INTRODUCTION

Thrombosis is a crucial pathological phenomenon in various cardiovascular diseases. An injured or denuded surface of the vessel attracts platelets and primary thrombus is formed. In this process, particularly in the formation of primary thrombus which mainly formed by platelets, it recently draws wide attention that a long chain macromolecule named von Willebrand factor (vWF) plays an important role to mediate the mechano-chemical interactions between the platelets and between the platelets and the vessel wall. The platelet has a variety of receptors on its surface, such as GP Ibα and GP IIb/IIIa, known to react with the von Willebrand factor and the Fibrinogen. Some of them also have an ability to sense the fluid shear stress. Thus the initiation of the thrombosis is now related to the hemodynamics.

In the present study, main aim is to analyze the mechanism of platelet adhesion by using a computational model of the receptors on the platelet, the vWF and the Fibrinogen. The computational method was a combination of the Discrete Element Method (DEM) and the Stokesian Dynamics Method (SDM) specially devised for this study. The computational analysis was performed to examine the influence of activation of the receptors in the thrombus formation process.

METHOD

Physiological mechanism of the thrombus formation

The platelet adhesion process on the vessel wall is generally thought to proceed as following. At the first stage, the collagen and the insoluble form of the vWF in the subendothelial layer is exposed to the blood on the damaged vessel wall, and the vWF is activated. Platelets carried in by the blood flow transiently bind with vWF mediated by the activated GP Ibα. When a platelet stops, it is caught on the denuded collagen surface and holds firmly through the collaboration of the GPIIb/IIIa, the vWF, and the Fibrinogen. Then, a thrombus will develop towards the intraluminal side of a vessel.

Simulation Model

The computational model was built to simulate the above physiological mechanism. First, the platelet was modeled by a circle in a 2-dimensional flow field. Two kinds of receptors were assumed to exist on the platelet (Figure 1(a)). As GP Ibα is known to react with vWF transiently, the model to GP Ibα was set up to be activated for a limited length of the time. On the other hand, GPIIb/IIIa model to as set to be activated when relative velocity between platelets becomes less than a specified value. The mechanical interactions of the platelets mediated by the vWF and the Fibrinogen are expressed by the Voigt model using a spring and a dashpot. (Figure 1 (b)(c)). This is an extension of the DEM. It was assumed that they have the ability to react with two kinds of receptors present on the platelet. The vWF binds GP Ibα and GPIIb/IIIa, and the Fibrinogen binds only GPIIb/IIIa.

Physical properties of the vWF, the Fibrinogen and the receptors, were assumed to have appropriate values.

Stokesian Dynamics Method (SDM)

In the SDM used in the present study, the flow field induced by the motion of particles are computed using analytical solution of the Stokes equation and the entire velocity fields is computed summing up the individual velocity field around the particles. The calculated induced velocity field is, in turn, used to estimate the fluid force acting on individual particles. Thus the influence of the motion of other particles is taken into account when the net force acting on a particle is computed as shown in the following;
\[ \mathbf{v}_d = \mathbf{y} \boldsymbol{\gamma} + \sum_{(i=1)}^{N} \left( \frac{1}{6} \sum_{(j=1)}^{N} \mathbf{F}_i \right) \mathbf{I} \mathbf{F}_j \]

In Eq. (1), the velocity \( v_d \) of a particle \( \alpha \) is induced by the force \( \mathbf{F}_i \) by the surrounding fluid of each particle, where \( a \) is the diameter of particle, \( \eta \) is the viscosity of fluid, \( \gamma \) is the local velocity gradient of the flow.

**Simulation method**

The analysis of the movement of a platelet particle was carried out using the DEM. From the particle location at the previous time step, the acceleration was calculated using the sum of the forces of inertia, collision, and binding of the vWF and the Fibrinogen to it. At the same time, the influence of the flow field induced by the particle was calculated using the SDM. Finally, the velocity of a particle is calculated from the direct forces of other particles and the surrounding fluid. The position of a particle can be thus calculated by repeating integration. The collective movements of the particle group were analyzed in this way.

**RESULTS AND DISCUSSION**

We analyzed four conditions in which two different receptors had two activation statuses. The formation process of the platelet thrombus on the vessel wall where the subendothelial layer was exposed due to injury was thus modeled. In Figure 2 (a), a snap shot from the simulation results for the case in which neither GP Ib \( \alpha \) nor GP IIb/IIIa were activated is shown. In this case, no adhesive forces are experienced by the platelets, therefore the platelets flow by the vessel wall without stopping and no aggregation occurs. On the other hand, Figure 2 (b) shows the case in which both GP Ib \( \alpha \) and GP IIb/IIIa were ready to be activated. In this case, the vWF caused platelets adhere on the vessel wall first, and the platelets were fixed by the Fibrinogen when they stopped moving. Consequently, platelets not only adhered to the vessel wall, but the thrombus was formed piling up in to the intraluminal space in the vessel.

The time course of the total length of the platelet adhesion on the vessel wall equivalent to the occupying area of the thrombus in 3-dimensional case, is shown in Figure 3. We can see that the platelet adhered to the vessel wall when the GPIIb/IIIa was activated. Figure 4 shows the time course of the growth of the platelet thrombus in the vertical direction, that is, the time change of the height of the thrombus. Only when both GP Ib \( \alpha \) and GPIIb/IIIa were activated, the platelet on the primary thrombus grew in the blood flow. We could observe that platelets were adhered on the vessel wall and the platelet thrombi were found to develop into the intraluminal space of the vessel, only when both two kinds of receptors could be activated.

It became possible to analyze the thrombus formation process within the blood flow by the introduction of the model of the vWF, the Fibrinogen and two kinds of receptors on the platelet surface, by using a combination method of the DEM and the SDM. Moreover, the importance of the collaboration of two different types of receptors was suggested by comparing the results of the time course of the platelet adhesion with different activation conditions of each type of receptor. By introducing the SDM in the present study, in cooperation with the DEM whose single application was already reported in previous studies, mechanical interactions between the platelets and the plasma macromolecules became appropriately modeled and the method was considered to be applied wider physiological and pathological phenomena in the future studies.