Effect of Different Oral Anticoagulant Intensities on Prothrombin Fragment 1+2 in Thai Patients with Mechanical Heart Valve Prostheses†

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Abstract

The minimal intensity of oral anticoagulant required for antithrombotic protection in patients with a mechanical heart valve is still debatable, and that of the Westerner may not be directly applied to Thai patients. Our preliminary clinical review suggested that International Normalized Ratio (INR) 2-3 might be enough but it needs further supporting evidence. Therefore, we studied the effect of different anticoagulant intensities, expressed as INR, on the *in vivo* coagulation activation by measuring prothrombin fragment 1+2 (F1+2) in 116 patients with mechanical heart valve replacements. The patients had received warfarin for not less than one month with different intensities. The mean ± S.D. of F1+2 level in 30 normal controls was 0.7±0.17 nmol/L. After excluding two outliers, the maximum linear correlation between INR and F1+2 was -0.658 ($p<0.001$) when only patients whose intensities were lower than INR3 were taken into account. Adding more data from the patients having higher intensities decreased the correlation coefficient. The patients were subsequently classified by INR values in the range INR 1.1-1.9, 2-3 and 3.1-4.2. The F1+2 in each group was 0.6±0.30, 0.28±0.13 and 0.24±0.13 nmol/L respectively. The F1+2 in the first group did not differ from normal ($p=0.119$) but was higher than the others ($p=0.000$). The latter two groups had no difference between them ($p=0.112$). Hence, from the laboratory point of view, we did not see additional benefit in the reduction of thrombin activation by the anticoagulant intensities higher than the range INR 2-3. The evidence supported that this therapeutic range might be enough for Thai patients with mechanical heart valves.

Thromboembolism is a well recognized complication in patients with a mechanical heart valve. All the patients with this condition have to be treated with life-long anticoagulant to prevent the thrombotic event[1]. Even though oral anticoagulant has been used in these patients for quite a

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long time, the optimal level of its intensity which provides protective effect against thrombosis with minimal bleeding problems is still debatable. The recommended ranges in Western patients (expressed in terms of the International Normalized Ratio [INR]) are different such as INR 3.6-4.8[2], 2.5-3.5[3], 3.0-4.5[4]. We have reviewed clinical data to evaluate the proper range in Thai patients with mechanical heart valve replacements[5]. The results suggest that INR 2-3 might be enough, however, the conclusion needs further supporting evidence.

A foreign surface, for example, a mechanical heart valve can activate the contact system of the intrinsic coagulation pathway[6]. The key reaction in blood coagulation is the conversion of prothrombin to thrombin by factor Xa. During this process the amino-terminal half of prothrombin is released as the inactive prothrombin fragment 1+2 (F1+2)[7].

Elisas for F1+2 are novel methods that are highly sensitive and quantitative determinations. Measurement of this fragment can be used as a marker of coagulation activation and an assessment of hypercoagulable or hypocoagulable state[8,9].

Plasma F1+2 level is reduced in patients receiving oral anticoagulant therapy[10-13], and has been proposed to be useful to estimate the anticoagulant effect that counteract the procoagulant activity in patients with high risk of thrombosis[13]. It may be applied in monitoring this medication [11,13-15].

The aim of this study was to determine the effect of different oral anticoagulant intensities, expressed as INR, on the *in vivo* thrombin generation using F1+2 as an indicator. The findings might be evidence taken into consideration of the minimum level of anticoagulation to prevent hypercoagulable state which leads to thromboembolism in a laboratory aspect, besides clinical trial[16].

MATERIAL AND METHOD

Patients

116 patients with mechanical heart valve replacements, who have been under stable oral anticoagulation, in the form of warfarin, for at least one month, were selected according to anticoagulant intensities.

The normal control group consisted of 30 healthy individuals (15 males and 15 females) with a mean age of 29 years, range 16-41 years.

Method

We used prothrombin time (PT) to measure the intensity of warfarin therapy. PTs were performed with human placental thromboplastin (Thromborel® S). The International Sensitivity Index (ISI) of this reagent is 1.1. The INRs were calculated according to the following formular: INR = (patient PT/ mean control PT)^{ISI}.

Prothrombin fragment 1+2 was determined with enzyme immunoassay (Enzygnost® F1+2 assay, Behringwerke AG, Germany).

Statistical analysis

The relationship between F1+2 and INR of anticoagulated patients was analyzed by linear regression with Statview statistical computer package. The difference in F1+2 levels between each patient group and normal control were calculated by unpaired *t*-test with statistical significant difference of each pair at $p < 0.01$ using software SPSS/PC for Windows. Since these were multiple comparisons, the ability to detect significant difference at least one pair of comparison only by chance (not the real difference) was less than 5.58 per cent.

RESULTS

We measured the intensities of warfarin therapy expressed as INR and the levels of pro-

Table 1. Patients' characteristics.

Characteristics	No. of patients
Total	116
Sex	
Male	47
Female	69
Valve position	
Mitral	81
Aortic	20
Mitral & Aortic	10
Mitral & Tricuspid	3
Unknown	2
Type of mechanical valve	
Medtronic-Hall	51
CarboMedics	15
Starr-Edwards	22
St. Jude	8
Bjork-Shiley	12
Combined*	2
Unknown	6
Mean age 44 years (range 13-74 years)	

* Bjork-Shiley & Medtronic-Hall, Bjork-Shiley & Starr-Edwards

thrombin activation fragment F1+2 in 116 patients with mechanical heart valves. The patients' characteristics are shown in Table 1. The mean ± S.D. of F1+2 concentration in normal control was 0.7±0.17 nmol/L (Table 2). Two patients who had very high F1+2 levels were considered as outliers and not included in calculation. One of them had INR 1.6 and F1+2 5.6 nmol/L. This patient was followed-up one week later and INR was 1.6 and F1+2 was 5 nmol/L. The other patient had INR 3.5 and F1+2 10 nmol/L. At that time he had clinical deterioration in cardiac function due to atrial fibrillation. The sample was repeatedly assayed and the result was nearly the same.

Table 2. Plasma F1+2 levels in normal and each group of patients classified by the intensities of oral anticoagulant expressed as International Normalized Ratio (INR). The significances were calculated with unpaired *t*-test.

Group	n	F1+2 (nmol/L) mean ± S.D.	range	Difference from normal group (p)
Normal	30	0.70 ± 0.17	0.41 - 1.15	NT
INR 1.1-1.9	33	0.60*± 0.30	0.17 - 1.40	0.119
INR 2-3	38	0.28 ± 0.10	0.06 - 0.54	0.000
INR 3.1-4.2	43	0.24 ± 0.13	0.10 - 0.70	0.000

NT = not tested.
*Significantly different from other patient groups (p=0.000)

After excluding the two cases, the relationship between F1+2 and INR is presented in Fig. 1. Overall correlation coefficient ($r1$) was -0.586 ($p<0.001$). There was maximum correlation when we analyzed the data in the range INR lower than 3 ($r2$ = -0.658, $p< 0.001$). If the data of the higher INR range were included, the correlation coefficient decreased (not shown).

We subsequently classified the patients by INR range into three groups i.e., INR 1.1-1.9, INR 2-3 and INR 3.1-4.2. The F1+2 levels in normal and each patient group are presented in Table 2. The concentration of F1+2 in the first group did not differ from normal, but was higher than the others. There was no difference between the latter two groups.

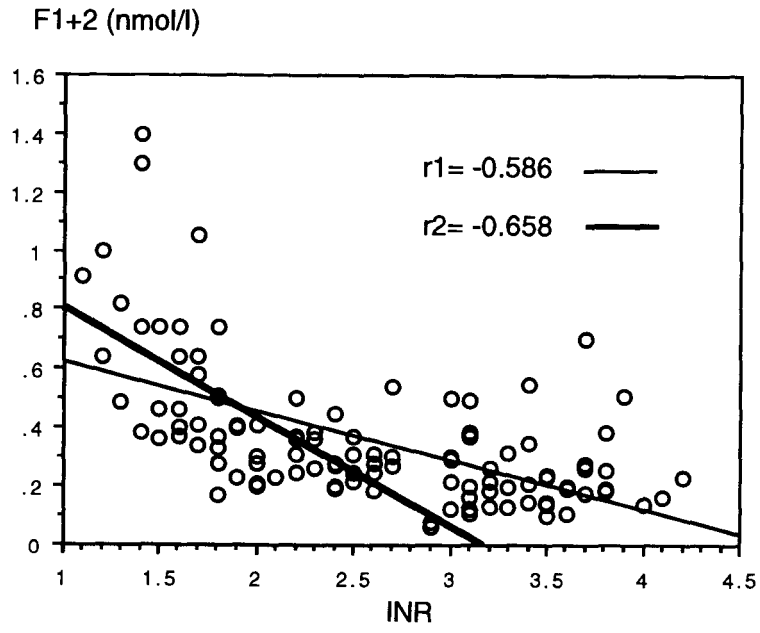

Fig. 1. Correlation between anticoagulant intensities expressed as International Normalized Ratio (INR) and prothrombin fragment 1+2 (F1+2) in 114 patients with mechanical heart valves, after excluding two outliers. "r1" and "r2" represent correlation coefficients when the data of all patients and only whose INR lower than 3 are analyzed respectively.

DISCUSSION

The mean ± S.D. of F1+2 in normal control is 0.70 ± 0.17 nmol/L corresponding to the report by Pelzer et al, that was 0.67 ± 0.19 nmol/L[8].

After excluding two outliers, there was inverted relationship between the *in vivo* thrombin generation indicated by F1+2 level and the intensities of anticoagulant in these patients. However, the correlation coefficient was decreased when the additional data of the patients that had INR of more than 3 were included. This suggested that the increment of intensity beyond a certain level might not give further linear reduction of thrombin activation proportionally. There was a study that showed no correlation between the degree of reduction in F1+2 levels. However, the intensities of oral anticoagulant were expressed in PT ratio and the patients had different conditions (venous and arterial thromboembolism)[13].

Plasma F1+2 in the group having INR 1.1-1.9 which were considered as an inadequate dose, did not have significant difference from normal control while the other two groups did. There is no clinical trial that has scrutinized the effect of different F1+2 levels on the clinical outcome of patients with mechanical heart valves. Therefore, we considered it with our preliminary study, in which the record of 125 Thai patients with this condition were reviewed. It showed that most thromboembolisms occurred in this range[5]. We surmised that the patient should be in hypocoagulable state compared with normal, in other words, have lower plasma F1+2. If the *in vivo* thrombin generation in the patients was equal to that of normal, when some changes were triggered by many conditions, it might become a hypercoagulable state which leads to thromboembolism.

F1+2 in the group INR 2-3 did not differ from that with higher INR. In a study, with results similar to ours, the residual thrombin activity in orally anticoagulated patients with mechanical heart valve prostheses was measured by using the level of F1+2 and thrombin-antithrombinIII complex and classified into three groups according to the INR range (INR 4.8-3.6, 3.5-2.5, 2.4-2.1). Differences of the two parameters between these groups of patients were small and negligible[15]. Hence, from the laboratory point of view, we did not see additional benefit in reduction of thrombin activation with higher intensities of oral anticoagulant, comparing INR 2-3 with INR more than 3.

In a recently clinical study, including 1,608 patients with mechanical heart valves, they concluded that the optimal intensity of anticoagulation with lowest incidence of thromboembolic complication and bleeding was INR between 2.5 and 4.9 and the recommended target INR was 3-4[17]. It is not certain that the therapeutic range in Westerners could be directly applied to Thai patients who have a lower incidence of thromboembolism in the general population, and practically, this range has not been used[18].

One of our patients who had very high F1+2 concentration had been receiving inadequate therapy. The value after the successive follow-up was nearly the same implying that the test is quite reliable. It's interesting that the other one with high intensity (INR 3.5) which should be considered as adequate, still had hypercoagubable state. We do not know whether the current illness was the cause of the individual variation in this case.

The results in this study support our previous retrospective clinical review that anticoagulant intensities in the range INR 2-3 might be enough to prevent embolic complication in Thai patients with mechanical heart valves. Since the risk of bleeding is related to the intensity of oral anticoagulant, to maintain the medication at a higher level (INR more than 3) may increase this side-effect and cause more expense[19,20]. However, the optimal intensity for this condition should be established by clinical trials, and measurement of F1+2 might be useful in monitoring the individual response to the medication.

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ผลของระดับยากันเลือดแข็งชนิดรับประทานในขนาดต่าง ๆ กันต่อโปรธรอมบิน แฟร็กเม้นท์ 1+2 ในผู้ป่วยไทยที่ได้รับการเปลี่ยนลิ้นหัวใจ แบบกล

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ระดับของขนาดยากันเลือดแข็งชนิดรับประทาน ที่น้อยที่สุดที่สามารถป้องกันการเกิดลิ่มเลือดอุดตันในผู้ป่วยที่ได้รับการเปลี่ยนลิ้นหัวใจแบบ mechanic ยังไม่มีข้อสรุปที่แน่นอน และขนาดการรักษาที่เหมาะสมในผู้ป่วยชาวตะวันตกอาจไม่สามารถนำมาใช้กับผู้ป่วยไทยได้โดยตรง คณะผู้วิจัยได้เคยศึกษาข้อมูลทางคลินิกย้อนหลังในผู้ป่วยกลุ่มนี้ พบว่าระดับยาที่เหมาะสมอาจจะอยู่ในช่วง International Normalized Ratio (INR) 2–3 แต่ข้อสรุปนี้ ยังต้องการหลักฐานสนับสนุนในการวิจัยครั้งนี้จึงได้ศึกษาผลของยาในระดับต่าง ๆ ซึ่งรายงานเป็น INR ที่มีต่อระดับการกระตุ้นระบบการแข็งตัวของเลือดในร่างกาย โดยใช้ prothrombin fragment 1+2 (F1+2) เป็นตัวชี้บ่ง ในผู้ป่วยไทยซึ่งได้รับการเปลี่ยนลิ้นหัวใจแบบ mechanic 116 ราย

ผลการศึกษาพบว่า ระดับของ F1+2 ในคนปกติ 30 ราย มีค่าเท่ากับ 0.7 ± 0.17 nmol/L ในกลุ่มผู้ป่วยเมื่อตัดสองรายซึ่งมีค่าสูงมากผิดปกติ (outliers) ออก จะพบความสัมพันธ์ระหว่าง INR และ F1+2 มากที่สุดในช่วงที่ผู้ป่วยมี INR น้อยกว่า 3 โดยค่า correlation coefficient เป็น −0.658 ($p < 0.001$) แต่เมื่อคำนวณรวมกับผู้ป่วยที่มี INR สูงกว่านี้ความสัมพันธ์จะลดลง เมื่อแบ่งผู้ป่วยออกเป็น 3 กลุ่มโดยใช้ค่า INR คือกลุ่มที่มี INR 1.1–1.9, 2–3 และ 3.1–4.2 พบว่าค่า F1+2 ในแต่ละกลุ่มเท่ากับ 0.6 ± 0.30, 0.28 ± 0.13, 0.24 ± 0.13 nmol/L ตามลำดับ F1+2 ในกลุ่มแรกไม่แตกต่างจากกลุ่มปกติ แต่สูงกว่าอีกสองกลุ่มอย่างมีนัยสำคัญทางสถิติ ส่วนสองกลุ่มหลังนั้นไม่แตกต่างกัน

โดยสรุป การศึกษาทางห้องปฏิบัติการสนับสนุนว่า ขนาดของยากันเลือดแข็งในช่วง INR 2–3 อาจเพียงพอสำหรับผู้ป่วยไทยซึ่งได้รับการเปลี่ยนลิ้นหัวใจแบบ mechanic โดยที่การเพิ่มขนาดยาที่มากกว่านี้ไม่ได้ประโยชน์เพิ่มขึ้นในแง่การลดการกระตุ้นระบบการแข็งตัวของเลือด

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