 K.U. after 12 hr, respectively, showing the significant decrease (Fig. 5).

Pre-treatment serum creatinine levels were 1.1 ± 0.3 mg/100 ml in group I and 1.0 ± 0.5 mg/100 ml in group II. The serum creatinine levels showed no significant change during 24 hr in both groups.

Tissue water content of the lung (WL)

The water content of the lung tissue (WL) was $(2.1\pm0.5)\times10^{-3}$ g/b.w. in

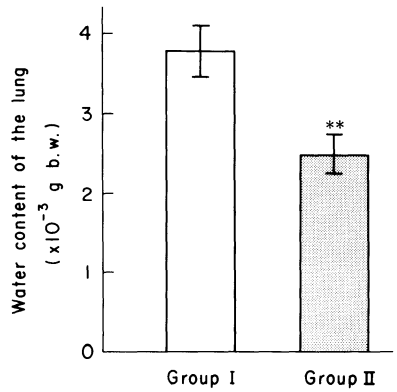

Fig. 6. Water content of the lung in both group I (open colum) and group II (meshed column). $^{**}p<0.01$ compared to water content of the lung in group I at 12 hr after the induction of pancreatitis.

normal rats. WL in group I were $(3.3\pm0.5)\times10^{-3}$ g/b.w. 6 hr after the induction of pancreatitis, and they were markedly elevated to $(3.7\pm0.2)\times10^{-3}$ g/b.w. 12 hr after the induction of pancreatitis. WL in group II were $(3.3\pm0.2)\times10^{-3}$ g/b.w. after 6 hr, almost equal to those in group I, and $(2.4\pm0.2)\times10^{-3}$ g/b.w. after 12 hr, demonstrating significantly reduced values (Fig. 6).

Histological changes in the liver, kidney and lung

Histological findings of the liver showed the feathery degeneration, but neither hepatocellular necrosis nor intralobular fibrogenesis in both group I and II. The difference between two groups could not be histologically elucidated. Histological findings of the kidney revealed no pathological changes in the renal tubules, glomeruli and capillaries 12 hr after the induction of pancreatitis in both group I and II. Gross appearance of the lung in group I revealed hyperemia and a slight hemorrhage, suspected of the edematous changes in the lung, however, colloid retention and marked inflammatory cellular infiltration were not observed. In group II, gross appearance showed only a slight hyperemia and no relevant changes histologically.

Discussion

Conventional therapeutic interventions such as peritoneal lavage (Wall 1965; Ranson and Spencer 1978) and operative peripancreatic drainage (Lawson et al. 1970) have been made to remove toxic substances in the abdominal cavity in the case of acute pancreatitis. However, these procedures failed to remove pancreatic enzymes and toxic substances from the blood stream because they are non-dializable macromollecular substances by peritoneal dialysis. Previously, we reported plasma exchange as a blood purification in severe acute pancreatitis (Yamauchi et al. 1985). Plasma exchange of four litters reduced levels of serum amylase, lipase and trypsin sequentially to the half of the pre-value in a patient with acute pancreatitis. Plasma exchange is useful for rapid removal of the pancreatic enzymes from the blood stream, however, basic studies such as prospective effect on multiple organ failure and the effect on the mortality rate are not yet clearly demonstrated. Thus, we conducted blood exchange transfusion as a blood purification in acute experimental pancreatitis model in rats, and elucidated its effects on survival rates and organ failure.

Our experimental results revealed significant effects of blood purification on survival rates and lung injury in the early period of acute panceatitis. In this model, 24 hr after the induction of pancreatitis only 13% of rats survived in non-treated control group, whereas, 78% of rats with exchange transfusion survived. The incidence of respiratory insufficiency is very common in the early stage of severe acute pancreatitis (Ranson et al. 1974), and lung injury is assumed to be caused by increasing vascular permeability (Takada et al. 1976) and pulmonaly capillary impairment (Ashbaugh and Uzawa 1968) due to pancreatic

enzymes such as kinin substances and free fatty acids. In this study, a marked increase in water content of the lung suggesting the progression of pulmonary edema was found in the non-treated control group. On the contrary, water content of the lung showed no increase in the exchange transfusion group.

Based upon these results blood exchange transfusion may be capable in alleviating the severity of acute pancreatitis by removing toxic substances from the blood stream as is done in the plasma exchange therapy. In addition, the blood or plasma exchange not only removes toxic substances but also supplements protease inhibitors (Balldin and Ohlsson 1979) and opsonic proteins (Pisano and Luzio 1970) being contained in the fresh blood or plasma, and this process may also be useful in preventing the progress of disease.

In conclusion, our experimental results suggest the beneficial effects of blood purification on survival rates and lung injury. When blood purification is performed combined with either peritoneal lavage or surgical drainage of the peripancreatic exudates in early stage, further benefits may be expected for the improvement of mortality rate in severe acute pancreatitis.

References

1) Ashbaugh, D.G. & Uzawa, T. (1968) Respiratory and hemodynamic changes after injection of free fatty acids. *J. Surg. Res.*, **8**, 417-423.
2) Balldin, G. & Ohlsson, K. (1979) Demonstration of pancreatic protease-antiprotease complexes in the peritoneal fluid of patients with acute pancreatitis. *Surgery*, **85**, 451-456.
3) Caraway, W.T. (1959) A stable starch substrate for the determination of amylase in serum and other body fluid. *Am. J. Clin. Pathol.*, **32**, 97-108.
6) Lawson, D.W., Dagget, W.M., Civetta, J.M., Corry, R.J. & Bartlett, M.K. (1970) Surgical treatment of acute necrotizing pancreatitis. *Ann. Surg.*, **172**, 605-617.
5) Pisano, J.C. & Luzio, N.R. (1970) Purification of an opsonic protein fraction from rat serum. *J. Reticuloendoth. Soc.*, **7**, 386-396.
6) Ranson, J.H.C. & Spencer, F.C. (1978) The role of peritoneal lavage in severe acute pancreatitis. *Ann. Surg.*, **187**, 565-575.
7) Ranson, J.H.C., Roses, D.F. & Fink, S.D. (1974) Respiratory complications in acute pancreatitis. *Ann. Surg.*, **179**, 557-566.
8) Takada, Y., Appert, H.E. & Howard, J.M. (1975) Vascular permeability induced by pancreatic exudate formed during acute pancreatitis in dogs. *Surg. Gynecol. Obstet.*, **143**, 779-783.
9) Yamauchi, H., Takeda, K., Sunamura, M., Miyagawa, K., Matsubara, S., Asanuma, Y. & Sato, T. (1985) Effect of plasma exchange on acute pancreatitis. *Tohoku J. Exp. Med.*, **145**, 103-104.
10) Wall, A.J. (1965) Peritoneal dialysis in the treatment of severe acute pancreatitis. *Med. J. Aust.*, **2**, 281-283.