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BRIEF REPORT

Growth differentiation factor-15 for prediction of bleeding in cancer patients

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Abstract

Background: Growth differentiation factor-15 (GDF-15) is a strong predictor for bleeding in patients with atrial fibrillation, but there are no data on cardiovascular outcomes for this biomarker in cancer patients. Bleeding risk assessment is important in cancer patients when considering primary thromboprophylaxis because it is associated with an increased bleeding risk.

Objectives: To evaluate GDF-15 as predictor for bleeding events in cancer patients previously enrolled in the AVERT trial.

Patients/Methods: In this trial, 574 participants were randomized to prophylactic apixaban or placebo and followed for 180 days for venous thromboembolism, major bleeding, clinically relevant nonmajor bleeding, and any bleeding. Plasma concentrations of GDF-15 were measured centrally with the Elecsys GDF-15 commercial assay kit (Roche Diagnostics GmbH).

Results: In apixaban recipients, the area under the receiver operator characteristic curve of GDF-15 for major bleeding was 0.73 (95% confidence interval [CI], 0.44–1.00). Compared with the lowest GDF-15 tertile (<1470 ng/L), major bleeding risk was significantly higher in the highest tertile (≥2607 ng/L; hazard ratio [HR] 3.19; 95% CI, 2.41–4.22), also when adjusting for sex, age, antiplatelet use, and gastrointestinal cancer (adjusted HR 2.80; 95% CI, 1.91–4.11). GDF-15 was also significantly associated with clinically relevant nonmajor bleeding (adjusted HR 1.67; 95% CI, 1.08–2.58) and any bleeding (adjusted HR 2.12; 95% CI, 1.38–3.25).

Conclusions: Although hypothesis generating, this is the first study to show that GDF-15 predicts bleeding in cancer patients receiving thromboprophylaxis.

KEYWORDS
biomarkers, hemorrhage, neoplasms, risk, venous thromboembolism

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1 | INTRODUCTION

Growth differentiation factor 15 (GDF-15) is a cell regulatory protein that plays a role in body weight regulation and chronic inflammation in cancer patients.[1,2] In patients with atrial fibrillation, GDF-15 is a very strong predictor for bleeding,[3–9] and it was the best predictor for bleeding in the recently introduced age, biomarkers, and clinical history (ABC) score, which can be used to predict bleeding in this population.[6,7]

Data on GDF-15 as a biomarker for predicting bleeding in cancer patients are lacking. Bleeding risk assessment is important for these patients, especially when considering primary thromboprophylaxis. The benefit-risk ratio of this preventive measure in cancer patients directly depends on the individual risk of venous thromboembolism (VTE) and bleeding. Although the Khorana score could be used to select cancer patients with the highest VTE risk,[10,11] clinicians are currently not able to identify cancer patients at high bleeding risk in whom thromboprophylaxis may be harmful.[12] Among cancer patients included in the AVERT randomized trial, a prophylactic dose of apixaban was associated with a lower 6-month risk of VTE (hazard ratio [HR] 0.41; 95% confidence interval [CI], 0.26–0.65), but this was offset by an increased risk of major bleeding (HR 2.00; 95% CI, 1.01–3.95) compared with placebo.[13]

We hypothesize that the benefit-risk ratio can be improved by bleeding risk stratification based on GDF-15 levels. We therefore evaluated the association of GDF-15 as well as the ABC score with bleeding events in cancer patients in the AVERT study.

2 | METHODS

2.1 | Study population and design

This was a post hoc analysis of AVERT, a randomized, double-blind, placebo-controlled trial in which 574 ambulatory cancer patients with an intermediate-to-high VTE risk according to the Khorana score (≥2 points), included between February 2014 and April 2018, were randomized to prophylactic apixaban (2.5 mg twice daily) or placebo.[13,14] Patients were eligible when initiating a course chemotherapy for a newly diagnosed cancer or progression of a known cancer after remission. Patients were considered ineligible in case of an increased bleeding risk. Patients were followed for 180 days for the occurrence of VTE, bleeding, or mortality confirmed by blinded adjudication. Methodology and results were described in detail previously.[13,14]

2.2 | GDF-15 and ABC score

Growth differentiation factor-15 levels were measured in citrate plasma collected 1 month after enrollment in AVERT. Samples were directly processed after blood withdrawal by centrifugation at 1500*g* for 15 min at room temperature. Plasma concentrations

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ESSENTIALS

- Bleeding risk assessment tools in cancer patients on thromboprophylaxis are lacking.
- We assessed GDF-15 as a biomarker for bleeding in cancer patients on thromboprophylaxis.
- GDF-15 appeared to predict major bleeding in cancer patients starting thromboprophylaxis.
- Larger studies are needed to evaluate its clinically utility as a biomarker in clinical practice.

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of GDF-15 were measured centrally with the Elecsys GDF-15 commercial assay kit (Roche Diagnostics GmbH) by laboratory personnel unaware of study outcomes. This assay was validated previously and showed an acceptable inter- and intra-assay coefficient of variation.[15]

The ABC score includes the following items: age (in years), previous bleeding, plasma hemoglobin level (in g/L), GDF-15 level (in ng/L), and high-sensitivity troponin T (in ng/L).[6,7] Because information on prior bleeding events was not captured during the AVERT trial, this item could not be used. Therefore, we calculated a modified ABC score without prior bleeding. This modified ABC score was calculated with the regression formula provided by the developers of this score.[6,7]

2.3 | Outcomes

The primary outcome of the present analysis was ISTH-defined[16] major bleeding after 1 month up to the end of the 180-day study period. Secondary outcomes were clinically relevant nonmajor bleeding (CRNMB), defined as any bleeding not meeting the criteria for major bleeding but leading to contact with a physician, and the composite of major bleeding and CRNMB.[13]

2.4 | Statistical analysis

The area under the receiver operating characteristic curve (AUROC) was calculated using the Mann-Whitney statistic along with Wald 95% CI. Patients were then grouped based on GDF-15 level tertiles. Cox regression analysis was used to calculate the crude HR for the highest vs lowest tertile with the robust sandwich variance estimator.[17] Second, this HR was adjusted for sex, age, antiplatelet use, and gastrointestinal cancer.

The modified ABC score was evaluated continuously and dichotomously by calculating the HR for a score higher than −0.463 compared with a lower score.[6,7] Additionally, the HRs for the individual items of the modified ABC score were calculated in a multivariable model including all items.

3 | RESULTS AND DISCUSSION

Plasma samples of 470 (82%) patients were available for analysis, of whom 235 (50%) received apixaban and 235 (50%) placebo. Of the 470 patients, eight (1.7%) developed major bleeding, 23 (4.9%) CRNMB, and 30 (6.4%) any first bleeding events during the study period. One patient had both major bleeding and, subsequently, a CRNMB. Baseline characteristics are given in Table 1. The median GDF-15 plasma level in the overall cohort was 1913 (interquartile range [IQR] 1182–3309 ng/L). The median GDF-15 plasma level was 3308 (IQR 1565–10 330 ng/L) in patients with major bleeding, 2186 (IQR 1329–3522 ng/L) in patients with CRNMB, and 1898 (IQR 1164–3248 ng/L) in patients with no bleeding (Figure 1).

The AUROC of GDF-15 plasma levels in predicting major bleeding in the apixaban group was 0.73 (95% CI, 0.44–1.00). The major bleeding risk was 1.3% (one event) in the lowest GDF-15 tertile (<1470 ng/L), 1.3% (one event) in the middle tertile (1470–2607 ng/L), and 3.8% (three events) in the highest tertile (≥2607 ng/L) (Figure 2). Compared with the lowest tertile, the major bleeding risk was significantly higher in the highest GDF-15 tertile group (HR 3.19; 95% CI, 2.41–4.22), also when adjusting for sex, age, antiplatelet use, and gastrointestinal cancer (HR 2.80; 95% CI, 1.91–4.11). GDF-15 was also significantly associated with CRNMB (adjusted HR 1.67, 95% CI 1.08–2.58) and any bleeding (adjusted HR 2.12, 95% CI 1.38–3.25) (Figure 2).

The AUROC of the modified ABC score for major bleeding in the apixaban group was 0.65 (95% CI, 0.28–1.00). Of the 117 patients with a high score (higher than −0.463), three (2.6%) had major bleeding compared with two (1.7%) of 118 patients with a lower score (HR 1.56; 95% CI, 0.39–6.27). In the multivariable model including all available ABC score items, GDF-15 levels were significant predictors for CRNMB (HR 1.73 per log ng/L increase; 95% CI, 1.28–2.35) and any bleeding (HR 2.13 per log ng/L increase; 95% CI, 1.45–3.14) but not for major bleeding, whereas hemoglobin concentration was a significant predictor for all bleeding outcomes (HR 0.68 per log g/dl increase; 95% CI, 0.47–0.99 for major bleeding). Troponin T concentration and age were not significant for any of the bleeding outcomes (Table 2).

This study showed that GDF-15 may be associated with bleeding in cancer patients initiating primary thromboprophylaxis. Overall, in the AVERT trial, the major bleeding incidence was 2.1% in the apixaban arm. If we would exclude patients with a GDF-15 level in the highest tertile, this would have been 1.3%, a relative risk difference of 38%.

During the 6-month follow-up period (excluding the first month), eight major bleeding events occurred, resulting in limited statistical power. Therefore, the results of this post hoc analysis must be primarily considered as hypothesis generating. AVERT was not designed nor powered for the current analysis. Still, GDF-15 measured at 1 month after start of thromboprophylaxis appeared to be significantly associated with future bleeding events in patients on apixaban, also after adjusting for several potential confounders. Because GDF-15 levels can differ across different groups of tumors, it would be interesting to stratify according to tumor type; however, this was not possible due to the limited number of events.[18] Nonetheless, we were able to adjust for gastrointestinal cancer, which is the group of tumors most associated with bleeding in cancer patients using direct oral anticoagulants.[19] Additionally, data on several cardiovascular risk factors, such as coronary artery disease or left ventricular dysfunction, which are known to be associated with increased GDF-15 values, were not routinely recorded in the AVERT trial. Therefore, the variables could not be included in the analysis. Future, larger studies should take these risk factors into account when assessing GDF-15 in cancer patients on thromboprophylaxis. Evaluation of the association between GDF-15 and VTE was beyond the scope of the present analysis. We were unable to use the item "previous bleeding" in the ABC score, which may have decreased the performance of the score. Discrimination of GDF-15 appeared to be higher for patients randomized to apixaban than for those randomized to placebo, although not statistically significant (difference in AUROC 0.24, 95% CI −0.28 to 0.76, p = .26).

Notably, because baseline samples were not available for this analysis, samples at 1 month after inclusion were used. As a consequence, bleeding events occurring before the 1-month sample were not included, hampering the generalizability of the results to

TABLE 1 Baseline characteristics of patients in the AVERT trial using apixaban

	Any Bleeding (n = 17)	No Bleeding (n = 218)
Age (years) (SD)	62.8 (17.2)	60.2 (11.6)
Male sex (%)	8 (47.1)	87 (39.9)
Weight (kg) (mean, SD)	80.2 (21.1)	81.2 (22.9)
Creatinine clearance ≤50 ml/min (%)	1 (5.9)	9 (4.1)
Tumor type (%)		
Brain	-	12 (5.5)
Lung	1 (5.9)	23 (10.6)
Testicular	-	1 (0.5)
Stomach	4 (23.5)	17 (7.8)
Pancreatic	2 (11.8)	24 (11.0)
Lymphoma	3 (17.7)	55 (25.2)
Myeloma	1 (5.9)	6 (2.8)
Gynecologic	5 (29.4)	62 (28.4)
Colon	-	1 (0.5)
Other	1 (5.9)	17 (7.8)
Body mass index ≥35 (%)	3 (17.7)	61 (28.0)
Leukocyte count >11 000/mm³ (%)	3 (17.7)	65 (29.8)
Hemoglobin <10 g/dl	4 (23.5)	50 (22.9)
Platelet count ≥350 000/mm³ (%)	7 (41.2)	89 (40.8)
Antiplatelet medication (%)	5 (29.4)	51 (23.4)

Abbreviation: SD, standard deviation.

FIGURE 1 Dot plot of GDF-15 values in patients with no bleeding, major bleeding, and clinically relevant nonmajor bleeding. Abbreviations: CRNMB, clinically relevant nonmajor bleeding; GDF, growth differentiation factor
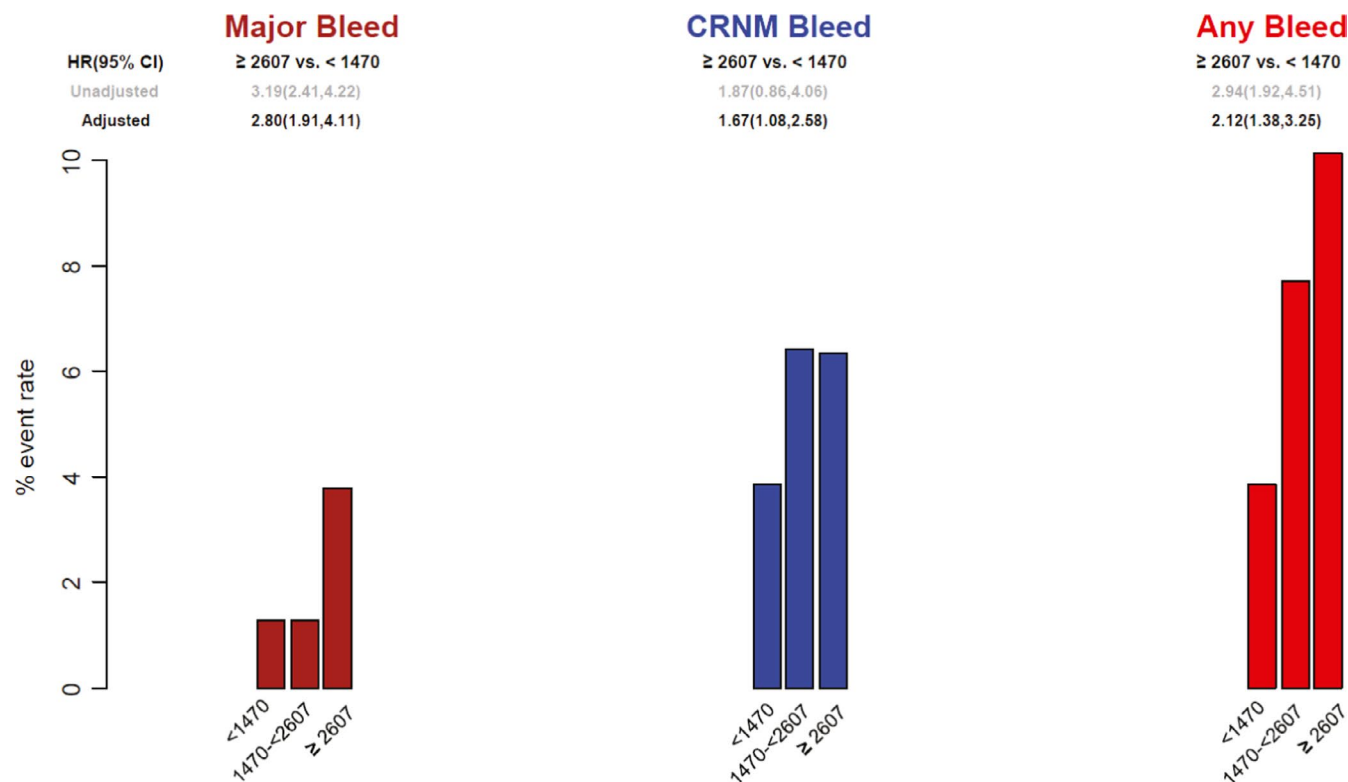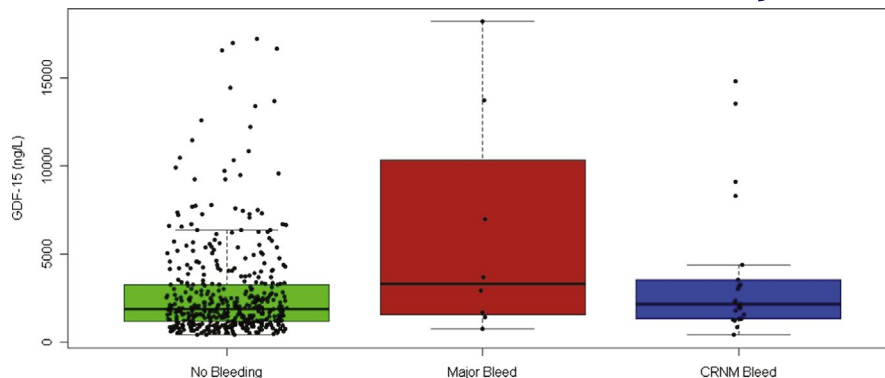

FIGURE 2 Bleeding events during the 180-day study period in cancer patients using apixaban by GDF-15 plasma level tertile in ng/L. *Hazard ratio for bleeding events adjusted for sex, age, antiplatelet use, and gastrointestinal cancer. Abbreviations: GDF, growth differentiation factor; HR, hazard ratio

this specific period. Although GDF-15 levels have been reported to change over time in patients experiencing cardiovascular events,[20] another large prospective cohort study of 813 community-dwelling elderly individuals showed that GDF-15 levels at baseline and at 5-year follow-up were strongly correlated ($r = 0.70$; $p < .001$) and only changed by 11%, from 1102 ng/L to 1238 ng/L.[21] These data suggest that there is little change in GDF-15 levels in individuals with no recent cardiovascular events, although we do not know to what extent GDF-15 levels at 1 month correlated with those at baseline in the present study.

This study indicates that GDF-15 potentially is a predictive biomarker for bleeding in cancer patients using apixaban for thromboprophylaxis. Additional larger studies are needed to confirm these results and prospectively evaluate its clinically utility as a biomarker, as well as the full ABC bleeding-risk score in clinical practice.

CONFLICT OF INTEREST

TABLE 2 Area under the receiver operator curve and hazard ratio[a] for major bleeding, CRNMB, and any bleeding

	Major Bleeding	CRNMB	Any Bleeding
Apixaban Group (N = 235)	(N = 5)	(N = 13)	(N = 17)[b]
GDF-15			
AUROC	0.73 (0.44–1.00)	0.61 (0.47–0.76)	0.67 (0.54–0.80)
HR high vs low tertile (unadjusted)	3.19 (2.41–4.22)	1.87 (0.86–4.06)	2.94 (1.92–4.51)
HR high vs low tertile (adjusted)	2.80 (1.91–4.11)	1.67 (1.08–2.58)	2.12 (1.38–3.25)
Total ABC score			
AUROC	0.65 (0.28–1.00)	0.60 (0.41–0.80)	0.65 (0.48–0.82)
HR (high vs low score)	1.56 (0.39–6.27)	1.74 (0.45–6.79)	1.96 (0.92–4.24)
ABC score individual components			
HR increase in age by 1 year	0.95 (0.85–1.07)	1.00 (0.92–1.09)	1.01 (0.93–1.09)
HR increase in log(troponin) by 1	1.17 (0.18–7.55)	1.07 (0.55–2.07)	1.04 (0.60–1.78)
HR increase in log(gdf15) by 1	3.62 (0.92–14.23)	1.73 (1.28–2.35)	2.13 (1.45–3.14)
HR increase in HGB by 1	0.68 (0.47–0.99)	0.79 (0.71–0.88)	0.79 (0.72–0.87)
Placebo group (N = 235)	**(N = 3)**	**(N = 10)**	**(N = 13)**
GDF-15			
AUROC	0.48 (0.05–0.91)	0.51 (0.32–0.70)	0.50 (0.33–0.67)
HR high vs low tertile (unadjusted)	2.31 (0.61–8.67)	1.10 (0.56–2.18)	1.35 (0.74–2.46)
HR high vs low tertile (adjusted)	1.34 (0.50–3.57)	1.38 (0.48–3.99)	1.27 (0.58–2.77)
Total ABC score			
AUROC	0.50 (0.05–0.94)	0.56 (0.37–0.74)	0.54 (0.37–0.71)
HR (high vs low score)	2.09 (0.58–7.58)	0.68 (0.28–1.68)	0.88 (0.40–1.93)
ABC score individual components			
HR increase in age by 1 year	0.97 (0.92–1.02)	1.06 (1.01–1.12)	1.04 (1.01–1.08)
HR increase in log(troponin) by 1	2.16 (0.32–14.77)	0.19 (0.12–0.29)	0.33 (0.20–0.53)
HR increase in log(gdf15) by 1	0.74 (0.23–2.37)	1.62 (1.05–2.52)	1.28 (0.79–2.08)

(Continues)

TABLE 2 (Continued)

Placebo group (N = 235)	(N = 3)	(N = 10)	(N = 13)
HR increase in HGB by 1	1.06 (0.89–1.27)	0.86 (0.59–1.24)	0.94 (0.73–1.21)
All patients (N = 470)	**(N = 8)**	**(N = 23)**	**(N = 30)[b]**
GDF-15			
AUROC	0.66 (0.43–0.89)	0.57 (0.45–0.68)	0.60 (0.49–0.71)
HR high vs low tertile (unadjusted)	5.51 (3.52–8.63)	1.69 (1.01–2.85)	2.26 (1.49–3.43)
HR high vs low tertile (adjusted)	4.55 (2.65–7.81)	1.75 (0.83–3.71)	1.85 (1.02–3.36)
ABC score			
AUROC	0.60 (0.33–0.86)	0.53 (0.39–0.67)	0.57 (0.44–0.69)
HR (high vs low score)	1.74 (0.76–3.98)	1.15 (0.50–2.66)	1.38 (0.78–2.43)
ABC score individual components			
HR increase in age by 1 year	0.96 (0.90–1.03)	1.02 (0.97–1.08)	1.02 (0.97–1.08)
HR increase in log(troponin) by 1	1.15 (0.27–4.90)	0.60 (0.32–1.12)	0.67 (0.42–1.08)
HR increase in log(gdf15) by 1	2.23 (1.42–3.49)	1.67 (1.30–2.16)	1.78 (1.47–2.15)
HR increase in HGB by 1	0.86 (0.74–0.99)	0.84 (0.70–1.01)	0.86 (0.75–1.00)

Abbreviations: AUROC, area under the receiver operator curve; CI, confidence interval; CNMB, clinically relevant nonmajor bleeding; GDF-15, growth-differentiation factor-15; HGB, hemoglobin; HR, hazard ratio.

[a]Hazard ratio for bleeding events adjusted for sex, age, antiplatelet use, and gastrointestinal cancer.

[b]One patient had both major bleeding and, subsequently, clinically relevant nonmajor bleeding. The HR of any bleeding was calculated using only the major bleeding, which occurred first.

AUTHOR CONTRIBUTIONS

Pieter Willem Kamphuisen, Nick van Es, and Frits I. Mulder were responsible for initiation and concept of the study. Ranjeeta Mallick performed the statistical analysis. Frits I. Mulder and Floris T. M. Bosch wrote the first draft. All authors critically revised the paper for important intellectual content, approved the final version, and agree with the submission.

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