Utility of Noninvasive Endothelial Function Test for Prediction of Deep Vein Thrombosis After Total Hip or Knee Arthroplasty

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Background: Venous thromboembolism (VTE) is a common and sometimes lethal postoperative complication of arthroplasty. Endothelial dysfunction is important in the pathogenesis of thrombus formation. Reactive hyperemia-peripheral arterial tonometry (RH-PAT) can noninvasively evaluate endothelial function. This study investigated the predictive value of RH-PAT for deep vein thrombosis (DVT) after lower limb arthroplasty.

Methods and Results: A prospective observational study of 126 osteoarthritic patients who underwent total knee arthroplasty (TKA) or hip arthroplasty (THA) was conducted. The RH-PAT index (RHI) was measured on the day before surgery, and presence of DVT was checked by ultrasonography or phlebography before and after surgery. Following arthroplasty, DVT was diagnosed in 51 patients (40.5%). RHI in the DVT group (0.58±0.25) was significantly lower than in the non-DVT group (0.71±0.25, P=0.004). RHI was a significant and independent predictor of postoperative DVT in multivariate logistic regression analyses and improved a net reclassification index (23.8%, P=0.022). Subgroup analyses according to operation site with adjustment for Qthrombosis score demonstrated that RHI significantly predicted postoperative DVT in the THA group (odds ratio per 0.1, 0.77; 95% confidence interval 0.60–0.98; P=0.03), but did not reach statistical significance in the TKA group.

Conclusions: Low RHI was significantly associated with DVT after lower limb arthroplasty. Endothelial dysfunction, as assessed by RH-PAT, is potentially useful for identifying patients at high risk for VTE especially after THA. (Circ J 2014; 78: 1723–1732)

Key Words: Arthroplasty; Endothelial function; Venous thromboembolism

Postoperative venous thromboembolism (VTE), which includes deep vein thrombosis (DVT) and pulmonary embolism (PE), is a common and sometimes lethal complication of surgery. The incidence of VTE is particularly high after orthopedic surgery. Despite the current use of prophylaxis in the clinical setting, subclinical venous thrombosis develops after surgery in 15–20% of patients who undergo total hip arthroplasty (THA) and in 40–60% of those who undergo total knee arthroplasty (TKA). The traditional risk factors for thrombosis often do not predict VTE events. Accurate risk
stratification is warranted to establish new prophylactic strategies and to improve prognosis.

Endothelial dysfunction is reported to be associated with increased risk of cardiovascular events. The endothelium is responsible for the fine tuning of vascular homeostasis and mirrors current vascular function through its control of coagulation, fibrinolysis, and platelet activation. Many risk factors for VTE overlap with those for atherosclerotic disease, and patients with VTE have a high risk of subsequent development of atherosclerotic disease. The vascular endothelium regulates the functional capabilities of the entire circulatory system; in patients with endothelial dysfunction of the peripheral arteries, the venous functional status may have deteriorated as well.

Reactive hyperemia-peripheral arterial tonometry (RH-PAT) is a noninvasive, automatic, and less operator-dependent clinical test that is used to evaluate peripheral endothelial function. In a community-based cohort, Hamburg et al reported a significant relationship between RH-PAT and cardiovascular risk factors.

The aim of this study was to determine whether peripheral arterial endothelial dysfunction, as assessed by RH-PAT, could predict the development of DVT after arthroplasty of the lower limb.

Methods

Study Design and Patients

The following criteria were used for inclusion of patients with osteoarthritis in this prospective observational study: (1) age ≥20 years, (2) negative for preoperative DVT, and (3) eligibility for THA or TKA at Yokohama City University Medical Center between February 2011 and January 2013. Exclusion criteria: treatment with anticoagulants, active systemic inflammatory disease, congenital deficiency of protein C or protein S, antiphospholipid antibody syndrome, and antithrombin deficiency.

RH-PAT was performed in the fasting state early in the morning on the day before surgery, at the same time as blood and urine samples were also taken. Ultrasonography or phlebography was scheduled for before and on postoperative day 8 to evaluate the occurrence of DVT. A tourniquet was applied to reduce bleeding and obtain a clear surgical field during the TKA operation. Supervised quadriceps exercises were initiated within 24 h after surgery in all patients. Patients were routinely mobilized to a wheelchair on the first day after surgery. We defined delayed rehabilitation as patients who required 2 or more days to rise from their beds. Thromboprophylaxis therapy was provided according to the American College of Chest Physicians Evidence-Based Clinical Practice Guidelines. We allowed surgeons to use edoxaban, which is noninferior with respect to treatment with anticoagulants, active systemic inflammatory disease, congenital deficiency of protein C or protein S, antiphospholipid antibody syndrome, and antithrombin deficiency.

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Ultrasonography and Phlebography

Bilateral ultrasonography was performed to evaluate the presence of DVT in all patients before and after surgery using a standardized technique. Phlebography was performed pre- and postoperatively in patients (n=20) whose lower limb veins could not be visualized satisfactorily by preoperative ultrasonography. The assessment of DVT was made by physicians and sonographers who were blinded to the results of RH-PAT. Ultrasonography was performed by experienced technicians using a 10-MHz linear probe and a 3.5-MHz convex probe. All patients were evaluated using compression ultrasound testing and Doppler ultrasonography of the entire leg. Iliac veins were visualized by direct imaging and Doppler flow. All venous segments over the entire length of the leg were examined in both the transverse and longitudinal axes. DVT was ruled out by negative results of the compression ultrasonography test and no visualized thrombus. DVT was diagnosed by lack of compressibility of a deep vein and, (eg, the iliac vein) by direct visualization of thrombus or the absence of Doppler flow according to a previous report. Ultrasonography was interpreted independently by the sonographer performing the examination and by the physicians. During the period of study, the examinations were performed by one of 2 sonographers. Bilateral ascending contrast phlebography was performed as reported previously. The diagnostic criteria for DVT were the presence of an intraluminal filling defect on at least 2 different projections or a venous segment that did not fill despite repeated injections of contrast material according to previous reports. Phlebography was independently interpreted by 2 physicians. In case of disagreement, consensus was made by 2 physicians who were blinded to the results of RH-PAT. Proximal DVT was defined as occurring in the popliteal vein or above, and distal DVT was defined as occurring below the popliteal vein.

Qthrombosis Score

The risk for VTE was assessed by the Qthrombosis score in each patient. Julia Hippisley-Cox et al developed and validated a new risk prediction algorithm designed to predict the absolute risk of VTE in a large representative primary care population. Independent risk factors included in the model for men and women are age, body mass index, smoking status, varicose veins, congestive cardiac failure, chronic kidney disease, cancer, chronic obstructive pulmonary disease, inflammatory bowel dis-
Coronary risk factors were defined as current smoking (smoking within 1 year), hypertension (>140/90 mmHg or taking antihypertensive medication), dyslipidemia (high-density lipoprotein cholesterol <40 mg/dl, low-density lipoprotein cholesterol ≥140 mg/dl, triglycerides ≥150 mg/dl, or taking medication for dyslipidemia), and diabetes mellitus (symptoms of diabetes plus a random plasma glucose concentration ≥200 mg/dl, fasting plasma glucose concentration ≥126 mg/dl, 2-h plasma glucose concentration ≥200 mg/dl during a 75-g oral glucose tolerance test, or taking medication for diabetes mellitus). The estimated glomerular filtration rate was determined using the equation proposed by the Japanese Society of Nephrology and based on the equation described in the Modification of Diet in Renal Disease Study.26 The urine albumin-creatinine ratio (in mg/g) was assessed in a morning urine specimen as the ratio between urinary albumin and urinary creatinine measured with a modified Jaffe method.27 The homeostasis model assessment of insulin resistance (HOMA-IR) was calculated by the following equation: HOMA-IR=fasting plasma glucose (mmol/L)*fasting plasma insulin (μU/ml)/22.5.28

Statistical Analysis
Variables that showed normal distribution are expressed as mean (standard deviation), whereas those with a skewed distribution are expressed as median (interquartile range). Continuous variables were analyzed by unpaired t-test or Mann-Whitney U test as appropriate. Categorical variables were analyzed using a chi-square test (and Fisher’s exact test). Spearman’s correlation coefficient (rho) was used for evaluation of a possible association between RHI and Framingham risk score. The association between the presence of DVT and other parameters was analyzed by multiple logistic regression analysis with the backward algorithm and the forced entry algorithm, and a Hosmer-Lemeshow goodness-of-fit statistic was calculated. Because the Qthrombosis risk score already includes age and other risk factors, we did not include it in the multivariate model to avoid overlapping adjustment as well as the Framingham risk score. We built multivariate regression models with forced entry algorithm using RHI and Framingham risk score, DVT-related traditional risk factors as reported previously (age, sex, height, weight, hypertension, dyslipidemia, diabetes mellitus, and current smoking)9 (model-1), and Qthrombosis score (model-2). Receiver-operating characteristics (ROC) curves were constructed. The area under the curve (AUC), sensitivity, and specificity were calculated to predict the presence of postoperative DVT, with AUC value of 0.50 representing no accuracy and a value of 1.00 indicating maximal accuracy. We defined the optimal thresholds of RHI by maximizing the sums of sensitivity and specificity. Positive and negative predictive values were also calculated. The C-statistics were estimated after addition of RHI to the DVT-related traditional risk factors9 and Qthrombosis risk score to compare AUC values using an algorithm suggested by DeLong et al.29 The incremental effects of adding RHI were also evaluated using the net reclassification index (NRI) as previously described.30 For the assessment of reclassification improvement, we defined 3 risk categories on the basis of traditional risk factors: low risk <30%, intermediate risk 30–50%, and high risk >50%. We assessed subgroup analyses according to operation site and delay in postoperative DVT evaluation. The priori power analysis was performed to detect the minimum number of patients required.

Risk Factors
Coronary risk factors were defined as current smoking (smoking within 1 year), hypertension (>140/90 mmHg or taking antihypertensive medication), dyslipidemia (high-density lipoprotein cholesterol <40 mg/dl, low-density lipoprotein cholesterol ≥140 mg/dl, triglycerides ≥150 mg/dl, or taking medication for dyslipidemia), and diabetes mellitus (symptoms of diabetes plus a random plasma glucose concentration ≥200 mg/dl, fasting plasma glucose concentration ≥126 mg/dl, 2-h plasma glucose concentration ≥200 mg/dl during a 75-g oral glucose tolerance test, or taking medication for diabetes mellitus). The estimated glomerular filtration rate was determined using the equation proposed by the Japanese Society of Nephrology and based on the equation described in the Modification of Diet in Renal Disease Study.26 The urine albumin-creatinine ratio (in mg/g) was assessed in a morning urine specimen as the ratio between urinary albumin and urinary creatinine measured with a modified Jaffe method.27 The homeostasis model assessment of insulin resistance (HOMA-IR) was calculated by the following equation: HOMA-IR=fasting plasma glucose (mmol/L)*fasting plasma insulin (μU/ml)/22.5.28
SUZUKI H et al.

Table 1. Clinical Characteristics of Osteoarthritic Patients Who Underwent Total Knee or Hip Arthroplasty

|                      | All group (n=126) | Postoperative DVT Yes (n=51) | No (n=75) | P value |
|----------------------|-------------------|------------------------------|-----------|---------|
| **Age, years**       | 70.4±9.7          | 73.7±7.5                     | 68.1±10.4 | 0.001   |
| **Male sex, n (%)**  | 18 (14.3)         | 6 (11.8)                     | 12 (16.0) | 0.51    |
| **Body mass index, kg/m²** | 24.6±4.5       | 24.5±3.2                     | 24.6±5.3  | 0.85    |
| **Current smoking, n (%)** | 14 (11.1)   | 1 (2.0)                      | 13 (17.3) | 0.007   |
| **Hypertension, n (%)** | 74 (58.7)       | 30 (58.8)                    | 44 (58.7) | 0.99    |
| **Diabetes mellitus, n (%)** | 17 (13.5)     | 8 (15.7)                     | 9 (12.0)  | 0.55    |
| **Dyslipidemia, n (%)** | 72 (57.1)       | 30 (58.8)                    | 42 (56.0) | 0.99    |
| **Coronary artery disease, n (%)** | 6 (4.8)        | 3 (5.9)                      | 3 (4.0)   | 0.63    |
| **Chronic obstructive pulmonary disease, n (%)** | 2 (1.6)        | 1 (2.0)                      | 1 (1.3)   | 0.78    |
| **eGFR, ml·min⁻¹·1.73m⁻²** | 72.6±16.7       | 70.8±16.2                    | 73.9±17.0 | 0.30    |
| **hsCRP, mg/L, median, IQR** | 0.7 (0.4–1.4) | 0.6 (0.4–1.3)                | 0.9 (0.4–1.5) | 0.53 |
| **BNP, pg/ml, median, IQR** | 23.9 (15.2–41.7)| 27.7 (17.8–42.4)           | 19.8 (11.9–38.9) | 0.066 |
| **HOMA-IR, median, IQR** | 1.8 (1.1–2.4)  | 1.8 (1.3–2.4)                | 1.7 (1.0–2.4) | 0.35   |
| **HbA1c, %, median, IQR** | 5.3 (4.9–5.6)   | 5.4 (5.2–5.6)                | 5.1 (4.8–5.6) | 0.053 |
| **UACR, mg/g, median, IQR** | 8.0 (5.4–13.4) | 8.1 (5.4–12.4)               | 7.5 (5.4–14.4) | 0.87   |
| **D-dimer, μg/ml, median, IQR** | 0.8 (0.5–1.3) | 0.9 (0.6–1.8)                | 0.7 (0.5–1.2) | 0.068  |
| **PAI-1, ng/ml, median, IQR** | 23.0 (14.5–31.0) | 24.0 (14.0–33.0)            | 22.0 (14.8–28.3) | 0.35   |
| **TAT, ng/ml, median, IQR** | 3.1 (2.2–4.7)   | 3.5 (2.3–4.7)                | 2.6 (2.1–4.5) | 0.12   |

**Medication, n (%)**
- Aspirin 13 (10.3)   7 (13.7) 6 (8.0) 0.30
- Statins 32 (25.4) 15 (29.4) 17 (22.7) 0.39
- Antihypertensive drugs 65 (51.6) 27 (52.9) 38 (50.7) 0.80
- NSAIDs 92 (73.0) 39 (72.5) 53 (70.7) 0.47

**Operative time, min, median, IQR**
- TKA (n=62) 69 (35–169) 61 (39–161) 79 (31–188) 0.93
- THA (n=64) 564 (321–840) 600 (384–889) 530 (300–806) 0.47

**Blood transfusion, n (%)**
- 63 (50.0) 20 (39.2) 43 (57.3) 0.050

**Prophylactic anticoagulation therapy, n (%)**
- Factor Xa inhibitor, n (%) 67 (53.2) 31 (60.8) 36 (48.0) 0.16
- Fondaparinux, n (%) 63 (50) 28 (54.9) 35 (46.7) 0.36
- Edoxaban, n (%) 48 (38.1) 20 (39.2) 28 (37.3) 0.83
- Low-molecular-weighted heparin, n (%) 15 (11.9) 8 (15.7) 7 (9.3) 0.28

**Graduated compression stockings, n (%)**
- 126 (100) 51 (100) 75 (100) >0.99

**Bilateral surgery, n (%)**
- 62 (49.2) 38 (74.5) 24 (32.0) <0.001

**Framingham risk score, %, median, IQR**
- 7.0 (3.0–11.0) 9.0 (3.0–12.0) 5.0 (3.0–10.0) 0.18

**Qthrombosis at 1 year, %, median, IQR**
- 2.1 (1.2–3.5) 2.6 (1.9–3.8) 1.8 (1.1–2.9) 0.004

**RHI**
- 0.66±0.26 0.58±0.25 0.71±0.25 0.004

*Data are mean±SD.
P values are for differences between the DVT and non-DVT groups.
Body mass index was calculated by dividing weight in kilograms by (height)² in meters. HOMA-IR is calculated using the following equation:
HOMA-IR = fasting plasma glucose (mmol/L) × fasting plasma insulin (μU/ml)/22.5.
BNP, B-type natriuretic peptide; DVT, deep vein thrombosis; eGFR, estimated glomerular filtration rate; HbA1c, hemoglobin A1c; HOMA-IR, homeostasis model assessment of insulin resistance; hsCRP, high-sensitivity C-reactive protein; IQR, interquartile range; NSAIDs, nonsteroidal antinflammatory drugs; PAI-1, plasminogen activator inhibitor-1; RHI, reactive hyperemia-peripheral arterial tonometry index; TAT, thrombin-antithrombin complex; THA, total hip arthroplasty; TKA, total knee arthroplasty; UACR, urine albumin-creatinine ratio.

To detect a significant difference in the RHI between patients with and without postoperative DVT. For 2-sided α=0.05, power=0.90, events rate of 35%, the required sample size was estimated as 119 patients to detect an odds ratio (OR) of 0.5 per 1 standard deviation of RHI under the normality assumption on the distribution of RHI. P<0.05 denoted statistical significance. Statistical analyses were performed using SPSS version 19.0 (SPSS Inc, Tokyo, Japan) and SAS version 9.2 (SAS Institute Inc, Cary, NC, USA).
Results

We recruited 127 patients hospitalized for elective THA or TKA; 1 patient with anti-phospholipid antibodies was excluded (Figure 1), so data for the remaining 126 patients (62 TKA, 64 THA) were analyzed. Of them, 33 patients underwent bilateral arthroplasty and 93 patients underwent unilateral arthroplasty. All patients used the intermittent pneumatic compression device, and 67 patients received pharmacologic prophylaxis (factor Xa inhibitor n=63; low-molecular-weight heparin n=3; direct thrombin inhibitor n=1). Although ultrasonography or phlebography was performed on postoperative day 8, the assessment of postoperative DVT was delayed in 25 patients because of their condition, examination availability or holidays. In total, 51 (40.5%) of the 126 patients developed postoperative DVT (4 proximal DVT, 47 distal DVT). Among the 93 patients who underwent unilateral arthroplasty, 27 developed DVT on the operative side, 7 developed DVT on both the left and right sides, 2 developed DVT on the side contralateral to the operation side, and 57 patients did not develop DVT. The DVT was asymptomatic in all cases. We did not perform routine surveillance for PE, but no patients presented clinical symptoms of PE. We performed contrast-enhanced chest computed tomography (n=3) or perfusion/ventilation scintigraphy (n=1) in all patients with proximal DVT (n=4), and there was no evidence of PE.

Baseline Characteristics (Table 1)

Patients diagnosed with DVT were significantly older than those who did not. Among the 14 current smokers, only 1 developed DVT. The incidence of DVT was higher in patients who underwent TKA than in those who underwent THA. DVT occurred in 38 (61.3%) of the 62 patients who underwent TKA and in 13 (20.3%) of the 64 patients who underwent THA (P<0.001). The Qthrombosis score was significantly higher in patients who developed DVT than in those who did not. Framingham risk score tended to be higher in patients who developed DVT than in those who did not, and was inversely related to RHI (rho=−0.257, P=0.004).

Other details of the medical history and clinical characteristics did not differ between the 2 groups, nor did the laboratory tests of blood coagulation and fibrinolysis, including D-dimer, plasminogen activator inhibitor-1, and thrombin-antithrombin complex levels.
Perioperative Period Conditions
There was no significant difference in the pharmacological thromboprophylaxis applied to both groups. Furthermore, the operative time, intraoperative blood loss, and blood transfusion were not significantly different between the 2 groups (Table 1). On the first day after surgery, 113 patients were mobilized to a wheelchair, while the remaining 13 patients took more than 1 day to rise from bed to a wheelchair. The interval until mobilization to a wheelchair did not correlate with the incidence of postoperative DVT. The prevalence of a delay in postoperative DVT evaluation was higher in patients with DVT.

RHI and Development of DVT
Figure 2 shows RH-PAT signals of representative patients who did and did not develop DVT. The recording in the patient with DVT showed a blunted finger PAT response during the reactive hyperemia phase (Figure 2B). The mean RHI of patients with DVT (0.58±0.25) was significantly lower than that of DVT-free patients (0.71±0.25, P=0.004, Table 1, Figure 2C). Simple logistic regression analysis demonstrated that older age, non-smoker, undergoing TKA, higher Qthrombosis score, delay in postoperative DVT evaluation, and lower RHI were significant predictors of the occurrence of DVT after arthroplasty (Table 2). In the multiple logistic regression analysis with backward algorithm, RHI and TKA were independent and significant predictors of the occurrence of DVT after arthroplasty (RHI: OR per 0.1, 0.75; 95% confidence interval (95% CI) 0.67–0.95; P=0.013; TKA: OR 9.50; 95% CI 3.51–25.7; P<0.001; Table 2). RHI was independently associated with the occurrence of DVT in multiple logistic regression analysis (Table 2), and in patients without a delay in postoperative DVT evaluation, RHI was a significant predictor in both single regression analysis and multivariate regression analysis model-1 (Table S1). In the forced entry multivariate regression model using the DVT-related traditional risk factors, RHI significantly and independently predicted postoperative DVT (OR per 0.1, 0.80; 95% CI 0.67–0.95; P=0.013; Table 2). The RHI was also associated with postoperative DVT independently of the Framingham risk score (OR per 0.1, 0.81; 95% CI 0.69–0.94; P=0.007), The Qthrombosis score (for 1 year) was a significant predictor of postoperative DVT (OR for increasing tertiles, 1.96; 95% CI 1.26–3.01, P=0.003; Table 2). The RHI was still a significant predictor of postoperative DVT after adjustment.

Table 2. Logistic Regression Analysis for the Development of DVT After Total Hip or Knee Arthroplasty

| Variable                        | Simple regression | Multiple regression (backward) | Multiple regression (Forced entry model 1) | Multiple regression (Forced entry model 2) |
|---------------------------------|-------------------|---------------------------------|--------------------------------------------|-------------------------------------------|
|                                 | OR 95% CI P value | OR 95% CI P value               | OR 95% CI P value                          | OR 95% CI P value                          |
| Age (per year)                  | 1.07 1.02–1.12 0.002 | Not selected                     | 1.08 1.02–1.15 0.009                       | –                                         |
| Sex (male)                      | 0.51 0.24–2.00 0.51 | 0.27 0.06–1.29 0.10              | 0.69 0.16–2.86 0.60                        | –                                         |
| Height (per cm)                 | 0.99 0.94–1.04 0.74 | 1.07 0.99–1.15 0.10              | 1.07 0.99–1.15 0.11                        | –                                         |
| Weight (per kg)                 | 0.99 0.97–1.02 0.66 | Not selected                     | 1.00 0.96–1.04 0.99                        | –                                         |
| Hypertension (yes)              | 1.01 0.49–3.81 0.55 | Not selected                     | 0.50 0.19–1.35 0.17                        | –                                         |
| Dyslipidemia (yes)              | 1.12 0.55–2.31 0.75 | Not selected                     | 1.51 0.61–3.73 0.37                        | –                                         |
| Diabetes mellitus (yes)         | 1.36 0.49–3.81 0.55 | Not selected                     | 2.43 0.70–8.82 0.16                        | –                                         |
| Current smoking (yes)           | 0.095 0.01–0.75 0.026 | 0.16 0.02–1.48 0.11              | 0.12 0.01–1.17 0.07                        | –                                         |
| Ln [BNP] (per 0.1)              | 1.08 0.98–1.18 0.11 | Not selected                     | –                                          | –                                         |
| Ln [hsCRP] (per 0.1)            | 0.97 0.90–1.05 0.48 | Not selected                     | –                                          | –                                         |
| Ln [UACR] (per 0.1)             | 0.98 0.90–1.07 0.64 | Not selected                     | –                                          | –                                         |
| Statin (yes)                    | 1.42 0.63–3.19 0.39 | Not selected                     | –                                          | –                                         |
| ACEI (yes)                      | 4.63 0.47–45.7 0.19 | Not selected                     | –                                          | –                                         |
| Delayed rehabilitation (yes)    | 1.30 0.41–4.11 0.66 | Not selected                     | –                                          | –                                         |
| Pharmacological prophylaxis (yes)| 0.60 0.29–1.23 0.16 | Not selected                     | –                                          | –                                         |
| Delay of postoperative DVT evaluation (yes) | 2.71 1.10–6.65 0.03 | 3.86 1.26–11.9 0.02 | – | – |
| TKA (yes)                       | 6.21 2.81–13.8 <0.001 | 9.50 3.51–25.7 <0.001          | –                                          | –                                         |
| Qthrombosis at 1 year (per tertiles) | 1.96 1.26–3.01 0.003 | – | – | 1.80 1.14–2.84 0.012 |
| RHI (per 0.1)                   | 0.81 0.69–0.94 0.005 | 0.75 0.63–0.91 0.003            | 0.80 0.67–0.95 0.013                        | 0.83 0.71–0.97 0.021                      |

Multivariate analysis was performed with the backward algorithm with significance set at 0.20. The Hosmer-Lemeshow goodness-of-fit chi-square and P value are 13.8 and 0.09, respectively. Forced inclusion model-1: adjusted for age, sex, height, weight, hypertension, dyslipidemia, diabetes mellitus, and current smoking. Model 2: adjusted for Qthrombosis risk score. ACEI, angiotensin converting enzyme inhibitor; CI, confidence interval; OR, odds ratio. Other abbreviations as in Table 1.
Endothelial Function and Post-Arthroplasty DVT

by Qthrombosis score (for 1 year: OR per 0.1, 0.83; 95% CI 0.71–0.97, P=0.021; Table 2).

ROC Analysis and NRI of RHI in Predicting DVT After Lower Limb Arthroplasty

ROC analysis demonstrated that the RHI was a significant predictor of DVT after surgery (AUC 0.65; 95% CI 0.56–0.75, P=0.004). Using a cutoff value of RHI <0.76, the sensitivity and specificity for prediction of DVT after arthroplasty were 80% and 45%, respectively, with positive and negative predictive values of 50% and 77%, respectively. Adding the RHI to the DVT-related traditional risk factors increased the C-statistics from 0.695 to 0.731 (Figure 3A), and the NRI was significant with the inclusion of the RHI (16.0% for patients without postoperative DVT, 7.8% for patients with postoperative DVT, and 23.8% for overall, P=0.022) (Table 3A). The addition of the RHI to the Qthrombosis score showed similar results (C-statistics: from 0.655 to 0.671; Figure 3B) (NRI: 18.7% for patients without postoperative DVT, 9.8% for patients with postoperative DVT, and 28.5% for overall, P=0.018; Table 3B).

Subgroup Analysis According to Operation Site

Although the number of patients was small, we performed subgroup analyses to confirm the trend of the association between the RHI and the occurrence of DVT in each group stratified by operation site. The AUC of the ROC curve by RHI to predict postoperative DVT was 0.71 with 95% CI 0.56–0.86 (P=0.019) in patients who underwent THA, and 0.62 with 95% CI 0.48–0.76 (P=0.112) in patients who underwent TKA. In patients who underwent THA, the RHI was significantly associated with postoperative DVT in single regression analysis and multivariate regression analysis model-2, but not in model-1 (Table 4). In patients who underwent TKA, the RHI was not significantly associated with postoperative DVT in logistic regression analyses (Table 4). NRI in the subgroup analyses showed the same tendency, but did not reach a significant level with the addition of RHI to the DVT-related traditional risk factors and Qthrombosis score in patients who either underwent TKA or THA (Table S2).

Discussion

The present study showed that endothelial dysfunction as assessed by RH-PAT predicted the development of DVT after arthroplasty of the lower limb, especially hip arthroplasty. Endothelial dysfunction was a significant predictor of DVT even after adjustment for DVT-related risk factors, Framingham risk score, and Qthrombosis score. Although there are no established predictors of postoperative VTE in the clinical setting, the present results suggest that RH-PAT is potentially useful for identifying patients at high risk for postoperative VTE. Although several measures reduce the risk of VTE, orthopedic surgeons are properly concerned about the potential hemorrhagic effect of pharmacological prophylaxis, which could be the cause of prolonged recovery, wound failure, and even periprosthetic infection. The benefit of using anticoagulants for the prevention of VTE is counterbalanced by the potential risk of bleeding. Thus, an important issue in the prevention of VTE is risk stratification (ie, whether some patients are at greater risk for VTE than others and therefore should receive different doses of anticoagulants or different treatment modalities). This study is the first to assess the relation between endothelial dysfunction and the development of VTE after surgery, and demonstrated that the RHI could be a useful screening test. The results of this study provide useful information about the assessment of patients at high risk of VTE, and that such patients

Figure 3. Receiver-operating characteristic (ROC) curve for the diagnostic value of the RHI for identification of DVT after orthopedic surgery. (A) ROC curves for the abilities of DVT-related traditional risk factors (blue, AUC=0.695) and the traditional risk factors+RHI (red, AUC=0.731) to predict postoperative DVT. (B) ROC curves for the abilities of the Qthrombosis risk score (blue, AUC=0.655) and Qthrombosis risk score+RHI (red, AUC=0.671) to predict postoperative DVT. AUC, area under the curve; DVT, deep vein thrombosis; RHI, reactive hyperemia-peripheral arterial tonometry index.
should be monitored carefully during follow-up and deserve a careful treatment strategy.

Traditionally, the pathogenesis of VTE is considered to be different from that of atherosclerotic cardiovascular disease. However, recent epidemiological studies have demonstrated an overlap of many risk factors for atherosclerotic diseases with venous thrombosis. Furthermore, microalbuminuria, which is a well-established risk factor of arterial thromboembolism and can reflect endothelial dysfunction, has been reported to be independently associated with VTE. Assessment of endothelial function could be useful in risk stratification not only for atherosclerotic disease but also for VTE. Migliacci et al found endothelial dysfunction in patients with spontaneous VTE, as assessed by flow-mediated vasodilatation (FMD), compared with age- and sex-matched controls. Their study was a cross-sectional, case-control study. To our knowledge, our study is the first prospective study to assess the risk of development of VTE after surgery using an endothelial function test. Because technical issues could influence the results of FMD, we used the RH-PAT to evaluate endothelial function, which is new, noninvasive and less operator-dependent, and well reflects metabolic risk factors.

Table 3. Reclassification by Adding the RHI to (A) DVT-Related Traditional Risk Factors and (B) the Qthrombosis Risk Score

| (A) Risk category by Traditional risk factors+RHI | New risk category by traditional risk factors+RHI |
|-----------------------------------------------|-----------------------------------------------|
| Patients without postoperative DVT           |                                              |
| Low risk                                     | 23                                            |
| Intermediate risk                            | 1                                             |
| High risk                                    | 0                                             |
| Patients with postoperative DVT              |                                              |
| Low risk                                     | 3                                             |
| Intermediate risk                            | 2                                             |
| High risk                                    | 0                                             |
| (B) Risk category by Qthrombosis risk score+RHI |                                              |
| Patients without postoperative DVT           |                                              |
| Low risk                                     | 0                                             |
| Intermediate risk                            | 22                                            |
| High risk                                    | 0                                             |
| Patients with postoperative DVT              |                                              |
| Low risk                                     | 0                                             |
| Intermediate risk                            | 6                                             |
| High risk                                    | 1                                             |

Low risk, <30%, intermediate risk 30–50%, high risk >50%. Abbreviations as in Tables 1,2.

(A) The net reclassification index was 16.0% (12 of 75 patients) for patients without postoperative DVT, 7.8% (4 of 51 patients) for patients with postoperative DVT, and 23.8% (95% CI: 3.5–44.1, P=0.022) overall.

(B) The net reclassification index was 16.7% (14 of 75 patients) for patients without postoperative DVT, 9.8% (5 of 51 patients) for patients with postoperative DVT, and 28.5% (95% CI: 4.9–52.1, P=0.018) overall.

Table 4. Logistic Regression Analysis for Development of DVT After THA or TKA According to Subgroups

| Subgroup | Patient underwent TKA* | Patient underwent THA** |
|----------|------------------------|-------------------------|
| RHI (per 0.1) | RHI (per 0.1) |
| Univariate    | 0.82                | 0.76                   |
| 95% CI     | 0.66–1.03           | 0.60–0.96             |
| P value   | 0.08                | 0.02                   |
| Multivariate (Model 1) | 0.79                | 0.74                   |
| 95% CI     | 0.61–1.02           | 0.54–1.00             |
| P value   | 0.068               | 0.052                  |
| Multivariate (Model 2) | 0.82                | 0.77                   |
| 95% CI     | 0.66–1.03           | 0.60–0.98             |
| P value   | 0.09                | 0.03                   |

Forced inclusion Model-1: adjusted for age, sex, height, weight, hypertension, dyslipidemia, diabetes mellitus, and current smoking. Model 2: adjusted for Qthrombosis risk score. Abbreviations as in Tables 1,2.

*No. of patients=62, no. of occurrence of DVT=38. **No. of patients=64, no. of occurrence of DVT=13.
The difference in the properties of FMD and RH-PAT is not completely understood and remains to be elucidated. FMD with occlusion at the forearm has been demonstrated to be mainly nitric oxide dependent. RH-PAT requires a proximal occlusion, and reflects changes in flow and digital microvessels dilatation, which is partly dependent on nitric oxide, and could be affected by other factors. Past studies, including the Framingham study, demonstrated that FMD and RH-PAT had different relationships with cardiovascular risk factors and were nearly uncorrelated with each other. FMD is particularly sensitive to being impaired by age and hypertension whereas RH-PAT is more sensitive to metabolic risk factors, especially body mass index, higher cholesterol, and diabetes mellitus. Exercise has been shown to improve FMD, but not RH-PAT in a recent study, albeit we also reported that lifestyle modification significantly improved RH-PAT in metabolic syndrome patients. This conflicting evidence highlights the need for further studies of the clinical and pathophysiological properties of the RH-PAT examination. In this study, the RHI showed a negative correlation with the Framingham risk score and was associated with postoperative DVT independently of the Framingham risk score, DVT-related traditional risk factors, and Qthrombosis risk score. We suspect that the RHI could be associated with VTE through cardiovascular disease risk factors, including unknown factors, and also through intrinsic properties of the venous endothelium. It has been reported that G>T polymorphism in the endothelial nitric oxide synthase gene is associated with endothelial dysfunction and the risk of VTE. Further studies are required to elucidate whether the RHI reflects these intrinsic venous endothelial properties.

In the present study, DVT was less common in smokers than in nonsmokers (7.1% vs. 44.6%, P<0.001), which is not consistent with past reports that have identified smoking as a risk factor for DVT. In the present study, however, smokers were significantly younger, with a higher percentage of males (age: 59 ±10.5 vs. 71.7±8.8 years, P<0.001, male: 7 (50.0%) vs. 11 (9.8%), P<0.001), which could alter the relationship between smoking and DVT. Smoking was not a significant predictor of DVT in either the multiple regression model or the age- and sex-adjusted model.

Study Limitations
First, the sample size was small, and it was a single-center study in Japan. In particular, the number of patients in the subgroup analyses was rather small. In patients with TKA, the RHI did not reach statistical significance, but tended to have an influence on postoperative DVT. The use of a tourniquet might cause local soft-tissue exposure, deep vein injury, and local circulatory disorder in patients with TKA, whereas a tourniquet was not used in patients with THA. Local factors caused by the tourniquet might account for the relatively low predictive value of the RHI in patients with TKA. Further study with a large number of patients is warranted to confirm our results. Second, the Qthrombosis score has not been validated in surgical patients, but there is no established risk score for VTE in such patients. Thus, we adopted the Qthrombosis score in this study as a reference. Third, we preferentially used ultrasonography for testing. Deep veins could not be visualized satisfactorily in 20 patients by ultrasonography, and they underwent phlebography. Fourth, although ultrasonography or phlebography was planned to be performed on postoperative day 8, postoperative evaluation of DVT was delayed in 25 patients. A delay in postoperative DVT evaluation potentially influences the incidence of DVT. In this study, the RHI was significantly associated with the incidence of postoperative DVT independent of a delay in postoperative DVT evaluation. Fifth, even if patients were asymptomatic, we performed further examination for PE in patients with proximal DVT. However, we did not routinely survey all of the study patients, and we cannot exclude the possibility of occult PE.

In conclusion, the present study demonstrated a significantly lower RHI in the DVT group compared with the non-DVT after lower limb arthroplasty, especially hip arthroplasty. Endothelial function, as assessed by the RHI, is potentially useful for the identification of patients at high risk for VTE after hip or knee arthroplasty.

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