

PROJET "SCAR FREE" PROGRAMME PHYSIO 2007

AN INTEGRATIVE APPROACH TO SCAR FREE HEALING BASED ON AN EMERGING FAMILY OF EXTRACELLULAR PROTEASES

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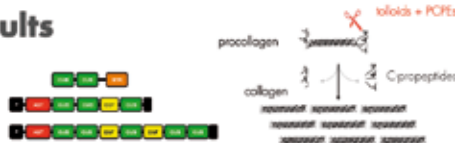
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Objectives

Fibrosis, or scarring, is one of the most important causes of mortality worldwide. Most major organs are affected (cardiovascular system, liver, lung, kidney, skin ...) including cornea, where wounding, chemical burns, infections and refractive surgery often result in partial or complete loss of vision through the appearance of corneal opacities, or haze. Fibrosis is characterised by excessive synthesis of extracellular matrix (ECM), whose composition and organisation differs markedly from normal tissue. The objectives of the SCAR FREE project are to study the role played by an emerging family of extracellular proteinases, called BMP-1/tolloid-like, in corneal wound healing, including biochemical and structural studies, with the aim of identifying new therapeutic targets for the prevention of corneal scarring.

Results

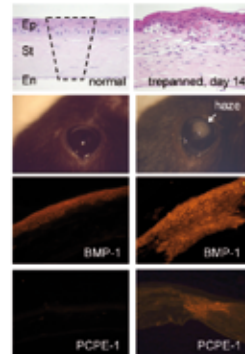
PCPE-1,2
BMP-1
mTD, mTL-1, 2



There are 4 forms of BMP-1/tolloid-like proteinase (left), where BMP-1 is the most abundant in cornea [6]. They cleave many substrates, involved mainly in ECM assembly and growth factor activation, including two new ones discovered during the course of this contract [2,10]. Substrates include procollagens (right), where C-propeptide cleavage, the rate limiting step in fibril formation, is specifically enhanced by ECM proteins called PCPEs. We also found that mepriins, which share a similar catalytic domain, also process procollagens [4].

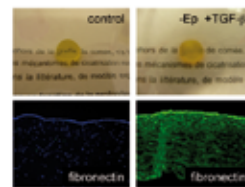


Left: Superstimulation of procollagen processing in the presence of heparan sulphate proteoglycans [3]. Right: Structure of the PCPE-1/C-propeptide complex [1,5].



Mouse model of corneal scarring [6], induced by full-thickness incision (zone outlined). Epithelium, stroma and endothelium are indicated. Scarring appears as corneal haze. Immunohistochemistry shows up-regulation of BMP-1 and PCPE-1 expression in the scar, which are also seen by Q-PCR, along with increased expression of collagen III.

Synthetic and naturally occurring inhibitors of tolloid proteinases are also being studied [8,9], as is a knockout model for PCPE-1 (collaboration D. Greenspan, University of Wisconsin, Madison, USA).



Human *ex vivo* model of corneal scarring [7], using donor corneas unsuitable for grafting. Absence of epithelium (with added TGF-β) results in corneal opacity and increased expression of e.g. fibronectin.

Impact

Our results give important new insights into the molecular mechanism of tolloid proteinase regulation. This information will be useful in the search for new therapeutic strategies aimed at preventing fibrosis. The *in vivo* mouse model validates tolloid proteinases and PCPEs as potential therapeutic targets. The development of a human *ex vivo* model provides new possibilities for the testing of anti-fibrotic drugs.

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5. J.M. Bourhis, D. Kronenberg, S. Vadon-Le Goff et al., submitted.
6. D. Massoudi, P. Fournié, A. Erraud et al., submitted.
7. H. Janin-Magnificat, M.R. Rovère, S. Galiacy et al., submitted.
8. S. Galiacy, C. Bijakowski; D. Massoudi et al., in preparation.
9. C. Bijakowski, F. Delolme, S. Vadon-Le Goff et al., in preparation.
10. F. Delolme, R. Capomaccio, I. Zanella-Cléon et al., in preparation.

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