Kardiologia Polska 2018; 76, 9: 1372–1375; DOI: 10.5603/KP.2018.0184

SHORT COMMUNICATION

Similar prevalence of platelet factor 4/heparin immunoglobulin G antibodies in patients following cardiac surgery and other patients suspected of heparin-induced thrombocytopaenia

Agata H. Bryk[1, 2], Piotr Mazur[1, 2], Joanna Zdziarska[3], Paweł Kuczia[4], Krzysztof Plens[5], Agata Leśniak-Sobelga[6], Ewa Wypasek[1, 2, 7], Anetta Undas[1, 2]

[1]Institute of Cardiology, Jagiellonian University Medical College, Krakow, Poland

[2]John Paul II Hospital, Krakow, Poland

[3]Department of Haematology, Jagiellonian University Medical College, Krakow, Poland

[4]Department of Medicine, Jagiellonian University Medical College, Krakow, Poland

[5]KCRI, Krakow, Poland

[6]Department of Cardiac and Vascular Diseases, Institute of Cardiology, Jagiellonian University Medical College, John Paul II Hospital, Krakow, Poland

[7]Faculty of Medicine and Health Sciences, Andrzej Frycz Modrzewski Krakow University, Krakow, Poland

INTRODUCTION

Heparin-induced thrombocytopaenia (HIT) is an acquired thrombophilia associated with high risk of venous thromboembolism (VTE) and arterial thrombosis, largely caused by platelet-activating immunoglobulin G (IgG) antibodies against platelet factor (PF)4/heparin complexes [1]. Unfractionated heparin (UFH) is more likely, compared with low-molecular-weight heparins (LMWH), to provoke synthesis of anti-PF4/heparin antibodies, with the highest risk following cardiac surgery with extracorporeal circulation [2]. The 4Ts scoring system, incorporating four typical features of HIT (magnitude of thrombocytopaenia, timing with respect to heparin exposure, thrombosis, and other causes of thrombocytopaenia) is commonly used to assess the pre-test probability of HIT [3]. The diagnostic pathway involves high-sensitivity tests, e.g. enzyme-immunoassays (EIAs), followed by confirmatory platelet activation assays (serotonin-release assay [SRA] and/or heparin-induced platelet activation [HIPA]) [4]. To our knowledge, very few case reports have described Polish patients with HIT, with no cases including subjects undergoing cardiac surgery [5]. We sought to assess the prevalence of anti-PF4/heparin IgG antibodies in the first series of Polish patients undergoing cardiac surgery versus other patients suspected of HIT.

METHODS

We performed a retrospective study of patients suspected of HIT, tested at the John Paul II Hospital, Krakow, Poland, between January 2012 and July 2017. Patients provided written consent to use their data. The Hospital Bioethical Committee was informed about the study. Demographic and clinical data, including 4Ts scores [6], were analysed. The anti-PF4/heparin IgG antibodies were determined by EIA test (Immucor GTI Diagnostics, Waukesha, WI, USA), with confirmation using excess heparin (100 U/mL), following the manufacturer's instructions. All patients with optical density (OD) values ≥ 0.4 and inhibition of a positive reaction by ≥ 50% in the presence of excess heparin were deemed positive for PF4/heparin antibodies [7].

Statistical analysis

Continuous variables were expressed as mean ± standard deviation or median and interquartile range. Normality was assessed by the Shapiro-Wilk test. Groups were compared using the Student t test or Mann-Whitney U test, as appropriate. Categorical variables were presented as numbers (percentages) and compared using the Pearson's χ^2 test or the Fisher exact test if 20% of cells had an expected count of less than five. Two-sided p-values < 0.05 were considered statistically

Address for correspondence:
Agata H. Bryk, MD, Institute of Cardiology, Jagiellonian University Medical College, ul. Prądnicka 80, 31–202 Kraków, Poland, tel: +48 12 614 30 04, fax: +48 12 614 21 20, e-mail: agata.bryk@uj.edu.pl

Received: 19.06.2018 **Accepted:** 17.07.2018

Table 1. Comparison of patients following cardiac surgery and other patients

Variable	All patients (n = 122, 100%)	Patients following cardiac surgery (n = 39, 32%)	Other patients (n = 83, 68%)	p
Age [years]	66.5 (55–75)	67 (54–76)	66 (56–74)	0.9
Male sex	75 (61.5)	26 (66.7)	49 (59.0)	0.4
Heparin exposure and platelet fall				
Heparin type:				< 0.001
Low-molecular-weight heparin	51 (41.8)	0 (0)	51 (61.4)	
Unfractionated heparin	11 (9.0)	0 (0)	11 (13.3)	
Both agents	60 (49.2)	39 (100)	21 (25.3)	
Platelet count fall [%]	59.3 (44.2–77.5)	67.5 (53.8–83.2)	57.6 (42.3–76.2)	0.04
Time from introducing heparin to platelet fall [days]	6 (3–11)	6 (4-11)	7 (3-11)	0.6
Anti-PF4-heparin IgG EIA result*:				
≥ 0.4 OD	37 (30.3)	10 (25.6)	27 (32.5)	0.4
≥ 1.0 OD	27 (22.1)	7 (18.0)	20 (24.1)	0.4
≥ 2.0 OD	19 (15.6)	6 (15.4)	13 (15.7)	1.0
4Ts score:				0.003
≤ 3 points	72 (59.0)	29 (74.4)	43 (51.8)	
4–5 points	36 (29.5)	10 (25.6)	26 (31.3)	
≥ 6 points	14 (11.5)	0 (0)	14 (16.9)	

Data are presented as number (percentage) or median (interquartile range). Groups were compared using Mann-Whitney U test for continuous variables, Fisher exact test for 2 × 2 tables and χ^2 test for other tables. IgG — immunoglobulin G; EIA — enzyme-immunoassays; OD — optical density; PF — platelet factor
*Three participants with OD ≥ 0.4 were not inhibited by high heparin, and thus 34 (27.9%) patients were considered positive for anti-PF4/heparin IgG EIA

significant. Analyses were performed with Statistica (v. 13.1, StatSoft, Inc, Tulsa, OK, USA) and JMP® version 14.0.0 (SAS Institute Inc., Cary, NC, USA).

RESULTS AND DISCUSSION

A total of 255 tests were performed in 236 patients. Due to incomplete data, the final analysis was performed in 122 (51.7%) patients (Table 1), with similar age and sex distribution as the remainder. The cardiac surgery group with extracorporeal circulation (n = 39, 32%) comprised patients after the following procedures: aortic valve replacement (n = 27, 69.2%), mitral valve replacement (n = 3, 7.7%), coronary artery bypass grafting (n = 3, 7.7%), heart transplantation (n = 3, 7.7%), and other surgeries (n = 3, 7.7%). A reference group comprised 39 (47.0%) medical patients, 16 (19.3%) subjects following non-cardiac surgery, 16 (19.3%) after percutaneous angioplasty, five (6.0%) after aortic stent graft implantation, and seven (8.4%) after other procedures.

The cardiac surgery group, in which all patients received UFH followed by LMWH, experienced greater median platelet fall compared with the reference group, mainly receiving LMWH, with no intergroup difference in the median time from heparin therapy onset to the platelet fall (Table 1). The proportion of patients with positive anti-PF4/heparin antibodies was comparable in both groups (10 [25.6%] vs. 24 [28.9%], p = 0.7), and the former value was similar compared with the rate reported recently [8]. The median OD value among patients positive for the anti-PF4/heparin IgG antibodies was 2.94 (0.69–3.90) in the cardiac surgery group and 1.70 (1.06–3.26) in the reference group (p > 0.05). A previous report showed that replacing UFH with LMWH decreases levels of anti-PF4/heparin antibodies [9]. Contrary to earlier studies [10], in our study the prevalence of positive anti-PF4/heparin IgG antibodies was similar in the cardiac surgery group and the reference group, probably due to a postoperative conversion from UFH to LMWH soon after the surgery. The reference group included patients treated for acute coronary syndrome, among whom UFH is still a more frequently used anticoagulant than LMWH [11].

The proportion of positive results according to the heparin type was: UFH, 9.0%; LMWH, 25.5%; and both agents, 33.3%. Enoxaparin (n = 84, 68.9%) was the most commonly used LMWH, as reported in various Polish patients [12], followed by nadroparin (n = 11, 9.0%), dalteparin (n = 6, 4.9%), and two different LMWHs used during therapy (n = 3, 3.6%); in seven (5.7%) patients a LMWH was unspecified.

Patients in the cardiac surgery group were less likely to have four points or more in 4Ts score compared with the reference group (10 [25.6%] vs. 40 [48.2%], p = 0.02), confirming the clinical relevance of this scoring system [3].

Fondaparinux was the most common non-heparin alternative (n = 19, 55.9%) among our patients with positive anti-PF4/heparin antibodies following heparin withdrawal, as it was among anticoagulants used off-label in Germany [13]. Other anticoagulants used in our cohort following positive results were bivalirudin (n = 1, 2.9%), vitamin K antagonists (n = 3, 8.8%), and rivaroxaban (n = 1, 2.9%), although the use of oral anticoagulants is contraindicated in patients suspected of HIT if the platelet count is low [14]; other non-heparin alternatives were unspecified. Argatroban was not used in any patient.

We identified 21 (17.2%) thromboembolic events, including 12 (57.1%) VTEs, six (28.6%) arterial thromboses, and three (14.1%) cases of acral necrosis, all of which occurred in the reference group. Among those 21 patients, there were more patients with OD ≥ 0.4, as compared with patients without thromboembolic events (14 [66.7%] vs. 23 [22.7%], p < 0.001). Similar observations held true for OD ≥ 1.0 (13 [61.9%] vs. 14 [13.9%], p < 0.001) and OD ≥ 2.0 (9 [42.9%] vs. 10 [9.9%], p < 0.001), respectively.

The most unusual patient in our series was a 31-year-old obese woman, with prior deep vein thrombosis at the age of 18 years while on oral contraceptives, with no family history of VTE, who was diagnosed with left iliofemoral thrombosis in the 10th week of her second pregnancy. After three days of UFH therapy followed by the use of enoxaparin for 32 days, progression of the disease and a decrease in platelet count from 337 × 10^9/L to 38 × 10^9/L (reference range, 140–440 × 10^9/L) were noted. As shown in **Supplemental Figure 1 (see journal website)**, the diagnosis of HIT was supported by highly elevated anti-PF4/heparin IgG antibodies. Fondaparinux 7.5 mg/d was initiated, and platelet count normalised. Thrombophilia screening showed a heterozygous factor V Leiden mutation. Pregnancy failed at the 24th week. Later, warfarin was introduced, and anticoagulation was stable. Unexpectedly, anti-PF4/heparin IgG antibodies remained elevated in the subsequent 4.5 years, exceeding 90 days to a negative test observed in most patients with HIT [15]. The HIPA test confirmed HIT-specific antibodies and their absence when the antibody test yielded a negative result (**Suppl. Fig. 1 — see journal website**). It is known that in the EIA test the OD values between 0.4 and 1.0 are weakly associated with positive functional test results, while OD values ≥ 2.0 correspond to a > 90% probability of SRA [3]. This case strongly encourages the use the SRA or HIPA tests to confirm HIT, especially if OD values are below 1.

The study limitations involve shortcomings typical of retrospective design and a lack of results of specific tests for all patients with elevated OD. The study strength lies in the fact that it is the first analysis of consecutive patients suspected of HIT referred for laboratory work-up in Poland, which confirms the need for a wider availability of specific tests to confirm positive anti-PF4/heparin antibodies, given the high proportion of negative final results at low 4Ts scores. This study supports rapid initiation of LMWH after cardiac surgery to reduce the risk of HIT and its life-threatening sequalae.

Acknowledgements

We would like to thank Professor Andreas Greinacher, MD for his invaluable help in the evaluation of patients with heparin-induced thrombocytopaenia and Magdalena Kopytek, MSc for her technical assistance.

Conflict of interest: *none declared*

References

1. Greinacher A. Heparin-Induced thrombocytopenia. N Engl J Med. 2015; 373(3): 252–261, doi: 10.1056/NEJMcp1411910, indexed in Pubmed: 26176382.
2. Warkentin TE, Sheppard JA, Horsewood P, et al. Impact of the patient population on the risk for heparin-induced thrombocytopenia. Blood. 2000; 96(5): 1703–1708, indexed in Pubmed: 10961867.
3. Cuker A, Gimotty PA, Crowther MA, et al. Predictive value of the 4Ts scoring system for heparin-induced thrombocytopenia: a systematic review and meta-analysis. Blood. 2012; 120(20): 4160–4167, doi: 10.1182/blood-2012-07-443051, indexed in Pubmed: 22990018.
4. Warkentin TE. How I diagnose and manage HIT. Hematology. 2011; 2011: 143–149, doi: 10.1182/asheducation-2011.1.143, indexed in Pubmed: 22160026.
5. Sikorska A, Tomaszewska M, Baran B, et al. Cerebral venous and sinus thrombosis complicated by heparin-induced thrombocytopenia. Hematologica. 2011; 2(4): 363–369.
6. Lo GK, Juhl D, Warkentin TE, et al. Evaluation of pretest clinical score (4 T's) for the diagnosis of heparin-induced thrombocytopenia in two clinical settings. J Thromb Haemost. 2006; 4(4): 759–765, doi: 10.1111/j.1538-7836.2006.01787.x, indexed in Pubmed: 16634744.
7. Pauzner R, Greinacher A, Selleng K, et al. False-positive tests for heparin-induced thrombocytopenia in patients with antiphospholipid syndrome and systemic lupus erythematosus. J Thromb Haemost. 2009; 7(7): 1070–1074, doi: 10.1111/j.1538-7836.2009.03335.x, indexed in Pubmed: 19291166.
8. Pishko A, Cuker A. Heparin-induced thrombocytopenia in cardiac surgery patients. Semin Thromb Hemost. 2017; 43(07): 691–698, doi: 10.1055/s-0037-1602664.
9. McGowan KE, Makari J, Diamantouros A, et al. Reducing the hospital burden of heparin-induced thrombocytopenia: impact of an avoid-heparin program. Blood. 2016; 127(16): 1954–1959, doi: 10.1182/blood-2015-07-660001, indexed in Pubmed: 26817956.
10. Pouplard C, May MA, Iochmann S, et al. Antibodies to platelet factor 4-heparin after cardiopulmonary bypass in patients anticoagulated with unfractionated heparin or a low-molecular-weight heparin: clinical implications for heparin-induced thrombocytopenia. Circulation. 1999; 99(19): 2530–2536, indexed in Pubmed: 10330384.
11. Wojtkowska I, Stępińska J, Stępień-Wojno M, et al. Current patterns of antithrombotic and revascularisation therapy in patients

hospitalised for acute coronary syndromes. Data from the Polish subset of the EPICOR study. Kardiol Pol. 2017; 75(5): 445–452, doi: 10.5603/kp.a2017.0034.
12. Krasiński Z, Krasińska B, Dzieciuchowicz Ł, et al. Heparins in cancer-associated venous thrombosis. Pol Arch Med Wewn. 2016; 126(6): 419–429, doi: 10.20452/pamw.3449, indexed in Pubmed: 27362395.
13. Schindewolf M, Steindl J, Beyer-Westendorf J, et al. Frequent off-label use of fondaparinux in patients with suspected acute heparin-induced thrombocytopenia (HIT)--findings from the GerHIT multi-centre registry study. Thromb Res. 2014; 134(1): 29–35, doi: 10.1016/j.thromres.2014.03.029, indexed in Pubmed: 24703295.
14. Linkins LA, Dans AL, Moores LK et al. Treatment and prevention of heparin-induced thrombocytopenia: Antithrombotic Therapy and Prevention of Thrombosis, 9th ed: American College of Chest Physicians Evidence-Based Clinical Practice Guidelines. Chest. 2012; 114(2 Suppl): e495S-e530S. doi: 10.1378/chest.11-2303, indexed in Pubmed: 22315270.
15. Mattioli AV, Bonetti L, Zennaro M, et al. Heparin/PF4 antibodies formation after heparin treatment: temporal aspects and long-term follow-up. Am Heart J. 2009; 157(3): 589–595, doi: 10.1016/j.ahj.2008.11.007, indexed in Pubmed: 19249435.

Cite this article as: Bryk AH, Mazur P, Zdziarska J, et al. Similar prevalence of platelet factor 4/heparin immunoglobulin G antibodies in patients following cardiac surgery and other patients suspected of heparin-induced thrombocytopaenia. Kardiol Pol. 2018; 76(9): 1372–1375, doi: 10.5603/KP.2018.0184.