Experimental Eye Research 142 (2016) 83–91

Contents lists available at ScienceDirect

Experimental Eye Research

journal homepage: www.elsevier.com/locate/yexer

Review

Current perspectives on the role of orbital fibroblasts in the pathogenesis of Graves' ophthalmopathy

Willem A. Dik*, Sita Virakul, Leendert van Steensel

Department of Immunology, Laboratory Medical Immunology, Erasmus MC, University Medical Center Rotterdam, The Netherlands

ARTICLE INFO

Article history:
Received 1 September 2014
Received in revised form
5 February 2015
Accepted in revised form 10 February 2015

Keywords:
Graves' ophthalmopathy
Orbital fibroblast
Adipocyte
Fibrocyte
Inflammation
Fibrosis
Cytokine
Growth factor
Hyaluronan

ABSTRACT

Graves' ophthalmopathy (GO) is an extra-thyroidal complication of Graves' disease (GD; Graves' hyperthyroidism) characterized by orbital tissue inflammation, expansion, remodeling and fibrosis. Although the initiating trigger of GO is still indistinct, excessive orbital fibroblast activity is at the heart of its pathogenesis. Orbital fibroblasts are activated by cellular interactions with immune cells and the soluble factors they secrete. Orbital fibroblasts, especially from GO patients, express the thyrotropin receptor (TSH-receptor; TSHR), and activation of the orbital fibroblast population by stimulatory autoantibodies directed against the TSHR may provide an important link between GD and GO. Furthermore, stimulatory autoantibodies directed against the insulin-like growth factor-1 receptor have been proposed to contribute to orbital fibroblast activation in GO. Activated orbital fibroblasts produce inflammatory mediators thereby contributing to the orbital inflammatory process in GO. Moreover, orbital fibroblasts exhibit robust proliferative activity and extracellular matrix (especially hyaluronan) synthesizing capacity and can differentiate into adipocytes and myofibroblasts with disease progression, thereby contributing to tissue expansion/remodeling and fibrosis in GO. Orbital fibroblasts, especially those from GO patients, exhibit a hyper-responsive phenotype when compared to fibroblasts from other anatomical regions, which may further contribute to GO pathogenesis. Fibrocytes have been identified as additional source of orbital fibroblasts in GO, where they may contribute to orbital tissue inflammation, adipogenesis and remodeling/fibrosis. This review addresses our current view on the role that orbital fibroblasts fulfill in GO pathogenesis and both established as well as less established not fully crystallized concepts that need future studies will be discussed.

1. Introduction

Graves' disease (GD; Graves' hyperthyroidism) is one of the most common autoimmune disorders and accounts for the majority of cases of hyperthyroidism. Hyperthyroidism is a pathological syndrome in which tissue is exposed to excessive amounts of thyroid hormone, causing typical symptoms as nervousness or anxiety, weight loss, palpitations, heat intolerability and fatique. Hyperthyroidism in GD is caused by specific autoantibodies that stimulate the thyrotropin receptor (TSH-receptor; TSHR), thereby mimicking the effect of pituitary thyroid stimulating hormone (TSH) (Cooper, 2003).

Graves' ophthalmopathy (GO), also referred to as thyroid eye disease, is an extra-thyroidal complication that develops in ~25–50% of patients with GD and is characterized by inflammation and extensive remodeling of the soft tissues surrounding the eyes (Bahn, 2010). Most patients exhibit extraocular muscle and adipose/connective tissue volume increase, while in some patients either extraocular muscle enlargement or adipose/connective tissue expansion may predominate (Bahn, 2010). Fibroblast and adipocyte numbers are increased in extraocular muscle and adipose/connective tissue from GO patients, leading to collagen and glycosaminoglycan accumulation between the muscle fibers and within the adipose/connective tissue (Smith et al., 1989a). Clinical symptoms of GO result from the increased orbital tissue volume within the noncompliant space-limited bony orbit and comprise of upper eyelid retraction, edema, erythema of the periorbital tissues and conjunctivae, and proptosis. Keratitis can occur in case of severe and prolonged proptosis, while optic neuropathy can result from optic nerve compression (Smith et al., 1989a; Bahn, 2010).

* Corresponding author. Department of Immunology, Laboratory Medical Immunology, Erasmus MC, University Medical Center Rotterdam, Wytemaweg 80, 3015 CN Rotterdam, The Netherlands.
E-mail address: w.dik@erasmusmc.nl (W.A. Dik).

http://dx.doi.org/10.1016/j.exer.2015.02.007
0014-4835/

Early active GO is characterized by infiltration of the extraocular muscles and adipose/connective tissue with mononuclear cells, primarily $CD4^+$ T-lymphocytes, some $CD8^+$ T-lymphocytes, monocytes, macrophages, B-lymphocytes and plasma cells (Weetman et al., 1989; Kahaly et al., 1994; Yang et al., 1999; Pappa et al., 2000; Eckstein et al., 2004; Bahn, 2010). Mast cells are more abundant in the late fibrotic disease phase (Smith et al., 1989a; Boschi et al., 2005; van Steensel et al., 2012a). These inflammatory cells activate orbital fibroblasts via the secretion of inflammatory mediators (e.g. cytokines) or by direct cellular interaction (Bahn, 2010). Moreover, orbital fibroblasts in GO may be activated by stimulatory autoantibodies directed against the TSHR and the insulin-like growth factor-1 receptor (IGF-1R) (Bahn, 2010; Smith et al., 2012). The activated orbital fibroblasts increase their proliferative activity, produce inflammatory mediators, differentiate into adipocytes and myofibroblasts and produce excess amounts of extracellular matrix (ECM) components. Thereby, orbital fibroblasts fulfill central roles in orbital inflammation and tissue remodeling in GO. This activation, combined with several unique properties and heterogeneity within the orbital fibroblast pool, has led to the concept that orbital fibroblasts represent the central cell type in the pathogenesis of GO. In this review important effector functions and characteristics of orbital fibroblasts that contribute to the pathogenesis of GO will be discussed.

2. Orbital fibroblasts contribute to orbital inflammation

The inflammatory environment within GO orbital tissue is determined by soluble and cellular components and strongly influences orbital fibroblast behavior. In early active GO, T-helper 1 (Th_1)-lymphocytes dominate and Th_1-like cytokines (including a.o. interferon (IFN)-γ, interleukin (IL)-2 and tumor necrosis factor (TNF)-α) that facilitate cell mediated immunity are abundantly present. Although less evident, Th_2-lymphocytes and associated cytokines (including IL-4 and IL-10) may dominate the later disease stage characterized by tissue remodeling and fibrosis (late GO), fitting the current paradigm that Th_2-like cytokine responses predominate in chronic inflammation and fibrosis (de Carli et al., 1993; Aniszewski et al., 2000; Hiromatsu et al., 2000; Wakelkamp et al., 2003; Bahn, 2010; Wick et al., 2013). Other inflammatory cell types, including monocytes, macrophages and mast cells also contribute to the increased orbital cytokine/growth factor levels in GO (Kumar and Bahn, 2003; van Steensel et al., 2012a).

The effect of several cytokines and growth factors elevated in GO orbital tissue on orbital fibroblast inflammatory activity has been examined. IFN-γ stimulates the production of chemokine (C—C motif) ligand (CCL)2, a chemotactic factor for monocytes, as well as T-lymphocyte chemoattractants such as chemokine (C-X-C motif) ligand (CXCL)9, CXCL10 and CXCL11, which is synergistically enhanced by TNF-α (Elner et al., 1998; Antonelli et al., 2006, 2009). Cytokines and growth factors such as IL-1β, TNF-α and platelet-derived growth factor (PDGF)-AA, PDGF-AB and PDGF-BB also stimulate orbital fibroblasts to produce cytokines/chemokines like CCL2, CCL5, CCL7, IL-6, IL-8, and IL-16 that are collectively involved in recruitment and activation of monocytes, T-lymphocytes, B-lymphocytes and mast cells (Elner et al., 1998; Pritchard et al., 2002; Chen et al., 2005; Hwang et al., 2009; van Steensel et al., 2010; van Steensel et al., 2012a). Moreover, IL-1β and leukoregulin stimulate prostaglandin E_2 (PGE_2) production by orbital fibroblasts (Wang et al., 1996; Han and Smith, 2006). PGE_2 stimulates B-lymphocyte maturation, activates mast cells and induces Th_2 skewing, but also stimulates IL-6 production by orbital fibroblasts (Betz and Fox, 1991; Roper et al., 1995; Raychaudhuri et al., 2010; Kuehn et al., 2011).

Leukocyte infiltration and activation in tissue not only depends on local chemokine gradients, but also requires expression of adhesion and co-stimulatory molecules on leukocytes, endothelial cells and tissue resident cells. Intercellular adhesion molecule (ICAM)-1 expression on orbital fibroblasts is upregulated by IL-1α, IL-1β, IFN-γ, TNF-α (Heufelder and Bahn, 1992; Li et al., 2000; Cawood et al., 2006). The co-stimulatory molecule CD40, highly expressed by orbital fibroblasts from GO patients, is further upregulated by IFN-γ stimulation (Hwang et al., 2009). CD40-CD154 ligation is involved in physical interactions between orbital fibroblasts and T-lymphocytes in GO and enhances ICAM-1 expression as well as cytokine and prostaglandin production (e.g. CCL2, IL-1α, IL-6, IL-8, PGE_2) by orbital fibroblasts (Cao et al., 1998; Sempowski et al., 1998; Hwang et al., 2009; Zhao et al., 2010).

Collectively these data illustrate that orbital fibroblasts, through the production of inflammatory molecules, are involved in regulating the orbital inflammatory process in GO where they orchestrate leukocyte recruitment and activation.

3. Orbital fibroblasts contribute to orbital tissue expansion

Proliferation, extracellular matrix production (especially hyaluronan) and differentiation of orbital fibroblasts into adipocytes and myofibroblasts are important determinants of orbital tissue volume expansion and fibrosis in GO (Smith et al., 1989a; Bahn, 2010) and will be discussed hereunder.

3.1. Orbital fibroblast proliferation

Fibroblast proliferation is an important contributor to tissue remodeling and fibrotic responses (Wynn, 2008). The basal proliferative activity of GO orbital fibroblasts was found to be higher than that from normal orbital fibroblasts (Heufelder and Bahn, 1994). In addition, cellular interactions such as CD40-CD154 ligation between T-lymphocytes and orbital fibroblasts, but also various cytokines and growth factors, including IL-4, insulin-like growth factor (IGF)-1, PDGF and transforming growth factor (TGF)-β more strongly increase the proliferation rate of GO orbital fibroblasts than that of control orbital fibroblasts (Heufelder and Bahn, 1994; Feldon et al., 2005). Still, studies on this are not always consistent, as it has also been described that PDGF-BB stimulates proliferation of GO and control orbital fibroblasts equally and that TGF-β has no effect on orbital fibroblast proliferation (van Steensel et al., 2009). PDGF-BB was found to be a stronger mitogen for orbital fibroblasts than PDGF-AB, which in turn is more potent than PDGF-AA (van Steensel et al., 2012a). This may be related to the more abundant expression of the PDGF-receptor β chains by orbital fibroblasts compared to the PDGF-receptor α chains, as PDGF-BB signals through all PDGF-receptor dimers (αα,αβ,ββ), while PDGF-AB activates αα,αβ receptor dimers and PDGF-AA only activates αα receptor dimers (van Steensel et al., 2012a). The picture that emerges is that GO orbital fibroblasts are extremely sensitive to mitogenic factors and that exaggerated proliferation by these cells contributes to orbital tissue expansion and fibrosis in GO.

3.2. Hyaluronan production by orbital fibroblasts

GO orbital tissue contains increased amounts of non-sulfated glycosaminoglycans (especially hyaluronan) as well as collagen, which are produced by orbital fibroblasts (Smith et al., 1989a). Hyaluronan is the ECM component mostly contributing to orbital tissue expansion in GO. Hyaluronan is estimated to occupy ~75000 times the volume of that of an equivalent weight of collagen, which is mainly related to its massive water binding capacity (Smith et al., 1989a). Hyaluronan synthesis is regulated by cell membrane expressed hyaluronan synthases (HASs), of which three different