



Deposited via The University of Leeds.

White Rose Research Online URL for this paper:

<https://eprints.whiterose.ac.uk/id/eprint/134783/>

Version: Accepted Version

Proceedings Paper:

Shah, P, Cordell, P, Yuldasheva, N et al. (2017) Insulin-Like Growth Factor Binding Protein-2: A Putative Agent for Therapeutic Angiogenesis Acting via Its Arginine-Glycine-Aspartic Acid (RGD) Domain. In: Circulation. Resuscitation Science Symposium, 11-15 Nov 2017, Anaheim, Calif., U.S.A.. American Heart Association. Article no: A16460. ISSN: 0009-7322. EISSN: 1524-4539.

https://doi.org/10.1161/circ.136.suppl_1.16460

(c) 2017 by American Heart Association, Inc. This is an author produced version of an abstract published in Circulation. Uploaded in accordance with the publisher's self-archiving policy.

Reuse

Items deposited in White Rose Research Online are protected by copyright, with all rights reserved unless indicated otherwise. They may be downloaded and/or printed for private study, or other acts as permitted by national copyright laws. The publisher or other rights holders may allow further reproduction and re-use of the full text version. This is indicated by the licence information on the White Rose Research Online record for the item.

Takedown

If you consider content in White Rose Research Online to be in breach of UK law, please notify us by emailing eprints@whiterose.ac.uk including the URL of the record and the reason for the withdrawal request.

Insulin-Like Growth Factor Binding Protein-2: A Putative Agent for Therapeutic Angiogenesis Acting via Its Arginine-Glycine-Aspartic Acid (RGD) Domain

Pooja Shah, Paul Cordell, Nadira Yuldasheva, Anna Skromna, Jess Smith, Alex Bruns, Natalie Haywood, Hema Viswambharan, Kirti Kain and Stephen Wheatcroft

Therapeutic angiogenesis is under investigation to restore tissue perfusion in ischaemic heart disease and peripheral arterial disease. Insulin-like growth factor binding protein-2 (IGFBP-2) has been attributed with pro-angiogenic activity but the molecular mechanisms have not been established. Structurally, IGFBP-2 possesses insulin-like growth factor (IGF) binding-, integrin recognition- (Arginine-Glycine-Aspartic Acid (RGD)) and heparin binding- sites and a nuclear localisation signal which may be implicated in its cellular actions.

We hypothesise that IGFBP-2 can be exploited to promote therapeutic angiogenesis through cellular actions mediated by one or more of its structural domains.

Restoration of limb perfusion was quantified by laser Doppler imaging following hind limb ischemia in mice expressing human IGFBP-2. Mechanistic studies were carried out in cultured human umbilical vein endothelial cells (HUVEC). Functional angiogenic responses were explored with tube formation and cell adhesion assays. Wild type IGFBP-2 (^{WT}IGFBP-2) and recombinant IGFBP-2 containing a non-functional IGF binding site (IGF Δ) or mutated RGD domain (RGD Δ) were generated to identify the structural domain responsible for the pro-angiogenic signalling mechanisms.

Transgenic expression of hIGFBP-2 in mice significantly enhanced restoration of blood flow to the limb at day 7 compared to WT littermates ($p < 0.05$). Both ^{WT}IGFBP-2 and IGF Δ IGFBP-2, significantly increased tube formation in HUVEC. Interestingly, RGD Δ IGFBP-2 stimulated tube formation was greatly decreased compared to ^{WT}IGFBP-2 ($p < 0.05$). ^{WT}IGFBP-2 increased HUVEC adhesion to fibronectin (1.6 fold); however this effect was absent with RGD Δ IGFBP-2. Stimulation of endothelial cells with ^{WT}IGFBP-2 induced Akt phosphorylation (1.3 fold). RGD Δ IGFBP-2 did not significantly activate Akt. Neither ^{WT}IGFBP-2 nor the mutants induced the canonical angiogenic signalling factors, such as eNOS, FAK or MAPK phosphorylation.

IGFBP-2 promotes angiogenesis in vitro and in vivo, supporting its further investigation for therapeutic angiogenesis in ischaemic disorders. For the first time, we demonstrate that the RGD domain of IGFBP-2 appears to play a critical role in angiogenic activity.