ELSEVIER

Blood 142 (2023) 3977

The 65th ASH Annual Meeting Abstracts

POSTER ABSTRACTS

321.COAGULATION AND FIBRINOLYSIS: BASIC AND TRANSLATIONAL

Factor VIII Is an Endothelial Factor That Promotes Vessel Stability

Cristina Olgasi, PhD[1], *Alessia Cucci*[1], *Simone Assanelli*[1], *Chiara Sgromo*[1], *Chiara Borsotti*[1], *Roberto Santi*[2], *Antonia Follenzi*[1,2]

[1] Department of Health Sciences, University of Piemonte Orientale, Novara, Italy
[2] Dipartimento Attività Integrate Ricerca Innovazione, SSD Medicina Traslazionale, Alessandria, Italy

Background:
Hemophilia A (HA) is a rare bleeding disorder caused by the absence or dysfunction of factor VIII (FVIII). Clinical manifestations include spontaneous bleedings that primarily consist of hemarthroses and intracranial hemorrhages. Attenuated microvascular endothelial functionality is observed in HA patients suggesting a dysfunction in endothelial cells (ECs). Current therapeutic practice consists of replacement therapy with recombinant FVIII (rFVIII), either as needed to treat acute hemorrhages, or as prophylaxis to prevent bleeds. It is well established that FVIII is largely secreted by ECs and in particular by sinusoidal endothelial cells in the liver. Healthy and HA ECs can be isolated from the peripheral blood as blood outgrowth endothelial cells (BOECs) or can be differentiated from induced pluripotent stem cells (iPSCs). Here we show that, in addition to its role in blood coagulation, FVIII plays a role in endothelial cell functionality.
Aims:
To elucidate the potential impact of FVIII on EC stability and to investigate the effect of different rFVIII products on the maintenance of EC functionality.
Methods:
BOECs isolated from HA patients and healthy donors were used. HA BOECs were treated *in vitro* with B-domain deleted (BDD; simoctocog alfa) or full-length (FL) rFVIII products, and EC functionality was evaluated by tubulogenic, migration, permeability and proliferation assays. The impact of the different rFVIII products on vessel formation and permeability was evaluated *in vivo* in NOD/SCID γ-Null (NSG) and NOD/SCID γ-Null HA (NSG-HA) mice.
Results:
In vitro results demonstrated a weakening in tubulogenesis, migration potential and permeability in HA ECs vs healthy ECs and a significant enhancing of tubule network formation in HA BOECs treated with BDD-rFVIII or FL-rFVIII. Quantification of nodes, junctions, branches and the total length of the vessel-like structures confirmed that treatment of HA BOECs with BDD-rFVIII or FL-rFVIII improved HA BOEC function. Moreover, migration assays of HA BOECs treated with BDD-rFVIII or FL-rFVIII showed that the migration ability of HA BOECs was improved after treatment with either of the rFVIII products. We also evaluated whether rFVIII could ameliorate cell permeability. When treated with either rFVIII, the HA BOEC monolayer was less permeable to fluorescein isothiocyanate (FITC)-labeled dextran compared with that from non-treated HA BOECs. Vessel permeability assays *in vivo* demonstrated a significant reduction of dye extravasation in NSG-HA mice treated with rFVIII, with an improvement in mice treated with BDD-rFVIII compared with FL-rFVIII.
Conclusion:
In conclusion, information about the possible extra-coagulative role of FVIII may be crucial to understand the key molecular targets missing in HA patients at the cellular level that impair EC functionality. Knowledge of the possible effect of different rFVIII products on ECs functionality can lead to new therapeutic approaches potentially resulting in safer and more efficient treatment of HA.

Disclosures Follenzi: *Sparks:* Research Funding.

https://doi.org/10.1182/blood-2023-185948