

Platelet activation and the protective effect of aprotinin in hepatolithiasis patients

Wen-Ming Feng, Ying Bao, Mao-Yun Fei, Qiu-Qiang Chen, Qi Yang and Chuang Dai
Huzhou, China

OBJECTIVE: To explore platelet activation and the protective effect of aprotinin in patients with hepatolithiasis.

METHODS: The count of platelets and levels of CD_{62P} and CD₆₃ were measured by flow cytometry in 38 patients with hepatolithiasis. Several measurements were carried out after treatment with aprotinin.

RESULTS: The levels of CD_{62P}, CD₆₃ in patients with hepatolithiasis were higher than those in patients with cholecystolithiasis ($P < 0.05$), but the count of platelets was lower ($P < 0.05$). After operation, the levels of CD_{62P}, CD₆₃ were significantly increased in patients with hepatolithiasis, but the count of platelets was lower ($P < 0.05$). Postoperative levels of CD_{62P}, CD₆₃ were significantly lower in patients treated with aprotinin than in normal controls ($P < 0.05$); but there was no significant change in the count of platelets in the two groups.

CONCLUSION: Platelet activation occurs in patients with hepatolithiasis, and may be inhibited by aprotinin.

(*HBDP Int 2003; 2; 602-604*)

Key words: hepatolithiasis; platelet membrane glycoproteins; aprotinin

Introduction

Patients with obstructive jaundice caused by hepatolithiasis may have secondarily enhanced fibrinolysis.^[1] Studies have revealed that patients with hepatolithiasis have a decreased amount of platelets and enhanced aggregation.^[2] The use of aprotinin results in reduction of intraoperative blood loss and less transfusion during operation.^[3] By measuring the expression levels of anti-alpha granule membrane glycoprotein (CD_{62P}) and anti-lysosomal integral membrane glycoprotein GP53 (CD₆₃) in peripheral vein blood, we attempted to assess the status of platelet activation in this study. In addition,

we administered aprotinin in some patients to observe the effect of platelet activation.

Methods

Patients

Thirty-eight patients with hepatolithiasis diagnosed according to the Diagnostic Criteria for Hepatolithiasis^[4] at our hospital from January 2001 to May 2002 were confirmed that part of patients were caused by biliary calculus through biliarygraphy in the procedure of surgery. The patients were randomly divided into groups I and II. The hepatolithiasis group I consisted of 19 patients (9 men and 10 women) aged from 36 to 67 years (mean 41.5 years). APACHE II value within the first 24 hours was 5.4 ± 2.1 , and the bilirubin level was 41.4 ± 10.9 . Group II who were treated with aprotinin (aprotinin 2×10^7 IU ivgtt. used one hour before operation until 5 days after operation) was composed of 19 patients (10 men and 9 women), aged from 32 to 68 years (mean 42.3 years).

From the Department of Hepatobiliary Surgery, First Hospital of Huzhou, Huzhou 313000, China (Feng WM, Bao Y, Fei MY, Chen QQ, Yang Q and Dai C)

Correspondence: Wen-Ming Feng, MD, Department of Hepatobiliary Surgery, First Hospital of Huzhou, Huzhou 313000, China (Tel: 86-572-2211208; Fax: 86-572-2023728; Email: d_fum@hotmail.com)

Table. Comparison of platelet, CD_{62P}, CD₆₃ in each group before and after surgery

Group	n	Platelet (10 ⁹ /L)		CD _{62P} (%)		CD ₆₃ (%)	
		Preoperation	Postoperation	Preoperation	Postoperation	Preoperation	Postoperation
I	19	87.4±19.2	77.3±11.3 *	1.39±0.51	1.95±0.53 *	4.02±1.33	5.62±0.90 *
II	19	87.8±18.7	81.1±13.1 *	1.34±1.08	1.62±0.37 *△	4.04±1.33	4.67±1.34 *△

* : Compared with themselves before and after surgery, *P* < 0.05; △: compared with group I, *P* < 0.05.

APACHE II value within the first 24 hours was 5.6±1.6, and the bilirubin level was 39.5±11.3. Twenty patients with simple cholecystolithiasis, 8 men and 12 women aged from 29 to 55 years (mean 43.0 years), served as controls in the same period.

Methods

The count of platelets was measured by fully automatic analyzing-machine for blood cells. The levels of CD_{62P} and CD₆₃ were measured by flow cytometry (Epics-XL type, USA) in every patient from 3 days before operation to 3 days after operation.

Special reagents, CD_{62P} and CD₆₃ monoclonal antibodies purchased from American Counter Company and used strictly according to the manufacturer's instructions.

Statistical analysis

The data were expressed as mean ± SD. The difference between each group was analyzed by Student's *t* test and the correlative significance was defined as a *P* value less than 0.05.

Results

Sex, age, APACHE II value within the first 24 hours were compared on admission and the bilirubin levels showed no significant difference between the groups I and II (*P* > 0.05).

The count of platelets groups I and II before exploration of the biliary tract was 87.6±18.7×10⁹/L, which was lower than that of the control group, while the levels of CD_{62P} and CD₆₃ (1.37%±0.84%, 4.03%±1.32% respectively) were significantly higher than those of the control group

(0.64%±0.49%, 1.77%±1.06% respectively). Three days after surgery, the count of platelets (79.2±12.4×10⁹/L) decreased markedly, and the levels of CD_{62P} and CD₆₃ were increased markedly (1.79%±0.48%, 5.15%±1.13%, respectively, *P* < 0.05).

Between groups I and II, platelet count, and CD_{62P} and CD₆₃ levels before operation were not significantly different. Three days after surgery, platelet count in group II was slightly higher than that of group I, but the difference was not statistically significant (*P* > 0.05), but the CD_{62P} and CD₆₃ levels decreased markedly (*P* < 0.05, Table).

Discussion

In this study, we found that the count of platelets in patients with hepatolithiasis was obviously lower than in patients with cholecystolithiasis, while the levels of CD_{62P} and CD₆₃ increased markedly. The results of previous studies^[1] and this study showed that these patients had had disturbance of coagulation and fibrinolysis before hospitalization. Coagulation and fibrinolysis are thought to be activated, while disturbing the physiological balance between clotting and fibrinolysis. In addition to injury of endothelial cells and platelets, all the above factors can lead to microthrombogenesis of the capillaries, tissues and organs. The causes for platelet activation in patients with hepatolithiasis are as follows: first, C₃ produced by the complement that is activated by endotoxin may activate platelet by combining it with C₃-receptor in platelet membrane;^[5] second, bile obstruction, infection and bacteria, toxemia, which may produce an inflammatory medium such as platelet activator, also can activate platelet; third, hypercholesterolemia is a

factor of platelet activation.^[6]

In this study the count of platelets decreased to some extent while the levels of CD_{62p} and CD₆₃ increased obviously. It is illustrated that surgery and trauma can further facilitate platelet injury and activation. Comparison of groups I and II showed the count of platelets decreased insignificantly in contrast to the levels of CD_{62p} and CD₆₃. These findings demonstrated that patients treated with aprotinin could not prevent the decrease of the count of platelets after operation but have some effect on the process of decrease. It is shown that aprotinin may prevent platelet from activating, protect platelet adhesiveness and aggregation, and improve coagulation function after operation.

Aprotinin inhibiting platelet activation may have the following mechanisms:^[7-10] (1) aprotinin could prevent platelet activation by protecting glycoprotein Ib of platelet adhesion receptor; (2) the activity of platelet activator can be lowered by inhibiting the complement system activation, thus indirectly protecting platelet function; (3) the protective effect of aprotinin is related to the inhibition of neutrophil activation since the activation between platelet and leucocyte can stimulate platelet activation; and (4) aprotinin can inhibit the release of CD_{62p} and CD₆₃ by stabilizing platelet membrane.

Bile obstruction can be relieved surgically or by use of antibiotics, but prevention and treatment is the key to increasing the rates of survival and prognosis. Use of aprotinin in hepatolithiasis patients one hour before surgery can stabilize the status of coagulation, inhibit the activity of fibrinolysis, protect platelet function, and decrease the occurrence of complications caused by coagulation disorder.

Competing interest

No benefits in any form have been received or will be received from a commercial party related directly or indirect-

ly to the subject of this article.

References

- 1 Feng WM, Chen QQ, Yang Q, et al. The value of D-dimer in patients with obstructive jaundice. *Chin J Hepatobiliary Surg* 2001;7:86.
- 2 Wang YS, Yang J. Clinical researches on changes in amount and aggregation of platelets in patients with hepatolithiasis. *Chin J Hepatobiliary Surg* 2000;6:190-191.
- 3 Kyriss T, Wurst H, Friedel G, et al. Reduced blood loss by aprotinin in thoracic surgical operations associated with high risk of bleeding. A placebo-controlled, randomized phase IV study. *Eur J Cardiothorac Surg* 2001;20:38-41.
- 4 Nomination and diagnosing criterion of hepatolithiasis diseases. *Chin J Surg* 1983;6:372-373.
- 5 Sakaguchi S, Kanda N, Hsu CC, et al. Lipid peroxidation formation and membrane damage in endotoxin poisoned mice. *Microbiol Immunol* 1981;25:229-232.
- 6 Ngai KC, Yeuny CY. Additive of tumor necrosis factor and endotoxin on bilirubin cytotoxicity. *Pediatr Res* 1999;1:526-530.
- 7 Cao B, Fang SQ, Li QQ, et al. The suppressive effect of aprotinin on platelet activation during cardiopulmonary bypass. *Chin J Thorac Cardiovasc Surg* 1997;13:25-27.
- 8 Li TH, Wang ZQ, Sun JL. In vitro effects of aprotinin on human blood platelets. *Chin J Anesthesiol* 1996;16:343-345.
- 9 Huang H, Ding W, Su Z, et al. Mechanism of the preserving effect of aprotinin on platelet function and its use in cardiac surgery. *J Thorac Cardiovasc Surg* 1993;106:11-18.
- 10 Wachtfogel YT, Kucich U, Hack CE, et al. Aprotinin inhibits the contact, neutrophil, and platelet activation systems during simulated extracorporeal perfusion. *J Thorac Cardiovasc Surg* 1993;106:1-4.

Received March 27, 2003

Accepted after revision August 12, 2003