EVALUATING RISK OF RECURRENT VENOUS THROMBOEMBOLISM DURING THE ANTICOAGULATION PERIOD IN PATIENTS WITH MALIGNANCY

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Table of Contents

Tables and Figures

ABSTRACT

Background

Current guidelines suggest that all cancer patients with venous thrombosis be treated with long-term low molecular weight heparin. Whether treatment strategies should vary according to clinical characteristics remains unknown.

Systematic review

A systematic review was performed to determine current understanding of the association between malignancy characteristics in patients with cancer-associated VTE and the risk of VTE recurrence. Four retrospective and 6 prospective studies were included. They suggest that lung cancer, metastases, and adenocarcinomas confer an increased the risk of recurrence and breast cancer a low risk.

Survey

I performed survey to evaluate thrombosis experts' opinion about the low risk of VTE recurrence they would consider acceptable for patients with cancer- associated thrombosis 103 specialists participated. 80% of respondents agreed that a risk of recurrent VTE during anticoagulation below 7% is low enough. 92% agreed that a CPR that categorizes risk of recurrence is relevant.

Retrospective Study

I performed a single retrospective cohort study to assess the feasibility of derivation of a CPR that stratifies VTE recurrence risk in patients with cancer–associated thrombosis. The study included 543 patients. A multivariate analysis selected

female, lung cancer and prior history of VTE as high risk predictors and breast cancer and stage I disease as low risk.

Conclusion

Patients with cancer-associated thrombosis do have varying risks of recurrent VTE depending on clinical characteristics.

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1.0 INTRODUCTION

1.1 Overall Aim

I sought to determine whether the current literature supports a variable susceptibility for venous thromboembolism (VTE) recurrence during the anticoagulation period in patients with malignancy-associated venous thrombosis.

I sought to determine potential predictors of recurrent VTE risk in patients with malignancy-associated thrombosis. Assuming I could identify potential predictors, it would then be important to gauge the interest amongst Thrombosis experts to derive a clinical prediction rule to identify low risk patients, to identify what threshold they would consider low risk and what trial design would be the most appropriate to conduct a prospective study for validation of the rule. Finally, I aimed to retrospectively derive a potential prediction rule from our own prospectively-enrolled patient population with VTE and cancer.

1.2 Epidemiology of Malignancy- associated Venous Thromboembolism and Statement of the Problem

Venous thromboembolism is a leading, though highly preventable, cause of morbidity and mortality (3). It is the third leading cause of cardiovascular mortality

in North America and has an annual incidence of 100-150 per 100,000 of the general population (3;4).

Thrombosis is considered to be the second most common cause of death in hospitalized patients with cancer, after death from cancer itself (3). However, the true incidence of VTE in cancer patients is uncertain due to the heterogeneity of this population and difficulties conducting large epidemiological studies in patients with diverse diseases and variable prognosis. A population-based case-control study performed among residents of Olmsted County yielded a combined annual incidence of VTE of 1 per 200 cancer patients. Individuals diagnosed with cancer experienced a 4.5 to 6-fold increase in the incidence of VTE compared with those without malignancy. This higher risk could be dependent upon additional external precipitants such as chemotherapy, prolonged immobilization or major surgery (5;6).

Although, no formal comparisons have been made with respect to VTE recurrence risk in patients with and without cancer, the literature suggests that patients with cancer –associated thrombosis appear to have a higher risk of recurrence than non-cancer patients with VTE. The risk of VTE recurrence during the anticoagulation period in the non-cancer population is approximately 1% (7;8). We recently published a systematic review of RCTs that compared vitamin K antagonists (VKA) versus low molecular weight heparin (LMWH) for treatment of cancer-

associated venous thrombosis and we found a VTE recurrence rate of 13 % in patients treated with VKA and 7% in patients treat with LMWH for 3 to 6 months with similar major bleeding rate of approximately 5% (9). In addition, the prospective cohort study conducted by Prandoni *et a.l* compared VTE recurrence risk in patients with and without cancer treated with vitamin K antagonists for a minimum of 3 months (10) . This study evaluated the rate of recurrent VTE in 181 patients with cancer and 661 patients without cancer. VTE recurrence was considered during anticoagulation and after completion of the standard thrombosis treatment strategy, using VKA for at least 3 months. The study demonstrated a 1–year cumulative incidence of recurrent VTE of 20.7% for cancer patients versus 6.8% for non-cancer patients (hazard ratio, 3.2; 95% CI 1.9-5.4). This study also demonstrated that major bleeding events were more frequent in patients with advanced metastatic malignancy and VTE than in patients with non-metastatic malignancy and VTE with 42.8% and 3.4% of major bleeding rates, respectively (10). Similar results were found in the prospective study conducted by Sallah and colleagues where patients with advanced malignancy and VTE presented a 2.4-fold higher risk of major bleeding complications than patients with limited malignancy (11). Other studies have demonstrated similar results with the additional information that patients with cancer-associated thrombosis treated with VKA are also at increased risk of bleeding complications (5;12). A pathophysiologic mechanism that could explain this association includes a higher

risk of bleeding at the site of cancer due to local increased vascularity with more friable, unorganized tissues surrounding the tumour.

Importantly, for many years management of VTE in cancer patients was similar to non-cancer patients. That is, initial therapy with LMWH or unfractionated heparin followed by VKA for at least 3 months (10;13-15). However, in 2003, the CLOT trial was published and changed practice (6). This open label, randomized controlled trial compared the efficacy (VTE recurrence) and safety (major bleeding) of 6 months of treatment with LMWH or VKA in patients with cancer-associated thrombosis. Nine percent (27 of 336) of patients in the LMWH arm developed a recurrent VTE whereas 17% (53 of 336) in the VKA arm developed a recurrent VTE during the anticoagulation period (hazard ratio, 0.48; p=0.002). Major bleeding events were similar between the 2 treatment arms (around 5%). With this study results, LMWH became the standard of care for treatment of cancer-associated thrombosis. Currently, oncology and thrombosis guidelines recommend that all cancer patients who develop a VTE receive the same anticoagulation approach, which is full-dose weight-based LMWH for one month, followed by a 75% dose for a minimum of 5 months or indefinitely should the patient remain with active malignancy or on active cancer treatment (16-18). These guidelines do conform with the AGREE collaboration for development and appraisal of guidelines (www.agreecollaboration.org). However, LMWH is expensive and

requires daily subcutaneous injections: expending scarce health care resources and possibly worsening the already fragile psychological status of the patient.

A treatment approach tailored to a patient's risk for the development of recurrent VTE while on anticoagulation would be ideal since it may enable variations on the types (e.g. VKA versus LMWH), durations or intensities of treatment to allow maximum benefit while minimizing risk and cost. Observational studies suggest that tumour site, histology and stage may influence the incidence of VTE and possibly the risk of recurrent VTE in patients with cancer (10;19-21). There has been no systematic review to summarize this risk data, neither there have been large prospective studies to evaluate the impact of the various tumour and patients' characteristics on VTE recurrence risk. In addition to the lack of data to guide therapy, to conduct a randomized controlled trial (RCT) for evaluation of VTE recurrence in patients with malignancy, randomizing patients to LMWH or VKA, without identification of a subset of patients likely to derive equal benefit from the use of VKA could be considered unethical given the recommended approach is LMWH for all patients.

To address these knowledge gaps a systematic review of the literature was performed attempting to summarize the most relevant tumour and clinical predictors of VTE recurrence risk in patients with malignancy-associated venous

thrombosis. The results of the systematic review helped with selection of the variables I used in the retrospective study (part 3 of this thesis) conducted to assess the potential to derive a clinical prediction rule (CPR) that stratifies patients' risk of recurrent VTE according to malignancy and patient's characteristics.

In addition, a survey was conducted to evaluate the opinion of thrombosis experts in North America and Europe regarding their opinion about the lowest risk of VTE recurrence that they would consider acceptable for patients with cancer- associated thrombosis and also to assess if they would value a clinical prediction rule (CPR) for stratification of VTE recurrence risk in patients with cancer-associated thrombosis. The survey also assessed their interest in participating in a future trial for derivation and validation of a definitive CPR. Therefore, if a CPR is deemed relevant according to the survey results and if I could derive, or demonstrate the potential to derive a rule from our patient data, future work could focus on development of a definitive rule in a prospective study.

2.0 BACKGROUND

2.1 Understanding Venous Thromboembolism

Venous thromboembolism is a common, potentially fatal, yet treatable, condition. It is classified as provoked, when triggered by a well defined, controllable risk factor

such as surgery, prolonged bed rest or immobilization; or it may be unprovoked, if not associated with known risk factors. A provoked VTE is usually safely treated with anticoagulation for 3 months whereas the unprovoked events need a minimum of 6 months of anticoagulation (22;23). Patients with malignancy-associated VTE do not fall in either category because even though malignancy is a well known risk factor for VTE, it is frequently an active, ongoing, poorly controlled risk factor and neither 3 months nor 6 months of anticoagulation appear to be enough to control and halt the malignancy-induced hypercoagulable state.

Most episodes of VTE occur in the lower or upper extremities or in the lungs (pulmonary embolism). However, 10% of episodes affect unusual sites, such as deep abdominal veins (i.e. portal vein, mesenteric veins), pelvic veins (i.e. ovarian vein thrombosis) or cerebral veins (24-27).

2.2 Pathophysiology of Malignancy and Venous Thromboembolism

Blood coagulation is a physiological process triggered whenever a vessel wall is injured (e.g. surgery, a skin cut) for appropriate vessel wall repair and re-establishment of haemostasis and homeostasis (blood clots prevent "never ending" bleeding). There are however, a few pathological states that excessively trigger the coagulation system and predispose to blood clot formation. A German physician

named Rudolf Virchow (1821-1902) described the pathophysiology of pulmonary embolism that included a triad of elements that predispose to thrombosis: diminished or interrupted blood flow (stasis); vessel wall injury (endothelial injury); and hypercoagulability. A few years later, the triad was named after him and it simplistically explains why patients with cancer have a higher risk for thrombosis. Patients with cancer are frequently immobile (stasis); have vessel wall injury through surgery and use of central venous catheters or are exposed to venous wall injury by chemotherapy and/or radiotherapy; and are hypercoagulable due to specific procoagulant proteins produced by the cancer cells that initiate and accelerate the coagulation cascade. Cancer cells interact with the coagulation system with the intent to perpetuate their growth by stimulating angiogenesis and metastasis (13).

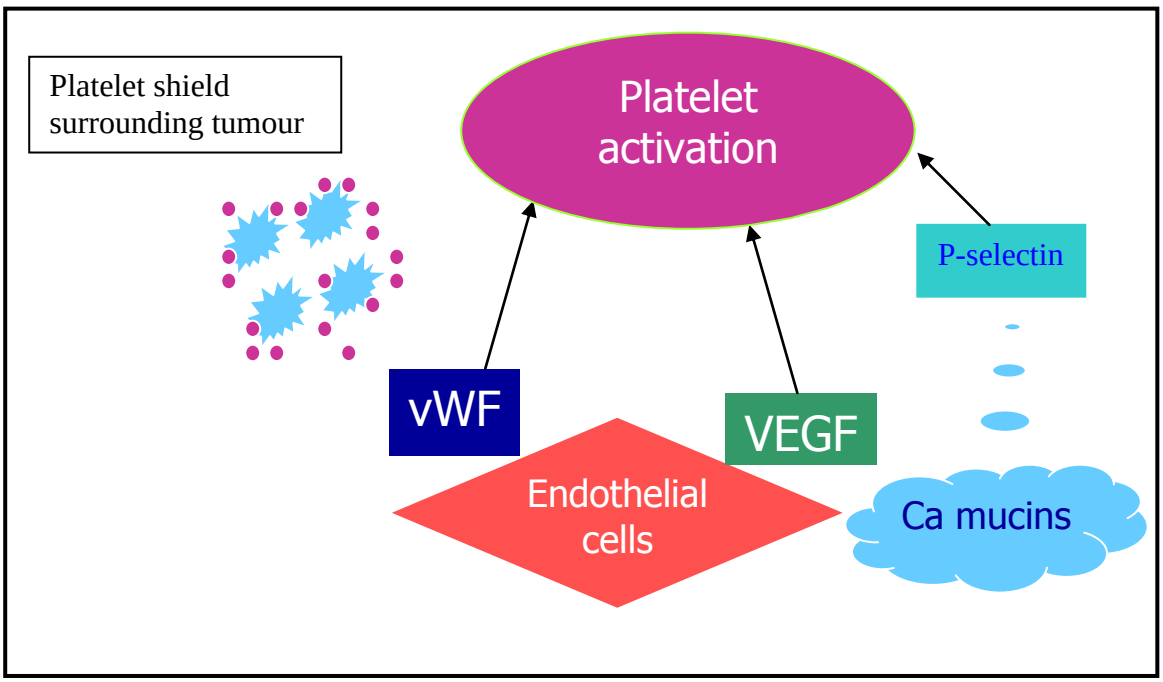
Altogether, the elements of the Virchow's triad trigger and upregulate the coagulation system. The coagulation system is divided in primary and secondary haemostatic systems that act primarily in the venous system, in a very balanced fashion to enable appropriate haemostasis in case of vessel wall injury. In the primary system, platelets and von Willebrand factor (vWF) play the major role. The secondary hemostatic system contains several procoagulant, anticoagulant and fibrinolytic proteins that work together for the production of the final fibrin clot and then, its physiological dissolution. In the context of malignancy however, this

balance is impaired through abnormal up regulation in the production of a set of procoagulant proteins and inhibition of the anticoagulant pathways leading to a constant hypercoagulable state.

Animal models suggest that the process begins with the activation of MET-protooncogenes that up regulate the translation and aberrant expression of procoagulant proteins by the tumour cell membrane, such as tissue factor, vWF and cancer procoagulant (28). Along with this process, patients with cancer are frequently subjected to chemotherapy and radiation therapy, as well as insertion of central venous catheters and surgical procedures; all of which predispose to vessel wall injury and consequently a procoagulant state (5;12).

In addition, several adenocarcinomas are known to produce mucins that will stimulate the activation of a protein, P-Selectin, by platelets. P-selectin induces the activation of platelets which in turn will produce and express vWF and Vascular Endothelial Growth Factor (VEGF) (29;30). The platelets will adhere at the same time to specific receptors in the tumour cell membrane and in the endothelium, making a shield that protects the malignant cell against the host's immune system enabling therefore, tumour cell migration and invasion through small vessels (29) (Figure 1).

Figure 1. Production of platelet shield for tumour cell protection against the host's immune system



P-selectin and VEGFstimulate platelet activation and aggregation

VEGF will not only promote neo-angiogenesis aiding with tumour viability and growth, but also will act in a positive feedback with platelets by promoting a continuous stimulation of their activation and by stimulating abnormal expression of Tissue Factor by endothelial cells, fibroblasts, monocytes and the tumour cell *per se* (31). Tissue Factor is the key initiator of the coagulation cascade and plays a pivotal role in cancer-induced hypercoagulability (Figure 2). It complexes with activated Factor VII and leads to the activation of Factor X that in turn activates prothrombin into thrombin which induces clot formation. Tissue factor expression in tumour cells has been shown in many cancers, including breast, lung, colorectal and pancreatic cancer. Elevated levels have been correlated with increased

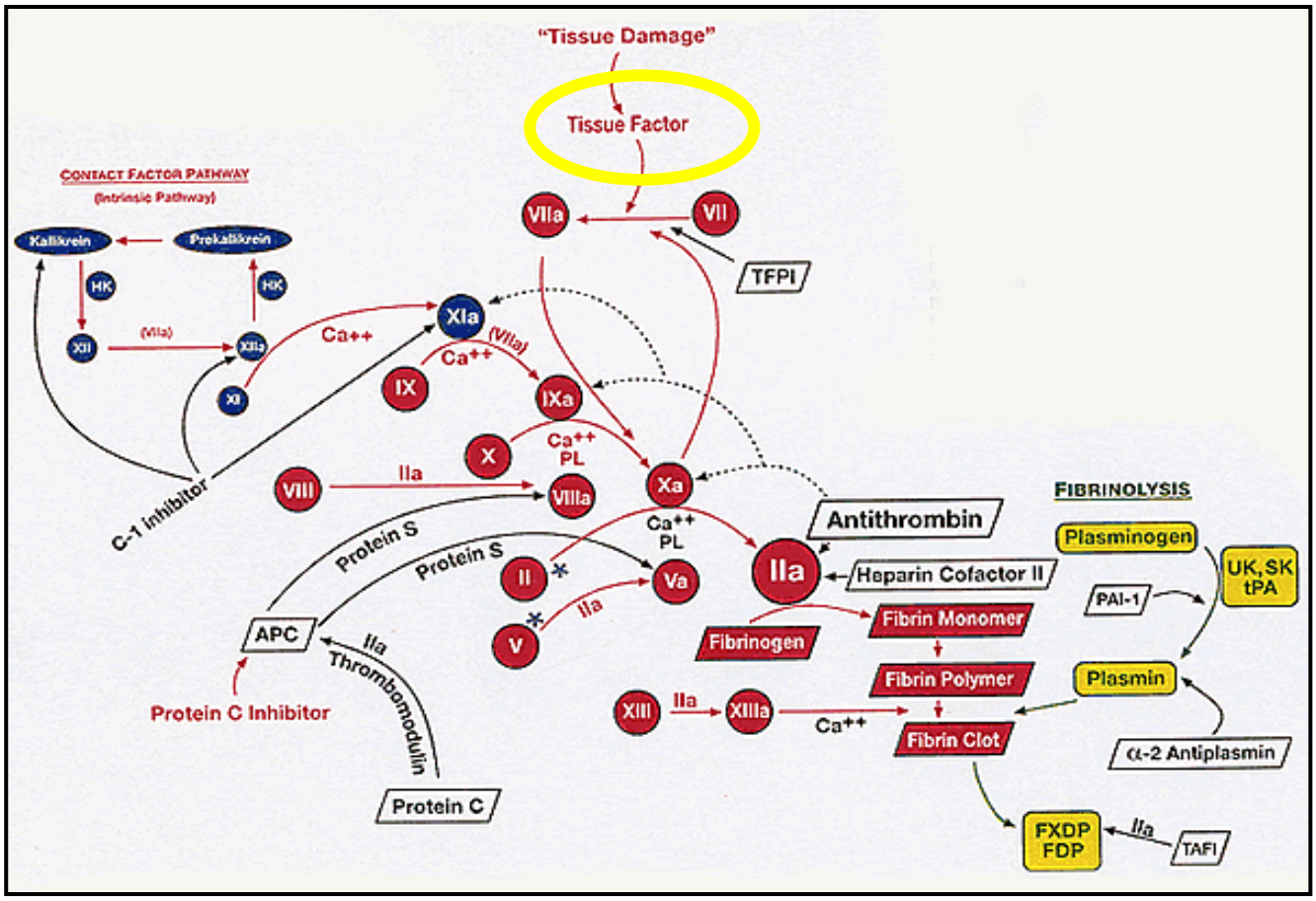
angiogenesis, increased vascular density, unfavourable prognosis and metastatic disease (32;33). Another prothrombotic protein produced by tumour cells is a cysteine proteinase cancer procoagulant. It acts independent from the Tissue Factor pathway and directly activates Factor X, facilitating even more thrombus (clot) formation. The thrombus will also be used as a shield to protect malignant cells from recognition by the host's immune system which facilitates cancer growth and spread.

The major component of a blood clot is fibrin. In normal haemostasis there is a terminating system to prevent ongoing clotting and to confine the fibrin clot to the site of vascular lesion. The fibrin clot is "dissolved" through the activation of the fibrinolytic system. Its main component is Plasminogen that is transformed into plasmin through plasmin activator proteins (urokinase and tissue plasminogen activator) that will lyse the fibrin clot. In the context of active malignancy this pathway is disrupted. Tumour cells are capable of expressing PAI- 1, plasminogen activator inhibitor, responsible for inhibiting clot lysis and consequently enabling the perpetuation of the hypercoagulable state. Furthermore, in the cancer setting there is disruption of the function of the natural anticoagulant proteins, such as antithrombin, protein C and protein S that act directly in the coagulation cascade by inhibiting the activation of clotting factors. In normal haemostasis, Protein C and its co-factor Protein S are responsible for cleaving the procoagulants Factor VIII and

Factor V thereby, inhibiting the perpetuation of the activation of the coagulation cascade. A few studies suggest that patients with cancer develop an acquired Protein C resistance (33;34). Protein C resistance occurs when a conformational abnormality in the protein C binding site to Factor VIII and Factor V occurs and leads to an inability of protein C to halt these factors' activity. However, the pathways through which this occurs are not established. Furthermore, there is speculation that a few commonly used chemotherapeutic agents (cyclophosphamide, metotrexate, 5- fluoruracil) may decrease levels of protein C and protein S.

The net result of the imbalance between the coagulation cascade and the fibrinolytic system induced by malignant cells is therefore, an increased risk of pathological blood clot formation. The extent to which the tumour (site, type and extent), the cancer treatment, or other patient characteristics influence this risk is not well defined.

Figure 2. Malignancy induced – thrombus formation



2.3 Efficacy and Safety of Different Anticoagulation Strategies

VKA has been the cornerstone of anticoagulation for many years. It works by inhibiting the production of vitamin K dependent coagulation factors (Factors II, VII, IX and X) by the liver. It also inhibits the production of the vitamin K dependent natural anticoagulants, protein C and S. VKAs are drugs not always easy to monitor. They interact with various food and drugs, require 2 to 3 times weekly blood tests for INR monitoring (the standard laboratory test for assessment of anticoagulant effect of VKA) in the first 2 weeks of treatment and at least

monthly thereafter. In addition, VKA cannot be administered as a single anticoagulant in the first 5 to 7 days of treatment due to the increased risk of rebound thrombosis because it not only inhibits the procoagulant factors but also the anticoagulants, protein C and S. Therefore, 5 to 7 days of concomitant use of LMWH or unfractionated heparin with VKA is necessary during the first week of anticoagulation (35). In the context of cancer-associated thrombosis the use of VKA can be particularly difficult because chemotherapy and other frequently used drugs, such as antibiotics may affect VKA pharmacokinetics and INR control leading to increased bleeding risk. Chemotherapy and radiation therapy-induced thrombocytopenia, erratic oral intake, and higher risk of liver dysfunction may also predispose these patients to bleeding complications. Management of VTE in patients with cancer using VKA can therefore, be a challenging task. Clearly, treatment for acute VTE which is initial therapy with LMWH or unfractionated heparin followed by long term-therapy with VKA is not ideal for all cancer patients.

Given the limitations of VKA, it had been suggested that cancer patients with VTE should receive special attention while on anticoagulation for VTE and the best therapeutic approach for this patient population has been a matter of debate. Several studies suggest that long-term fixed-dose LMWH may be at least as safe and efficacious as VKA for cancer and non-cancer patients (36-39). One RCT investigated the efficacy of long-term LMWH in patients with cancer- associated

thrombosis and it did not find any benefit of LMWH over VKA (1). However, as explained in section 1.3, the largest study to date (the CLOT trial) demonstrated superiority of LMWH over VKA (absolute risk reduction in VTE recurrence risk of 52% for LMWH in comparison to VKA) and as such, LMWH is considered the standard of care for treatment of VTE in patients with malignancy (6).

I recently performed a systematic review and meta-analysis of the published RCTs that investigated the efficacy and safety of long-term treatment with LMWH or VKA in adult patients with cancer-associated thrombosis (9). We included 5 RCTs that in total encompassed 1158 patients. Patients were anticoagulated with either VKA or LMWH for 3 months in 3 studies (38;40;41) and for 6 months in 2 studies (1;6) . We found that the use of LMWH resulted in a 53 % (95% CI, 0.36-0.76; *p*= 0.0007) reduction in the relative risk of VTE recurrence during the anticoagulation period when compared to patients using VKA. Overall, 73 of 565 patients (12.9%) recurred in the VKA group and 40 of 593 (6.7%) recurred in the LMWH group. A subgroup analysis evaluating time of anticoagulation (3 or 6 months) was performed and demonstrated no significant difference between the use of VKA or LMWH in the 3-month group (RR=0.62, 95% CI:0.27-1.46; p=0.26). The 6-month group showed a significant difference between the 2 anticoagulant groups (RR=0.52; 95% CI: 0.34-0.79; *p*=0.002) (9). There was no difference in major bleeding risk (around 7% in each group).The results of the review were largely

driven by the results of the CLOT trial (6) , since it was the only trial adequately powered to evaluate differences between the use of VKA and LMWH in patients with cancer-associated thrombosis. Even so, the ideal length of time for anticoagulation and whether different therapeutic strategies should be used for the various types and different stages of malignancy is a question that remains unanswered.

Furthermore, LMWH treatment requires daily subcutaneous injections which can be a hassle for many patients and it is an expensive drug (around CND$ 30.00 per injection versus CND$ 0.30 for each 5 mg VKA tablet). In Canada, the cost LMWH treatment for cancer-associated thrombosis is covered by the government which enables full access of patients with cancer-associated thrombosis to the drug, but this certainly increases the burden of its already fragile health care system.

2.4 Association between Malignancy Characteristics and the Risk of VTE

Thrombosis is one of the major complications of cancer. However, large epidemiological studies that evaluate risk of recurrent VTE according to malignancy and patient's characteristics are lacking. The available literature is derived from large database studies or retrospective studies that evaluated risk of an index (first) VTE in patients with active cancer and the risk factors for index VTE

may be different than risk for treatment failure and recurrent VTE. If so, the risk factors that predispose to recurrence deserve attention, so that tailoring of VTE treatment can be improved.

2.4.1 Tumour Stage

A few observational studies that evaluated the incidence of a first VTE in patients with active malignancy found a 2 to 19-fold higher incidence among patients with distant metastasis than in patients with localized disease (19;20;42;43). Blom *et al.* conducted a case-control, record linkage study to evaluate the risk of a first VTE in more than 5,000 patients with diverse types of malignancy with and without metastasis. Although they found an increased risk of VTE in both groups, patients with metastatic disease had a significantly higher risk [metastasis: odds ratio:67.7 (95%CI, 9.4-486.6); no metastasis: odds ratio 3.7 (95%CI, 2.4-5.7)] (44). Later on, Blom *et al.* prospectively evaluated 2149 patients with lung cancer and found an increased relative risk for a first VTE associated with malignancy of 1.9 (95%CI, 1.9-2.3) for patients with metastasis in comparison to patients without metastasis (45).

2.4.2 Primary Tumour Site

Two large administrative database studies suggest that the most common malignancies associated with development of a first VTE are lung cancer, colorectal cancer, breast cancer and lymphomas (46;47). One evaluated over 7 thousand patients with cancer over a total cohort of 10 million hospitalized patients of the USA Medicare database (46). The rate of a first VTE was not clearly reported but the study stated that renal cancer, GI, brain and ovary cancer as well as lymphomas were the most frequent types of malignancies that predisoposed to a first VTE compared to head/neck, bladder and breast with RR of DVT/PE of 4.13 (95%CI; 3.82 – 4.45). Another large administrative database study evaluated 34,000 records of patients with various malignancies and it suggested that the concomitant diagnosis of VTE and cancer is much more prevalent in patients with lung (17%) and gastrointestinal cancer (17%) than in patients with breast (3.6%) or prostate cancer (7%) (47). One retrospective cohort study reviewed the charts of 529 patients with cancer-associated DVT. They found similar results with lymphomas presenting with the highest rates of an index (first) VTE at 15% followed by breast cancer (13%), lung and gastrointestinal malignancies (11% each) (48). An accurate rate of VTE recurrence according to primary tumour site is not established.

2.4.3 Tumour Histology

A handful of studies evaluated the impact of tumour histology on the risk of VTE with conflicting results. The retrospective study by Blom and colleagues evaluated 537 patients with lung adenocarcinoma or non-adenocarcinoma tumours. The Hazard Ratio for development of a first VTE was 2.8 (95% CI, 1.2-6.4) fold higher in patients with lung adenocarcinomas than in patients with non-adenocarcinomas (20). On the other hand, Tagalakis and colleagues retrospectively compared the rate of a first VTE in 254 patients with adenocarcinoma and 105 non-adenocarinoma of the lung and found no statistically significant difference in the incidence of VTE between the two histological types (non-adenocarcinoma: 17 VTE in 105 (16.2%)patients; and adenocarcinoma: 40 VTE in 254 (15.7%) patients; p=0.99)(43).

The study conducted by Descourt and colleagues evaluated the rate of recurrent VTE in patients with malignancy-associated venous thrombosis. They prospectively compared the outcomes of 147 patients with adenocarcinoma or other histological types of malignancy and VTE, and reported that patients with adenocarcinoma presented a 4-fold higher risk of VTE recurrence on oral anticoagulation at therapeutic levels when compared to non-adenocarcinoma patients, suggesting that histology may play an important role in the development of VTE recurrence (49).

3.0 ORGANIZATION OF THIS PROJECT

3.1 Systematic Review

We conducted a systematic review to identify potentially important risk factors for VTE recurrence in patients with malignancy-associated venous thrombosis to aid in the selection of the potentially most relevant predictors for my retrospective clinical prediction rule derivation.

3.2 Survey Research

We conducted a survey to evaluate the opinion of thrombosis experts in North America and Europe with regards to the minimum acceptable threshold for VTE recurrence risk in patients with cancer-associated thrombosis that they would be willing to accept to consider using VKA instead of LMWH in the treatment of a first venous thromboembolic event in patients with cancer. The survey also assessed the specialists' expectations regarding the relevance of the derivation of a CPR for stratification of risk of recurrent VTE in patients with malignancy-associated thrombosis and their interest in participating in a future prospective study for derivation/validation of such a rule.

3.3 Retrospective Study

We conducted a retrospective cohort study to investigate malignancy and clinical characteristics of patients who developed a recurrent VTE during the anticoagulation period, diagnosed and/ or followed at the Thrombosis Unit of the Ottawa Hospital for collection of potentially relevant data for evaluation of the feasibility to derive a clinical prediction rule that will stratify risk of VTE recurrence in patients with active cancer and venous thrombosis.

4.0 SYSTEMATIC REVIEW

4.1 Rationale for the Systematic Review

As detailed above, there is lack of guideline advice regarding the best management of malignancy –associated venous thrombosis taking into consideration malignancy and patient's characteristics, suggesting a lack of evidence. In addition, our clinical experience and some prospective studies suggest that effectiveness of treatment of VTE in patients with active malignancy is still a concern, since even patients with cancer-associated thrombosis treated with LMWH still have a much higher recurrence rate than non-cancer patients with VTE, which is about 2% during the anticoagulation period (6;38;40;41). Tumour characteristics such as stage and histology, and certain patient characteristics may importantly influence the risk of recurrent VTE. No publication has systematically evaluated the literature and summarized data on factors that influence the risk of recurrence. Therefore, the

results of this review will allow a better understanding of the most important variables to be addressed in the retrospective study.

4.2 Systematic Review Methods

4.2.1 PICOS Question and Objectives

4.2.1.1 PICOS Question

Population: adult patients with malignancy- associated venous thromboembolism on long-term anticoagulation with LMWH or VKA (minimum of 3 months)

Intervention: none

Comparator: none [note: there is no formal comparator since there is no intervention. However we did compare different levels of malignancy characteristics such as malignancy stage (localized versus metastatic disease), histology (adenocarcinoma versus non-adenocarcinoma), and the various primary tumour sites]

Outcome: VTE recurrence during the anticoagulation period

Study Design: observational studies, registries and clinical trials (randomized-controlled or quasi-experimental)

PICOS Question: *Do adult patients with active cancer and VTE have different risk of VTE recurrence during the anticoagulation period according to tumour or*

patient's characteristics such as tumour type and histology or easily obtained clinical parameters (e.g. gender, age)?

4.2.1.2 Objectives

4.2.1.2.1 Primary objective

To evaluate observational and experimental studies that investigated the impact of cancer characteristics such as histology, tumour stage and primary tumour site in the risk of VTE recurrence in patients with cancer-associated VTE.

4.2.1.2.2 Secondary Objective

1) To determine whether malignancy characteristics influence the efficacy (recurrence risk) and safety (bleeding risk) of different treatment strategies

2) To determine overall mortality

4.2.2 Data Sources and Searches

Following our systematic review protocol, a systematic literature search strategy was conducted to identify potential studies on MEDLINE (1950 to September week 3 2009), EMBASE (1980 to 2009 week 29), the Cochrane Register of Controlled Trials (1st quarter 2008) and MEDLINE in-Process and other non-indexed citations

(Feb 12, 2008) using the OVID interface. Adjustments were made to the search strategy to account for the differences in indexing between databases. A search of the "grey literature" was performed using SIGLE up to 2005 (After 2005 SIGLE was no longer updated). Hand-search of journals and of the American Society of Clinical Oncology (ASCO) and the American Society of Hematology (ASH) Conference proceedings (2001-2009) was also performed. The systematic search strategy is documented in Table 1. The reference lists of identified studies were reviewed to identify additional articles. There was no restriction to language, publication year or type of publication for identification of studies.

4.2.3 Study Selection

We used a structured question format (PICOS) to aid our literature search strategy. We identified studies that satisfied all of the following criteria set *a priori*: 1) adult patients (18 years of age or older) with active malignancy and objectively diagnosed acute VTE (DVT, PE or both) on active anticoagulation therapy. To be considered as VTE associated with active malignancy, we required the diagnosis of cancer (other than basal-cell or squamous-cell carcinoma of the skin) to be within 6 months before or after VTE. Moreover, the patient had to be on active treatment, or have received anti-cancer treatment within the previous 6 months, or had progressive malignancy on palliative care. Objectively proven DVT was defined as

lower extremity DVT confirmed by compression ultrasound or contrast venography with evidence of thrombus in the trifurcation of calf veins as they join the popliteal vein or more proximal veins; and PE was defined by high probability on ventilation-perfusion lung scan or by segmental or more proximal vascular filling defects, or multiple subsegmental filling defects on computerized tomography pulmonary angiography; 2) studies must have reported at least one of the following: tumour type, tumour site or stage at study enrolment and in the patients with recurrent VTE.

Studies were excluded if: 1) treatment did not involve anticoagulation; 2) they were published in language other than English, French, Portuguese or Spanish; 3) they evaluated only non-lower limb DVT sites; 4) they were case reports.

4.2.4 Outcome Measures

The primary outcome measure was VTE recurrence risk during the anticoagulation period according to potential predictor (e.g. tumour histology, primary tumour site, gender and age) in patients with malignancy-associated venous thrombosis.

Secondary outcome measures included VTE recurrence rate according to anticoagulant strategy; major and minor bleeding events; overall mortality rate. Solid tumours classified as stage I or II were described as localized malignancy and

Stage III or IV as metastatic malignancy (50). We attempted to define bleeding events according to the International Society of Thrombosis and Haemostasis criteria (51). Major bleeding was considered when the patient had a fatal bleeding; or had a fall in haemoglobin level of 2 g/l or more; or if patients had to receive 2 or more packs of red blood cell transfusion ; or if bleeding occurred in a critical organ (e.g. retroperitoneal, intracranial, intraspinal, intraocular).

4.2.5 Data Extraction and Quality Assessment

Prior to broad screening, we performed a calibration exercise with two reviewers (ML and HM) using ten records. Subsequently, each reviewer independently applied the inclusion criteria to articles identified from the initial search strategy. Evaluation of potentially eligible studies to confirm eligibility and methodological quality were performed independently by three reviewers (ML, HM and VD) and summary information of eligible articles were extracted. Data abstraction was performed using a spread sheet (Microsoft Excel®). Disagreements were resolved by discussion and by consulting a fourth reviewer (PW). The methodological quality of the selected observational studies was assessed according to the Newcastle-Ottawa scale for cohort studies and case-control studies (72). This is the scale recommended by the Cochrane Non-Randomized Studies Methods Working Group. The methodological quality of selected RCTs would have been done according to the Cochrane Collaboration – Cochrane Handbook for Systematic

reviews of Interventions (http://www.cochrane.org/resources/handbook/). However, no RCTs fulfilled our inclusion criteria. Risk of bias was assessed for individual studies at study level. This systematic review was reported according to the QUOROM statement criteria (52).

Table 1. Systematic Literature Search Strategy

1 . exp Neoplasms/
2 . (cancer or neoplasm$ or tumour$ or tumour$ or malignan$).tw.
3. (carcinoma$ or adenocarcinoma$ or sarcoma$).tw.
4 . epithelioma$.tw.
5. or/1-4
6. exp Venous Thrombosis/
7. (dvt or vte).tw.
8. Venous Thromboembolism/
9. thrombosis/ or thromboembolism/
10. (thrombos$ or thromboemboli$ or thrombotic).tw.
11. or/6-10
12. exp risk/ or risk.tw.
13. Incidence/ or incidence.tw.
14. Recurrence/
15. (recurrence or recurrent).tw.
16. or/12-15
17. 5 and 11 and 16
18. cohort studies/ or longitudinal studies/ or follow-up studies/ or prospective studies/
19. (cohort or longitudinal or follow-up or prospective).tw.

20. Retrospective Studies/ or retrospective.tw.
21. comparative study.pt.
22. or/18-21
23. 17 and 22
24. from 23 keep 1-999
25. 23 not 24
26. from 25 keep 1-999

4.2.6 Data Synthesis and Analysis

Comparisons between patients with and without VTE recurrence would be attempted for different anticoagulation strategies and for the various potential predictors using formal meta-analysis methods with Relative Risk (RR) as the primary measurement with 95% confidence intervals (CIs). Pooled measurements were calculated using a random-effects model. RR> 1.0 suggests increased risk of the outcome measured and RR< 1.0 suggests reduced risk of the outcome measured. Individual trial estimates and pooled estimates were performed with the Review Manager® software (Cochrane Collaboration's Information Management System). Between and within study heterogeneity was assessed using the Cochrane Q/chi-square tests. Comparison of binomial variables was analyzed with Fisher's exact test or chi square test, as appropriate, with a 2- sided alpha of 0.05.

4.3 Results of the Systematic Review

4.3.1 Identification of Relevant Studies

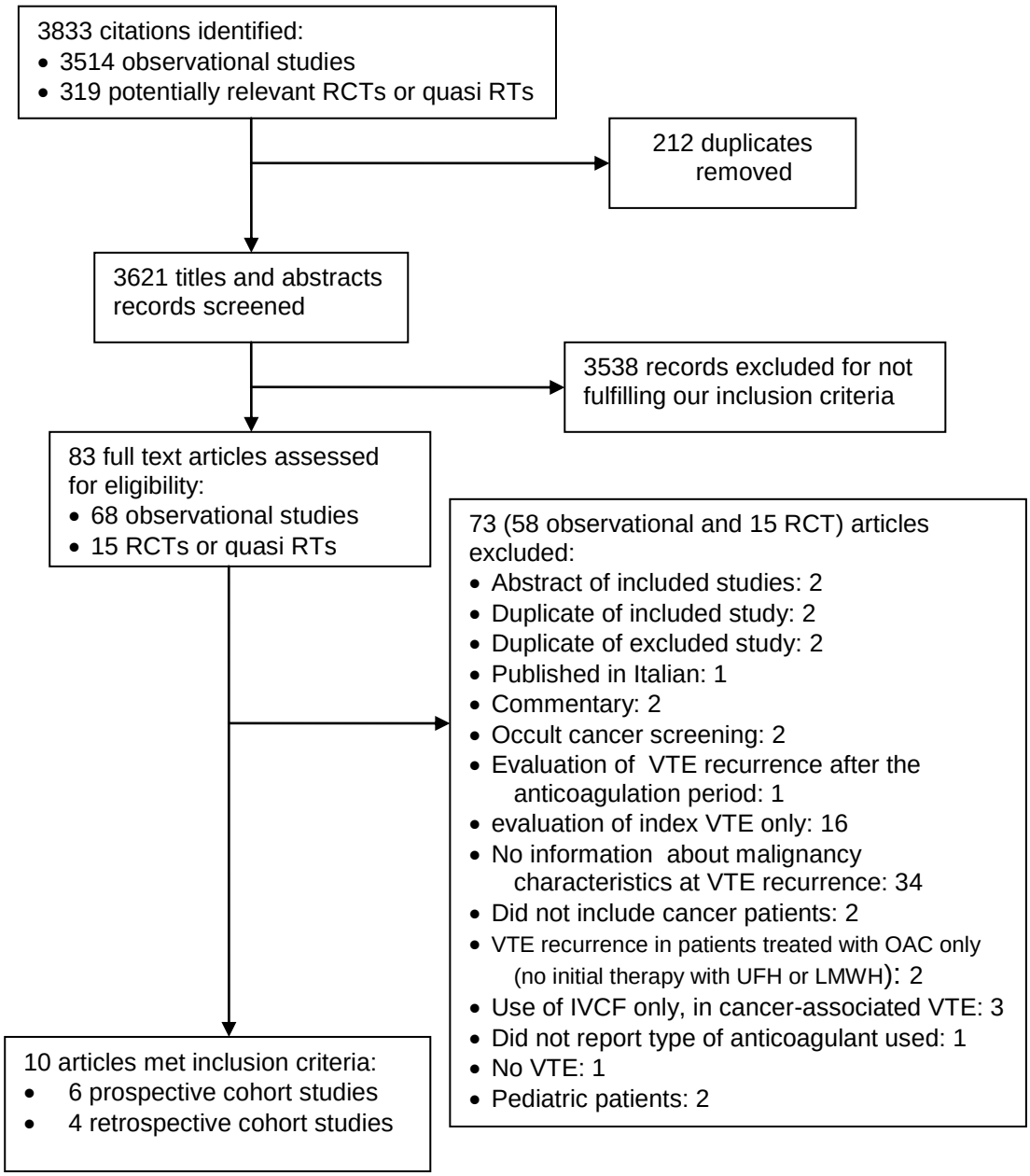
In total, 3833 citations were identified by our systematic search of the literature, including 212 duplicates. Details of literature search strategy are presented in Figure 3.The corresponding authors of 10 potentially eligible RCTs and 6 observational studies were contacted, but they did not reply or were unable to provide the necessary data for information about tumour characteristics. Four retrospective studies (53-56) and 6 prospective studies (10;11;21;49;57;58) were selected with no discrepancies between the reviewers with regards to study eligibility.

4.3.2 Quality Assessment

Quality assessment was done using the Newcastle-Ottawa quality assessment scale (Table 2) (74). Three retrospective (56) and 2 prospective studies (21;57) evaluated a selected group of patients whereas the others evaluated diverse types of malignancy (10;58). Overall, studies were somewhat representative of the average in the community. Only 1 study had an exposed (cancer and VTE) and a non-exposed cohort (no cancer and VTE) and therefore, comparability of cohorts could not be assessed (9). All but one study provided secure record of outcome with objectively confirmed VTE recurrence (54). Follow-up information during the anticoagulation period or until death was provided by all studies. However, 2

studies did not provide data on patients lost to follow up (53;54). Tumour stage, primary tumour site and histology were not uniformly reported at study entrance or at VTE recurrence in any of the included studies. One study that included 147 patients reported that 18 had the diagnosis of malignancy confirmed after the first acute VTE (49). It did not report specifically if this was within the first 6 months after VTE diagnosis. We elected to include these patients since they comprised only 12% of this study population.

Figure 3. Flow diagram summarizing the identification Process of Relevant Articles



4.3.3 Baseline characteristics of included studies

4.3.3.1 Retrospective Studies

Four studies with a total of 227 patients (range: 31-74) were evaluated (53-56). The mean age of patients and gender distribution were similar between studies. Primary tumour site was described by all studies and one also reported histology (53). One hundred patients had localized disease and 127 had metastasis. Initial VTE site was uniformly described: 117 isolated lower limb DVT, 41 PE and 69 patients with both DVT and PE. Details of malignancy characteristics at presentation and anticoagulation strategy are depicted in Table 2. In 3 studies, initial anticoagulation was unfractionated heparin (UFH) or LMWH followed by VKA (target INR = 2.0 to 3.0; or prothrombin time (PT) between 1.5 and 2 times the baseline PT) (54-56). The fourth study used UFH for long-term anticoagulation but the dose was not specified (53). In the study by Schiff *et al*. 9 patients did not receive anticoagulation therefore, they were excluded from the review (55).

4.3.3.2 Prospective Studies

Six prospective cohort studies were identified. Five included only patients with cancer (11) and 1 study evaluated patients with and without cancer (10). There were 4573 (range 14- 3805) adult patients with cancer-associated VTE. Mean age of participants and gender distribution were similar between studies.

Table 2. Methodological Quality of included studies - Newcastle-Ottawa Scale

Author, year	**Selection**				**Comparability Of cohorts**	**Exposure**		
	Represent tiveness of Exposed cohort	Representa tiveness of nonexposed cohort	Ascertainment of Exposure	Outcome Not present at beginning Study		Assessment of outcome	Was follow-enough?	Adequacy of follow-up
Schiff, 1994	*	-	*	*	-	*	*	*
Constantini, 1991	*	-	*	-	-	-	*	-
Clarke-Pearson, 1983	*	-	*	*	-	-	*	*
Debourdeu, 1996	*	-	-	*	-	*	*	-
De Stefano, 2005	*	-	*	*	**	*	*	*
Descourt, 2006	*	-	*	*	-	*	*	*
Sallah, 2004	*	-	*	*	-	*	*	*
Prandoni, 2002	*	*	*	*	*	*	*	*
Monreal, 2004	*	-	*	*	-	-	*	*
Trujillo-Santos, 2008	*	-	*	*	-	*	*	*

†A study can be awarded a maximum of 1 star for each numbered item in the selection and outcome categories. A maximum of 2 stars can be given for comparability

Malignancy characteristics at study enrolment were reported by all studies. Primary tumour site was described in 4 studies (10;11;21;58) and histology in 1 study (49;57). Of the 5 studies that included solid tumours(10;11;21;49;58), 4 reported on disease stage: 2294 with localized and 2118 with metastatic malignancy(10;58). Initial VTE site was reported in 4 studies: 2282 DVT, 1887 isolated PE and 34 cases of both PE and DVT, but it was not clear whether all cases of PE had investigation for DVT. Baseline characteristics are presented in Table 3.

UFH or LMWH were the anticoagulants of choice for initial anticoagulation, followed by VKA (target INR= 2.0 to 3.0) in 3 studies (10;21;49;58). LMWH was continued in 2 studies (21;57). In the study by Trujillo-Santos, 1643 patients received VKA and 1855 patients received LMWH for long-term anticoagulation but the data on recurrence by treatment strategy was not accurately reported (58).

Table 3. Baseline malignancy characteristics and anticoagulation strategy of included studies

Author, year (country)	Study Design	Population (n)	Age Years (range)	Gender M/F	Primary Tumour Site	Reported Histology	Stage	Long Term anticoagulation	Duration of treatment (months)
Schiff, 1994 (USA)	Retrospective	51	58 (25-81)	30/21	various	NO	Metastasis only	VKA	3
Constantini, 1991 (Israel)	Retrospective	31	52 (38-66)	NR	CNS*	NO	Various	UFH	3
Clarke-Pearson, 1983 (USA)	Retrospective	74	62 (23-91)	0/74	Gyneco logical	NO	Various	VKA	3
Debourdeu, 1996 (France)	Retrospective	71	60 (22-91)	37/34	various	NO	Various	VKA	3
De Stefano, 2005 (USA)	Prospective	14	49 (18-70)	7/7	Hemato logical	YES	Not applicable	LMWH	6
Descourt, 2006 (France)	Prospective	147	71.5	88/74	various	YES	Various	VKA	6
Sallah, 2004 (USA)	Prospective	223	57 (29-81)	111/112	various	NO	Various	VKA	6
Prandoni, 2002(Italy)	Prospective	181	65 (54-76)	84/97	various	NO	Various	VKA	3
Monreal, 2004 (Spain)	Prospective	203	65 (36-96)	132/71	various	NO	Metastasis only	LMWH	3
Trujillo-Santos, 2008 (Spain)	Prospective	3805	NR	2086 /1729	various	NO	Various	LMWH or VKA	3
Total		4800	60 (18-96)	2475 /2219					

NR=not reported; * CNS= central nervous system

4.3.4 Primary Outcome Measure

4.3.4.1 Retrospective Studies

VTE recurrence and malignancy characteristics

Twenty-eight of 218 (13%) patients recurred during the 3-month anticoagulation period. There were 16 DVT, 11 PE and 1 patient had both DVT and PE. Three studies with a total of 113 patients reported on VTE recurrence according to tumour stage: 13 of 79 (16%) patients with metastatic disease and 8 of 34 (24%) with localized malignancy recurred (p= 0.4319) (53-55). A formal meta-analysis of these studies was not possible because one of the studies evaluated only metastatic disease (55). Evaluation of primary tumour site was possible in the 2 studies that enrolled patients with brain malignancies (53;55). Seven of 73 (10%) patients presented with a recurrent VTE. Information regarding tumour histology was not provided by any study.

4.3.4.2 Prospective Studies

VTE recurrence and malignancy characteristics

Three hundred and three of 4573 (7%) patients had a recurrent VTE. There were 135 (45%) patients with recurrent DVT and 109 (36%) with PE. Fifty-nine (19%) recurrent events were not specified (unknown whether DVT or PE or both). Four studies comprising 4559 patients reported recurrence according to malignancy stage. Recurrent VTE developed in 157 of 2011 (8%) patients with metastatic malignancy

and 102 of 2548 (4%) with localized malignancy ($p < 0.001$) (10;21;49;58). A formal meta-analysis concerning the association between VTE recurrence risk and malignancy stage could only be calculated pooling the data of 2 studies (10;21;49;58) because one study did not provide TNM staging data at enrolment (49) and the other evaluated patients with metastatic disease only (21). For the 2 studies meta-analyzed, VTE recurrence rate for patients with metastasis was 7 % (120/1605) and with localized malignancy was 4.5 % (101/2220). The relative risk for VTE recurrence was RR=1.41 (95% CI, 1.09-1.83; p=0.009) suggesting a statistically significant higher risk for patients with advanced malignancy. The Higgins test for heterogeneity was not significant ($I^2 = 45.4_{\%;}$ p= 0.18). The association between VTE recurrence risk and primary tumour site was described in 3 studies (10;58). However, the studies did not provide the absolute number of patients with and without recurrence according to primary tumour site. Therefore, we report only ratios (hazard ratios or odds ratios, depending on the study) and frequencies provided by the included studies. For Prandoni *et al.* the highest hazard ratios for VTE recurrence were 6.9 (95%CI, 3.0-15.9) for lung and 5.1 (95%CI, 2.3-11.3) for gastrointestinal (GI) malignancies; whereas the lowest hazard ratio of recurrent VTE was reported in patients with breast cancer (0.7; 95%CI, 0.1-4.9). Trujillo-Santos *et al.* reported the highest VTE recurrence rate for genitourinary (GU) and lung malignancies with 23.5% (45/190) and 22.5% (43/190), respectively (58). The lowest VTE recurrence rate occurred in patients with breast (13/190) or haematological malignancies (9/190), approximately

4% each. Monreal *et al.* reported that GI malignancies had a 2.4 higher risk while breast (OR=0.0; 95% CI= 0.0-3.4) and lung (OR=0.7; 95% CI= 0.2-2.4) had the lowest risk of recurrence (21). Another characteristic that seemed to be associated with VTE recurrence risk in these studies was age. In the studies by Trujillo-Santos and Monreal, patients 65 years of age or younger appeared to have up to 3-fold higher risk for recurrence [Trujillo-Santos (< 65 years): OR=3.0 (95% CI; 1.9-4.9); Monreal (> 65years): 0.7 (95% CI; 0.3 – 2.1)] (10;58).

The evaluation of recurrence according to histology was only reported by one study that demonstrated that among 17 patients with active adenocarcinoma and venous thrombosis treated with VKA for 6 months, 12 (71%) had a recurrent VTE versus 2 of 11(18%) non-adenocarcinoma patients (p=0.018) (49).

4.3.5 Secondary Outcome Measures

4.3.5.1 Retrospective Studies

Bleeding events were reported in all studies (53-56). Forty-two of 218 (19%) had major bleeding and 8 (4%) had minor bleeding events during the anticoagulation period. Amongst the major bleeding events INR, PT or PTT were above the therapeutic range in 23 of 52 (44%) events, similar to the general VTE population (59). There were 7 (10%) intracranial bleeding haemorrhage (ICH) in 73 patients with brain malignancies who received anticoagulation compared to 1 event in the other 145

(0.007%) patients without primary or secondary brain malignancy (p = 0.002). A formal meta-analytic technique to evaluate bleeding risk according to malignancy characteristics was not feasible because studies did not provide this information. Furthermore, all studies used long-term VKA; therefore, bleeding risk evaluation would not have a comparator for meta-analysis regarding treatment strategy.

Sixty-four of 218 (29%) patients died. Thirty-five (55%) due to malignancy progression, 15 (23%) due to PE, 9 (14%) due to major bleeding and 5 (8%) due to other clinical causes. ICH was responsible for 8 of 9 (89%) deaths that occurred due to major bleeding. In the 2 studies that evaluated only brain malignancies, death due to ICH comprised 7 of 23 (30%) of all deaths (53;55) versus 2 of 41(0.5%) in the other studies (p = 0.011) (54;56).

4.3.5.2 Prospective Studies

Bleeding events were reported in all but one study (57). Two hundred and five of 4559 (5%) patients developed major bleeding events during the anticoagulation period, 14% of which were ICH (21;58). Minor bleeding events occurred in 31 of 4559 (0.7%) patients. A formal meta-analytic technique to evaluate bleeding risk according to malignancy characteristics was not feasible because studies did not provide enough information. Even so, major bleeding risk was assessed with respect to treatment strategy whenever possible. We evaluated 3 studies [10;16;32] that treated patients

with VKA and 1 study that used LMWH [15]. The relative risk of major bleeding was similar between the 2 treatment modalities [RR=1.641 (95% CI= 0.886 – 3.083; *p*= 0.131)].

Of the studies reporting cause of death, 531 of 1035 (51%) were judged to have been due to malignancy progression, 54 (5 %) due to PE, 48 (4%) due to major bleeding, and 405 (39%) from other causes [16;15;32;46].

4.3.6 Pooling all the Studies

Altogether the 10 studies comprised 331 of 4791 (7%) VTE recurrences. When I compared VTE recurrence rate according to tumour stage I found a statistically significant increased risk for patients with advanced malignancy when compared to patients with localized malignancy with a RR of 1.36 (95%CI, 1.06-1.74; p=0.01). I was not able to pool data for evaluation of the impact of VTE recurrence according to tumour site and histology.

I attempted to evaluate VTE recurrence risk according to treatment strategy (VKA or LMWH). I pooled the data of 6 studies that used VKA (10;11;49;54-56) and compared them to the 2 studies that used LMWH (21;57). One study that used UFH (53) and another study that did not report separate outcomes for the VKA and LMWH groups (58) were excluded from the analysis. Overall, VTE recurrence rate was 16%

(115/738) for patients treated with VKA and 11% (24/217) for patients treated with LMWH in these studies. The RR for VTE recurrence was 0.68 (95%CI 0.45-0.99; p=0.05), suggesting benefit of LMWH in comparison to VKA for the treatment of cancer-associated thrombosis. A sensitivity analysis stratifying patients according to duration of anticoagulation suggested that for patients treated for 3 months there was a significant benefit of LMWH over VKA [RR= 0.575 (95%CI= 0.362 – 0.904; p= 0.02)]. However, for patients treated for 6 months this benefit was lost [RR= 1.344 (95% CI= 0.464 – 3.111; p= 0.481)].

Overall, patients who were treated with VKA remained within the therapeutic range in 64% of the time in the retrospective studies (54-56) and 79% of the time in the prospective studies (10;21;49;58).

Assessment of publication bias for evaluation of recurrent VTE rate and tumour stage using a funnel plot was not possible since I would have only 3 studies available for inclusion in the funnel plot (11;54;58) .

4.4 Discussion of the Systematic Review

In this study, I systematically reviewed the available literature that evaluated the rate of VTE recurrence in patients with cancer-associated venous thrombosis attempting to find a link between malignancy characteristics and VTE recurrence rate. When I

combined all evaluable studies I found a modest but significantly higher risk of VTE recurrence in patients with metastasis [RR=1.36 (95%CI, 1.06-1.74; p=0.01)]. The evaluation of tumour site suggested that patients with lung or GI malignancies may present a higher risk for recurrent VTE and patients with breast cancer and haematological malignancies, a lower risk (10;21;58). I was not able to ascertain the relevance of tumour histology in the risk for VTE recurrence due to the lack of data information in the included studies.

A few observational studies that evaluated the incidence of a first VTE in patients with active cancer found a 2 to 19-fold higher incidence among patients with distant metastasis than in patients with localized malignancy (19;20;42;43). My less impressive results may be due to the fact that I was unable to pool all the studies for a formal meta-analysis or because risk of anticoagulant failure is due to different mechanisms / biology than those that cause a 1st VTE. Furthermore, in the retrospective studies, VTE recurrence rate was higher in patients with localized malignancy. This may have been simply due to chance or due to reporting bias, since the total number of participants was much smaller in the retrospective studies (n=218) than in the prospective studies (n=4573).

Very little data was available for evaluation of the relevance of primary tumour site and tumour histology. Nevertheless, the study conducted by Descourt *et al*. suggested

that histology may play an important role on the development of VTE recurrence. The available literature is scarce in this matter and presents with conflicting results (45). We were not able to pool the data regarding primary tumour site. However, the role of tumour site in VTE recurrence risk found in the individual evaluation of the included studies suggested that lung, GI and GU malignancies may predispose to a higher risk for recurrent VTE whereas breast and haematological malignancies may represent a lower risk (10;21;58). A large population-based study that evaluated more than 10,000 discharge summaries of patients admitted to hospital due to malignancy-associated VTE reported that lung cancer accounted for 21% of cases, colon cancer for 18% and prostate cancer for 17%; but it is unknown what proportion were first VTE and recurrent VTE (2). Another large retrospective study suggested that the concomitant diagnosis of VTE and cancer is much more prevalent in patients with lung, pancreas and colorectal cancer than in patients with breast or prostate cancer (47). Recently, a *post-hoc* analysis of the CLOT trial was presented in an international meeting (73). This data suggested that lung cancer [HR= 3.51 (1.62 - 7.62)]; unknown primary [HR= 3.63 (1.36 - 9.65)]; younger age [HR= 0.77 (0.06 - 0.90)] and metastasis [HR= 2.59 (1.29 - 5.60)] were independent predictors of VTE recurrence in the context of cancer-associated VTE. Conversely, Breast cancer [HR= 0.59 (0.17 - 2.01)] showed a trend towards being low risk for recurrent VTE.

Seventy percent of the included studies used VKA for the treatment of cancer-associated venous thrombosis. Overall, VTE recurrence occurred in the context of a therapeutic INR in more than 60% of cases. These results are supported by previously reported trials (6;40) and they may suggest that VTE recurrence risk in patients with malignancy-associated VTE treated with VKA, may not simply be related to a more difficult management of the oral anticoagulant. Rather, it may relate to underlying biochemical abnormalities that in certain malignancies may trigger a more aggressive hypercoagulable state that will promote a less effective response to anticoagulation with VKA and will ultimately lead to VTE recurrence regardless of therapeutic INR. When I compared different treatment strategies I found an overall trend towards a protective effect with the use of LMWH. However, the sensitivity analysis demonstrated discrepant results when studies were separated according to duration of anticoagulation. This result contradicts the literature (9), perhaps due to the heterogeneity of the included studies, but it may suggest further research is required.

Although my primary focus was VTE recurrence, I also evaluated bleeding complications associated with anticoagulation. The most common causes of major bleeding were supra- therapeutic levels of anticoagulation and the presence of an intracranial lesion. Intracranial haemorrhage occurred in 11% of patients that presented with major bleeding complication while on anticoagulants. This frequency is significantly higher than the 3 to 4% overall major bleeding rate observed in

previous studies that evaluated efficacy and safety of different anticoagulant strategies in patients with VTE (60-63). In these studies the main cause of major bleeding was GI haemorrhage, while in our review, it was intracranial haemorrhage. This suggests that the presence of intracranial malignant lesion may be an important risk factor for major bleeding in patients with VTE on anticoagulants, regardless of anticoagulant used. We also compared the rates of major bleeding between VKA and LMWH. The incidence of bleeding events did not seem to be influenced by the type of anticoagulant. Our results are supported by a recently published systematic review of RCTs that evaluated the impact of VKA or LMWH in patients with cancer-associated VTE. There was no difference on major bleeding rates among patients that were treated with either anticoagulant (9).

Malignancy was the main cause of death in all studies analysed. It was almost 10-fold higher than death from PE, the second most common cause of death. Our results are consistent with previously reported trials (1;6;40;41). This may suggest that overall survival rates are poor for patients who sustain a venous thrombotic complication in the context of active cancer regardless of the type of anticoagulant used. Although major bleeding was not the main cause of death, it is important to highlight that intracranial bleeding was responsible for 89% of deaths that occurred due to major bleeding in the retrospective studies. Furthermore, amongst the studies that evaluated only brain malignancies, it was the cause of death in 30% of all deaths.

4.5 Limitations and Strengths of the Systematic Review

The results of this review must be interpreted cautiously. One limitation of the study is that I was unable to establish the relevance of immobilization, surgery, chemotherapy and radiation therapy with VTE recurrence risk since these variables were not systematically reported in any of the included studies. Moreover, this review could be criticized for the fact that it included only observational studies, that evaluated a heterogeneous population of cancer patients and some of the studies were small. I elected not to impose limit to the number of included participants per study because I anticipated that it would be difficult to find a large number of studies that would provide detailed information about malignancy characteristics both at study enrolment and at VTE recurrence. In fact, the heterogeneity regarding study design and population was such, that we were basically limited to describe the results of the included studies and not able to pool the data and perform formal meta-analysis to evaluate the impact of malignancy characteristics in VTE recurrence risk. Meta-analysis was feasible for the evaluation of tumour stage only. This result was likely driven by the results of one single study that evaluated over 3800 patients (51). Although this study was part of a registry, participants were prospectively and consecutively enrolled. Furthermore, all the prospective studies were adequately designed and it is unlikely that the results of this registry would have severely impacted on the overall results of the prospective studies since results about tumour

histology, and evaluation of efficacy and safety of LMWH or VKA were very much in keeping with the literature and did not include this study.

One of my major findings was that 34 potentially relevant studies, including both observational and intervention studies were not included in our analysis due to the paucity of data about tumour characteristics in the patients with a recurrent VTE during anticoagulation. With respect to the included studies, malignancy characteristics were not completely reported, which limited our ability to uniformly evaluate the relationship between cancer characteristics and recurrent VTE. With the extensive literature search that I performed, which included published and unpublished data, it is unlikely that publication bias would severely affect this study results.

This systematic review is not the first one to address the issue of the impact of malignancy characteristics in the risk of VTE recurrence. The study conducted by Noble *et al*. attempted to evaluate the management of VTE in patients with advanced malignancy. They found that patients with advanced malignancy have a 2-fold increased risk of VTE recurrence compared to patients with limited stage malignancy. However,this study differs from ours since it included 4 RCTs and 4 observational studies that did not provide information about malignancy stage for patients with recurrent VTE (64). They considered all patients with VTE recurrence as having

advanced malignancy and they included 1 study that treated patients solely with VKA, with no initial anticoagulation with UFH or LMWH. Our review was based on very strict inclusion and exclusion criteria and we only included studies that provided accurate information regarding malignancy characteristics and objective diagnosis of VTE recurrence.

4.6 Conclusion of the Systematic Review

In summary, this review suggests that no definitive conclusions can be drawn from the published literature regarding the association between malignancy and patient's characteristics and VTE recurrence risk because reporting of malignancy characteristics in patients with cancer and recurrent VTE during the anticoagulation period is scarce. Strikingly, this was our most important finding. With respect to our primary outcome, at most, the literature suggests that younger patients seem to be at higher risk of recurrent VTE as well as patients with metastatic malignancy or lung cancer. The impact of tumour histology on VTE recurrence risk remains debatable since only 1 study to date has addressed this issue appropriately. Future studies evaluating cancer-associated thrombosis should clearly state malignancy characteristics at study enrolment and at VTE recurrence so that a better understanding of the diverse cancer characteristics can ultimately be linked with the risk of VTE recurrence. This may enable the development of new and more specific treatment options for cancer-associated VTE.

5.0 SURVEY RESEARCH

5.1 Rationale for the Survey

A survey is an important form of scientific inquiry that merits rigorous design and analysis. A survey is an important part of this thesis because it helped me anchor retrospective study findings with my future aspiration of research in the field of malignancy and thrombosis.

5.2 Objectives

5.2.1 Primary Objective

To conduct a survey with thrombosis experts in North America and Europe to determine what is the lowest risk of VTE recurrence that thrombosis specialists would consider acceptable for patients with cancer- associated thrombosis

5.2.2 Secondary Objectives

i) To evaluate how important thrombosis specialists would consider a clinical prediction rule is for categorization of patients with malignancy-associated thrombosis into low risk and high risk of VTE recurrence

ii) To determine whether Thrombosis specialists would be willing to participate in a randomized control trial (RCT) to compare different treatment strategies and for the validation of the CPR in patients with cancer-associated VTE

iii) To determine what level of VTE recurrence risk they would consider high enough so that new therapeutic interventions should be evaluated

5.3 Methods

We conducted an internet-based self-administered survey, with questions presented in a single scrolling page. SurveyMonkey® was the electronic software used for the questionnaire development and analysis.

I followed the Basic Dillman's Approach to assess potential respondents with the intention to maximize our response rate and we also used personal contact of recipients whenever possible (personal e-mail) (65) (see *appendix 1*). A cover letter explaining the objectives and the rationale for the survey, as well as the importance of each recipient's participation was provided. An estimate of the time to be spent answering the questionnaire was also highlighted.

5.3.1 Sampling Frame

My target population was the thrombosis specialists from North America and Europe. We selected the experts from our own list of collaborators and from the membership list of the Canadian Haematology Association and the membership list of the ISTH (International Society in Thrombosis and Haemostasis).

5.3.2 Question format

Question stems contained between 20 and 40 words in the maximum, so that they could be easy to understand and interpret. We also provided background information and examples within the question structure to aid the understanding of each question stem. The questionnaire consisted of 5 questions.

5.3.3 Response format

Answers were provided as multiple-choices with one possible answer. I provided 5 structured possible responses and a "other (please specify)" option with an appropriate field for comments of 50 words in the maximum.

5.3.4 Pre-testing, Pilot testing and Sensibility testing

A pre-test and a pilot test with the thrombosis specialists at the Thrombosis Unit of the Ottawa hospital were conducted to ascertain the clarity and interpretation of each question in the questionnaire, the acceptability of the questionnaire, its flow,

evaluation of possible redundant or unclear questions or answers, and to record the time spent to fill out the full questionnaire.

The pre-test comprised 4 steps:

1. Review of questionnaire by knowledgeable colleagues (Dr. Marc Rodger and Dr. Marc Carrier) within the thrombosis clinic of the Ottawa hospital to assess if questions were well designed and concise; and whether questions needed to be added or removed
2. Sensibility test: interviews with 10 thrombosis specialists of the Ottawa hospital to assess the comprehensiveness, clarity, face and content validity of the questionnaire, addressing issues such as whether response formats were easily understood, whether there were items inappropriate or missing and how likely the questionnaire was to address the survey objective.
3. Pilot study: evaluated whether there were any skewed responses, to evaluate the likely response rate and if some questions were being skipped
4. New set of eyes: 1 person who was not a thrombosis specialist (Dr.Tim Ramsay) evaluated the questionnaire looking for "dumb" mistakes, such as grammatical errors.

Changes to the format of the questions or answers were done according to the pre-test results.

5.3.5 Data Analysis

Frequency distribution was generated for all partially or fully responded questions. I expected a response rate ranging between 50 and 55% as per previously reported studies (66;67). We did not test for inter-rate reliability by formulating questions that are negative mirror image because the answers were expected to be simple and we wanted to ensure a high response rate. For the same reason we did not feel that it was necessary to conduct a test-retest reliability, posing the same questions to the same individuals in a different period of time.

5.4 Results of the Survey

In the pre-test section of the survey, overall thrombosis experts of the Ottawa Hospital agreed that the survey had face validity. However, they suggested that I modify the stem of questions 2 and 5, by inserting examples in these questions in order to improve their clarity.

One hundred and three thrombosis specialists from North America and Europe were identified through our own list of collaborators, or from the membership lists of the Canadian Hematology Association or the ISTH. Overall, 60 (58.2%) respondents started the survey and 58 (96.7%) completed it. Thirty-two (58.3%) respondents started or completed the survey after the first questionnaire was electronically sent

out. The remaining 28 participants responded between Day 7 and Day 35. Questionnaire details are shown in Table 4.

Nearly 92% of participants agreed that a clinical prediction rule that would enable categorization of risk of recurrence in patients with malignancy-associated VTE into low and high risk of VTE recurrence and prior to initiation of therapy, would be important and may influence the current management of these patients. When questioned about what would be an acceptable VTE recurrence risk low enough that they would consider treating an index VTE in a patient with active cancer, with VKA instead LMWH, 80% of respondents agreed that a risk below 7% would be an acceptable threshold. Moreover, nearly 80% of participants said they would participate in a treatment study that utilizes a CPR for stratification of patients into low and high risk of VTE recurrence. All respondents that stated they would agree to participate in such a study preferred a RCT instead of a prospective cohort study. Finally, all respondents agreed that patients with a very high risk of recurrence in spite of appropriate treatment with LMWH deserve a novel therapeutic strategy (such as 120% weight-adjusted LMWH for 6 months or use of inferior vena cava filter together with LMWH) to be compared to LMWH alone, the current standard of care. Sixty-three percent of respondents considered that a risk of recurrence high enough to try some novel therapeutic strategy would be between 13 and 17%.

Table 4. Questionnaire for Thrombosis specialists

Questionnaire	*Answers*	*Response Percent*	*Response Count*
1. Patients with cancer and VTE frequently develop recurrent VTE despite, and while on anticoagulant therapy. **Do you think that a clinical prediction rule that would enable you to categorize the risk of recurrence in these patients, prior to initiation of therapy, into low and high risk of VTE recurrence, would be important and may influence your current management of these patients?**	***YES***	***91.7%***	***55***
	NO	***8.3%***	***5***
2. In patients with cancer and VTE after using LMWH for the first week of treatment, VKA or LMWH are options for the long-term phase of therapy. Many physicians prefer LMWH due to data that suggests recurrence rates are lower with LMWH (7%) compared to VKA (13%). However, some patients may be at low risk of recurrence regardless of using LMWH or VKA. For example, a VTE that develops in a patient with early stage colon cancer after curative resection may be at low risk of recurrence. If bleeding risks were the same, **at what risk of VTE recurrence (during and while on the first 6 months of anticoagulant treatment) would you consider treating an index VTE in a patient with active cancer, with VKA instead LMWH, thereby avoiding the high cost and inconvenience of injections?**	***1%***	***0.0%***	***0***
	3%	***23.3%***	***14***
	5%	***31.7%***	***19***
	7%	***25.0%***	***15***
	9%	***8.3%***	***5***
	Other	***11.7%***	***7***
3. In question 2 we asked what recurrence rate would be acceptable to use VKAs. **If a prediction rule could identify patients with the recurrence rate you found acceptable in question 2, would you participate in a treatment study that utilizes the prediction rule?**	***YES***	***79.7%***	***47***
	NO	***20.3%***	***12***
	Skip question		***1***
4. If you answered 'Yes' in question 3, **would you prefer a cohort study using VKA, as a first step to validate the rule or would you rather go directly to a randomized controlled trial that compares VKA to LMWH for 6 months for**	***Cohort***	***24.0%***	***12***
	RCT	***76.0%***	***38***

validation of such a clinical prediction rule? If you answered 'No' in question 3, please, skip to question 5.			
5. It is possible that some patients with cancer are at higher risk then the overall VTE recurrence rate of 7% we quoted above. For example, an elderly 75 year old female with previous VTE, metastatic pancreatic cancer who develops VTE prior to chemotherapy may have a recurrence risk as high as 25%. **If we could identify patients at high risk of recurrence despite and while on optimal LMWH therapy, at what level of recurrence would you consider evaluating in a RCT, a novel therapeutic strategy versus LMWH alone for 6 months?**	***9%*** ***13%*** ***17%*** ***22%***	***16.7%*** ***36.7%*** ***26.7%*** ***10.0%***	***10*** ***22*** ***16*** ***6***

5.5 Discussion of the Survey

The survey aimed to evaluate thrombosis specialists' interest in a clinical prediction rule for stratification of patients' risk of a recurrent VTE in the cancer setting. It also sought to evaluate the minimum acceptable threshold of risk at which the specialist would consider treating low risk patients with VKA. This information would aid in the calculation of the absolute risk threshold for VTE recurrence to be adopted in a possible future prospective study.

Overall, specialists considered that a CPR is very relevant and would be a useful tool to better tailor treatment for patients with malignancy-associated VTE. Furthermore, the majority of respondents would agree to participate in an intervention study to further validate the CPR and to evaluate different treatment strategies for patients considered to be low or high risk of recurrent VTE. There are very little data comparing different treatment strategies in patients with malignancy-associated thrombosis (1;6;38;40;41). These studies evaluated patients with diverse types of malignancy and VTE and they did not perform a sub-group analysis to test for difference in susceptibility of VTE recurrence risk according to malignancy characteristics. Altogether, these studies encompass a little more than 1000 patients and they are the only evidence that LMWH may be a better strategy than VKA for

patients with cancer and VTE. Importantly, only 1 study had adequate power to assess for VTE recurrence risk (6).

5.5.1 Limitations and Strengths of the Survey

This survey was performed with the intention to gather information from thrombosis specialists regarding the relevance of a CPR and possible novel treatments strategies for patients with malignancy-associated thrombosis.

One limitation of the survey is that it was not based on pre-standardized and validated questionnaires. It was elaborated by myself and another thrombosis specialist (Dr. Phil Wells) based on the lack of evidence regarding patients' risk of recurrent VTE according to the diverse types of malignancy; and the need to evaluate other specialists' opinion with respect to this important matter. Therefore, we cannot ascertain the reproducibility of the survey. Another limitation relies on the fact that respondents considered that 7% and 13% were recurrence risks respectively low and high enough that they would consider participating in a treatment trial to test different therapeutic strategies for patients with cancer and VTE. It is important to highlight though, that these frequencies (7 and 13%) relate to the results of the study we referenced in the cover letter and may reflect the specialists' opinions but we cannot discard the possibility of recall bias.

I intentionally made the survey simple, with very few but relevant questions to try to optimize response rate. In fact, response rate was very good and above the expected 50 to 55% (66;67). The electronic format with multiple-choice answers made it easy for the respondents to comply and complete the full questionnaire. In addition, the Dilman's approach was a fairly efficient tool. I had 50% of specialists responding after the first questionnaire was sent out. The repeated questionnaires preceded by a cover letter that emphasized the relevance of the survey improved the response rate (another 28 (48.7%) respondents between D7 and D35 questionnaires).

5.6 Conclusion of the Survey

The survey results indicate that thrombosis specialists believe that 7% or less is the lowest acceptable risk of VTE recurrence to consider treating patients with cancer-associated thrombosis with VKA instead of LMWH. Moreover, the experts agree with my belief that more clinical research should be performed to better stratify patients risk and to better evaluate anticoagulation strategies to avoid the hazards of VTE recurrence in the patient with active malignancy and VTE.

6.0 RETROSPECTIVE STUDY

6.1 Methodological Standards for Clinical Prediction Rules (CPR) and Rationale for the study

6.1.1 Methodological Standards for Clinical Prediction Rules (CPR)

Wasson (68) published the methodological standards for Clinical Prediction Rules and later those standards were updated (69). A CPR is an algorithmic decision-making tool that ideally, is derived from original research using strict methodological guidelines. CPRs are appealing because they offer several potential benefits for practitioners, patients and the Health Care System such as, reduction in the clinical uncertainty at the bedside, improvement of quality of care for patients and may decrease exposure to costly and potentially hazardous or unnecessary procedures (67). In short, developing a CPR should include the following (75):

1) The outcome or diagnosis to be predicted must be clearly defined, clinically important and the assessment of the outcome must be blinded (i.e. the final arbiter of outcome must have no prior knowledge of potential predictive variables under study).

2) The clinical findings to be used as predictive variables must be clearly defined, standardised, and their assessment must be done without knowledge of the outcome (i.e. blinded).

3) The reproducibility of the clinical findings used as predictive variables must be demonstrated and the reproducibility of the rule must be demonstrated.

4) The patients in the study should be selected without bias and should represent a wide spectrum of clinical and demographic characteristics to increase the generalisability of the study results.

5) The statistical techniques used to derive the rule must be identified and valid.

6) The accuracy of the prediction rule in classifying patients with the outcome (i.e. sensitivity) and without the outcome (i.e. specificity) should be demonstrated.

7) Prospective validation in a second independent set of patients is an essential test of a prediction rule's accuracy and clinical utility (i.e. the effects of clinical use of the rule should be prospectively measured).

8) Clinical prediction rules should be sensible i.e. have a clear purpose, be relevant, demonstrate content validity, be concise, and be easy to use in the intended clinical application. The use of the rule should provide a probability of disease and should imply a course of action.

6.1.2 Rationale for the study

The current standard of care for patients with cancer-associated VTE is long-term LMWH (17;18). However, the association between VTE recurrence risk and treatment management according to malignancy characteristics is largely unknown. VTE is highly prevalent among the cancer population with a 4 to 6-fold increased risk when compared to the non-cancer population (3). Moreover, the risk of VTE recurrence during appropriate anticoagulation ranges between 7 and 13% depending on the type of anticoagulant used (9). A better understanding of the different malignancy

characteristics that influence the risk VTE recurrence is needed, so that the practitioner may offer a better tailored treatment approach for the patient with cancer-associated VTE without exposing the patient to unnecessary risk of bleeding and to the high psychological and financial cost of the prolonged use of LMWH.

The systematic review performed as part of this thesis suggests that patients with youger age (less than 65 years old), or metastatic malignancy, or lung malignancies sustain the greatest risk for recurrent VTE during the anticoagulation period. Most importantly, the systematic review revealed that the available literature does not accurately evaluate potential predictors of recurrent VTE in patients with malignancy-associated venous thrombosis. This important information aligns with our goal to investigate which characteristics would be the best independent predictors for VTE recurrence in patients with cancer-associated VTE since the current standard of care is LMWH. For an accurate prediction rule, to conduct a randomized controlled trial (RCT) or a prospective cohort study to evaluate VKA treatment for low risk patients with malignancy will be required. This subset of patients should be likely to derive equal benefit from the use of VKA or such a study could be considered unethical. Furthermore, patients with high risk of recurrent VTE, who fail standard treatment with LMWH, should be evaluated for a more aggressive anticoagulation approach (e.g. higher dose of LMWH or LMWH + antiplatelet agents).

As suggested by the survey, a clinical prediction rule for stratification of patient's risk for the development of a recurrent VTE during the anticoagulation period is needed and welcome by thrombosis specialists. Therefore, even though the retrospective design is not the best for derivation of a clinical prediction rule, it may be a useful tool for collection of potentially relevant data regarding risk of VTE recurrence in patients with cancer-associated thrombosis. For the moment, trying to ascertain the feasibility of a CPR with a retrospective dataset seems to be a reasonable approach, so that in the future, a definitive CPR may be prospectively derived and validated.

6.2 Objectives

6.2.1 Primary Objective

To assess the feasibility for derivation of a clinical prediction rule that stratifies risk of VTE recurrence in patients with cancer and venous thrombosis through identification and evaluation of characteristics of malignancy (e.g. primary tumour site, histology, and metastasis) along with other clinical characteristics (e.g. age, gender).

6.2.2 Secondary Objectives

1) To determine whether type of anticoagulation influences VTE recurrence risk

2) To determine if rates of major and minor bleeding vary by VTE recurrence risk (classification according to the Internal Society of Thrombosis and Homeostasis criteria) (51) (*see appendix 2*)

3) To determine if rates of major bleeding are dependent on type of anticoagulation (VKA or LMWH)

4) To determine overall mortality during the anticoagulation period

6.3 Patients and Methods

6.3.1 Study Design

A chart review (electronic and hardcopy charts) was conducted to collect data from patients with cancer and VTE who were diagnosed and/or followed at the Thrombosis Unit from January 2002 to December 2004 and from January 2007 to July 2008. These treatment periods were selected because it was expected that the vast majority of patients would have been treated with VKA for cancer-associated VTE in the first period, as was the standard of care at that time; and with LMWH in the second period, the current standard of care.

The end point for collection of data was determined as follows:

1. The patient developed a recurrent VTE during the anticoagulation period, OR
2. The last time patient was seen at the Ottawa Hospital provided that he was still on anticoagulation, OR

3. Time of death , OR

4. When anticoagulation was terminated for any reason

We included all the recurrent VTEs that occurred at any time during these observation periods, provided patients were on anticoagulation, however, we divided the recurrence period into: a) within the first 6 months after the start of anticoagulation or b) after 6 months of anticoagulation. From all patients we collected potentially relevant data for the development of a clinical prediction rule that would stratify the risk for VTE recurrence.

6.3.1.1 History

Demographic characteristics were collected including age and gender. Tumour characteristics included primary tumour site, histology and stage of solid tumours (according to the TNM classification) (50). Secondary risk factors for VTE recurrence such as major surgery, use of chemotherapy and/or hormone therapy within the first 3 months of VTE diagnosis were collected. A history of previous VTE was also collected. Characteristics of index and recurrent VTE as well as type of anticoagulant strategy used were recorded.

6.3.2 Selection of Participants

6.3.2.1 Inclusion criteria

We included data from adult patients (18 years of age or older) with active malignancy and objectively diagnosed index VTE [pulmonary embolism (PE), proximal deep venous thrombosis (DVT) of the legs or arms, PE + DVT (both); unusual site thrombosis].

1. Active cancer is defined as diagnosis of cancer, other than basal-cell or squamous-cell carcinoma of the skin, within 6 months before VTE diagnosis, any treatment for cancer within the previous 6 months, or recurrent or metastatic cancer, or concomitant diagnosis of VTE and cancer (within 6 months after VTE diagnosis).
2. Objectively proven DVT is defined as proximal DVT of the lower extremities confirmed by compression ultrasound or contrast venography with evidence of thrombus in the calf trifurcation or more proximal veins. Acute proximal DVT of the arms and/ or neck is defined as objectively proven DVT confirmed by compression ultrasound or contrast venography with evidence of thrombus in the axillary vein, subclavian vein, internal jugular vein, superior vena cava or innominate vein. PE is defined by high probability on ventilation-perfusion lung scan (V/Q scan) with filling defects in segmental vessels or larger vessels, or multiple subsegmental filling defects on Computerized Tomography Pulmonary angiography (CTPA). For this study, single subsegmental filling defects were not considered to represent PE, unless a concomitant DVT was diagnosed. Unusual site thrombosis is defined as filling defect present at any site other than

arms, legs or lungs diagnosed through Computerized tomography, or Magnetic resonance imaging (MRI) or Ultrasound of the abdomen, pelvis or head together with constitutive symptoms or not (*e.g*. cerebral sinus thrombosis; portal vein thrombosis, ovarian vein thrombosis).

6.3.2.2 Exclusion criteria

Patients with index arterial thrombosis were excluded from the analysis.

6.3.3 Primary Outcome measure

The primary outcome measure is VTE recurrence during the anticoagulation treatment period. For DVT, diagnosis required confirmation by imaging techniques (compression ultrasound or contrast venography) and for PE confirmation required CTPA or V/Q lung scan. For unusual site thrombosis confirmation required Computerized tomography, or Magnetic resonance imaging (MRI) or Ultrasound of the affected area.

VTE Recurrence is defined as:

Deep Vein Thrombosis

1. Compression ultrasound revealing a new (compared to baseline ultrasound) area of non compressibility of a venous segment from the trifurcation area or

above or an increase of the noncompressibility ≥ 4mm were considered diagnostic of recurrent deep vein thrombosis of the leg OR

2. Venography demonstrating a constant intraluminal filling defect in the deep veins above the trifurcation of the popliteal vein was considered diagnostic of a deep vein thrombosis OR

3. Compression ultrasound revealing a new (compared to baseline ultrasound) area of non compressibility of a venous segment from the axillary vein or above or an increase of the noncompressibility ≥ 4mm were considered diagnostic of recurrent deep vein thrombosis of arm OR

4. Venography demonstrating a constant intraluminal filling defect in the axillary vein and above was considered diagnostic of recurrent DVT of the arm

Pulmonary Embolism

1. Recurrent PE was considered when a new mismatched large, segmental or greater, perfusion defect was documented in the V/Q scan OR
2. If the CTPA demonstrated a central intraluminal filling defect in a segmental or greater vessel in an area normal on the baseline study.

Unusual site thrombosis

Recurrent unusual site thrombosis was considered when extension of a previous filling defect occurred or a new venous filling defect was present at any site other than arms, legs or lungs diagnosed through Computerized tomography, or MRI or

Ultrasound as appropriate; of the abdomen, pelvis or head (*e.g.* cerebral sinus thrombosis; portal vein thrombosis, ovarian vein thrombosis; arterial thrombosis).

6.3.4 Secondary Outcome Measures

1. D-Dimer levels before and after recurrence according to the IL-test (Instrumental Laboratory, Lexington, USA) parameters
2. Major and minor bleeding events (*see Appendix 2*)
3. All cause mortality

6.3.5 Statistical analysis

6.3.5.1 Sample Size

The methodological criteria for the development of clinical prediction rules states that a minimum of 5 to 10 patients per predictor studied are required in the smallest outcome category (68). We suspected that age, gender, stage of malignancy, histology, tumour site and site of initial VTE would be relevant variables and our systematic review helped us determine whether we should include other variables. To develop a clinical prediction model we needed between 50 to 100 events (VTE recurrence cases) to include 10 variables in the final logistic regression model.

6.3.5.2 Primary Analysis

SAS 9.2® was used for the analysis. Baseline characteristics of participants were analyzed by means of descriptive statistics. Demographic and clinical characteristics of study patients by recurrence status were compared using a two-sample t-test for continuous variables and a two-way contingency table using Chi square or Fisher's exact test for categorical variables, as appropriate. The risk of VTE recurrence over time was estimated according to the Kaplan-Meier method.

For the clinical prediction rule, I analysed only the patients who had a recurrent VTE within the first 6 months of anticoagulation. Six months is the minimum standardized treatment approach for patients with cancer-associated VTE (6;16;17). I performed univariate analysis to determine the strength of association between each potential predictor and VTE recurrence (dependent variable). This process aided the selection of the best variables for the multivariate analysis. The appropriate univariate technique was chosen according to the type of data: for nominal data, the chi-square test with continuity correction; for ordinal variables, Wilcoxon rank-sum test; and, for continuous variables, the unpaired 2-tailed t-test, using pooled or separate variance estimates as appropriate. All potential predictor variables ($p<0.25$) were evaluated in a Logistic regression model. First, a likelihood ratio test was performed to compare the full model to the full model without interaction terms. If the likelihood test was significant ($p<0.05$) we would then perform backward elimination of the individual

interaction terms to identify significant interaction terms. All significant interactions would have been retained in the model.

Age was evaluated both as a continuous variable and as a categorical variable, since it was dichotomized (cut-off point= 65 years) to make the model simpler and easy to use, in the case age would be included in the final model. Goodness of fit of the possible models was evaluated using the likelihood test and Hosmer-Lemeshow test.

I would test for confounding by removing all potential confounding factors from the models and re-evaluating the chi-square of the analysis of maximum likelihood estimates results. Should it be significantly changed from the full model, a confounding factor would be identified and removed from the model.

I also performed a sensitivity and specificity analysis using Recursive Partitioning Analysis for the selection of the most appropriate variables. The decision tree was performed using R Statistic® software because Enterprise Miner, the appropriate software for decision tree analysis in SAS 9.2® is not currently available in the University of Ottawa package. Recursive partitioning allows a heterogeneous group with and without a specified outcome to be split in increasingly homogenous strata (nodes). It starts partitioning the groups by using the predictor variables with the most

significant chi-square. The chi-squares are adjusted for multiple comparisons by the method of Bonferroni.

Interobserver reliability assessment was performed using the variables that were significant in the univariate analysis to evaluate the suspected recurrent venous thromboembolic events. It was performed by myself (ML) and the second observer was Dr.Vi Dao. The interobserver reliability of each potential predictor variable was determined by calculating a two rater unweighted Kappa statistic. Kappa is defined as: $K=P_0-P_e$ /1-P_e; where P_0 is the actual probability of agreement and P_e is the expected agreement by chance. A kappa score above 0.8 is considered excellent reliability, a kappa score above 0.6 is considered good reliability and a kappa score below 0.4 is considered poor reliability. For continuous and ordinal variables the optimal cut-point was determined at the univariate data analysis stage and kappas were calculated for the optimal cut-point. Only those predictors with good reliability (Kappa >0.6) or better were retained for inclusion in the logistic models being tested.

The regression coefficients of each model were used to weight the predictors. Each predictor variable in the models was assigned an integer point score by rounding its associated regression coefficient to the nearest integer, and the best predictive point score cut-off for each model was identified. Rounding the regression coefficients to the nearest integer would make the clinical prediction rule easy to use and remember.

Next, the patient's predicted state at the best point score cut-off for each model was compared with the true state by 2x2 contingency tables to determine the risk of VTE recurrence for each group: low risk group, intermediate risk group, high risk group and the rate of low risk excluded proportion [(a+b)/(a+b+c+d)] (*Appendix 3*); as well as specificity, sensitivity and negative predictive values (true negatives). Based on these parameters, the classification performance of competing logistic models was assessed in order to identify the best combination of predictor variables to produce a safe and useful candidate clinical prediction rule that accurately stratifies patients with cancer-associated VTE into high, intermediate and low risk of recurrent VTE.

The final model should have the largest number of low risk group (highest excluded proportion), the lowest recurrence rate in the low risk group (at most 7% during the anticoagulation period, according to our survey results), and the highest recurrence rate in the high risk group (over 13% during the anticoagulation period, according to our survey results). At the same time, the rule should have face validity, and a practical number of predictor variables that would be easy to remember and apply.

6.3.5.3 Secondary analysis

Relative risks with correspondent 95% confidence intervals for death from all causes, major bleeding events and all bleeding rates were calculated according to VTE recurrence risk and type of anticoagulant used (VKA or LMWH).

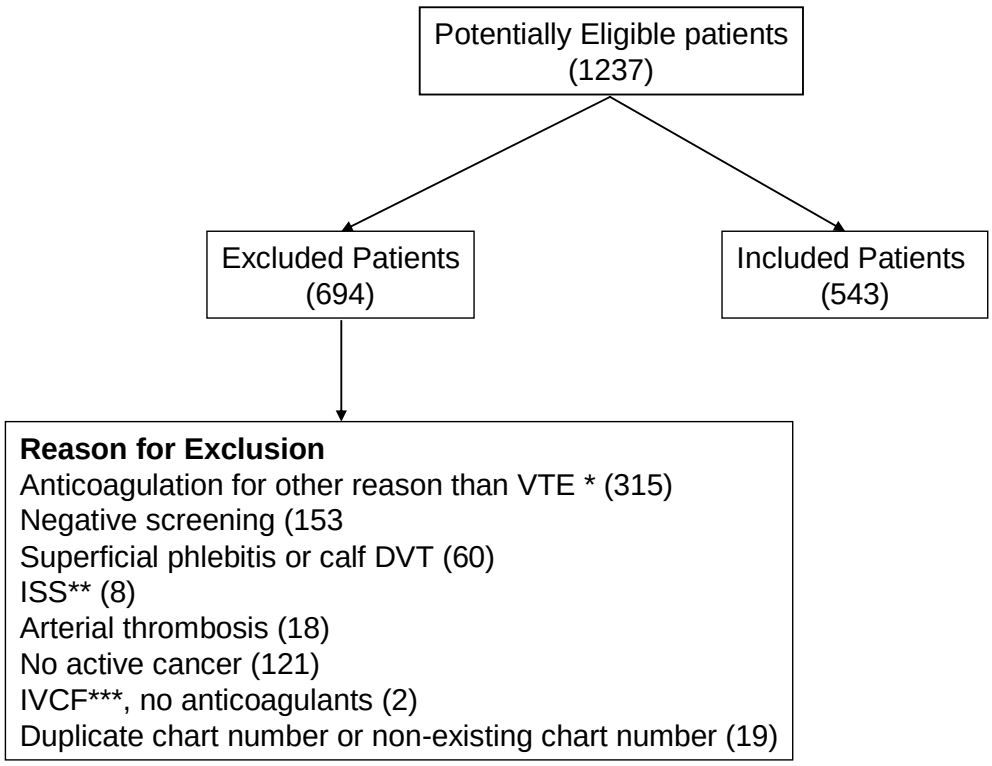
6.3.6 Research Ethics Board Approval

The retrospective cohort study conducted as part of this thesis project was submitted for evaluation by the Ottawa Hospital Research Ethics Board and approved.

6.4 Results of the Retrospective Study

This retrospective single center study evaluated the electronic/hard copy chart of all cancer patients followed at the Thrombosis Unit of the Ottawa Hospital from January/2002 to December/2004 and January/ 2007 to July/ 2008. In total, there were 1237 evaluable patients, of whom 694 did not fulfill our inclusion criteria, leaving 543 patients for inclusion in this analysis (Figure 4).

Figure 4. Flowchart of included and excluded patients



Anticoagulation for other reason than VTE*: perioperative management of patients with cancer on anticoagulants due to atrial fibrillation or prosthetic heart valves; ISS**: isolated subsegmental PE; IVCF***: inferior vena cava filter

When type of long-term anticoagulation was analyzed according to date of diagnosis of index VTE we found the following:

- From 2002 to 2004, one hundred and forty-two patients were evaluated. Among them 110 (77.5%) patients used VKA for long-term anticoagulation and LMWH was the long-term therapy of choice in 32 patients (22.5%) (p= 0.0001)
- From 2005 to 2008, four hundred and one patients were diagnosed and followed at the Thrombosis Unit of the Ottawa Hospital. Among them 89 (22.4%) patients used VKA for long-term anticoagulation and 312 (77.8%) used LMWH (p= 0.0001)

Therefore, in the entire study population, VKA was the long-term treatment of choice in 200 (36.6%) patients and LMWH in 343 (63.3%) patients (p= 0.0001).

6.4.1 Baseline Characteristics of participants

Baseline characteristics of included patients are depicted in Table 5. There were 240 (44.2%) males. Mean age of participants was 63 years. There were 58 (10.7%) patients with haematological malignancies and 485 (89.3%) patients with solid tumours. The most common primary tumour site was GI malignancy [140 (25.8%)] followed by lung [96(17.9%)] and breast cancer [85 (15.6%)]. Of the 85 patients with breast cancer, 1 was a male. One hundred and sixty-three (30.0%) patients were classified as miscellaneous [bone (4); brain (27); genito-urinary (47); gynaecological (50); hepato-biliary (4); larynx (1); musculo-skeletal (10); mesothelioma (1); neuroendocrine(2); oropharynx (5); skin (4); thymoma (4); unknown primary (4)]. Although genito-urinary and gynaecological malignancies were relatively frequent,

they comprised less than 10% of the total study population and we elected to put them in the miscellaneous group. At VTE presentation, 238 (43.8%) patients had distant metastasis. Adenocarcinoma was present in 306 (56.4%) patients with solid tumours.

Two hundred and eight (38.3%) patients had lower limb DVT, 141 (26.0%) had a PE and 67 (12.3%) had both. Upper limb DVT was the initial event in 106 (19.5%) patients among which 64 (60.4%) had catheter-related thrombosis and 20 (18.8%) had extrinsic compression by tumour bulk. Unusual site thrombosis was present in 21(3.9%) patients. Details of malignancy characteristics in patients with upper limb depicted in table 6.

Table 5. Baseline Characteristics of Patients

Characteristics		VTE Recurrence		
		NO (n= 465)	**YES (n= 78)**	**Total (n=543)**
Gender	**M/F (%)**	211/254 (45.4/54.6)	29/49 (36.4/63.6)	240/303 (44.20/55.80)
Mean Age	**Years(SD)**	63.5(13.5)	60.4 (13.2)	63.0 (13.5)
Age category	**< 65**	235 (50.5)	46(58.4)	281 (55.1)
	> 65	230 (49.5)	32(41.6)	262 (44.9)
Primary tumour site n(%)	**Haematological**	51(10.9)	7(8.9)	58(10.7)
	Lung	71(15.3)	25(32.1)	96(17.7)
	GI	123(26.5)	17(22.1)	140(25.8)
	Breast	79(17.0)	6(7.8)	85(15.6)
	Miscellaneous	141(30.3)	23(29.5)	164(30.2)
Histology n(%)	**Adenocarcinoma**	266 (57.0)	40(52.0)	306(56.4)
	Non-adenocarcinoma	151(32.8)	31(38.9)	183 (33.5)
	Not applicable*	48(10.2)	7 (9.1)	55 (10.1)
TNM Stage n(%)	**1**	59(12.6)	2(2.6)	61(11.2)
	2	64(13.7)	10(13.0)	74(13.6)
	3	72(15.4)	12(15.6)	84(15.5)
	4	195(41.9)	42(54.6)	237(43.7)
	Other**	75 (16.3)	12(14.2)	87(16.0)
Previous VTE n(%)	**Yes**	35(7.5)	11 (14.3)	46 (8.4)
	No	430 (92.5)	67 (85.7)	497 (91.6)
Major Surgery n(%)	**Yes**	106 (22.8)	16 (20.5)	122 (13.9)
	No	359 (77.2)	62 (79.5)	421 (14.)
Chemotherapy/ Hormonetherapy n(%)	**Yes**	245(52.7)	35 (44.7)	280 (51.5)
	No	220 (47.3)	43(55.3)	263 (48.5)
Index VTE n(%)	**Leg DVT**	177 (38.1)	31 (38.1)	208 (38.3)
	PE	123 (26.5)	18 (23.7)	141 (26.0)
	Both	59 (12.6)	8 (10.6)	67 (12.3)
	Arm/neck DVT	86 (18.4)	20 (26.3)	106 (19.5)
	Unusual site	20(4.3)	1(1.3)	21 (3.9)
Symptomatic PE	**Yes**	149(82.5)	25 (96.2)	174 ()
	No	33(17.5)	1(3.8)	34 ()
Long-term	**VKA**	169(36.2)	31(39.7)	200 (36.7)

Anticoagulation n(%)	**LMWH**	296 (63.8)	47 (60.3)	343 (63.3)
Choice of initial anticoagulant n (%)	**Dalteparin**	330 (71)	53 (67.5)	383 (70.5)
	Tinzaparin	121(26)	19(24.7)	140(25.8)
	UFH	5(1.1)	3(3.89)	8 (1.5)
	Enoxaparin	6(1.3)	2(2.6)	8 (1.5)
	Danaparoid	3(0.6)	1(1.3)	4 (0.7)
Duration of anticoagulation in weeks Mean (range)	**LMWH**	44.9 (1- 220)	15.0 (1 - 156)	42.0 (1 - 220)
	VKA	81.0 (1 - 520)	37.4 (1 - 172)	74.2 (1 - 520)

Not applicable*: haematological malignancy; Other:** hematological and brain malignancy; [£] History of VTE before the event the occurred in the context of cancer

Table 6. Characteristics of patients with upper limb DVT

Characteristics		**Index DVT**
Gender	**M/F (%)**	40 /66 (38/62)
Mean Age	**Years (SD)**	59 (13.5)
Primary tumour site n(%)	**Haematological**	14 (13)
	Lung	21 (20)
	GI	25 (24)
	Breast	33 (31)
	Miscellaneous	13 (12)
TNM Stage n(%)	**I**	10 (10)
	II	14 (13)
	III	20 (19)
	IV	48 (45)
	Not applicable**	14 (13)
Catheter	**Yes**	64 (60)
	No	42 (40)
Extrinsic Compression		20 (19)

Type of Catheter*	**PICC**	27 (25)
	Porth-a cath	35 (33)
	Hickman	1 (1)
	No catheter	43 (41)

*PICC: peripherally inserted central catheter; Porth-a-cath: totally implanted central venous catheter; Hickman-Broviak: semi-implanted central venous catheter
** Not applicable: haematological or brain malignancy

6.4.2 Primary Outcome

Details of VTE recurrence according to patient and malignancy characteristics are presented in Table 5.

6.4.2 .1 Patients' characteristics and anticoagulation strategy

Seventy-eight (14.4%) patients recurred during the anticoagulation period. Females were slightly more susceptible to recurrence than males [49/303 (16.2%) versus 29/241(12.0%) (p= 0.0584)]. Fifty-five (70.5%) patients developed recurrent VTE within the first 6 months of anticoagulation whereas 23 (29.5%) recurred after the first six months of anticoagulation (p= 0.0002) (Figure 4). Among these, 10 (12.8%) patients recurred after 1 year of anticoagulation (1 patient using LMWH and 9 patients using VKA). Overall, VTE recurrence occurred in 31 (15.5%) patients using VKA and 47 (13.7%) patients using LMWH. The relative risk for VTE recurrence was not significantly different between patients who used VKA or LMWH [RR= 1. 13 (95%CI, 0.743 - 1.711; p= 0.565)]. I performed a sensitivity analysis considering only the patients who recurred within the first 6 months of anticoagulation. At VTE

recurrence 19 (9.5%) patients were using vitamin K antagonists and 36 (10.5%) patients were using LMWH. The relative risk of recurrent VTE according to anticoagulant strategy remained not significantly [RR=1.04 (95% CI, 0.626-1.758; p= 1.000]. Levels of VKA as measured by the INR were subtherapeutic in 35.6% of patients at the time of recurrence. With respect to VTE recurrence site, 40 of 78 (51%) of patients developed a recurrent VTE in the same site as the index event. Details of VTE recurrence site are depicted in Table 7.

External precipitants, such as major surgery or use of chemotherapy or hormone therapy in the 3 months prior to the index VTE did not seem to contribute to recurrence risk. Index VTE occurred after major surgery in 121 patients and despite appropriate anticoagulation, VTE recurred in 17 (13.1%) patients in this subgroup. For the 422 patients who did not undergo surgery, 61 (14.7%) had a recurrent VTE (RR= 0.97 (95%CI= 0.587 – 1.574) *p*= 1.000). When we evaluated VTE recurrence risk in the surgery group, according to anticoagulation strategy there was no statistically significant difference between the use of VKA or LMWH [VKA: 8 of 48 (16.7%) patients; LMWH: 9 of 73 (12.4%) patients; RR= 1.35 (95%CI 0.568 – 3.191) (*p*= 0.595)].

Figure 5. Kaplan-Meier Survival estimate for VTE recurrence during the anticoagulation period

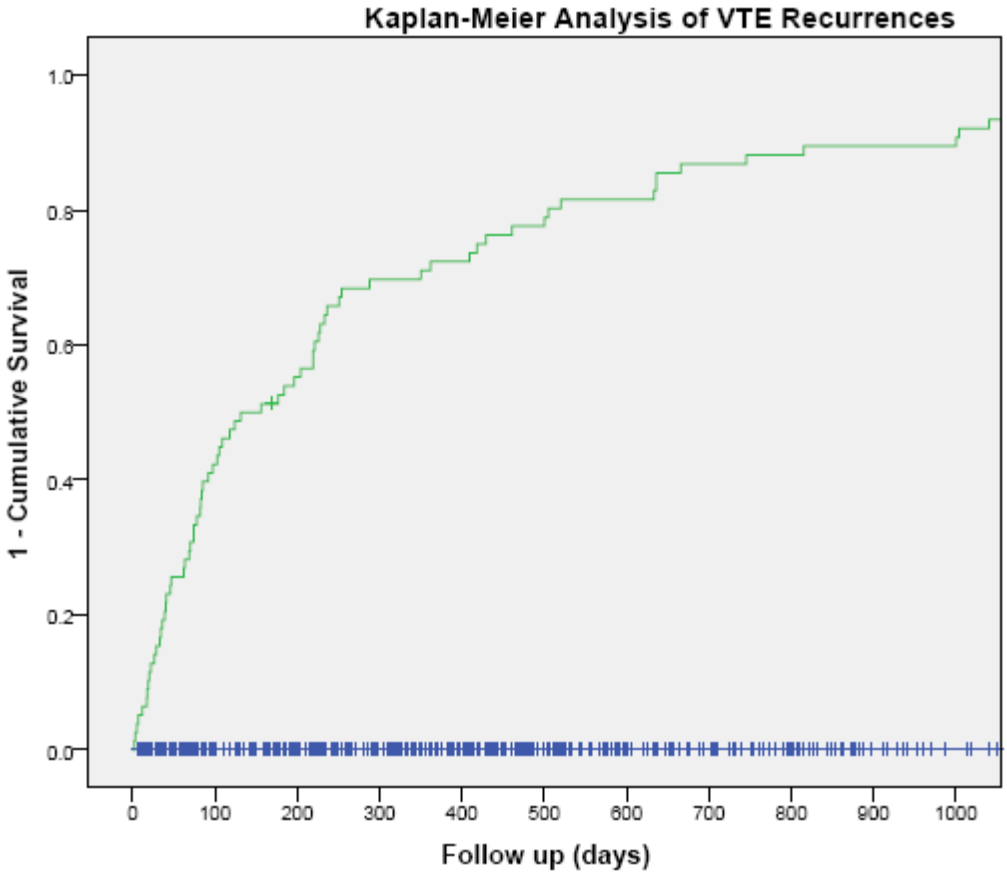


Table 7. VTE recurrence site

VTE recurrence site N (%)					
Index VTE Site	**Arm**	**Leg**	**PE**	**DVT+PE**	**Unusual Site**
Arm (n= 20)	14 (70.0)	3 (15.0)	3 (15.0)	0 (0.0)	0 (0.0)
Leg (n= 29)	1 (0.35)	19 (65.5)	8(24.1)	2 (6.9)	0 (0.0)
PE (n= 18)	1 (0.56)	9 (50.0)	6 (33.3)	2 (11.1)	0 (0.0)
Leg DVT+ PE (n= 9)	3 (33.3)	6 (66.7)	0 (0.0)	0 (0.0)	0 (0.0)
Unusual site (n= 1)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (100)

6.4.2.3 Tumour Characteristics

6.4.2 .3.1 Primary Tumour Site

Patients with lung cancer were the ones who most frequently developed a recurrent VTE [25 of 96 (26.1%)]. For GI and haematological malignancies, 12.1% of patients recurred (17 of 140 for GI and 7 of 58 for haematological). Only 7.0% (6 of 85) of patients with breast cancer had a recurrent VTE. In the miscellaneous subgroup, 23 of 164 patients recurred (14.0%). Among those, the most common tumour site was brain with 5 of 27 (18.5%) patients developing a recurrence, followed by gynaecological with 7 of 50 (14.0%) and genito-urinary with 6 of 47 (12.8%) recurrences. All other tumour sites in this subgroup had 1 case of recurrent VTE. Although VTE recurrence rate was lower in patients with breast cancer, females presented with a slightly higher recurrence rate than males [females: 49 of 303 (16.2%); males: 29 of 240 (12.1%); p= 0.217].

6.4.2.3.2 Presence of Distant Metastasis

Excluding haematological malignancies and brain tumours that cannot be classified according to the TNM staging system, the incidence of VTE recurrence was significantly lower in patients with stage I disease [2 of 61 (3.3%)] compared to stages II, II and IV [stage II=10 of 74 (13.5%); stage III=12 of 84 (14.3%); stage IV= 42 of 237 (9.6%)] (p= 0.004).

6.4.2.3.3 Histology

Among the 489 patients with solid tumours 71 had a recurrent VTE. Among those, there was no difference in recurrence rate with respect to tumour histology: 40 of 306 (13.1%) patients with adenocarcinoma and 31of 183 (16.9%) with non-adenocarcinoma tumours recurred (*p*= 0.2301).

6.5 Univariate Analysis

For the univariate and logistic regression analysis we evaluated only the 55 patients who had a VTE recurrence within the first 6 months of anticoagulation. The results of the univariate analysis are shown in Table 8. *P*-values less or equal to 0.25 were considered statistically significant for this analysis to select potentially relevant predictor variables for the multivariate analysis. Age as a continuous variable and age dichotomized into > or >/= 65 years old, gender, presence of lung cancer, breast cancer, histology, stage I disease, history of previous VTE and surgery presented as potentially relevant predictors.

Table 8. Univariate Analysis

Variables	Odds Ratio	95%CI	P-value
Female gender	**1.663**	**0.919 - 3.009**	**0.0929**
Age	0.840	0. 477 - 1.479	0.5465
Tumour Histology	1.179	0.646 - 2.152	0.5913
Primary Tumour Site			
***Haematological* ***	1.353	0.476 - 3.850	0.5707
Lung cancer*	**2.524**	**1.124 - 5.664**	**0.0248**
Breast cancer*	0.733	0.246 - 3.850	0.5776
Miscellaneous*	1.181	0.524 - 2.662	0.6890
Tumour Stage[§]	**0.135**	**0.018 – 0.994**	**0.0493**
Previous VTE	**1.719**	**0.728 - 4.056**	**0.2166**
Chemo/hormone therapy	0.859	0.490 - 1.508	0.5967
Surgery	**0.572**	**0.262 – 1.247**	**0.1603**

*Reference: GI malignancy; [§] TNM stage I versus stage II, III and IV

6.5.1 Inter-Observer Reliability of Significant Univariate Predictors

Fifty patients were randomly selected for physicians assessment performed by myself and a second observer (VD). The final Kappa was 0.857 (95% CI= 0.771-0.942) suggesting an excellent reproducibility of historical data. Details in table 9.

Table 9. Inter-observer reliability of significant univariate predictors

Variable	Kappa	95% CI*
Primary Tumour Site	0.904	0.774 – 1.034
Tumour Stage	0.758	0.558 – 0.958
Tumour Histology	0.953	0.861 – 1.044
Previous VTE[£]	1.000	1.000 – 1.000

***** 95% Confidence interval**; £** History of VTE before the event the occurred in the context of cancer

6.6 Logistic Regression

There were 5 variables with a *p*-value equal or lower than 0.25 in the univariate analysis that were tested in the multivariate model (Table 10). The variables were gender, primary tumour site, stage, history of previous VTE and surgery. We had a total of 55 VTE recurrences. This gave us 11 patients per predictor, a number more than adequate for the multivariate analysis. Although age was not selected in the univariate analysis, we also tested it in the Logistic Regression Model. We evaluated age both as a continuous variable and as a categorical variable (age < or = 65 years and age > 65 years) in separate models. This age cut-off value has been compared in previous studies suggesting that younger patients with cancer and VTE have a higher risk for VTE recurrence(21;58) . The multivariate model suggested that gender, primary tumour site, tumour stage and history of previous VTE were statistically

significant ($p<0.05$) (Table 10). Then, I applied the backward selection to evaluate the best model and it considered that tumour stage, history of previous VTE and surgery should be removed from the model, leaving only gender and primary tumour site as significant variables.

Table 10. Logistic regression for evaluation of VTE recurrence risk according to significant univariate predictors

Variables	Odds Ratio	95%CI	*P*-value
Gender	**2.077**	**1.106 - 3.901**	**0.0230**
Primary Tumour Site*			
Haematological	1.037	0.372 – 3.187	0.9472
Lung	**2.245**	**0.968 - 5.208**	**0.0595**
Breast	0.528	0.168 – 1.662	0.2747
Other	1.140	0.496 - 2.623	0.7573
TNM Stage I [§]	**0.143**	**0.019 - 1.084**	**0.0598**
Previous VTE	**2.091**	**0.842 - 5.193**	**0.1121**
Surgery	0.642	0.282 – 1.463	0.2919

*Reference: GI malignancy; [§] TNM stage I versus stage II, III and IV

6.6.1 Testing for Confounding and Interaction

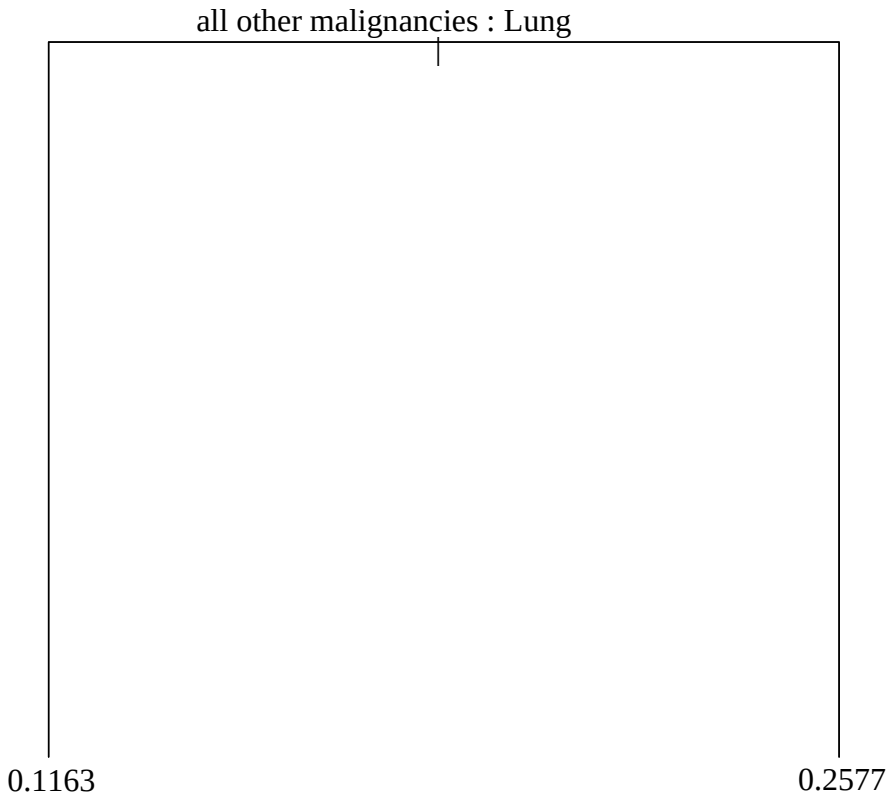
There is evidence that in patients without cancer who sustain a venous thromboembolic event, age is an important risk factor for recurrence, increasing its risk (67;69). Our assumption was that age could act as a confounding factor in the cancer setting because the incidence of several types of cancer (e.g.; colon, lung, prostate) tends to be higher in the elderly. However, in this study age was not statistically significant in the multivariate analysis, both as a continuous variable and as a categorical variable. Therefore, we did not need to test it as a confounding factor since it would not be included in the feasibility model. Nevertheless, we did test gender as a confounding factor because females presented with a higher rate of

recurrent VTE and this was not related to breast cancer, as one could expect. However, the adjustment for this potential confounding did not affect the magnitude of the coefficient estimate for our predictors of interest: tumour site, stage and previous history of VTE. We also tested the interaction between gender and tumour site, stage and previous history of VTE; as well as a interaction between stage and primary tumour site. None of the interaction terms were statistically significant; therefore they were removed from the model.

6.6.2 Recursive Partitioning

We entered our dataset, including the outcome variable (VTE recurrence or no VTE recurrence) and the 4 independent variables identified in the logistic regression (gender, primary tumour site, tumour stage and previous history of VTE) in the recursive partitioning process. The only predictor variable that had a chi-square high enough to be considered for splitting was primary tumour site (Figure 5). The decision tree demonstrated that 25.8% of patients with lung cancer had recurrent VTE versus 11.6% of patients with any other malignancy (p= 0.003) and that this was the only significant independent variable to predict VTE recurrence risk. We did not adjust the data to force the formation of other nodes in the decision tree.

Figure 4. Results of Recursive Partitioning



6.6.3 Goodness of Fit of Candidate Models

Although only gender and primary tumour site were considered significant predictors in the logistic regression analysis and /or in the decision tree analysis, we decided to evaluate other independent variables of primary interest in this study, considered to be important significant potential predictors. Therefore, an evaluation of the best model fit was applied comparing 4 different models:

Model 1: gender and tumour site (assigned by the selection procedure)

Model 2: gender, tumour site and stage

Model 3: gender, tumour site and previous VTE

Model 4: gender, tumour site, stage and previous VTE

A model fits when the summary measures of the distance between the observed and the estimated values are small, as well as when the contribution of each pair of observed and predicted values to these summary measures is random and small, relative to the error structure of the model. We performed the likelihood ratio test and the Hosmer-Lemeshow test:

A) Likelihood Ratio

The likelihood ratio test is a statistical test for making a decision between two hypotheses based on the value of the ratio between them and it requires that the number of observations in the model is the same. The comparison between model 1 with models 2, 3 and 4 confirmed that at least the addition of the variable "tumour stage" significantly improved the model fit (**1x2:** p= 0.0149; **1x3:** p=0.2637; **1x4:** p=0.0189).

B) Hosmer-Lemeshow test (H-S)

The H-S test is not sensitive to a number of misspecifications such as missing values or type II error and consequently lacks power in these situations. It is a crude way to screen for fitting and should not be used as a definitive fitting test (76). We did not

have missing values in our data. When the Hosmer-Lemeshow test was applied, there was no difference regarding the model fit for any of the 4 models (Model 1: *p*= 0.9740; Model 2= 0.95676; Model 3: *p*= 0.4598; Model 4: p= 0.4811). These results suggest that the *p*-value was not significant in either model, meaning that in all cases there was no rejection of the test's null hypothesis (there is the same frequency of observed and expected outcome in the various groups tested: groups of 10) which implies a good fit.

6.7 Choosing the Final Model

The goodness of fit test and the Hosmer-Lemeshow test suggested that models that included the variables gender, tumour site, tumour stage and previous VTE in various combinations had a good fit; therefore we evaluated the classification performance of the 4 models attempting to find the safest and the most clinically relevant model. Details are presented in Tables 11 and 12. Within the 4 models I tested different sensitivity and specificity values within the various models. I evaluated the probable 3 best models (model 1, 2 and 3) with a sensitivity level of 100% and 98%. The lower level of sensitivity allows an improvement in specificity and in the negative likelihood ratio, but at the expense of a decrease in the sensitivity itself and in the negative predictive value (NPV) (Table 12). Details of the 4 models are also available in Appendix 3. After reviewing the candidate models myself and with my supervisors, we concluded that models 2, 3 and 4 are extremely similar. However, to avoid the

exclusion of any significant variable the consensus was that model 4 had the best classification performance with the highest total recurrence rate (19.7%), 98.1% negative predictive value, 48.1% low risk excluded proportion and a good negative likelihood ratio of 15.5%; therefore, it was selected as the final model. In this model, the score sum ranges between -3 and +3 score points. Patients with a score equal or less than 0 have low risk (< or = 4.5%) for VTE recurrence. Patients with a score equal or above 1 have high risk (> 19%) for VTE recurrence (Tables 13, 14and 15).

Table 11. Steps in model building using logistic regression with predictors entered in order of strength of association

Model	N	DF*	Deviance (-2 log L)	Δ Deviance χ 2, P>0.005
Model 1 (Gender and tumour site)	543	5	427.056	19.8634>0.00013
Model 2 (Gender, tumour site and stage)	543	7	419.605	27.3147>0.003
Model 3 (Gender, tumour site and previous VTE)	543	6	424.014	22.9049>0.0008
Model 4	543	8	415.071	31.8480<0.001

(Gender, tumour site, stage previous VTE)				

* DF=degrees of freedom

Table 12. Classification performance of the 4 models

	Model 1	**Model 2**	Model 3	**Model 4**
# variables	2	**3**	3	**4**
Sensitivity	1.000	**1.000**	1.000	**1.000**
1 - Specificity	0.997	**0.946**	0.997	**0.912**
True negative proportion	0.3%	**5.4%**	0.3%	**8.8%**
NPV	100%	**100%**	100%	**100%**
% recVTE (low risk group)	5.6	**4.3**	4.1	**4.5%**
% recVTE (high risk group)	17.5	**19.1**	17.1	**19.7%**
Low Risk Excluded Proportion	46.8%	**48.9%**	43.3%	**48.1%**
Positive Likelihood ratio	1.00	**1.06**	1.00	**1.10**
Negative Likelihood ratio	0	**0**	0	**15.5%**

* NPV= negative predictive value

Table 13. Final Model with Regression coefficients and odds ratios of selected variables and assigned score points

		Regression coefficient	OR	95%CI lower	95%CI upper	**Model points**
Gender	**Female**	0.59	1.82	1.069	3.095	**1**
Primary Tumour Site	**Lung**	0.94	2.55	1.273	5.108	**1**
	Breast	- 0.76	0.46	0.167	1.268	**-1**
Malignancy Stage	**Stage I**	- 1.74	0.75	1.312	24.679	**-2**
History of previous VTE	**Yes**	0.40	2.42	1.114	5.264	**1**

Table 14. Evaluation of the Clinical Probability of VTE Recurrence using the final model (gender, primary tumour site, stage and history of previous VTE)

Sum of points according to patien malignancy's characteristics	**Patients (n)**	**VTE recurrence (n)**	**Frequency of VTE recurrence**
-3	**0**	**0**	**0**
-2	**33**	**0**	**0**
-1	**24**	**1**	**4.2 %**
0	**215**	**10**	**4.7 %**
1	218	34	15.6 %
2	49	9	18.4 %
3	4	1	25.0 %
Clinical Probability			
Low (< or = 0) **High (> or = 1)**			**-3 to 0** **1 to 3**

Table 15. The Final Clinical Prediction Rule

Predictor	Points
Female	1
Lung cancer	1
Breast Cancer	-1
TNM Stage I	-2
Previous VTE	1
Clinical Probability **Low (< or = 0)** **High (> or = 1)**	 **-3 to 0** **1 to 3**

6.8 Secondary Outcomes

The rates of major and minor bleeding as well as mortality were evaluated as secondary outcomes. We were not able to pull enough data for evaluation of D-dimer test results at index event and at recurrence, since less than 12% (63 of 543) of patients had a D-Dimer test performed during their follow up at the Thrombosis unit.

6.8.1 Bleeding events

Details of bleeding events are depicted in Table 15. Overall, bleeding complications occurred in 77 (12.0 %) of 543 patients. Major bleeding events occurred in 37 (6.8%) patients and minor bleeding in 40 (5.2%) patients. Different anticoagulation strategies did not impact on the frequency of major bleeding [LMWH= 23 of 344 (6.7%) and

VKA= 14 of 200 (7.0%)] with a non-significant relative risk of 1.05 (95% CI= 0.552, 1.988; *p*=0.8803). Among patients who developed major bleeding, only 1 had a concomitant VTE recurrence.

6.8.2 Mortality

Forty-seven (8.6%) patients died during the anticoagulation period. Twenty-nine (62%) died due to malignancy progression. The second most common cause of death was sepsis (28%). VTE and major bleeding were the cause of death in 4% and 6% of patients, respectively. Details of cause of death in Table 16.

Table16. Bleeding Events and All Cause Mortality

		VTE Recurrence		
Outcomes		**NO [n (%)]**	**YES [n (%)]**	**Total [n (%)]**
Bleeding Events	**Major**	36 (7)	1 (1)	37 (6.8)
	Minor	38 (8)	3 (34)	40 (5.2)
Death		45 (10)	2 (2)	47 (8.6)
Cause of Death	**Malignancy**	28 (62)	1 (50)	29 (62.0)
	VTE	1(2)	1 (50)	2 (4.0)
	Major Bleeding	3 (7)	0	3 (6.0)
	Sepsis	13 (29)	0	13 (28.0)

6.9 Discussion of the Retrospective Study

This retrospective study sought to determine the clinical predictors that are independently associated with risk of recurrent VTE in patients with malignancy-associated venous thrombosis during active anticoagulation for derivation of a clinical prediction rule that stratifies patients into high or low risk of recurrent VTE. Four possible independent predictors were identified: gender, primary tumour site, tumour stage and previous history of VTE; with female gender, lung cancer and history of previous VTE posing an increased risk for VTE recurrence; and stage I malignancy and breast cancer posing low risk. The final model stratifies patients with cancer-associated thrombosis into low or high risk of recurrent VTE. It includes easy to obtain malignancy and patient's characteristics; and it is very easy to calculate. Patients scoring zero or less had a low risk for VTE recurrence and patients scoring 1 or more had a high risk.

The independent predictors found in this retrospective study are in keeping with the *post-hoc* analysis of the CLOT trial recently presented at the 2009 ISTH meeting (73) and with the results of the systematic review we performed as part of this thesis project. In these 2 studies, lung cancer and the presence of distant metastasis conferred a high risk of recurrence (6;21;58). In the Lee study, unknown primary tumour site was also an independent predictor for high risk of VTE recurrence; and in the

systematic review, GI and GU malignancies also seemed to promote a greater risk. In addition, these studies suggest that each 10 years increase in age and the presence of breast cancer seemed to confer a low risk for recurrent VTE (6;10;21;58).

In the retrospective study, unknown primary tumour site and GU malignancies were pooled in 1 single group and their overall risk of VTE recurrence was not significant. Although age was not a significant predictor in this study population, increase in age showed a trend towards a low risk for VTE recurrence [OR= 0.788 (CI =0.471 - 1.319; *p*= 0.3643)]. The suggestion that younger age confers a greater risk for recurrence in patients with cancer-associated venous thrombosis is striking. There is a fair amount of evidence suggesting that in the non-cancer population with unprovoked VTE, age above 65 is an important risk factor for recurrence (23;70). The explanation for this diversity in VTE recurrence risk may be centered in the fact that the mortality rate (VTE - related and cancer treatment-related) may be higher in older patients. I hypothesize that older patients with cancer frequently receive only palliative anticancer treatment, are not clinically fit and tend to have a higher mortality rate. Therefore, older patients with cancer would be less predisposed to a first VTE or a recurrent VTE. Another hypothesis is that patients with inherited thrombophilias such as Factor V Leiden and Prothrombin gene mutation (G20210A) tend to present with recurrent VTE at earlier age. Perhaps, younger patients with cancer who recur also carry a thrombophilia; however this was not evaluated in this study.

As I expected, the majority of patients treated for VTE between 2002 and 2004 received VKA, whereas patients treated between 2007 and 2008 received LMWH. Interestingly, the overall VTE recurrence risk according to anticoagulant strategy was similar with both regimens [RR= 1.13 (95%CI, 0.736-1.716; p=0.609)] even when I analysed only patients who had a recurrent VTE within the first 6 months of anticoagulation [RR=1.04 (95% CI, 0.626-1.758; p= 1.000]. These results are different from previously published trials that suggested that LMWH is the best treatment strategy with up to 53% absolute risk reduction in VTE recurrence rate when compared to VKA. The study by Lee *et al*. is the largest published RCT that compared the efficacy of VKA or LMWH in patients with malignancy-associated thrombosis and it provides the strongest evidence that LMWH is the best treatment approach for patients with cancer-associated venous thrombosis (6). However, it is necessary to emphasize that in this trial 40 % of patients in the VKA group who presented with a recurrent VTE had a sub-therapeutic INR at the time of recurrence, suggesting that VKA management was suboptimal. In my thesis study patients presented with a similar rate of subtherapeutic INRs of 35.6%, but we were unable to collect INR data from all patients who used VKA and developed a VTE recurrence (22 of 30 (73.4%). Most likely we were underpowered to assess VTE recurrence risk according to anticoagulation strategy. Roughly, I would have needed a sample size of 772 patients with a 1:1 ratio between VKA and LMWH to have 80% power to

demonstrate a difference between treatment strategies and VTE recurrence risk similar to the available literature (9).

The results of my secondary outcomes were overall in keeping with the literature. A systematic review we recently published demonstrated major bleeding rates around 7.0% for patients with cancer-associated thrombosis treated with either VKA or LMWH (9). In the retrospective study we found the same rate. Moreover, 3 of 36 (8%) of the major bleeding events led to death, a much higher rate than the 3 to 4% observed in previous studies that evaluated efficacy and safety of different anticoagulant strategies in non-cancer patients with VTE (60-63).I collected information about bleeding events by grouping medical reports together with blood tests results, radiology tests and records of patient's admission to the emergency department of the Ottawa Hospital. Therefore, I believe that precision regarding this information is reliable.

In this study patients were censored when: they developed a recurrent VTE or; when anticoagulant was discontinued or; when they were last seen by the Thrombosis unit or; when they died. Close to 9% of patients died during the follow-up period. This rate is much lower than the published literature that reports a 40% mortality rate for patients with cancer-associated thrombosis on anticoagulation (9). Overall mortality was not accurately ascertained in this study because, on many occasions, the last

medical report concerning the patient stated that he/she was being sent to palliative care due to progressive, untreatable malignancy. In case death had occurred, there was no registration of its date or cause in the electronic or hardcopy chart of those patients. Furthermore, the patients studied were derived from our outpatient clinic. Those patients who were admitted and not discharged are likely to have higher mortality rates secondary to recurrent VTE. Therefore, mortality rate may have been underestimated. Finally, the unexpected finding that patients with cancer-associated thrombosis who undergo surgery have a similar rate of VTE recurrence to patients with cancer and thrombosis who do not undergo surgery is striking and deserves further evaluation due to the well recognised association between the temporary risk factor surgery and VTE. In surgical patients with VTE the risk of recurrent VTE is negligible (1%) after a short course of anticoagulants (7;16;22).

6.10 Limitations and Strengths of the Retrospective Study

One advantage of a retrospective study is its ability to analyse data obtained from a relatively large number of subjects with a comparatively small investment of time and at a modest cost. However, a retrospective study is hypothesis generating, which *per se* limited our ability to derive a clinical prediction rule. Ideally, a CPR should be derived in a prospective study with blinded outcome assessment, which was not possible with a retrospective design. However, our goal was to assess the feasibility of derivation of a rule by identifying possible independent predictors of VTE recurrence

in patients with malignancy-associated VTE. In this manner, I was able to identify 4 independent predictors (gender, primary tumour site, previous VTE and tumour stage). Among them, the backward selection in the logistic regression analysis pointed only gender and tumour site (lung cancer) as being important predictors. In addition, the recursive partitioning only selected lung cancer as a significant variable influencing on VTE recurrence risk. Regardless, I decided to go on and evaluate alternative models that included not only the variables selected by the logistic regression and recursive partitioning analyses, but also variables I considered of primary interest for the CPR. As such, I included breast cancer, history of previous VTE and tumour stage along with gender and lung cancer for evaluation of various competing models. Although haematological malignancy also seemed to confer low risk for VTE recurrence, I decided not to include this variable due to the small number of patients with this type of malignancy in the study and the consequent wide 95% confidence intervals in the logistic regression's odds ratio for this variable. I also decided to exclude age because of its possible feature as a confounding factor. The relevance of age in the CPR should most likely be re-evaluated in a future prospective study. I believe that this model has face validity, since it does include variables considered relevant for evaluation of VTE recurrence risk in patients with malignancy-associated thrombosis; however I cannot ascertain content validity because the design in which the clinical prediction rule was derived was not ideal and a larger number of prospectively enrolled patients with cancer-associated thrombosis

will be needed for a more appropriate analysis. Even so, the results of the retrospective study adds to the results of the CLOT trial *post-hoc* analysis (73) and this thesis systematic review results, and suggest that patients with cancer-associated thrombosis do indeed have varying VTE recurrence risk influenced by malignancy characteristics (primary tumour site, tumour stage) and patients' characteristics (gender, history of previous VTE).

Reporting bias cannot be excluded from the analysis since selection of patients could not be random, consecutive and concealed with respect to treatment allocation but the fact that the vast majority (if not all) of patients with VTE at The Ottawa Hospital are referred to our clinic makes reporting bias less likely with respect to ascertainment of VTE recurrence. However, the presence of reporting bias is likely responsible for the discrepant results we found regarding mortality rate in the study population compared to the literature results, since we were unable to collect precise data regarding occurrence of death. One important aspect is that patients who used VKA took a longer time to develop a VTE recurrence. Their overall follow-up time was twice as long as patients who used LMWH. However, only (10 of 78)12.8% of patients developed VTE recurrence after 1 year of anticoagulation. In fact, 70.5 % of patients recurred within the first 6 months of anticoagulation. Although, I evaluated all VTE recurrences, in the CPR I included only the patients who recurred within the first 6 months of anticoagulation. Therefore, it is unlikely that the longer follow-up time of patients on VKA would have severely biased my results.

I was unable to collect detailed data regarding type of anti-cancer treatment patients used such as type of chemotherapy regimen and number of cycles. Therefore, these parameters were not tested in the model. Moreover, the evaluation of D-Dimer test both at the time of index VTE and at recurrence was rarely requested by the attending physician. For this reason, we could not evaluate a possible implication of this test as an adjunct tool for prediction of VTE recurrence. Immobilization is another external VTE precipitant that we did not evaluate. Immobilization is a soft outcome and we included only outpatients in this study. Although some of them may have been "immobile", in home care, it would not be feasible to accurately ascertain this parameter retrospectively.

7.0 FUTURE DIRECTIONS

The CPR presently derived should be evaluated in a future prospective study, so that its reproducibility, generalizibilty, validation and/ or improvement can be assessed. Once this is accomplished, a RCT should be conducted for further external validation of the CPR and for evaluation of the best treatment strategy for patients stratified as low or high risk for recurrent VTE. For low risk patients I should compare standard treatment (LMWH) with VKA for 6 months. For high risk patients I will need to consider a more intensive anticoagulant approach (e.g. 120% dose of LMWH) compared to standard dose LMWH.

8.0 CONCLUSION

In conclusion, the systematic review conducted as part of this thesis admitted potential predictors of recurrent VTE in patients with cancer-associated thrombosis. The survey confirmed that Thrombosis specialists in North America and Europe agree that patients with cancer-associated venous thrombosis appear to have varying risk for VTE recurrence; therefore, a better way to stratify risk and tailor treatment for patients with cancer-associated-venous thrombosis is very relevant. A clinical prediction rule that stratifies patients' risk of VTE recurrence will therefore be very welcome by thrombosis specialists.

In the retrospective study, I was able to derive a simple and easy to calculate preliminary clinical prediction rule that stratifies patients with cancer-associated thrombosis into low or high risk of recurrent VTE. The first step for a probable important improvement in the care of these patients has been made. Hopefully, I will be able to demonstrate in the near future, that this rule is reproducible, generalizable, safe and a useful tool for clinicians to help them improve care for patients with cancer-associated venous thromboembolism.

Appendix 1- Survey letters according to the Dillman's approach

Pre-notification letter

Dear colleagues

As you know, at the Ottawa Hospital/University of Ottawa, we run a research intensive Thrombosis Program. An increasing number of our patients have cancer – associated venous thrombosis and we all appreciate how difficult it can be to manage these patients.

We seek to develop a clinical prediction rule that will predict those at low risk of venous thromboembolism (VTE) recurrence during the anticoagulation period in the hope that these patients can be treated less expensively with long term vitamin K antagonists rather than low molecular weight heparin. The primary rule will be derived retrospectively from our database of patients with cancer-associated VTE followed at the Thrombosis Unit in the past six years.

We would appreciate if you, as a physician with interest and expertise in thrombosis would participate in a 5-question survey that will ultimately disclose how relevant you think this clinical prediction rule might be and what would be the best study design to prospectively validate such a rule. Please note, you will be assisting Dr Martha Louzada with her master's thesis when you complete this simple questionnaire.

To ensure your anonymity we have no way to know if you completed the survey and we will therefore, have to remind everyone on the e-mail out about the survey at a 1-week interval for 4 weeks. If you let us know you have completed the survey, we will not have to send you a reminder. We sincerely appreciate your assistance with this small project.

We will send you the electronic survey on March 30th, 2009.

Thank you, in advance, for your time.

Cover letter

Dear colleagues

We hope you had a chance to read the pre-notification letter we have sent you last week. Attached is the link to the survey which will enable us to understand your expectations, as a thrombosis expert, of a clinical prediction rule that stratifies venous thromboembolism (VTE) recurrence risk in patients with cancer-associated thrombosis.

The motivation for the development of such a rule is that evidence exists that some cancer patients are low risk for recurrent VTE. A meta-analysis we recently published established that in patients with cancer-associated VTE , recurrent VTE rates were 7% when low-molecular-weight-heparin (LMWH) was used and 13% when vitamin K antagonists (VKA) was used. Bleeding risks were similar (Louzada,ML. Thrombosis Research, 2008; DOI 10.1016/j.thrombres.2008.09.002).

Many people now consider LMWH the standard of care for this population. However, some data suggest that certain cancer patients (e.g. those with secondary VTE, localised disease, certain tumour types) may have a low risk of VTE recurrence on treatment and could be treated with VKA. With a clinical prediction rule that predicts those at low risk of recurrence, these patients could be treated with the less expensive and less invasive usual strategy of 1 week of LMWH followed by 6 months of VKA.

We would be very grateful if you would click on the link below and take 3 minutes to complete our online survey. Please, send us the completed survey until April 6th, 2009.

Once again, if you let us know you have completed the survey, we will not have to send you the weekly reminders. The first reminder will be sent out on April 7th, 2009.

We appreciate your time and attention to this!

Thank you note to participants who responded

Dear colleagues

Thank you very much for completing the survey "Evaluating VTE recurrence risk during the anticoagulation period in patients with cancer –associated thrombosis".

Your expert opinion is of great value to us.

Kind regards,

D7: Thank you note to participants who responded and reminder 1

Dear colleagues

Thank you very much for participating in the survey "Evaluating VTE recurrence risk during the anticoagulation period in patients with cancer –associated thrombosis".

For those who did not have a chance to complete it yet, this is our first reminder.

Please, click on the link below and take 3 minutes to complete this 1-page, 5-question survey and help us define the best research strategy to develop a clinical prediction rule that stratifies patients with cancer-associated VTE into low risk of VTE recurrence.

If you no longer wish to receive our weekly reminders, simply let us know that you have completed the survey or that you do not wish to participate on it. Please, complete the survey until April 13th. The second reminder will be sent out on April 14th, 2009.

http://www.surveymonkey.com/s.aspx?sm=nPM4nFIFLf3RbW435eYfOQ_3d_3d

Kind regards,

D14: second reminder

Dear colleagues

We hope you had a chance to read the pre-notification letter we have sent you 2 weeks ago. Attached is the link to the survey which will enable us to understand your expectations, as a thrombosis expert, of a clinical prediction rule that stratifies venous thromboembolism (VTE) recurrence risk in patients with cancer-associated thrombosis.

The motivation for the development of such a rule is that evidence exists that some cancer patients are low risk for recurrent VTE. A meta-analysis we recently published established that in patients with cancer-associated VTE , recurrent VTE rates were 7% when low-molecular-weight-heparin (LMWH) was used and 13% when vitamin K antagonists (VKA) was used. Bleeding risks were similar (Louzada,ML. Thrombosis Research, 2008; DOI 10.1016/j.thrombres.2008.09.002).

Many people now consider LMWH the standard of care for this population. However, some data suggest that certain cancer patients (e.g. those with secondary VTE, localised disease, certain tumour types) may have a low risk of VTE recurrence on treatment and could be treated with VKA. With a clinical prediction rule that predicts those at low risk of recurrence, these patients could be treated with the less

expensive and less invasive usual strategy of 1 week of LMWH followed by 6 months of VKA.

We would be very grateful if you would click on the link below and take 3 minutes to complete this 1-page, 5- question online survey. Please, send us the completed survey until April 20th, 2009. Once again, if you let us know you have completed the survey, we will not have to send you our last reminder on April 21st. http://www.surveymonkey.com/s.aspx?sm=nPM4nFIFLf3RbW435eYfOQ_3d_3d
We appreciate your time and attention.

Kind regards,

D35: Last reminder - special plea

Dear colleagues
Thank you very much for participating in the survey "Evaluating VTE recurrence risk during the anticoagulation period in patients with cancer –associated thrombosis".

For those who did not have a chance to complete it yet, this is our **last reminder**.
Please, click on the link below and **take 3 minutes to complete this 1-page, 5-question survey** and help us define the best research strategy to develop a clinical prediction rule that stratifies patients with cancer-associated VTE into low risk of VTE recurrence.
http://www.surveymonkey.com/s.aspx?sm=nPM4nFIFLf3RbW435eYfOQ_3d_3d

Kind regards,

Appendix 2 - Definition of Major and Minor bleeding according to the International Society of Thrombosis and Haemostasis

Major bleeding:

- Associated with a fall in hemoglobin of 2g/dL or more, or;
- Leading to a transfusion of 2 or more units of packed red blood cells or whole blood, or;
- Occurring in a critical site: intracranial, intraspinal, intraocular, pericardial, intra-articular, intramuscular with compartment syndrome, retroperitoneal; or contributing to death.

Minor bleeding:

- Any bleeding not fulfilling the criteria for major bleeding

Appendix 3. Two-by-two tables of competing Models

Model 1:gender + tumour site

Score points	Total Patients	VTE recurrence	Frequency(%)
-1	1	0	0
0	268	15	5.6
1	233	30	12.9
2	41	9	22.0

	No VTE Recurrence	VTE Recurrence
Low prob (cutoff < or =0)	254	15
High prob (cutoff > 0)	235	39

Low risk excluded proportion= 46.8
Low risk= 5.6%
High risk= 17.5%

Model 2: gender + tumour site + stage

Score points	Total Patients	VTE recurrence	Frequency(%)
-3	0	0	0
-2	41	1	2.4
-1	18	0	0
0	230	14	6.1
1	216	31	14.4
2	38	9	23.7

	No VTE Recurrence	VTE Recurrence
Low prob (cutoff < or = 0)	290	18
High prob (cutoff >0)	218	37

Low risk excluded proportion= 50.5%
Low risk= 4.3%
High risk= 19.1%

Model 3: gender + tumour site + previous VTE

Score Points	Total patients	VTE recurrence	Frequency
-1	1	0	0
0	244	10	4.1
1	240	35	14.6
2	53	9	17.0
3	5	1	20.0

	No VTE Recurrence	VTE Recurrence
Low prob (cutoff < or = 0)	235	10
High prob (cutoff >0)	254	44

Low risk excluded proportion= 43.3%
Low risk= 4.1%
High risk= 17.1%

Model 4: gender + tumour site + stage + previous VTE

Score points	Total patients	VTE Recurrence	Frequency
-3	0	0	0
-2	33	0	0
-1	24	1	4.2 %
0	215	10	4.7 %
1	218	34	15.6 %
2	49	9	18.4 %
3	4	1	25.0 %

	No VTE Recurrence	VTE Recurrence
Low prob (Cutoff <or = 0)	261	11
High prob (cutoff > 0)	227	44

Low risk excluded proportion= 48.1
Low risk= 4.7%
High risk= 19.7%%

9.0 REFERENCE LIST

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