Title: Probing the platelet lipidome from lipid structure to function

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Introduction: Platelet adhesion and activation are essential for primary hemostasis and critical for the development of acute thrombotic occlusion, the major pathophysiological mechanism underlying myocardial infarction or ischemic stroke. Platelets are activated by several prothrombotic mediators, including subendothelial collagen and thrombin. After the stimulation lipids are involved in key aspects of the subsequently following signaling cascade, appearing morphological changes, fusion with the plasma membrane (PM), cell-membrane scrambling, phosphatidylserine (PS) exposure at the outer leaflet of the PM and degranulation. Therefore, lipids are essential for platelet integrity and function and playing a fundamental role for the platelet lifespan and senescence. Various lipids have been described in resting and activated platelets. Nevertheless, no comprehensive study sequencing and describing the platelet lipidome from a quantitative perspective is available. Knowledge of concentrations is essential for comparing lipids between species or cross species, and to fully understand the mechanisms underlying initiation and propagation of platelet activation. Quantification of membrane and signaling lipids is clearly required for correct modeling of membrane fluidity, stiffness, or signaling.

Methods: Quantitative lipidomics, bioinformatics and functional platelet studies

Results: Platelet integrity and function critically depend on lipid composition. However, the lipid inventory in platelets was hitherto not quantified. Here, we examined the lipidome of murine platelets using lipid-category tailored protocols on a quantitative lipidomics platform. We could show that the platelet lipidome comprises almost 400 lipid species and covers a concentration range of 7 orders of magnitude. A systematic comparison of the lipidomics network in resting and activated murine platelets, validated in human platelets, revealed that <20% of the platelet lipidome is changed upon activation, involving mainly lipids containing arachidonic acid. Sphingomyelin phosphodiesterase-1 (Smpd1) deficiency resulted in a very specific modulation of the platelet lipidome with an order of magnitude upregulation of lysosphingomyelin (SPC), and subsequent modification of platelet activation and thrombus formation. In conclusion, this first comprehensive quantitative lipidomic analysis of platelets sheds light on novel mechanisms important for platelet function, and has therefore the potential to open novel diagnostic and therapeutic opportunities.

Conclusion: Platelet lipidome displays a huge dynamic range, the lipidome is dominated by 15 lipids in resting platelets, PI 18:0-20:4 is the major precursor for AA, the platelet lipidome is stable, changes < 20%, SPC 18:1;2 is bioactive and compromised platelet function

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