Plasma Levels of Platelet-Derived Microparticles are Increased After Anaerobic Exercise in Healthy Subjects

It is well known that aerobic exercise is effective for reducing the risk factors for diabetes mellitus and hyperlipidemia, and the incidence of cardiovascular events\(^1\). On the other hand, the sudden death of trained and apparently healthy athletes during exercise has become a problem in recent years. Strenuous exercise induces hypercoagulability and activates platelet function\(^2\). Recently, platelet-derived microparticles (PDMPs) have become a frequently used marker of platelet activation, and can be measured using an enzyme-linked immunosorbent assay (ELISA). Although flow cytometry is the most widely used method for measuring PDMPs, the major disadvantage of flow cytometry is that MPs < 0.5 \(\mu\)m are difficult to distinguish from cellular debris. In contrast, it has been considered that ELISA is an easier and more reproducible PDMP assay than flow cytometry. Plasma PDMP levels have been measured by ELISA in patients with various diseases, including acute coronary syndrome, arteriosclerosis\(^3\), and obstructive sleep apnea syndrome\(^4\). We herein show that the plasma levels of PDMPs were significantly increased after anaerobic exercise, and were prolonged for up to 1 h after exercise.

The study population consisted of 18 healthy volunteers (9 males and 9 females; aged 21 \(\pm\) 0.3 years, mean \(\pm\) SD). Treadmill testing was performed using the Bruce protocol\(^5\). The point when the subject reached 85% of his or her target maximum heart rate \([220 - \text{subject age}] \times 0.85\) was assumed to be the endpoint. Blood samples were taken before the exercise test, and immediately and 1 hour after the exercise test. The plasma levels of PDMP were determined by an ELISA kit (Jimro Co. Ltd.). To examine fibrinolysis and coagulation, the plasma levels of t-PA antigen (Assay Pro), plasminogen activator inhibitor type 1 (PAI-1) antigen and soluble fibrin (SF) (Mitsubishi Chemical Medience) were measured. The study followed the institutional guidelines of the University of Kanazawa, and informed consent was obtained from all patients according to the Declaration of Helsinki.

The systolic blood pressure and plasma levels of lactic acid were significantly increased after exercise (Fig. 1A; \(p<0.05\)). PDMPs have a negatively charged phospholipid surface, and rapidly bind activated coagulation factors\(^6\). Moreover, a previous study reported that PDMPs have 50- to 100-fold higher specific procoagulant activity than activated platelets\(^7\). PDMPs activate monocytes, and promote the production of monocyte-derived MPs (MDMPs). MDMPs are associated with the development of platelet- and fibrin-rich thrombi through the recruitment of cells and accumulation of TF\(^8\); therefore, the PDMPs that are increased after anaerobic exercise are not only involved in the activation of platelets, but also may promote clot formation.

The plasma levels of t-PA antigen increased immediately after exercise and returned to the baseline after 1 h (Fig. 1B; \(p<0.05\)). In contrast, PAI-1 antigens did not change immediately, but were decreased after 1 h (Fig. 1C; \(p<0.05\)); therefore, these results suggest that anaerobic exercise might increase fibrinolytic activity. Our results are in agreement with a previous report\(^9\). Additionally, a previous study showed that strenuous exercise increased plasma catecholamine levels\(^8\), and catecholamine enhanced the release of t-PA from endothelial cells and inhibited PAI-1 mRNA and production in human fat\(^9\). Thus, PAI-1 antigens might decrease after 1 h by increasing plasma catecholamine levels.

The plasma levels of SF did not significantly change after exercise in our study (data not shown). SF is an intermediary product formed during the process of converting fibrinogen to fibrin. Bartsch P. et al. have reported that prothrombin fragment 1 + 2 and thrombin-antithrombin complex were increased by exercise, but fibrinopeptide A did not significantly change. They suggested that the generated thrombin

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<th>Table 1. Systolic blood pressure and plasma levels of lactic acid in 18 healthy subjects before (pre) and after (post) exercise</th>
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<td>systolic blood pressure (mmHg)</td>
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<td>plasma levels of lactic acid (mg/mL)</td>
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Data are presented as the mean \(\pm\) SD. *\(p<0.05\) versus pre-exercise.
might be fully inactivated by antithrombin, and was therefore unable to give rise to fibrin formation\(^{10}\). Our results appear to be consistent with their findings.

In this study, we could not directly show that anaerobic exercise activated the coagulation system. Furthermore, this result might be different from research which targeted middle-aged or elderly patients with arteriosclerosis since this study targeted healthy young subjects. In further studies, we plan to investigate plasma PDMPs after exercise in middle-aged or elderly patients with arteriosclerosis.

**Fig. 1.** Effects of exercise on plasma levels of PDMPs (A), t-PA antigen (B), and PAI-1 antigen (C). Values are the mean ± SD.

**References**


7) Sinauridze EI, Kireev DA, Popenko NY, Pichugin AV, Panteleev MA, Krymskaya OV, Ataullakhanov FI: Platelet microparticle membranes have 50- to 100-fold higher specific procoagulant activity than activated platelets. Thromb Heamost, 2007; 97: 425-434


9) Halleux CM, Declerck PJ, Tran SL, Detry R, Brichard SM: Hormonal control of plasminogen activator inhibitor-1 gene expression and production in human adipose
tissue: stimulation by glucocorticoids and inhibition by catecholamines. J Clin Endocrinol Metab, 1999; 84: 4097-4105


Keiko Maruyama¹, Tadaaki Kadono² and Eriko Morishita¹

¹Department of Clinical Laboratory Science, Kanazawa University Graduate School of Medical Science, Kanazawa, Japan
²Department of Clinical Laboratory, Kanazawa University Hospital, Kanazawa, Japan

Address for correspondence: Eriko Morishita, Department of Clinical Laboratory Science, Kanazawa University Graduate School of Medical Science, 5-11-80 Kodatuno Kanazawa, Ishikawa 920-0942, Japan
E-mail: eriko@med3.m.kanazawa-u.ac.jp