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Identification and characterization of zebrafish thrombocytes.

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Abstract

To analyse primary haemostasis in the zebrafish we have identified and characterized the zebrafish thrombocyte by morphologic, immunologic and functional approaches. Novel methods were developed for harvesting zebrafish blood with preservation of thrombocytes, and assaying whole blood adhesion/aggregation responses in microtitre plates. Light and electron microscopy of the thrombocyte illustrated morphological characteristics including the formation of aggregates, pseudopodia, and surface-connected vesicles analagous to the platelet canalicular system. Immunostaining with polyclonal antisera versus human platelet glycoproteins demonstrated the presence of glycoprotein Ib and IIb/IIIa-like complexes on the thrombocyte surface. Whole blood assays for adhesion/aggregation and ATP release showed ristocetin-induced adhesion without ATP release, and platelet agonist (collagen, arachidonic acid) induced aggregation with ATP release. Blood harvested from zebrafish treated with aspirin demonstrated inhibition of arachidonic acid induced aggregation and agonist induced ATP release, consistent with at least partial dependence on an intact cyclo oxygenase pathway. The combined morphologic immunologic and functional evidence suggest that the zebrafish thrombocyte is the haemostatic homologue of the mammalian platelet. Conservation of major haemostatic pathways involved in platelet function and coagulation suggests that the zebrafish is a relevant model for mammalian haemostasis and thrombosis.