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**Review**

**Biomarkers for the (cancer-related) risk of thrombosis in the veins: microRNAs**

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**Abstract**

Disease conditions often include dysregulation of microRNAs (miRNAs), which are short noncoding RNAs with gene regulatory activities. MiRNAs are attractive biomarkers for the diagnosis and prediction of cancer and cardiovascular disorders because they are reasonably stable, readily quantified, and accessible from plasma or other bodily fluids. The third most frequent form of cardiovascular illness, venous thromboembolism (VTE) is associated with a high rate of morbidity and death globally. More evidence is emerging to support the concept that microRNAs (miRNAs) have a role in controlling the pathophysiology of VTE and serving as VTE biomarkers. Cancer patients have a higher incidence of venous thromboembolism (VTE) than the general population. However, existing risk prediction models for cancer-associated thrombosis (CAT) perform suboptimally, and innovative biomarkers are thus urgently required to determine which patients may benefit the most from thromboprophylaxis. The pathophysiology of VTE will be reviewed, beginning with the mechanistic role played by miRNAs. Then, miRNAs’ potential as prognostic biomarkers for VTE in cancer-free people is discussed, and finally, CAT is the topic of extensive attention. Differential regulation of some of the CAT-associated miRNAs was also seen in VTE, providing more insight into CAT’s pathogenesis. Our findings suggest that future research should use sufficiently powered methods to identify the miRNA panel that most accurately predicts VTE and CAT. To determine if miRNAs, either on their own or as part of established risk models, have promise as VTE and CAT biomarkers, validation studies employing similar patient groups are needed.

**Keywords:** Cancer, microRNA, venous thromboembolism, biomarker, cancer-associated thrombosis.

1. **Introduction**

Deep vein thrombosis and pulmonary embolism together make up venous thromboembolism (VTE), which is the third most frequent form of cardiovascular illness globally with an annual incidence rate of 1-2 per 1000 people [1]. The quality of life is severely diminished by VTE, and it is also associated with an increase in mortality and morbidity. Post-thrombotic syndrome and persistent thromboembolic pulmonary hypertension are two of the most serious consequences of venous thromboembolism [2]. Risk factors for venous thromboembolism (VTE) include age, obesity, diabetes, fractures, trauma, non-O blood group, factor V Leiden mutation, thrombophilia, oral contraceptives, pregnancy, a history of thrombosis, and long-distance travel [2,3].

One of the biggest risk factors for VTE is cancer, with an average 9-fold greater chance of developing VTE in the first year following cancer diagnosis, however the precise risk is depending on patient-specific risk factors [4]. Of all VTE occurrences, around 20% to 30% are related with malignancy [5]. Major surgery, chemotherapy, tumor type, and cancer stage are all associated with a higher risk of venous thromboembolism (VTE) in cancer patients [5]. Additionally, the death rate is increased by a factor of 30 compared to disease-free participants and by a factor of 4 compared to patients...
with cancer alone [5]. Overexpression of prothrombotic factors (tissue factor [TF] or podoplanin [PDPN]) or somatic mutations (e.g. STK11 or KRAS) are other tumor-intrinsic traits that may contribute to the development of venous thromboembolism [6,7].

In order to determine which cancer patients might gain the most from thromboprophylaxis, many risk prediction models have been created. The Khorana score takes into account cancer kind, body mass index, and blood parameters including hemoglobin level, platelet count, and leukocyte count to determine a patient’s risk of developing cancer-associated thrombosis (CAT). The Khorana score was not as effective as expected in external validation investigations [8,9], despite its widespread clinical endorsement. While the Khorana score was developed to select high-risk ambulatory patients undergoing chemotherapy, many validation studies have used other inclusion criteria, such as chemo-naïve patients or a different distribution of tumor types, which may account for the subpar performance [8]. When compared to individuals with a low Khorana score, those with a high score only had a 1.6-fold greater chance of getting VTE, according to a recent comprehensive meta-analysis of 54 trials [10]. In order to more accurately identify those at high risk for developing CAT, we need a deeper knowledge of the underlying processes that contribute to the disease. Despite their potential, miRNAs have just lately begun to be explored as a unique class of biomarkers in health and illness.

2. miRNAs

microRNAs (miRNAs) are non-coding RNAs that are around 21–23 nucleotides in length and are single-stranded. Their seed region, a 6- to 8-nucleotide sequence that binds to messenger RNAs, regulates gene expression [11]. Figure 1 shows how RNA polymerase II converts miRNA genes into the primary miRNA (pri-miRNA) precursors described in reference [12]. Precursor miRNA (pre-miRNA) is formed when the Drosha-DGCR8 complex recognizes the double-stranded RNA [12]. The RNase III enzyme DICER then cleaves the pre-miRNAs in the cytoplasm [12]. This process requires exportin-5 to transport the pre-miRNAs from the nucleus to the cytoplasm. The miRNA strand may originate from either the 5′ or 3′ arm of the pre-miRNA, and the two strands join together via hydrogen bonding to produce a mature miRNA:miRNA duplex [12]. Although the 3′ arm typically generates few functional miRNAs, both the 5′ arm and the 3′ arm may be useful [12].

Each miRNA may interact with many mRNAs via complementary base-pairing, controlling gene expression at the posttranscriptional level, either by inhibition of translation or by mRNA destruction [3]. Intriguingly, the expression of numerous genes may be controlled by a single miRNA, and conversely, individual genes can be controlled by several miRNAs [3,13]. Furthermore, miRNAs seem to be appropriate biomarkers for predicting illness for a number of reasons. MiRNAs are readily isolated for study since they are released into the extracellular environment via several carriers [3]. Microvesicles (MVs) generated from circulating blood cells (platelets and leukocytes) contain miRNAs and, together with exosomes, make up the subpopulations of EVs [14]. MiRNAs may be detected in a wide variety of bodily fluids, not only blood (plasma and serum). Because of their resistance to nuclease digestion, miRNAs that are released from cells may be tested repeatedly [11]. This is especially true when they are enclosed in EVs. Given these features, miRNAs are emerging as a promising class of biomarkers for several illnesses, including cancer and cardiovascular conditions.

Hemostasis is only one of the many biological processes in which miRNAs play a role [3]. It has not yet been demonstrated in vivo [15,16] that dysregulation of miRNAs disrupts physiological processes and may disrupt the hemostatic balance, resulting in the development of VTE via multiple pathways. First, dysregulation of miRNAs directly influences gene expression via autocrine control and impairs the function of cells involved in hemostasis [15]. Second, circulating miRNAs in EVs released by donor cells elicit paracrine effects [15]. Various cell-specific mechanisms of how miRNAs alter hemostasis have been reported in tumor cells, platelets, neutrophils and endothelial cells, have been characterized. The known effects of miRNAs on variables involved in hemostasis and thrombosis are
outlined in Table 1. Factors II, V, VII, IX, X, and XII; protein S; and protein C are only some of the coagulation factors that are produced by hepatocytes in the liver [17]. Coagulation factor expression may be influenced by miRNAs [16,18,19,20, and 21] that are expressed by these cells or adopted from EVs.

Thrombotic illness is linked to abnormal regulation of TF, a transmembrane protein that is widely expressed and acts as the first step in the extrinsic coagulation cascade. Interestingly, numerous miRNAs have been demonstrated to control TF expression [(22), (23), (24), (25), (26), (27), (28), (29)]. Hemostasis and thrombosis are mostly primary processes, although miRNAs may also influence platelets and inflammatory proteins [(30), (31), (32), (33), (34), (35), (36), (37)]. miRNAs may also influence platelet activity, count, and aggregation [(38), (39), (40), (41)]. Platelets are often investigated in the context of arterial thrombosis, although their potential involvement in venous thrombosis has also been proposed [42]. At the opposite end of the range, miRNAs control genes involved in fibrinolysis, such as plasminogen activator inhibitor-1 (PAI-1), to aid in blood clot resolution [43].

The clinical trials that evaluated miRNAs as a biomarker for VTE are first discussed in this narrative review. Several research looked at miRNA plasma levels and their possible diagnostic or prognostic function for VTE. Many studies that identified miRNAs as diagnostic miRNAs may reflect changes in miRNA expression driven by thrombosis. Since it is more important to determine which miRNAs contribute to the development of thrombosis and serve as possible biomarkers for this objective, only studies that focused on predictive miRNAs are reviewed here. Second, we examine the significance of microRNAs (miRNAs) in predicting VTE in cancer patients and emphasize their involvement in this context.

Embase and PubMed were used to locate all included studies, and all of them focused on classical VTE (including deep vein thrombosis and/or PE). All inclusion and exclusion criteria for the systematic search are shown in a flowchart (Figure 2), and the specific search words are provided in the Additional files. On July 25, 2022, we found 612 hits for venous thromboembolism, of which 4 were deemed papers on predicting miRNAs in overall venous thromboembolism in a noncancer environment [20,44,45,46]. For CAT, we discovered 222 hits on July 25, 2022, of which 6 were regarded research on predictive miRNAs in CAT after a careful assessment of the title and abstract [(47), (48), (49), (50), (51), (52)]. Pathogenesis of VTE involves dysregulation of several cellular processes, including those involved in hemostasis. The expression of many pro- and anticoagulant factors involved in hemostasis, as well as platelet activation and aggregation, are all regulated by miRNAs, as outlined above and shown in Table 1. We looked at research evaluating miRNAs that have been shown to be predictive of either a first or subsequent VTE (Table 2) [3,16,53]. Two studies examined first-time VTE occurrences, while two examined VTE recurrence.

The miRNA profiles of 52 patients with VTE and 52 controls were analyzed in a research by Rodriguez-Rius et al. [20] that included 35 families from Spain. After a VTE episode, blood was taken for testing while the patient was on a break from anticoagulation. RT-qPCR was used to examine the miRNA profiles, and the results were then verified in-house. The following 4 miRNAs were elevated in patients with VTE after multiple correction testing: miR-126-3p, miR-885-5p, miR-194-5p, and miR-192-5p [20], of which 2 (miR-126-3p and miR-194-5p) have been previously found in a diagnostic VTE scenario [54]. The four miRNAs, together with age and sex, were used to create a risk model that outperformed previously established genetic risk scores [20], with an area under the receiver operating characteristic curve of 0.77. In a large population-based cohort, the Tromsø project, Starokiva et al. chose 20 first unprovoked VTE patients and age- and sex-matched healthy controls [44]. After the VTE episode, blood samples were taken to analyze miRNA expression using RT-qPCR and compare it to that of healthy individuals. Nine microRNAs were found to be upregulated or downregulated in VTE patients. However, no adjustment was made for repeated testing. We observed no conserved miRNAs between our analysis and those of Rodriguez-Rius et al. [20]. Possible causes include variations in sampling strategies and techniques for isolating miRNAs.
Wang et al. [45,55] conducted a nested case-control (NCC) investigation in 2019 to look for new predictive biomarkers for recurrent VTE, using participants drawn from a prospective population-based cohort (the Malmo Thrombophilia project). An unprovoked VTE occurred again within 2 weeks after stopping anticoagulant treatment, and this was considered a recurrence. In order to compare patients with and without recurrent VTE, plasma samples were collected after the first VTE incident while patients were on a treatment hold for anticoagulants. After accounting for multiple testing, logistic regression analysis revealed that 12 miRNAs were substantially linked to VTE recurrence probability.

The risk of VTE recurrence was evaluated using next-generation sequencing to profile plasma miRNAs in the biggest prospective trial to date, conducted by Thibord et al. [46]. Blood samples were collected after the first VTE incident, when the patients were on suspension from anticoagulant therapy, but before the recurrent VTE, from a preexisting large prospective cohort (MARseille THrombosis Association study). Multiple testing correction failed to reveal a statistically significant association between plasma levels of miR-370-3p, miR-27b-3p, and miR-222-3p with the risk of recurrent VTE. The last two, miR-27b-3p and miR-222-3p, interestingly corroborated the results of Wang et al. [45] in patients with recurrent VTE, indicating that these miRNAs play a similar function in the onset of both de novo and recurrent VTE. MiR-222-3p (upregulated) and miR-27b-3p (downregulated) were the only two miRNAs shared across the two recurrent VTE investigations.

MiR-103a-3p (downregulated), miR-27b-3p (downregulated), miR-320a (downregulated), miR-320b (downregulated), miR-191-5p (upregulated), and miR-222-3p (upregulated) were all identified as being associated with VTE across all 4 studies (Table 2, Figure 3). While several miRNAs were discovered, there was little agreement across investigations. Importantly, in the studies examining initial VTE (Starikova et al. [20] and Rodrigues-Riuz et al. [44]), blood was sampled after the occurrence of VTE in a case-control setting, raising the question of whether the miRNA profiles found are rather a consequence than causally involved in the development of VTE. After the first VTE episode but before recurrent VTE, Wang et al. [45] and Thibord et al. [46] utilized NCC and prospective designs, respectively, in which anticoagulant therapy was temporarily discontinued. Since large prospective cohort studies are so time-consuming, costly, and resource-intensive for biomarker-finding purposes, NCC designs or subsampling prospective cohorts are acceptable alternatives [56,57]. NCC designs are less expensive to implement and have a lower risk of bias, particularly for infrequent outcomes [56,57]. The question of whether miRNAs are a cause or a consequence can now be answered with certainty since blood was collected before the end occurred. MiR-222-3p and miR-27b-3p were among the overlapping miRNAs detected in both the Wang et al. [45] and Thibord et al. [46] research, respectively, indicating that these miRNAs may be potential targets for further mechanistic discovery studies or predictive biomarker possibilities.

The various miRNA profiles seen in the 4 studies may be due to confounding factors such as differences in blood sampling circumstances, the duration between the VTE episode and blood sample, and the makeup of the patient group. miRNA expression levels are also reliant on the experimental kits and processes used to identify and quantify miRNAs [20]. Only Wang et al. [45] utilized a different isolation kit (Exiqon miRCURY RNA Isolation Kit—Biofluids), whereas Starikova et al. [44], Rodrigues-Riuz et al. [20], and Thibord et al. [46] all used the Qiagen miRNeasy Mini Kit. Both RT-qPCR and NGS were employed for quantification in the aforementioned investigations, however NGS is more powerful in terms of discovery because of its scalability and capacity to identify novel sequences. Patient variables including comorbidities, race, ethnicity, genetics, and health care systems might be predicted to be distributed differently throughout the aforementioned studies due to their utilization of samples from distinct patient cohorts (Norway, Sweden, France, and Spain). Some of the discrepancies in findings between studies [3,58] may be due to sampling conditions (time of blood collection after VTE event and type of anticoagulant in which blood is drawn), small sample sizes contributing to low statistical power, and different statistical methods. One example may be the use of multiple correction testing, which was not undertaken in the research by Starikova et al. [44]. Furthermore, as indicated by Oto et al., accurate and repeatable internal miRNA normalizers are essential to limit the amount of discrepancies across investigations [59]. Using miRNAs as biomarkers in a diagnostic or clinical environment may be
more viable and attractive if the lack of consistency in laboratory techniques can be improved by reaching an agreement on which reference miRNAs to utilize. Overall, the use of miRNA plasma levels as predictors of VTE in clinical practice has to be further developed via external validation using bigger cohorts to explain some of the contradictions.

4. CAT microRNA profiling

Only three papers and three abstracts have discussed miRNAs’ potential as prognostic biomarkers for CAT so far (Table 3). Oto et al. [49] reported in 2020 on a prospective cohort analysis of 121 patients with PDAC or distal extrahepatic cholangiocarcinoma (DECC), of which 10 (8.3%) developed CAT. Blood was collected at the time of cancer diagnosis, before to the onset of VTE, from 5 patients with CAT who were chosen from this cohort based on the quality of their RNA and 5 patients with cancer but no CAT in an NCC design. Eleven microRNAs (miRNAs) were shown to be potential predictors of CAT using RT-qPCR and an elastic-net logistic regression model [49]. Seven of the previously discovered miRNAs (miR-486-5p, miR-106b-5p, miR-let-7i-5p, miR-let-7g-5p, miR-144-3p, miR-19a-3p, and miR-103a-3p) exhibited a substantial correlation with CAT in a validation stage including 10 CAT patients and 22 non-CAT patients [54]. At the time of cancer diagnosis, this model had an AUC (area under the curve) of 0.95 (95% confidence interval [CI]: 0.98-1).

In addition to collecting plasma samples at 3-month intervals throughout the follow-up until VTE occurred, the authors compared the plasma levels at the time of inclusion with that of the last plasma sample collected just before the VTE event, in the hopes that doing so would shed light on the triggers of the underlying mechanism. The following 7 miRNAs were discovered to be downregulated: miR-30e-3p, miR-let-7i-5p, miR-let-7g-5p, miR-144-3p, miR-199a-3p, miR-101-3p, and miR-15a-5p [54]. The authors hypothesized that miR-144-3p and miR-let-7g-5p would be relevant targets for future exploration since they showed the highest variation in expression levels between the inclusion and pre-VTE time points [54]. The same authors reported in 2020 a prospective trial cohort of 59 and 93 patients with glioma and meningioma, respectively [50]. Patients had brain surgery and were monitored until the onset of postsurgical pulmonary edema (PE), for which blood was collected at the time of inclusion. Cases and controls were drawn from this prospective cohort in an NCC. Using RT-qPCR and 8 miRNAs in a prediction model for PE in 50 glioma patients. Subsequent external validation resulted in a receiver operator curve (ROC) of 0.78 for 6 of 8 miRNAs [50], including miR-363-3p, miR-93-3p, miR-22-5p, miR-451a, miR-222-3p, and miR-140-3p. A similar method was used to create and externally evaluate a prediction model using 6 miRNAs in 50 meningioma patients, with a ROC of 0.69 [50]. These miRNAs included miR-29a-3p, miR-660-5p, miR-331-3p, miR-126-5p, miR-23a-3p, and miR-23b-3p.

Third, in an abstract presented at the 2019 congress of the International Society of Thrombosis and Hemostasis (ISTH), Oto et al. compared 16 prospectively followed patients with lung cancer till thrombosis, whose plasma samples were acquired at the time of inclusion. Differential regulation was observed for 14 miRNAs [60]. The plasma levels of miR-let-7i-5p, miR-let-7g-5p, miR-140-3p, and miR-363-3p were identified in more than one study, suggesting that these miRNAs are possible candidate CAT biomarkers for multiple cancer types (Oto et al., 2017; lung, brain, and pancreatic carcinoma). Using data from the Troms research [44,47], Starikova et al. [47] presented an abstract at the 2017 congress of the ISTH on the identification of predictive miRNAs in plasma separated from patients with cancer (breast, bronchus, lung, ovary, pancreatic, spinal cord, and kidney). Within the first year of blood collection, a pilot NCC research was conducted with 6 patients with CAT and 8 controls [47]. Three microRNAs, miR-324-3p (up), miR-133a-3p (down), and miR-421 (down), were shown to be significant predictors of venous thromboembolism using an elastic-net penalized logistic regression model [47]. In addition, target genes identified by software were revealed; some of these genes were involved in coagulation [47].

In Mojdeh Mahmoudian and colleagues’ study, it was discovered that certain microRNAs showed increased expression in BC tumor compared to the adjacent tissues. Specifically, hsa-miR-25-3p, -29a-5p, -105-3p, and -181b1-5p were upregulated, while hsa-miR-335-5p and -339-5p were downregulated. The upregulation or downregulation of these
candidate microRNAs was found to be associated with TNM stages, except for hsa-miR-339-5p. Additionally, with the exception of hsa-miR-105-3p, each candidate microRNA correlated with HER-2 status. Furthermore, the analysis of ROC curves revealed that the combination of these six microRNAs could potentially serve as a biomarker to differentiate between tumor and non-tumor breast tissue samples.

Kim et al. [51] presented the results of an NCC study at the 2019 American Society of Hematology conference. Cases and controls were drawn from a previously established prospective colorectal cancer cohort and were followed for 6 months. 9 miRNAs were substantially downregulated (false discovery rate 0.2) in the plasma of patients compared to controls [51], out of a total of 2426 distinct miRNAs tested. The patients and controls for our most recent NCC research on miRNAs were drawn from a prospective colorectal cancer cohort. Cases and controls had tumor tissues removed before the onset of VTE [52]. There were a total of 547 different miRNAs found. Nineteen tumor miRNAs were found to be differently regulated between VTE patients and controls, with hsa-miR-3632, hsa-miR-92b-5p, and hsa-miR-10,394-5p being the most substantially downregulated using a false discovery rate of 0.1 and a 1.5-fold difference. Of the 19 miRNAs found, seven were shown to modulate the GnRH receptor pathway [52].

These 3 published research and 3 abstracts are the only ones to date that have focused on finding miRNAs in CAT; nevertheless, in 2 of these studies (both by Oto et al.), the miRNA profile was successfully utilized to predict the likelihood of future VTE in a subset of cancer patients. However, limited repeatability was seen among the miRNAs discovered. Similar to the VTE research discussed in the preceding section, there were discrepancies in study designs, which could have impacted the findings. The NCC research design was utilized in two studies by Oto et al. [49, 50], where blood was obtained at the time of cancer diagnosis prior to the VTE occurrence, and the prospective study design was used in a third study [60], which was otherwise comparable. An NCC design was also employed by Kim et al. [51] and Anijs et al. [52], with blood collection or tissue resection occurring at the time of cancer diagnosis prior to the occurrence of VTE. Starikova et al. did not specify when blood was obtained in the abstract of their research [47], therefore it is unclear if the miRNAs they detected are more a result than a cause of VTE. While Oto et al. and Starikova et al. evaluated miRNA levels in plasma using RT-qPCR, Kim et al. utilized NGS to analyze miRNA levels in plasma samples from cancer patients, while Anijs et al. used NGS to analyze tumor tissue isolations. Since NGS has more discovery power than RT-qPCR, this difference in results might be explained. Type I and II mistakes and the margin of error are exacerbated by small sample numbers and discrepancies across techniques (such multiple testing correction). Additionally, several isolation kits were used, and there were variations in the isolation procedures for miRNA. While part of the identified miRNAs likely originate from platelets, blood cells, and endothelial cells, reflecting a more general hypercoagulable state in patients with cancer, another part of the identified miRNAs could be tumor-secreted miRNAs and represent tumor-specific miRNAs that differ per tumor. Finally, for future research or deployment of these miRNA panels, external validation was undertaken in just the 2 published papers by Oto et al.

Four microRNAs (miR-140-3p, miR-363-3p, miR-let-7i-5p, and miR-let-7g-5p) were found in many investigations (Table 3, Figure 3). It’s remarkable that one research finds upregulation of miR-let-7i-5p and miR-let-7g-5p while another finds downregulation of these same miRNAs. In spite of this, miR-140-3p and miR-363-3p were shown to be downregulated in the related investigations, suggesting a possible thrombotic function for genes that these miRNAs target and designating these miRNAs as most promising candidate CAT biomarkers employed in a larger cancer patient group.

In both cancer and noncancer situations, the following microRNAs (miRNAs) were shown to be predictive VTE biomarkers: miR-126-3/5p, miR-30a/c-5p, miR-199a/b-3p, miR-301a-3p, miR-15a-5p, miR-106-5p, miR-222-3p, miR-885-5p, miR-184, and miR-451a (Figure 3). Interestingly, these miRNA have been suggested to individually regulate the expression of coagulation factors, such as TF (miR-126, miR-106-5p, miR-451a) [24,29,61], FVIII (miR-30) [21], tissue plasminogen activator and SERPINE1/2 (miR-199, miR-301a-3p) [43,62], and protein S (miR-885-5p) [20], and regulate the recruitment of platelets (miR-15a-5p) [63], but none have yet been functionally validated in thrombosis models in
Reduced binding of miR-126 to the 3′UTR region of the F3 mRNA in vitro, as a consequence of miR-126 suppression by hairpin inhibitors, resulted in enhanced production of TF isoforms and increased TF-mediated cellular thrombogenicity [24]. In addition, human microvascular endothelial cells were shown to produce more TF in both non-inflammatory and inflammatory circumstances when miR-126 was inhibited [24]. In vitro studies using leiomyosarcoma cells showed that miR-106 acted posttranscriptionally by interacting with the 3′UTR of TF mRNA [61].

To control vascular endothelial dysfunction and a prothrombotic condition, HUVECs miR-451a aimed targeting the inflammatory cytokine receptor IL6R/CD126, activating the JAK/STAT/TF signaling cascade [29].

In vitro studies showed that miR-30c inhibited F8 expression by focusing on its 3′UTR [21]. Transfecting miR-30c into lymphoblastoid and HUVEC cell lines led to reduced FVIII expression, and adding an inhibitor specific for miR-30c resulted in a 2-fold rise in FVIII levels (as measured by immunofluorescence and immunosorbent assays; ref. 21).

Increased thrombin activity was seen in endothelial cells when miR-199a-3p was present due to its ability to inhibit SERPINE2 via its 3′UTR, hence promoting coagulation [62]. Overexpression of miR-199a-3p in vitro lowered tPA expression, demonstrating that miR-199a-3p inhibited the tPA pathway, for which SERPINE2 is an upstream regulator [62]. In vitro, miR-301a inhibited tPA and urokinase plasminogen activator (u-PA) by binding to conserved regions in the 3′UTR of SERPINE1 mRNA, encoding protein plasminogen activator inhibitor - 1 (PAI-1) [43]. There was a correlation between plasma miR-885-5p levels and protein S concentrations, which in turn predicted the ability to generate thrombin [20].

miR-15a-5p was proved to control platelet signaling by interfering with the GPVI pathway in megakaryocyte cells, but no conclusive association with VTE has been identified [63]. Although miR-222 has been implicated in cardiovascular illnesses such atherosclerosis and peripheral artery disease [64], no molecular pathways relevant to coagulation or VTE have yet been identified for miR-222-3p or miR-184.

Since it has been shown in every research that several miRNAs are differently regulated, lending credence to the concept of a multifactorial illness phenotype, it stands to reason that miRNAs should be investigated collectively in a disease model. In contrast, three of the ten overlapping miRNAs have been shown to control TF, the activator of the extrinsic coagulation pathway, indicating a pivotal function for TF in CAT. Notably, none of these miRNAs currently have a proven function, alone or in combination, with others in an in vivo thrombosis situation, and hence, careful interpretation of these data is required. A deeper knowledge of CAT pathophysiology and individualized CAT risk prediction may also be possible via the discovery of particular miRNAs that may be tumor-specific. Better estimates on the potential of these miRNAs as predictive biomarkers for VTE and CAT may be established once future research focus on overcoming the aforementioned constraints, such as proper study designs, external validation of findings, and higher statistical power. As a follow-up, functional validation studies of the miRNA profiles obtained should be done in vivo and ex vivo seeking to uncover the molecular pathways of VTE and CAT, as this would assist in the search and creation of predictive biomarkers.

6. Conclusions

The use of miRNAs as biomarkers to better predict VTE risk in cancer patients and those without the disease is an exciting new development. Understanding the role of miRNAs in the progression of VTE and CAT may lead to the development of innovative therapeutic options and the identification of new biomarkers. Understanding and foreseeing VTE is greatly aided by knowledge of the functions of microRNAs in controlling the expression of several proteins involved in hemostasis and malignancy. Few clinical research have looked at miRNAs and their ability to predict VTE in cancer-free and cancer patients. However, the vast majority of research had inadequate methods and little attempt at external validity. Several miRNAs shown to be differently regulated in the plasma of cancer-free individuals with VTE provide insight into the pathophysiological processes underpinning CAT. But there are several hurdles and blind spots in biomarker discovery that need to be overcome. Predicting VTE and CAT in a certain...
population requires first designing studies with large enough sample sizes to draw meaningful conclusions. In order to determine if miRNAs represent a promising catch among the pool of potential VTE and CAT biomarkers, validation studies are required.

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