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Clinical Research Article

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Impact of Adrenal Function on Hemostasis/ Endothelial Function in Patients Undergoing Surgery

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Abbreviations: ACTH, adrenocorticotropic hormone; DHEA-S, dehydroepiandrosterone sulfate; dHPA, deficient hypothalamic-pituitary-adrenal; FVIII, factor VIII; HPA, hypothalamic-pituitary-adrenal; nHPA, normal hypothalamic-pituitary-adrenal; PAI-1, plasminogen activator inhibitor-1; sVCAM-1, soluble vascular cell adhesion molecule-1; vWF, von Willebrand factor antigen

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Abstract

Context: Glucocorticoids regulate hemostatic and endothelial function, and they are critical for adaptive functions during surgery. No data regarding the impact of adrenal function on hemostasis and endothelial function in the perioperative setting are available.
Objective: We assessed the association of adrenal response to adrenocorticotropic hormone (ACTH) and markers of endothelial/hemostatic function in surgical patients.
Methods: This prospective observational study, conducted at a tertiary care hospital, included 60 patients (35 male/25 female) undergoing abdominal surgery. Adrenal function was evaluated by low-dose ACTH stimulation test on the day before, during, and the day after surgery. According to their stimulated cortisol level (cutoff ≥ 500 nmol/L), patients were classified as having normal hypothalamic-pituitary-adrenal (HPA)-axis function (nHPA) or deficient HPA-axis function (dHPA). Parameters of endothelial function (soluble

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vascular cell adhesion molecule-1, thrombomodulin) and hemostasis (fibrinogen, von Willebrand factor antigen, factor VIII [FVIII]) were measured during surgery.
Results: Twenty-one patients had dHPA and 39 had nHPA. Compared with nHPA, patients with dHPA had significantly lower peak cortisol before (median 568 vs 425 nmol/L, $P < 0.001$) and during (693 vs 544 nmol/L, $P < 0.001$) surgery and lower postoperative hemoglobin levels (116 g/L vs 105 g/L, $P = 0.049$). FVIII was significantly reduced in patients with dHPA in uni- and multivariable analyses; other factors displayed no significant differences. Coagulation factors/endothelial markers changed progressively in relation to stimulated cortisol levels and showed a turning point at cortisol levels between 500 and 600 nmol/L.
Conclusions: Patients with dHPA undergoing abdominal surgery demonstrate impaired hemostasis which can translate into excessive blood loss.

Key Words: Adrenal insufficiency, hemostasis, endothelial function, abdominal surgery

Surgery represents a major stress to the body, leading to a variety of homeostatic responses to defend and protect the organism in this setting. Apart from effects on volume, electrolyte, and pH balance, surgical interventions may have an impact on hemostasis (ie, by increasing coagulation proteins and platelet reactivity and by decreasing coagulation inhibitors and fibrinolysis) [1-3] and arterial endothelial function [4, 5]. Some of these adaptive reactions are mediated through stress hormones (ie, glucocorticoids and catecholamines) [6].

Glucocorticoids are critical for vascular function/reactivity and they exert a permissive function on catecholamine release and action during the stress response [7], effects that are promoted through genomic and nongenomic pathways [8, 9]. Glucocorticoids act on the coagulation system in various ways. Their impact is mainly known from states with endogenous cortisol excess or exogenous therapeutic administration of glucocorticoids [10]. Glucocorticoids promote an increase in clotting factors (ie, factor VIII [FVIII], von Willebrand factor antigen [vWF]) [11], thrombin/antithrombin complexes, plasminogen activator inhibitor-1 [PAI-1] and fibrinogen levels [12].

Apart from their well-known effects on the vasculature, catecholamines also stimulate clotting factors (ie, FVIII and vWF) and fibrinolytic molecules (ie, tissue plasminogen activator) [13] and they represent important cofactors for platelet activation/aggregation [14, 15]. Adrenocortical and adrenomedullary function are closely linked together by a complex system of paracrine regulation [16]. For example, glucocorticoids induce enzymes for catecholamine biosynthesis and stimulate the release of catecholamines [17].

Therefore, the integrity of the adrenocortical and adrenomedullary unit in the form of an adequate response of the hypothalamic-pituitary-adrenal (HPA) axis to stress (ie, the rapid increase of the cortisol secretion and a sufficient catecholamine secretion capacity) is critical to maintain volume/electrolyte balance and to control inflammatory cascades, vascular tone, endothelial function, and hemostatic functionality in the peri- and postoperative period.

Surgery is a potent activator of the HPA axis [18, 19], with the degree of the activation being dependent on several factors, including severity of surgical stress [20], administered medications [21-23], and type of anesthesia [24]. Untreated adrenocortical insufficiency in the perioperative setting is associated with increased morbidity and mortality, mainly due to arterial hypotension and volume-refractory shock [25, 26]. Furthermore, the perioperative regulation of the volume balance and the amount of blood loss correlate with the adequacy of the adrenal function. Patients with adrenal insufficiency undergoing coronary artery bypass graft procedures show altered perioperative outcomes, such as increased blood loss and impaired volume balance [27].

Currently, no data are available regarding the impact of adrenal function on hemostasis and platelet function in the perioperative setting. Therefore, the aim of this study was to evaluate the association of the adrenal response to adrenocorticotropic hormone (ACTH) and markers of endothelial and hemostatic function in patients undergoing abdominal surgery. Based on the assumption that patients with deficient HPA-axis function (dHPA) have impaired adrenal catecholamine secretion, we hypothesize that

a) Glucocorticoid and catecholamine deficiency are associated with lower levels of coagulation factors (ie, FVIII and vWF) and probably lower levels of markers of endothelial function.
b) Combined glucocorticoid and catecholamine deficiency may have the profoundest impact on coagulation markers whose secretion is dependent on glucocorticoid *and* catecholamine action (FVIII and vWF).

Methods

Participants

This prospective observational study was conducted at the Luzerner Kantonsspital, a large urban hospital. Consecutive

patients scheduled for various types of abdominal surgery were recruited (Fig. 1). We excluded patients with a history of glucocorticoid treatment (topical, inhaled, intraarticular, or systemic application during the last 6 months), endocrine diseases (ie, Cushing disease, thyroid dysfunction), acute illness, and induction of anesthesia with etomidate.

The study conformed to the declaration of Helsinki and was specifically approved by the local ethics committee (Ethikkommission Nordwest- und Zentralschweiz). Written informed consent was obtained from all patients.

Study Procedures

The adrenal function was evaluated by low-dose (1 µg) ACTH stimulation test in all patients on the day before surgery (8 AM), during surgery (30 minutes after the first incision, the time of the stimulation test depending from the time of scheduled surgery) and on the first postoperative day (8 AM). Dehydroepiandrosterone sulfate (DHEA-S) as an additional parameter for adrenal function was measured before surgery. Parameters of endothelial function and hemostasis were measured during surgery (30 minutes after the first incision).

Study Measurements

Evaluation of adrenal function: low-dose ACTH stimulation test, measurement of DHEA-S

For the low-dose (1 µg) ACTH stimulation test, a bolus intravenous injection of 1 µg (1–24)-corticotropin (tetracosactrin, Synacthen, Novartis Pharma, Berne, Switzerland) was administered. One vial of 250µg tetracosactrin was diluted in sterile saline solution to a concentration of 1µg/mL, filtered in plastic syringes, and stored at 4 °C as described elsewhere [28]. Blood was taken immediately before (basal cortisol level) and 30 minutes after (stimulated cortisol level) injection of the corticotropin bolus. A stimulated cortisol level of ≥500 nmol/L [29] was defined as a normal adrenal response to corticotropin and normal HPA-axis function (nHPA).

Parameters of endothelial function and hemostasis: soluble vascular cell adhesion molecule-1, thrombomodulin, fibrinogen, vWF, FVIII

Soluble vascular cell adhesion molecule-1 (sVCAM-1) and thrombomodulin were measured as markers of endothelial function. Fibrinogen, vWF, FVIII, and thrombocytes were measured to evaluate hemostasis.

Statistical Analysis

Baseline characteristics and laboratory measurements were analyzed descriptively using medians and interquartile ranges for continuous variables or numbers and percentages for categorical variables. Comparisons were performed between groups of persons with nHPA and dHPA, respectively, using the Kruskal-Wallis (continuous variables) or Chi-square (categorical variables) tests. Of note, 3 persons had missing measurements of preoperative hemoglobin (all in the nHPA group, and there were 14 postoperative hemoglobin measurements (6 in the dHPA group and 8 in the

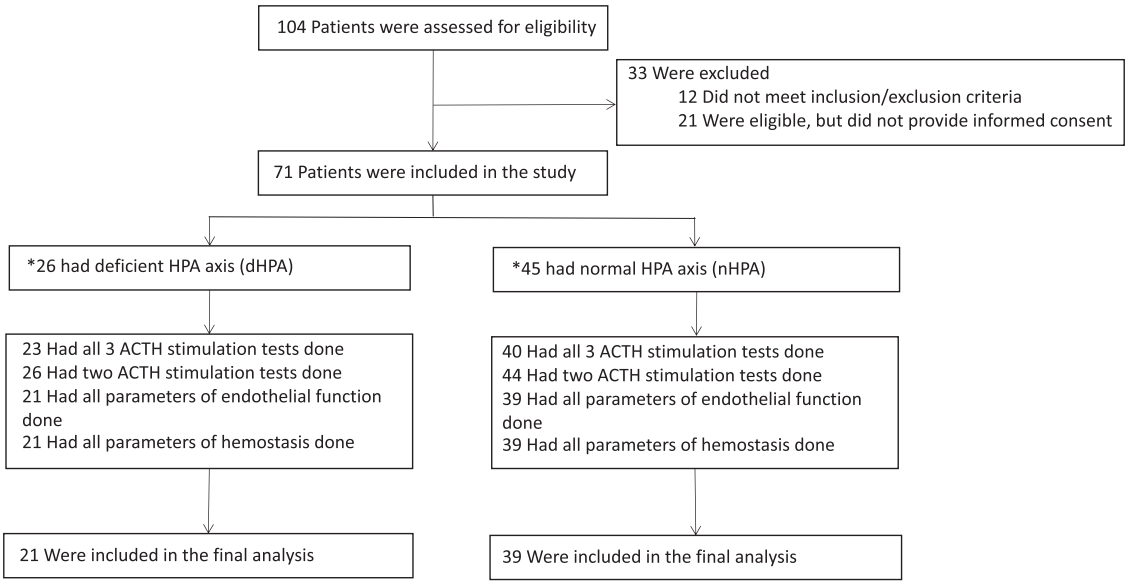

Figure 1. Patient flow chart depicting the number of patients included in the study and in the final analysis, respectively.

nHPA group). Persons with missing laboratory parameters were excluded from the respective univariable analysis.

Multivariable linear regression analyses were performed to test for differences between the nHPA and dHPA groups, using the parameters of endothelial function and hemostasis as outcomes, adjusted for a priori determined confounders of age, sex, weight, intraoperative volume balance, and intubation time. Because some persons had missing information for weight (deficient group n = 1, normal group n = 3), intubation time (deficient group n = 2, normal group n = 5), and volume balance (deficient group n = 4, normal group n = 7), the missing values were replaced by the overall means of the respective variables in order to maintain them in regression analyses. Regression assumptions were visually checked using residual-versus-fitted plots (not shown).

Additionally, the binary classification of preoperative, stimulated cortisol into deficient and normal in the main analysis was scrutinized in an exploratory analysis. Based on the multivariable linear regression models described above, the "deficient or normal" groups were replaced by a restricted cubic spline transformation of preoperative stimulated cortisol levels with 3 knots. Next, we plotted model-predicted coagulation parameters as a function of stimulated cortisol values, without making any assumptions regarding the shape of the association (plotting marginal effects with all other values fixed at the mean).

Analyses were performed using Stata 13 (Stata Corp., College Station, TX, USA). Statistical significance was defined as P values < 0.05 (2-tailed). We did not adjust the analysis for multiple comparisons.

Results

Baseline Characteristics

A total of 60 patients (35 male / 25 female) were included in the study; median age was 66 years. A deficient adrenal response to ACTH on the day before surgery was diagnosed in 21 patients (35%, dHPA group), whereas 39 patients (65%, nHPA group) had intact adrenocortical function. The 2 groups were well balanced with respect to sex, age, surgery type and characteristics (ie, open vs laparoscopic procedure, elective vs urgent procedure), use of intraoperative noradrenalin and pre-/postoperative systolic blood pressure. However, the dHPA group tended to be heavier, have a shorter intubation time, and a higher blood volume balance, although differences did not reach statistical significance (Table 1).

Impact of Adrenal Function on Hemostasis

Univariable analyses of cortisol levels indicated that—compared to patients with normal HPA-axis function—patients with deficient HPA-axis function had a significantly lower peak cortisol level after stimulation with ACTH before (median 425 vs 568 nmol/L, $P < 0.001$) and during (median 544 vs 693 nmol/L, $P < 0.001$) surgery. Stimulated cortisol levels in the postoperative setting also tended to be higher in the nHPA-axis group, but the difference did not reach statistical significance (median 566 vs 650 nmol/L, $P = 0.066$) (Fig. 2/Table 2).

Patients in both groups had similar preoperative hemoglobin levels (median of 139 g/L in both groups), but patients in the dHPA group had lower postoperative levels (median 105 vs 116 g/L in the deficient and normal groups, respectively, $P = 0.049$) suggesting greater blood loss (Table 3). The higher operative blood loss is also reflected by the nominally higher amount of infused solutions in the dHPA group (median 2500 mL vs 2350 mL in the normal group, $P = 0.136$) (Table 1).

The univariable analysis of biochemical and coagulation parameters during surgery revealed that patients with deficient HPA-axis function had significantly reduced concentrations of FVIII (median 125% vs 184% in the normal HPA-axis group, $P = 0.000$). In contrast, there were no significant group differences in concentrations of vWF, thrombomodulin, sVCAM-1, and fibrinogen during surgery (Table 3). FVIII was also significantly lower in the multivariable regression analysis adjusted for age, sex, weight, intraoperative volume balance, and intubation time (Fig. 3).

The exploratory, restricted cubic spline analyses suggest that most coagulation parameters do indeed show a turning point around the cutoff at around 500 (indicated by black line in Fig. 4) to 600 nmol/L, thus validating our initially chosen cutoff for deficient vs normal cortisol. The only exception was thrombomodulin, which exhibited a linear association with stimulated preoperative cortisol.

Discussion

An insufficient response of plasma cortisol to ACTH is common in patients undergoing abdominal surgery and the rate in our study is comparable to other studies [27]. However, the reasons for this high proportion of patients with a deficient HPA-axis function remain unclear. We have excluded patients with possible causes (ie, history of treatment with glucocorticoids or induction of general anesthesia with etomidate) of primary or secondary/tertiary adrenal insufficiency. There seems to be a dynamic in cortisol secretion patterns: Patients with deficient HPA-axis function had significantly lower stimulated cortisol levels before and during surgery.

Table 1. Baseline characteristics

Parameter	dHPA	nHPA	*P* value
Male, n (%)	13 (61.9%)	22 (56.41%)	0.681
Caucasians, n (%)	21 (100%)	37 (94%)	0.924
Age, years	66 [59; 72]	69 [53; 76]	0.914
Weight, kg	78 [67; 85]	73 [64; 80]	0.118
BMI, kg/m^2	26.7 [24.9; 27.6]	25.9 [23.3; 27.0]	0.068
Characteristic of surgery			
Laparoscopic, n (%)	13 (62%)	24 (61%)	0.977
Open surgery, n (%)	8 (38%)	15 (39%)	0.977
Elective surgery, n (%)	18 (85%)	34 (88%)	0.873
Emergency operation, n (%)	3 (14%)	5 (12%)	0.873
Type of abdominal surgery			
Cholecystectomy lap.-scop./open	4/1	6/1	
Sigma resection	0	7	
Ileostomy reversal	1	5	
Hernioplasty	6	6	
Rectum amputation	3	2	
Hemicolectomy	3	4	
Gastrectomy	2	2	
Esophagus resection	0	1	
Pancreas resection	0	2	
Adhesiolysis	1	3	
Preoperative medication (n)			
Alpha blocker	0	1	
Beta blocker	2	5	
Calcium antagonists	2	3	
ACE-inhibitors/sartans	3	6	
NSAID	1	4	
Antidepressants	1	1	
ASA class I; II; III; IV (n)	7; 10; 3; 1	13; 16; 7; 2	
Perioperative anesthesia drugs (n)			
Etomidate	0	0	
Propofol	16	34	
Thiopentone	5	5	
Isoflurane in air	21	39	
Volume balance (mL)	2500 [2250; 3000]	2350 [1200; 3450]	0.136
Intubation time (min)	150 [115; 245]	202.50 [125; 282]	0.194
Use of intraoperative noradrenaline (n)	2	5	0.704
Systolic blood pressure preop. (mmHg)	131 [122; 142]	124 [110; 131]	0.061
Diastolic blood pressure preop. (mmHg)	78 [70; 94]	70 [63; 80]	0.007
Systolic blood pressure postop. (mmHg)	130 [117; 145]	130 [120; 140]	0.729
Diastolic blood pressure postop. (mmHg)	80 [76; 84]	80 [70; 84]	0.580

Baseline characteristics in patients undergoing abdominal surgery with deficient (n = 21, dHPA) and normal HPA-axis function (n = 39, nHPA). Apart from gender, data are represented as median [IQR].

Abbreviations: ACE, angiotensin converting enzyme; ASA, American Society of Anesthesiologists; BMI, body mass index; dHPA, deficient hypothalamic-pituitary-adrenal; IQR, interquartile range; nHPA, normal hypothalamic-pituitary-adrenal; NSAID, nonsteroidal anti-inflammatory drug; postop., postoperative; preop., preoperative.

It could be assumed that patients with deficient HPA-axis had subnormal stimulated cortisol levels only by chance or confounding factors (ie, altered binding protein levels) interfered with the cortisol assay leading to false low values. However, patients with deficient HPA-axis function demonstrated (1) distinct alterations in the coagulation system, even after correction for multiple confounders (age, sex, weight, intraoperative volume balance, and intubation time) and (2) coagulation factors and endothelial markers changed progressively in relation to stimulated serum cortisol levels (as could be shown in the restricted cubic spline transformation). Most of these parameters show a turning

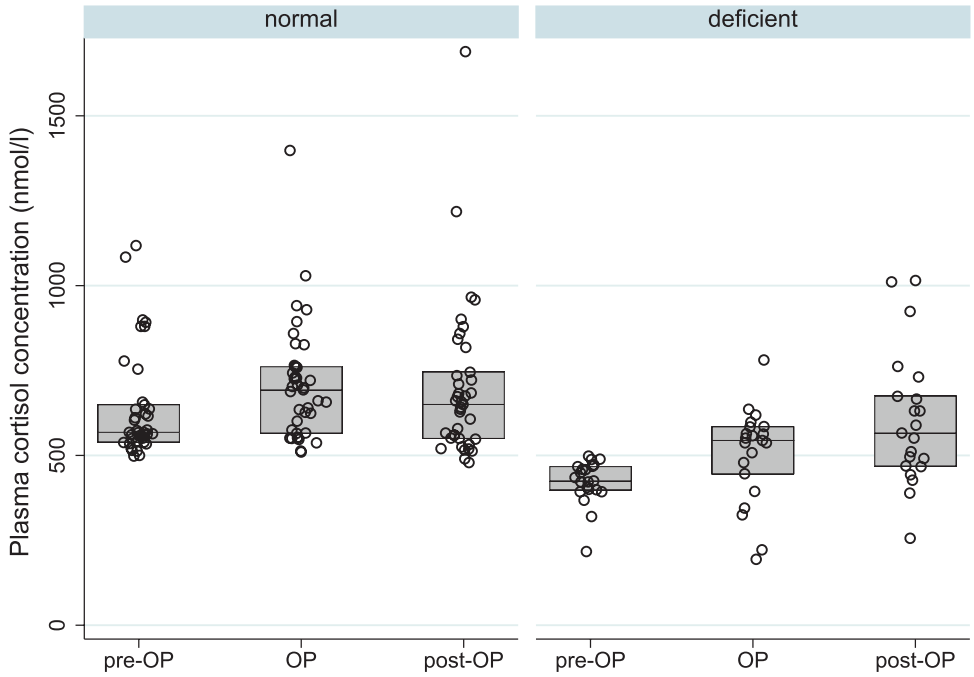

Figure 2. Stimulated plasma cortisol levels that were evaluated at different time-points (preoperatively, during surgery, postoperatively) in patients with normal and deficient HPA-axis function.

Table 2. Cortisol levels

Parameter	dHPA	nHPA	*P* value
Preop. basal	209 [164; 226]	258 [222; 389]	0.005
Preop. stimulated	425 [399; 467]	568 [540; 649]	0.000
During OP basal	314 [182; 361]	350 [250; 467]	0.114
During OP stimulated	544 [446; 584]	693 [566; 761]	0.000
Postop. basal	406 [337; 465]	393 [278; 492]	0.822
Postop. stimulated	566 [469; 674]	650 [550; 745]	0.066

Cortisol levels [nmol/L] basal and after stimulation with ACTH before, during and after surgery in patients with deficient (n = 21) and normal HPA-axis function (n = 39). Data are represented as median [IQR].

Abbreviations: ACTH, adrenocorticotropic hormone; dHPA, deficient hypothalamic-pituitary-adrenal; IQR, interquartile range; nHPA, normal hypothalamic-pituitary-adrenal; OP, operative procedure; postop., postoperative; preop., preoperative.

point at cortisol levels between 500 and 600 nmol/L which corresponds to the generally accepted cutoff for the classification of HPA-function (normal vs deficient). With these points taken together, we conclude that these patients had a clinically existent and relevant dysfunction of the adrenal gland.

In the postoperative setting, levels of basal and stimulated cortisol demonstrated no difference between the 2 groups. Therefore, the deficiency of the adrenal function in the setting of acute illness/surgery could be a temporary phenomenon reflecting a delayed response to stress with downstream effects on the hemostasis system. This finding is supported by the fact that levels of DHEA-S, which are a sensitive indicator for the presence of chronic adrenal insufficiency [30-32], were not different in patients with nHPA and dHPA.

Our main finding is that the lack of an adequate adrenal response is associated with alterations in the coagulation system: patients with a deficient HPA-axis displayed significantly lower levels of FVIII during surgery, whereas other parameters remained unchanged (vWF antigen, thrombomodulin, and sVCAM-1).

On a clinical basis, these alterations in hemostasis translated to an increased intraoperative blood loss, probably an increased amount of infusions, and a lower postoperative hemoglobin level. By analogy, an increased rate of bleeding complications is seen in patient with congenital (hemophilia A) and acquired FVIII deficiency, and, historically, epinephrine, although not norepinephrine, was used to stimulate FVIII clotting activity in patients with hemophilia [33]. Reduced permissive action of cortisol on catecholamine release during surgery could thus be one explanation for

Table 3. Biochemical and coagulation parameters

Parameter	dHPA	nHPA	*P* value
Hemoglobin preoperative (g/L)	139 [125; 146]	139 [132; 143.50]	0.843
Hemoglobin postoperative (g/L)	105 [93; 114]	116 [105; 128]	0.049
Thrombocytes (g/L)	221 [191; 258]	213 [179; 284]	0.110
DHEA-S (µmol/L) preopoperative	1.73 [0.97; 3.49]	1.78 [0.99; 3.70]	0.914
High-sensitive CRP (mg/L) preoperative	0.77 [0.43; 2.24]	1.58 [0.89; 3.71]	0.079
Albumin (g/L) preoperative	29 [23; 32]	29 [24; 33]	0.658
Fibrinogen Clauss (g/L) during surgery	2.51 [1.92; 2.76]	2.31 [2.07; 2.96]	0.895
Factor VIII (%) during surgery	125 [98; 140]	184 [135; 244]	0.000
vWF antigen (%) during surgery	115 [95; 148]	139 [97; 177]	0.215
sVCAM1 (ng/mL) during surgery	350 [270; 431]	377 [299; 473]	0.577
Thrombomodulin (ng/mL) during surgery	29.20 [25.30; 34.60]	28.20 [24; 32.50]	0.271

Biochemical and coagulation parameters in patients with deficient (n = 21, dHPA) and normal HPA-axis function (n = 39, nHPA). Data are represented as median [IQR].

Abbreviations: CRP, C-reactive protein; DHEA-S, dehydroepiandrosterone sulfate; sVCAM-1, soluble vascular cell adhesion molecule-1; vWF, von Willebrand factor antigen.

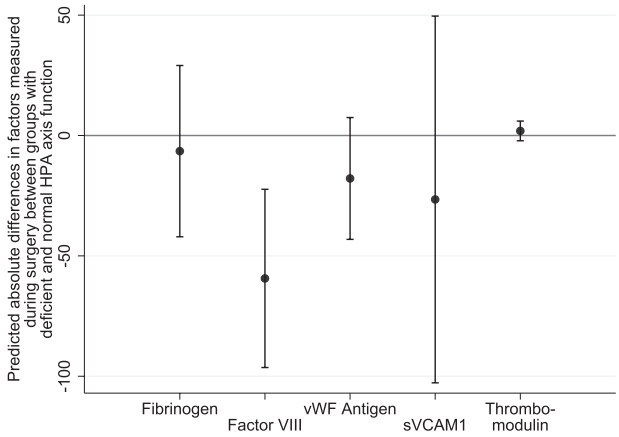

Figure 3. Multivariable regression analysis (adjusted for age, gender, weight, volume balance, and intubation time) displaying differences in clotting factors and endothelial markers measured during surgery between groups with deficient and normal HPA-axis function. Data are displayed as model-derived predicted absolute differences and 95% confidence intervals.

why patients with dHPA mounted a relatively lower FVIII response. FVIII increase during stress is mediated by beta-adrenergic receptors [34], which are also involved in vWF antigen release from endothelial cells during the stress response [35]. Indeed, compared to patients with nHPA, those with dHPA showed also lower absolute levels of VWF antigen. However, FVIII seems to show a relatively stronger stress response than VWF antigen [36], which together with limited statistical power, likely prevented the group difference in VWF antigen from becoming statistically significant. In turn, to our knowledge, it is currently unknown to what extent and by which mechanisms catecholamines are involved in the human stress response of sVCAM-1 and thrombomodulin, which may explain the absence of a group difference.

The hemostasis/coagulation system is a complex and balanced system simultaneously protecting the body against excessive blood loss and preventing hypercoagulability and thrombosis. It involves several key components: endothelial function, cellular factors (ie, platelet aggregation and activation), a coagulation cascade (ie, vWF, coagulation factors) that itself is regulated by proteins (protein C, protein S, antithrombin), fibrinolysis, and inhibition of fibrinolysis respectively (ie, by PAI-1). The crosstalk between the endocrine system and hemostasis/coagulation is well known and supported by a plethora of data from ex vivo and in vivo studies in healthy persons, patients with endocrine diseases, and patients taking exogenous glucocorticoids [37]. For example, acquired vWF disease and decreased FVIII levels can occur with hypothyroidism [38] and are described in patients with hypopituitarism [39].

Endogenous Cushing syndrome—the "pathologic counterpart" of adrenal insufficiency—is associated with a hypercoagulable state and increased risk of thromboembolic complications [40, 41]. Endogenous Cushing syndrome affects every component of the hemostatic system: increased platelet count [42, 43] and activation/aggregation [44], increased thrombin/antithrombin complexes [12, 45], and increased PAI-1 and fibrinogen levels [11, 12, 43, 46]. Alterations in the coagulation cascade have also been described. In all studies, FVIII consistently seems to be elevated in endogenous Cushing syndrome [10, 11, 46-50]. There is an increase in vWF levels [11, 12, 45, 47, 48, 51, 52] in patients with endogenous Cushing syndrome, as well as a positive correlation between serum cortisol levels and vWF concentrations [52], and patients present usually with large vWF multimers [48].

Elevated levels of glucocorticoids stimulate the endothelial production of vWF, probably by a direct effect of

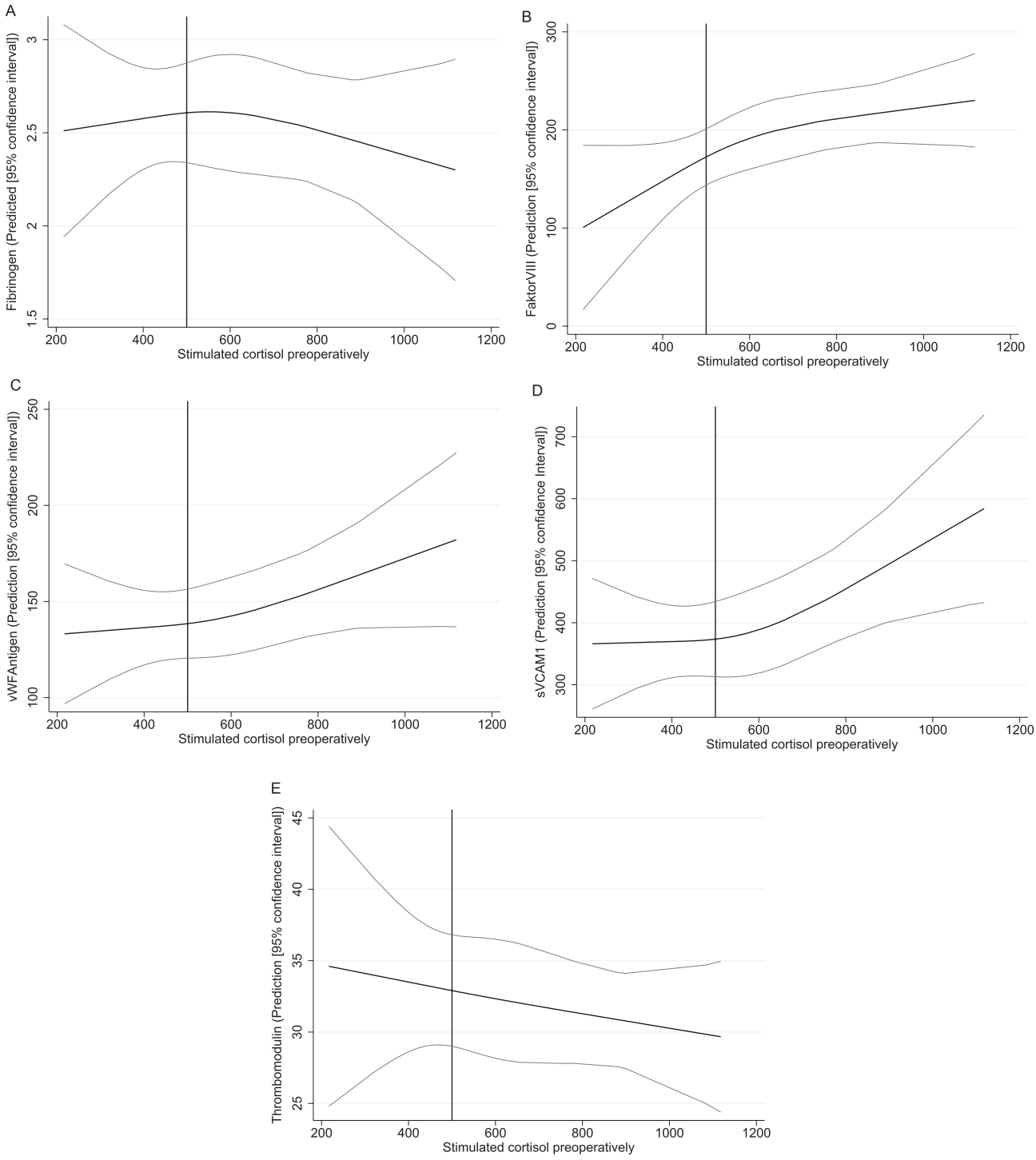

Figure 4. Association of clotting factors and endothelial markers with preoperative stimulated cortisol, based on multivariable linear regression models including cortisol as a restricted cubic spline transformation. The figures show marginal effects, that is, predicted clotting factors when keeping all other variables included in the model constant at their respective overall average values. Stimulated cortisol preoperatively in nmol/L, the vertical black line indicates cutoff for stimulated serum cortisol at 500 nmol/L. **A,** Fibrinogen; **B,** Factor VIII; **C,** vWF antigen; **D,** sVCAM-1; **E,** Thro mbomodulin.

glucocorticoids on vWF gene expression [53]. Higher vWF levels promote a rise in FVIII [54] in patients with Cushing syndrome. Thereby treatment of endogenous cortisol excess is associated with a decrease and normalization of vWF and FVIII [47, 48], indicating the dynamic impact of hypercortisolism on the coagulation system.

On the other side there is a paucity of data describing the direct impact of adrenal insufficiency on the hemostatic

system [37, 55]. It is well known that Addison disease can be associated with anti-phospholipid syndrome and recurrent thromboembolic events [56] and anti-phospholipid syndrome itself can lead to bilateral adrenal hemorrhage/infarction and acute adrenal insufficiency [57, 58].

Derex et al described the case of a young woman with previously undiagnosed primary adrenal insufficiency and a spontaneous intracerebral hemorrhage due to severe hypofibrinogenemia and a deficit of vitamin K-dependent coagulation factors [59]. The hemostatic abnormalities resolved after treatment with hydrocortisone. Peacey et al demonstrated that increased hydrocortisone doses in patients with hypopituitarism did not significantly affect levels of PAI-1, tissue plasminogen activator, and fibrinogen [60].

To the best of our knowledge, this study shows for the first time the consequences of a reduced cortisol secretion on FVIII. Referring to the above-mentioned mechanism of glucocorticoids on endothelial vWF production, we should have expected lower levels of vWF in our HPA-axis deficient patients. These had a trend to lower vWF levels, but the difference was statistically not significant.

Our study has limitations: the number of studied subjects was rather small, and we evaluated only a part of the complex coagulation system addressing thrombocyte and endothelial function, respectively. Assuming a close relationship between cortisol and catecholamine effects on the coagulation system, measuring catecholamines perhaps would have elucidated further mechanisms. However, this was not done in this study. We did not assess hemostasis parameter before surgery, which would have allowed a more precise pathophysiological classification of low FVIII and vWF levels in patient with dHPA (preexisting factor deficiency vs inability to mount an adequate FVIII response). The definition of normal and deficient HPA-axis function was made solely on the low-dose ACTH stimulation test.

Therefore, we emphasize the need to extend the studies and to also include persons with established and chronic adrenal insufficiency, respectively, in comparison to persons with nHPA and dHPA.

In conclusion, our data indicate that persons undergoing abdominal surgery with subnormal secretion of cortisol and probably decreased sympatho-adrenal medullary function have reduced levels of clotting factors whose release may critically relate to catecholaminergic function, particularly FVIII. This may have important clinical consequences, such as excessive blood loss.

Additional Information

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Disclosures: The authors have nothing to disclose.

Data Availability: All datasets generated and analyzed during the current study are not publicly available but are available from the corresponding author on reasonable request.

References

1. Collins GJ Jr, Barber JA, Zajtchuk R, Vanek D, Malogne LA. The effects of operative stress on the coagulation profile. *Am J Surg*. 1977;**133**(5):612-616.
2. Aillaud MF, Juhan-Vague I, Alessi MC, et al. Increased PA-inhibitor levels in the postoperative period–no cause-effect relation with increased cortisol. *Thromb Haemost*. 1985;**54**(2):466-468.
3. Sandset PM, Høgevold HE, Lyberg T, Andersson TR, Abildgaard U. Extrinsic pathway inhibitor in elective surgery: a comparison with other coagulation inhibitors. *Thromb Haemost*. 1989;**62**(3):856-860.
4. Gokce N, Keaney JF Jr, Hunter LM, Watkins MT, Menzoian JO, Vita JA. Risk stratification for postoperative cardiovascular events via noninvasive assessment of endothelial function: a prospective study. *Circulation*. 2002;**105**(13):1567-1572.
5. Hu YJ, Wei AN, Chook P, et al. Impact of non-cardiovascular surgery on reactive hyperaemia and arterial endothelial function. *Clin Exp Pharmacol Physiol*. 2013;**40**(7):466-472.
6. Rosenfeld BA, Faraday N, Campbell D, Dise K, Bell W, Goldschmidt P. Hemostatic effects of stress hormone infusion. *Anesthesiology*. 1994;**81**(5):1116-1126.
7. Sapolsky RM, Romero LM, Munck AU. How do glucocorticoids influence stress responses? Integrating permissive, suppressive, stimulatory, and preparative actions. *Endocr Rev*. 2000;**21**(1):55-89.
8. Yang S, Zhang L. Glucocorticoids and vascular reactivity. *Curr Vasc Pharmacol*. 2004;**2**(1):1-12.
9. Lee SR, Kim HK, Youm JB, et al. Non-genomic effect of glucocorticoids on cardiovascular system. *Pflugers Arch*. 2012;**464**(6):549-559.
10. Kastelan D, Dusek T, Kraljevic I, et al. Hypercoagulability in Cushing's syndrome: the role of specific haemostatic and fibrinolytic markers. *Endocrine*. 2009;**36**(1):70-74.
11. Boscaro M, Sonino N, Scarda A, et al. Anticoagulant prophylaxis markedly reduces thromboembolic complications in Cushing's syndrome. *J Clin Endocrinol Metab*. 2002;**87**(8):3662-3666.
12. Manetti L, Bogazzi F, Giovannetti C, et al. Changes in coagulation indexes and occurrence of venous thromboembolism in patients with Cushing's syndrome: results from a prospective study before and after surgery. *Eur J Endocrinol*. 2010;**163**(5):783-791.
13. von Känel R, Dimsdale JE. Effects of sympathetic activation by adrenergic infusions on hemostasis in vivo. *Eur J Haematol*. 2000;**65**(6):357-369.
14. Lanza F, Beretz A, Stierlé A, Hanau D, Kubina M, Cazenave JP. Epinephrine potentiates human platelet activation but is not an aggregating agent. *Am J Physiol*. 1988;**255**(6 Pt 2):H1276-H1288.
15. Spalding A, Vaitkevicius H, Dill S, MacKenzie S, Schmaier A, Lockette W. Mechanism of epinephrine-induced platelet aggregation. *Hypertension*. 1998;**31**(2):603-607.
16. Ehrhart-Bornstein M, Hinson JP, Bornstein SR, Scherbaum WA, Vinson GP. Intraadrenal interactions in the regulation of adrenocortical steroidogenesis. *Endocr Rev*. 1998;**19**(2):101-143.

17. Critchley JA, Henderson CG, Moffat LE, Ungar A, Waite J, West CP. Proceedings: The release of catecholamines from perfused canine adrenal glands by corticosteroids. *J Physiol*. 1976;**254**(1):30P-31P.
18. Udelsman R, Norton JA, Jelenich SE, et al. Responses of the hypothalamic-pituitary-adrenal and renin-angiotensin axes and the sympathetic system during controlled surgical and anesthetic stress. *J Clin Endocrinol Metab*. 1987;**64**(5):986-994.
19. Donald RA, Perry EG, Wittert GA, et al. The plasma ACTH, AVP, CRH and catecholamine responses to conventional and laparoscopic cholecystectomy. *Clin Endocrinol (Oxf)*. 1993;**38**(6):609-615.
20. Chernow B, Alexander HR, Smallridge RC, et al. Hormonal responses to graded surgical stress. *Arch Intern Med*. 1987;**147**(7):1273-1278.
21. Rittmaster RS, Cutler GB Jr, Sobel DO, et al. Morphine inhibits the pituitary-adrenal response to ovine corticotropin-releasing hormone in normal subjects. *J Clin Endocrinol Metab*. 1985;**60**(5):891-895.
22. Masala A, Satta G, Alagna S, et al. Effect of clonidine on stress-induced cortisol release in man during surgery. *Pharmacol Res Commun*. 1985;**17**(3):293-298.
23. Lacoumenta S, Paterson JL, Burrin J, Causon RC, Brown MJ, Hall GM. Effects of two differing halothane concentrations on the metabolic and endocrine responses to surgery. *Br J Anaesth*. 1986;**58**(8):844-850.
24. Engquist A, Brandt MR, Fernandes A, Kehlet H. The blocking effect of epidural analgesia on the adrenocortical and hyperglycemic responses to surgery. *Acta Anaesthesiol Scand*. 1977;**21**(4):330-335.
25. Kehlet H. *Clinical Course and Hypothalamc-Pituitary-Adrenocortical Function in Glucocorticoid-Treated Surgical Patients*. Copenhagen: FADL's Forlag. Published online 1976.
26. Salem M, Tainsh RE Jr, Bromberg J, Loriaux DL, Chernow B. Perioperative glucocorticoid coverage. A reassessment 42 years after emergence of a problem. *Ann Surg*. 1994;**219**(4):416-425.
27. Henzen C, Kobza R, Schwaller-Protzmann B, Stulz P, Briner VA. Adrenal function during coronary artery bypass grafting. *Eur J Endocrinol*. 2003;**148**(6):663-668.
28. Dickstein G, Shechner C, Nicholson WE, et al. Adrenocorticotropin stimulation test: effects of basal cortisol level, time of day, and suggested new sensitive low dose test. *J Clin Endocrinol Metab*. 1991;**72**(4):773-778.
29. Thaler LM, Blevins LS. The low dose (1-microg) adrenocorticotropin stimulation test in the evaluation of patients with suspected central adrenal insufficiency. *J Clin Endocrinol Metable* 1998;**83**(8):2726-2729.
30. Laureti S, Candeloro P, Aglietti MC, et al. Dehydroepiandrosterone, 17alpha-hydroxyprogesterone and aldosterone responses to the low-dose (1 micro g) ACTH test in subjects with preclinical adrenal autoimmunity. *Clin Endocrinol (Oxf)*. 2002;**57**(5):677-683.
31. Nasrallah MP, Arafah BM. The value of dehydroepiandrosterone sulfate measurements in the assessment of adrenal function. *J Clin Endocrinol Metab*. 2003;**88**(11):5293-5298.
32. Fischli S, Jenni S, Allemann S, et al. Dehydroepiandrosterone sulfate in the assessment of the hypothalamic-pituitary-adrenal axis. *J Clin Endocrinol Metab*. 2008;**93**(2):539-542.
33. Ingram GI. Increase in antihaemophilic globulin activity following infusion of adrenaline. *J Physiol*. 1961;**156**:217-224.
34. Ingram GI, Jones RV. The rise in clotting factor 8 induced in man by adrenaline: effect of alpha- and beta-blockers. *J Physiol*. 1966;**187**(2):447-454.
35. von Känel R, Kudielka BM, Helfricht S, et al. The effects of aspirin and nonselective beta blockade on the acute prothrombotic response to psychosocial stress in apparently healthy subjects. *J Cardiovasc Pharmacol*. 2008;**51**(3):231-238.
36. Jern C, Eriksson E, Tengborn L, Risberg B, Wadenvik H, Jern S. Changes of plasma coagulation and fibrinolysis in response to mental stress. *Thromb Haemost*. 1989;**62**(2):767-771.
37. Isidori AM, Minnetti M, Sbardella E, Graziadio C, Grossman AB. Mechanisms in endocrinology: The spectrum of haemostatic abnormalities in glucocorticoid excess and defect. *Eur J Endocrinol*. 2015;**173**(3):R101-R113.
38. Franchini M, Lippi G, Manzato F, Vescovi PP, Targher G. Hemostatic abnormalities in endocrine and metabolic disorders. *Eur J Endocrinol*. 2010;**162**(3):439-451.
39. Setian N, Tanaka CM, Damiani D, Dichtchekenian V, Carneiro JD, D'Amico EA. Hypopituitarism, deficiency of factors V and VIII and von Willebrand factor: an uncommon association. *J Pediatr Endocrinol Metab*. 2002;**15**(3):331-333.
40. Miljic P, Miljic D, Cain JW, Korbonits M, Popovic V. Pathogenesis of vascular complications in Cushing's syndrome. *Hormones (Athens)*. 2012;**11**(1):21-30.
41. Van Zaane B, Nur E, Squizzato A, et al. Hypercoagulable state in Cushing's syndrome: a systematic review. *J Clin Endocrinol Metab*. 2009;**94**(8):2743-2750.
42. Sato T, Hiramatsu R, Iwaoka T, Fujii Y, Shimada T, Umeda T. Changes of platelets, serum lactic dehydrogenase, gamma-glutamyltranspeptidase, choline esterase and creatine phosphokinase levels in patients with Cushing's syndrome. *Tohoku J Exp Med*. 1984;**142**(2):195-200.
43. Erem C, Nuhoglu I, Yilmaz M, et al. Blood coagulation and fibrinolysis in patients with Cushing's syndrome: increased plasminogen activator inhibitor-1, decreased tissue factor pathway inhibitor, and unchanged thrombin-activatable fibrinolysis inhibitor levels. *J Endocrinol Invest*. 2009;**32**(2):169-174.
44. Karamouzis I, Berardelli R, D'Angelo V, et al. Enhanced oxidative stress and platelet activation in patients with Cushing's syndrome. *Clin Endocrinol (Oxf)*. 2015;**82**(4):517-524.
45. Fatti LM, Bottasso B, Invitti C, Coppola R, Cavagnini F, Mannucci PM. Markers of activation of coagulation and fibrinolysis in patients with Cushing's syndrome. *J Endocrinol Invest*. 2000;**23**(3):145-150.
46. van der Pas R, de Bruin C, Leebeek FW, et al. The hypercoagulable state in Cushing's disease is associated with increased levels of procoagulant factors and impaired fibrinolysis, but is not reversible after short-term biochemical remission induced by medical therapy. *J Clin Endocrinol Metab*. 2012;**97**(4):1303-1310.
47. Dal Bo Zanon R, Fornasiero L, Boscaro M, Cappellato G, Fabris F, Girolami A. Increased factor VIII associated activities

in Cushing's syndrome: a probable hypercoagulable state. *Thromb Haemost*. 1982;**47**(2):116-117.
48. Casonato A, Pontara E, Boscaro M, et al. Abnormalities of von Willebrand factor are also part of the prothrombotic state of Cushing's syndrome. *Blood Coagul Fibrinolysis*. 1999;**10**(3):145-151.
49. Patrassi GM, Dal Bo Zanon R, Boscaro M, Martinelli S, Girolami A. Further studies on the hypercoagulable state of patients with Cushing's syndrome. *Thromb Haemost*. 1985;**54**(2):518-520.
50. Ikkala E, Myllylä G, Pelkonen R, Rasi V, Viinikka L, Ylikorkala O. Haemostatic parameters in Cushing's syndrome. *Acta Med Scand*. 1985;**217**(5):507-511.
51. Patrassi GM, Sartori MT, Viero ML, Scarano L, Boscaro M, Girolami A. The fibrinolytic potential in patients with Cushing's disease: a clue to their hypercoagulable state. *Blood Coagul Fibrinolysis*. 1992;**3**(6):789-793.
52. Chopra A, Kumar R, Kishore K, et al. Effect of glucocorticoids on von Willebrand factor levels and its correlation with von Willebrand factor gene promoter polymorphism. *Blood Coagul Fibrinolysis*. 2012;**23**(6):514-519.
53. Kerachian MA, Cournoyer D, Harvey EJ, et al. Effect of high-dose dexamethasone on endothelial haemostatic gene expression and neutrophil adhesion. *J Steroid Biochem Mol Biol*. 2009;**116**(3-5):127-133.
54. Trementino L, Arnaldi G, Appolloni G, et al. Coagulopathy in Cushing's syndrome. *Neuroendocrinology*. 2010;**92**(Suppl 1):55-59.
55. Squizzato A, Gerdes VE, Ageno W, Büller HR. The coagulation system in endocrine disorders: a narrative review. *Intern Emerg Med*. 2007;**2**(2):76-83.
56. Elebrashy I, Yousief E, Saif A. Primary anti-phospholipid antibody syndrome causing recurrent venous thrombosis and thrombocytopenia in a patient with Addison's disease. *JRSM Open*. 2014;**5**(12):2054270414562985.
57. Satta MA, Corsello SM, Della Casa S, et al. Adrenal insufficiency as the first clinical manifestation of the primary antiphospholipid antibody syndrome. *Clin Endocrinol (Oxf)*. 2000;**52**(1):123-126.
58. Ramon I, Mathian A, Bachelot A, et al. Primary adrenal insufficiency due to bilateral adrenal hemorrhage-adrenal infarction in the antiphospholipid syndrome: long-term outcome of 16 patients. *J Clin Endocrinol Metab*. 2013;**98**(8):3179-3189.
59. Derex L, Giraud P, Hanss M, Riche H, Nighoghossian N, Trouillas P. Spontaneous intracerebral hemorrhage revealing Addison's disease. *Cerebrovasc Dis*. 1998;**8**(4):240-243.
60. Peacey SR, Wright D, Aye M, Moisey R. Glucocorticoid replacement therapy and fibrinolysis in patients with hypopituitarism. *Clin Endocrinol (Oxf)*. 2012;**77**(1):94-98.