Circulating biomarkers in chronic thromboembolic pulmonary hypertension

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Abstract

Chronic thromboembolic pulmonary hypertension (CTEPH) is a serious condition characterized with chronic organized thrombi that obstruct the pulmonary vessels, leading to pulmonary hypertension (PH) and ultimately right heart failure. Although CTEPH is the only form of PH that can be cured with surgical intervention, not all patients with CTEPH will be deemed operable. Some CTEPH patients still have a poor prognosis. Therefore, the determination of diagnostic and prognostic biomarkers of CTEPH is of great importance for the early intervention to improve prognosis of patients with CTEPH. Several markers related to multiple mechanisms of CTEPH have been recently identified as circulating diagnostic and prognostic biomarkers in these patients. However, the existing literature review of biomarkers of CTEPH is relatively sparse. In this article, we review recent advances in circulating biomarkers of CTEPH and describe future applications of these biomarkers in the management of CTEPH.

Introduction

Chronic thromboembolic pulmonary hypertension (CTEPH) is classified as group 4 pulmonary hypertension (PH) according to the World Health Organization (WHO) classification. CTEPH is a serious condition characterized by chronic organized thrombi that obstruct the pulmonary vessels, with an estimated incidence rate in the range of 0.57–3.8% after acute pulmonary embolism (PE).[1,2] More recently, our meta-analysis has revealed that the overall incidence of CTEPH after acute PE is 3.13% (95% confidence interval [CI] = 2.11–4.63).[3] Advanced CTEPH leads to an increase in pulmonary artery pressure (PAP) and pulmonary vascular resistance (PVR), subsequently resulting in progressive PH and right heart failure.[4] The pathophysiology of CTEPH is summarized in Fig. 1.

CTEPH is distinct among PH types in that it can be cured by pulmonary endarterectomy (PEA), which has become the principal treatment of choice for CTEPH.[5] However, not all patients with CTEPH are deemed operable. For patients with inoperable CTEPH, medical therapy and balloon pulmonary angioplasty (BPA) are considered alternatives to PEA.[5] Although tremendous improvement has been observed in CTEPH treatment, some patients still have a poor prognosis. Condliffe et al. reported that the one- and three-year survival rates of patients with inoperable CTEPH were only 82% and 70%, respectively.[6] Therefore, the determination of diagnostic and prognostic biomarkers of CTEPH is of great importance for the early intervention and improving prognosis of patients with CTEPH.

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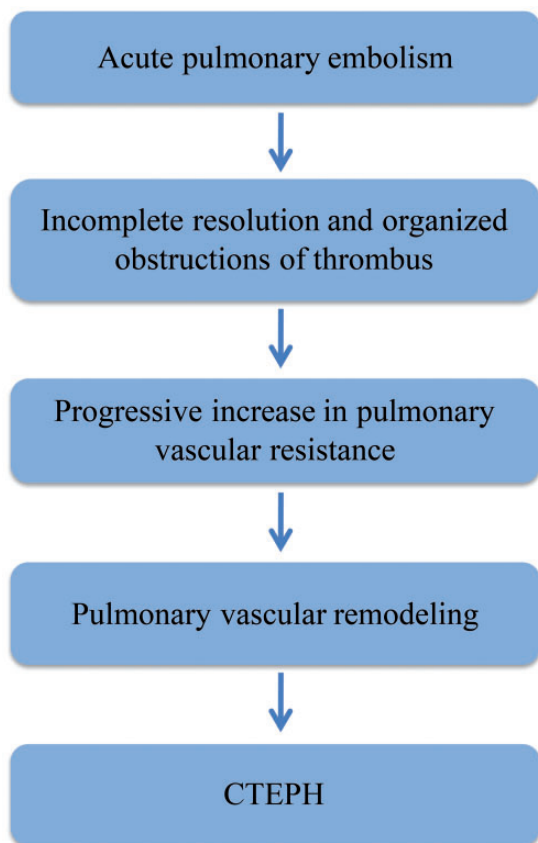Email: cyh-birm@263.net

Fig. 1. The pathophysiology of CTEPH. CTEPH, chronic thromboembolic pulmonary hypertension.

Circulating biomarkers testing has advantages as an approach to population-based disease screening, because it is non-invasive, inexpensive, and time-saving. Several markers related to multiple mechanisms of CTEPH have been recently identified as circulating diagnostic and prognostic biomarkers in these patients. Although the pathogenesis of CTEPH has not been completely elucidated, various mechanisms leading to incomplete thrombus resolution and pulmonary vascular remodeling have been shown to participate in the development of CTEPH, such as the abnormalities in coagulation and fibrinolysis, inflammation, oxidative stress, endothelial dysfunction, and excessive proliferation of pulmonary arterial smooth muscle cells (PASMC). The existing literature review of biomarkers of CTEPH is relatively sparse. The present review will focus on the current knowledge on circulating biomarkers of CTEPH that are linked to aforementioned mechanisms and describe the potential applications of biomarkers in the management of patients with CTEPH. The candidate biomarkers discussed in this article are summarized in Fig. 2.

Biomarkers of coagulation and fibrinolysis

CTEPH has been considered to result from incomplete thrombus resolution after acute PE or recurrent PE.

Increased coagulation and decreased fibrinolysis have been shown to be related to the development of CTEPH. Several thrombotic factors involved in the coagulation cascade and platelet activation have been identified as biomarkers of CTEPH in previous studies. Biomarkers related to coagulation and fibrinolysis are summarized in Fig. 3.

Plasma Factor VIII and von Willebrand factor

Plasma Factor VIII (FVIII) is the first prothrombotic factor identified in a large proportion of patients with CTEPH.[7] Bonderman et al. showed that the CTEPH patients had higher FVIII levels than healthy controls and pulmonary arterial hypertension (PAH) patients.[7] Stability and activity of FVIII are highly dependent on von Willebrand factor (vWF), a glycoprotein-mediating platelet adhesion to subendothelium. Elevated plasma concentration of vWF has also been reported in anecdotal cases of CTEPH. However, there was no association between FVIII-VWF and WHO functional status in the CTEPH group.[8]

Tissue factor and tissue factor pathway inhibitor

Tissue factor (TF) is a 47 kDa transmembrane cell surface glycoprotein responsible for triggering the extrinsic coagulation pathway. The TF pathway plays a significant role in several bleeding and clotting disorders.[9,10] Tissue factor pathway inhibitor (TFPI) is the primary inhibitor of TF-induced coagulation in vivo. And TFPI cannot only inhibit activated factor Xa directly, but also inhibit VIIa/TF activity.[11] Enhancements of TF expression have been observed in monocytes from the blood of patients with CTEPH.[12] Due to a high consumption of TF in CTEPH patients, plasma levels of TF were lower than the healthy controls, while TFPI levels were increased. Their results indicated that extrinsic coagulation pathway factors, such as TF and TFPI, may play a critical role in the pathogenesis of CTEPH.[13] Larger studies are needed to validate the roles of TF or TFPI in CTEPH and determine the utility of TF or TFPI as CTEPH biomarkers that predicts disease severity and/or response to treatment.

Antiphospholipid antibodies

Antiphospholipid antibodies (APAs) can interfere with the function of phospholipid-binding proteins in the coagulation cascade; binding of APAs induces activation of endothelial cells and platelets. The presence of APAs in CTEPH has long been studied in previous studies. Wolf et al. observed high frequency of APAs both in patients with CTEPH and primary pulmonary hypertension (PPH). In PPH, antibodies were reported only in low titer; however, half of the patients with CTEPH had high APAs titers. In PPH, these antibodies reflect endothelial dysfunction, whereas the high antibody titer in CTEPH is thought to
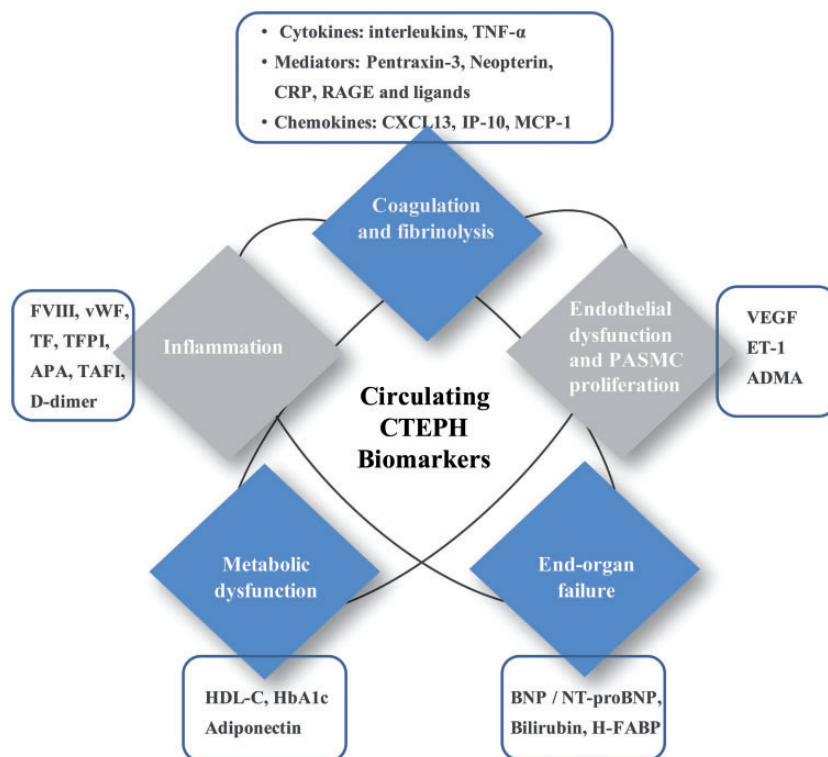
Fig. 2. A summary of circulating biomarkers in CTEPH. CTEPH, chronic thromboembolic pulmonary hypertension; CRP, C-reactive protein; MCP-1, monocyte chemoattractant protein-1; RAGE, receptor for advanced glycation end products; HMGB1, high mobility group box-1; CXCL13, chemokine CXC ligand 13; IP-10, interferon-γ-induced protein-10; ADMA, asymmetric dimethylarginine; BNP/NT-pro-BNP, brain natriuretic peptide/N-terminal-pro-brain-type natriuretic peptide; H-FABP, heart-type fatty acid-binding protein; RDW, red blood cell distribution width; FVIII, factor VIII; vWF, von Willebrand factor; TF, tissue factor; TFPI, tissue factor pathway inhibitor; APA, antiphospholipid antibody; TAFI, thrombin-activatable fibrinolysis inhibitor; ET-1, endothelin-1; VEGF, vascular endothelial growth factor; HDL-C, high-density lipoprotein cholesterol; HbA1c, glycosylated hemoglobin A1c.

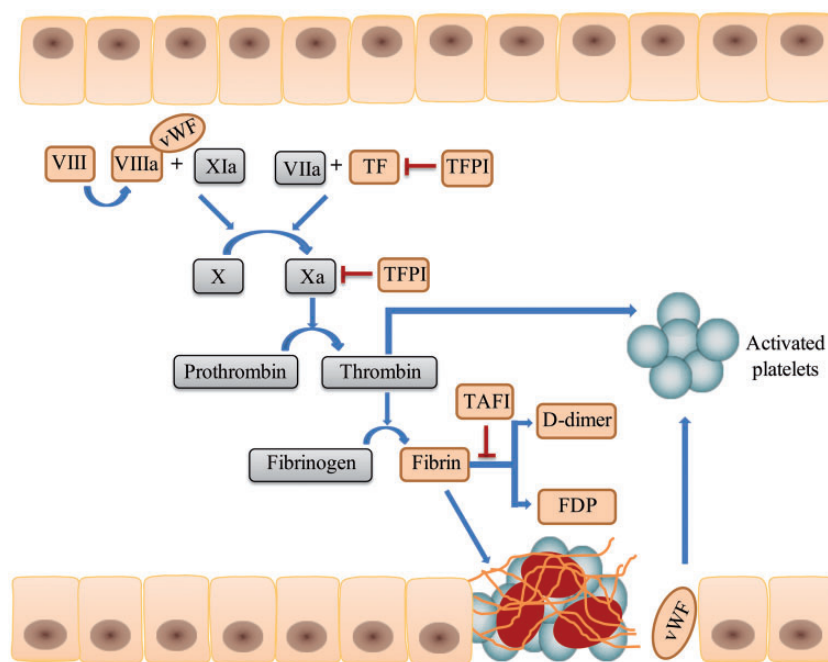**Fig. 3.** Biomarkers of coagulation and fibrinolysis in CTEPH. Orange boxes indicate biomarkers have been reported in CTEPH. CTEPH, chronic thromboembolic pulmonary hypertension; FVIII, factor VIII; vWF, von Willebrand factor; FX, factor X; FXI, factor XI; APA, antiphospholipid antibody; TAFI, thrombin-activatable fibrinolysis inhibitor; TF, tissue factor; TFPI, tissue factor pathway inhibitor; FDP: fibrinogen degradation products.

be associated with lupus anticoagulant, indicative of the critical role of thrombosis in the pathophysiology of CTEPH.[8] Similarly, Bonderman et al. also reported the association between elevated APAs levels and CTEPH. However, Wong et al. failed to reproduce the high incidence of APAs in patients with CTEPH.[7]

Fibrinogen

Fibrinogen abnormalities related to incomplete fibrinolysis have been reported in several previous studies. Fibrinogen, a 340-kDa plasma glycoprotein, comprises two pairs each of Aα, Bβ, and γ chains arranged as a rod-like protein. Elevated fibrinogen is a well-established marker of fibrinolysis abnormality. In a retrospective study, fibrinogen plasma concentrations were significantly higher in patients with CTEPH and PAH than in control patients. A significant correlation between fibrinogen and WHO functional class was reported. Fibrinogen plasma concentration is an independent marker of hemodynamic impairment in CTEPH.[14] High plasma fibrinogen levels and low plasminogen activity have been reported to be associated with poor long-term outcomes in medically treated patients with CTEPH.[15] In addition, abnormal variants of fibrinogen have been related to CTEPH. A marked difference was demonstrated in fibrinogen (Fg)-Aa Thr312Ala genotype and allele frequencies between CTEPH patients and healthy controls.[16,17] Besides fibrinogen (Fg)-Aa Thr312Ala, other variants of fibrinogen have been observed in CTEPH patients. In the study by Morris et al., five fibrinogen variants with corresponding heterozygous gene mutations were observed in 5/33 CTEPH patients: Bβ P235L/γ R375W; Bβ P235L/γ Y114H; Bβ P235L; Aα L69H; and Aα R554H. These results suggested that differences in variants of fibrinogen may play an important role in the development of CTEPH.[18]

D-dimer

D-dimer is a degradation product of cross-linked fibrin. It has been shown to be increased in hypercoagulability and thrombotic events. Previous studies failed to show the correlation of higher levels of D-dimer and prognosis of CTEPH. A recent study by Skoro-Sajer et al. observed that baseline D-dimer levels correlated with hemodynamics and WHO functional status of CTEPH patients. In addition, D-dimer independently predicted the survival of lung transplantation in these patients.[19] Several studies have also evaluated the diagnostic values of D-dimer in CTEPH. Gong et al. found that D-dimer could be considered an independent risk factor of CTEPH after acute PE.[20] The study by Klok et al. did not show the similar tendency. CTEPH patients exhibited plasma D-dimer levels similar to the symptomatic patients without PH.[21] The results from another study indicated that D-dimer was an insensitive and non-specific test for the diagnosis of CTEPH.

Despite the high negative predictive value, D-dimer alone could not be used to rule out CTEPH.[22]

Thrombin-activatable fibrinolysis inhibitor

Thrombin-activatable fibrinolysis inhibitor (TAFI) is a carboxypeptidase inhibitor synthesized by the liver and known to inhibits fibrinolysis. General review suggested that TAFI played a pivotal role in thrombus formation and mediated the pulmonary vascular inflammation.[23] In comparison with PAH patients or controls, CTEPH patients showed significantly elevated levels of TAFI.[24] Additionally, TAFI can be easily detected in outpatient clinic. Hence, TAFI has been potential to be a promising biomarker and a therapeutic target of CTEPH.

Biomarkers of inflammation

There is accumulating evidence supporting the involvement of the inflammation in the pathogenesis of CTEPH. Inflammatory cytokines and chemokines contribute directly to further recruitment of inflammatory cells and proliferation of PASMCs and endothelial dysfunction. A variety of both pro-inflammatory and anti-inflammatory cytokines and chemokines have been reported as potential biomarkers of CTEPH (Fig. 4).

Cytokines

Cytokines have been regarded as critical mediators of inflammation in several pulmonary diseases.[25–27] Higher levels of interleukins (ILs) and TNF-α were detected in the serum of CTEPH patients.[12,28–31] In the study by Soon et al., CTEPH patients exhibit elevated levels of IL-1β, IL-2, IL-4, IL-6, IL-8, and IL-10. Of these, IL-6 and IL-8 performed well in predicting long-term response to PEA.[31] Kimura et al. detected the circulating levels of IL-1β and TNF-α of CTEPH patients. However, IL-1β and TNF-α failed to show any correlation with pulmonary hemodynamics.[32] In another study, authors reported a significantly elevated level of IL-8 and IL-6 in the serum of CTEPH patients compared to health controls, while only IL-6 changes were associated with hemodynamic parameters and exercise capacity.[30] In addition, the study by Langer et al. evaluated the expression of pro-inflammatory and anti-inflammatory cytokines in CTEPH patients undergoing PEA. They found that CTEPH patients exhibited elevated TNF-α, IL-6, and IL-10 levels before surgery. In addition, elevated levels of IL-10 and IL-6 exhibited a marked peak immediately after surgery, while elevated levels of TNF-α decreased significantly within the first 24 postoperative hours.[29] Taken together, although various cytokines have been identified as potential biomarkers of CTEPH, it is to be noted that these biomarkers are still in the investigative phase and more pre-clinical and clinical studies are demanded.
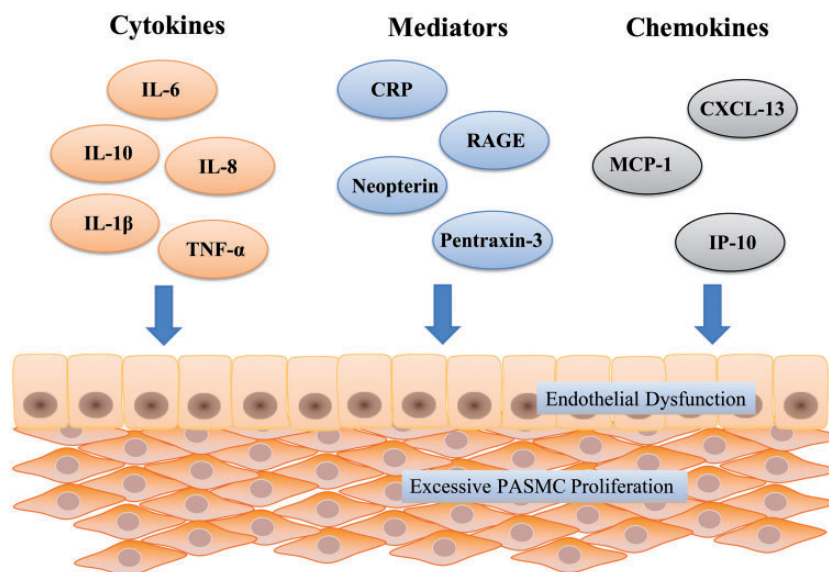
Fig. 4. Biomarkers of inflammation in CTEPH. CTEPH, chronic thromboembolic pulmonary hypertension; PASMC: pulmonary arterial smooth muscle cell; IL: interleukin; CRP, C-reactive protein; MCP-1, monocyte chemoattractant protein-1; RAGE, receptor for advanced glycation end products; HMGB1, high mobility group box-1; CXCL13, chemokine CXC ligand 13; IP-10, interferon-γ-induced protein-10; BNP/NT-pro-BNP, brain natriuretic peptide/N-terminal-pro-brain-type natriuretic peptide; H-FABP, heart-type fatty acid-binding protein; RDW, red blood cell distribution width; PASMC, pulmonary arterial smooth muscle cell.

C-reactive protein

C-reactive protein (CRP) is a well-known biomarker of inflammation, widely recognized as a predictor of various cardiovascular diseases. CRP could contribute to CTEPH through the promotion of the pulmonary vascular remodeling, endothelial function, and in situ thrombosis.[33] CRP is one of the first identified inflammatory factors involved in CTEPH. The work by Quarck et al. showed that the patients with CTEPH displayed increased levels of CRP levels, which significantly decreased after 12 months of PEA. The close correlation between plasma CRP levels and neutrophil and macrophage accumulation has been reported.[28,34] Additionally, in CTEPH patients, plasma levels of CRP were associated with TF antigen, reflecting the crosstalk of inflammation and thrombosis in the pathogenesis CTEPH.[12] Similarly, the study by Skoro-Sajer et al. also implicated the roles of inflammation and thrombosis in the pathobiology of CTEPH. The results showed that D-dimer and CRP levels both were the independent predictors of clinical outcome, which decreased distinctively after PEA.[19] Kolk et al. developed a diagnostic model for ruling out CTEPH in symptomatic patients after acute PE. They included 82 consecutive patients with confirmed CTEPH and 160 consecutive patients with a history of PE that were suspected to have CTEPH but were ruled out for CTEPH. The circulating levels of CRP were reported to be significantly higher in patients with CTEPH in this study.[21] However, a potential limit for the use of CRP in CTEPH is that CRP elevation occurs in many different clinical situations.

Receptor for advanced glycation end products and its ligands

Receptor for advanced glycation end products (RAGE), a member of the immunoglobulin protein family of cell surface molecules, may bind to a variety of structurally diverse ligands, including advanced glycation end products, S100/calgranulin proteins, and high mobility group box-1 (HMGB1). RAGE is expressed at a low basal level in the majority of healthy adult tissues. The upregulation of RAGE has been implicated in many cardiovascular diseases.[35,36] Studies have shown that RAGE and its ligands play evident roles in the pathogenesis of PH via promoting PASMC proliferation and migration.[37] Moser et al. detected the concentrations of RAGE and HMGB1 in a cohort of 26 patients with CTEPH and 15 patients with idiopathic pulmonary arterial hypertension (IPAH). Interestingly, these authors reported the increased levels of soluble RAGE (sRAGE) and endogenous secretory RAGE (esRAGE) in the serum of patients with CTEPH and IPAH, whereas high levels of HMGB1 were only detected in CTEPH group. However, serum sRAGE changes in CTEPH patients showed no association with PAP and no difference before and after PEA could be found.[38] Another study subsequently reported that the significantly higher levels of sRAGE, not HMGB1, in CTEPH group than in healthy individuals, indicating that sRAGE level is superior to HMGB1 level in reflecting the pathological condition of patients with CTEPH. Besides, sRAGE levels showed a notable decrease

after BPA.[39] These inconsistent results warrant further investigations to explore the potential roles of RAGE and its ligands in CTEPH.

Pentraxin 3

Several new pentraxins (PTXs) have been discovered in recent years. Of these, PTX3 was the first long PTX to be discovered. PTX3 is produced by multiple cell types, but most prominently by endothelial cells and mononuclear phagocytes in response to stimulation of various inflammatory cytokines. PTX3 has been considered as a useful biomarker for PAH, especially in patients with connective tissue disease.[40] Naito et al. found that PTX3 levels were elevated in patients with CTEPH compared with healthy controls, whereas PTX3 failed to show any correlation with hemodynamic severity and the response to PEA in CTEPH. Furthermore, higher levels of PTX3 may serve as a potential indicator for the early diagnosis of CTEPH in patients after acute PE.[41]

Neopterin

Neopterin (NP), an early biomarker of cellular immune response, is secreted by macrophages and dendritic cells after stimulation with interferon-γ (IFN-γ) and has been shown to be elevated in multiple cardiovascular diseases including stable angina pectoris, myocardial infarction, and coronary atherosclerotic lesions.[42,43] Elevated NP concentrations were closely correlated with adverse clinical outcomes and hemodynamic parameters. NP may participate in the development of CTEPH via oxidative stress enhancement and act as a potential prognostic biomarker of CTEPH.[44]

Monocyte chemoattractant protein-1

Monocyte chemoattractant protein-1 (MCP-1), as a potent chemotactic factor for monocytes, belongs to the C-C chemokine family. MCP-1 is predominantly produced by macrophages and endothelial cells. Several studies have documented the elevated levels of circulating MCP-1 in patients with CTEPH.[12,28] In addition, MCP-1 levels closely correlate with hemodynamic parameters.[12,32] In a previous study, the circulating levels of MCP-1 were significantly associated with PVR in patients with CTEPH.[32] In a more recent study, Yang et al. confirmed the correlation between MCP-1 levels and hemodynamics.[12] Restricted to the small sample sizes of these studies, larger studies are warranted to characterize the potential role of MCP-1 for the assessment of CTEPH severity.

Chemokine CXC ligand 13

Chemokine CXC ligand 13 (CXCL13) has been implicated in perivascular inflammation and pulmonary vascular remodeling in patients with IPAH. Olsson et al. have reported that higher concentrations of CXCL13 were detected in the serum of patients with IPAH and CTEPH, suggesting the potential role of CXCL13 in the pathogenesis of CTEPH. However, considering the weak associations between serum CXCL13 and markers of disease severity and outcome, the authors suggested that CXCL13 was unlikely to become a promising biomarker in these patient populations.[45]

Interferon-γ-induced protein-10

Increased levels of interferon-γ-induced protein-10 (IP-10) have been detected in patients with CTEPH and IPAH. The study by Zabini et al. showed elevated levels of IP-10 messenger RNA and protein levels in PEA tissues. Elevated levels of IP-10 showed correlation with the poor hemodynamic and clinical outcomes. This study has further implicated the potential roles of IP-10 in the pathophysiology of CTEPH, including promotion of fibroblast migration and endothelial dysfunction.[30]

Red blood cell distribution width

Red blood cell distribution width (RDW) is a measurement of the heterogeneity in size of circulating erythrocytes. Higher RDW levels indicate the presence of anisocytosis, reflecting an underlying chronic inflammation.[46,47] Recent studies have identified RDW as a biomarker for the evaluation of clinical outcome in patients with several cardiovascular and pulmonary diseases.[48,49] RDW levels are elevated in different subtypes of PH and correlated with the mortality in patients with PH;[50–52] furthermore, RDW had a better prognostic value than NT-pro-BNP.[50] In another study of 77 patients with PAH and inoperable CTEPH, baseline RDW levels showed no correlation with the prognosis; however, gradually increasing RDW values during follow-up were significantly associated with poor prognosis. RDW levels in survivors that had received the targeted therapies showed a distinctive decrease.[51] Similar results have been reported in the study by Wang et al.[52] In addition, RDW has been identified as an independent risk factor of CTEPH in patients after acute PE and may serve as a potential marker for CTEPH prediction.[53]

Biomarkers of endothelial dysfunction and PASMC proliferation

Although the pathogenesis of CTEPH has not been completely elucidated, it is widely accepted that CTEPH shares similar pathological characteristics of vascular remodeling with PAH including endothelial dysfunction and excessive PASMC proliferation. Biomarkers related to endothelial dysfunction and PASMC proliferation are summarized in Fig. 5.
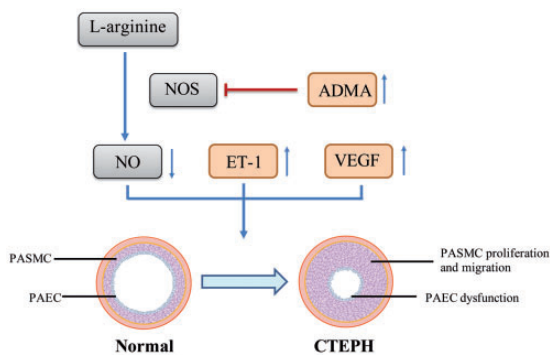
Fig. 5. Biomarkers of endothelial dysfunction and PASMC proliferation in CTEPH. Orange boxes indicate biomarkers have been reported in CTEPH. CTEPH, chronic thromboembolic pulmonary hypertension; PASMC: pulmonary arterial smooth muscle cell; PAEC: pulmonary arterial endothelial cell; ADMA, asymmetric dimethylarginine; ET-1, endothelin-1; VEGF, vascular endothelial growth factor; NO: nitric oxide; NOS: nitric oxide synthase.

Endothelin-1

Endothelin-1 (ET-1) is the most potent vasoconstrictor predominantly produced and released predominantly by vascular endothelial cells. ET-1 is involved in cardiovascular homeostasis and respiratory development.[54] Aside from its role as a potent vasoconstrictor, ET-1 promotes PASMC proliferation and migration.[55,56] It is well established that the endothelin-receptor antagonists exert multiple beneficial effects in the treatment of PAH and is commonly used in PAH. In patients with non-thromboembolic PH, plasma ET-1 concentrations are detectable at high levels, correlating with hemodynamic severity.[56,57] In a previous study, a positive correlation was observed between preoperative big ET-1 levels and the clinical severity of CTEPH. Preoperative big endothelin-1 levels may be a potential predictor of hemodynamic outcome after PEA.[58] In agreement with this study, a larger cohort study also indicated the prognostic value of ET-1 in the hemodynamic outcome after PEA. It was suggested that ET-1 could be used to identify the patients at risk for persistent or residual PH after PEA.[59]

Vascular endothelial growth factor

Vascular endothelial growth factor (VEGF) has multiple isoforms, including VEGF-A, -B, -C, and -D, all of which are known to mediate angiogenesis and vasculogenesis. In a recent study, the plasma concentrations of angiogenic and inflammatory biomarkers were evaluated in different types of PH. VEGF-A levels were significantly elevated in patients with CTEPH compared with the controls, whereas no association was reported between VEGF-A and hemodynamics.[60]

Asymmetric dimethylarginine

Asymmetric dimethylarginine (ADMA) is characterized as an endogenous and competitive nitric oxide (NO) synthase inhibitor. ADMA concentrations are associated with reduced activity of NO synthase. Increased ADMA plasma levels have been reported in several different types of PH.[61–64] In patients with IPAH, elevated ADMA levels correlate significantly with unfavorable pulmonary hemodynamics and survival.[65] In patients with chronic obstructive pulmonary disease, ADMA may help in the determination of the presence of PH.[63] In patients with CTEPH, elevated ADMA levels were associated with mean right atrial pressure, cardiac index, and mixed venous oxygen saturation.[62] Zhang et al. observed similar tendency in their study, wherein patients with both CTEPH and PAH had significantly higher ADMA levels than the control group.[64]

Biomarkers of end-organ failure

In CTEPH, pulmonary vascular remodeling leads to increase in the PAP and PVR, which results in right heart failure. Low cardiac output subsequently leads to tissue hypoxia and organ damage. Biomarkers of end-organ failure are summarized in Fig. 6.

Brain natriuretic peptide/ N-terminal-pro-brain-type natriuretic peptide

Natriuretic peptides are released from cardiac myocytes in response to pressure and volume overload. Brain natriuretic peptide (BNP) or N-terminal-pro-brain-type natriuretic peptide (NT-pro-BNP) levels were demonstrated to be increased in response to right ventricular remodeling and dysfunction.[66–68] BNP or NT-pro-BNP has been identified as the first blood-derived marker of PH. There is accumulating evidence that circulating elevated levels of BNP or NT-pro-BNP have been reported in different types of PH.[67,69,70] In patients with CTEPH, plasma BNP may not only reflect the hemodynamic severity of disease in patients with CTEPH, but also help evaluate the effect of PEA.[70] In addition, BNP/NT-pro-BNP was also an independent risk factor of CTEPH in patients after acute PE.[21,71] Circulating levels of NT-pro-BNP were significantly higher in patients with CTEPH than in patients ruled out for CTEPH. A diagnostic model, including ECG criteria and NT-pro-BNP levels, had a relatively high sensitivity and specificity and may be used to rule out CTEPH in patients after acute PE.[21]

Heart-type fatty acid-binding protein

Heart-type fatty acid-binding protein (H-FABP), a low-molecular-weight cytosolic protein, is abundantly present in the cytosol of cardiomyocytes. H-FABP has already been considered as an early and sensitive biomarker of acute coronary syndromes.[72,73] H-FABP has long been described as an excellent biochemical cardiac marker in patients with PE.[74] Lankeit et al. investigated the use of H-FABP for risk assessment of CTEPH. The findings of this study demonstrated H-FABP as an independent
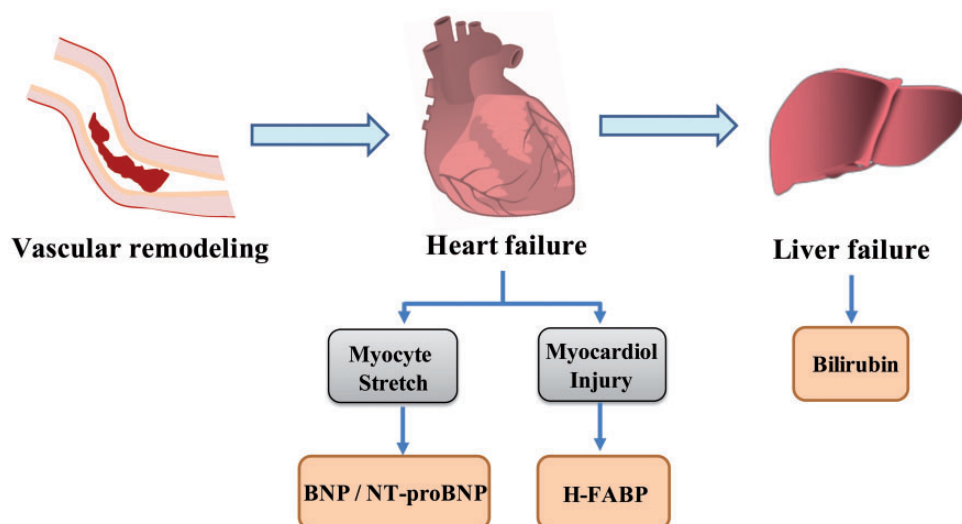
Fig. 6. Biomarkers of end-organ failure in CTEPH.[89,90] Orange boxes indicate biomarkers have been reported in CTEPH. CTEPH, chronic thromboembolic pulmonary hypertension; BNP/NT-pro-BNP, brain natriuretic peptide/N-terminal-pro-brain-type natriuretic peptide; H-FABP, heart-type fatty acid-binding protein.

predictor of adverse outcomes (CTEPH-related death, lung transplantation, or persistent PH after PEA) in multivariable analysis. Moreover, the results also indicated the superiority of H-FABP over troponin T in the diagnostic and prognostic evaluation of CTEPH.[75] Although these early data are encouraging, further studies involving larger populations are needed to confirm these findings.

Bilirubin

Chronic heart failure results in a broad range of liver abnormalities, the most common being elevated serum bilirubin levels.[76] In a prospective cohort study by Gong et al., elevated levels of total bilirubin showed significantly correlation with mortality, suggestive of its potential as a potential predictor for poor prognosis in inoperable CTEPH.[77]

Biomarkers of metabolic dysfunction

An emerging "metabolic theory" of PH suggests that metabolic dysfunction may underlie abnormalities in pulmonary vascular pathologies. Increasing evidence indicated that various components of metabolic syndrome, such as insulin resistance, hyperglycemia, and dyslipidemia were implicated in the development of PAH.[78–80] Although studies on the role of metabolic dysfunction in CTEPH are still lacking, some biomarkers related to metabolic dysfunction have been reported to correlate with the development of CTEPH, indicating that metabolic reprogramming may also play an important role in CTEPH (Fig. 7).

High-density lipoprotein cholesterol

High-density lipoprotein cholesterol (HDL-C) is an established marker of cardiovascular diseases. Several studies have reported decreased levels of HDL-C in patients with PAH and its association with worse clinical outcomes.[81,82] In a retrospectively cohort study of 90 CTEPH patients, high levels of HDL-C showed a strong correlation with decreased right ventricular dilation and considerable decrease in postoperative PVR.[83]

Adiponectin

Adiponectin is a plasma protein exclusively expressed in the adipose tissue. Adiponectin has a protective role in various pathological conditions. In a study of 30 patients with CTEPH that underwent interventional therapy of BPA or PEA, serum adiponectin levels positively correlated with BNP and hemodynamic parameters. There were significant improvements in adiponectin levels after BPA or PEA. Therefore, serum adiponectin changes in association with clinical outcome and hemodynamic severity suggest its role as a prognostic biomarker of CTEPH. Given the fact that the mechanism of adiponectin contributing to CTEPH has not been completely understood, further studies are needed to identify all potential roles of adiponectin in CTEPH.[84]

Hemoglobin A1c

Studies have revealed the relationship between insulin resistance and PH.[79,85–87] Although measurement of hemoglobin A1c (HbA1c) does not quantitate insulin resistance, elevated HbA1c could indicate abnormal glucose metabolism. Insulin resistance and glucose intolerance have been noted in PAH.[79,86,87] In patients with PAH, high HbA1c levels correlate with poor prognosis.[86,87] Richter et al. evaluated impaired glucose metabolism, as assessed by HbA1c, in a prospective cohort of 102 patients with CTEPH. Baseline HbA1c levels were significantly associated with cardiac
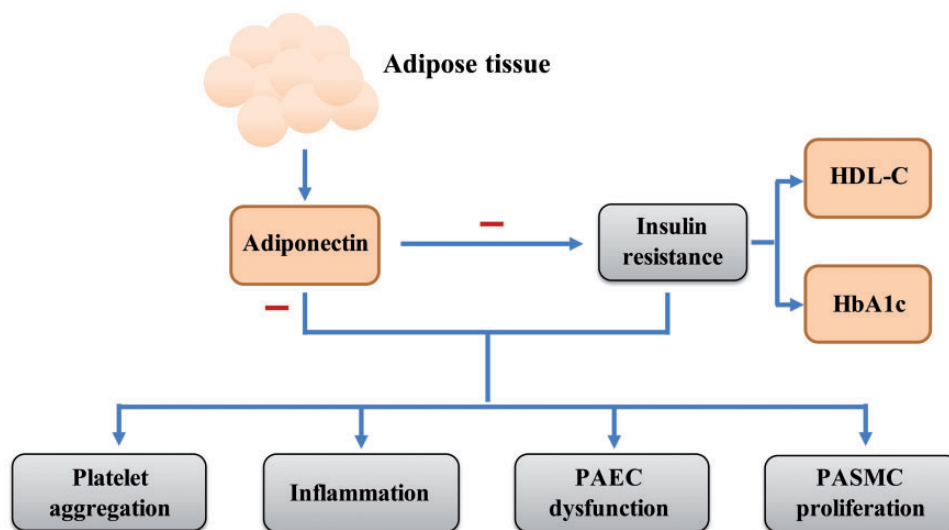
Fig. 7. Biomarkers of metabolic dysfunction in CTEPH. Orange boxes indicate biomarkers have been reported in CTEPH. CTEPH, chronic thromboembolic pulmonary hypertension; PASMC: pulmonary arterial smooth muscle cell; PAEC: pulmonary arterial endothelial cell; HDL-C: high-density lipoprotein cholesterol; HbA1c: hemoglobin A1c.

index, right atrial pressure, peak oxygen uptake, and change in 6-min walking distance, whereas no correlation was reported with hemodynamic outcome one year after PEA.[88]

Summary

The present article reviewed published studies on potential circulating biomarkers in CTEPH. A summary of circulating biomarkers for diagnosis and prognosis of CTEPH in Table 1. Although a wide variety of biomarkers have been explored in the field, none of the circulating biomarkers identified in CTEPH are specific or effective as a routine screening test in a clinical setting. Most studies only report the serum changes of circulating biomarkers in CTEPH, but failed to show the significant correlations with diagnosis and prognosis. Presently, BNP/NT-pro-BNP remains the only biomarker that is widely used in the routine practice of PH. They have been shown to be the potential markers of heart failure caused by any of cardiovascular diseases. 2015 ESC guideline suggested that BNP/NT-pro-BNP could be used for the risk assessment of PAH; however, it did not provide explicit recommendations for the use of BNP/NT-pro-BNP in patients with CTEPH.[5] Further efforts are required to identify the reliable cut points of the BNP/NT-pro-BNP levels in the risk stratification and assessment for the follow-up of CTEPH. In addition, considering the complexity of CTEPH, it is likely that no single biomarker will accurately represent all the relevant information required for an individual patient; a combination of several biomarkers in a more personalized manner may be more effective.

Our review indicated that although various biomarker reports for CTEPH have been published, the majority of these have studied small numbers of patients or has been

Table 1. A summary of circulating biomarkers for diagnosis and prognosis of CTEPH.

Biomarker	Diagnosis	Prognosis	Correlation with disease severity	Correlation with treatment effect
Coagulation and fibrinolysis				
FVIII[7,8]	+	+	–	NA
vWF[7,8]	NA	NA	–	NA
TF[13]	NA	NA	NA	NA
TFPI[13]	NA	NA	NA	NA
APAs[7,8]	+	NA	–	NA
Fibrinogen[14–17]	+	+	+	–
D-dimer[19–21]	+	+	+	+
TAFI[24]	NA	NA	NA	–
Inflammation				
IL-1β[31,32]	NA	–	–	–
IL-6[29–31]	NA	+	+	+
IL-8[28–31]	NA	+	+	+
IL-10[28,29,31]	NA	+	–	+
TNF-α[12,29,32]	NA	+	–	+
CRP[12,19,21,28,34]	+	+	+	+
MCP-1[12,28,32]	NA	NA	+	NA
RDW[51–53]	+	+	+	+
RAGE[38,39]	NA	NA	–	+
HMGB1[38,39]	NA	NA	NA	NA
CXCL13[45]	NA	–	+	–
IP-10[30]	NA	NA	+	NA
Pentraxin 3[41]	+	NA	+	–
Neopterin[44]	NA	+	+	–
Vascular remodeling				
ET-1[58,59]	–	+	+	+
VEGF[60]	NA	NA	–	NA

(continued)

Table 1. Continued

Biomarker	Diagnosis	Prognosis	Correlation with disease severity	Correlation with treatment effect
ADMA[62]	NA	+	+	+
End-organ failure				
BNP/NT-pro-BNP[21,70,71]	+	+	+	+
H-FABP[75]	NA	+	+	NA
Bilirubin[77]	NA	+	+	–
Metabolic dysfunction				
HDL-C[83]	NA	–	+	+
Adiponectin[84]	NA	+	+	+
HbA1c[88]	NA	NA	+	–

CTEPH, chronic thromboembolic pulmonary hypertension; NA, not assessed; IL, interleukin; TNF-α, tumor necrosis factor-a; CRP, C-reactive protein; MCP-1, monocyte chemoattractant protein-1; RAGE, receptor for advanced glycation end products; HMGB1, high mobility group box-1; CXCL13, chemokine CXC ligand 13; IP-10, interferon-γ-induced protein-10; ADMA, asymmetric dimethylarginine; BNP/NT-pro-BNP, brain natriuretic peptide/N-terminal-pro-brain-type natriuretic peptide; H-FABP, heart-type fatty acid-binding protein; RDW, red blood cell distribution width; FVIII, factor VIII; vWF, von Willebrand factor; TF, tissue factor; TFPI, tissue factor pathway inhibitor; APAs, antiphospholipid antibodies; TAFI, thrombin-activatable fibrinolysis inhibitor; ET-1, endothelin-1; VEGF, vascular endothelial growth factor; HDL-C, high-density lipoprotein cholesterol; HbA1c, glycosylated hemoglobin A1c.

limited to retrospective studies. Some biomarkers that have performed well to date will need to be validated in larger patient populations. Therefore, some studies of circulating biomarkers in CTEPH have reported conflicting results or have failed to fulfill earlier expectations. More prospective analyses and larger cohort studies should be carried out for validation and future research.

Proteomic approaches may uncover the disease-specific targets and biomarkers, providing promising diagnostic and prognostic information. However, proteomic data from patients with CTEPH was relatively less. Morris et al. used proteomic analysis to detect serum biomarkers by evaluating the serum profiles of low-molecular weight peptides in CTEPH patients. They found that fibrinogen Aα chain fragment may be a potential diagnostic biomarker for CTEPH.[18] Further focused research in this field is imperative to identify novel biomarkers in patients with CTEPH.

Conflict of interest
The author declare that there is no conflict of interest.

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