

Department of Cardiology
Helsinki University Central Hospital
Helsinki, Finland

Venous thromboembolism: acute diagnostic assessment and follow-up

Mia Laiho

ACADEMIC DISSERTATION

To be presented, with the permission of the Faculty of the University of Helsinki,
for public examination in Auditorium room 3, Meilahti Hospital,
on 7th February 2014, at 12 noon.

Helsinki 2014

Supervisors

Docent Veli-Pekka Harjola
Department of Medicine, Division of Emergency Care
Helsinki University Central Hospital
Helsinki, Finland

MD Pirjo Mustonen
Department of Medicine, Division of Cardiology
Jyväskylä Central Hospital
Jyväskylä, Finland

Reviewers

Docent Matti Mänttari
Department of Medicine, Division of Cardiology
Helsinki University Central Hospital
Helsinki, Finland

Docent Heikki Ukkonen
Department of Medicine, Division of Cardiology
Turku University Central Hospital
Turku, Finland

Opponent

Professor h.c, Matti Halinen
Docent of Internal Medicine, Emeritus
University of Eastern Finland
Kuopio, Finland

ISBN 978-952-10-9700-3 (paperback)

ISBN 978-952-10-9701-0 (PDF)

<http://ethesis.helsinki.fi>

Helsinki University Printing House

Helsinki 2014

To my family

CONTENTS

| | |
|--|----|
| ABBREVIATIONS | 6 |
| LIST OF ORIGINAL PUBLICATIONS | 7 |
| ABSTRACT | 8 |
| 1. INTRODUCTION | 12 |
| 2. REVIEW OF THE LITERATURE | 13 |
| 2.1 Pathophysiology | 13 |
| 2.2 Risk factors of venous thromboembolism (VTE) | 14 |
| 2.3 Clinical findings and diagnosis of VTE | 15 |
| 2.3.1 Clinical presentation and assessment of clinical probability | 15 |
| 2.3.2 D-dimer assay | 18 |
| 2.3.3 Compression ultrasonography | 19 |
| 2.3.4 Arterial blood gas analysis | 19 |
| 2.3.5 Electrocardiogram | 20 |
| 2.3.6 Computed tomography | 20 |
| 2.3.7 Echocardiography | 21 |
| 2.3.8 Cardiac biomarkers | 22 |
| 2.3.9 Lung function parameters | 23 |
| 2.4 Prognostic stratification of PE | 23 |
| 2.5 Treatment of VTE | 26 |

| | |
|---|-----------|
| 2.5.1 Acute treatment of VTE | 26 |
| 2.5.2 Duration of anticoagulation | 28 |
| 2.6 Long-term sequelae and follow-up of VTE | 29 |
| 2.6.1 Post-thrombotic syndrome (PTS) | 29 |
| 2.6.2 Chronic thromboembolic pulmonary hypertension (CTEPH) | 30 |
| 3. AIMS OF THE STUDY | 32 |
| 4. PATIENTS AND METHODS | 33 |
| 5. RESULTS | 40 |
| 6. DISCUSSION | 47 |
| 7. SUMMARY AND CONCLUSIONS | 54 |
| 8. ACKNOWLEDGEMENTS | 55 |
| 9. REFERENCES | 57 |
| ORIGINAL PUBLICATIONS | 79 |

ABBREVIATIONS

ABBREVIATIONS

| | |
|----------------|---|
| APE | Acute pulmonary embolism |
| BNP | Brain natriuretic peptide |
| CEAP | Clinical, etiological, anatomical, pathophysiological |
| DL,CO | Diffusing capacity for carbon monoxide |
| CT | Computed tomography |
| CTPA | Computed tomography pulmonary angiography |
| CTEPH | Chronic thromboembolic pulmonary hypertension |
| CUS | Compression ultrasonography |
| DL | Diffusing capacity |
| DM | Diffusing capacity of alveolocapillary membrane |
| DVT | Deep venous thrombosis |
| ECG | Electrocardiogram |
| ELISA | Enzyme-linked immunosorbent assay |
| LMWH | Low molecular weight heparin |
| PE | Pulmonary embolism |
| POC | Point-of-care |
| PTS | Post-thrombotic syndrome |
| Rt-PA | Recombinant tissue plasminogen activator |
| RV | Right ventricular |
| RVD | Right ventricular dysfunction |
| TTE | Transthoracic echocardiography |
| VA | Alveolar volume |
| V _c | Pulmonary capillary blood volume |
| VC | Vital capacity |
| VTE | Venous thromboembolism |

LIST OF ORIGINAL PUBLICATIONS

This thesis is based on the following original publications, referred to in the text by their Roman numerals:

I

Von Lode P, Rainaho J, Laiho MK, Punnonen K, Peltola O, Harjola VP, Pettersson K. Sensitive and quantitative, 10 min immunofluorometric assay for D-Dimer in whole blood. *Thromb Res* 2006;118:573-85.

II

Laiho MK, Harjola VP, Graner M, Piilonen A, Raade M, Mustonen P. Helical computerized tomography and NT-proBNP for screening of right ventricular overload on admission and at long term follow-up of acute pulmonary embolism. *Scand J Trauma, Resusc Emerg Med* 2012;20:33.

III

Piirilä P, Laiho M, Mustonen P, Graner M, Piilonen A, Raade M, Sarna S, Harjola VP, Sovijärvi A. Reduction in membrane component of diffusing capacity is associated with the extent of acute pulmonary embolism. *Clin Physiol Funct Imaging* 2011;31:196-202.

IV

Laiho MK, Oinonen A, Sugano N, Harjola V-P, Lehtola A, Roth W-D, Lepäntalo M. Preservation of venous valve function after catheter-directed and systemic thrombolysis for deep venous thrombosis. *Eur J Vasc Endovasc Surg* 2004;28:391-6.

These publications have been reprinted with the permission of their copyright holders.

ABSTRACT

Introduction. Venous thromboembolism (VTE), including deep vein thrombosis (DVT) and pulmonary embolism (PE), is a common condition with significant rates of morbidity and mortality. VTE can lead to such complications as post-thrombotic syndrome (PTS) and chronic thromboembolic pulmonary hypertension (CTEPH). Accurate and prompt diagnosis of PE is the prerequisite for therapies to reduce the morbidity and mortality in PE. In recent years, guidelines have given diagnostic strategies based on clinical probabilities, D-dimer levels, ultrasonography of the legs, and computed tomography. The diagnostic tools have become more precise, and increasingly rapid diagnostic methods are being sought. However, late effects and consequences of VTE have rarely been the focus of interest in publications.

Aims of the study. We aimed to develop a sensitive, fully quantitative, and automated POC (point-of-care) testing assay for D-dimer in whole blood with the shortest possible assay time. A second aim was to determine the value of CT and NT-proBNP in detection of right ventricular dysfunction (RVD) in the acute phase and during follow-up of non-high-risk acute pulmonary embolism in clinical practice. Third, the membrane and pulmonary capillary blood volume components of diffusing capacity at the acute stage of PE was studied to identify which parameters are associated with right ventricular overload. Concerning the long-term effects of DVT, a fourth aim was to determine the prevalence of reflux patterns in patients with iliofemoral venous thrombosis 2-3 years after systemic or catheter-directed thrombolysis.

Patients and methods. The study consists of four projects.

1. The blood samples of patients (n=525) and healthy volunteers (n=101) were involved. The plasma samples were analyzed either fresh or stored at -20°C or -70°C until used. The simple, automated assay procedure comprised a 1:50 sample dilution, one-step incubation, washing, and time-resolved fluorometric measurement directly from the wet assay wells. The Roche Diagnostic TinaQuant® D-dimer (Knecht MF et al. 1997) and the Biopool

ABSTRACT

Auto Dimer ® (Bozic M et al. 2003) assays were used as comparison methods in the study.

2. Sixty-three non-high-risk APE patients were studied. RVD was assessed at admission to the emergency department by computed tomography (CT), transthoracic echocardiography (TTE), cardiac biomarkers (NT-proBNP and cardiac troponin T), blood gases, electrocardiogram (ECG), and clinical evaluation. TTE, cardiac biomarkers, and clinical evaluation were repeated 7 months later.

3. Forty-seven patients with acute non-high-risk PE confirmed by CT were included. The extent of PE was assessed by scoring mass of embolism. Measurements of diffusing capacity for carbon monoxide (DL_{CO}), diffusing capacity of alveolocapillary membrane (DM), pulmonary capillary blood volume (V_c), and alveolar volume (V_A) were measured at the acute event and 7 months later. RVD was evaluated with TTE and ECG. Fifteen healthy subjects were included as controls.

4. Thirty-seven patients with ileofemoral deep venous thrombosis treated with either systemic or catheter-directed thrombolysis were evaluated 2-3 years after the thrombotic event. The evaluation was performed with CEAP (clinical, etiological, anatomical, pathophysiological) classification and color duplex ultrasound. Patients were also asked to fill in the questionnaire concerning risk factors, duration of anticoagulation therapy, and use of compression stockings.

Results.

1. Achieving the steady-state in the one-step assay took 60 min. Of the steady-state values approximately 45% were reached in 10 minutes incubation time. The analytical detection limit of the assay was 0.046 mg/l (calculated as the mean background signal +3 SD). The within- and between-assay imprecision was similar for whole-blood and plasma samples, ranging from 8.3% to 13% below and from 3.8% to 7.7% above the cut-off level of 0.6 mg/l. The within- and between-assay imprecision was also calculated for commercial D-dimer controls from Bio-Rad and Biopool and showed a corresponding imprecision. When using the cut-off values of 0.6 and 1.0 mg/l, clinical sensitivities and specificities were 98.7% and 64.4%, and 62.2 % and 81.0%, respectively.

2. Right ventricular overload was detected with CT in 37 (59%) of 63 patients. Twenty-eight patients (76%) in the RVD-positive group also had signs of RVD in TTE. Nine of the RVD-positive patients (24%) showed no echocardiographic alterations in RV

ABSTRACT

morphology or function. Altogether findings of RVD in CT were congruent with those in echocardiography ($p < 0.0001$). Patients who had RVD in CT had elevated NT-proBNP relative to patients with no RVD (87% vs. 27%, $p < 0.0001$). In the RVD-positive group, 32 (86.5%) of 37 patients had signs of RVD in ECG, whereas 8 patients (13%) had such findings in the RVD-negative group ($p < 0.0001$). In the 7-month follow-up, RVD was detected in 6 (10%) of 63 patients. These patients had also elevated NT-proBNP levels ($p < 0.0001$). All patients who were found to have persistent RVD had RVD in both CT and TTE on admission.

3. DL,CO, DL, DM, vital capacity (VC) and VA were lower in patients with acute PE than in healthy controls ($p < 0.001$). DM correlated inversely with central mass of embolism ($r = -0.312$; $p = 0.047$), whereas Vc did not. DM, DL,CO, VC, and VA improved significantly within 7 months. In all patients ($p = 0.001$, $p = 0.001$) and persistent RVD patients ($p = 0.020$, $p = 0.012$), DM, DL,CO remained significantly lower than in healthy controls at follow-up. DM was inversely related to central mass of embolism.

4. In both catheter-directed thrombolysis and systemic lysis-treated patient groups, 38% of the patients were using warfarin permanently at the follow-up visit. In the rest of the patients in both groups, the mean duration of warfarin treatment was 8 months. Risk factors, including genetic defects, were found in 50% of catheter-directed and 75% of systemic-treated patients ($p = 0.25$). Fifty percent of catheter-directed lysis and 38% of systemic lysis-treated patients had used or were still using compression stockings ($p = 0.41$). Valvular competence was preserved in 44% of patients treated with catheter-directed thrombolysis compared with 13% of those treated systemically ($p = 0.01$). Any deep reflux was present in 44% of catheter-directed lysis-treated patients compared with 81% of systemic-treated patients ($p = 0.03$). Any superficial reflux was observed in 25% and 63% of the patients, respectively ($p = 0.03$). Patients treated with catheter-directed thrombolysis tended to have a better clinical status based on evaluation by CEAP classification at follow-up.

Conclusions

1. The rapid and sensitive 10-min D-dimer assay seems to be suitable for excluding VTE in POC settings. Based on the high preliminary sensitivity and negative predictive value obtained here, the assay could potentially be used as a stand-alone test given that a sufficiently low cut-off level was selected. Alternatively, combined with clinical

ABSTRACT

probability assessment, a higher cut-off level could be used to increase the specificity of the assay.

2. The study of 63 patients with APE shows that CT is a practical and valuable imaging method for detection of RVD in non-high-risk APE patients. All patients found to have RVD at the 7-month follow-up had RVD also in the acute phase CT and TTE. According to these results, a follow-up protocol is suggested. Patients with RVD on admission CT should be rescreened with natriuretic peptide assessment at follow-up and if elevated an echocardiography should be performed. Timing for screening is reasonable at the visit where warfarin is to be discontinued. Alternatively, if anticoagulation therapy is deemed continuous at discharge, then biomarker screening might be done during a routine follow-up visit 6-12 months after APE. The suggested protocol needs to be validated in a larger patient population.

3. The extent of PE and subsequent signs of RVD were associated with a decrease in diffusion capacity of membrane in both the acute phase and in the resolution phase of PE independent of lung volume changes. In patients with permanent RVD, also the total diffusing capacity remained decreased in the follow-up. The diffusing capacity recovered during the 7-month follow-up, but did not reach the level of healthy controls or the normal reference range. Study data provide new knowledge of consequences of PE on lung function, indicating that sustained reduction in diffusing capacity may remain despite treatment of PE according to current guidelines.

4. Catheter-directed thrombolysis for DVT at the acute event reduced both deep and superficial reflux at the 2- to 3-year follow-up.

1. INTRODUCTION

Venous thromboembolism (VTE) is a multifactorial disease with both environment- and genetic-related risk factors. VTE presents clinically as deep venous thrombosis (DVT) and pulmonary embolism (PE), with serious outcome in both men and women. PE is the third most common cause of death from cardiovascular disease after heart attack and stroke (Naess IA et al. 2007).

Over the past two decades, new diagnostic and prognostic tools (e.g. D-dimer, computer tomography, cardiac biomarkers) have been developed, improving the care of patients with suspected or diagnosed VTE. Clinical probability assessment nowadays helps to identify patients with a high or intermediate risk and those in need of anticoagulation treatment while awaiting the results of the tests. Low-risk patients can be identified as well. The probability can be assessed with prediction rules or scores during the acute phase.

VTE can be diagnosed by various cost-effective diagnostic algorithms in combination with non-invasive imaging techniques. Pharmacological and non-pharmacological interventions can be used to prevent and manage these conditions. The combination of both interventions may ease the process of prevention and treatment of VTE (Moheimani F et al. 2011).

There are several earlier studies examining short-term effects of PE. A paucity of literature also exists concerning long-term course of PE (Carson JL et al. 1992, Ribeiro A et al. 1999, Sharma GV et al. 2000, Poulsen SH et al. 2001, Pengo V et al. 2004, Becattini G et al. 2005), and most of these studies have investigated mortality as the study endpoint.

In spite of the uncomplicated clinical course of most VTE patients, VTE also can cause significant complications, including recurrent thrombotic events, PTS, and CTEPH. PTS can be seen in about one-third of DVT patients (Prandoni P et al. 1996). Pulmonary hypertension, in turn, is detected in 4-5% of PE sufferers (Pengo V et al. 2004). Early identification of patients at risk for CTEPH is important so that these patients can be referred for further evaluation of relevant diagnostic and therapeutic strategies.

2. REVIEW OF THE LITERATURE

2.1 Pathophysiology

PE and DVT are parts of the same process, venous thromboembolism. DVT can be found in about 70% of patients with PE. Rarely, the source of emboli can be upper extremity veins, iliac veins, renal veins, or right heart (Riedel 2001). The mortality of untreated PE is estimated to be approximately 30%; adequate treatment reduces this figure to 2-8% (Goldhaber et al. 1999).

The annual incidence of VTE is 100 persons per 100 000 in the United States, rising exponentially with age. Despite anticoagulation therapy, the 6-month recurrence rate of VTE was approximately 6% in the California Patient Discharge data set (Murin S et al. 2002). Death occurs in 6% of DVT patients and 12% of PE patients within one month of diagnosis, and early mortality after VTE diagnosis is strongly associated with presentation as PE, advanced age, cancer, and underlying cardiovascular disease (White RH 2003). The risk of fatal PE is 2-to 3-fold higher after an episode of PE than DVT (Douketis JD et al. 1998, Heit JA et al. 1999, Murin S et al. 2002).

Hemodynamic consequences of PE appear when over 30-50% of the pulmonary arterial bed is occluded by thromboemboli (McIntyre KM et al. 1971). The effects of an embolus depend on the extent to which it obstructs the pulmonary circulation, the localization over which the obstruction accumulates, and the pre-existing condition of the patient. As a result, pulmonary hypertension may develop disproportionate to the amount of vasculature that is mechanically occluded. Most emboli are multiple. As both the extent and chronicity of obstruction vary widely, PE can produce markedly differing clinical pictures (Riedel M 2001).

2.2 Risk factors of venous thromboembolism (VTE)

The proportion of patients with idiopathic or unprovoked PE was about 20% in the International Cooperative Pulmonary Embolism Registry (ICOPER) (Goldhaber SZ et al 1999). A prior episode of lower extremity DVT is the greatest risk factor for a subsequent episode of DVT (Kakkar VV et al. 1970). VTE can be seen as a result of a interaction between patient-related and setting-related risk factors (Heit JA et al. 2002, Alikhan R et al. 2004). Setting-related predisposing factors are quite often temporary, whereas patient-related factors are usually permanent.

Predisposing factors for VTE are listed in Table 1. It is important to recognize that the predictive values of predisposing factors are not equal. In assessing the need for prophylaxis, both the strength of individual risk factors and the cumulative weight of all risk factors should be considered.

Table 1. Risk Factors for VTE (adapted from Anderson et al. 2003).

| |
|--|
| Strong risk factors (odds ratio >10) |
| Fracture (hip or leg) |
| Hip or knee replacement |
| Major general surgery |
| Major trauma |
| Spinal cord injury |
| Moderate risk factors (odds ratio 2–9) |
| Arthroscopic knee surgery |
| Central venous lines |
| Chemotherapy |
| Congestive heart or respiratory failure |
| Hormone replacement therapy |
| Malignancy |
| Oral contraceptive therapy |
| Paralytic stroke |
| Pregnancy, postpartum |
| Previous venous thromboembolism |
| Thrombophilia |
| Weak risk factors (odds ratio <2) |
| Bed rest >3 days |
| Immobility due to sitting (e.g. prolonged car or air travel) |
| Increasing age |
| Laparoscopic surgery (e.g. cholecystectomy) |
| Obesity |
| Pregnancy, antepartum |
| Varicose veins |

A spectrum of inherited risk factors, thrombophilia, contributes to VTE (De Stefano et al. 1996, Anderson FA 2003, Rosendaal FR et al. 2009). They are also broken down into strong, medium, and weak genetic risk factors (Roosendaal FR et al. 2009). Deficiencies of some natural coagulation inhibitors, including antithrombin, protein C, and its cofactor protein S, insufficiency in such anticoagulants as tissue factor pathway inhibitor, thrombomodulin, and endothelial protein C receptor, and an elevated level of factor VIII are examples of strong genetic risk factors. Moderate genetic risk factors consist of a mutation in factor V Leiden, causing resistance to activated protein C (APC resistance), and a mutation in part of the prothrombin gene (prothrombin 20210A) (Moheimani F et al. 2011).

Most hospitalized patients should receive pharmacological prophylaxis to minimize the risk of VTE (Cohen AT et al. 2008, Dentali F et al. 2007, Guyatt GH et al. 2012, Tapson VF et al. 2007). Although a large number of medical and surgical patients have risk factors for DVT, prophylaxis often remains underused. In the multicenter cross-sectional ENDORSE study, 52% of the enrolled patients were at moderate risk for developing VTE. Surgical patients received more often prophylaxis than medical patients, 58% vs. 40% (Cohen et al 2008). It also has been proposed that additional lifestyle risk factors, such as cigarette smoking, overweight, metabolic syndrome, hypertension, high red meat consumption, and hyperlipidemia, should be considered in preventing both myocardial infarction and VTE (Goldhaber SZ 2010). An association has been shown between atherosclerotic disease and spontaneous venous thrombosis (Prandoni P et al. 2003). Shared risk factors and pathophysiology help to understand the increased risk of VTE in patients with atherothrombosis and the greater frequency of atherothrombotic events in patients with VTE (Piazza G et al. 2010).

2.3 Clinical findings and diagnosis of VTE

2.3.1 Clinical presentation and assessment of clinical probability

Notably, three-quarters of patients who present with suspected DVT have non-thrombotic causes of leg pain (O'Donnell T et al. 1980). DVT most often initiates in the calf veins. The majority of distal DVT likely resolves spontaneously with no symptoms; patients

more often become symptomatic when distal DVT extends to the popliteal and femoral veins and other proximal veins (Kearon C et al. 2003).

Commonly reported symptoms in patients with suspected DVT include leg pain, swelling, and other signs such as pitting edema, warmth, dilated superficial veins, and erythema (Haeger K et al. 1969, O'Donnell T et al. 1980, Hull RD et al. 1984).

The clinical symptoms and signs of DVT and PE are often non-specific. A clinical model for predicting probabilities for DVT has been suggested, where different clinical features can be scored (Wells PS et al. 1997, Le Gal G et al. 2006). By using these rules, the proportion of PE is 10% in the lower and 30-65% in the higher clinical probability category (Torbicki A et al. 2008). About two-thirds of patients presenting with suspected DVT or PE do not have these conditions. Sometimes PE presents in such dramatic way that the diagnosis is obvious and treatment is started immediately (Riedel M 2001). Typical symptoms of PE are dyspnea, chest pain, cough, hemoptysis, and syncope. Clinical signs and symptoms are not, however, very useful since they are not sensitive or specific for PE (Torbicki A et al. 2008).

Clinical probability assessment aims to identify patients with a high or intermediate clinical probability of needing immediate treatment. In patients with a low clinical probability, VTE can be ruled out solely with a normal D-dimer test (Figure 1).

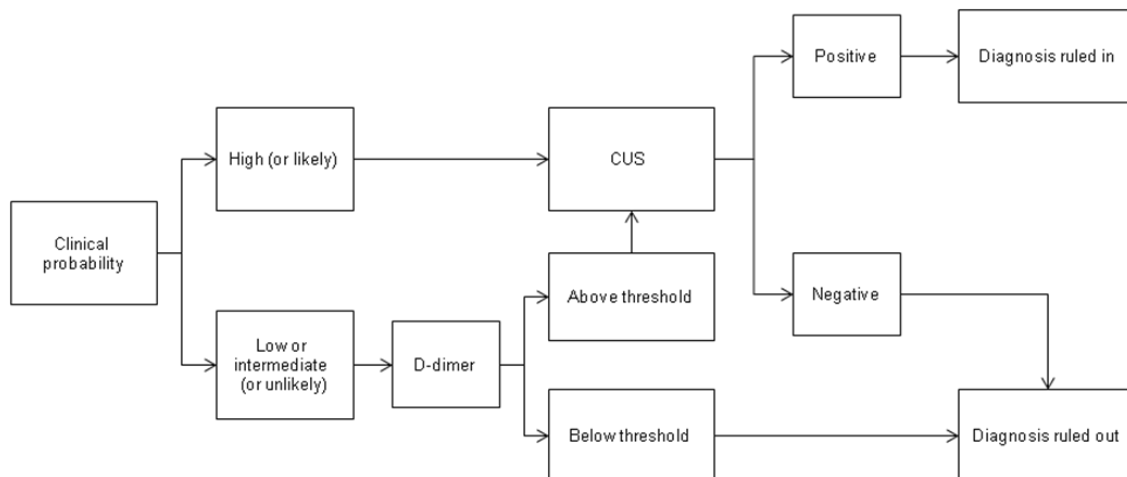


Figure 1. A diagnostic algorithm for clinically suspected DVT or PE.

Use of compression ultrasonography (CUS) with suspected DVT (adapted from Goldhaber et al. 2012).

REVIEW OF THE LITERATURE

Clinical probability includes clinical history as well as symptoms, signs, and abnormalities of oxygen saturation, chest radiography, and ECG. There are two widely used scores: the Wells prediction score (Wells PS et al. 2000) and the revised Geneva scores (Le Gal G et al. 2006). Scoring systems can help to assess the probability of suspected DVT or PE (Table 2).

Table 2. *Clinical probability assessment*

| | Points |
|--|--------|
| Wells score for DVT* | |
| Cancer | +1 |
| Paralysis or recent plaster cast | +1 |
| Bed rest >3 days or surgery <4 weeks | +1 |
| Pain on palpation of deep veins | +1 |
| Swelling of entire leg | +1 |
| Diameter difference on affected calf >3 cm | +1 |
| Pitting oedema (affected side only) | +1 |
| Dilated superficial veins (affected side) | +1 |
| Alternative diagnosis at least as probable as DVT | -2 |
| Wells score for PE† | |
| Previous PE or DVT | +1.5 |
| Heart rate >100 beats per min | +1.5 |
| Recent surgery or immobilisation | +1.5 |
| Clinical signs of DVT | +3 |
| Alternative diagnosis less likely than PE | +3 |
| Haemoptysis | +1 |
| Cancer | +1 |
| Revised Geneva score for PE‡ | |
| Age >65 years | +1 |
| Previous DVT or PE | +3 |
| Surgery (under general anaesthesia) or fracture (of the lower limbs) within 1 month | +2 |
| Active malignancy (solid or haematological malignancy, currently active or considered as cured since less than 1 year) | +2 |
| Unilateral leg pain | +3 |
| Haemoptysis | +2 |
| Heart rate 75–94 beats per min | +3 |
| Heart rate ≥95 beats per min | +5 |
| Pain on deep vein palpation in leg and unilateral oedema | +4 |

Scoring systems used to assess probability of suspected DVT or PE on the basis of items and assigned points. DVT=deep vein thrombosis. PE=pulmonary embolism.

** Patients with a score of 0 are low risk, 1–2 are intermediate risk, and ≥3 are high risk.*

† For the initial rule, patients with a score of 0–1 are low risk, 2–6 are intermediate risk, and ≥7 are high risk; for the dichotomized rule, patients are unlikely or likely to have PE if they have scores ≥4 and ≤4, respectively.

‡ Patients with a score <2 are low risk, 2–6 are intermediate risk, and ≥6 are high risk (with permission of Elsevier, Goldhaber SZ et al. 2012).

The evidence strongly supports the use of clinical prediction rules for establishing the pretest probability of DVT or PE in a patient before more definitive testing. The pretest probability can then be used in interpreting subsequent test results. The use of D-dimer assay with a clinical prediction rule may even have a negative predictive value high enough to obviate the need for imaging studies in many low-risk patients (Segal JB et al. 2007).

2.3.2 D-dimer assay

D-dimer is a degradation product of cross-linked fibrin. It is elevated in conditions where fibrin is formed and degraded (VTE, surgery, trauma, hemorrhage, ischemic heart disease, cerebrovascular event, infections, malignancy, peripheral arterial disease and aneurysms, pregnancy, advanced age of patients, extensive burns) (Palareti G 2006, Adam SS 2009). There are a numerous D-dimer assays available nowadays (Stein PD 2004, Di Nisio M 2007).

ELISA and ELISA-derived assays have a sensitivity of over 95% and specificity of approximately 40%. A negative ELISA can rule out PE in up to 30% of patients (Perrier A et al. 1999, Wells PS et al. 2001, Perrier A et al. 2004).

D-dimer assays have restricted specificity and are less useful than other measures in some patient groups, including patients with high clinical probability, patients admitted to hospital for some other reason but in whom the suspicion of PE is raised during hospital stay, patients older than 65 years, and pregnant women (Righini M et al. 2005, Righini M et al. 2008). A normal D-dimer assay can be used in low-to high-risk patients as a reliable tool for the exclusion of VTE (Stein PD et al. 2004, Adam SS et al. 2009).

In clinical diagnostics, there are two ways to organize laboratory services: automation and centralization in central laboratories (Hoffmann GE et al. 1998, Wheeler MJ 2001) or testing in local laboratories near the patients (St-Louis P. 2000, Price CP et al. 2004). The total process of clinical testing using a central laboratory often contains many additional steps. In answer to the growing need for rapid diagnostics, point-of-care testing (POCT) has been developed in recent years. POCT is defined any testing near the patient (St-Louis P 2000, Price CP et al. 2004). It is performed by personnel with minimal practice and experience in laboratory procedures. The tests are mainly available within 2-20 minutes. The settings for the POCT include primary care settings, ambulances and other transport

settings, emergency units, and other hospital units (Hicks JM et al. 2001, Hardy RW et al. 2004, Kirkbride RE 2004). In a study of diagnostic accuracy of POCT for thromboembolic events in primary care, the D-dimer assay showed value in excluding VTE (Tomonaga Y et al. 2011). POCT can be more expensive than testing performed in a central laboratory. Nevertheless, POCT technologies can improve emergency department measures such as length of stay (Lee-Lewandrowski E et al. 2009).

2.3.3 Compression ultrasonography

Compression ultrasonography (CUS) as a simple, non-invasive procedure is the method of choice in evaluating lower limb veins for DVT (Fraser JD et al.1999, Palareti G et al. 2006, Tan M et al. 2009). CUS has a sensitivity of over 90% for proximal DVT and a specificity of about 95% (Perrier A et al.1998, Kearon C et al. 1998). Use of ultrasound in diagnosing symptomatic thrombosis in the proximal veins of the lower limb is recommended for patients whose pretest probability of disease falls in the category of high risk of DVT under the Wells prediction rule. Ultrasound is less sensitive in patients who have DVT limited to the calf veins; the sensitivity of venous ultrasound is only 73% (Kearon C et al. 1998). Therefore, a negative ultrasound does not rule out DVT in these patients. A repeated ultrasound may be required for patients who have suspected calf vein DVT and a negative ultrasound and for patients who have suspected proximal DVT and an ultrasound that is technically inadequate or equivocal (Qaseem A et al. 2007).

2.3.4 Arterial blood gas analysis

Arterial blood gas analysis is one of the clinical examinations in patients with suspected PE. It can help in evaluation of gas exchange and acid-base status of a suspected PE patient. Hypoxemia or hypocapnia in arterial blood gas analysis can be detected, but results may also be normal, especially in younger patients without previous cardiopulmonary disease (Stein PD 1991). PE is usually associated with hypoxemia, but 20% of patients with PE have a normal arterial oxygen pressure (Stein PD et al. 1996). In the setting of a normal or near-normal chest radiograph and significant unexplained hypoxemia, PE should be considered (Tapson VF et al. 2012). In conclusion, blood gases

may heighten the suspicion of PE and contribute to the clinical assessment, but they are of insufficient discriminative value to confirm or exclude the diagnosis of PE.

2.3.5 Electrocardiogram

The electrocardiogram (ECG) can show in patients with APE inverted T-waves in leads V1-V4, QR-pattern in lead V1, S1Q3T3 pattern, and incomplete or complete right bundle-branch block. Especially if the findings are new at onset of APE, ECG can be helpful (Rodger M et al. 2000, Geibel A et al. 2005). However, ECG findings in APE are non-specific (Rodger M et al. 2000). The S1Q3T3 pattern may be present on the ECG, but it is non-specific. With extensive emboli, a right ventricular strain pattern may be present, which also can be considered with regard to determining the level of aggressiveness of therapy in proven PE (Tapson VF 2012).

2.3.6 Computed tomography

Computed tomography pulmonary angiography (CTPA) has become the first-line examination for the detection of APE (Costello P et al. 2000, Schoepf UJ et al. 2002, Schoepf UJ et al. 2003, Schoepf UJ et al. 2004). CTPA is available in most hospitals nowadays, and it is fast and easy to perform (British Thoracic Society Standards of Care Committee Pulmonary Embolism Guideline Development Group 2003). New-generation CT scans are also able to visualize subsegmental arteries (Raptopoulos V et al. 2001, Schoepf UJ et al. 2002). While multidetector CT is quite sensitive, small, subsegmental emboli are sometimes difficult to visualize. If a study is suboptimal or if findings are uncertain, additional ultrasound of the leg or lung imaging should be considered (Tapson VF 2012). The quantitative assessment of CTPA correlates well with clinical severity (Bankier AA et al. 1997, Mastora I et al. 2003). CTPA is also useful in demonstrating alternative diagnoses when PE is excluded. Furthermore, several studies have suggested that CTPA is helpful in detecting RVD (Araoz PA et al. 2003, Schoepf UJ et al. 2004, WU AS et al. 2004, Quiroz R et al. 2004, Van der Meer RW et al. 2005, Ghaye P et al. 2006, Araoz PA et al. 2007). Signs of RVD include a high ratio of the right ventricle to the left ventricle on CTPA (Reid JH et al. 1998, Contractor S et al. 2002, Araoz PA et al. 2002), a

bowing interventricular septum, and reflux of contrast medium into the inferior vena cava (Collomb D et al. 2003, Aviram G et al. 2008).

2.3.7 Echocardiography

RVD has been consistently detected with TTE in approximately 50% of APE patients (Kasper W et al. 1997, Ribeiro A et al. 1997, Goldhaber SZ 1999, Kreit JW et al. 2004). Echocardiographic criteria used for RVD have varied in different studies. The most commonly used qualitative echocardiographic assessment of RVD is wall motion (Goldhaber SZ et al. 1993, Wolfe MW et al. 1994, Ribeiro A et al. 1997, Goldhaber SZ et al. 1999), which is evaluated as normal or mildly, moderately, or severely hypokinetic. The basic quantitative criterion is RV dilatation (Kasper W et al. 1997, Grifoni S et al. 2000). The presence of pulmonary hypertension has also been applied as a diagnostic criterion (Kasper W et al. 1997, Ribeiro A et al. 1997, Grifoni S et al. 2000). Continuous wave Doppler is used to estimate the pulmonary artery systolic pressure by measuring the peak velocity of the tricuspid valve and pulmonary valve. The gradient across the tricuspid valve can be calculated by using the modified Bernoulli formula (Dabestani A et al. 1987). Estimated right atrial pressure is added to the gradient to estimate pulmonary artery pressure. Hepatic venous flow can also be used to estimate right atrial pressure (Ommen SR et al. 2000). A relatively new method is the Doppler-derived index (TEI index) to assess overall RV function. The index is defined as the isovolumic contraction time and isovolumic relaxation time divided by the ejection time (Tei et al. 1996). An increased RV TEI index is associated with either LV diastolic abnormalities or pulmonary hypertension (Tei et al. 1995, Giunta A et al. 2000, Vonk MC 2007).

Many authors have based the diagnosis of RVD on various combinations of these findings. It is unknown which of these criteria is the most sensitive indicator of PE-induced RVD since data on the issue are limited (McConnell MV 1996, Kurzyna M et al. 2002, Kreit JW et al. 2004). In any case, peak of tricuspidal gradient is the most commonly used method to assess pulmonary artery systolic pressure in clinical practice (Chan KL et al. 1987).

In patients in critical condition with shock or hypotension, bedside TTE can help in making management decisions. The absence of signs of RVD excludes PE as a cause of hemodynamic instability. In these clinical situations, TTE can be helpful in differential

diagnosis of other causes of shock by, for example, detecting cardiac tamponade, acute valvular dysfunction, or acute myocardial infarction. On the other hand, if there are signs of RVD in a hemodynamically stable patient with suspected PE, echocardiographic diagnostic tools can justify aggressive treatment because of the patient's critical condition. The main role of TTE in non-high-risk PE is further prognostic stratification to the intermediate- or low-risk category (Torbicki A et al. 2008).

2.3.8 Cardiac biomarkers

Increased myocardial stretch leads to the release of brain natriuretic peptide (BNP) and cardiac troponins. High levels of BNP, pro-BNP, and cardiac troponin have been associated with an elevated risk of death in patients with APE (Kostrubiec M et al. 2005, Becattini C et al. 2007). Elevated natriuretic peptides BNP and NT-proBNP are predictive of adverse short-term outcome in patients with APE (Tulevski II et al. 2001, Kucher N et al. 2003, Pruszczyk P et al. 2003, Ten Wolde M et al. 2003, Krüger S et al. 2004, Pieralli F et al. 2006, Kostrubier M et al. 2007, Alonso-Martinez II et al. 2009). However, although elevated BNP and pro-BNP levels are associated with worse outcome, their positive predictive value is quite low (12-26%) (Perrier A et al. 1999, Kruip MJ et al. 2002, Perrier A et al. 2004, Van Belle A et al. 2006). Low levels of BNP or NT-pro-BNP, by contrast, can be utilized for identification of patients with a good prognosis concerning short-term mortality or clinical outcome (Kucher N et al. 2003, Pruszczyk P et al. 2003, Ten Wolde M et al. 2003, Binder L et al. 2005, Kostrubier M et al. 2007). A systematic review of evaluating the accuracy of BNP or NT-proBNP for the diagnosis of RVD has indicated that BNP and NT-proBNP are associated with the diagnosis of RVD in patients with acute PE and are significant predictors of all-cause in-hospital or short-term mortality in these patients (Cavallazzi et al. 2008). The results of another meta-analysis concerning troponin-based risk stratification showed that the prognostic value of troponin levels in normotensive APE patients depends greatly on the cut-off points used (Jimenez D et al. 2009).

Some novel biomarkers, such as heart-type fatty acid-binding protein (H-FABP), an early marker of myocardial injury, have also been demonstrated to have prognostic value (Kaczynska A et al. 2006, Puls M et al. 2007, Dellas C et al. 2010).

2.3.9 Lung function parameters

APE has been shown to be associated with decreased pulmonary diffusing capacity, reflecting deterioration of gas exchange capacity of the lungs (Sharma GV et al. 1980). Lung diffusing capacity refers to the ability of the lungs to transfer gases between the alveolar and pulmonary capillary compartments and is most frequently quantified by the rate of uptake of carbon monoxide (CO) during breath-holding or rebreathing maneuvers (Sackrer M et al. 1975, Meyer M et al. 1990, Hsia CC et al. 2002, Stam H et al. 1983, Snyder EM et al. 2005).

The simplified model described by Roughton and Forster is widely accepted as the standard for quantifying lung diffusing capacity. It describes a series of resistances: the diffusion of the gas across the alveolocapillary membrane, the transfer into the plasma and across the red blood cell membrane, and the chemical reaction of the gas with hemoglobin (Roughton FJ et al. 1957). Measurement of diffusing capacity (DL) is informative for pathophysiologic diagnoses of lung diseases, and serial measurements are used to follow the course of disease (Macintyre N et al. 2005). A few studies have attributed the low DL to a loss of diffusing capacity of alveolocapillary membrane (DM) rather than vital capacity (Vc) (Bernstein RJ et al. 1996, Steenhuis LH et al. 2000). The decreased DL in association with decreased DM and Vc has been described in chronic heart failure patients in stable clinical condition (Agostoni P et al. 2006). There is also growing evidence that left ventricular diastolic function may become independently compromised in pulmonary artery hypertension (PAH) (Puwanant S et al. 2010 Tonelli AR et al. 2012). This may contribute to the reduced DL, DM, and Vc. In a recent study with PAH patients, findings indicated that lower than normal gas transfer in PAH is due to loss of both DM and Vc, but deterioration of DM/Vc over time is related to worsening membrane diffusion (Farha S et al. 2013).

2.4 Prognostic stratification of PE

Acute treatment of PE is planned in accordance with mortality risk based on prognostic stratification. Both ESC (European Society of Cardiology) and ACCP (American College of Chest Physicians) guidelines suggest evaluating treatment of PE according to clinical

REVIEW OF THE LITERATURE

risk (Kearon MB et al. 2008, Torbicki A et al. 2008). Risk markers useful for risk stratification can be classified into three groups: clinical markers (shock, hypotension), markers of RVD, and markers of myocardial injury. Clinical assessment based on these clinical markers allows stratification into high-risk and non-high-risk PE. Classification can be utilized in evaluating the choice of optimal diagnostic strategy and initial treatment (Torbicki A et al. 2008). High-risk patients need an immediate diagnostic and treatment strategy since the short-term mortality rate in this group is over 15% (Kasper W et al. 1997). Non-high-risk patients are normotensive. As normotensive patients often present with RVD, non-high-risk PE patients can be further stratified according to the presence of RVD into intermediate- and low-risk PE (Torbicki et al. 2008). Moreover, accurate and objective models of prognosis could help clinicians to determine the appropriateness of early hospital discharge or complete ambulatory treatment for patients with acute symptomatic PE. The Pulmonary Embolism Severity Index estimates the risk of 30-day mortality in patients with acute PE. The Pulmonary Embolism Severity Index and its simplified version (Table 3) (Aujesky D et al. 2005, Jimenez D et al. 2010) can also be helpful in evaluating stratification on a clinical basis.

Table 3. *The Pulmonary Embolism Severity Index and its simplified version (modified from Aujesky D et al. 2005, Jimenez D et al. 2010).*

| Variable | Score | |
|---|----------------------------|------------------------------|
| | Original PESI ^a | Simplified PESI ^b |
| Age > 80 years | Age in years | 1 |
| Male sex | +10 | |
| History of cancer | +30 | 1 |
| History of heart failure | +10 | 1 ^c |
| History of chronic lung disease | +10 | |
| Pulse ≥ 110 beats/min | +20 | 1 |
| Systolic blood pressure < 100 mm Hg | +30 | 1 |
| Respiratory rate ≥ 30 breaths/min | +20 | |
| Temperature < 36°C | +20 | |
| Altered mental status | +60 | |
| Arterial oxyhemoglobin saturation level < 90% | +20 | 1 |

a) Total point score for a given patient is obtained by summing the patient's age in years and the points for each predictor present. Scores correspond to the following risk classes: 65 or less, class I; 66 to 85, class II; 86 to 105, class III; 106 to 125, class IV; and more than 125, class V. Patients in risk classes I and II are defined as being at low risk.

REVIEW OF THE LITERATURE

b) Total point score for a given patient is obtained by summing the points. Scores correspond to the following risk classes: 0, low risk; 1 or more, high risk. Empty cells indicate that the variable was not included.

c) Variables (history of heart failure and chronic lung disease) were combined into a single category of chronic cardiopulmonary disease.

Different treatment procedures exist for patients with PE. High-risk patients (representing about 5% of all symptomatic patients) should be treated aggressively with thrombolytic agents or embolectomy (Kucher N et al. 2006). Low-risk patients (most patients with PE) might benefit from early discharge or even outpatient treatment (Aujesky D et al. 2011). Intermediate-risk patients (about 30% of all symptomatic patients) should probably be admitted to hospital and may benefit from thrombolytic agents. Recent results from the Hestia Study also suggest that certain APE patients could be selected for outpatient treatment with predefined criteria (Zondag W et al. 2011). For patients with low-risk PE and adequate home circumstances, early discharge was recommended in recent ACCP guidelines (Guyatt GH et al. 2012).

Echocardiography or elevated levels of troponin or pro-brain natriuretic peptide might aid in prognostic stratification (Agnelli G et al. 2010, Goldhaber SZ et al. 2012).

In recent years, studies involving multiple prognostic variables (so-called hybrid studies) demonstrate that combinations of RVD, elevated natriuretic peptides, or elevated troponin indicate adverse prognosis (Binder L et al. 2005, Kostrubier M et al. 2005, Scridon T et al. 2005, Hsu IT et al. 2006, Kline JA et al. 2006, Logeart D et al. 2007, Maziere F et al. 2007, Tulevski II et al. 2007, Jimenez D et al. 2008, Kline JA et al. 2008, Palmieri V et al. 2008, Bova C et al. 2009, Vuilleumier N et al. 2009). Although the techniques as described have utility for predicting prognosis in acute PE, clinical judgment is required to determine which of these is appropriate for each patient. American Heart Association (Jaff M et al. 2011) has proposed a definition for submassive PE: acute PE without systemic hypotension (blood pressure ≥ 90 mmHg) but with either RVD or myocardial necrosis. RVD refers to the presence of at least one of the following:

1. RV dilatation (apical 4-chamber RV diameter divided by LV diameter >0.9) or RV systolic dysfunction on echocardiography
2. RV dilatation (4-chamber RV diameter divided by LV diameter >0.9) on CT

3. Elevation of BNP (>90 pg/ml), elevation of N-terminal pro-BNP (>500 pg/ml) or electrocardiographic changes (new complete or incomplete right bundle-branch block, anteroseptal ST elevation or depression, or anteroseptal T-wave inversion)
4. Elevation of troponin I (>0.4 ng/ml) or elevation of troponin T (>0.1 ng/ml)

2.5 Treatment of VTE

2.5.1 Acute treatment of VTE

The goal of therapy for lower-extremity DVT is to prevent extension of thrombus and PE in the short term and to prevent recurrent events in the long term (Scarvelis D et al. 2006).

The ACCP consensus statement on VTE recommends initial treatment with low molecular weight heparin (LMWH) subcutaneously once or twice daily as an outpatient if possible. Initiation of vitamin K antagonist is started on the same day. With unfractionated heparin (UFH) or LMWH, the dose should be weight-based. If there is a high clinical suspicion of DVT, treatment with anticoagulants should be initiated while awaiting the outcome of diagnostic tests. In selected patients with extensive acute proximal DVT (iliofemoral DVT, symptoms for < 14 days, good functional status, life-expectancy ≥ 1 year) with low risk for bleeding, catheter-directed thrombolysis followed by balloon angioplasty and stents may be used to reduce acute symptoms and post-thrombotic morbidity. Initial treatment with LMWH, UFH, or the synthetic pentasaccharide fondaparinux is recommended to continue for at least 5 days and until the INR is ≥ 2.0 for over 24 h (Kearon C et al. 2008).

Placement of an inferior vena cava filter in addition to anticoagulation therapy has not been found to prolong survival among patients with DVT. A retrievable filter is indicated when there is a contraindication to anticoagulation therapy (recent hemorrhage, impending surgery) in patients with newly diagnosed proximal DVT (Scarvelis D et al. 2006). In catheter-directed thrombolysis, thrombolytic drugs are delivered using catheter-based techniques to achieve a higher concentration in the venous thrombus, thereby enabling clot lysis with reduced thrombolytics (Semba CP et al. 1994, Vedantham S et al. 2006). The underlying veins are evaluated after thrombolysis by venography, and balloon angioplasty or stent placement is performed if needed. Limitations of the technique are the fairly long

REVIEW OF THE LITERATURE

infusion time required to lyse extensive DVT and the healthcare resources used (Popuri R et al. 2011).

Systemic thrombolytic therapy has been shown to reduce the incidence of post-thrombotic sequelae compared with anticoagulant therapy alone (Elliot MS et al. 1979, Arnesen H et al. 1982, Killewich LA et al. 1989). Whether early canalization results in lower rates of PTS in the long run remains unknown (Forster AJ 2001). Recently, catheter-directed thrombolysis has overtaken systemic thrombolysis. Data from the National Venous Registry and subsequent studies on the role of catheter-directed thrombolysis for treatment of acute lower-extremity DVT indicate that the technique is safe and effective (Semba PC et al. 1994, Mewissen MW et al. 1999, Mewissen MW 2010).

Daily use of graduated elastic compression stockings for 2 years after a proximal DVT has been reported in several trials to significantly reduce the rate of PTS (Brandjes DP et al. 1997, Prandoni P et al. 2004). For a patient who has had a symptomatic proximal DVT, the use of an elastic compression stocking is recommended. Compression therapy, which includes the use of bandages acutely, should be started as soon as feasible after commencing anticoagulant therapy and continued for a minimum of 2 years, longer if patients have symptoms of PTS (Kearon C et al. 2008).

All PE patients should undergo rapid risk stratification. For patients with evidence of hemodynamic compromise, the use of thrombolytic therapy unless there are major contraindications owing to bleeding risk should be considered. In selected high-risk patients without hypotension who are judged to have a low risk of bleeding, administration of thrombolytic therapy is suggested as well. The decision to use thrombolytic therapy depends on the clinician's assessment of PE severity, prognosis, and risk of bleeding (Kearon C et al. 2008). Thrombolytic therapy should not be used in patients with low-risk PE. Catheter embolectomy or fragmentation of proximal pulmonary arterial clots may be considered in high-risk PE patients if thrombolysis is contraindicated or has failed (Torbicki et al. 2008).

2.5.2 Duration of anticoagulation

The duration of anticoagulation has been debated for years. ACCP, AHA (American Heart Association), and ESC have each published their own guidelines, which are mainly in line with each other.

For patients with DVT secondary to a transient (reversible) risk factor, ACCP guidelines recommend anticoagulation for 3 months rather than treatment for shorter periods. For patients with unprovoked DVT, anticoagulation is recommended for at least 3 months. All patients with unprovoked DVT should be evaluated for the risk-benefit ratio of long-term therapy. For patients with a first unprovoked VTE that is a proximal DVT, for whom risk factors for bleeding are absent and for whom good anticoagulant monitoring is achievable, long-term treatment is recommended. For patients with PE and cancer, LMWH is recommended for 3-6 months as long-term anticoagulant therapy. For these patients, anticoagulant therapy is recommended indefinitely or until the cancer is resolved. If the patient has a second episode of unprovoked VTE, long-term anticoagulation should be considered (Kearon C et al. 2008). The length of the anticoagulation therapy depends on the patient's VTE risk factors, presence of recurrent VTE, bleeding risk factors, and the patient's opinion (Prandoni P et al. 1996, Heit IA et al. 2000).

D-dimer is an indirect marker of residual thrombosis (Bockenstedt P 2003). In a systematic review of patients with a first episode of unprovoked VTE, a negative D-dimer after a 2-year follow-up was associated with a 3.5% annual risk for recurrent disease, whereas a positive D-dimer result was associated with an 8.9% annual risk (Verhovsek M et al. 2008). These results were confirmed in a systematic review (Douketis J et al. 2010). In the multicenter, prospective PROLONG study, patients with an abnormal D-dimer level one month after the discontinuation of anticoagulation had a significant incidence (15%) of recurrent VTE (Palareti G et al. 2006). These findings suggest that D-dimer assay may have a role when determining appropriate duration of anticoagulation.

The therapeutic effectiveness and safety of warfarin depends on maintaining the international normalized ratio (INR) in the therapeutic range. Because of the dose response variability and interactions with drugs and diet, warfarin therapy can be troublesome and time-consuming to manage (Ansell J et al. 2008). Most patients are treated with oral warfarin as a first-line long-term anticoagulant with initial LMWH

therapy for a minimum of 5 days and until INR is ≥ 2.0 for at least 24 h (Hull R et al. 1979, Levine MN et al. 1995, Kearon C et al. 2003, Ridker PM et al. 2003).

A number of new oral anticoagulants are currently under evaluation (Mavrakanas T et al. 2011). These orally administered direct inhibitors of FXa (rivaroxaban, apixaban) or thrombin (dabigatran) avoid most of the drawbacks of heparin and require no dose monitoring. In certain situations, it could be beneficial to monitor their effects such as in the case of suspected overdose, patients who need emergency surgery, or patients with a thromboembolic or hemorrhagic event (Lindhoff-Last E et al. 2010, Samama M et al. 2011). Dabigatran has been shown to be as safe and effective as warfarin for acute VTE (Schulman S et al. 2009, Einstein Investigators 2010). Rivaroxaban was the first drug to be accepted for the treatment of VTE. It is expected that other new oral anticoagulants, such as dabigatran, apixaban, and edoxaban, will be available in the near future, as the final phases and results of clinical trials are completed (Agnelli G et al. 2013, Schulman S et al. 2011, Schulman S et al. 2013, Raskob G et al. 2013). The major problem/drawback with these new drugs is the lack of an effective antidote and the lack of long-term experiences. However, these newer anticoagulants have shorter half-lives and the need for reversal agents is less critical. In healthy volunteers, a four-factor prothrombin concentrate was effective in reversal of rivaroxaban activity (Eerenberg E et al. 2011). Recent preclinical studies have shown promise for agents in the reversal of anticoagulant activity (Zhou W et al. 2011). Administration of prothrombin complex concentrate or activated prothrombin complex concentrate may be considered in addition to supportive measures for patients with severe or life-threatening bleeding. Target-specific antidotes are being developed, but require further investigations (Siegal DM et al. 2013).

2.6 Long-term sequelae and follow-up of VTE

2.6.1 Post-thrombotic syndrome (PTS)

Approximately two-thirds of patients with ileofemoral DVT will develop post-thrombotic syndrome (Strandness D et al. 1983, Landefeld CS 1993, Branjes DP et al. 1997). PTS is characterized by leg swelling, pain, darkened skin color, and sometimes ulcers.

Manifestation of PTS becomes apparent within 1-5 years after acute thrombosis, in most patients within the first 2 years (Prandoni P et al. 1996, Brandjes DP 1997).

Diagnosis of PTS is mainly based on clinical findings for patients with a history of DVT. Villalta PTS scale (Villalta S 1994) and the CEAP (clinical, etiologic, anatomic, and pathophysiologic) classification for chronic venous disease (Porter JM et al. 1995) are frequently used to measure PTS in clinical practice.

When carefully supervised and proper elastic compression stockings are worn, more than 50% of patients either remain quiescent or improve during long-term follow-up (Pesavento R et al. 2010). The placement of iliac vein stents to reduce PTS symptoms and heal venous ulcers in patients with advanced PTS can be considered. The adequacy of anticoagulation treatment therapy should be evaluated as well (Jaff M et al. 2011). Clinical predictors of PTS are proximally extensive DVT, prior ipsilateral DVT, persistent venous symptoms one month after DVT, obesity, and older age. The likelihood of developing PTS after DVT should be discussed with patients, and symptoms and signs of PTS should be monitored during clinical follow-up (Kahn SR et al. 2009).

2.6.2 Chronic thromboembolic pulmonary hypertension (CTEPH)

Pulmonary hypertension is a hemodynamic and pathophysiological condition that is defined as an increase in mean pulmonary arterial pressure ≥ 25 mmHg at rest, as assessed by right heart catheterization (Galie N et al. 2009).

CTEPH is a late complication of PE. The natural history of most emboli is to undergo total or almost total resolution with restoration of normal pulmonary hemodynamics (Moser KM et al. 1990). In patients with CTEPH, the original embolic material is replaced over a period of months to years with fibrous tissue. This material may extend from the main pulmonary trunk to the segmental and subsegmental branches of the pulmonary arteries. Partial recanalization or total occlusion of the involved pulmonary artery vasculature may occur (Moser KM et al. 1990, Galie N et al. 2009). The consequence is an increased pulmonary vascular resistance, resulting in pulmonary hypertension and right heart failure (Hoepfer MM et al. 2006).

Follow-up studies have detected RVD in 6-20% of patients (Kline JA et al. 2009, Golpe R et al. 2010). Pengo V et al. (2004) reported symptomatic CTEPH in 4% of

REVIEW OF THE LITERATURE

patients 2 years after the first episode of symptomatic pulmonary embolism. Thus, the occurrence of CTEPH is not rare.

Patients with a history of DVT or PE who present with dyspnea, exercise intolerance, or clinical evidence of right heart failure should undergo diagnostic evaluation for CTEPH (Jaff R et al. 2011). The diagnosis of CTEPH is based on echocardiography, perfusion scintigraphy, CT, right heart catheterization, and pulmonary angiography (Coulden R et al. 2006). Medical therapy aims to treat right heart failure and to lower pulmonary artery resistance. The efficacy of the medical therapy is limited by the morphological substrate of pulmonary artery obstruction. Pulmonary endarterectomy has provided fairly good results. Drugs targeting the pulmonary circulation of patients in whom surgery is not possible or has failed are undergoing clinical trials (Torbick A et al. 2008). CTEPH patients should receive lifelong anticoagulation to prevent recurrence of thromboembolic events (Hoeper MM et al. 2006).

3. AIMS OF THE STUDY

The main purpose of this study was to determine the easiest way to diagnose right ventricular dysfunction in emergency department acute non-high-risk patients and to identify those patients likely to benefit from follow-up. A further aim was to evaluate the long-term clinical outcome of patients with earlier PE or DVT.

Specific aims of the four studies were as follows:

1. To develop a sensitive, fully quantitative and automated POC testing assay for D-dimer in whole blood with the shortest possible assay time.
2. To determine the value of CT and NT-proBNP in detection of right ventricular dysfunction in the acute phase and during follow-up of non-high-risk acute pulmonary embolism in clinical practice.
3. To study the membrane and pulmonary capillary blood volume components of diffusing capacity at the acute stage of pulmonary embolism to determine which parameters are associated with right ventricular overload.
4. To compare the reflux patterns in patients with iliofemoral venous thrombosis 2-3 years after systemic or catheter-directed thrombolysis.

4. PATIENTS AND METHODS

Study I: Rapid measurement for D-dimer in VTE patients

This prospective study comprised patients (n=525) from the University Central Hospitals of Helsinki, Turku, and Kuopio. The diagnoses for DVT and PE were performed with duplex ultrasound examination and helical-CTscan according to accepted protocols in clinical use. Samples from age-adjusted healthy volunteers (n=101) were collected among the staff at the Department of Biotechnology, University of Turku, and at Innotrac Diagnostics, Turku, Finland.

All whole-blood samples were analyzed fresh, and plasma samples were analyzed either fresh or stored frozen at -20°C or -70°C until use. The samples were stable at both freezing temperatures. The simple, automated assay procedure comprises a 1:50 sample dilution, one-step incubation, washing, and time-resolved fluorometric measurement directly from the wet well surface. The Roche Diagnostics TinaQuant® D-Dimer (Knecht MF et al. 1997) and the Biopool Auto Dimer® (Bozic M et al. 2003) assays were used as comparison methods. A cut-off value of 0.5 mg/l was employed in both methods. Commercial D-dimer control preparations were purchased from Bio-Rad Laboratories and from Biopool and were used as instructed by the manufacturers.

The study protocol was approved by the Ethics Committee of Helsinki University Central Hospital, and written informed consent was obtained from all participants.

Study II: Right ventricular overload in patients with acute pulmonary embolism: role of helical computerized tomography (CT) and N-terminal pro-BNP in screening and follow-up in routine patient care

This was a single-center prospective follow-up study carried out at the Emergency Department of Helsinki University Central Hospital between January 2003 and August 2004. Sixty-three consecutive patients were included in the study. The main inclusion

PATIENTS AND METHODS

criterion was CT-confirmed APE. Subjects with massive APE (hemodynamically unstable patients), pulmonary disease with regular medication, previous APE, ongoing anticoagulation therapy, and end-stage cancer (estimated life-expectancy of less than 7 months) were excluded. No randomization of treatment was performed. Management of patients was decided by the clinician in charge.

On admission, right ventricular overload was assessed by the following four different methods: 1) CT, 2) TTE, 3) cardiac biomarkers (NT-proBNP and cardiac troponin T), and 4) clinical evaluation including ECG. TTE, cardiac biomarkers, and clinical evaluation were repeated 7 months later. A detailed medical history, including risk factors for thromboembolic events (age, gender, immobilization (within 3 months), hormone replacement or contraception therapy, family history of venous thromboembolic events, active cancer, varicose veins, smoking, weight, and height), was collected from all patients. Patient characteristics and risk factors are presented in Table 4.

Table 4. Characteristics and risk factors of 63 patients.

| Parameter | Patients, n (%) |
|--|-----------------|
| Age >75 years | 10 (16) |
| Gender | |
| Male | 30 (48) |
| Female | 33 (52) |
| BMI (Body Mass Index) >25 kg/m² | 50 (79) |
| Immobilization | 28 (44) |
| (prolonged immobility, hospitalization, surgery within 3 months) | |
| Hormone therapy | 13 (21) |
| (Hormone replacement therapy n=7) | |
| Contraception n=6 | |
| History of DVT* | 7 (11) |
| Family history of VTE** | 5 (8) |
| Thrombophilias*** | 9 (14) |
| Homocysteinemia (> 12µmol/l) | 11 (17) |
| Active cancer | 3 (5) |
| Varicose veins | 27 (43) |
| Diabetes mellitus | 5 (8) |
| Asthma (mild, no regular medication) | 5 (8) |
| Smoking | |
| Smokers | 11 (17) |
| Ex-smokers | 19 (30) |
| Non-smokers | 33 (52) |

* DVT= Deep venous thrombosis

** VTE= Thromboembolic disease

*** Known previously or detected in clinical laboratory tests

PATIENTS AND METHODS

Blood samples for plasma NT-proBNP, cardiac troponin T (cTNT), and D-dimer were collected at presentation. D-dimer >0.5 mg/l and cTNT >0.03 ug/l were assessed as abnormal. NT-proBNP value ≥ 350 ng/l was regarded as elevated. The criteria of RVD in CTPA were RV/LV ratio over 1.0 and/or deviation (straightening or bowing) of the interventricular septum (Schoepf UJ et al. 2004, Araoz PA et al. 2003). The diameters of the ventricles were measured on the axial image where the ventricles were the most distorted. Thirty-six CT examinations were performed with an 8-slice scanner (GE Light Speed Ultra), 22 with a 4-slice scanner (22 with Philips Mx 8000, one with GE High Speed), and 5 with a single-slice scanner (GE High SpeedLX/i).

TTE was performed within 24 h of the diagnosis of APE using the Vivid Digital Ultrasonography System (GE Vivid 5 or Vivid 7, Horten, Norway). Recordings were stored on an M-mode, two-dimensional color-flow disc, and Doppler data were stored on a magneto-optic disc. Examination was performed both in a left lateral and in a flat supine position. All studies were performed by experienced cardiologists blinded to the results of biochemical assays. TTE was repeated 7 months later by the same cardiologist as on admission. Presence of RVD was assessed using the following established criteria: 1) increased ratio of RV and left ventricular end-diastolic diameter (RV/LV) above 0.9, 2) presence of paradoxical septal wall motion, and 3) mean peak velocity greater than 2.8 m/s. At least one of these criteria had to be positive.

All statistical analyses were performed with SPSS 15.0 for Windows (SPSS Inc., Chicago, IL, USA). The subjects were categorized into two groups: RVD positive and RVD negative. Data are presented as frequencies or percentages for categorical variables and as mean (SD) for continuous variables, unless otherwise noted. Normality of continuous variables was checked by the Kolmogorov-Smirnov test. Between-group differences were assessed by the Mann-Whitney U-test. Categorical data were compared by the Chi-square test or Fischer's exact test. P-values <0.05 were considered statistically significant.

The study protocol was approved by the Ethics Committee of Helsinki University Central Hospital, and written informed consent was obtained from all participants.

Study III: Reduction of membrane component of diffusing capacity is associated with the extent of acute pulmonary embolism

Forty-seven consecutive patients (24 women, 23 men) with acute non-high-risk PE confirmed by computerized tomography (CT) at the Emergency Department of Helsinki Meilahti University Central Hospital were enrolled in the study. Patients were recruited between January 2003 and August 2004. Exclusion criteria comprised clinically massive PE (hemodynamically unstable patients), chronic pulmonary disease requiring regular medication, previous PE, anticoagulation therapy, and terminal cancer (with estimated life-expectancy of less than 7 months). In addition, patients arriving at the hospital between Friday 18:00 and Sunday 12:00 were excluded because the services of the laboratory of clinical physiology were unavailable at that time. The measurements of DL, CO with DM and Vc were scheduled within 24 h of diagnosis of PE. DL, CO, Vc, DM, and VA were measured by using the single breath method with carbon monoxide and oxygen (Viljanen AA 1982) applying the European Respiratory Society recommendations (McIntyre N et al. 2005) at both the acute phase and 7 months later. The measurements were done with Jaeger MasterScreen PFT equipment (Würzburg, Germany). Values of diffusing capacity were corrected with actual hemoglobin concentration in blood. Flow-volume spirometry was performed at the 7-month control to exclude patients with pulmonary obstruction. At the acute phase, spirometry was not performed for safety reasons. Forced vital capacity (FVC), forced expiratory volume in one second (FEV1), and forced expiratory flow at a level where 50% of FVC remains to be exhaled (MEF50) were recorded.

The extent of PE was assessed by scoring mass of embolism. Mass of embolism was calculated using the method of Mastora (Mastora I et al. 2003), in which CT severity score is based on the percentage of the obstructed surface of each central and pulmonary arterial section using a 5-point scale. Echocardiography was also performed. Patients were given antithrombotic treatment according to guidelines in force at the time of the study. Most of the patients were treated initially with low molecular weight heparin, followed by warfarin for 6 months. Seven months later, diffusing capacity measurements were repeated and flow-volume spirometry was performed. Echocardiography was also repeated to identify patients with RVD. Fifteen healthy, age-matched controls (9 women, 6 men) were also

PATIENTS AND METHODS

studied; for 12 of them (three did not come to the second measurement), the measurements were repeated later, at times corresponding to those of the patients.

Lung function parameters were compared between patients and controls with independent samples t-test. Correlation methods were used to determine the covariants, and analysis of covariance (ANCOVA) was applied to adjust the results according to covariants. For parameters indicated as percentages of predicted values, the results were adjusted for years and also for weight if weight was not calculated in the reference value; for parameters indicated as absolute values, the results were adjusted for height, weight, age, and smoking. In multiple comparisons, Bonferroni correction was used.

Partial correlation was used to adjust the lung function parameters for age, height, weight, and smoking.

Table 5. Gender and anthropometric data of patients with PE and healthy control subjects.

| | Patients with PE | Healthy controls | Singificance in t-test |
|-------------------------------|------------------|------------------|------------------------|
| Gender, male/female (persons) | 23/24 | 6/9 | |
| Weight (kg) | 88.9 (18.1) | 73.0 (11.2) | 0.002 |
| Height (cm) | 172.0 (10.4) | 173.0 (11.1) | 0.730 |
| Age (years) | 56.1 (16.3) | 55.0 (16.5) | 0.881 |
| Smoking (pack-years) | 3.92 (7.6) | 2.67 (6.0) | 0.469 |
| Number (percent) of | 30 (63.8) | 11 (73.3) | 0.761 * |
| non-smokers | 7 (14.9) | 2 (13.3) | |
| smokers | 10 (21.3) | 2 (13.3) | |
| ex-smokers | | | |

* *Chi-square test*

The change in the parameters during follow-up was analyzed with paired t-test, or with Wilcoxon's pairwise test when the variable was not normally distributed. To evaluate the influence of lung volume on the observed changes, the difference in VA between the acute and recovery phases of PE was compared in regression analysis with the corresponding difference in DL, CO, DL, CO/VA, DM, or Vc between the examination phases. SPSS Windows version 15.0 was used in all calculations.

The study protocol was approved by the Ethics Committee of Helsinki University Central Hospital, and written informed consent was obtained from all participants.

Study IV: Comparison of venous valve function after catheter-directed and systemic thrombolysis for deep venous thrombosis

Patients with iliofemoral deep venous thrombosis treated with either systemic or catheter-directed thrombolysis were evaluated in this retrospective study 2-3 years after the thrombotic event. A total of 60 patients with a diagnosis of deep iliofemoral DVT were collected from the Helsinki University Central Hospital Emergency Care Data Registry over a one-year period, from July 1999 to the end of June 2000. Of 60 patients, 12 had died since the acute event and 11 were treated with low molecular weight heparin and anticoagulation without thrombolysis. Altogether 37 patients treated with thrombolytic for iliofemoral DVT participated in the study. The patients had overt clinical signs and symptoms of DVT, and the initial diagnosis had been established with compression two-dimensional B-mode ultrasound. Of the 37 patients, 16 had been treated with catheter-directed thrombolysis with recombinant tissue plasminogen activator (rt-PA) and 16 with systemically administered thrombolysis either with streptokinase or rt-PA. Furthermore, there were 5 patients who received catheter-directed thrombolysis after an initial systemic thrombolysis had failed to produce an adequate clinical result. This group of 5 patients was not included in the final analysis because of its small size. The study sample thus included 32 patients. Three of the 16 patients in the catheter-directed lysis-treated group had residual stenosis after lysis and were treated with percutaneous balloon angioplasty (PTA) (1 patient), PTA and stenting (1 patient), and catheter thrombectomy (1 patient). All patients also received treatment with low weight molecular heparin and anticoagulation.

The clinical status of the affected leg was classified from 0 to 6 according to the clinical, etiologic, anatomic, and pathophysiologic (CEAP) classification (Rutherford RB et al. 2000, Porter J et al. 1995), and disability score was calculated at follow-up 2-3 years after the acute event. A detailed systemic examination of the entire venous system from the groin to the ankle was performed for the limb with previous DVT by an experienced vascular surgeon who was blinded to the previous treatment. Reflux was assessed with color duplex ultrasound (Hewlett Packard M2410A scanner using 5 MHz probe), and standardized distal pneumatic cuff release (Venopulse, STR Teknik, Norway) was used to provoke venous reflux.

PATIENTS AND METHODS

Patients were also asked to fill in the questionnaire concerning risk factors, duration of anticoagulation therapy, and use of compression stockings. No significant baseline difference between these two groups was found (Table 6).

Table 6. *Patient characteristics.*

| Variables | CATHETER-DIRECTED LYSIS | SYSTEMIC LYSIS |
|---|------------------------------------|---|
| No. of patients | 16 | 16 |
| No. of males | 8 | 6 |
| Age (years) mean and range | 56 (21 to 86) | 61 (35 to 81) |
| Thrombus age (days) mean and range | 3 (1 to 8) | 4 (1 to 14) |
| Thrombolytic agents | | |
| Alteplase (n) | 16 | 4 |
| Streptokinase (n) | 0 | 14 |
| Total dose of thrombolytic | | |
| Alteplase (mg) mean and range | 73 (20 to 142) | 229 (100 to 343) |
| Streptokinase (IU) mean and range | | 6.5x10 ⁶ (2.7 to 7.4x10 ⁶) |
| Duration of thrombolytic therapy (hours) mean and range | 33 (12 to 72) | 62 (33 to 72) |
| Bleeding | | |
| Major | 13% (2/16) | 6% (1/16) |
| Minor | 25% (4/16) | 38% (6/16) |
| Intracranial haemorrhage | 0 | 0 |
| Concomitant pulmonary embolism | 13% (2/16) | 31% (5/16) |
| Genetic risk factor | 25% (4/16) | 13% (2/16) |
| Risk factors * | 50% (8/16) | 75% (12/16) |
| Use of compressing stockings | 50% (8/16) | 38% (6/16) |
| Warfarin treatment | | |
| Permanent warfarin usage | 38% (6/16) | 38% (6/16) |
| Duration of warfarin (months) mean and range | 8 (3 to 24) | 8 (3 to 15) |

* Presence of one or more of the following risk factors: genetic thrombophilias, previous deep venous thrombosis or pulmonary embolism, obesity, hormones, smoking, surgery, traveling, immobilization in the 3-month period before onset.

Statistical measurements were made with the χ^2 method with no continuity correction. Statistical significance was defined as a P-value of less than 0.05

The study protocol was approved by the Ethics Committee of Helsinki University Central Hospital, and written informed consent was obtained from all participants.

5. RESULTS

Study I: Rapid measurement of D-dimer in VTE patients

The analytical detection limit of the assay, calculated as the mean background signal +3 SD, was 0.046 mg/l. The within- and between-assay imprecision was similar for whole-blood and plasma samples, ranging from 8.3% to 13% below and from 3.8% to 7.7% above the cut-off level of 0.6 mg/l. The estimate of 0.2 mg/l for the limit of quantification appeared to be correct also based on the precision studies. The within- and between-assay imprecision was also calculated for commercial D-dimer controls from Bio-Rad and Biopool and showed a corresponding imprecision. Measurable concentrations of D-dimer were found in most citrated blood samples taken from apparently healthy volunteers. Both the whole-blood and plasma fractions were measured, and the results obtained in whole blood were corrected for the individual's hematocrit values. For persons <50 years of age (n=66), the median D-dimer concentrations were 0.19 and 0.24 mg/l in whole blood and plasma, respectively, with the highest measured concentrations of 0.51 and 0.57 mg/l. The upper reference limits based on the 95th percentile were 0.41 mg/l in whole blood and 0.45 mg/l in plasma. For individuals \geq 50 years of age (n=35), however, the reference values were notably higher. The median values for this age group were 0.36 mg/l in both whole blood and plasma, with the highest measured concentrations of 1.0 and 0.93 mg/l. The upper reference limits based on the 95th percentile were 0.84 and 0.82 mg/l in whole blood and plasma. In the entire population of apparently healthy volunteers (n=101), the median D-dimer concentrations in whole blood and plasma were 0.24 and 0.29 mg/l, with upper reference limits (95th percentile) of 0.68 and 0.56 mg/l. The 10-min D-dimer assay was compared with the Roche Diagnostics TinaQuant D-dimer assay.

RESULTS

Table 7. Characteristics of the 10-min D-dimer assay using two different clinical decision limits (mg/l cut-offs).

| | Cut-off value | |
|----------------------------------|---------------------|--------------------|
| | 0.6 mg/l | 1.0 mg/l |
| Sensitivity (95% CI), % | 98.7 (93.0 – 100.0) | 92.2 (83.8 – 97.1) |
| Specificity (95% CI), % | 64.4 (56.8 – 71.5) | 81.0 (74.4 – 86.6) |
| NPV ^a , % | 99.1 | 95.9 |
| PPV ^b , % | 55.1 | 68.3 |
| Test efficiency ^c , % | 74.9 | 84.5 |

^aNPV=negative predictive value

^bPPV=post-predictive value

Of the 77 samples obtained from patients with DVT and/or PE, all but one (which was negative in the Tina Quant assay as well) gave a result above the cut-off level of 0.6 mg/l (98.7%), and 71 (92.2%) gave a result ≥ 1.0 mg/l; the median D-dimer value of the whole group was 6.67 mg/l. However, although the median D-dimer level in the group of unselected outpatients with a wide range of other diseases was only 0.43 mg/l (n=174), a significant portion of these samples were above the cut-off levels of 0.6 mg/l (62 samples, 35.6%) and 1.0 mg/l (30 samples, 17.2%), demonstrating the low specificity of the assay in a typical outpatient population. The reasons for the increased D-dimer concentrations included cardiovascular disease (n=7), kidney disease (n=6), atrial fibrillation (n=5), erysipelas (n=4), cancer (n=4), pneumonia (n=2), hepatitis (n=1), other infection/inflammatory disease (n=7), other thrombotic disorder (n=6), and multiple concurrent diseases (n=4); the reason was unknown or unreported in 16 cases.

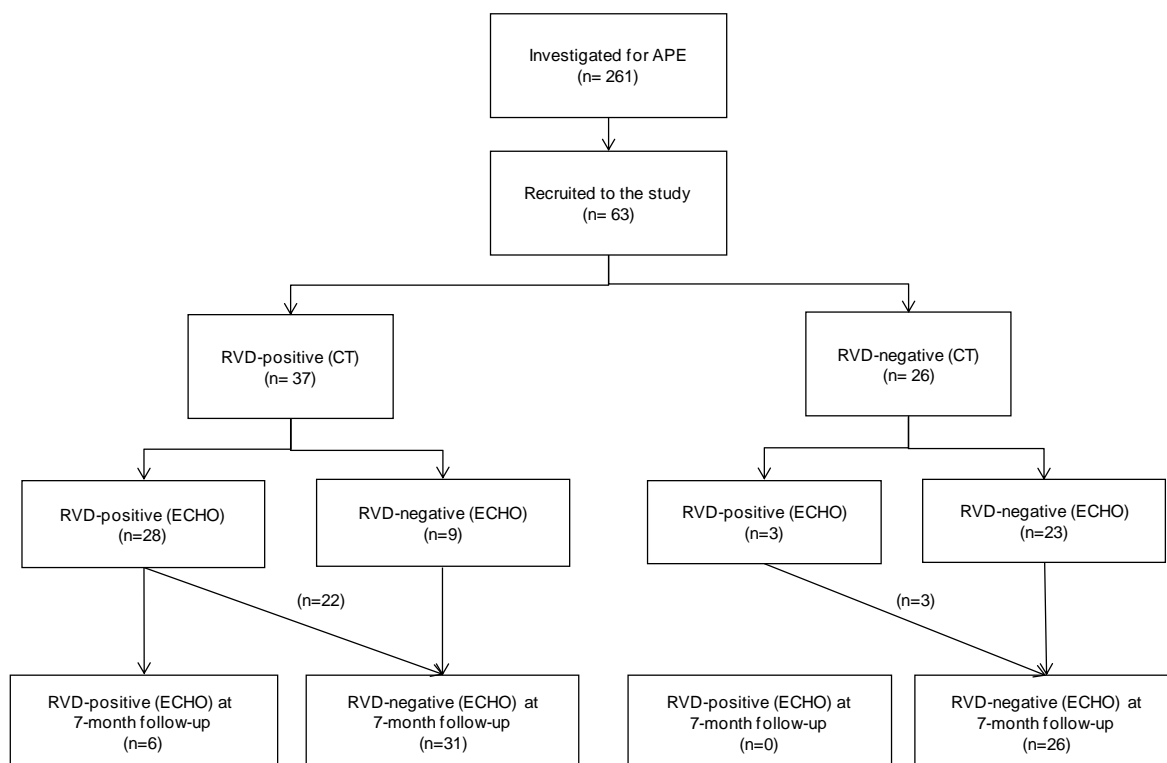
Study II: Right ventricular overload in patients with acute pulmonary embolism: role of helical computerized tomography (CT) and N-terminal pro-BNP in screening and follow-up in routine patient care

Right ventricular overload was detected with CT in 37 (59%) of 63 patients (Figure 2). Twenty-eight patients (76%) in the RVD-positive group also had signs of RVD in TTE performed within 24 h of the diagnosis. Nine of the RVD-positive patients (24%) showed no echocardiographic alterations in RV morphology or function. Overall, findings of RVD in CT were congruent with those of echocardiography (p<0.0001). Findings suggesting

RESULTS

RVD were found with TTE in three of the initially RVD-negative patients. These patients had as the only sign of RVD mean tricuspid regurgitation peak velocity of 2.9-3.1 m/s.

Figure 2. Evaluation of right ventricular dysfunction with CT



The RVD diagnosis was based on both right ventricular and left ventricular ratio, and septum deviation was detected in most (73%) of the patients. Detailed information of RVD findings in CT of these 37 RVD patients is presented in Table 8.

Table 8. CT findings of RVD patients and elevated pro-BNP in the criteria groups.

| | CT findings | | pro-BNP over 350 ng/l | |
|-------------------------------|-------------|------|-----------------------|------|
| | n (%) | n=37 | n (%) | n=32 |
| RV/LV >1 (only) | 3 (8.1) | | 2 (66.7) | |
| septum deviation (only) | 7 (18.9) | | 6 (85.7) | |
| RV/LV >1 and septum deviation | 27 (73.0) | | 24 (88.9) | |

RV= right ventriculum

LV= left ventriculum

Of patients who had RVD in CT, 32 (86%) also had NT-proBNP \geq 350 ng/l, whereas in 7 patients (27%) with no RVD NT-proBNP was over the limit ($p < 0.0001$). TnT was above

RESULTS

the reference value in 11 (17%) of 63 patients; 9 (82%) of these patients were RVD-positive and 2 (18%) were RVD-negative ($p<0.082$). At the 7-month follow-up, RVD was detected in 6 (10%) of 63 patients. These patients also had elevated NT-proBNP levels ($p<0.0001$). All patients who were found to have persistent RVD had RVD in both CT and TTE on admission.

Signs of RV overload in admission ECG were detected in 33 patients. In the RVD-positive group, 32 (86.5%) of 37 patients had signs of RVD in ECG, whereas 8 patients (13%) had such findings in the RVD-negative group ($p<0.0001$). Findings of ECG (one or more) are presented in Table 9.

Table 9. *Electrocardiogram (ECG) in APE patients*

| Criteria of ECG changes | Total | RVD-positive | RVD-negative |
|--|-------|--------------|--------------|
| Sinus tachykardia (pulse rate > 100/min) | 9 | 7 | 2 |
| T-wave inversion in leads V1-V3 | 26 | 20 | 6 |
| Complete or incomplete right bundle branch block | 4 | 3 | 1 |
| S1Q3T3 configuration | 3 | 1 | 2 |
| P-pulmonale | 1 | 1 | 0 |

Arterial hypoxemia was detected in 56 (93%) and hypocapnia in 27 (45%) of 60 patients. RVD-positive patients ($n=37$) had significantly more often hypoxia and hypocapnia ($n=36$) than patients in the RVD-negative group ($p<0.0001$). Elevated D-dimer levels were detected in all 63 patients. The levels of D-dimer did not differ between the RV-positive and RV-negative groups ($p<0.102$).

Study III: Reduction of membrane component of diffusing capacity is associated with the extent of acute pulmonary embolism

The total score for mass of embolism varied between 0 and 110 (mean, SD; 54.1, 32.4), corresponding to a 0-71% obstruction of the total pulmonary artery bed. Forty patients had both central and peripheral emboli. Seven patients had only peripheral emboli and one subsegmental emboli. The score for central mass of embolism was 0-34 (mean, SD; 17.5, 1.8), corresponding to a 0-62% obstruction of the central pulmonary artery bed. The scores for peripheral mass of embolism ranged from 0 to 82 (37.0, 22.5), thus, a 0-82%

RESULTS

obstruction of the peripheral pulmonary artery bed.

DL, CO, Vc, DM, and VA were lower in patients with acute PE than in healthy controls ($p < 0.001$). DM correlated inversely with central mass of embolism ($r = -0.312$; $p = 0.047$), whereas Vc did not.

DM, DL, CO, VC, and VA improved significantly within 7 months. It is notable that in all patients ($p = 0.001$, $p = 0.001$), also persistent RVD patients ($p = 0.020$, $p = 0.012$), DM, DL, and CO remained significantly lower than in healthy controls at follow-up. DM was inversely related to central mass of embolism. Reduction in DM mainly explains the sustained decrease in DL and CO in PE after 7 months despite modern and adequate treatment of PE (Table 10).

Table 10. Lung function data of patients and healthy controls during the acute phase and at the 7-month follow-up.

| Variable | Patients, acute phase, n=47 | Controls, acute phase, N=15 | Significance between patients and controls, acute phase #** | Patients, 7 month follow-up, n=47 | Significance, patients acute vs. 7 month follow-up ## | Controls, 7 month follow-up N=12 | Significance between patients and controls, 7 month follow-up #** |
|--------------------------|-----------------------------|-----------------------------|---|-----------------------------------|---|----------------------------------|---|
| VC (l) | 3.5 (1.1)* | 4.4 (1.1) | <0.001 | 3.80 (1.19) | < 0.001 | 4,25 (1.17) | < 0.001 |
| VC (%) | 83.3 (15.5) | 103.9 (14) | <0.001 | 90.3 (16.4) | < 0.001 | 104.1 (15.2) | 0.062 |
| VA (l) | 4.3 (1.1) | 5.5 (1.3) | <0.001 | 4.7 (1.2) | < 0.001 | 5.1 (1.15) | < 0.001 |
| DLCO (mmol/min/kPa) | 5.9 (1.8) | 8.3 (2.5) | <0.001 | 6.6 (2.1) | < 0.001 | 8.2 (2.5) | < 0.001 |
| DLCO (%) | 73.7 (13.6) | 103.7 (15.2) | <0.001 | 82.3 (14.4) | <0.001 | 101.1 (15.59) | < 0.001 |
| DLCO/VA (mmol/min/kPa/l) | 1.39 (0.2) | 1.48 (0.2) | 0.259 | 1.30 (0.24) | 0.568 | 1.47 (0.27) | < 0.001 |
| DLCO/VA (%) | 94.6 (14.5) | 104.4 (13.7) | 0.025 | 96.3 (14.4) | 0.278 | 104.8 (17.2) | 0.023 |
| Dm (mmol/min/kPa) | 9.1 (3.0) | 13.9 (4.7) | <0.001 | 10.3 (3.8) | 0.001 | 13.6 (4.9) | < 0.001 |
| Dm (%) | 49.3 (15.6) | 72.3 (22.8) | <0.001 | 55.0 (19.3) | 0.003 | 71.7 (28.7) | 0.014 |
| Qc (ml) | 53.6 (16.1) | 63.9 (22.6) | <0.001 | 56.6 (16.5) | 0.159 | 69.3 (19.5) | < 0.001 |
| Qc (%) | 68.7 (17.2) | 79.3 (19.4) | 0.047 | 72.0 (16.5) | 0.173 | 85.8 (18.1) | 0.041 |
| Dm/Qc relation | 0.18 (0.09) | 0.22 (0.06) | 0.116 | 0.19 (0.09) | 0.770 | 0.21 (0.11) | 0.250 |

* Means and standard deviations are presented.

**Level of significance according to Bonferroni correction is 0.004.

non-paired ANCOVA

paired ANCOVA

At 7 months, the size and function of the right ventricle had normalized in most patients with RVD at baseline (24/47, 51%), but 5/47 patients (10.6%) still had RVD in echocardiography. In patients with persistent RVD at 7 months, DM was significantly lower than in those without RVD or in healthy controls. Also DL and CO remained

RESULTS

significantly lower in those with RVD than in healthy controls. In patients with permanent RVD, the correlations between VA and DL, CO, DL, CO/VA, DM, or Vc were not significant.

Study IV: Comparison of venous valve function after catheter-directed and systemic thrombolysis for deep venous thrombosis

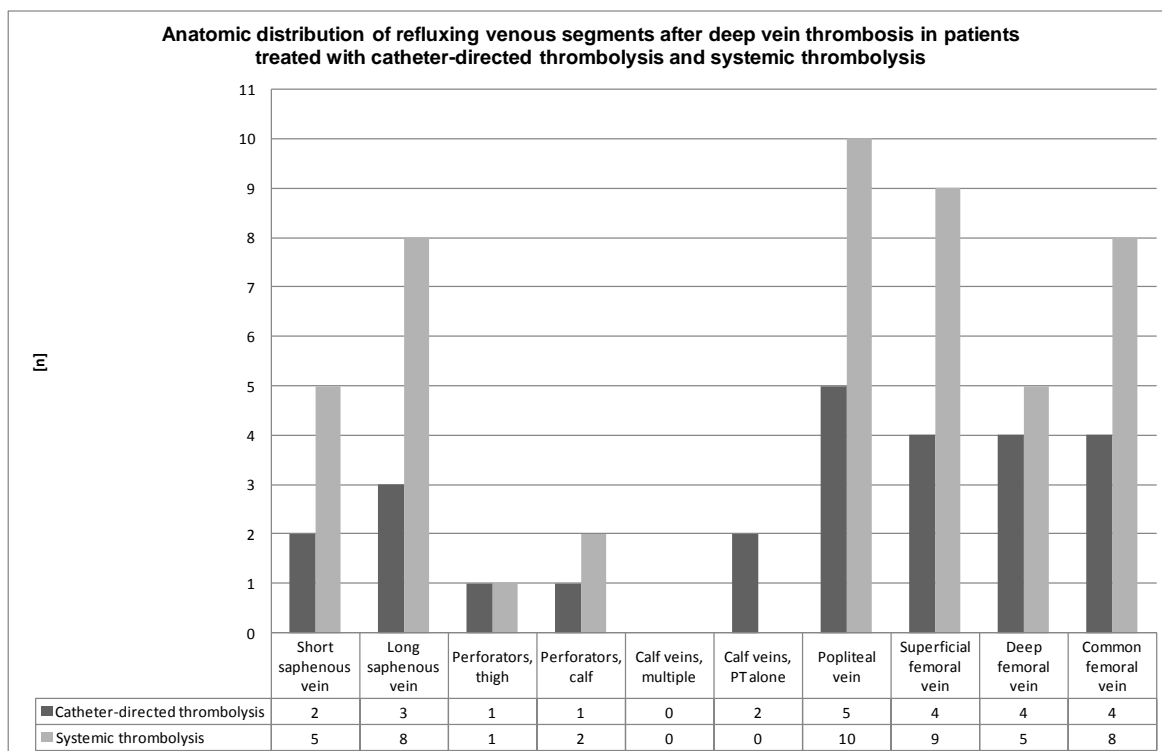
In this non-randomized retrospective study of both catheter-directed and systemic thrombolysis-treated patients, 38% were using warfarin permanently at the follow-up visit. In the rest of the patients in both groups, the mean duration of warfarin treatment was 8 months.

Risk factors, including genetic defects, were found in 50% of catheter-directed and 75% of systemic-treated patients ($p=0.25$). Fifty percent of the catheter-directed lysis patients and 38% of the systemic lysis-treated patients had used or were still using compression stockings ($p=0.41$).

Valvular competence was preserved in 44% of patients treated with catheter-directed thrombolysis compared with 13% of those treated systemically ($p=0.01$). Any deep reflux was present in 44% of catheter-directed lysis-treated patients compared with 81% of systemic-treated patients ($p=0.03$). Any superficial reflux was observed in 25% and 63% of the patients, respectively ($p=0.03$). No significant differences emerged in other variables (Figure 3).

RESULTS

Figure 3. Anatomic distribution of refluxing venous segments after deep vein thrombosis in patients treated with catheter-directed thrombolysis (black column) or systemic thrombolysis (light gray column).



Patients treated with catheter-directed thrombolysis were recorded to have better clinical status at follow-up (Table 11).

Table 11. Clinical classification at follow up.

| Clinical class * | Catheter-directed thrombolysis | | Systemic thrombolysis | |
|------------------|--------------------------------|----|-----------------------|----|
| | n | % | n | % |
| C0 | 7 | 44 | 2 | 13 |
| C1 | 2 | 13 | 1 | 6 |
| C2 | 1 | 6 | 2 | 13 |
| C3 | 3 | 19 | 6 | 38 |
| C4 | 3 | 19 | 5 | 31 |
| C5 | 0 | 0 | 0 | 0 |
| C6 | 0 | 0 | 0 | 0 |

*C0 = No visible signs of venous disease, C1 = Teleangiectases or reticular formation, C2 = Varicose veins, C3 = Edema, C4 = Skin changes, C5 = Skin changes + healed ulcer, C6 = Skin changes + active ulcer

6. DISCUSSION

General view and summary of results

This thesis includes a sample of VTE patients who underwent clinical, radiological, or biochemistry evaluation over the course of their illness. The main results of the four studies are as follows:

1. The rapid and sensitive 10-min immunofluorometric D-dimer assay showed utility in rapid exclusion of VTE in outpatients.
2. CT proved to be a valuable and sensitive imaging method to detect RVD in APE. RVD was detected in 37 (59%) of the patients with non-high-risk APE. Of patients who had RVD in CT, 32 (86%) also had NT-proBNP ≥ 350 ng/l, whereas in 7 patients with no RVD (27%) NT-proBNP was over the limit ($p < 0.0001$). At the 7-month follow-up, RVD was detected in 6 (10%) of 63 patients, and all of these patients also had elevated NT-proBNP levels ($p < 0.0001$).
3. The diffusing capacity (DL, CO) and especially its alveolocapillary membrane component (DM) are decreased in acute PE. The extent of PE and the severity of RVD were inversely associated with DL, CO and DM; the larger the PE, the lower the DL, CO or DM. Although DL, CO and DM increased at the 7-month follow-up, they still remained lower than in healthy control subjects, especially in patients with persistent RVD. The reduction in DM seemed to be independent of lung volume in persistent RVD.
4. Comparison of reflux pattern after catheter-directed and systemic thrombolysis revealed that local thrombolysis of ileofemoral deep venous thrombosis is associated with less venous reflux 2-3 years later. By reducing the incidence of venous reflux, the likelihood of developing PTS diminishes.

Value of rapid immunofluorometric D-dimer assay for exclusion of VTE in outpatients

We were able to develop a rapid and sensitive D-dimer method in POC settings using a non-competitive assay design with two monoclonal antibodies (dry-reagent assay chemistry and time-resolved fluorometric detection).

The analytical efficiency characteristics of this assay, including detection limit, limit of quantification, linearity, assay range, and precision, were quite similar to those of the immunoturbidimetric assays (Knecht MF et al. 1997, Bozic M et al. 2003) employed as comparison methods here. Nevertheless, the correlation of the new assay to these was only moderate (Roche TinaQuant, $r=0.726$) or poor (Biopool Auto Dimer, $r=0.190$). The samples obtained from patients with VTE typically gave a higher concentration in the 10-min assay than in either of the comparison methods, which is a positive feature of the new assay. Because of the lack of standardization and correlation between the different methods, assessment of diagnostic accuracy must be performed separately for each D-dimer method based on the clinical status of the tested patients.

Preliminary values for sensitivity, specificity, NPV, and PPV were established by comparing the D-dimer concentrations obtained in outpatients with DVT and/or PE. This was a group of unselected outpatients with various other diseases, therefore representing well the population in which the assay is to be used. Because the aim here was solely to verify the clinical functionality of the novel assay, a more thorough clinical evaluation study is required to better assess the assay's clinical performance characteristics. However, considering the wide range of diseases and the large number of increased D-dimer concentrations encountered in the comparison population, the preliminary calculations are not likely to be unduly optimistic. D-dimer is less useful in some groups of patients, including those with high clinical probability of VTE, patients older than 65 years, pregnant women, and patients admitted to hospital for another reason and in whom the suspicion of PE is raised during hospital stay (Righini M et al. 2005, Righini M et al. 2008).

Two different cut-off values, 0.6 and 1.0 mg/l, compared well with the detection limits typically employed in sensitive automated assays and rapid manual assays. The sensitive assays are sometimes criticized as being too unspecific, generating a large number of positive results that require further studies. Nevertheless, the cut-off level of the more

DISCUSSION

sensitive methods can be freely selected, and when used in combination with clinical probability assessment, VTE can be excluded in a significantly larger number of patients.

In recent years, the highly sensitive D-dimer assays have been used to exclude PE in patients whose clinical probability for the condition is not high (Djurabi RK et al. 2009, Legnani C et al. 2010, Pasha SM et al. 2010, Gosselin RC et al. 2012). In our study, the rapid immunofluorometric assay showed value in rapid exclusion of VTE in outpatients. When using a quantitative ELISA or some automated turbidimetric assays, D-dimer is highly sensitive (more than 95%) in excluding acute DVT or PE, frequently below a threshold of 500 ng/l. In other studies, concentrations even lower than this have ruled out VTE in patients with low or intermediate clinical probability (Righini M et al. 2008, Goldhaber SZ et al. 2012). Within the past few years, discussion has also centered around how POC testing can be used to improve the efficiency of emergency department operations. The POC D-dimer test has been reported to be associated with a shorter emergency department length of stay and fewer hospital admissions (E.Lee-Lewandrowski et al. 2009).

Role of helical computerized tomography (CT) in detection of RVD in APE patients and N-terminal pro-BNP in screening and follow-up in routine patient care

RVD assessed on TTE has been reported to be one of the strongest predictors of early mortality in non-massive PE (Goldhaber SZ 2002, Kreit JW 2004). TTE examination is not available around the clock at all institutions, which is a limitation to its use. In recent years, CTPA has become the diagnostic gold standard for PE. In this prospective study (III), CT proved to be a practical and valuable method for detection of RVD in clinical practice. The incidence of RVD in this study is in line with earlier reports of APE patients in whom RVD was detected in CT in 58-64% (Schoepf UJ et al. 2004, Van der Meer RW et al. 2005, He H et al. 2006). In this study, NT-proBNP was a strong indicator of RVD. A normal pro-BNP level on admission excluded the development of persistent pulmonary hypertension during follow-up. NT-proBNP values did not have an influence on survival, which probably can be explained by the non-high-risk patient population. The role of NT-

DISCUSSION

pro-BNP in predicting mortality risk could not be evaluated here. However, NT-proBNP on admission predicted future morbidity. Survival in this subject pool was good, even in the RVD-positive group. The restricted number of study patients does not, however, allow further conclusions about survival to be drawn.

Numerous papers exist concerning mortality from PE, whereas the late consequences of PE have received little attention. It remains unclear which patients should be followed after acute hospital stay and standard treatment. This study revealed that normal findings in CT as well as an NT-proBNP level ≤ 350 ng/l excluded RVD both in the acute phase and at the 7-month follow-up. This thesis may be an aid for clinicians when considering which patients might benefit from follow-up.

The cut-off level of ≤ 350 ng/l was chosen, although in our laboratory the references vary according to age and sex group, because values exceed 350 ng/l in all groups. Therefore, 350 ng/l is not a suitable cut-off point between “true” normal and abnormal values, instead indicating a slightly elevated value still low enough to detect patients who will benefit from a more intensive follow-up. Earlier studies of APE have used 500 ng/l (Pruszczyk P 2005) and 1000 ng/l (Binder L et al. 2005) as a cut-off for RVD.

It is notable that some clinical methods have retained a strong position in clinical practice despite the newer diagnostic technologies. Clinical probability assessment, including several symptoms, signs, and risk factors, has a central role in PE diagnostics. ECG and arterial blood gas analysis are still today valuable clinical tools in assessing the probability of RVD in APE.

CT and TTE were not performed simultaneously, which can be seen as a limitation of our study. However, the real-life situation is often the same. We wanted to investigate the role of immediate assessment of RVD by CT. The short-term prognostic significance of echocardiography in detecting RVD is under debate, even though several studies suggest that these patients are at higher risk for adverse events (Grifoni S et al. 2000, Ten Wolde M et al. 2004, Fremont B et al. 2008). The main purpose of detecting RVD in non-high-risk PE might be prognostic stratification to the low- or intermediate-risk groups (Torbicki A et al. 2008). A recent study suggests that patients with a severe thromboembolic episode might constitute a high-risk group for the development of CTPH, and these patients should be subjected to strict follow-up with echocardiography (Otero R et al. 2011). ESC guidelines recommend that all APE patients showing signs of RVD at any time during their hospital stay should receive a follow-up echocardiography usually 3-6 months after

DISCUSSION

discharge (Galiè N et al. 2011). AHA guidelines recommend that patients should be evaluated with echocardiography as early as 6 weeks after an acute PE to screen for persistent pulmonary hypertension (Jaff M et al. 2011).

Recently, CT has been used as a single procedure for diagnosis and risk stratification. Patients without right ventricular dysfunction at CT have a low risk of in-hospital adverse outcome (Becattini C et al. 2011). CT has also been useful in identifying high-risk patients associated with elevated biomarkers in APE where RVD/LVD showed a correlation with the level of NT-proBNP (Seon HJ et al. 2011). In this study, CT seems to be a reliable imaging method to screen RVD also in non-high-risk APE patients. All patients found to have chronic thromboembolic pulmonary hypertension at the 7-month follow-up had RVD in acute-phase CT. This indicates that when APE is diagnosed by CT, CT could be used also as a screening method to select patients who need to have a new RVD assessment later. After the 7-month follow-up, RVD was normal in the majority of patients (57/63 patients, 90%). Normal NT-proBNP had a good negative predictive value of RVD at the acute phase and at 7 months. Our study results show that CT tends to be a practical imaging method to detect RVD in APE. Furthermore, elevated NT-proBNP appears to be a good and valuable tool in assessing RVD in patients with APE both in the acute phase and during follow-up.

Based on our results, we suggest the following follow-up protocol. Patients with RVD on admission CT should be rescreened with natriuretic peptide assessment at follow-up and if elevated an echocardiography should be performed. Timing for screening seems reasonable at the visit when warfarin is to be discontinued. Alternatively, if anticoagulation therapy is to be continuous at discharge, then biomarker screening might be done during a routine follow-up visit 6-12 months after APE. The suggested protocol needs to be validated in a larger patient population.

Reduction of membrane component of diffusing capacity is associated with the extent of acute pulmonary embolism

DL, CO and especially DM are decreased in acute PE. At the 7-month follow-up, they remained lower than in healthy control subjects, especially in patients with persistent RVD. To the best of our knowledge, this was also the first study to use the CT-based

DISCUSSION

scoring of mass of embolism in relation to the components of diffusing capacity. This study yielded new information about the effects of PE on lung function.

A previous study (Fennerty et al.1988) used a ventilation-perfusion isotope scan with only a crude estimation of the size of PE. In the present study, a more exact characterization of the embolism mass was obtained. The results indicate that also the area of the alveolocapillary membrane affected by the thrombus, i.e. the extent of PE, is responsible for a reduction in DL, CO at the acute phase of PE, in addition to the effect of a reduction in lung volume. Loss of lung volume is important in the mechanism of decreased DL, CO explained by atelectasis, bronchoconstriction, and local pulmonary edema occurring in terminal respiratory units corresponding to the site of PE (Nadel JA et al. 1964).

DM/Vc was lower in patients with RVD than in those without RVD, suggesting a role for DM/Vc in the pathophysiology of RVD. Earlier studies (Oppenheimer BW 2006) have proposed that a reduction in the capillary volume in PE causes vascular dilatation and leads to a lower DM/Vc ratio. In this study, there were improvements in both DM and Vc, and thus, their relation might not change significantly at follow-up.

Catheter-directed thrombolysis of ileofemoral deep venous thrombosis reduces venous reflux

In 2003, when Study IV was performed, catheter-directed thrombolysis was a fairly new treatment method for DVT. We were living in a transition period, moving from systemic thrombolysis towards catheter-directed thrombolysis. Both methods were in use in clinical practice, which offered us an excellent opportunity to compare their late effects. The long-term goal of treatment of an acute DVT episode is mainly the prevention of PTS. Rapid thrombus resolution may offer potential for prevention of PTS based on its known favorable effect on the preservation of venous valvular function (Janssen et al. 2005). The study results showed that patients treated with catheter-directed thrombolysis in the acute phase had significantly less reflux at follow-up 2-3 years later.

In this retrospective, non-randomized study, the baseline status of patients was comparable. However, potential differences between the two groups cannot be excluded. Valvular competence was confirmed better in patients with catheter-directed thrombolysis

DISCUSSION

than in systemic thrombolysis-treated patients. Retrospective patient sampling did not allow a detailed analysis of imaging data before the treatment. A venography was performed on patients in the catheter-directed lysis group both before and after lysis. In the systemic lysis group, the imaging method was compression ultrasound, and a control imaging was only performed if the result of the lysis seemed clinically inadequate. Patients in the catheter-directed lysis-treated group were evaluated more precisely. The control venography also offered an immediate possibility for the interventional radiologist to detect an underlying lesion and use intravenous procedures, such as venous angioplasty and stenting, when needed.

Significantly less deep reflux and superficial reflux were observed in catheter-treated patients than in systemic-treated patients. These results are in line with other studies. The data of the systematic review of systemic and local-regional thrombolytic therapy suggest catheter-directed thrombolytic therapy decreases the incidence of post-thrombotic syndrome (Alesh I et al. 2007). In another systematic review and meta-analysis, besides a decrease in the incidence of post-thrombotic syndrome and venous obstruction, the results suggested that surgical thrombectomy seems to decrease the incidence of PTS and venous reflux (Casey ET et al. 2012). Clinical outcome at 2-3 years after catheter-directed or systemically administered thrombolytic therapy for iliofemoral DVT did not differ between the two treatment groups. However, a trend emerged towards less symptoms in the catheter-directed treatment group.

In recent years, the growing interest in endovascular technologies has led to the development of a variety of minimally invasive, catheter-based strategies to deal with venous thrombi. These technologies combine catheter-directed thrombolytic infusion, venous angioplasty, percutaneous mechanical thrombectomy, mechanical fragmentation, or ultrasound energy to remove the intraluminal thrombus (Lin PH et al. 2008, Nazir SA et al. 2009, Lin PH et al. 2010). Patients in whom thrombolytic treatment is being considered should undergo careful evaluation for contraindications of thrombolytic treatment and risks for bleeding as well as clinical evaluation of the severity of DVT. Selecting the patients, especially those at low risk of bleeding and with a high proximal DVT, who can benefit the most from aggressive thrombolytic treatment improves outcome, making the intervention more cost-effective (Enden T et al. 2013). Endovascular thrombolytic therapy procedures often require centralization to centers where an experienced interventional radiologist is available.

7. SUMMARY AND CONCLUSIONS

The main conclusions of the present studies can be summarized as follows:

1. The rapid and sensitive 10-min D-dimer assay seems to be suitable for excluding VTE in POC settings. Due to the high preliminary sensitivity and NPV obtained here, the assay could potentially be employed as a stand-alone test given that a sufficiently low cut-off level was selected. Alternatively, combined with clinical probability assessment, a higher cut-off level could be employed to increase the specificity of the assay.
2. CT is a practical and valuable imaging method for detection of RVD in non-high-risk APE patients. All patients found to have RVD at the 7-month follow-up had RVD also in the acute-phase CT and TTE. Furthermore, normal RV findings in CT as well as normal NT-proBNP level excluded RVD both in the acute phase and at the 7-month follow-up. According to study results, a follow-up protocol is suggested. Patients with RVD on admission CT should be rescreened with natriuretic peptide assessment at follow-up and if elevated an echocardiography is recommended.
3. The extent of PE and the consecutive signs of RVD were associated with a decrease in diffusion of membrane at both the acute phase and the resolution phase of PE independent of lung volume changes. In patients with permanent RVD, also the total diffusing capacity remained decreased in the follow-up. While the diffusing capacity recovered somewhat during the 7-month follow-up, it did not reach the level of healthy controls or the normal reference range. The data provide new knowledge of the consequences of PE on lung function and indicate that sustained reduction of diffusing capacity may remain despite treatment of PE according to current guidelines.
4. Catheter-directed thrombolysis for DVT at the acute event reduced both deep and superficial reflux at the 2- to 3-year follow-up. By reducing the incidence of venous reflux, the likelihood of developing PTS diminishes as well.

8. ACKNOWLEDGEMENTS

This work was carried out at the Department of Medicine, Division of Cardiology, Helsinki University Central Hospital.

I am sincerely grateful to the main supervisor of this thesis, my supervisor for about these years, Docent Veli-Pekka Harjola. Without his advice and support, I would not have completed this work. His expertise, enthusiasm, and never-ending patience have been the driving forces behind this work. I am also grateful to my other supervisor, Pirjo Mustonen. Her friendly support and constructive criticism during the review of the manuscripts and thesis have been invaluable. I also want to express my gratitude to Professor Markku S. Nieminen for giving me the opportunity to carry out this research at the department of Cardiology, and his support and interest to my work.

My gratitude is owed to Docent Matti Mänttari and Docent Heikki Ukkonen, the official referees of this thesis, for their extremely thorough review of my work, greatly improving the final product.

I am deeply grateful to my coauthor of the original publications, Marit Graner, who performed part of the echocardiograms and shared her expertise in statistics. I am indebted to Docent Päivi Piirilä for demanding analyses of clinical physiology and to Anneli Piilonen and Merja Raade for radiological expertise. I am grateful to Piia von Lode from the Department of Biotechnology, University of Turku, for being the driving force behind the immunofluorometric assays. I thank Professor Mauri Lepäntalo and Docent Aarno Lehtola for their expertise in venous diseases and Annamari Oinonen for good collaboration. I also thank all of the other coauthors of the original publications: Professor Anssi Sovijärvi, Norihide Sugano, Jarmo Rainaho, Professor Kari Punnonen, Olli Peltola, Professor Kim Pettersson, Wolf-Dieter Roth, Docent Pekka Keto, and Professor Seppo Sarna.

I am grateful to Professor Riitta Lassila for sharing her expertise in coagulation disorders. I thank Carol Ann Pelli for editing the English language of this manuscript.

I am grateful to all of the patients who volunteered in the study, making this work possible.

ACKNOWLEDGEMENTS

I owe my sincerest gratitude to my colleagues and the personnel of the Department of Cardiology and Emergency Care of Meilahti Hospital for helping me gather the study patients even in the middle of the night. I extend my gratitude to the staff of Helsinki University Central Hospital Laboratory, especially to Ulla Wartiovaara-Kautto for sharing her expertise in laboratory practice and Ellen Saarela for her friendly help in sample logistics and storing.

I am indebted to my former chiefs Antti Kohvakka and Päivi Koivuranta-Vaara for their flexibility and courage in making this work possible during this project. I am also grateful to my current employer Matti Toivola and Hannu Juvonen at the City of Helsinki Department of Social Services and Health Care for their support and interest in this study. My warmest thanks are owed to my colleagues and the staff at Helsinki Emergency Services. In particular, I thank Katariina Jantunen, Anne Weide and Marja Sippola-Soininen for supporting me through this project when I was combining clinical work and science.

I want to thank all my friends, especially my long-time friend Minna Aaltonen for her support and useful advice throughout the years.

I warmly thank my parents Kaarina and Seppo for their endless love and support throughout my life. I thank my sister Titta and her family, and my brother Jari for their generous love and support always. Your support in our everyday life has been essential for this study.

Finally, I owe my warmest grateful and love to my wonderful sons Patrik, Henrik, and Eerik for their patience with their mum. You have kept me attached to real life and have given me so much love and happiness over the years. I owe my thanks and love to Kari, who faithfully encouraged me to write my thesis and and took care of the daily routines while I was buried in work.

Financial support via grants from the Helsinki University Central Hospital Research Foundation, the Helsinki Social Services and Health Care Department, the Finnish Society for Angiology, the Aarne Koskelo Foundation, and the Orion Research Foundation is gratefully acknowledged.

Helsinki, February 2014

Mia Laiho

9. REFERENCES

- Adam SS, Key NS, Greenberg CS. D-Dimer antigen: Current concepts and future prospects. *Blood* 2009;113: 2878-87.
- Agnelli G, Becattini C. Acute pulmonary embolism. *N Engl J Med* 2010;363:266-274.
- Agnelli G, Buller HR, Cohen A, AMPLIFY-EXT Investigators et al. Apixaban for extended treatment of venous thromboembolism. *N Engl J Med*.2013;368:699-708.
- Agostoni P, Bussotti M, Cattadori G, Margutti E, Contini M, Muratori M, Marenzi G, Fiorentini C. Gas diffusion and alveolar-capillary unit in chronic heart failure. *Eur Heart J* 2006;27:2538–2543.
- Alesh I, Kayali F, Stein PD. Catheter-directed thrombolysis (intrathrombus injection) in treatment of deep venous thrombosis: a systematic review. *Catheter Cardiovasc Interv* 2007;70:143-8.
- Alikhan R, Peters F, Wilmott R, Cohen AT. Fatal pulmonary embolism in hospitalized patients: a necropsy review. *J ClinPathol* 2004;57:1254-1257.
- Alonso-Martínez JL, Urbieto-Echezarreta M, Anniccherico-Sánchez FJ, Abínzano-Guillén ML, Garcia-Sanchotena JL. N-terminal pro-B-type natriuretic peptide predicts the burden of pulmonary embolism. *Am J Med Sci* 2009;337:88-92.
- Anderson FA Jr, Spencer FA. Four topics in Venous Thromboembolism. *Circulation* 2003;107:I-9-I-16.
- Ansell J, Hirsch J, Hylek E, Jacobson A, Crowther M, Polaret G. Pharmacology and management of vitamin K antagonists. American College of Chest Physicians evidence-based clinical practice guidelines. 8thedn. *Chest* 2008;133:160S-98S.
- Araoz PA, Gotway MB, Harrington JR, Harmsen WS, Mandrekar JN. Pulmonary embolism: Prognostic CT findings. *Radiology* 2007;242:889-897.
- Araoz PA, Gotway MB, Trowbridge RL, Bailey RA, Auerbach AD, Reddy GP, Dawn SK, Webb WR, Higgins CB. Helical CT pulmonary angiography predictors of in-hospital morbidity and mortality in patients with acute pulmonary embolism. *J Thoracic Imaging* 2003;18:207-216.
- Arnesen H, Hoiseth A, Ly B. Streptokinase or heparin in the treatment of deep vein thrombosis: follow up results of a prospective study. *Acta Med Scand* 1982;211:65-71.

REFERENCES

- Aujesky D, Obrosky DS, Stone RA, Auble TE, Perrier A, Cornuz J, Roy PM, Fine MJ. Derivation and validation of a prognostic model for pulmonary embolism. *Am J Respir Crit Care Med* 2005;172:1041-46.
- Aujesky D, Roy PM, Verschuren F, Righini M, Osterwalder J, Egloff M, Renaud B, Verhamme P, Stone RA, Legall C, Sanchez O, Pugli NA, N`gako A, Cornuz J, Hugli O, Beer HJ, Perrier A, Fine MJ, Yealy DM. Outpatient versus inpatient treatment for patients with acute pulmonary embolism: an international, open-label, randomized, non-inferiority trial. *Lancet* 2011;378:41-48.
- Aviram G, Rogowski O, Gotler Y et al. Real-time risk stratification of patients with acute pulmonary embolism by grading the reflux of contrast medium into the inferior vena cava on computerized tomographic pulmonary angiography. *J Thromb Haemost* 2008;6:1488-1493.
- Bankier AA, Janata K, Fleischmann D, Kreuzer S, Mallek R, Frossard M, Domanovits H, Herold CJ. Severity assessment of acute pulmonary embolism with spiral CT: evaluation of two modified angiographic scores and comparison with clinical data. *J Thorac Imaging*. 1997;12:150-8.
- Becattini C, Agnelli G, Prandoni P, Silingardi M, Salvi R, Taliani MR, Poggio R, Imberti D, Ageno W, Pogliani E, Porro F, Casazza F. A prospective study on cardiovascular events after acute pulmonary embolism. *Eur Heart J* 2005;26:77-83.
- Becattini C, Agnelli G, Vedovati MC, Pruszczyk P, Casazza F, Grifoni S, Salvi A, Bianchi M, Douma R, Konstantinides S, Lankeit M, Duranti M. Multidetector computed tomography for acute pulmonary embolism: diagnosis and risk stratification in a single test. *Eur Heart J* 2011;32:1657-1663.
- Becattini C, Vedovati MC, Agnelli G. Prognostic value of troponins in acute pulmonary embolism: meta-analysis. *Circulation* 2007;116:427-33.
- Bernstein RJ, Ford RL, Clausen JL, Moser KM. Membrane diffusion and capillary blood volume in chronic thromboembolic pulmonary hypertension. *Chest* 1996;110:1430-1436.
- Binder L, Pieske B, Olschewski M, Geibel A, Klostermann B, Reiner C, Konstantinides S. N-terminal pro-brain natriuretic peptide or troponin testing followed by echocardiography for risk stratification of acute pulmonary embolism. *Circulation* 2005;112:1573-1579.

REFERENCES

- Bockenstedt P. D-Dimer in venous thromboembolism. *N Engl J Med* 2003;349:1203-1204.
- Bova C, Pesavento R, Marchiori A, Palla A, Enae I, Pengo V, Visonà A, Noto A, Prandoni P; TELESIO Study Group. Risk stratification and outcomes in hemodynamically stable patients with acute pulmonary embolism: a prospective multicentre, cohort study with 3 months of follow-up. *J Thromb Haemost* 2009;7:938-944.
- Bozic M, Stegnar M. Validation of an automated immunoturbidimetric assay for measurement of plasma D-Dimer. *Clin Chem Lab Med* 2003;41:958-62.
- Brandjes DP, Buller HR, Heijboer H, Huisman MV, de Rijk M, Jagt H, Ten Cate JW. Randomised trial of effect of compression stockings in patients with symptomatic proximal-vein thrombosis. *Lancet* 1997;349:759-62.
- British Thoracic Society Standards of Care Committee Pulmonary Embolism Guideline Development Group. British Thoracic Society guidelines for the management of suspected acute pulmonary embolism. *Thorax* 2003;58:470-483.
- Carson JL, Kelley MA, Duff A. The clinical course of pulmonary embolism. *N Engl J Med* 1992;326:1240-1245.
- Casey ET, Murad MH, Zumata-Garcia M, Elamin MB, Shi Q, Erwin PJ, Montori VM, Glociczki P, Meissner M. Treatment of acute iliofemoral deep vein thrombosis. *J Vasc Surg* 2012;55:1463-73.
- Cavallazzi R, Nair A, Vasu T, Marik PE. Natriuretic peptides in acute pulmonary embolism: a systematic review. *Intensive Care Med* 2008;34:2147-56.
- Chan KL, Currie PJ, Seward JB, Hagler DJ, Mair DD, Tajik AJ. Comparison of three Doppler ultrasound methods in the prediction of pulmonary artery pressure. *J Am Coll Cardiol* 1987;9:549-54.
- Cohen AT, Tapson VF, Bergmann JF, Goldhaber SZ, Kakkar AK, Deslandes B et al. Venous thromboembolism risk and prophylaxis in the acute hospital care setting (ENDORSE study): a multinational cross-sectional study. *Lancet*. 2008;371:387-94.
- Collomb D, Paramelle PJ, Calaque O, Bosson JL, Vanzetto G, Barnaoud D, Pison C, Coulomb M, Ferretti G. Severity assessment of acute pulmonary embolism: Evaluation using helical CT. *Eur Radiol* 2003;13:1508-1514.

REFERENCES

- Contractor S, Maldjian PD, Sharma VK, Gor DM. Role of helical CT in detecting right ventricular dysfunction secondary to acute pulmonary embolism. *J Comput Assist Tomogr* 2002;26:587-591.
- Costello P, Gupta KB. CT angiography gains acceptance in diagnosis of pulmonary emboli. *Diagn Imaging* 2000;22:43-45,49,87.
- Coulden R. State-of the art imaging techniques in chronic thromboembolic pulmonary hypertension. *Proc Am Thorac Soc* 2006;3:577-583.
- Dabestani A, Mahan G, Gardin JM, Takenaka K, Burn C, Allfie A, Henry WL. Evaluation of pulmonary artery pressure and resistance by pulsed Doppler echocardiography. *Am J Cardiol* 1987;59:662-8.
- Dellas C, Puls M, Lankeit M, Schäfer K, Cuvv M, Berner M, Hasenfuss G, Konstantinides S. Elevated heart-type fatty acid-binding protein levels on admission predict on adverse outcome in normotensive patients with acute pulmonary embolism. *J Am Coll Cardiol* 2010; 55:2150-2157.
- Dentali F, Douketis JD, Gianni M, Lim W, Crowther MA. Meta-analysis: anticoagulant prophylaxis to prevent symptomatic venous thromboembolism in hospitalized medical patients. *Ann Intern Med.*2007;146:278-88.
- De Stefano V, Finazzi G, Mannucci PM. Inherited thrombophilia: pathogenesis, clinical syndromes and management. *Blood* 1996;87(9):3531-44.
- Di Nisio M, Squizzato A, Rutjes AW, Büller HR, Zwinderman AH, Bossuyt PM. Diagnostic accuracy of D-Dimer test for exclusion of venous thromboembolism: a Systematic review. *J Throm haemost* 2007;5:292-304.
- Djurabi RK, Klok FA, Nijkeuter M, Kaasjager K, Kamphuisen PW, Kramer MH, Kruip MJ, Leebek FW, Büller HR, Huisman MV. Comparison of the clinical usefulness of two quantitative D-Dimer tests in patients with a low clinical probability of pulmonary embolism. *Thromb Res* 2009;123:771-4.
- Douketis JD, Kearon C, Bates S, Duku EK, Ginsberg JS. Risk of fatal pulmonary embolism in patients with treated venous thromboembolism. *JAMA* 1998;279:458-462.
- Douketis J, Tositto A, Marcucci M et al. Patients-level meta-analysis: effect of measurement timing, threshold, and patient age on ability of D-Dimer testing to assess recurrence risk after unprovoked venous thromboembolism. *Ann Intern Med* 2010;153:523-531.

REFERENCES

- Eerenberg E, Kamphuisen P, Sijpkens M, Meijers J, Buller H, Levi M. Reversal of rivaroxaban and dabigatran by prothrombin complex concentrate: a randomized, placebo-controlled, crossover study in healthy subjects. *Circulation* 2011;124:1573-1579.
- The Einstein investigators. Oral Rivaroxaban for symptomatic venous thromboembolism. *N Engl Med* 2010;363:2499-510.
- Elliot MS, Immelmann ES, Jeffrey P, Benatar SR, Funston MR, Smith JA, Shepstone BJ, Ferguson AD, Jacobs P, Walker W, Louw JH. A comparative randomized trial of heparin versus streptokinase in the treatment of acute proximal venous thrombosis: an interim report of a prospective trial. *Br J Surg* 1979;66:838-41.
- Enden T, Resch S, White C, Wik HS, Kloew NE, Sandset PM. Cost-effectiveness of additional catheter-directed thrombolysis for deep vein thrombosis. *J Thromb Haemost* 2013;11:1032-42.
- Farha S, Laskowski D, George D, Park MM, Tang WH, Dweik RA, Erzurum SC. Loss of alveolar membrane diffusing capacity and pulmonary capillary blood volume in pulmonary arterial hypertension. *Respir Res* 2013;14:6.
- Fennerty AG, Gunawardena KA, Smith AP. The transfer factor and its subdivisions in patients with pulmonary emboli. *Eur Respir J* 1981;98-101.
- Forster AJ, Wells PS. Tissue plasminogen activator for the treatment of deep venous thrombosis of the lower extremity: a systematic review. *Chest* 2001;119:572-9.
- Fraser JD, Anderson DR. Deep venous thrombosis: recent advances and optimal investigation with US. *Radiology* 1999;211:9-24.
- Galiè N, Hoeper MM, Humbert MM, Torbicki A, Vachjery JL, Barbera JA, Beghetti M, Corris P, Gaine S, Gibbs JS, Gomez-Sanchez MA, Jondeau G, Klepetko W, Opitz C, Peacock A, Rubin L, Zellweger M, Simonneau G. Guidelines for the Diagnosis and Treatment of Pulmonary Hypertension. The Task Force for the Diagnosis and Treatment of Pulmonary Hypertension of the European Society of Cardiology (ESC) and the European Respiratory Society (ERS), endorsed by the International Society of Heart and Lung Transplantation (ISHLT). *European Heart Journal* 2009;30:2493-2537.
- Geibel A, Zehender M, Kasper W, Olschewski M, Klima C, Konstantinides SV. Prognostic value of the ECG on admission in patients with acute major pulmonary embolism. *Eur Respir J* 2005;25:843-848.

REFERENCES

- Ghaye B, Ghuysen A, Bruyere PJ, D'orio V, Dondelinger RF. Can CT pulmonary angiography allow assessment of severity and prognosis in patients presenting with pulmonary embolism? What the radiologist needs to know. *Radiographics* 2006;26:23-39.
- Ghaye B, Ghuysen A, Willems V, Lambermont B, Grard P, D'Orío V, Gevenois PA, Dondelinger RF. Severe pulmonary embolism: Pulmonary artery clot load scores and cardiovascular parameters as predictors of mortality. *Radiology* 2006;239:884-891.
- Giannitsis E, Muller-Bardoff M, Kurowski V, Weidtmann B, Wiegand U, Kampmann M, Katus HA. Independent prognostic value of cardiac troponin T in patients with confirmed pulmonary embolism. *Circulation* 2000;102: 211-7.
- Giunta A, Tirri E, Maione S, Cangianiello S, Mele A, De Luca A et al. Right ventricular diastolic abnormalities in systemic sclerosis. Relation to left ventricular involvement and pulmonary hypertension. *Ann Rheum Dis* 2000;59:94-8.
- Goldhaber SZ, Bounameaux H. Pulmonary embolism and deep vein thrombosis. *Lancet* 2012;379:1835-46.
- Goldhaber SZ. Critical pathways in cardiology 2001. Pulmonary embolism chapter 19.
- Goldhaber SZ. Echocardiography in the management of pulmonary embolism. *Ann Intern Med* 2002;136:691-700.
- Goldhaber SZ, Haire WD, Feldstein ML, Miller M, Toltzis R, Smith JL, Taveira da Silva AM, Come PC, Lee RT, Parker JA. Alteplase versus heparin in acute pulmonary embolism: randomized trial assessing right-ventricular function and pulmonary perfusion. *Lancet* 1993;341:507-11.
- Goldhaber SZ. Risk factors for venous thromboembolism. *J Am Coll Cardiol* 2010; 56:1-7.
- Goldhaber SZ, Visani L, De Rosa M. Acute pulmonary embolism: clinical outcomes in the International Cooperative Pulmonary Embolism Registry (ICOPER). *Lancet* 1999;353:1386-1389.
- Golpe R, Perez-de-Llano LA, Castro-Añón O, Vázquez-Caruncho M, Conzález-Juanatey C, Veres-Racamonde A, Iglesias-Moreira C, Fariñas MC. Right ventricle dysfunction and pulmonary hypertension in haemodynamically stable pulmonary embolism. *Respir Med* 2010;104:1370-6.
- Gosselin RC, Wu JR, Kottke-Marchant K, Peetz D, Christie DJ, Muth H, Panacek E. Evaluation of the Stratus® CS Acute Care™ D-Dimer assay (DDMR) using the

REFERENCES

- Stratus® CS STAT Fluorometric Analyzer: A prospective multisite study for exclusion of pulmonary embolism and deep vein thrombosis. *Thromb Res* 2012;130:274-8.
- Grifoni S, Olivotto I, Cecchini P, Pieralli F, Camaiti A, Conti A, Agnelli G, Berni G. Short-term clinical outcome of patients with acute pulmonary embolism, normal blood pressure, and echocardiographic right ventricular dysfunction. *Circulation* 2000;101:2817-2822.
- Guyatt GH, Akl EA, Crowther M, Gutterman DD, Schünemann HJ; American College of Chest Physicians Antithrombotic Therapy and Prevention of Thrombosis Panel. Executive summary: Antithrombotic therapy and prevention of thrombosis, 9th ed: American College of Chest Physicians Evidence-Based Clinical Practice Guidelines. *Chest* 2012;141(Suppl):7S-47S.
- Haeger K. Problems of acute deep venous thrombosis. I. The interpretation of signs and symptoms. *Angiology* 1969;20:219-223.
- Hardy RW, Hortin GL. Point-of-care testing in patient transport settings. In: Point-of-care testing, 2nd ed. Price CP, St John A, Hicks JM(eds) pp. 221-5. AACC Press, Washington 2004.
- Heit JA, Melton LJ, III, Lohse CM, Pettersson TM, Silverstein MD, Mohr DN, O'Fallon WM. Incidence of venous thromboembolism in hospitalized patients vs. community residents. *Mayo Clin Proc* 2001;76:1102–10.
- Heit JA, Mohr DN, Silverstein MD, Pettersson TM, O'Fallon WM, Melton LJ 3rd. Predictors of recurrence after deep vein thrombosis and pulmonary embolism: a population-based cohort study. *Arch Intern Med* 2000;160:761-768.
- Heit JA, Silverstein MD, Mohr DN, Pettersson TM, O'Fallon WM, Melton LJ 3rd. Predictors of survival after deep vein thrombosis and pulmonary embolism: a population-based cohort study. *Arch Intern Med* 1999;159:445-453.
- Hicks JM, Haeckel R, Price CP, Lewandrowski K, Wu AH. Recommendations and opinions for the use of point-of-care testing for hospitals and primary care: summary of a 1999 symposium. *Clin Chim Acta* 2001;303:1-17.
- Hirsh J. Comparison of the relative efficacy and safety of low molecular weight heparin and unfractionated heparin for the treatment of deep venous thrombosis. *Semin Hematol* 1997;34:20-5.
- Hoeper MM, Mayer E, Simonneau G, Rubin L. Chronic Thromboembolic Pulmonary Hypertension. *Circulation* 2006;113: 2011-2020.

REFERENCES

- Hoffman GE. Concepts for the third generation of laboratory systems. *Clin Chim Acta* 1998;278:203-16.
- He H, Stein M, Zalta B, Haramati L. Computed tomography evaluation of right heart dysfunction in patients with acute pulmonary embolism. *J Comput Assist Tomogr* 2006;30:262-266.
- Hsia CC. Recruitment of lung diffusing capacity: update of concept and application. *Chest* 2002;122: 1774–1783.
- Hsu JT, Chu CM, Chang ST, Cheng HW, Cheng NJ, Ho WC, Chung CM. Prognostic role of alveolar-arterial oxygen pressure difference in acute pulmonary embolism. *Circ J*. 2006;70:1611-6.
- Hull R, Delmore T, Genton F, Hirsch I, Gent M, Sackett D, McLoughlin D, Armstrong P. Warfarin sodium versus low-dose heparin in the long term treatment of venous thrombosis. *N Engl Med* 1979;301:855-858.
- Hull RD, Raskob GE, Leclerc J, Jay RM, Hirsh J. The diagnosis of clinically suspected venous thrombosis. *Clin Chest Med* 1984;5:439-452.
- Hyers TM, Agnelli G, Hull RD, Morris TA, Samama M, Tapson V, Weg JG. Antithrombotic therapy for venous thromboembolic disease. *Chest* 2001;119:S176-93.
- Jaff M, McMurtry S, Archer S, Cushman M, Goldenberg N, Goldhaber SZ, Jenkins JS, Kline JA. A Scientific Statement From the American Heart Association. Management of Massive and Submassive Pulmonary Embolism, Iliofemoral Deep Vein Thrombosis, and Chronic Thromboembolic Hypertension. *Circulation* 2011;123:1788-1830.
- Janssen MC, Wollersheim H, Schutze-Kooli LJ, Thien T. Local and systemic thrombolytic therapy for acute DVT. *The Neth J Med* 2005;63:81-90.
- Jiménez D, Aujesky D, Moores L, Gómez V, Martí D, Briongos S, Monreal M, Barrios V, Konstantinides S, Yusen RD. Simplification of the pulmonary embolism severity index for prognostification in patients with acute symptomatic pulmonary embolism. *Arch Intern Med* 2010;170:1383-89.
- Jiménez D, Diaz G, Molina I, Martí D, Del Rey I, García-Rull, Escobar C, Vidal R, Sueiro A, Yusen RD. Troponin I and risk stratification of patients with acute non-massive pulmonary embolism. *Eur Respir J* 2008; 31:847-853.
- Jiménez D, Uresandi F, Otero R, Lobo JL, Monreal M, Martí D, Zamora J, Muriel A, Aujesky D, Yusen R. Troponin-based risk stratification of patients with acute

REFERENCES

- nonmassive pulmonary embolism. Systematic review and meta-analysis. *Chest* 2009;136:974-982.
- Kahn SR. How I treat postthrombotic syndrome. *Blood* 2009;114: 4624-4631.
- Kaczynska A, Pelsers MM, Bochowicz A, Kostrubier M, Glatz IF, Pruszczvk P. Plasma heart-type fatty acid binding protein is superior to troponin and myoglobin for rapid risk stratification in acute pulmonary embolism. *Clin Chim Acta* 2006;371:117-123.
- Kakkar VV, Howe CT, Nicolaidis AN, Renney JT, Clarke MB. Deep vein thrombosis of leg. Is there a "high risk" group? *Am J Surg* 1970;120:527-30.
- Kasper W, Konstantinides S, Geibel A, Tiede N, Krause T, Just H. Prognostic significance of right ventricular afterload stress detected by echocardiography in patients with clinically suspected pulmonary embolism. *Heart* 1997;77:346-9.
- Kearon C, Ginsberg JS, Hirsh J. The role of venous ultrasonography in the diagnosis of suspected deep venous thrombosis and pulmonary embolism. *Ann Intern Med* 1998;129:1044-1049.
- Kearon C, Ginsberg JS, Kovacs MI, Anderson DR, Wells P, Julian JA, MacKinnon B, Weitz II, Crowther MA, Dolan S, Turpie AG, Geerts W, Solvmoss S, van Nouven P, Demers C, Kahn SR, Kassis I, Rodger M, Hambleton I, Gent M: Extended Low-Intensity Anticoagulation for Thrombo-Embolism Investigators. Comparising of low-intensity warfarin therapy with conventional-intensity warfarin therapy for long-term prevention of recurrent venous thromboembolism. *N Engl J Med* 2003;349:631-639.
- Kearon C, Julian JA, Newman TE, Ginsberg JS. Non invasive diagnosis of deep vein thrombosis. *Mc Master Diagnostic Imaging Practice Guidelines Initiative. Ann Intern Med.* 1998;128:663-677.
- Kearon C, Kahn SR, Agnelli G, Goldhaber SZ, Raskob GE, Comerota AJ. Antithrombic therapy in neonates and children: American College of Chest Physicians Evidence-Based Clinical Practice Guidelines (8th edition). *Chest* 2008;133:454S-545S.
- Kearon C. Natural history of venous thromboembolism. *Circulation* 2003;107:122-130.
- Killewich LA, Bedford GR, Beach KW, Strandness DE Jr. Spontaneous lysis of deep venous thrombi: rate and outcome. *J Vasc Surg* 1989;9:89-97.
- Kirkbride RE. Point-of-care testing in community. In: *Point-of-care testing*, 2nd ed. Price CP, St John A, Hicks JM (eds) pp. 353-60. AACC Press, Washington, 2004.

REFERENCES

- Kline JA, Hernandez-Nino, Rose GA, Norton HI, Camargo CA Jr. Surrogate markers for adverse outcomes in normotensive patients with pulmonary embolism. *Crit Care Med* 2006;34:2773-2780.
- Kline JA, Zeitouni R, Marchick MR, Hernandez-Nino, Rose GA. Comparison of 8 biomarkers for prediction of right ventricular hypokinesis 6 months after submassive pulmonary embolism. *Am Heart J* 2008;156:308-314.
- Kline JA, Steuerwald MT, Marchick MR, Hernandez-Nini J, Rose GA. Prospective evaluation of right ventricular function and functional status 6 months after acute submassive pulmonary embolism. *Chest* 2009;136:1202-1210.
- Knecht MF, Heinrich F. Clinical evaluation of an immunoturbidimetric D-Dimer assay in the diagnostic procedure of deep vein thrombosis and pulmonary embolism. *Thromb Res* 1997;88:413-7.
- Kostrubiec M, Pruszczyk P, Bochowicz A, Pacho R, Szulc M, Kaczynska A, Styczynski G, Kuch-Wocial A, Abramczyk P, Bartoszewicz Z, Berent H, Kuczynska K. Biomarker-based risk assessment model in acute pulmonary embolism. *Eur Heart J* 2005;26:2166-72.
- Kostrubier M, Pruszczyk P, Kaczynska A, Kucher N. Persistent NT-pro-BNP elevation in acute pulmonary embolism predicts early death. *Clin Chim Acta* 2007;382:124-128.
- Kreit JW. The impact of right ventricular dysfunction on the prognosis and therapy of normotensive patients with pulmonary embolism. *Chest* 2004;125:1539-45.
- Kruip MJ, Slob MJ, Schijen JH, van der HC, Buller HR. Use of a clinical decision rule in combination with D-Dimer concentration in diagnostic workup of patients with suspected pulmonary embolism: a prospective management study. *Arch Intern Med* 2002;162:1631-1635.
- Krüger S, Graf I, Merx MW, Koch KC, Kunz D, Hanrath P, Jansses U. Brain natriuretic peptide predicts right heart failure in patients with acute pulmonary embolism. *Am Heart J* 2004;147:60-65.
- Kucher N, Printzen G, Doernhoefer T, Windecker S, Meier B, Hess OM. Low pro-brain natriuretic peptide levels predict benign clinical outcome in acute pulmonary embolism. *Circulation* 2003;107:1576-78.
- Kucher N, Printzen G, Goldhaber SZ. Prognostic role of brain natriuretic peptide in acute pulmonary embolism. *Circulation* 2003;107:2545-47.

REFERENCES

- Kucher N, Rossi E, De Rosa M, Goldhaber SZ. Massive pulmonary embolism. *Circulation* 2006;113:577-82.
- Kurzyna M, Torbicki A, Pruszczyk P, Burakowska B, Fijalkowska A, Kober J, Oniszh K, Kuca P, Tomkowski W, Burakowski J, Wawrzyńska L. Disturbed right ventricular ejection pattern as a new Doppler echocardiographic sign of acute pulmonary embolism. *Am J Cardiol* 2002;90:507-511.
- Landefeld CS, Hanus P. Economic burden of venous thromboembolism. Prevention of venous thrombosis. New York; Dekker.1993; 69-85.
- Lee-Lewandrowski E, Lewandrowski K. Perspectives on cost and outcomes for point-of-care testing. *Clin Lab Med* 2009;29:479-89.
- Lee-Lewandrowski E, Nichols J, Van Cott E, Grisson R, Louissaint A, Benzer T, Lewandrowski K. Implementation of a rapid whole blood D-Dimer test in the emergency department of an urban academic medical center. *Am J Clin Pathol* 2009;132:326-331.
- Le Gal G, Righini M, Roy PM et al. Prediction of pulmonary embolism in the emergency department: the revised Geneva score. *Ann Intern Med* 2006;144:165-71.
- Legnani C, Cini M, Scarvelis D, Toulon P, Wu JR, Palareti G. Multicenter evaluation of a new quantitative highly sensitive D-Dimer assay, the Hemosil D-Dimer HS 500, in patients with clinically suspected venous thromboembolism. *Thromb Res* 2010; 125:398-401.
- Levine MN, Hirsch J, Gent M, Turnie AG, Weitz J, Ginsberg J, Geerts W, LeClerc J, Neemeh I, Powers P, Piovella F. Optimal duration of oral anticoagulant therapy: a randomized trial comparing four weeks with three months of warfarin in patients with proximal deep vein thrombosis. *Thromb Haemost* 1995;74:606-611.
- Lindhoff-Last E, Samama M, Ortel T, Weitz J, SpiroT. Assays for measuring rivaroxaban: their suitability and limitations. *Ther Drug Monit* 2010;32:673-679.
- Lin PH, Barshes NR, Annambhotla S, Koungias P, Huynh TT. Advances in endovascular interventions for deep vein thrombosis. *Expert Rev Med Devices* 2008;5:153-66.
- Lin PH, Ochoa LN, Duffy P. Catheter-directed thrombectomy and thrombolysis for symptomatic lower-extremity deep vein thrombosis: a review of current interventional treatment strategies. *Perspect Vasc Surg Endovasc Ther* 2010;22:152-63.

REFERENCES

- Logeart D, Lecuver I, Thabut G, Tabet IY, Tartiere IM, Chavelas C, Bonnin F, Stievenart II, Solal AC. Biomarker-based strategy for screening right ventricular dysfunction in patients with non-massive pulmonary embolism. *Intensive Care Med* 2007;33:286-292.
- Macintyre N, Crapo RO, Viegi G, Johnson DC, van der Grinten CP, Brusasco V, Burgos F, Casaburi R, Coates A, Enright P, Gustafsson P, Hankinson J, Jensen R, McKay R, Miller MR, Navajas D, Pedersen OF, Pellegrino R, Wanger J. Standardisation of the single-breath determination of carbon monoxide uptake in the lung. *Eur Respir J*. 2005;26:720–735.
- Markel A, Manzo RA, Bergelin RO, Strandness DE Jr. Incidence and time of occurrence of valvular incompetence following deep vein thrombosis. *Wien Med Wochenschr* 1994;144:216-20.
- Mastora I, Remy-Jardin M, Masson P, Galland E, Delannoy V, Bauchart JJ, Remy J. Severity of acute pulmonary embolism: evaluation of a new spiral CT and angiographic scores in correlation with echocardiographic data. *Eur Radiol* 2003;13:29-35.
- Mavrakanas T, Bounameaux H. The potential role of new oral anticoagulants in the prevention and treatment of thromboembolism. *Pharmacol Ther* 2011;130:46-58.
- Maziere F, Birolleau S, Medimagh S, Arthaud M, Bennaceur M, Riou B, Rav P. Comparison of troponin I and N-terminal pro B-type natriuretic peptide for risk stratification in patients with pulmonary embolism. *Eur J Emerg Med* 2007;14:207-211.
- McConnell MV, Salomon SD, Ryan ME, Come PC, Goldhaber SZ, Lee RT. Regional right ventricular dysfunction detected by echocardiography in acute pulmonary embolism. *Am J Cardiol* 1996;78:469- 473.
- McIntyre KM, Sasahara AA. The hemodynamic response to pulmonary embolism in patients without prior cardiopulmonary disease. *Am J Cardiol* 1971;28:288-294.
- Mewissen MW, Seabrook GR, Meissner MH, Cynamon J, Labropoulos N, Haughton SH. Catheter-directed thrombolysis for lower extremity deep venous thrombosis: report of a national multicenter registry. *Radiology* 1999;211:39-49.
- Mewissen MW. Thrombolysis for lower-extremity deep vein thrombosis. *Semin Vasc Surg* 2010;23:228-34.
- Meyer M, Schuster KD, Schulz H, Mohr M, Piiper J. Pulmonary diffusing capacities for nitric oxide and carbon monoxide determined by rebreathing in dogs. *J Appl Physiol* 1990;68: 2344–2357.

REFERENCES

- Moheimani F, Jackson DE. Venous thromboembolism: classification, risk factors, diagnosis and management. International Scholarly Research Network. ISRN Hematology 2011, article ID 124610, 7 pages.
- Moser KM, Auger WR, Fedullo PF. Chronic major-vessel thromboembolic pulmonary hypertension. *Circulation* 1990;81:1735-1743.
- Murin S, Romano PS, White RH. Comparison of outcome after hospitalization for deep vein thrombosis or pulmonary embolism. *Thromb Haemost* 2002;88:407-414.
- Nadel JA, Colebatch HJH, Olsen CR. Location and mechanism of airway constriction after barium sulphate microembolism. *J Appl Physiol* 1964;19:387-394.
- Naess IA, Christiansen SC, Romunstad P, Cannegieter SC, Rosendaal FR, Hammerstrøm J. Incidence and mortality of venous thrombosis: a population-based study. *Journal of Thrombosis and Haemostasis* 2007;5:692-699.
- Nazir SA, Ganeshan A, Nazir S, Uberoi R. Endovascular treatment options in the management of lower limb deep venous thrombosis. *Cardiovasc Intervent Radiol* 2009;32:861-76.
- O'Donnell T, Abbott W, Athanasoulis C, Millan V, Callow A. Diagnosis of deep venous thrombosis in the outpatient by venography. *Surg Gynecol Obstet* 1980;150:69-74.
- Omnen SR, Nishimura RA, Hurrell DG, Klarich KW. Assessment of right atrial pressure with 2-dimensional and Doppler echocardiography: a simultaneous catheterization and echocardiographic study. *Mayo Clin Proc* 2000;75:24-9.
- Oppenheimer BW, Berger KI, Hadjiangelis NP, Norman RG, Rapoport DM, Goldring RM. Membrane diffusion in diseases of the pulmonary vasculature. *Respir Med* 2006;100:1247-53.
- Otero R, Oribe M, Ballaz A, Jimenénez D, Uresandi F, Nauffal D, Conget F, Rodriguez C, Elias T, Jara L, Cayuela A, Blanco I, Barberá J. Echocardiographic assessment of pulmonary arterial pressure in the follow-up of patients with pulmonary embolism. *Thromb Res* 2011;127:303-8.
- Palareti G, Cosmi B, Legnani C. Diagnosis of deep vein thrombosis. *Semin Thromb Hemost* 2006;32:659-72.
- Palareti G, Cosmi B, Legnani C, Tositto A, Brusi C, Lorio A, Pengo V, Ghirarduzzi A, Pattacini C, Testa S, Lensing AW, Tripodi A. D-Dimer testing determine the duration of anticoagulant therapy. *N Engl J Med* 2006;355:1780-9.

REFERENCES

- Palmieri V, Gallotta G, Rendina D, De Bonis S, Russo V, Postiglione A, Martino s, Di Minno MN, Celetano A. Troponin I and right ventricular dysfunction for risk assessment in patients with nonmassive pulmonary embolism in the emergency department in combination with clinically based risk score. *Intern Emerg Med* 2008;3:131-138.
- Pasha SM, Klok FA, Snoep JD, Mos IC, Goekoop RJ, Rodger MA, Huisman MV. Safety of excluding acute pulmonary embolism based on an unlikely clinical probability by the Wells rule and normal D-Dimer concentration: a meta-analysis. *Thromb Res* 2010; 125:123-7.
- Pengo V, Lensing AW, Prins MH, Machiori A, Davidson BL, Tiozzo F, Albanese P, Biasiolo A, Pegoraro C, Iliceto S, Prandoni P, and for the thromboembolic pulmonary hypertension Study Group. Incidence of chronic thromboembolic pulmonary hypertension after pulmonary embolism. *N Engl Med* 2004;350:2257-2264.
- Perrier A, Bounameaux H. Ultrasonography of leg veins in patients suspected of having pulmonary embolism. *Ann Intern Med* 1998;128:243-245.
- Perrier A, Desmarais S, Miron MJ, de Moerloose P, Lepage R, Slosman D, Didier D, Unger PF, Junod A, Patenaude JV, Bounameaux H. Non-invasive diagnosis of venous thromboembolism in outpatients. *Lancet* 1999;353:190-195.
- Perrier A, Roy PM, Aujesky D, Chagnon I, Howarth N, Gourdier AL, Leftheriotis G, Barghouth G, Cornuz J, Hayoz D, Bounameaux H. Diagnosing pulmonary embolism in outpatients with clinical assessment, D-Dimer measurements, venous ultrasound, and helical computed tomography: a multicenter management study. *Am J Med* 2004;116:291-299.
- Pesavento R, Villalta S, Pandoni P. The postthrombotic syndrome. *Intern Emerg Med* 2010; 5:185-92.
- Piazza G, Goldhaber SZ. Venous Thromboembolism and atherothrombosis: an integrated approach. *Circulation* 2010;121:2146-50.
- Pieralli F, Olivotto I, Vanni S, Conti A, Camaiti A, Targioni G, Grifoni S, Berni G. Usefulness of bedside testing for brain natriuretic peptide to identify right ventricular dysfunction and outcome in normotensive patients with acute pulmonary embolism. *Am J Cardiol* 2006;97:1386-1390.
- Popuri RK, Vedantham S. The role of thrombolysis in the clinical management of deep vein thrombosis. *Arterioscler Thromb Vasc Biol* 2011;31:479-484.

REFERENCES

- Porter JM, Moneta GL, and an international Concensus Committee on Chronic Venous Disease. Reporting standards in venous disease: an update. *J Vasc Surg* 1995;21:635-645.
- Poulsen SH, Noer I, Moller JE, Knudsen TE, Frandsen JL. Clinical outcome of patients with suspected pulmonary embolism. A follow-up study of 588 consecutive patients. *J Intern Med* 2001;250:137-143.
- Prandoni P, Bilura F, Marchiori A, Bernardi E, Petrobelli F, Lensing AW, Prins MH, Girolami A. An association between atherosclerosis and venous thrombosis. *N Engl J Med* 2003;348:1435-1441.
- Prandoni P, Lensing AW, Cogo A, Cuppini S, Villalta S, Carta M, Cattelan AM, Polistena P, Bernardi F, Prins MH. The long-term clinical course of acute deep venous thrombosis. *Ann Intern Med* 1996;125:1-7.
- Prandoni P, Lensing AW, Prins MH, Frulla M, Marchiori A, Bernardi E, Tormene D, Mosena L, Pagnan A, Girolami A. Below-knee elastic compression stockings to prevent the post-thrombotic syndrome. *Ann Intern Med* 2004;141:249-256.
- Price CP, St John A, Hicks JM. Point-of-care testing: what, why and where? In: Point-of-care testing, 2nd ed. Price CP, St John A, Hicks JM (eds) pp 3-9. AACC Press, Washington 2004.
- Pruszczyk P, Kostrubier M, Bochowicz A, Styczynski G, Szulc M, Kurzwina M, Fialkowska A, Kuch-Wocial A, Chlwicka I, Torbicki A. N-terminal pro-brain natriuretic peptide in patients with acute pulmonary embolism. *Eur Respir J* 2003;22:649-653.
- Pruszczyk P. N-terminal Pro-Brain natriuretic peptide as an indicator of right ventricular dysfunction. *J Card Fail* 2005;11:65-69.
- Puls M, Dellas C, Lankeit M, Olschewski M, Binder I, Geibel A, Reiner C, Schäfer K, Hasenfuss G, Konstantinides S. Heart-type fatty acid-binding protein permits early risk stratification of pulmonary embolism. *Eur Heart J* 2007;28:224-229.
- Puwanant S, Park M, Popovic ZB, Tang WH, Farha S, George D, Sharp J, Puntawangkoon J, Loyd JE, Erzurum SC, Thomas JD. Ventricular geometry, strain, and rotational mechanics in pulmonary hypertension. *Circulation* 2010;121:259-266.
- Qaseem A, Snow V, Barry P, Hornbake EH, Rodnick JE, Tobolic T, Irdand B, Segal JB, Bass EB, Weiss KB, Green L, Owens DK. Joint American Academy of Family Physicians/American College of Physicians panel on deep venous

REFERENCES

- thrombosis/pulmonary embolism. Current Diagnosis of venous thromboembolism in primary care: A clinical practice guideline from the American Academy of Family Physicians and the American College of Physicians. *Ann Fam Med* 2007;5:57-62.
- Quiroz R, Kucher N, Schoepf UJ, Kipmueller F, Solomon SD, Costello P, Goldhaber SZ. Right ventricular enlargement on chest computed tomography: Prognostic role in acute pulmonary embolism. *Circulation* 2004;109:2401-2404.
- Raptopoulos V, Boiselle PM. Multi-detector row spiral CT pulmonary angiography: comparison with single-detector row spiral CT. *Radiology* 2001;221:606-13.
- Raskob G, Buller H, Prins M et al. The Hokusai-VTE Investigators. Edoxaban for the long-term treatment of venous thromboembolism: rationale and design of the Hokusai-VTE study-methodological implications for clinical trials. *J Thromb Haemost*. 2013;11:1287-94
- Rav P, Maziere F, Medimagh S, Lefort Y, Arthaud M, Duquet A, Teixeira A, Riou R. Evaluation of B-type natriuretic peptide to predict complicated pulmonary embolism in patients aged 65 years and older: Brief report. *Am J Emerg Med* 2006;24:603-607.
- Reid JH, Murchison JT. Acute right ventricular dilatation: a new helical CT sign of massive pulmonary embolism. *Clin Radiol* 1998;53:694-698.
- Ribeiro A, Lindmarker P, Johnsson H, Juhlin-Dannfelt A, Jorfeldt L. Pulmonary embolism: one year follow-up with echocardiography Doppler and five-year survival analysis. *Circulation* 1999;99:1325-1330.
- Ribeiro A, Lindmarker P, Juhlin-Dannfelt A, Johnsson H, Jorfeldt L. Echocardiography Doppler in pulmonary embolism: right ventricular dysfunction as a predictor of mortality rate. *Am Heart J* 1997;134:479-87.
- Ridker PM, Goldhaber SZ, Danielson F, Rosenberg Y, Fhv CS, Deitcher SR, Cushman M, Moll S, Kessler CM, Elliot CG, Paulson R, Wong T, Bauer KA, Schwartz BA, Miletich JP, Bounameaux H, Glynn RJ; PREVENT Investigators. Long-term, low-intensity warfarin therapy for the prevention of recurrent venous thromboembolism. *N Engl J Med* 2003;348:1425-1434.
- Riedel Marin. *Heart* vo 85, 1. Feb 2001: 229-240.
- Righini M, Le Gal G, Perrier A, Bounameaux H. The challenge of diagnosing pulmonary embolism in elderly patients: influence of age on commonly used diagnostic tests and strategies. *J Am Geriatr Soc* 2005;53:1039-45.

REFERENCES

- Righini M, Perrier A, De Moerloose P, Bounameaux H. D-Dimer for venous thromboembolism diagnosis: 20 years later. *J Thromb Haemost* 2008;6:1059-71.
- Rodger M, Makropoulos D, Turek M, Quevillon J, Raymond F, Rasuli P, Wells PS. Diagnostic value of the electrocardiogram in suspected pulmonary embolism. *Am J Cardiol* 2000;86:807-9, A10.
- Rosendaal FR, Reitsma PH. Genetics of venous thrombosis. Review. *J Thromb Haemost*. 2009;7:301-4.
- Roughton FJ, Forster RE. Relative importance of diffusion and chemical reaction rates in determining rate of exchange of gases in the human lung, with special reference to true diffusing capacity of pulmonary membrane and volume of blood in the lung capillaries. *J Appl Physiol* 1957;11:290–302.
- Rutherford RB, Padberg FT, Camerota AJ, Kistner RL, Meissner MH, Moneta GL. Venous severity scoring: adjunct to venous outcome assessment. *J Vasc Surg* 2000;21:1307-1312.
- Sackner MA, Greenelch D, Heiman MS, Epstein S, Atkins N. Diffusing capacity, membrane diffusing capacity, capillary blood volume, pulmonary tissue volume, and cardiac output measured by a rebreathing technique. *Am Rev Respir Dis* 1975;111:157–165.
- Samama M, Guinet C. Laboratory assessment of new anticoagulants. *Clin ChemLab Med* 2011;49:761-772.
- Scarvelis D, Wells PS. Diagnosis and treatment of deep-vein thrombosis. Review. *CMAJ* 2006;175(9):1087-92.
- Schoepf UJ, Becker CR, Hofmann LK, Das M, Flohr T, Ohnesorge BM, Baumert B, Rolnick J, Allen JM, Raptopoulos V. Multislice CT angiography. *Eur Radiol* 2003;13:1946-1961.
- Schoepf UJ, Costello P. CT angiography for diagnosis of pulmonary embolism: State of the art. *Radiology* 2004;230:329-337.
- Schoepf UJ, Goldhaber SZ, Costello P. Spiral computed tomography for acute pulmonary embolism. *Circulation* 2004;109:2160-2167.
- Schoepf UJ, Holzknicht N, Helmberger TK, Crispin A, Hong C, Becker CR, Reiser MF. Subsegmental pulmonary emboli: improved detection with thin-collimation multi-detector row spiral CT. *Radiology* 2002;222:483-490.

REFERENCES

- Schoepf UJ, Kucher N, Kipfmüller F, Quiroz R, Costello P, Goldhaber S. Right ventricular enlargement on chest computed tomography. A predictor of early death in acute PE. *Circulation* 2004;110:3276-3280.
- Schulman S, Kakkar AK, Schellong SM et al. Randomized Trial of Dabigatran versus warfarin in the treatment of acute venous thromboembolism (RE-COVER II) *Blood* 2011;118:95-96.
- Schulman S, Kearon C, Kakkar AK, Mismetti P, Schellong S, Eriksson H, Baanstra D, Schnee J, Goldhaber SZ; RE-COVER Study Group. Dabigatran versus warfarin in the treatment of acute venous thromboembolism. *N Engl J Med* 2009;361:2342-2352.
- Schulman S, Kearon C, Kakkar AK, RE-MEDY trial investigators et al. Extended use of dabigatran, warfarin, or placebo in venous thromboembolism. *N Engl J Med* 2013;368:709-718.
- Scridon T, Scridon C, Skali H, Alvarez A, Goldhaber SZ, Solomon SD. Prognostic significance of troponin elevation and right ventricular enlargement in acute pulmonary embolism. *Am J Cardiol* 2005;96:303-305.
- Segal JB, Eng J, Tamariz LJ, Bass EB. Review of the evidence on diagnosis of deep venous thrombosis and pulmonary embolism. *Ann Fam Med* 2007;5:63-73.
- Semba CP, Dake MD. Iliofemoral deep venous thrombosis: aggressive therapy with catheter-directed thrombolysis. *Radiology* 1994;191:487-494.
- Seon HJ, Kim KH, Lee WS, Choi S, Yoon HJ, Ahn Y, Kim Y-H, Jeong MH, Cho JG, Park JC, Kang JC. Usefulness of computed tomographic pulmonary angiography in the risk stratification of acute pulmonary embolism. *Circ J* 2011;75:428-436.
- Sharma GV, Burleson VA, Sasahara AA. Effect of thrombolytic therapy on pulmonary-capillary blood volume in patients with pulmonary embolism. *N Engl J Med* 1980;303:842-845.
- Sharma GV, Folland ED, McIntyre KM, Sasahara AA. Long-term benefit of thrombolytic therapy in patients with pulmonary embolism. *Vasc Med* 2000;5:91-95.
- Siegel DM, Cuker A. Reversal of novel oral anticoagulants in patients with major bleeding. *J Thromb Thrombolysis* 2013;35:391-8.
- Snyder EM, Johnson BD, Beck KC. An open-circuit method for determining lung diffusing capacity during exercise: comparison to rebreathe. *J Appl Physiol* 2005;99:1985-1991.

REFERENCES

- Stam H, Versprille A, Bogaard JM. The components of the carbon monoxide diffusing capacity in man dependent on alveolar volume. *Bull Eur Physiopath Respir* 1983;19:17–22.
- Steenhuis LH, Groen HJ, Koeter GH, van der Mark TW. Diffusion capacity and haemodynamics in primary and chronic thromboembolic pulmonary hypertension. *Eur Respir J* 2000;16:276–281.
- Stein PD, Golhhaber SZ, Henry JW, Miller AC. Arterial blood gas analysis in the assessment of suspected acute pulmonary embolism. *Chest* 1996;109:78-81.
- Stein PD, Hull RD, Patel KC, Olson RE, Ghali WA, Brant R, Biel RK, Bharadia V, Kalra NK. D-Dimer for the exclusion of acute venous thrombosis and pulmonary embolism: a systematic review. *Ann Intern Med.* 2004;140:589-602.
- Stein PD, Terrin ML, Hales CA, Palevsky HI, Saltzman HA, Thompson BT, Weg JG. Clinical, laboratory, roentgenographic, and electrocardiographic findings in patients with acute pulmonary embolism and no pre-existing cardiac or pulmonary disease. *Chest* 1991;100:598-603.
- St-Louis P. Status of point-of-care testing: promise, realities, and possibilities. *Clin Biochem* 2000;33:427-40.
- Strandness DE Jr, Langlois Y, Cramer M, Randlett A, Thiele BL. Long term sequelae of acute venous thrombosis. *JAMA* 1983;250:1289-1292.
- Tan M, van Rooden CJ, Westerbeek RE, Huisman MV. Diagnostic management of clinically suspected acute deep vein thrombosis. *Br J Haematol* 2009;146:347-60.
- Tapson VF. Advances in the diagnosis and treatment of acute pulmonary embolism. *F1000 Med Rep* 2012; 4:9.
- Tapson VF, Decousus H, PiniM, Chong BH, Froehlich JB, Monreal M etal. Venous thromboembolism prophylaxis in acutely ill hospitalized medical patients: findings from the International Medical Prevention Registry on Venous Thromboembolism. *Chest* 2007;132:936-45.
- Tei C, Seward JB, Tajik AJ, Minagoe S, Toyama Y. Index of myocardial performance- a simple and reproducible measurement of global right-ventricular myocardial function. *Circulation* 1995;92:2834.
- Ten Wolde M, Söhne M, Quak E, MacGillavry MR, Büller HR. Prognostic value of echocardiographically assessed right ventricular dysfunction in patients with pulmonary embolism. *Arch Intern Med* 2004;164:1685-9.

REFERENCES

- Ten Wolde M, Tulevski II, Mulder IW, Sohne M, Boomsma F, Mulder BI, Büller HR. Brain natriuretic peptide as a predictor of adverse outcome in patients with pulmonary embolism. *Circulation* 2003;107:2082-2084.
- Tomonaga Y, Gutwiller F, Lüscher TF, Riesen WF, Hug M, Diemand A, Schwenkglenks M, Szucs TD. Diagnostic accuracy of point-of-care testing for acute coronary syndromes, heart failure and thromboembolic events in primary care: a cluster-randomised controlled trial. *BMC Fam Pract* 2011;12:12.
- Tonelli AR, Plana JC, Heresi GA, Dweik RA. Prevalence and prognostic value of left ventricular diastolic dysfunction in idiopathic and heritable pulmonary arterial hypertension. *Chest*. 2012;141:1457–1465.
- Torbicki A, Perrier A, Konstantinides S, Agnelli G, Galiè N, Pruszczyk P, Bengel F, Brady AJ, Ferreira D, Janssens U, Klepetko W, Mayer E, Remy-Jardin M, Bassand JP; ESC Committee for Practice Guidelines (CPG). Guidelines on the diagnosis and management of acute pulmonary embolism: the Task Force for the Diagnosis and Management of Acute Pulmonary Embolism of the European Society of Cardiology. *European Heart Journal* 2008;29:2276-2315.
- Tulevski II, Hirsch A, Sanson BI, Romkes H, Van der Wall FF, van Veldhuisen DJ, Büller HR, Mulder BI. Increased brain natriuretic peptide as a marker for right ventricular dysfunction in acute pulmonary embolism. *Thromb Haemost* 2001;86:1193-1196.
- Tulevski II, ten Wolde M, van Veldhuisen DI, Mulder IW, van der Wall FF, Büller HR, Mulder BI. Combined utility of brain natriuretic peptide and cardiac troponin T may improve rapid triage and risk stratification in normotensive patients with pulmonary embolism. *Int J Cardiol* 2007;116:161-166.
- Van Belle A, Buller HR, Huisman MV, Huisman PM, Kaasjager K, Kamphuisen PW, Kramer MH, Kruip MJ, Kwakkel-van Erp JM, Leebeck FW, Nijkeuter M, Prins MH, Sohne M, Tick LW: Christopher Study Investigators. Effectiveness of managing suspected pulmonary embolism using an algorithm combining clinical probability, D-Dimer testing and computed tomography. *JAMA* 2006; 295:172-179.
- Van der Meer RW, Pattynama PM, van Strijen MJ, van der Berg-Huisman AA, Hartmann IJ, Putter H, de Roos, Huisman MV. Right ventricular dysfunction and pulmonary obstruction index at helical CT: Prediction of clinical outcome during 3-month follow-up in patients with acute pulmonary embolism. *Radiology* 2005;235:798-803.

REFERENCES

- Van Ramshorst B, van Bemmelen PS, Hoeneveld H, Eikelboom BC. The development of valvular incompetence after deep vein thrombosis: a follow-up study with duplex scanning. *J Vasc Surg* 1994;19:1059-66.
- Vedantham S, Grassi CJ, Ferral H, Patel WH, Thorpe PE, Antonacci VP, Janne d` Othée BM, Hofmann LV, Cardella JF, Kundu S, Lewis CA, Schwartzberg MS, Min RJ, Sacks D: for the Technology Assessment Committee of the Society of Interventional Radiology. Reporting Standards for endovascular treatment of lower extremity deep vein thrombosis. *J Vasc Interv Radiol* 2006;17:417-434.
- Verhovsek M, Douketis JD, Yi Q, Shrivastava S, Tait RC, Baglin T, Poli D, Lim W. Systematic review: D-Dimer to predict recurrent disease after stopping anticoagulant therapy for unprovoked venous thromboembolism. *Ann Intern Med* 2008;149:481-490,W94.
- Viljanen AA. Reference values for spirometric, pulmonary diffusing capacity and body plethysmographic studies. *Scand J Clin Invest*1982;42:1-50.
- Villalta S, Bagatella P, Piccioli A, Lensing A, Prins M, Prandoni P. Assessment of validity and reproducibility of a clinical scale for the postthrombotic syndrome. *Haemostasis* 1994;24:158a.
- Vonk MC, Sander MH, van den Hoogen FH, van Riel PL, Verheugt FW, van Dijk AP. Right ventricle Tei-index: A tool to increase the accuracy of non-invasive detection of pulmonary arterial hypertension in connective tissue diseases. *Eur J Echocardiography* 2007;8:317-321.
- Vuillemier N, Le Gal G, Verschuren F, Perrier A, Bounameaux H, Turck N, Sanchez IC, Mensi N, Perneger T, Hochstrasser D, Righini M. Cardiac biomarkers for risk stratification in non-massive pulmonary embolism: a multicenter prospective study. *J Thromb Haemost* 2009;7:391-398.
- Wells PS, Anderson DR, Bormanis J, Guy F, Mitchell M, Gray L, Clement C, Robinson KS, Lewandowski B. Value of assessment of pretest probability of deep vein thrombosis in clinical management. *Lancet* 1997;350:1795-8.
- Wells PS, Anderson DR, Rodger M, Stiell I, Dreyer JF, Barnes D, Forgie M, Kovacs G, Ward J, Kovacs MJ. Excluding pulmonary embolism at the bedside without diagnostic management of patients with suspected pulmonary embolism presenting to the emergency department by using a simple clinical model and D-Dimer. *Ann Intern Med* 2001;135:98-107.

REFERENCES

- Wells PS, Anderson DR, Rodger M, Ginsberg JS, Kearon C, Gent M, Turpie AG, Bormanis J, Weitz J, Chamberlain M, Bowie D, Barnes D, Hirsh J. Derivation of a simple clinical model to categorize patients probability of pulmonary embolism: increasing the models utility with the SimpliRED D-Dimer. *Thromb Haemost* 2000; 83:416-20.
- Wells P, Hirsh J, Anderson D, Lensing A, Foster G, Kearon C. Accuracy of clinical assessment of deep-vein thrombosis. *Lancet* 1995;345:1326-1330.
- Wheeler MJ. Automated immunoassay analysers. *Ann Clin Biochem* 2001;38:217-29.
- White RH. The Epidemiology of Venous Thromboembolism. *Circulation* 2003;107:I-4-I8.
- Wolfe MW, Lee RT, Feldstein ML, Parker JA, Come PC, Goldhaber SZ. Prognostic significance of right ventricular hypokinesis and perfusion lung scan defects in pulmonary embolism. *Am Heart J* 1994;127:1371-1375.
- Wu AS, Pezzullo JA, Cronan JJ, Hou DD, Mayo-Smith WW. CT pulmonary angiography: Quantification of pulmonary embolus as a predictor of patient outcome- initial experience. *Radiology* 2004;230:831-835.
- Zhou W, Schwarting S, Illanes S, Liesz A, Middelhoff M, Zorn M, Bendszus M, Heiland S, van Ryn J, Veltkomp R. Hemostatic therapy in experimental intracerebral hemorrhage associated with the direct thrombin inhibitor dabigatran. *Stroke* 2011;42:3594-3599.
- Zondag W, Mos IC, Creemers-Schild D, Hoogerbrugge AD, Dekkers OM, Dolsma J, Eijsvogel M, Faber LM, Hofstee HM, Hovens MM, Jonkers GJ, van Kralingen KW, Kruip MJ, Vlasveld T, deVreede MJ, Huisman MV; Hestia Investigators. Outpatient treatment in patients with acute pulmonary embolism: the Hestia Study. *J Thromb Haemost* 2011;9:1500-7.

ORIGINAL PUBLICATIONS

ORIGINAL PUBLICATIONS