VASCERN HHT

Workshop on Immunity, injury, and inflammation in HHT and HHT vessels, Dubrovnik, Croatia, June 2017

Claire L. Shovlin and Luisa M. Botella, Imperial College London, UK; CSIC Madrid, Spain.

These relatively new topics for the HHT field were discussed in an interactive, joint clinical and scientific workshop. The known HHT pathogenic gene variants (in *ENG*¸ *ALK1* and *SMAD4*) affect proteins expressed on endothelial cells, but it is often overlooked that all three proteins are also co-expressed by other cell types, including the hemangioblasts [1] that give rise to endothelial, myeloid and lymphoid lineages, and in macrophages following the process of monocyte-macrophage differentiation [2].

IMMUNITY: The first workshop section considered *immunity*. In an illustrative exercise, HHT pathogenic variants in *ENG* and *ALK1* were initially postulated to modify immune responses in different ways based on the higher prevalence of brain abscesses and other unusual infections in HHT1/*ENG* patients.

The importance of recognizing potential confounders was then emphasized:

(i) Brain abscesses and other deep-seated infections are predominantly found in HHT patients with PAVMs [3,4] and PAVMs are more common and severe in HHT1 patients;

(ii) Bacteremia (infected blood) is normal after dental and other procedures [5] with PAVM-associated abscesses attributed to impaired pulmonary capillary removal of infected blood-borne particles [3,4]

(iii) In the general population, bacterial infections are more severe in the setting of high iron levels [6], which, counterintuitively, are often found in HHT patients who use iron treatments and have transient iron overload states [4].

Nevertheless, the infections observed are unusual, and the discussion concluded with laboratory data demonstrating lesser magnitude responses by lipopolysaccharide (LPS)-stimulated macrophages from a myeloid-specific ENG knockout model (Engfl/fl LysMCre mice): The ENG-deficient macrophages demonstrated lower expression of proinflammatory cytokines IL-1, IL-12, IL-6, CCL-20, and thrombospondin 1 compared to those with normal ENG expression [7].

The workshop presentations increased the proportion of participants who considered that HHT immunity was weaker than normal from 19 to 37%, although the most common response from patients was that in their day-to-day experience, their immune systems seemed stronger.

INJURY: The second section reviewed laboratory data that *injury* increases endothelial expression of ENG and ALK1 [8,9] and that vascular repair is abnormal if ENG or ALK1 are deficient [8].

(i) HHT-independent injuries (such as external trauma [9] mechanical stretching of vessels during respiration and peristalsis; gastrointestinal tract acidity; infection), and

(ii) HHT-specific injuries were discussed. The latter include locally modified flow through HHT vessels, generally increased flow through all vessels in response to the high cardiac outputs [10] and the emerging evidence that therapeutic iron treatments required by most HHT patients may directly injure the endothelium [11] In unbiased, replicate surveys, approximately 1 in 20 HHT patients using the treatments reported that iron tablets or infusions precipitate nosebleeds[12,13]

The presentations increased the proportion of participants considering HHT patients respond less well to injury from 44 to 66%, but the most common response from patients was that their responses were no different to normal.

INFLAMMATION: In the final workshop section, *inflammation* was discussed in more detail.

Impaired resolution of inflammation in pan-ENG heterozygous mice, in a chronic colitis model [14] is now supplemented by similar findings in mice with myeloid-specific ENG deficiency [7] ENG-deficient macrophages from Engfl/fl LysMCre knockout mice displayed reduced cytokine expression in response to peritoneal LPS (as noted above), and reduced phagocytic activity [7] While the ENG-deficient mice were more likely to develop spontaneous infections by opportunistic bacteria, they also demonstrated better survival in the LPS/septic shock model, attributed to less exuberant inflammatory responses [7]

The take-home messages from the workshop were

- the importance of future immunophenotyping of HHT patients, *and*
- incorporation of the discussed processes into HHT pathogenic models:

85% of attendees thought that immunity and inflammation would influence the development of abnormal blood vessels in HHT.

References

1. Zhang L, Magli A, Catanese J, Xu Z, Kyba M, Perlingeiro RC (2011) Modulation of TGF-beta signaling by endoglin in murine hemangioblast development and primitive hematopoiesis. Blood 118:88-97

2. Lastres P, Bellon T, Cabanas C, Sanchez-Madrid F, Acevedo A, Gougos A, Letarte M, Bernabeu C (1992) Regulated expression on human macrophages of endoglin, an Arg-Gly-Asp-containing surface antigen. Eur J Immunol 22:393-397

3. Shovlin CL (2014) Pulmonary arteriovenous malformations. Am J Respir Crit Care Med 190:1217-1228

4. Boother EJ, Brownlow S, Tighe HC, Bamford KB, Jackson JE, Shovlin CL (2017) Cerebral abscess associated with odontogenic bacteremias, hypoxemia, and iron loading in immunocompetent patients with right-to-left shunting through pulmonary arteriovenous malformations. Clin Infect Dis

5. Limeres Posse J, Alvarez Fernandez M, Fernandez Feijoo J, Medina Henriquez J, Lockhart PB, Chu VH, Diz Dios P (2016) Intravenous amoxicillin/clavulanate for the prevention of bacteraemia following dental procedures: a randomized clinical trial. J Antimicrob Chemother 71:2022-2030

6. Bruhn KW, Spellberg B (2015) Transferrin-mediated iron sequestration as a novel therapy for bacterial and fungal infections. Curr Opin Microbiol 27:57-61

7. Ojeda-Fernandez L, Recio-Poveda L, Aristorena M, Lastres P, Blanco FJ, Sanz-Rodriguez F, Gallardo-Vara E, de Las Casas-Engel M, Corbi A, Arthur HM, Bernabeu C, Botella LM (2016) Mice Lacking Endoglin in Macrophages Show an Impaired Immune Response. PLoS Genet 12:e1005935

8. Botella LM, Sanz-Rodriguez F, Komi Y, Fernandez-L A, Varela E, Garrido-Martin EM, Narla G, Friedman SL, Kojima S (2009) TGF-beta regulates the expression of transcription factor KLF6 and its splice variants and promotes co-operative transactivation of common target genes through a Smad3-Sp1-KLF6 interaction. Biochem J 419:485-495

9. Park SO, Wankhede M, Lee YJ, Choi EJ, Fliess N, Choe SW, Oh SH, Walter G, Raizada MK, Sorg BS, Oh SP (2009) Real-time imaging of de novo arteriovenous malformation in a mouse model of hereditary hemorrhagic telangiectasia. J Clin Invest 119:3487-3496

10. Shovlin CL (2015) Circulatory contributors to the phenotype in hereditary hemorrhagic telangiectasia. Front Genet 6:101

11. Mollet IG, Patel D, Govani FS, Giess A, Paschalaki K, Periyasamy M, Lidington EC, Mason JC, Jones MD, Game L, Ali S, Shovlin CL (2016) Low Dose Iron Treatments Induce a DNA Damage Response in Human Endothelial Cells within Minutes. PLoS One 11:e0147990

12. Shovlin CL, Gilson C, Busbridge M, Patel D, Shi C, Dina R, Abdulla FN, Awan I (2016) Can Iron Treatments Aggravate Epistaxis in Some Patients With Hereditary Hemorrhagic Telangiectasia?. Laryngoscope 126:2468-2474

13. Shovlin CL, Patel T, Jackson JE (2016) Embolisation of PAVMs reported to improve nosebleeds by a subgroup of patients with hereditary haemorrhagic telangiectasia. ERJ Open Res 2:00035-2016. eCollection 2016 Apr

14. Peter MR, Jerkic M, Sotov V, Douda DN, Ardelean DS, Ghamami N, Lakschevitz F, Khan MA, Robertson SJ, Glogauer M, Philpott DJ, Palaniyar N, Letarte M (2014) Impaired resolution of inflammation in the Endoglin heterozygous mouse model of chronic colitis. Mediators Inflamm 2014:767185

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