Sebia Hydragel 5 von Willebrand Multimers - A new and rapid von Willebrand factor (VWF) multimer screening method to aid subtyping of type 2 von Willebrand disease (VWD)

Goodfellow KJ[1], Bowyer AE [1], Nouadje G [2], Beaulieu G [2], Kitchen S [1], Makris M [1]
[1]Sheffield Haemophilia and Thrombosis Centre, UK [2] Sebia R&D, France

Background

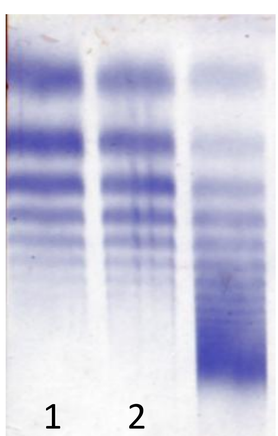
Analysis of VWF multimers is essential for diagnosis and classification of VWD. Reduction of high molecular weight multimers (HMWM) is observed in types 2A and 2B VWD whilst a normal pattern is seen in types 2M and 2N. The suitability of the Hydragel 5 von Willebrand multimers (Sebia, France) for subtyping of type 2 VWD was evaluated as part of a larger ongoing comparison study

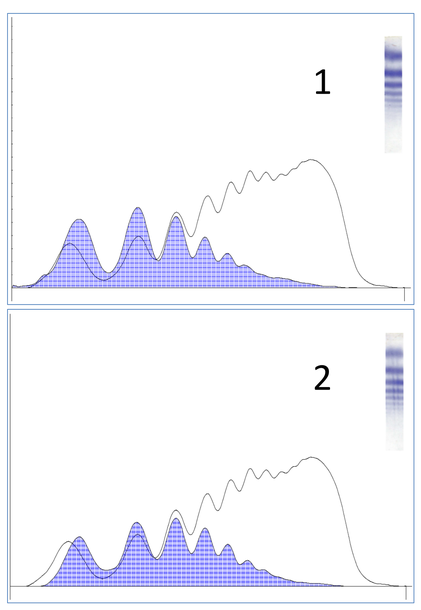
Methods

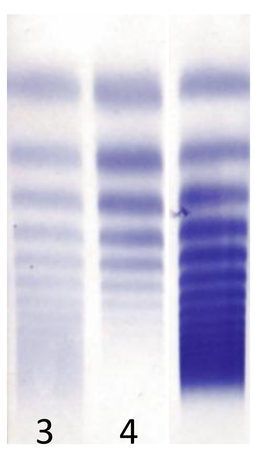
The multimer pattern of 54 samples from previously diagnosed VWD patients (Types 2A n= 25, 2B n= 6, 2M n= 18, type 2N n= 5) were analysed by two methods. The semi-automatic Hydragel 5 von Willebrand Multimers, using Hydrasys 2 Scan instrumentation involved electrophoresis on agarose gel, immunofixation with anti-VWF, visualisation using peroxidase-labelled antibody and densitometry of the multimers. The in-house method involved agarose gel electrophoresis and visualisation using alkaline phosphatase-conjugated antibody

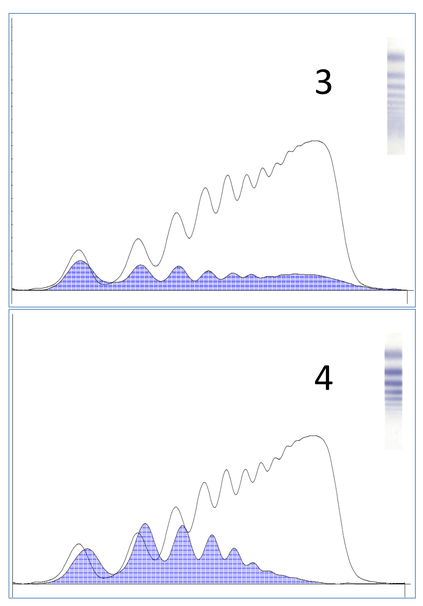
Results

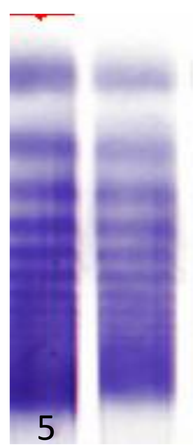
All type 2A samples showed a reduction in HMWM with Hydragel 5 von Willebrand multimers. One individual produced a normal multimer pattern using the in-house method, whereas other family members showed a reduction in both.17 type 2M samples had normal multimers, however, a number of these showed flattened HMWM peaks with densitometry. One 2M sample with a very low VWF level showed normal pattern with the in-house method but reduction of HMWM with the Hydragel 5 von Willebrand Multimers. All type 2B and type 2N samples produced expected multimer patterns with both methods.

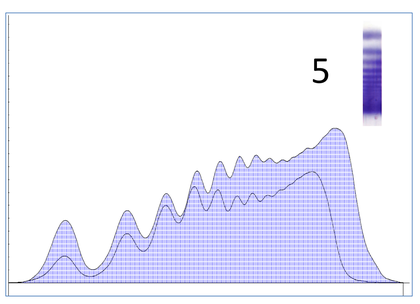












Track	VWF AG	VWF AC	RATIO ACT/AG	VWD Type
1	45	13	0,29	2A
2	/	9	/	2A

Track	VWF AG	VWF AC	RATIO ACT/AG	VWD Type
3	31	13	0,42	2B
4	36	10	0,28	2M
5	88	94	1,1	2N

Conclusions

52/54 gave the expected multimer pattern with the Hydragel 5 von Willebrand multimers. Densitometry, availableith this commercial test, highlighted heterogeneity in HMWM concentration between family members with type 2A. The densitometry of some of the 2M samples appeared to have a slight reduction of HMWM with flattened peaks. This may prove to be a be a unique pattern for some type 2M individuals using the Hydragel 5 von Willebrand multimers and may be related to the genetic mutation present. The Hydragel 5 von Willebrand multimers is suitable for use in the classification of type 2 VWD with the helpful addition of densitometry.