Mean Platelet Volume May Represent a Predictive Parameter for Overall Vascular Mortality and Ischemic Heart Disease

Georg Slavka, Thomas Perkmann, Helmuth Haslacher, Stefan Greisenegger, Claudia Marsik, Oswald F. Wagner, Georg Endler

Objective—An increased mean platelet volume (MPV), as an indicator of larger, more reactive platelets resulting from an increased platelet turnover, may represent a risk factor for overall vascular mortality, including myocardial infarction. We intended to identify patients at higher risk of dying from vascular disease in a large, hospital-based cohort.

Methods and Results—A total of 206,554 first-ever transmittals to the Allgemeines Krankenhaus Wien for determination of MPV between January 1996 and July 2003 were included. Primary end points were overall vascular mortality and death due to ischemic heart disease. Multivariate Cox regression adjusted for sex, age, and platelet count was applied for analysis. MPV values were categorized into quintiles, with the lowest quintile serving as the reference category. Compared with individuals with lower MPV (<8.7 fL), hazard ratios for overall vascular mortality gradually increased to 1.5 in the highest category (≥11.01 fL). The relationship of MPV to ischemic heart disease was even stronger and increased from 1.2 (8.71 to 9.60 fL category) to 1.8 in the highest category (≥11.01 fL).

Conclusion—Our results indicate that patients with an increased MPV (≥11.01 fL) are at higher risk of death due to ischemic heart disease, with hazard ratios comparable to those reported for obesity or smoking. (Arterioscler Thromb Vasc Biol. 2011;31:00-00.)

Key Words: cardiovascular disease prevention ■ ischemic heart disease ■ platelets ■ cardiovascular mortality ■ mean platelet volume

Platelets are known to have a major effect on the formation of atherosclerotic plaques and therefore play an essential role in the pathogenesis of atherothrombosis. Larger and hyperreactive platelets accelerate the formation of an intra-coronary thrombus, leading to a cascade of clinical events, such as acute coronary syndrome. An increase in platelet aggregability is associated with unstable angina and myocardial infarction. Platelet size and activity are correlated, and mean platelet volume (MPV) was found to be increased before acute myocardial infarction.

Also, patients with a severe stroke significantly more often have higher MPV levels on admission to the hospital. There is evidence that causal occlusion of the coronary artery in unstable angina is mediated by a platelet-rich thrombus. In stable conditions, it has been shown that MPV, which is the most accurate measure of the size of platelets, is inversely associated with platelet count, indicating the establishment of constant hemostasis. MPV is a simple and accurate marker of the functional status of platelets. As described previously, larger platelets are more reactive. Platelet size is determined at the level of the progenitor cell (ie, the megakaryocyte), and studies have reported that cytokines, such as interleukin-3 or interleukin-6, influence megakaryocyte ploidy and can lead to the production of more reactive, larger platelets. Thus, platelet volume has been proposed as an indirect marker of increased platelet reactivity. The majority of patients with suspected or known coronary artery disease are on aspirin. However, aspirin has no effect on platelet size.

An increased MPV, as an indicator of larger, more reactive platelets resulting from an increased platelet turnover, may therefore represent a risk factor for overall vascular mortality, including myocardial infarction. Our goal was to identify patients at high risk of dying from vascular disease in large, hospital-based cohort. All patient samples were collected and processed at admission of the patient at the Allgemeines Krankenhaus Wien. The role of high MPV as a potential risk factor for arterial thrombosis has been a matter of intense debate and the subject of several studies in the past. Because of the limited sample size of the studies, as well as variations of the chosen end points, several studies led to...
rather less significant results. Until now, the impact of high MPV levels on all-cause cardiovascular mortality has not been evaluated a hospital-based cohort of this size; previously, smaller study sizes and the lack of objectively defined end points made it impossible to accurately estimate risk. We evaluated the relevance of MPV as a potential predictor for vascular mortality in a large-scale cohort, via record linkage with the Austrian death registry.

Methods
A total of 218 836 first-ever transmittals to the Allgemeines Krankenhaus Wien for determination of MPV between January 1996 and July 2003 were analyzed, of whom 206 554 individuals fulfilled the inclusion criteria of our study. The median follow-up time was 4.7 years (interquartile range [IQR], 2.9 to 6.4 years), resulting in a total of 957 890 person-years at risk.

Patients
Inclusion criteria were a valid MPV, age >18 years, and complete patient data, including sex, name, and date of birth, required for successful record linkage. Exclusion criteria were incomplete patient data or admission from an intensive care unit, to exclude critical diseases that might influence MPV in total.

MPV values were collected from our laboratory software, Molis. Record linkage was performed via database query of the Austrian death registry, resulting in date of death (if it occurred between January 1990 and December 2004) and cause of death, encoded according to either the International Code of Diseases, version 9 (ICD9) (before 2002) or ICD10. The Austrian death registry comprises all deaths within Austria and the deaths of Austrian citizens in foreign countries, if reported to Austrian officials. According to Austrian laws, all deaths have to undergo post mortem examination, if the final cause of death is not evident from the patients’ history, resulting in an overall post mortem frequency of 58% in our study. For statistical analysis, only anonymized data were used, containing no personal information except age in years and sex. The study was approved by the local ethics committee.

MPV Determination
EDTA blood samples drawn at admission of the patient were analyzed in an automated hematology analysis system (Sysmex NE 8000 autoanalyzer, Sysmex Europe GmbH, Norderstedt, Germany) (O’Malley T, Ludlam et al, 1996) that measures platelet size using aperture-impedance technology. Daily quality controls showed an intraassay coefficient of variation of 2.5% and an interassay coefficient of variation of 3.0%.

All patient samples were processed within 2 hours after venipuncture as recommended in the literature to avoid bias due to excessive platelet swelling. Previous studies reported that MPV values increase because of platelet swelling when EDTA is used as anticoagulant; however, a recent study demonstrated that this increase of platelet size amounts to approximately 0.5 fL when the analysis is performed within 2 hours after venipuncture. Probably, the reported platelet swelling in EDTA was due to different amounts of EDTA in the blood tubes. In our study, every participating hospital used the same standardized blood tubes, and all blood samples were analyzed within 2 hours after blood sampling. The range of expected values for MPV in our laboratory is 7 to 13 fL.

Determination of Outcome Variables and Statistical Analysis
The primary end points, determined by record linkage with the Austrian death registry, were overall vascular mortality and death due to ischemic heart disease. To facilitate analysis, MPV values were categorized into quintiles, with the lowest quintile serving as the reference category.

| Table 1. Patient Characteristics of the Study Population |
|---------------------------------|------------------|
| Characteristic                  | Value (n=206 554) |
| Sex (male/female)               | 89 743/116 811 (43%/57%) |
| Median age, years (IQR)         | 50 (34 to 65) |
| Median MPV, fL (IQR)            | 10.5 (9.9 to 11.2) |
| Median platelet count, g/L (IQR)| 233 (193 to 280) |
| Median observation period, years (IQR)| 4.6 (2.8 to 6.4) |
| All cause vascular mortality, no. of cases (%) | 8188 (4.0%) |
| Mortality due to ischemic heart disease, no. of cases (%) | 3987 (1.9%) |
| Mortality due to cerebrovascular disease, no. of cases (%) | 1207 (0.6%) |

The main outcome variable was all-cause vascular mortality, defined as death occurring after MPV determination and before December 31, 2004, due to any vascular disease. All-cause vascular mortality was considered present in case of ICD9 codes 390 to 459, or the corresponding ICD10 codes of groups 100 to 199. The main contributors to overall vascular mortality were death due to ischemic heart disease (48%), including myocardial infarction; nonischemic heart disease (25%), including arrhythmia and cardiomyopathy; cerebrovascular disease (14%); and other arteriosclerotic diseases (5%), including aneurysmatic disease. Mortality due to ischemic heart disease was defined as ICD9 groups 410 to 416 and ICD10 groups I20 to I25, and death due to cerebrovascular disease was defined as ICD9 groups 430 to 438 and ICD10 groups I60 to I69. Observation time was calculated in years from MPV determination until death, or until the end of the observation time (December 31, 2004) in the case of survivors. Age was calculated at the time of MPV determination.

We intended to evaluate potential interactions of MPV with age and sex concerning the cardiovascular risk as secondary outcome variables. The influence of MPV levels on our main outcome variable, all-cause mortality, was assessed in a multivariate Cox regression, adjusting for sex, age, and platelet count as possible confounders. Regression diagnostics were performed according to standard recommendations. The logistic assumption was checked for continuous variables, an analysis of residuals was performed, and global goodness of fit testing was performed using the Hosmer Lemeshow test. A 2-sided probability value <0.05 was considered statistically significant.

Unless otherwise stated, all continuous variables are given as median and IQR, and categorical variables are given as counts and percentages.

Results
A total of 206 554 individuals were included in our study (43.4% men and 56.6% women). Median age was 50 years (ranging from 18 to 100 years) at the time of analysis, and the median observation period was 4.6 years, resulting in a total of 957 890 person-years at risk (Table 1). A total of 8188 deaths due to vascular disease were recorded, of which 49.1% were male and 50.9% were female. Median age at the time of death was 77 years. In a multivariate Cox regression adjusted for sex, age, and platelet count as possible confounders, MPV was significantly associated with vascular mortality. Compared with individuals with lower MPV (lowest quintile [MPV, <8.7 fL]), hazard ratios for overall vascular mortality gradually increased to 1.5 (95% CI, 1.3 to 1.8) in the highest category (≥11.01 fL; Table 2; Figure). Furthermore, MPV was associated with ischemic heart disease as the cause of death. The relationship of MPV to ischemic heart disease was
even stronger and increased to 1.8 (95% CI: 1.4 to 2.3) in the highest category (≥11.01 fL). Interestingly, higher MPV could not be associated with higher risk of death due to cerebrovascular disease, possibly as a result of a too-low number of cases (0.6%) (Table 2).

No significant interactions of MPV and sex or age were observed in our study. Adjustment for platelet count in the Cox regression model did not alter the results.

Discussion
Several studies indicate that high MPV levels and high platelet reactivity are associated with overall vascular mortality, including myocardial infarction.11–14 Larger and hyperactive platelets accelerate the formation of an intracoronary thrombus and therefore represent a major risk factor for atherothrombosis. During the formation of an atherosclerotic plaque, eg, in a coronary artery, platelets play an essential role during the subsequent thrombus assembly, leading to myocardial infarction. MPV is a simple and accurate marker of the functional status of platelets. An increased MPV may represent a risk factor for overall vascular mortality, including myocardial infarction.

In our study, increased MPV, acting as a stand-alone risk factor, was associated with a high risk in patients experiencing an acute ischemic cardiovascular event. Patients within the highest quintile of MPV had a 1.5-fold higher hazard ratio for overall vascular mortality and an up to 1.8-fold higher risk in association with ischemic heart disease compared with patients within the lowest quintile. In comparison, the ranges of relative risks for cardiovascular disease, including coronary events, stroke, or both, are 1.4 (men) to 2.2 (women) in smokers and 1.2 (men) to 2.1 (women) in obese subjects.19

Knowledge regarding the effects of various drugs on platelet size is weak. Previous in vitro studies found no effect of aspirin on platelet size.20,21 However, it is known that clopidogrel significantly inhibits the ADP-induced increase in MPV in vitro.22 We have not found any clinical data that show an association of MPV with various platelet inhibitors.

Because patients with high MPV can easily be identified during routine hematologic analysis, early monitoring and control of all risk factors, including hypertension, hyperlipidemia, and smoking, could be initiated.

Strengths and Limitations of This Study
To our knowledge, this is the first study that has evaluated the association between MPV and vascular mortality in a large population. The chosen record linkage approach of linking laboratory data with the death registry offers the unique opportunity to evaluate the outcome of risk markers in a large population (n=206,554) over a long time period, yielding an observation period of 957,851 person-years. Because Austrian laws require that all deaths be recorded in the central death registry, this approach will allow an almost complete follow-up of all patients. The only losses might occur due to spelling errors in names, which result in faulty record linkage, or disappeared persons, who are not recorded as deaths until 50 years after the date of disappearance. Overall, we estimate that these losses affect less than 1% of the study population and are negligible for statistical analysis. In contrast to

Table 2. MPV and Causes of Death

<table>
<thead>
<tr>
<th>MPV in Quintiles</th>
<th>P Value</th>
<th>Hazard Ratio</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>All-cause vascular mortality</td>
<td>&lt;8.70&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.05</td>
<td>1.00</td>
</tr>
<tr>
<td>8.71 to 9.60</td>
<td>0.03</td>
<td>1.21</td>
<td>1.02</td>
</tr>
<tr>
<td>9.61 to 10.30</td>
<td>0.01</td>
<td>1.26</td>
<td>1.06</td>
</tr>
<tr>
<td>10.31 to 11.00</td>
<td>&lt;0.01</td>
<td>1.49</td>
<td>1.26</td>
</tr>
<tr>
<td>≥11.01</td>
<td>11.01</td>
<td>0.01</td>
<td>1.28</td>
</tr>
</tbody>
</table>

Mortality due to ischemic heart disease

<table>
<thead>
<tr>
<th>MPV in Quintiles</th>
<th>P Value</th>
<th>Hazard Ratio</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>≤8.70&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.01</td>
<td>0.92</td>
<td>0.19</td>
</tr>
<tr>
<td>8.71 to 9.60</td>
<td>0.04</td>
<td>1.02</td>
<td>0.92</td>
</tr>
<tr>
<td>9.61 to 10.30</td>
<td>0.01</td>
<td>1.06</td>
<td>1.02</td>
</tr>
<tr>
<td>10.31 to 11.00</td>
<td>&lt;0.01</td>
<td>1.38</td>
<td>1.26</td>
</tr>
<tr>
<td>≥11.01</td>
<td>11.01</td>
<td>0.01</td>
<td>0.81</td>
</tr>
</tbody>
</table>

Mortality due to cerebrovascular disease

<table>
<thead>
<tr>
<th>MPV in Quintiles</th>
<th>P Value</th>
<th>Hazard Ratio</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>≤8.70&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.01</td>
<td>0.81</td>
<td>2.05</td>
</tr>
<tr>
<td>8.71 to 9.60</td>
<td>0.04</td>
<td>0.78</td>
<td>0.92</td>
</tr>
<tr>
<td>9.61 to 10.30</td>
<td>0.01</td>
<td>0.78</td>
<td>0.92</td>
</tr>
<tr>
<td>10.31 to 11.00</td>
<td>&lt;0.01</td>
<td>1.26</td>
<td>1.02</td>
</tr>
<tr>
<td>≥11.01</td>
<td>11.01</td>
<td>0.01</td>
<td>0.81</td>
</tr>
</tbody>
</table>

Hazard ratios for MPV quintiles were calculated for the outcome variables all-cause vascular mortality, death due to ischemic heart disease, and cerebrovascular mortality in a Cox regression analysis adjusted for sex, age, and platelet counts. Interestingly, the association between MPV and higher risk remained essentially unchanged in all outcome variables.

<sup>a</sup>Reference category.
clinical diagnoses, which are subject to examiner bias and usually vary because of different diagnostic criteria, death is usually reliably recorded, and misdiaognoses rarely occur. As described by Schottenfeld et al., as well as by Karwinski and Hartveit, clinical assessments of the cause of death in death certificates lead to wrong diagnosis in 12% to 15% of all cases. Because of the high autopsy frequency (58%) in our study providing a correct diagnosis in the majority of all cases, we estimate that diagnoses leading to death are recorded correctly in ~90% of the cases.

We have insufficient data regarding the reasons for which the patients had been hospitalized. Furthermore, patients admitted to MPV determination are not a representative sample of the healthy Austrian population and might be preselected to worse outcome. Thus, based on admissions to the hospital, our study population may therefore be subject to Berkson’s bias. Unfortunately, we have no further information regarding comorbidities because all data collected from our laboratory software were linked with the Austrian death registry without individual survey of the 206,554 patients. We are well aware that several comorbidities, such as hypertension, heart failure, and atrial fibrillation, have been associated with an increased MPV; however, these might as well be independent risk factors for cardiovascular death. Because clinical information is not available for our data set, we cannot determine whether the increased MPV merely acts as risk marker or is causally involved in atherogenesis. Furthermore, information about possible previous intake of aspirin is missing. However, aspirin does not affect platelet size.

The retrospective study design is prone to bias, and we are unable to adjust for risk factors other than sex and age in the Cox regression model. Furthermore, the association of MPV with vascular mortality does not necessarily imply a causal relationship. However, this is beyond the scope of our study, because we aimed to evaluate whether MPV is a risk marker for all-cause mortality.

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Disclosures
None.

References
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