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Effect of ulinastatin combined rivaroxaban on deep vein thrombosis in major orthopedic surgery

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ABSTRACT

Objective: To explore the effect of ulinastatin (UTI) continuous infusion combined Rivaroxaban on the deep vein thrombosis in patients undergoing major orthopedic surgery. **Methods:** Forty-five patients undergoing major orthopedic surgery were randomly divided into three groups: ulinastatin continuous infusion (Uc) group, ulinastatin single injection (Us) group and control (C) group. All patients received patient-controlled intravenous analgesia (PCIA) after operation, and took Rivaroxaban 10 mg orally 12 hours after operation. Ulinastatin (5 000 U/kg) was given intravenously to both Uc and Us groups preoperatively. Group C was given isometric normal saline, group Uc was pumped UTI continuous intravenously at the end of surgery (10 000 U/kg) to 48 hours through PCIA pump. The values of hematocrit (HCT), thrombomodulin (TM), Interleukin (IL-6), thrombin-antithrombin complex (TAT), D-Dimer (D-D) were normally tested before surgery (T1), at the end of the surgery (T2), 12 hours (T3), 24 hours (T4) and 48 hours (T5) after surgery. **Results:** Compared with T1, there was an upward tendency in TM, IL-6, TAT, and D-D after operation in group C group ($P < 0.05$). The values of them were significantly increased from nearly 24-hour after surgery in Us group ($P < 0.05$). In group Uc, there were no significant changes in these indices after operation ($P > 0.05$). **Conclusions:** During the perioperative period, ulinastatin continuous infusion combined Rivaroxaban can correct blood hypercoagulability through different approaches in patients undergoing major orthopedic surgery.

1. Introduction

Deep vein thrombosis (DVT) is the most common and serious complication after the major orthopedic surgery, which includes total hip replacement (THR), total knee replacement (TKR) and hip fractures surgery (HFS). Despite active anticoagulant therapy is given after the surgery, the fatality rate of the DVT can reached 3.8%, and the fatality rate of the pulmonary thromboembolism (PTE) can reached 38.9%[1].

Although compared to traditional anticoagulants, the new oral anticoagulant- rivaroxaban has advantages of safe dose rang and noninvasive administration, but the single mode of

anticoagulation and unpredictable blood drug concentration is limiting its application. Ulinastatin, an acid glycoprotein which contained in the fresh urine of healthy humans, is well known to inhibit the activity of inflammation. The recent researches show that UTI could inhibit coagulation and fibrolysis by inhibiting the leukocyte activity. Therefore, the combination therapy of rivaroxaban and UTI perhaps advance the development of anticoagulant regimen.

2. Materials and methods

2.1. Participants and inclusion criteria

After local ethics committee approval and written informed consent, 45 ASA I or III patients, aged 40–90 yr, scheduled for elective total hip replacement or hip fracture surgery, were enrolled in this study. Patients with such coexisting diseases or conditions as preoperative bleeding, clotting disorders, an abnormal coagulation test, thrombocytopenia,

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and severe renal, hepatic or heart disease were excluded. Aspirin and other antiplatelet agents were discontinued 7 days before the scheduled procedure.

Patients were randomly divided into three groups: ulinastatin continuous infusion (Uc) group, ulinastatin single injection (Us) group and control (C) group.

2.2. Treatments

After enter the operating room and before the anesthesia induction, Uc and Us group were premedicated with UTI 5 000 U/kg[2], which was dissolved in normal saline 100 mL, Group C was given isometric normal saline. Spinal anesthesia was performed at the L₂-L₃ interspace as the patient in lateral position. All patients received a patient-controlled intravenous analgesia (PCIA) for 48 h postoperatively. The PCIA setting was as follows: background infusion fentanyl 20 μg/kg and ondansetron 8 mg, which was dissolved in NS to 100 mL, at a rate of 2 mL/h, bolus dose 0.5 mL, lockout interval 15 min. The patients in Uc group were received UTI continuous infusion of 10 000 U/kg to 48 hours with the help of infusion pump. If visual analog scale (VAS) >3 in spite of treatment, a second dose of analgesic (fentanyl 0.05 mg) was given.

All the operations were conducted by the same surgery and anesthesia team. All the patients took rivaroxaban 10 mg orally 12 hours after the operation. Tests would be discontinued if the tendency of bleeding occurred.

2.3. Clinical evaluation and laboratory examination

The operation time, blood loss and the infusion were recorded.

Blood sample were obtained for analysis of Hematocrit (HCT), thrombomodulin (TM), Interleukin-6 (IL-6), thrombin-antithrombin complex (TAT), D-Dimer (D-D) levels at 5 intervals: before surgery (T1), at the end of the surgery (T2), 12 hours (T3), 24 hours (T4), 48 hours (T5) after the surgery. HCT were detected with hematology analyzer (SysmexXE-2100). Serum IL-6, TAT, D-D, TM concentrations were assayed by ELISA (R&D company, America).

2.4. Statistical analysis

Statistical analysis was performed with the χ² test of the sex and type of surgery. One-way factorial analysis of variance (ANOVA) was used in analysis of other demographic data. P value <0.05 was considered as statistically significant difference.

3. Results

3.1. General observation

The general data compared with treatment group and control group as follows (Table 1), and there was no significantly differences (P>0.05) in sex, age, type of surgery, etc. No significant change of the HCT was found between three groups in each time point, which excluded the effect of blood dilution on the experimental results (Table 2).

Table 1 Comparison of background variables.

Issues	Uc	Us	C
Sex (M/F)	8/7	9/6	8/7
Age	68.27±13.26	69.80±11.97	66.73±13.89
BMI	22.31±1.55	21.92±1.47	21.51±1.60
Type (THR/HFS)	11/4	12/3	11/4
OR time(min)	79.00±23.69	80.33±28.31	80.33±26.35
Blood loss(mL)	124.00±61.04	150.67±78.96	142.33±72.97
Infusion(mL)	973.33±187.91	923.33±273.77	1 093.33±405.26

Table 2 Changes of hematocrit in three groups (n=15, %, mean±sd).

Indexes		Uc	Us	C
HCT	T1	38.31±3.93	36.34±4.75	38.36±3.65
	T2	33.75±4.38	31.63±4.32	33.37±3.30
	T3	34.3±5.66	33.5±5.77	33.30±3.17
	T4	32.42±5.93	31.57±3.36	32.25±3.27
	T5	30.91±5.50	29.76±3.24	30.85±2.52

3.2. TM scores

As shown in Table 3, TM score was gradually increased in C group till 48 hours after the surgery (P<0.05). However, it was declined in Uc group (P<0.05). The TM score was significantly increase 24 hours after single injection (P<0.05).

3.3. IL-6 scores

As shown in Table 4, after the surgery, both ulinastatin single injection group and ulinastatin continuous infusion group showed no conspicuous change in IL-6 (P>0.05). However, the level of IL-6 in control group was significantly increased (P<0.05). But 24 hours after single injection, the IL-6 score in Uc group were significant increase as in C group (P<0.05). No outstanding change was observed in Uc group till 48 hours after the surgery (P>0.05).

Table 3 Changes of TM in three groups (n=15, μg/L, mean±sd).

Groups	T1	T2	T3	T4	T5
Uc	47.36±15.07	40.42±9.37●▲	40.60±8.80●▲	34.82±6.19●▲	43.98±13.14●▲
Us	41.93±11.42	42.35±8.45●	44.18±8.98●	50.50±14.33▲	57.00±9.52▲
C	44.33±8.00	51.62±7.66▲	53.86±7.47▲	54.72±8.47▲	57.10±8.31▲

●Compared with group C, P<0.05; ▲ Compared with T1, P<0.05.

3.4. TAT scores

From Table 5, it showed that TAT exhibited roughly increasing tendency after the surgery for 48 hours in C group ($P<0.05$). However, Uc group and Us group showed no significant changes ($P>0.05$).

3.5. D–D scores

As shown in Table 6, 12 hours after the surgery, no remarkable changes was found in D–D serous concentration of both Ulinastatin single injection group and ulinastatin continuous infusion group ($P>0.05$). Nevertheless, the control group showed very significant increase in D–D level ($P<0.05$). Also after 12 hours, D–D concentration in Uc group was significantly lower than that of C group ($P<0.05$). Although the D–D level in Us group was higher compared with C group ($P<0.05$), and no significant difference was observed ($P>0.05$).

3.6. Comparison of clinical adverse reaction

Both three groups showed no hemorrhagic tendency.

4. Discussion

Patients with trauma and orthopedic surgery can cause local tissue swelling, inflammation infiltration, endothelial cells damage at the same time, which promote the blood platelet aggregation and adhesion. In perioperative period the restrict mobility, the using of bone cement and tourniquet in the surgery, and general anesthesia, obesity, etc, are all risk factors for DVT occurred^[3,4]. Once DVT was

diagnosed from imageology, it is hard to be cured. So the early prophylaxis and discovery of thrombophilia is the key to prevent DVT.

Previous studies showed that low molecular heparin injected subcutaneously could be used to prevent and cure DVT effectively. Meanwhile, the potent anticoagulant effect could cause bleeding tendency, and it is one typical side effect. Moreover, the way of invasive administration reduces the clinical compliance of patients. Recently, it has been reported that one kind of new oral anticoagulant rivaroxaban is replacing its anticoagulation status gradually as a result of simple and noninvasive application. Rivaroxaban plays a role in restraining the process of blood coagulation as a kind of highly selective Fxa inhibitor, and it has been widely used in clinic currently. But there are some scruples with its using process, for example, the drug lacks of individualized treatment plan^[5], and it still induces bleeding tendency^[6].

Ulinastatin, an acid glycoprotein which was contained in the fresh urine of healthy humans, is well known to inhibit the activity of inflammation. The recent researches showed that UTI could inhibit coagulation and fibrolysis by inhibiting the leukocyte activity^[7]. This function is very important in regulating coagulation system. So it is hypothesized that Ulinastatin may take effect on preventing DVT.

Because of this, during the treatment, we especially emphasize on changing patients' hypercoagulable state, not just aim to theirs' single anticoagulation function. The treatment target is to improve patients' medication safety by assisting in regulating the coagulation system.

The results in this study suggest that the combination of UTI and rivaroxaban can significantly decrease the incidence of postoperative DVT as detection by laboratory examination. Even in those patients with the highest tendency to develop

Table 4

Changes of IL-6 in three groups ($n=15$, ng/L, mean \pm sd)

Groups	T1	T2	T3	T4	T5
Uc	478.82 \pm 124.01	468.86 \pm 112.99	455.24 \pm 84.37 [●]	493.31 \pm 66.12 [●]	499.26 \pm 107.78 [●]
Us	538.09 \pm 162.15	537.71 \pm 122.96	585.67 \pm 87.92	641.45 \pm 111.81 [▲]	635.61 \pm 101.65 [▲]
C	471.83 \pm 69.12	528.50 \pm 114.09 [▲]	546.35 \pm 102.79 [▲]	574.47 \pm 93.21 [▲]	601.32 \pm 150.73 [▲]

● Compared with group C, $P<0.05$; ▲ Compared with T1, $P<0.05$.

Table 5

Changes of TAT in three groups ($n=15$, ng/L, mean \pm sd).

Groups	T1	T2	T3	T4	T5
Uc	640.95 \pm 120.73	620.34 \pm 73.38	577.86 \pm 94.73 [●]	632.43 \pm 97.47 [●]	644.06 \pm 120.21 [●]
Us	636.99 \pm 87.23	639.55 \pm 61.93	617.04 \pm 49.63	675.41 \pm 109.81	710.31 \pm 113.13
C	605.91 \pm 67.11	693.63 \pm 121.11 [▲]	664.35 \pm 86.78 [▲]	752.56 \pm 100.95 [▲]	736.97 \pm 71.15 [▲]

● Compared with group C, $P<0.05$; ▲ Compared with T1, $P<0.05$.

Table 6

Changes of D–D in three groups ($n=15$, μ g/L, mean \pm sd).

Groups	T1	T2	T3	T4	T5
Uc	763.91 \pm 121.34	763.30 \pm 176.93	729.28 \pm 151.75 [●]	656.90 \pm 176.05 [●]	705.69 \pm 193.90 [●]
Us	760.32 \pm 112.21	799.80 \pm 121.28	877.62 \pm 152.11 [▲]	1 008.53 \pm 250.67 [▲]	1 141.09 \pm 218.54 [▲]
C	764.37 \pm 129.71	881.75 \pm 132.10 [▲]	870.60 \pm 97.17 [▲]	938.17 \pm 110.67 [▲]	1 065.70 \pm 145.44 [▲]

● Compared with group C, $P<0.05$; ▲ Compared with T1, $P<0.05$.

thrombosis, such as elderly patients having major orthopedic operations. There was no complication occurrence after administrating the drugs.

Previously, Virchow stated that venous thrombosis was caused by changes in one or more of three important factors— blood flow, venous endothelium, and the clotting mechanism^[8]. And “Virchow’s triad” is of prime importance in the prevention of thrombosis.

In our experiment, though rivaroxaban was routinely given for anticoagulation therapy postoperatively in C group, a rising curve of L-6, TAT, D-D and TM occurred and lasted from the completion of the surgery to 48 hours after the surgery. UTI can inhibit the rise of these parameters. This finding was consistent with those of several precious reports. Sympathetic excitation and further vasoconstriction resulted from Trauma and surgery, the use of alkaline anesthetics, the ischemia–reperfusion injury caused by tourniquet are all considered to play a part in the endothelial cells disorder. TM, a type of membrane receptor locating on the surface of vascular endothelial cells, would be released into the blood when the cell was damaged. This experiment suggests that UTI has a role on protecting endothelial cell and contrarily less role effect on rivaroxaban; in single injection group, endothelial cells are observed to start damaging 24 hours after the surgery. The primary mechanisms of the UTI on the protection of endothelial cells may not only depending on inhibiting the activity of elastase secreted by neutrophils, but also directly inhibit the secretion of the enzyme itself^[9].

TAT is induced by thrombin and has a rapid turnover. Thus, it is a sensitive parameter of the latent activation of the clotting pathway. The rise of TAT suggests that the thrombin is generating and the antithrombin is being depleted continuously. So TAT is the direct evidence of the activation of promoting coagulation and the consumption of inhibitor. As we all know, there are reciprocating relations between thrombosis and inflammation. In our study, the level of IL-6 was significantly increased after the surgery till 2 days in C group, but the same changes of IL-6 was not observed in UTI group. It was demonstrated that the enhancement of IL-6 had been inhibited by UTI. Similarly, TAT could increase with the surgery only using rivaroxaban, and UTI could inhibit this augmentation. The mechanism of UTI on coagulation system is mainly through the intrinsic coagulation pathway, the Kunitz structural domain of UTI can influence the coagulation factors, especially the activity of factor X^[10].

D-dimer is a kind of small protein fragment presented in the blood after the blood clot degraded by fibrinolysis. Hence, its increase in blood indicates the progression of fibrinolysis. 10 000 U/kg UTI can reduce the D-D in patients who undergoing hip joint replacement^[11]. The results in our research showed that 5×10^4 U/kg UTI has effect on fibrinolytic system in human, while the effect of single injection can only last less than 12 hours on D-dimer. However, Rivaroxaban has no such an effect.

Our experiment excluded the influence of the blood dilution by testing HCT. So there was no significant difference in the blood flow in and between three groups.

Although different studies were designed to investigate

the low molecular fragment originated from active pharmacological reaction still have effect, this suggests that in theory, if single dose of UTI was administered at the beginning of the surgery, the effect would maintain for a long time^[12]. In this experiment, the results show that the effect of UTI just lasted for 12–24 hours if single injection, while all the investigated indices were kept stable in condition of continuous injection of UTI. It means that UTI combined with rivaroxaban is a satisfactory method of preventing the DVT.

In this study, we explored the effect of combined UTI with rivaroxaban on preventing DVT. The conclusion was drawn that the combination of UTI and Rivaroxaban was more effective than using rivaroxaban only, and it is also important for UTI to be intravenously injected continuously. However, this study is lack of large-scale and multicenter clinical verification. Moreover, we just observed the changes of indices in 48 hours after the surgery. Thus, further studies are still needed to search for a reasonable drug administration time and the suitable dosage.

Conflict of interest statement

We declare that we have no conflict of interest.

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