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ORIGINAL ARTICLE

A seesaw-type device to measure the coagulation time of blood: application to assessment of anticoagulant effect of direct oral anticoagulants (DOACs) in patients

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Abstract A simple seesaw-type method using a sphere-shaped capsule as a blood testing container was developed, which is able to determine the time of onset of coagulation (coagulation time) of blood. The method is based on when a capsule containing a blood sample ceases rolling down a slope in a series of periodic, repetitive runs. From the elapsed time that the capsule stops rolling, the coagulation time can be determined. This method enables concurrent measurement of coagulation times for multiple blood samples. Optimal experimental conditions including the angle of slope and amount of blood were determined using cattle blood. The mean value of coagulation time for normal human donors was 19.6 ± 3.5 min, while that for patients taking direct oral anticoagulants (DOACs) was quite large. Addition of a coagulation accelerant (APTT reagent) to blood obtained from a normal donor and a DOAC-treated patient suffering from atrial fibrillation brought about dramatic shortening of the coagulation time. In addition, coagulation of the patient was slightly delayed compared with that of the normal donor. In this paper, it is shown that the present technique may be useful for assessing the anticoagulant effect of DOACs in patients as well as for determining the coagulation time of blood.

Keywords seesaw-type method, sphere-shaped polypropylene capsule, coagulation of blood, anticoagulant effect, APTT

1. Introduction

The clinical assessment of hypercoagulability and hypocoagulability of blood would be very important for knowing the coagulation functions in patients, such as the risk of thrombogenesis, risk of bleeding through surgery and treatment effect with anticoagulants [1–4]. To evaluate blood coagulation reactions in a clinical test, the activated partial thromboplastin time (APTT) and the prothrombin time (PT) measurements are commonly carried out [5, 6], with plasma centrifuged from whole blood.

Although the coagulation cascade of blood is very complicated, the coagulation takes place by fibrinogen-fibrin transformation through the catalytic action of thrombin and the subsequent polymerization and network formation of fibrin, that is gelation [7]. During coagulation of blood, the viscosity and rigidity of blood increase with the formation of a fibrin gel. Therefore, rheological measurements make it possible to investigate the mechanism of blood coagulation and the properties of fibrin gels. Most *in vitro* investigations have been carried out by use of forced oscillation-type rheometers such as the Weissenberg rheogoniometer [8], the Thrombelastograph (TEG) [9], and the Viscoelastorecorder [10]. In recent laboratory tests, ROTEM [11] and Sonoclot [12] that are also forced-oscillation type rheometers have been employed. ROTEM is an improved-type of TEG.

In our earlier studies, the mechanism of blood coagulation was examined using a damped oscillation rheometer [13, 14]. From rheological and biochemical analysis using blood obtained from normal controls and from patients, it was suggested that this device could assess the risk of venous thrombus formation as well as coagulation time [15, 16]. In clinical situation, a convenient device has been desired to assess the procoagulant activity of blood. In particular, there is need for a simple system capable of measuring the coagulation times of several blood samples at once.

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Therefore, we tried to develop a very simple method to ascertain the coagulation times of a set of blood samples, where sphere-shaped polypropylene capsules containing blood are put on parallel-arrayed V-shaped gutters. The gutters move in a seesaw fashion, and coagulation time of blood is determined from the time that the capsule stops rolling down the gutter slope. We show some fundamental data and discuss the potential of the present technique for assessing the treatment effect with anticoagulants (DOACs) as well as for determining the coagulation time of blood.

2. Materials

To determine optimal experimental conditions, cattle blood was obtained from Japan Lamb Hokkaido Farm (Hokkaido, Japan), in accordance with the farm's guidelines for the care and use of animals. Blood was taken using 3.8% tri-sodium citrate solution as an anticoagulant (9:1 v/v).

Human blood was obtained from adult volunteers in accordance with the guideline of each clinic, and donors gave their informed consent. They include normal donors and outpatients treated with DOACs (factor Xa inhibitor) including rivaroxaban (Xarelto®, Bayer Yakuhin Ltd) and apixaban (Eliquis®, Bristol-Myers Squibb Co). Blood was collected by venipuncture and anticoagulated with 1/10 vol of 3.8% tri-sodium citrate solution. Blood was immediately transferred into a polypropylene tube. Platelet–poor plasma (PPP) was prepared by centrifuging whole blood at 1,600 g for 15 min. Platelet-free plasma (PFP) was prepared by centrifuging PPP at 16,000 g for 15 min at 4°C.

Activated partial thrombin time (APTT) reagent (APTT-SLA) that accelerates the intrinsic coagulation reactions was obtained from Sysmex Corporation (Kobe, Japan).

3. Seesaw-type device

Figure 1 shows a schematic picture of hollow capsule, which was obtained from Japan Polypro Corporation (Tokyo, Japan). A transparent sphere-shaped capsule (MA3H; diameter, 25 mm; wall thickness, 1.5 mm; inner volume, 5.6 ml) is made of polypropylene. The capsule can be separated into hemispheres for sample loading. After blood sample is put in one part of hemispheres, the two parts are rejoined. A small pore (diameter, 1 mm) was drilled in a hemisphere, which is effective to prevent a leakage of blood from the joint part of hemispheres, owing to the slight increase in pressure inside the capsule. Prior to measurements, the inner surface of capsule was thoroughly washed with isopropyl alcohol and then distilled water, and dried.

A newly designed seesaw-type apparatus consists of a V-shaped gutter (angle 90°) made of stainless plate (Fig. 2). The present version has five parallel-arrayed gutters so that a total of five samples can be run concurrently. The gutter length is 82 mm in early experiments and 36 mm after improving the device. A reflection-type optical sensor (EE-

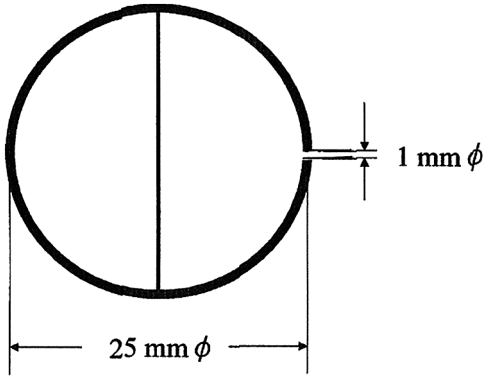

Fig. 1 A schematic picture of polypropylene capsule and its dimension.

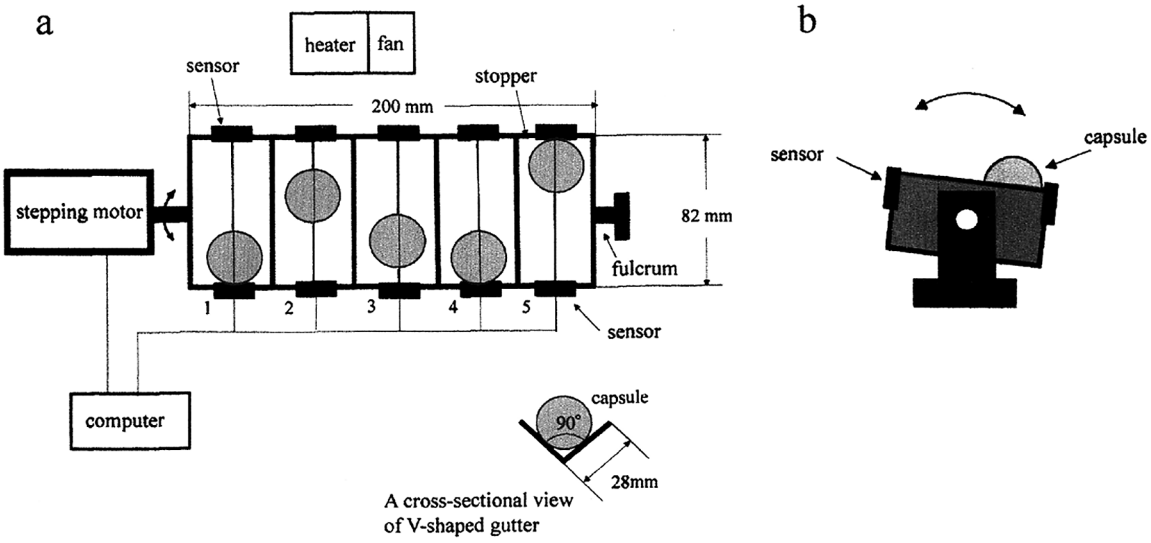

Fig. 2 A schematic diagram of the measuring system consisting of five parallel-arrayed V-shaped gutters. a, a view from above; b, a view from right side.

SY671, Omron Corporation, Kyoto, Japan) to detect a capsule is attached to stoppers at both gutter ends. The gutter arrays joined to each other move in a seesaw fashion about the fulcrum, which are driven by a stepping motor (KH4234-B90101, Nidec Servo Corporation, Gunma, Japan). The slope of gutter arrays could be varied from 3 to 10° and the interval of rocking from 10 to 120 s. A capsule containing blood sample is placed at one end of test section to begin measurement. The gutters are then moved and rolling of capsules to the other end of the test section is observed. Rocking is repeated at pre-determined interval for those samples that continue to roll. Coagulation time (Ti) of blood is determined from the time that the sensor at the lower position during seesaw motion becomes undetectable the capsule. The apparatus is totally controlled by a microcomputer and automatic data collection can be made. Temperature is kept at 37°C by circulating warm air.

4. Results

4.1 Effects of gradient of slope and amount of blood on capsule rolling

Optimal experimental conditions were determined using cattle blood. Whether a capsule containing uncoagulated blood rolls down the gutter slope was examined under the different gradients of slope and different amounts of blood (Table 1). Magnitudes of slope above 5° except for sample amounts of 0.3 and 0.5 ml provided sufficient torque to ensure movement of the capsule. At 0.3 and 0.5 ml, the capsule rolled down slowly.

Whether a capsule containing coagulated cattle blood rolls down is summarized in Table 2. These measurements were carried out after the completion of coagulation. Except for a sample amount of 0.3 ml and magnitudes of slope above 6°, the capsule did not roll down although there was a case in which the capsule stopped rolling in the middle of gutter slope or rolled down slowly.

From these results, if the magnitude of slope is 6° and sample amounts of 0.5, 1.0 and 1.5 ml, the coagulation time could be found by this method. When the slope of seesaw changed in the opposite direction, the time required for the capsule to reach the lower end stopper was less than 1 s at the gutter length of 82 mm. Therefore, the coagulation time was determined from when the sensor did not detect the capsule within 3 s.

The effects of rocking interval and gutter length on the coagulation time of blood were examined using normal human blood at rocking intervals of 10 and 60 s and gutter lengths of 36 and 82 mm. The rocking interval and gutter length had little effect on the coagulation time (data not shown). Thus, measurements were carried out at the rocking interval of 60 s and the gutter length of 36 or 82 mm for blood without added APTT, and 10 s and 36 mm for with added APTT.

4.2 Reproducibility of measurements

Table 3 shows some typical examples of the value of Ti, which were measured using blood obtained from 6 normal human donors. After adding $CaCl_2$ solution to blood obtained from a donor, the blood sample was transferred into 5 capsules, and the measurements were carried out at once. The mean value of Ti for 5 capsules was from 12.6 to 32.0 min in 6 donors, and the difference between the maximum and minimum values of Ti (ΔTi) from 0 to 6.3 min.

4.3 Coagulation of human blood

The values of Ti for blood from normal donors and patients taking DOACs (rivaroxaban or apixaban) are summarized in Table 4. In all donors, coagulation of PFP did not occur within the experimental time period (in excess of 60 min). Conversely, coagulation of whole blood did occur, and the mean value of Ti for normal donors was 19.6 ± 3.5 min. On the other hand, the values of Ti for patients taking DOACs were quite large. In particular, the coagulation of blood for patients taking apixaban was quite delayed.

Table 1 Determination of optimal experimental conditions by a series of roll tests using uncoagulated cattle blood, varying the gradient of slope and amount of blood

Amount of blood (ml)	Gradient of slope (°)					
	3	4	5	6	7	8
0.3	–	–	+(s)	+	+	+
0.5	–	–	+(s)	+	+	+
1.0	–	–	+	+	+	+
1.5	–	+(s)	+	+	+	+

–, capsule did not roll down ; +, capsule rolled down; +(s), capsule rolled down slowly.

Table 2 Determination of optimal experimental conditions by a series of roll tests using coagulated cattle blood

Amount of blood (ml)	Gradient of slope (°)					
	3	4	5	6	7	8
0.3	–	–	–	+	+	+
0.5	–	–	–	–	(–)	(–)
1.0	–	–	–	–	–	–
1.5	–	–	–	–	–	–

After adding 170 µl of 0.25 M $CaCl_2$ solution to 2.0 ml of blood, a capsule containing the blood sample of pre-determined amount was placed at stasis until coagulation was completed (about 30 min), and the capsule was put on the gutter. –, capsule did not roll down; +, capsule rolled down; (–), capsule stopped on the way of gutter slope or rolled down slowly.

Table 3 Comparison of the values of Ti for blood from 6 normal human subjects, measured using 5 gutters

Donor	Ti, (min)						
	Gutter number					Mean Ti	ΔTi
	1	2	3	4	5		
A	12.6	12.6	12.6	12.6	12.6	12.6	0
B	16.2	16.2	16.2	15.2	15.2	15.8	1.0
C	21.0	19.8	19.8	18.5	19.8	19.8	2.5
D	20.4	20.4	20.4	19.3	20.4	20.2	1.1
E	22.4	24.6	23.5	21.4	20.3	22.4	4.3
F	32.3	31.0	34.8	33.5	28.5	32.0	6.3

Prior to measurements, blood samples were incubated for 5 min at 37°C. After adding 425 µl of 0.25 M $CaCl_2$ solution to 5.0 ml of blood from a donor, the blood sample was transferred into 5 capsules, and the measurements were carried out at once. Time zero corresponds to addition of $CaCl_2$ solution to the blood sample. The gradient of gutter slope was 6°, the gutter length 82 mm and the rocking interval 60 s. ΔTi means the difference between the maximum and minimum values of Ti.

Table 4 The values of Ti for blood from normal donors and patients taking DOACs

Blood sample	Ti (min)
Platelet-free plasma (PFP)	>60
Whole blood of normal donors	19.6 ± 3.5 (n = 22)
Whole blood of patients taking rivaroxaban (Xarelto 15 mg)	36.1 ± 4.8 (n = 6)
Whole blood of patients taking apixaban (Eliquis 5 mg)	60.0 ± 9.1 (n = 8)

After adding 255 µl of $CaCl_2$ solution to 3.0 ml of blood, the blood sample was transferred into 3 capsules, and measurements were carried out at once. The value of Ti in one experimental run (n = 1) is the mean value of 3 capsules. In patients taking DOACs, measurements were carried out at 2–3 h after taking DOAC. The gutter length was 82 mm for normal donors and 36 mm for patients, and the rocking interval 60 s.

Table 5 Effect of the amount of APTT solution added to blood on the value of Ti

Amount of APTT (µl)/ 0.5 ml of blood	Ti (min)	
	Normal donor	Patient taking apixaban
0	19.4 ± 2.7	59.7 ± 11.2
30	2.31 ± 0.15	3.63 ± 0.26
40	1.84 ± 0.14	3.32 ± 0.20
50	1.61 ± 0.12	2.86 ± 0.19
60	1.24 ± 0.01	2.17 ± 0.00

Blood was obtained from a normal donor and an apixaban (5 mg)-treated patient suffering from atrial fibrillation. Prior to measurement, the blood sample was incubated for 5min at 37°C. Then APTT solution of a pre-determined amount was added to the blood sample, and incubated for 1 min. After adding 85 µl of $CaCl_2$ solution to 1.0 ml of blood sample, the sample was transferred into 2 capsules, and the measurements were carried out at once. The value of Ti in one experimental run is the mean value of 2 capsules. 5 or 6 experiments were repeated at each APTT amount. The gutter length was 36 mm and the rocking interval 10 s.

Table 6 Effects of elapsed time after taking apixaban on the values of Ti for blood with and without added APTT solution

Blood sample	Ti (min)
at 2–3 h after taking	
without APTT	61.9 ± 10.7 (n = 9)
with APTT	2.94 ± 0.16 (n = 9)
at 6–7 h after taking	
without APTT	42.0 ± 9.4 (n = 8)
with APTT	2.65 ± 0.08 (n = 8)

Blood was obtained from the same patient as explained in Table 5. The amount of APTT added was fixed at 50 µl per 0.5 ml of blood. The gutter length was 36 mm, and the rocking interval 10 s for blood with APTT and 60 s for without APTT.

We tried to measure the value of Ti in a short time, where APTT reagent, an accelerant of blood coagulation, was added to blood samples. Table 5 shows the relationships between the amount of APTT solution added and the values of Ti for blood from a normal donor and an apixaban-treated patient with atrial fibrillation. In both blood samples, the coagulation of blood occurred rapidly. In addition, the values of Ti for the patient were slightly larger than for the normal donor at any amount of APTT reagent. In the following experiments, APTT amount of 50 µl was used for the experimental convenience.

The effect of elapsed time after taking apixaban on the value of Ti was examined (Table 6). The mean value of Ti measured at 6–7 h after taking was shortened compared to that at 2–3 h. Addition of APTT reagent to the blood samples brought about a significant decrease in the value of Ti, at both elapsed times.

Figure 3 shows the relationship between the values of Ti for blood with and without added APTT reagent, of which blood was obtained from normal donors and a patient taking apixaban. Good correlation was observed between them.

5. Discussion

Whether the capsule containing blood sample rolls down the gutter slope would depend on the gradient of slope, the amount of blood sample, and the viscosity or viscoelasticity of blood sample. At a certain fixed gradient of slope and amount of blood, for example 6 ° and 0.5 or 1.0 ml, a capsule containing uncoagulated blood always rolls down the slope because movement of blood on the inner surface of capsule produces a steady rotational force of capsule. On

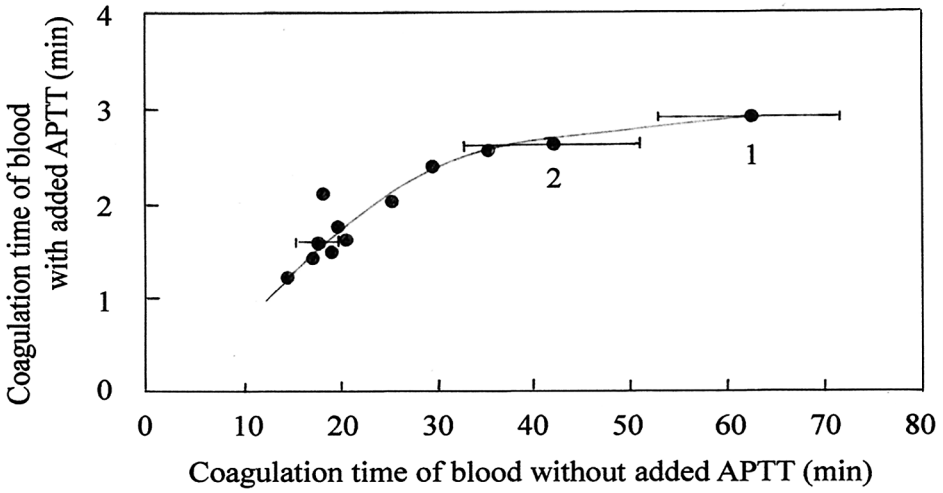

Fig. 3 The coagulation time (Ti) of blood samples with added APTT reagent plotted against that without APTT. The amount of APTT was fixed at 50 μl per 0.5 ml of blood. 1, an apixaban-treated patient suffering from atrial fibrillation, measured at 2–3 h after taking; 2, at 6–7 h; the others, normal donors. The experimental procedures were the same as those explained in Table 4 for blood samples without added APTT and in Table 5 for with added APTT. The points (1 and 2) represent the mean ± SD from 6 independent experiments, and the others represent the mean values of 2 or 3 capsules, measured at once.

the other hand, once the coagulation of blood occurs, the capsule stops rolling because an initial slight rotation of the capsule causes a rotational force in an opposite direction. Then the capsule stops rolling at a balanced position where a perpendicular line toward the inclined plane from the center of gravity of the capsule and the contact point of the capsule with the inclined plane would coincide.

In Tables 1 and 2, although the reasons for intermediate behaviors of capsules such as slow rolling or stopping on the way of gutter are not clear what is going on in blood, one of the reasons may be due to the difference in a subtle balance between a steady rotational force and a rotational force in an opposite direction.

In Table 3, the values of Ti of blood vary considerably in individuals, and the values of ΔTi in donors E and F are larger than those in donors A, B and D. It seems that the delay in coagulation causes a large difference in the value of Ti in different capsules. The reactions to produce thrombin are quite slow in the present devise because the enzymatic reactions in blood coagulation are influenced by many factors including the coagulant ability of erythrocytes described below, the concentration of coagulation factors, age, hematocrit and propagation reactions on blood cell membranes [17–19]. Therefore, a slight difference in the rates of sequential activation of coagulation factors would bring about the large difference in coagulation time.

Coagulation of PPP did not occur within the experimental time period, whereas coagulation of whole blood occurred (Table 4). This suggests that the intrinsic and extrinsic coagulation pathways are not initiated in a polypropylene capsule, that is, the polypropylene surface used offers an inert surface against blood coagulation [17]. We have found that coagulation of whole blood is initiated through the activation of factor IX by an enzyme (erythroelastase-IX) that exists in normal human erythrocyte membrane [19]. The coagulation ability of factor IX activated by the enzyme was estimated to be approximately 1/10 as high as that of factor IX activated by factor XIa [19]. Therefore, the coagulation of blood in the polypropylene capsule would occur slowly (Table 3).

Assessment of anticoagulant effects of DOACs would be helpful for the patients with a risk of bleeding or suspected thromboembolism [20, 21]. APTT and PT measurements are used to assess the anticoagulant efficacy in the clinical situations [22, 23], but the assays are time-consuming because the plasma sample centrifuged from whole blood is used. Rheological methods are considered to be a useful method for assessment of the anticoagulant effect of DOACs [24, 25] because measurements can be carried out using whole blood, and the rapid acquisition of data is expected. In addition, the measurement using whole blood would give useful information based on interactions between blood cells and components in plasma. A disadvantage in the present method, which is time-consuming to determine the value of Ti, could solve by adding APTT reagent to blood (Tables 5 and 6). It is reported that the time required for apixaban to reach the maximum concentration in circulating blood is 3–4 h and the elimination half-time 8–15 h [26]. In Table 6, the shortening of coagulation time with the elapsed time after taking apixaban seems to represent well the efficacy of medicine.

In Fig. 3, when the coagulation of blood without added APTT reagent occurs within 30 min, the values of Ti for blood with added APTT decreased almost linearly with the decrease in those without added APTT reagent. Previously, we had reported that the coagulation time of blood from a patient with lower leg deep vein thrombosis was approximately 12 min [16]. Therefore, it is expected from Fig. 3

that if APTT reagent is added to blood of patients with a high level of hypercoagulability, the coagulation may occur rapidly, i.e. within 1 min. This suggests that the technique used in this work may be useful for a rapid risk assessment for venous thrombogenesis although further investigation needs to be continued in many patients.

6. Conclusion

The present device would be a new method to measure the coagulation time of blood. It is expected that the present technique would be useful for examining the coagulability of blood and for assessing the anticoagulant effect in patients taking an anticoagulant drug such as DOACs. Further study focusing on more physiological aspects must still be done before this method can be used in clinical practice.

Conflict of interest The authors have no conflicts of interest to declare.

Patent The present method is protected by JAPAN PATENT (PATENT NUMBER 6239603).

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References

1. Chee YL, Greaves M. Role of coagulation testing in predicting bleeding risk. Hematol J. 2003; 4(6): 373–8.
2. Naess IA, Christiansen SC, Romundstad P, Cannegieter SC, Rosendaal FR, Hammerstrom J. Incidence and mortality of venous thrombosis: a population-based study. J Thromb Haemost. 2007; 5(4): 692–9.
3. Rhee AJ, Kahn RA. Laboratory point-of-care monitoring in the operating room. Curr Opin Anaesthesiol. 2010; 23(6): 741–8.
4. Douxfils J, Ageno W, Samama C-M, Lessire S, Cate HT, Verhamme P, et al. Laboratory testing in patients treated with direct oral anticoagulants: a practical guide for clinicians. J Thromb Haemost. 2018; 16(2): 209–19.
5. Korte W, Clarke S, Lefkowitz JB. Short activated partial thromboplastin times are related to increased thrombin generation and an increased risk for thromboembolism. Am J Clin Pathol. 2000; 113(1): 123–7.
6. Bajaj SP, Joist JH. New insights into how blood clots: implications for the use of APTT and PT as coagulation screening tests and in monitoring of anticoagulant therapy. Semin Thromb Hemost. 1999; 25(4): 407–18.
7. Kirkpatrick JP, McIntire LV, Maoke JL, Peterson DL. Dynamic rheological studies of coagulation and fibrinolysis. J Rheology. 1979; 23(6): 769–87.
8. McIntire LV. Dynamic materials testing: biological and clinical applications in network-forming systems. Ann Rev Fluid Mechanics. 1980; 12: 159–79.
9. Hartert H. Thrombelastography: physical and physiological aspects. In: Copley AL, Stainsby G, editors. Flow properties of blood and other biological systems. Oxford, London, New York, Paris: Pergamon Press; 1960. p.186–98.
10. Kaibara M. Rheological studies on blood coagulation and network formation of fibrin. Polym Gels and Networks. 1994; 2(1): 1–28.
11. Luddington RJ. Thrombelastography/thromboelastometry. Clin Lab Haematol. 2005; 27(2): 81–90.
12. Hett DA, Walker D, Pilkington SN, Smith DC. Sonoclot analysis. Brit J Anaesth. 1995; 75(6): 771–6.
13. Kaibara M, Kawamoto Y. Rheological measurement of blood coagulation in vascular vessel model tube consisting of endothelial cell monolayer. Biorheology. 1991; 28(3–4): 263–74.
14. Kawakami S, Kaibara M, Kawamoto Y, Yamanaka K. Rheological approach to the analysis of blood coagulation in endothelial cell-coated tubes: activation of the intrinsic reaction on the erythrocyte surface. Biorheology. 1995; 32(5): 521–36.
15. Iwata H, Kaibara M. Activation of factor IX by erythrocyte membranes causes intrinsic coagulation. Blood Coagul Fibrinoly. 2002; 13(6): 489–96.
16. Kaibara M. Rheological study on coagulation of blood with special reference to the triggering mechanism of venous thrombus formation. J Biorheology. 2009; 23(1): 2–10.
17. Kaibara M, Iwata H, Ujiie H, Himeno R, Kaibara M. Rheological analyses of coagulation of blood from different individuals with special reference to procoagulant activity of erythrocytes. Blood Coagul Fibrinoly. 2005; 16(5): 355–63.
18. Ujiie H, Kawasaki M, Suzuki Y, Kaibara M. Influence of age and hematocrit on the coagulation of blood. J Biorheology. 2009; 23(2): 111–4.
19. Iwata H, Kaibara M, Dohmae N, Takio K, Himeno R, Kawakami S. Purification, identification, and characterization of elastase on erythrocyte membrane as factor IX-activating enzyme. Biochem Biophys Res Commun. 2004; 316(1): 65–70.
20. Shoeb M, Fang MC. Assessing bleeding risk in patients taking anticoagulants. J Thromb Thrombolysis. 2013; 35(3): 312–9.
21. Conway SE, Hwang AY, Ponte CD, Gums JG. Laboratory and clinical monitoring of direct acting oral anticoagulants: what clinicians need to know. Pharmacotherapy. 2017; 37(2): 236–48.
22. Harenberg J, Erdle S, Marx S, Kramer R. Determination of rivaroxaban in human plasma samples. Semin Thromb Hemost. 2012; 38(2): 178–84.
23. Lippi G, Ardissino D, Quintavalla R, Cervellin G. Urgent monitoring of direct oral anticoagulants in patients with atrial fibrillation: a tentative approach based on routine laboratory tests. J Thromb Thrombolysis. 2014; 38(2): 269–74.
24. Dais JD, Norem K, Doorneweerd DD, Thurer RL, Popovsky MA, Omert LA. Use of thromboelastography (TEG) for detection of new oral anticoagulants. Arch Pathol Lab Med. 2015; 139(5): 665–73.
25. Chojnowski K, Gorski T, Robak M, Trelinski J. Effects of rivaroxaban therapy on ROTEM coagulation parameters in patients with venous thromboembolism. Adv Clin Exp Med. 2015; 24(6): 995–1000.
26. Haas S, Bode C, Norrving B, Turpie AGG. Practical guideline for using rivaroxaban in patients with atrial fibrillation: balancing benefit and risk. Vas Health Risk Manag. 2014; 10: 101–14.