Immunological Disorders and Immunotherapy

Mitsui, et al., Immunol Disord Immunother 2016, 1:1

Short Communication open access

P-Selectin is a Key Molecule Underlying the Pathophysiology of Aspirin-Exacerbated Respiratory Disease

Chihiro Mitsui[1,3], Keiichi Kajiwara[1], Hiroaki Hayashi[1], Jun Ito[1], Haruhisa Mita[1], Emiko Ono[1], Noritaka Higashi[1,2], Yuma Fukutomi[1], Kiyoshi Sekiya[1], Takahiro Tsuburai[1], Kazuo Akiyama[1], Kazuhiko Yamamoto[3] and **Masami Taniguchi[*1]**

[1]*Clinical Research Center for Allergy and Rheumatology, National Hospital Organization, Sagamihara National Hospital, Kanagawa, Japan*

[2]*Japanese Red Cross Kumamoto Health Care Center, Kumamoto, Japan*

[3]*Department of Allergy and Rheumatology, Graduate School of Medicine, University of Tokyo, Tokyo, Japan*

[*]**Corresponding author:** Masami Taniguchi, Clinical Research Center for Allergy and Rheumatology, National Hospital Organization, Sagamihara National Hospital, Kanagawa, Japan, Tel: +81 (42) 742-8311; FAX: +81 (42) 742-7990; E-mail: m-taniguchi@sagamihara-hosp.gr.jp

Rec date: March 1, 2016, **Acc date:** March 31, 2016, **Pub date:** April 4, 2016

Abstract

Recently, the evidence for platelet association with allergic diseases has been established by many researches. Activated platelets contribute to airway hyper reactivity, bronchoconstriction, airway inflammation and airway remodeling in asthmatic patients. We aimed to clarify platelet activation and key molecules on platelets, which are mainly associated with the mechanism underlying aspirin-exacerbated respiratory disease.

Keywords: P-selectin; Adhesion; Aspirin-exacerbated respiratory disease; Asthma; Cysteinyl leukotriene; Platelet

Abbreviations

AERD: Aspirin-Exacerbated Respiratory Disease; COX: Cyclooxygenase; PAFL: Persistent Airflow Limitation; CysLT: Cysteinyl Leukotriene; LT: Leukotriene; uLTE4: Urinary Leukotriene E4; PGE2: Prostaglandin E2; EP2: E Prostanoid 2; AHR: Airway Hyperreactivity; ATA: Aspirin-Tolerant Asthma; PSGL-1: P-Selectin Glycoprotein Ligand 1; CD40L: CD40 Ligand; sP-selectin: Soluble P-selectin; sCD40L: Soluble CD40L; CEP: Idiopathic Chronic Eosinophilic Pneumonia; L-ASA: Lysine-Aspirin; LTC4S: LTC4 Synthase; 5-LO: 5-Lipoxygenase

Aspirin-exacerbated respiratory disease (AERD) is characterized by the triad of asthma, nasal polyposis, and hypersensitivity to aspirin and other cyclooxygenase (COX)-1 inhibitors [1]. Aspirin intolerance is one of the risk factors for severe asthma [1-3], particularly in patients with persistent airflow limitation (PAFL) [4]. The overproduction of cysteinyl leukotriene (cysLT) is the biochemical hallmark of AERD. Urinary leukotriene (LT) E4 (uLTE4) levels are 3-4 fold higher in AERD patients than in ATA patients, which further increase after aspirin-induced reaction in AERD patients [5]. Decreased levels of prostaglandin E2 (PGE2) were also a characteristic feature of AERD patients. Impaired E prostanoid 2 (EP2) expression and resistance to PGE2 were detected in nasal polyp fibroblasts from AERD patients [6]. However, the mechanism underlying the development of AERD has not been clarified in detail.

In addition to their role in hemostasis, platelets have the capacity to mediate immune responses during inflammation and to facilitate granulocyte recruitment into airways. Platelet activation has been detected in several allergic diseases, including asthma. Platelets play an important role in asthma, which contributes to airway hyper reactivity (AHR), bronchoconstriction, airway inflammation and airway remodeling [6,7]. Recently, Laidlaw and Boyce have demonstrated that platelet activity is strongly associated with the pathophysiology of AERD [7,8]. We have also hypothesized that platelets might be related to the pathogenesis of AERD because AERD patients can be caused respiratory reactions to aspirin and be desensitized with low-dose aspirin [8-12], which irreversibly inhibits COX-1 in platelets but not COX-2 in endothelial cells and leukocytes [12-14]. As circulating platelets lack a nucleus and ability of mRNA synthesis, it is generally assumed that thromboxane A2 synthesis is mainly dependent on COX-1 [14]. Refractory period to aspirin after an aspirin challenge test, which is almost equivalent to platelet lifespan, is also characteristic of such patients [15].

In this study, we recently assessed reported comparison of platelet activation in between patients with AERD and aspirin-tolerant asthma (ATA) under stable disease condition and during the aspirin challenge test [16]. Activated platelets bind to leukocytes via adhesion molecules, including P-selectin (CD62P)-P-selectin glycoprotein ligand 1 (PSGL-1), GPIIb/IIIa-Mac-1, and CD40 ligand (CD40L)-CD40 [17,18]. Considering this, we assessed platelet activation markers in AERD patients (n = 30) and ATA patients (n = 21) under stable disease condition, including the expression levels of P-selectin (CD62P), CD63, CD69, and GPIIb/IIIa (PAC-1) on peripheral platelets, the percentage of circulating platelet-adherent leukocytes, and the levels of soluble P-selectin (sP-selectin) and soluble CD40 ligand (sCD40L) in plasma. Ten idiopathic chronic eosinophilic pneumonia (CEP) patients (n = 10), who also showed airway eosinophilia and cysLT overproduction, and normal controls (n = 14) were also included in the study.

As a result, AERD patients showed higher expression levels of all surface markers, P-selectin, CD63, CD69, and PAC-1, on platelets, and higher levels of plasma sP-selectin and sCD40L than ATA patients. In the comparison between AERD patients and CEP patients, the expression levels of CD63 and CD69, and the level of plasma sCD40 L in AERD patients were higher than those in CEP patients. Furthermore, the percentage of platelet-adherent eosinophils was significantly higher in AERD patients than in ATA patients and controls, although there was no difference between AERD patients and CEP patients.

We also assessed plasma levels of sP-selectin and sCD40L platelet activation markers in 24 AERD patients and 7 ATA patients during the aspirin challenge test. Blood samples were collected at 0-1, 1-3, 3-6, and 9-24 h after ingestion of lysine-aspirin (L-ASA) at a dose that produced a positive reaction in AERD patients or after ingestion of the last dose of L-ASA in ATA patients. The positive reaction was induced approximately 0.5-1 h after the ingestion of the last dose, and blood 0-1 h after the last dose was collected within 5-20 min from the occurrence of the reaction in AERD patients. Additionally, surface markers on platelets were also assessed at 0-1, 1-3, and 9-24 h in 8 AERD patients. Urine samples were also collected for measurements of LTE4 concentration at the following periods: 0-3, 3-6, 6-9, and 9-24 h.

There were no changes in the plasma levels of platelet activation markers and plasma markers, and the expression levels of the surface markers on platelets during the aspirin challenge test in both AERD and ATA patients. Other studies showed that platelets are activated after a mite challenge test in the early phase; however, the activation persisted until late phase after the allergen challenge test in some patients [19,20]. Our findings suggest that platelet activation did not occur in the early phase of aspirin-induced reaction. It is possible that platelets are activated in patients whose symptoms persistent for 9-24 h after aspirin-induced reaction.

When we considered the relationship of the markers with clinical characteristics, the levels of some of the platelet activation markers correlated with uLTE4 level and pulmonary function. In particular, the level of plasma sP-selectin well correlated with uLTE4 level and pre- and post-bronchodilator pulmonary function. Platelets lack 5-lipoxygenase (5-LO); however, adherent platelets contribute to the transcellular metabolism of LTs from leukocyte-derived LTA4 via LTC4S in platelets [21,22]. Laidlaw et al. demonstrated that almost 70% of the LTC4S activity in the granulocytes from AERD patients was platelet-derived, and uLTE4 concentration correlated strongly with the percentage of platelet-adherent leukocytes in peripheral blood [23].

Platelets in both mice and human subjects express the type 1 receptor for cysteinyl leukotrienes (cysLT1R) and cysLT2R, and can be stimulated by cysLT. The P2Y12 receptor was found as a novel receptor of LTE4, and mice lacking this receptor do not show allergic responses to LTE4. cysLT2R signaling by LTC4 subsequently causes an autocrine ADP-mediated response through the P2Y12 receptor, which induces the expression of P-selectin on the surface of murine platelets [24]. However, LTC4 cannot induce P-selectin expression on platelets from allergic patients [25]. It remains to be clarified neither platelets might be further activated by cysLT and serve in a positive feedback loop for inflammation in AERD patients.

Another study also indicated that P-selectin on the platelet surface is a key molecule in pulmonary eosinophil recruitment in patients with allergic asthma [26]. Platelet-adherent leukocytes, which highly express integrin, are recruited to airways by their firm adhesion to the bronchial endothelium [7]. In this study, AERD patients characteristically show severe eosinophil infiltration into the upper and lower airways and PAFL. Platelet adhesion to leukocytes particularly via P-selectin/PSGL-1 might mediate severe eosinophilia in airways of AERD patients.

In conclusion, P-selectin was found to be the key molecule related to cysLT overproduction and PAFL in AERD patients. P-selectin might be a therapeutic target molecule in AERD patients. Furthermore, other antiplatelet therapies, blockade of the thromboxane A2 and P2Y12 receptors, would also be therapeutically useful in AERD patients.

Acknowledgements

We thank Ms. Itsuko Ito for excellent technical assistant.

This study is supported by the Health labour sciences research grant on allergic disease and immunology from the Ministry of Health Labour and Welfare of Japan (Grant Number: H26 – ITAKU (MEN) – IPPAN – 002).

References

1. Stevenson DD, Szczeklik A (2006) Clinical and pathologic perspectives on aspirin sensitivity and asthma. J Allergy Clin Immunol 118: 773-786.
2. Szczeklik A, Nizankowska E, Duplaga M (2000) Natural history of aspirin-induced asthma. AIANE Investigators. European Network on Aspirin-Induced Asthma. Eur Respir J 16: 432-436.
3. Fukutomi Y, Taniguchi M, Tsuburai T, Tanimoto H, Oshikata C, et al. (2012) Obesity and aspirin intolerance are risk factors for difficult-to-treat asthma in Japanese non-atopic women. Clin Exp Allergy 42: 738-746.
4. Mascia K, Haselkorn T, Deniz YM, Miller DP, Bleecker ER, et al. (2005) Aspirin sensitivity and severity of asthma: evidence for irreversible airway obstruction in patients with severe or difficult-to-treat asthma. J Allergy Clin Immunol 116: 970-975.
5. Christie PE, Tagari P, Ford-Hutchinson AW, Charlesson S, Chee P, et al. (1991) Urinary leukotriene E4 concentrations increase after aspirin challenge in aspirin-sensitive asthmatic subjects. Am Rev Respir Dis 143: 1025-1029.
6. Idzko M, Pitchford S, Page C (2015) Role of platelets in allergic airway inflammation. J Allergy Clin Immunol 135: 1416-1423.
7. Laidlaw TM, Boyce JA (2015) Platelets in patients with aspirin-exacerbated respiratory disease. J Allergy Clin Immunol 135: 1407-1414.
8. Kowalski ML, Grzelewska-Rzymowska I, Rozniecki J, Szmidt M (1984) Aspirin tolerance induced in aspirin-sensitive asthmatics. Allergy 39: 171-178.
9. Hope AP, Woessner KA, Simon RA, Stevenson DD (2009) Rational approach to aspirin dosing during oral challenges and desensitization of patients with aspirin-exacerbated respiratory disease. J Allergy Clin Immunol 123: 406-410.
10. Stevenson DD, Pleskow WW, Simon RA, Mathison DA, Lumry WR, et al. (1984) Aspirin-sensitive rhinosinusitis asthma: a double-blind crossover study of treatment with aspirin. J Allergy Clin Immunol 73: 500-507.
11. Fruth K, Pogorzelski B, Schmidtmann I, Springer J, Fennan N, et al. (2013) Low-dose aspirin desensitization in individuals with aspirin-exacerbated respiratory disease. Allergy 68: 659-665.
12. Bertele V, Falanga A, Tomasiak M, Dejana E, Cerletti C, et al. (1983) Platelet thromboxane synthetase inhibitors with low doses of aspirin: possible resolution of the "aspirin dilemma". Science 220: 517-519.
13. Capone ML, Tacconelli S, Sciulli MG, Grana M, Ricciotti E, et al. (2004) Clinical pharmacology of platelet, monocyte, and vascular cyclooxygenase inhibition by naproxen and low-dose aspirin in healthy subjects. Circulation 109: 1468-1471.
14. Patrono C, Coller B, Dalen JE, FitzGerald GA, Fuster V, et al. (2004) Platelet-active drugs: the relationships among disease, effectiveness, and side effects. Chest 119: 39S-63S.
15. Taytard A, Guenard H, Vuillemin L, Bouvot JL, Vergeret J, et al. (1986) Platelet kinetics in stable atopic asthmatic patients. Am Rev Respir Dis 134: 983-985.
16. Mitsui C, Kajiwara K, Hayashi H, Ito J, Mita H, et al. (2016) Platelet activation markers overexpressed specifically in patients with aspirin-exacerbated respiratory disease. J Allergy Clin Immunol 137: 400-411.
17. Jawien J, Lomnicka M, Korbut R, Chlopicki S (2005) The involvement of adhesion molecules and lipid mediators in the adhesion of human platelets to eosinophils. J Physiol Pharmacol 56: 637-648.
18. Li N (2008) Platelet-lymphocyte cross-talk. J Leukoc Biol 83: 1069-1078.

19. Kowal K, Pampuch A, Kowal-Bielecka O, DuBuske LM, Bodzenta-Lukaszyk A (2006) Platelet activation in allergic asthma patients during allergen challenge with Dermatophagoides pteronyssinus. Clin Exp Allergy 36: 426-432.
20. Kowal K, Pampuch A, Kowal-Bielecka O, Iacoviello L, Bodzenta-Lukaszyk A (2006) Soluble CD40 ligand in asthma patients during allergen challenge. J Thromb Haemost 4: 2718-2720.
21. Maclouf JA, Murphy RC (1988) Transcellular metabolism of neutrophil-derived leukotriene A4 by human platelets. A potential cellular source of leukotriene C4. J Biol Chem 263: 174-181.
22. Patrignani P, Dovizio M (2012) Inside platelet-leukocyte cross-talk. Blood 119: 3649-3650.
23. Laidlaw TM, Kidder MS, Bhattacharyya N, Xing W, Shen S, et al. (2012) Cysteinyl leukotriene overproduction in aspirin-exacerbated respiratory disease is driven by platelet-adherent leukocytes. Blood 119: 3790-3798.
24. Cummings HE, Liu T, Feng C, Laidlaw TM, Conley PB, et al. (2013) Cutting edge: Leukotriene C4 activates mouse platelets in plasma exclusively through the type 2 cysteinyl leukotriene receptor. J Immunol 191: 5807-5810.
25. Paruchuri S, Tashimo H, Feng C, Maekawa A, Xing W, et al. (2009) Leukotriene E4-induced pulmonary inflammation is mediated by the P2Y12 receptor. J Exp Med 206: 2543-2555.
26. Ulfman LH, Joosten DP, van Aalst CW, Lammers JW, van de Graaf EA, et al. (2003) Platelets promote eosinophil adhesion of patients with asthma to endothelium under flow conditions. Am J Respir Cell Mol Biol 28: 512-519.