

Optical control of intercellular communication between platelets during activation

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Motivation and Aim: Platelet activation is considered a cornerstone of hemostasis and thrombosis. One of the important features of this process is the feedback mechanism which allows platelets to activate each other. Current research is dedicated to the local platelet activation and investigation of activation spreading patterns between cells. In this study we used a previously developed method of activation via UV light [1]. Our hypothesis is that platelet activation in regions not illuminated by UV light occurs due to a positive feedback mechanism by ADP released from already activated platelets. Therefore, it is expected that the closer cells to the light source the faster will be an activation and the fluorescence intensity changes correspondingly.

Methods and Algorithms: In this study the “caged” analog of adenosine-diphosphate (CADP) is used to initiate reaction by releasing free ADP into the platelet-rich plasma sample triggered by UV light. Ca^{2+} -sensitive fluorescent label Fluo-4 is used to monitor changes in cytoplasmic calcium which increases during activation. A simple algorithm for estimating the activation rate was developed and tested. The procedure of estimating includes division of the frame into even rectangles, which subsequently constitute a matrix where each element is processed separately. Processing relies on obtaining the dependence of mean intensity on time and linear approximation of the fluorescence kinetics after the UV flash.

Results: Received coefficients have the meaning of speed of intensity change and agrees well with our hypothesis. On the basis of this data we can conclude that the closer to the boundary of non-illuminated region platelets are the lesser their activation rate will be. It can be assumed that platelets that were activated by cleavage of CADP become a secondary source of ADP.

Conclusion: The results obtained in the experiment clearly show intercellular communication between platelets. Further research will focus on mathematical model formulation, improvement of sample preparation and modification of the current set up system.

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References

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