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Original Research

A combination of platelet features allows detection of early-stage cancer

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Abstract ***Background:*** Detection of early-stage cancer significantly improves patient survival. As platelets play an important role in cancer progression, we aimed to investigate whether platelets can be used for the discovery of early-stage cancer.
Methods: Patients with lung (n = 86) or head of pancreas (n = 42) cancer were included, as were healthy sex- and age-matched controls (n = 92). Blood was collected before initiation of treatment. Platelet count, volume and activation status were quantified in whole blood. Next, concentrations of vascular endothelial growth factor, platelet-derived growth factor, platelet factor 4, thrombospondin-1 and connective tissue–activating peptide III were measured in both platelets and plasma. Using the results, two multivariable diagnostic models were developed and internally validated.
Findings: Multiple platelet features, including platelet count, volume and protein content, were significantly changed in lung and head of pancreas cancer patients. However, the pattern

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of changes differed between both groups. The diagnostic model developed for lung cancer discriminated very well between patients and controls (AUC = 88.7%). Addition of smoking as a variable significantly increased the AUC of the model to 94.5%. The diagnostic model for head of pancreas cancer also performed well (AUC = 82.7%). Both models were internally validated, resulting in optimism-corrected AUC's of 86.8% and 80.8%, respectively.
Interpretation: In patients with lung or head of pancreas cancer, several platelet characteristics are changed compared to healthy sex- and age-matched controls. A cancer type-specific combination of these platelet features can be used to discriminate between patients with early-stage cancer and healthy individuals.

1. Introduction

Detection of cancer in its early stages radically improves the effectiveness of available treatment and overall prognosis of patients [1]. Up to now, studies searching for biomarkers are mostly based on blood plasma or serum parameters. A limitation of this approach is that platelets and their content are neglected [2].

Circulating platelets contain numerous proteins, including growth factors, chemokines and proteases, which are synthesised by megakaryocytes or absorbed from the blood by the platelets themselves [3]. Therefore, the presence of a growth factor-producing tumour can influence platelet content. Concentrations of angiogenic factors, like vascular endothelial growth factor (VEGF) and platelet-derived growth factor (PDGF), and the angiostatic platelet factor 4 (PF4) have been reported to be higher in platelets of patients with cancer than in platelets of healthy individuals [4,5]. Another recent study suggests that platelet mRNA profiles are also altered in patients with cancer, enabling discrimination between cancer patients and healthy individuals [6]. In addition, platelets may become activated systemically or within the tumour, potentially resulting in release of platelet content into the circulation [3]. Next to platelet content and platelet activation, platelet count is also frequently changed in cancer patients [7]. Tumours can increase platelet production by secretion of thrombopoietic cytokines, leading to paraneoplastic thrombocytosis [7,8].

Altogether, data from literature suggest that several platelet characteristics are affected in cancer patients. These features, either alone or in combination, may be useful tools in the detection of (early stages of) cancer. It was the aim of the present study to investigate, in two different groups of cancer patients, whether and how platelet features are changed in the presence of a tumour. In addition, we succeeded to combine these features into two internally validated diagnostic models, one for lung cancer and another one for head of pancreas cancer, to discriminate between patients and healthy individuals.

2. Methods

2.1. Study design and participants

This study was performed in accordance with the Declaration of Helsinki and approved by the medical ethical committee of Maastricht University Medical Center+. Informed consent was obtained from all participants. Patients with clinically established and histologically proven untreated primary lung (n = 86) or head of pancreas cancer (n = 42, including pancreas head cancer [n = 28], distal cholangiocarcinoma [n = 8] and duodenumcarcinoma [n = 6]), that were eligible for surgical resection, were included between July 2012 and October 2014. Exclusion criteria were previous history of cancer, neo-adjuvant chemotherapy or radiotherapy, use of platelet-influencing drugs such as aspirin, blood or platelet transfusion during the previous 14 days, active inflammatory disease, non-healing ulcers or fractures. Staging was performed in accordance with the tumour-node-metastasis (TNM) classification (version 7) of the Union for International Cancer Control [9]. A sex- and age-matched healthy control population for both cancer groups was included as well (Table 1).

2.2. Procedures

Blood from all patients and healthy individuals was collected. In case of cancer patients, sampling occurred within 1 week before initiation of treatment. To prevent platelet activation during blood collection and sample preparation, blood was collected as described before [10].

The procedures for quantification of platelet count, volume and activation status, as well as concentrations of various growth factors and chemokines in platelets and plasma, are described extensively in the Appendix. In short, platelet count and volume were determined in whole blood with a Beckman coulter counter, and platelet activation was quantified in whole blood using flow cytometry. The remaining blood was centrifuged several times to obtain platelet-free plasma (PFP) and