

P009 Expression and functional characterization of the G1 domain from human versican.

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Versican forms link protein-stabilized complexes with HA that contribute to the elasticity of blood vessels. HA binding is mediated through its N-terminal G1 domain (VG1), which is composed of an Ig domain and two contiguous Link modules. Currently no high-resolution structure is available for VG1 or for Type C HA-binding domains. Previously, VG1 was expressed in *Drosophila* S2 cells as two different N-glycoforms (dVG1) and shown to interact with HA but that this was unaffected by deglycosylation. Initial crystallization attempts were limited by low yields (~0.5 mg/litre) and perhaps glycosylation. To generate material more suitable for structural studies VG1 was expressed in *E. coli* (eVG1), refolded, and purified by ion exchange and gel filtration chromatography. Intrinsic fluorescence analysis and a microtitre plate assay indicated that eVG1 and dVG1 are structurally equivalent and exhibit identical HA-binding properties. Thus, we have developed a method of producing correctly folded VG1 in high yields (~7 mg/litre), which is facilitating further crystallization trials and enabling us to test, using MALLS, the hypothesis that the versican-HA interaction displays all-or-none cooperativity. We have also shown that biotinylated-VG1 can be used to visualise HA in tissue sections.