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Redox mechanisms in autoimmune thyroid eye disease

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ABSTRACT

Thyroid eye disease (TED) is an autoimmune condition affecting the orbit and the eye with its adnexa, often occurring as an extrathyroidal complication of Graves' disease (GD). Orbital inflammatory infiltration and the stimulation of orbital fibroblasts, triggering de novo adipogenesis, an overproduction of hyaluronan, myofibroblast differentiation, and eventual tissue fibrosis are hallmarks of the disease. Notably, several redox signaling pathways have been shown to intensify inflammation and to promote adipogenesis, myofibroblast differentiation, and fibrogenesis by upregulating potent cytokines, such as interleukin (IL)-1β, IL-6, and transforming growth factor (TGF)-β. While existing treatment options can manage symptoms and potentially halt disease progression, they come with drawbacks such as relapses, side effects, and chronic adverse effects on the optic nerve. Currently, several studies shed light on the pathogenetic contributions of emerging factors within immunological cascades and chronic oxidative stress. This review article provides an overview on the latest advancements in understanding the pathophysiology of TED, with a special focus of the interplay between oxidative stress, immunological mechanisms and environmental factors. Furthermore, cutting-edge therapeutic approaches targeting redox mechanisms will be presented and discussed.

1. Introduction

Thyroid eye disease (TED) is a disfiguring and profoundly debilitating condition, with the potential to lead to irreversible visual impairment, making it a matter of significant concern for both patients and society. The etiology of this disease is multifactorial, closely associated with Graves' disease (GD), and characterized by a complex network of cytokines and immune-mediated processes that sustain chronic inflammation, immune cell recruitment, activation of orbital fibroblasts (OFs), and their differentiation into adipocytes or myofibroblasts. Alongside immune signaling, oxidative stress plays a central role, capable of triggering OF activation, leading to cell proliferation and adipogenesis.

In this context, our objective is to provide an updated and comprehensive overview of TED's pathophysiology, with a particular focus on recent advances in the understanding of redox molecular signaling. Additionally, we discuss cutting-edge pharmacological strategies targeting redox signaling mechanisms aimed at improving patient outcomes in the management of this complex disorder.

2. Pathophysiology of thyroid eye disease

Thyroid dysfunction is critically implicated in the onset and progression of TED. Thyrotropin receptor (TSH-R) autoantibodies (TSH-R-Ab) are the pivotal players during GD. Hyperthyroidism and their cascades lead to wide escalation of autoimmune reactions against molecules expressed in both orbit and thyroid, finally causing chronic inflammation, deposition of extracellular matrix, myofibroblast differentiation and adipogenesis, in the eye specifically stimulating OFs [1–3]. In GD, these antibodies are routinely evaluated through competitive immune binding assays and thereby are also called TSH-R-binding inhibitory immunoglobulins (TBII) [4]. Importantly, cell-based assays allow a distinction within TSH-R-Ab, into three subgroups: (1) TSH-R stimulating (TSAb), or thyroid-stimulating immunoglobulins (TSI); (2) TSH-R blocking (TBAb), also called thyroid blocking immunoglobulins (TBI); and (3) TSH-R neutral antibodies, without a functional effect on the TSH-R and a still unclear clinical relevance [3–6]. In fact, neutral autoantibodies may trigger oxidative stress-related signaling cascades, inducing apoptotic effects and escalation of the autoimmune response in

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GD [4,7]. Serum levels of TSH-R-Abs in general, and TSAb in particular are associated with clinical disease activity and severity of TED, thus emphasizing their role as a crucial diagnostic and prognostic tool [4,8–10]. TSAb are also present in the vast majority of patients with autoimmune Hashimoto's thyroiditis and associated TED [11]. Thyroid dysfunction with increased serum TSH levels (hypothyroidism) activates the TSH-R expressing orbital target cells leading to enhanced release of hydrophilic mucopolysaccharides, local edema, swelling of eye muscles causing proptosis [2,12]. A recent work has illuminated the association between the immunoglobulin G4-related disease (IgG4-RD) and severe TED, with elevated IgG4 representing a risk factor for TED onset [13].

Detection of serum TSAb positivity in TED and/or GD subjects is regarded as a fundamental diagnostic tool, effective in suggesting management, timing of interventions, differential diagnosis, prognosis, and further being predictor, in case of elevated autoantibody levels, of limited therapeutic outcomes and of higher relapse rates [4,8–10,14–16]. TSH-R-Ab in general and TSAb in particular exhibit a robust correlation with the phenotype, activity, and severity of TED [15,17,18]. Notably, elevated TSAb titers can serve as an indicative marker for patients experiencing recent onset of DON, necessitating prompt intervention [19]. In contrast, TBAbs have demonstrated comparatively lower predictive potential [19–21]. However, the measurement of TBAb may prove beneficial in evaluating patients with suspected autoimmune-induced hypothyroidism [22]. It is noteworthy that approximately 6–7% of TED subjects are either hypothyroid, due to concomitant autoimmune thyroiditis or euthyroid without any present thyroid pathology [23,24].

In general, the key pathomechanism driving the abnormal alterations in TED is the stimulation of OFs by TSAbs. This stimulation sets off a cascade of signaling pathways in both immune cells and OFs, culminating in the synthesis of various inflammatory mediators. These mediators play a crucial role in adipogenesis, the deposition of extracellular matrix, and the enlargement of extraocular muscles. Fig. 1 offers a schematic representation of the primary alterations resulting from autoimmune stimulation on OFs induced by TSAbs. This process involves muscle tissues, adipose tissues, and connective tissues in the retrobulbar area, with specific cytokines playing a pivotal role in instigating the aberrant autoimmune reactivity.

In the following sections, we highlight the complex immune- and redox-related pathomechanisms occurring in TED, with a particular emphasis on emerging players of redox-related molecular signaling.

2.1. Immune Pathomechanisms

2.1.1. Orbital fibroblasts as pivotal actors in the autoimmune pathogenesis

The autoimmune pathogenesis of TED revolves around dysfunctional immunoreactivity linked with GD. The central player in this interplay is the TSH-R, found in both thyroid cells and OFs. Upon interaction with TSH or TSAb, TSH-R activates intracellular cascades, crucial for regulating thyroid cell functions [3,25,26].

The breakdown of self-tolerance for TSH-R triggers antigen-presenting cells (APCs) to recognize TSH-R epitopes as autoantigens. This process leads to T cell activation, stimulating B cells and culminating in the production of plasma cells and thyroid autoantibodies such as TSAb [4,27]. These autoantibodies hyperactivate the TSH-R, causing abnormal thyroid hormone secretion and intensifying thyroid cell activation via multiple signaling pathways [28].

Additionally, insulin-like growth factor-1 receptor (IGF1-R) activation by thyroid autoantibodies contributes to the inflammatory status. When triggered, IGF1-R activates molecular transductions responsible for increased fibrogenesis, glycosaminoglycan synthesis, and extracellular matrix deposition [29]. The role of specific anti-IGF1-R autoantibodies in this process is debated [3,30,31].

Crucially, the Forkhead box O (FOXO) transcription factors, modulated by various signaling pathways, play a significant role in regulating extracellular matrix deposition and adipogenesis [6,32]. Active B and T cells from the peripheral blood reach the orbit, initiating abnormal immunoreactivity within OFs, a pivotal step in TED development [26,33,34]. These processes establish the connection between GD and the underlying mechanisms of TED.

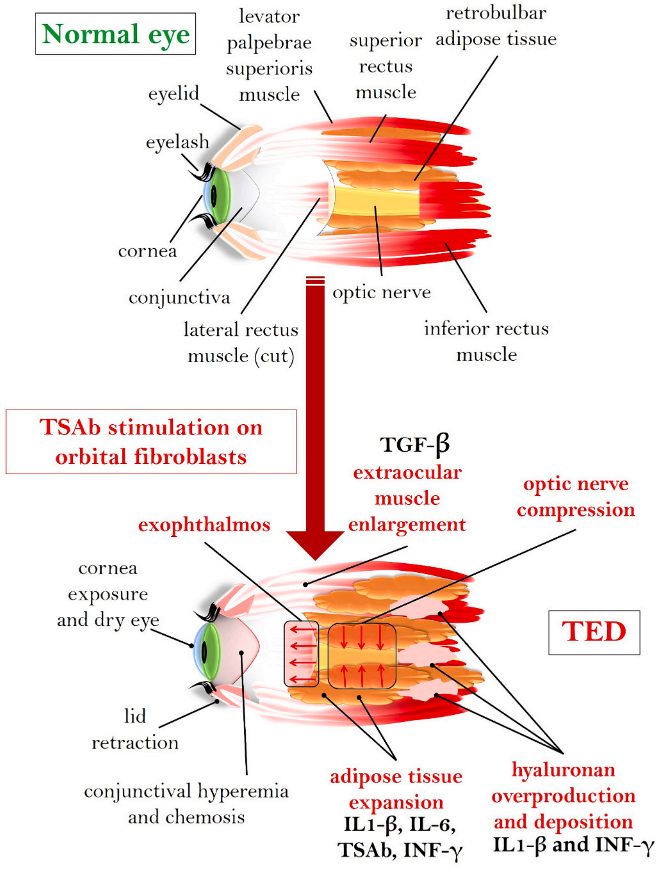

Fig. 1. Scheme on the autoimmune stimulation on orbital fibroblasts and manifestations in TED (color figure). TGF-β is mainly responsible of the myofibroblast transdifferentiation. IL-1β, TSAb, IL-6 and INF-γ elicit adipogenesis in retrobulbar tissues. Further, IL-1β and INF-γ are the principle triggers for the hyaluronan overproduction and the deposition of extracellular matrix. IL: interleukin; INF: interferon gamma; TED: thyroid eye disease; TGF: transforming growth factor; TSAb: TSH-R stimulating antibody.

Fig. 2 summarizes the initial pathophysiological processes in TED.

2.1.2. T helper cell functions

Naïve T cells differentiate into T helper (Th) or cytotoxic T cells (Tc), with CD4$^+$ cells supporting humoral immunity and CD8$^+$ cells regulating cell-mediated immune responses [35–37]. In TED, there is a notable increase in the CD4/CD8 ratio, potentially indicating a decline in CD8$^+$ T cells [33,38,39].

Within CD4$^+$ T cells, distinct subsets exist:

a) Th1: Produces proinflammatory cytokines like IL-2, TNF-α, IL-1β, and INF-γ, likely influencing early and active inflammation [40–43].
b) Th2: Releases cytokines supporting B cell activation and plasma cell differentiation, crucial for humoral response in later stages [33,42–44].
c) Th17: Identified as significant in fibrogenesis, secretes IL-21, IL-22, and IL-17 A, intensifying inflammation in the orbit [34,45–47].

Th1 cells predominantly produce INF-γ and IL-1β, stimulating expression of molecules like CD40 and MHC II on OFs. This interaction activates NF-kB and triggers an upsurge in inflammatory cytokines and matrix-related factors like HA production [26,34,44,48–51]. INF-γ

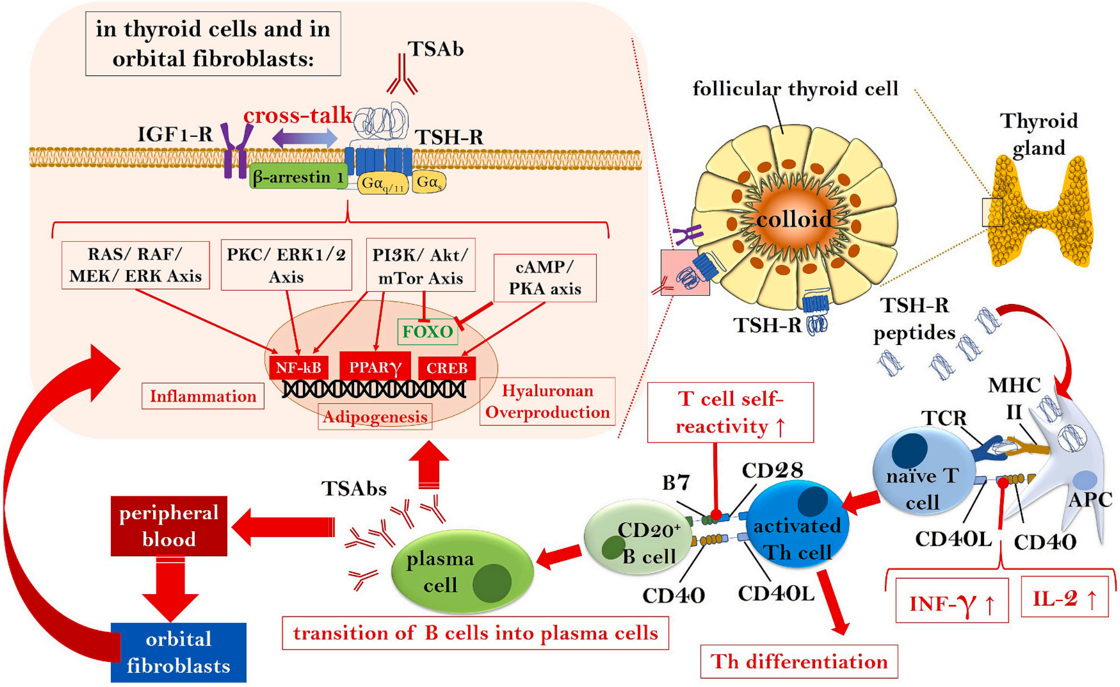

Fig. 2. Illustration depicting the initial pathophysiological events in the thyroid and orbit during TED (color figure). Akt: Ak strain transforming (also known as protein kinase B); APC: antigen presenting cell; cAMP: cyclic adenosine monophosphate; CD: cluster of differentiation; CREB: cAMP response element binding; ERK: extracellular-signal-regulated kinase; FOXO: Forkhead box O; IGF1-R: insulin-like growth factor 1 receptor; MHC: major histocompatibility complex; mTor: mammalian target of rapamycin; NF-kB: nuclear factor ‘kappa-light-chain-enhancer’ of activated B-cells; PI3K: Phosphoinositide 3-kinase; PK: protein kinase; PPARγ: peroxisome proliferator activated receptor γ; TCR: T cell receptor; Th: T helper cell; TSAb: thyroid stimulating antibody; TSH-R: thyroid stimulating hormone receptor.

blocks myofibroblast differentiation and suppresses α-SMA expression [52,53].

Th2-derived IL-4 primarily activates CD20$^+$ B cells and induces humoral responses, similarly inhibiting myofibroblast differentiation and influencing matrix modulation [33,53]. Both INF-γ and IL-4 intricately regulate extracellular matrix dynamics in TED, impacting tissue remodeling [34]. These processes significantly amplify the initial immune response, exacerbating pathological progression TED and fostering immune cell recruitment [34].

The Th17 subset, notably driven by IL17A, influences the differentiation of orbital fibroblasts (OFs) and CD34$^+$ fibrocytes, contributing to their recruitment and activation in TED.

[33,34,47,54–56]. This interaction between fibrocytes and Th17 cells sets up a loop, further fueling inflammation and OF differentiation [55,56]. Remarkably, OFs exist in subsets: CD90$^+$ and CD90$^-$ OFs, influenced differently by cytokines like IL-1β, IL-6, and TSAbs, driving adipogenesis in CD90$^-$ OFs and myofibroblast differentiation in CD90$^+$ OFs [3,43,44,47].

TGF-β prompts myofibroblast transformation in CD90$^+$ OFs via multiple pathways, including SMAD and p38 MAPK signaling [3,57].

Additionally, IL17A serves as a key factor in OF differentiation, stimulating fibrosis in CD90$^+$ OFs while impeding adipogenesis in CD90$^-$ OFs [47]. IL-23, released during inflammation, plays a pivotal role in Th17 cell differentiation, amplifying its signaling and contributing to the proliferation of Th17 cells [58,59]. This IL-23/IL-23R pathway, along with the PGE_2-EP_2/EP_4-cAMP-PKA pathway, influences Th17 differentiation, providing potential targets for intervention [47,59]. IL-38, identified as a suppressor of IL-23/IL-17 A axis, shows promise in downregulating inflammation in TED by disrupting this feedback loop [60].

2.1.2.1. Significance of CD4$^+$ T regulatory cells. CD4$^+$ regulatory T cells (Tregs) are under scrutiny in TED due to their role in regulating autoimmunity. While some studies indicate a potential deficit or dysfunction in Tregs linked to TED progression, others show increased Treg levels in TED patients, even after treatment, suggesting their role as a marker for therapy response [61–66].

Research on the nuclear receptor NR4A2 points to its involvement in the imbalance between Th17 and Treg populations in GD, providing insights into the pathogenesis of TED [67].

2.1.2.2. Emerging role of macrophages in early infiltrations. Alongside T and B cells, and activated OFs, also orbit-infiltrating CD68$^+$ macrophages, CD117$^+$ mast cells, and CD14$^+$ monocytes have been detected in TED [68–72]. In this context, a study conducted in a murine model of TED revealed high concentrations of macrophages infiltrating the orbital regions early on. This was followed by an overabundance of CD3$^+$ cells (a marker of total T cells), increased CD8$^+$ T cell proliferation, a decrease in Tregs, and a prevalence of inflammatory mediators like IFN-γ and TNF-α [73]. Lu and colleagues recently identified the immunohistochemical phenotype of macrophages in TED, highlighting prevailing CD86$^+$ M-1 like macrophages in active phases and CD163$^+$ M-2 like macrophages in stable TED, stimulated by IL-6 and TGF-β, respectively [74]. They emphasized the pivotal role of IL-6 signaling, activating orbit-infiltrating macrophages to stimulate OFs, amplifying fibrosis and inflammation [74].

Moreover, a recent study by Hai et al. in orbital tissue from active TED patients found elevated levels of various markers, correlating positively with TSAbs and CAS, and negatively with TED duration. The study suggested upregulations of TSH-R, IGF-1R, and CD40-CD40L binding as fundamental pathophysiological mechanisms for TED development. Another study identified IL-12, IL-35, and IL-27 as potential biomarkers in TED [75,76].

Fig. 3 illustrates a summarizing scheme on immune cascades and molecular signaling occurring via Th1, −2, −17, and a subsequent OF differentiation during TED.

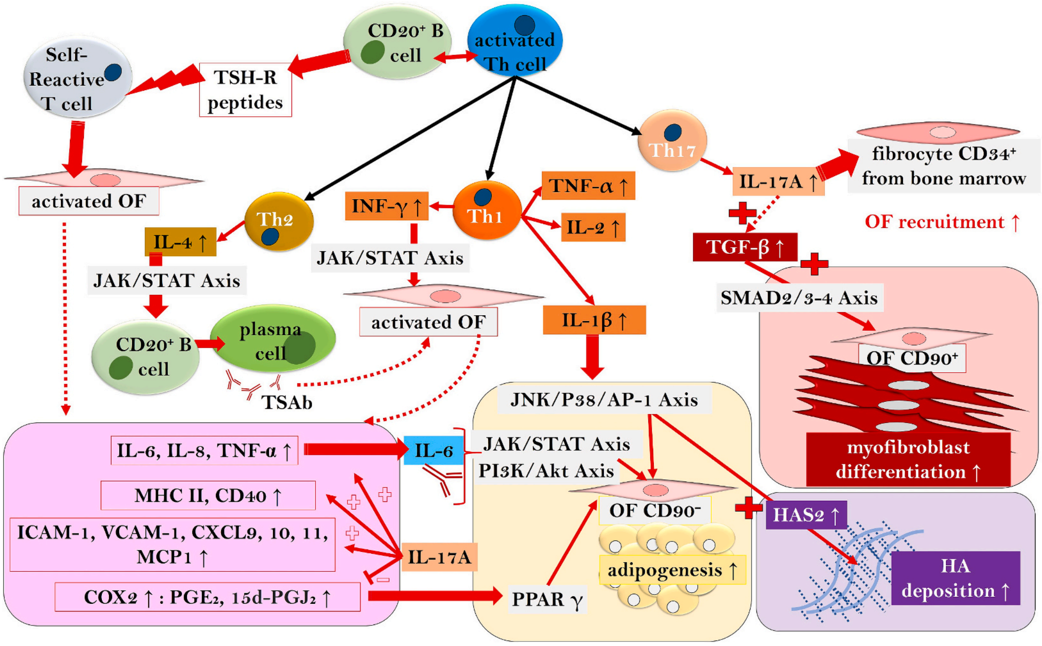

Fig. 3. Schematic representation of the main immune signaling and molecular pathways in the differentiation of T helper cells and orbital fibroblasts during TED (color figure). 15d-PGJ_2: 15-deoxy-Δ12,14-prostaglandin J2; Akt: Ak strain transforming (also known as protein kinase B); AP-1: activator protein 1; CD: cluster of differentiation; COX2: cyclooxygenase 2; CXCL: chemokine C-X-C motif ligand; ERK: extracellular-signal-regulated kinase; HA: Hyaluronan; HAS2: Hyaluronan synthase 2; ICAM: intercellular adhesion molecule 1; IGF1-R: insulin-like growth factor 1 receptor; IL: interleukin; JAK: janus kinase; JNK: c-Jun N-terminal kinase; MHC: major histocompatibility complex; OF: orbital fibroblast; PGE_2: prostaglandin E2; PI3K: Phosphoinositide 3-kinase; PK: protein kinase; PPARγ: Peroxisome proliferator-activated receptor γ; STAT: signal transducer and activator of transcription; TGF-β: transforming growth factor β; Th: T helper cell; TNF-α: tumor necrosis factor α; TSAb: thyroid stimulating antibody; TSH-R: thyroid stimulating hormone receptor; VCAM: vascular adhesion molecule 1.

2.2. Disruption of redox homeostasis in thyroid eye disease

2.2.1. Sources of oxidative stress in autoimmune thyroid disorders: Mitochondria

Mitochondria serve as pivotal intracellular organelles in energetic metabolism, with their activity strongly influenced by thyroid hormone levels, recognized as major modulators of metabolic efficiency [77,78]. Thyroid hormones exert regulatory actions on mitochondrial function, stimulating mitophagy, mitochondrial biogenesis, and enhancing the electron transport chain (ETC) to produce adenosine triphosphate (ATP), while collaterally inducing generation of reactive oxygen species (ROS) through single-electron reductions leaking from the ETC [79–84]. Consequently, due to proton leaks from the ETC and the subsequent excessive ROS formation, mitochondria are acknowledged as a principal source of oxidative stress in mammalian cells [85]. Given this background, hyperthyroidism, often associated with TED, is linked to an overabundance of ROS, increased intracellular ATP consumption, and notably, heightened ETC activity, collectively escalating oxidative stress [86,87]. Hence, it is not surprising to observe oxidative stress in individuals with hyperthyroidism, as supported by clinical investigations demonstrating elevated rates of lipid peroxidation, a marker of oxidative damage [88–90].

Conversely, a state of hypothyroidism, albeit in a smaller proportion of patients, can also be detected in TED, presenting similarly high levels of ROS [91–93]. The association between hypothyroidism and oxidative stress may be attributed to the decreased activity of the endogenous antioxidant system due to low thyroid hormone levels. Normally, thyroid hormones induce the expression of genes encoding antioxidative agents; however, in the absence of such stimulation, ROS accumulation can occur [87,94].

In summary, current literature indicates that mitochondrial oxidative stress plays a major pathogenetic role in TED, akin to other autoimmune disorders such as rheumatoid arthritis and systemic lupus erythematosus, where excess ROS is recognized as a central pathophysiological driver [95–98].

2.2.2. Tobacco smoking: A significant environmental factor exacerbating oxidative stress

In addition to oxidative stress caused by hyperactivated mitochondria or an insufficient antioxidant system, environmental factors in TED can exacerbate ROS overabundance. Among these factors, tobacco smoking emerges as a pivotal external contributor [99,100]. This assertion is supported by a wealth of evidence. Epidemiologically, individuals with GD and TED are notably more likely to be smokers [101]. Among those with TED, smokers tend to experience more severe manifestations of the disease compared to non-smokers, leading to poorer treatment outcomes [102–104]. Moreover, the severity of the disease shows a positive correlation with the amount of tobacco consumed [105]. While the direct causal link between smoking and TED remains not entirely proven [104], prevailing theories suggest that smoke triggers the activation of hypoxia-inducible factor 1 α (HIF-1α), an excessive generation of ROS, and the release of proinflammatory cytokines like interleukin 1 (IL-1). These factors affect OF and contribute to significant tissue remodeling, potentially initiating or exacerbating the disease [103,106].

2.2.3. Systemic oxidative biomarkers and oxidative stress in orbital fibroblasts

The persistent imbalance between the generation of ROS and their elimination by antioxidant agents, resulting in an excess of ROS, plays a pivotal role in the pathophysiology of GD and TED [107]. This surplus of ROS wreaks havoc on intracellular biomolecules, causing profound structural alterations and critical damage to proteins, DNA, and lipids. Diana et al. explored the impact of TSAbs on oxidative stress in GD patients, linking high levels of TSAbs to increased urinary malondialdehyde (MDA) levels (a marker of lipid peroxidation), elevated 8-hydroxy-2′-deoxyguanosine (8-OHdG) (a marker of oxidative DNA damage), and elevated serum levels of nicotinamide adenine dinucleotide phosphate (NADPH) oxidase subtype 2 (NOX2) —an enzyme crucial in ROS production. These findings strongly suggest that TSAbs trigger heightened ROS production and lipid peroxidation in GD [108]. Multiple studies have identified elevated markers of oxidative damage in TED patients' specimens, including blood, urine, and tears [109]. Blood samples from TED patients have shown increased concentrations of hydrogen peroxide (H_2O_2) and lipid hydroperoxides (ROOH), along with higher activities of antioxidative enzymes like superoxide dismutase (SOD) and catalase (CAT), while other antioxidants like glutathione peroxidase (GPx) and glutathione reductase exhibit reduced functionality [110,111]. A recent investigation by Acibucu et al. demonstrated altered serum thiol (SH)-disulphide (SS) levels in TED patients with proptosis, indicating decreased SH (an antioxidant) and increased SS (a marker of protein oxidative damage), revealing a connection between proptosis and SH-SS levels [112]. Additionally, markers of oxidative damage like MDA and 8-OHdG were detected in TED patients' tears, particularly linked to disease activity [113]. Urine analysis in TED patients revealed elevated 8-OHdG levels associated with active disease stages, where tobacco smoking notably impacted the increase in 8-OHdG levels [114,115]. Moreover, studies investigating the impact of glucocorticoids on the redox status of TED patients have shown a decrease in oxidative stress biomarkers following glucocorticoid treatment [116,117].

Beyond systemic markers, several in vitro studies have highlighted increased levels of 8-OHdG, MDA, and intracellular ROS in OFs from TED patients. One study comparing TED-OFs with control fibroblasts unveiled higher levels of 8-OHdG, MDA, as well as elevated concentrations of ROS such as $O_2^{\bullet -}$ and H_2O_2 in TED-OFs [118]. Another study indicated increased SOD activity and reduced GPx activity in TED-OFs, together with a reduced ratio between glutathione (GSH) and its oxidized form, glutathione disulphide (GSSG), suggesting elevated ROS concentrations within these cells [119]. Consistently, Hondur et al. reported heightened SOD activity and lower GSH levels in TED-OFs, further suggesting a negative correlation between low GSH and disease activity [120].

In summary, numerous studies provide evidence of altered systemic redox status in TED, indicating a potentially significant role of ROS in its etiopathogenesis.

2.2.4. *Redox signaling in the orbit*

2.2.4.1. Molecular pathways triggered by ROS in orbital fibroblasts. Efforts to delineate the impact of excessive ROS on OFs in TED have led to investigations into the molecular pathways influenced by ROS. For example, stimulation with H_2O_2 in TED-affected OFs has been observed to upregulate heat shock protein-72 (HSP-72), a significant contributor to the autoimmune reactivity seen in TED [121]. Additionally, studies have indicated that $O_2^{\bullet -}$ can enhance the proliferation of TED-OFs [122]. Furthermore, Lu et al. highlighted that ROS formation, particularly through IL-1β induction, triggers GAG production in TED-OFs [123]. Moreover, pathways related to H_2O_2 have been linked to the overexpression of essential molecules like HLA-DR and ICAM-1, pivotal in T-cell recognition and recruitment processes [124]. This ROS-driven positive feedback loop intensifies inflammation by upregulating potent cytokines such as IL-1β and IL-6 [43,49,125,126]. Notably, IL-6 plays a crucial role in early macrophage activation and adipogenesis, while TGF-β contributes to fibrogenesis and myofibroblast differentiation. Tsai et al. revealed that H_2O_2 induces an overexpression of both IL-1β and TGF-β in TED-OFs [126]. An emerging endogenous source of ROS, endoplasmic reticulum (ER) stress, has garnered attention in the scientific community [127]. Recent studies unveiled that the protein kinase RNA-like endoplasmic reticulum kinase (PERK)/activating transcription factor 4 (ATF4)/CCAAT-enhancer-binding protein-homologous protein (CHOP) axis, known to escalate ROS generation during ER stress, is significantly more active in TED-OFs than in control OFs. Intriguingly, silencing PERK resulted in the inhibition of both oxidative stress and adipogenesis in TED-OFs [128].

2.2.4.2. Role of tobacco smoke and hypoxia in orbital fibroblasts and adipocytes. As previously highlighted, tobacco smoking emerges as a predominant environmental risk factor in TED, notably exacerbating oxidative stress levels in TED patients who smoke compared to non-smoking counterparts [114,129,130]. Studies conducted in vitro on TED-affected OFs exposed to cigarette smoke extracts (CSE) revealed an abnormal surge in oxidative stress alongside elevated levels of TGF-β, IL-1, fibronectin, and connective tissue growth factor (CTGF) [131]. Additionally, exposure of OFs to CSE demonstrated an augmentation in GAG production, synergistically promoting adipogenesis in conjunction with IL-1 [132]. Yoon et al. further delineated the adipogenic effect of CSE mediated by heightened intracellular ROS generation in both TED-OFs and control fibroblasts [133]. Crucially, in TED, smoking has been reported to induce tissue hypoxia, upregulating HIF-1α expression, possibly through NF-kB activation [106]. Elevated HIF-1α levels have shown correlation with disease activity [106]. Investigating the role of hypoxia in TED progression, Görtz et al. suggested that hypoxic signaling in TED potentially accelerates the disease by inducing TNF-α, leading to the production of chemokines like CCL2, CCL5, and CCL20. This process stimulates the release of HIF-1α and, notably, an interaction between $CD68^+$ macrophages and OFs further promotes inflammation and adipogenesis [134]. Studies focusing on orbital adipocytes and extraocular muscle cells in TED have revealed augmented oxidative stress markers and increased expression of antioxidant agents, such as peroxiredoxin 5, indicating a cellular response to counteract oxidative stress [135]. Further investigations demonstrated increased HIF-1α expression and subsequent downregulation of caveolin 1 (Cav-1) in TED adipocytes. This downregulation affects glucose metabolism via the glucose transporter type 4 (Glut-4) and modulates the activity of NOX2 and endothelial nitric oxide synthase (eNOS). Moreover, an upregulation of deiodinase 3 (DIO3), a regulator of T3 metabolism, was noted, emphasizing the pivotal roles of HIF-1α, Cav-1, and DIO3 in oxidative stress processes within TED adipocytes [125]. Recent studies highlighted the involvement of microRNAs as epigenetic regulators in GD and TED orbital adipocytes. They emphasized the NOX4/HIF-1α/VEGF-A axis as a crucial player in oxidative stress and neoangiogenesis in TED [136]. In an ex vivo model, Hikage et al. investigating fibrosis mechanisms in TED suggested the upregulation of the HIF-2α-lysyl oxidase pathway in TED-OFs, correlating with excessive extracellular matrix deposition and proposing this axis as a potential therapeutic target in TED [137].

Collectively, current literature indicates a strong association between smoking, hypoxic conditions, ROS formation, aberrant neoangiogenesis, and adipogenesis in TED, with significant implications for disease progression. HIF-1α, TNF-α, VEGF-A, and NOX emerge as pivotal factors in this context.

Fig. 4 provides an illustration of the main redox molecular signaling occurring in OFs and orbital adipocytes during TED.

3. Emerging approaches targeting redox mechanisms

3.1. *Naturally occurring antioxidants*

Exploratory studies have unveiled the potential benefits of certain natural antioxidants in managing mild TED [127,138]. Selenium emerges as a prime candidate in this category, backed by randomized clinical trials advocating its use as the sole antioxidant for TED treatment [139].

This naturally occurring mineral, found in foods like eggs, cereals, and meat, exhibits anti-inflammatory and antioxidative properties [140,141]. In vitro studies on TED-affected OFs have shown that selenium suppresses ROS generation, ROS-related GAG synthesis, reduces

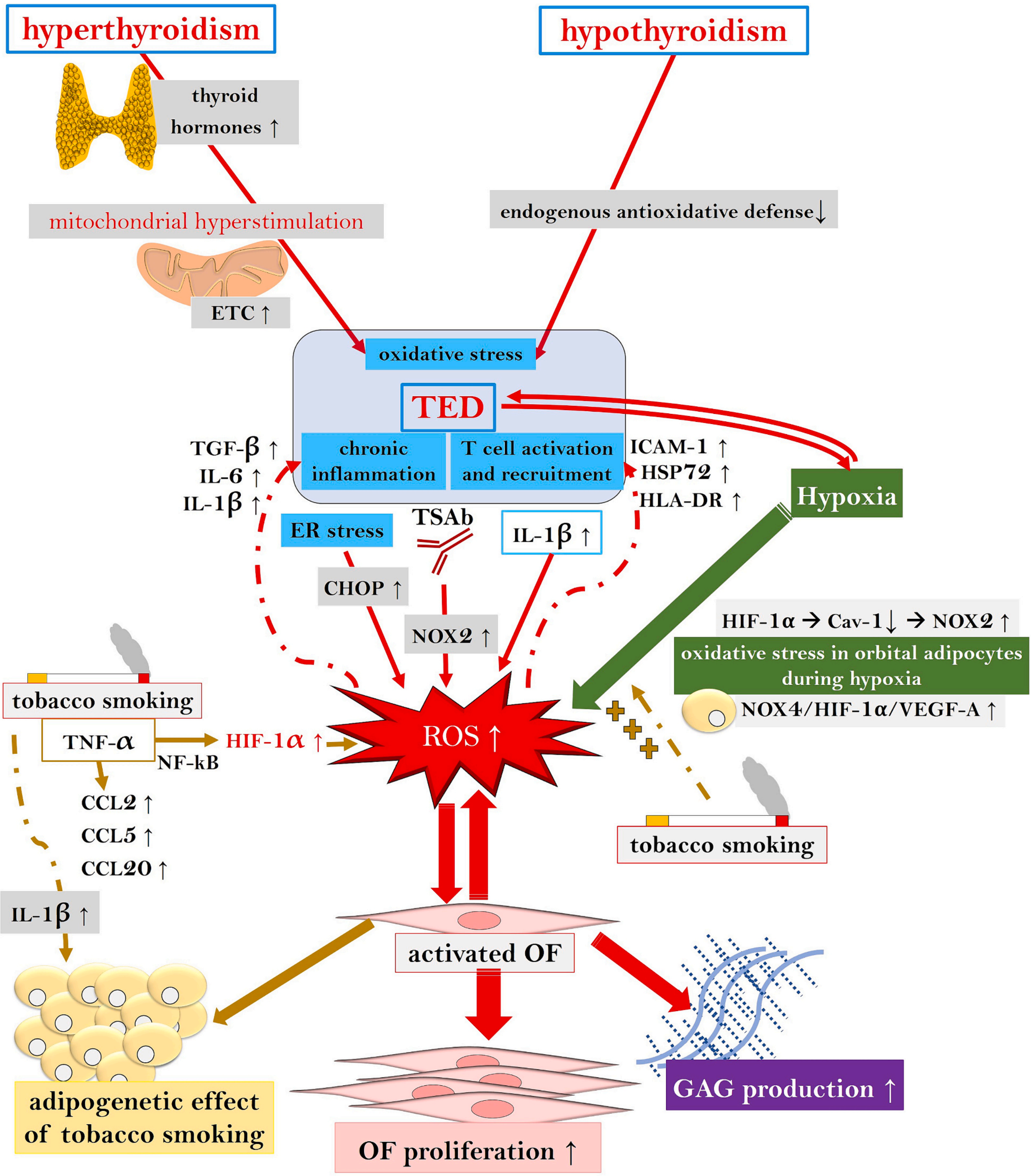

Fig. 4. Scheme on the main redox signaling pathways occurring in TED (color figure). Cav-1: caveolin 1; CCL: chemokine (C-C motif) ligand; CHOP: CCAAT-enhancer-binding protein homologous protein; ER: endoplasmic reticulum; ETC: electron transport chain; GAG: glycosaminoglycan; HIF-1α: hypoxia inducible factor 1α; HLA-DR: human leucocyte antigen DR isotype; HSP: heat shock protein; ICAM-1: intercellular adhesion molecule 1; IL: interleukin; NF-kB: nuclear factor ‘kappa-light-chain-enhancer’ of activated B-cells; NOX: nicotinamide adenine dinucleotide phosphate oxidase; OF: orbital fibroblasts; ROS: reactive oxygen species; TED: thyroid eye disease; TGF-β: transforming factor β; TNF-α: tumor necrosis factor α; TSAb: thyroid stimulating antibody; VEGF: vascular endothelial growth factor.

proinflammatory cytokines such as TNF-α, and inhibits fibroblastic proliferation [142,143]. The EUGOGO clinical trial in 2011 demonstrated the efficacy of selenium supplementation in mild TED, resulting in reduced ocular disease severity, improved quality of life, and slowed disease progression compared to pentoxifylline [144]. Tailoring selenium treatment according to regional dietary selenium intake is crucial [141], emphasizing the need for pre-treatment serum selenium detection to avoid potential side effects [145,146].

Preclinical investigations have explored the effects of ascorbic acid in combination with *N*-acetyl-L-cysteine (NAC) and melatonin on TED-OFs. NAC and ascorbic acid demonstrated a reduction of cell proliferation, IL-1β, IFN-γ signaling, and GAG synthesis, while melatonin showed similar results without inhibiting cell proliferation [147]. Consistent with these findings, a prior preclinical study described that NAC can reverse the increased proliferation of TED-OFs, as well as the production of TGF-β1, IL-1β, and $O_2^{\bullet -}$ [126]. It is worth noting that while the antioxidant effect of NAC has been previously demonstrated due to GSH biosynthesis, facilitating detoxification and removal of free radicals [148], the reported in vitro studies have not addressed the upstream mechanistic targets involved in the anti-proliferative and anti-inflammatory effects on TED-OFs, necessitating further research in this area. However, considering the antagonistic action of NAC against inflammation and the increased proliferation, a plausible explanation for its anti-proliferative and anti-inflammatory effects in TED may be linked to its inhibitory activity on the mammalian target of rapamycin (mTOR) signaling pathway. Activation of mTOR is responsible for inflammation and increased fibroblast proliferation in TED, and this pathway has been shown to be blocked by NAC in other autoimmune disorders such as systemic lupus erythematosus and anti-phospholipid syndrome [149–152]. Furthermore, β-carotene has displayed effective antioxidant effects on TED-OFs, reducing cell proliferation [153]. Quercetin, found in vegetables and fruits [154], exhibits anti-inflammatory and antioxidant properties, reducing the IL-1β pathway, GAG synthesis, adipogenesis, and ROS generation in TED-OFs exposed to cigarette smoke extracts [133,155]. It also counteracts the actions of IL-1 and TNF-α, as well as matrix metalloproteinases MMP-2 and MMP-9, displaying anti-fibrotic effects [156–158].

Curcumin, derived from *Curcuma longa*, offers anti-inflammatory, antioxidative, anti-adipogenic, anti-fibrotic, and anti-angiogenic features [159,160]. Studies on TED-OFs showed curcumin's efficacy in blocking IL-1β signaling, reducing synthesis of proinflammatory mediators like IL-6, IL-8, MCP-1, and ICAM-1, and decreasing adipogenesis [160,161].

Resveratrol, a polyphenol found in foods like berries or peanuts, is known for its antioxidant abilities [162]. This compound activates sirtuin 1 (SIRT1), a nuclear NAD^+-dependent deacetylase capable of overexpressing the Nrf2/ARE axis [163,164]. Resveratrol reduced ROS formation and blocked adipogenesis in TED-OFs, possibly by regulating the activities of ERK, JNK, and NF-kB [165].

Gypenosides from *Gynostemma pentaphyllum* extracts reduce TGF-β1-related fibrogenesis and IL-1β-related expression of pro-inflammatory mediators in TED-OFs, improving inflammation and fibrotic events. Additionally, they exhibit antioxidative effects through the Nrf2/ERK/HO-1 axis [159,166,167].

Several other naturally occurring molecules found in herb extracts have shown promise in mitigating TED by exerting antioxidant and anti-inflammatory activities in vitro, including berberine, astragaloside IV, icariin, polydatin, and tanshinone IIA [159,168–174].

3.2. *Synthetic medications as antioxidants*

Various synthetic medications have been investigated for their potential antioxidant roles in treating TED. Enalapril, a commonly prescribed antihypertensive medication possessing inherent antioxidant activity [175,176], has shown promise in dedicated in vitro studies on TED-OFs. Research suggests it can reduce cell proliferation, glycosaminoglycan (GAG) production, adipogenesis, and the TGF-β1 pathway [177]. A clinical trial involving 12 patients with mild TED treated with enalapril for six months demonstrated favorable outcomes regarding disease course and progression [178].

Statins, primarily used to manage hypercholesterolemia and prevent cardiovascular disorders by inhibiting 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase, possess critical antioxidant properties that modulate NOX activity [179]. These medications have shown efficacy in reducing the risk of developing TED in patients with in GD [180]. The ongoing "Statins for Graves' Orbitopathy" (STAGO) trial phase II (NCT03110848) has highlighted the benefit of atorvastatin as supplementary therapy to steroids in improving outcomes for moderate-to-severe TED cases accompanied by hypercholesterolemia [181].

Other existing drugs tested for their antioxidant potential in TED treatment include nicotinamide in combination with allopurinol, typically used to manage gout, and pentoxifylline, employed for peripheral vascular disorders [182,183]. These compounds have exhibited beneficial effects in preclinical and clinical investigations [139], reducing oxidative stress in TED-OFs and slowing disease progression [101,184–187].

Overall, while considering preclinical results and limited clinical findings, antioxidant compounds show promise as additional TED treatments when used alongside steroids, potentially slowing disease progression and severity [127,146]. It is noteworthy that while numerous promising preclinical findings exist, only a few antioxidant agents have demonstrated efficacy in both preclinical and clinical settings [139]. Currently, selenium remains the only antioxidant recommended in specific circumstances. Thus, further preclinical and clinical research on these antioxidants in TED is crucial to establish their effectiveness.

4. Conclusion & perspective

New treatment approaches have aimed to develop potent drugs, either as alternatives to or in conjunction with current therapies like steroids. Future medications, such as highly effective monoclonal antibodies or inhibitory small peptides, targeting autoantigens like TSH-R and/or IGF-1R, show promise in halting the autoimmune response. Pioneering translational research suggests the potential role of antioxidants as supplementary drugs, with selenium currently the sole recommended antioxidant for TED. Despite promising preclinical results, only a few antioxidant agents have progressed to clinical trials.

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CRediT authorship contribution statement

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Declaration of competing interest

None.

Data availability

No data was used for the research described in the article.

References

[1] Smith TJ, Hegedüs L. Graves' disease. N Engl J Med 2016;375:1552–65.

[2] Bartalena L. Prevention of Graves' ophthalmopathy. Best Pract Res Clin Endocrinol Metab 2012;26:371–9.

[3] Gupta V, Hammond CL, Roztocil E, Gonzalez MO, Feldon SE, Woeller CF. Thinking inside the box: current insights into targeting orbital tissue remodeling and inflammation in thyroid eye disease. Surv Ophthalmol 2022;67:858–74.

[4] George A, Diana T, Längericht J, Kahaly GJ. Stimulatory thyrotropin receptor antibodies are a biomarker for graves' orbitopathy. Front Endocrinol 2021;11.

[5] Diana T, Kahaly GJ. Thyroid stimulating hormone receptor antibodies in thyroid eye disease-methodology and clinical applications. Ophthal Plast Reconstr Surg 2018;34:S13–s19.

[6] Lee ACH, Kahaly GJ. Pathophysiology of thyroid-associated orbitopathy. Best Pract Res Clin Endocrinol Metab 2023;37:101620.

[7] Morshed SA, Ando T, Latif R, Davies TF. Neutral antibodies to the TSH receptor are present in Graves' disease and regulate selective signaling cascades. Endocrinology 2010;151:5537–49.

[8] Nicolì F, Lanzolla G, Mantuano M, Ionni I, Mazzi B, Leo M, et al. Correlation between serum anti-TSH receptor autoantibodies (TRAbs) and the clinical feature of Graves' orbitopathy. J Endocrinol Invest 2021;44:581–5.

[9] Kahaly GJ, Wüster C, Olivo PD, Diana T. High titers of thyrotropin receptor antibodies are associated with orbitopathy in patients with graves disease. J Clin Endocrinol Metabol 2019;104:2561–8.

[10] Roos JCP, Paulpandian V, Murthy R. Serial TSH-receptor antibody levels to guide the management of thyroid eye disease: the impact of smoking, immunosuppression, radio-iodine, and thyroidectomy. Eye 2019;33:212–7.

[11] Kahaly GJ, Diana T, Glang J, Kanitz M, Pitz S, König J. Thyroid stimulating antibodies are highly prevalent in Hashimoto's thyroiditis and associated Orbitopathy. J Clin Endocrinol Metab 2016;101:1998–2004.

[12] Bartalena L, Piantanida E, Gallo D, Lai A, Tanda ML. Epidemiology, natural history, risk factors, and prevention of Graves' Orbitopathy. Front Endocrinol 2020;11.

[13] Olejarz M, Szczepanek-Parulska E, Dadej D, Sawicka-Gutaj N, Domin R, Ruchała M. IgG4 as a biomarker in graves' orbitopathy. Mediators Inflamm 2021; 2021:5590471.

[14] Jeon H, Lee JY, Kim YJ, Lee MJ. Clinical relevance of thyroid-stimulating immunoglobulin as a biomarker of the activity of thyroid eye disease. Eye 2023; 37:543–7.

[15] Diana T, Ponto KA, Kahaly GJ. Thyrotropin receptor antibodies and Graves' orbitopathy. J Endocrinol Invest 2021;44:703–12.

[16] Rui ZY, Yin X, Wang X, Shen J, Zheng W. A retrospective study of clinical features and prognosis of patients with Graves' disease and ophthalmopathy. Nucl Med Commun 2023;44:137–41.

[17] Sarić Matutinović M, Kahaly GJ, Žarković M, Ćirić J, Ignjatović S, Nedeljković Beleslin B. The phenotype of Graves' orbitopathy is associated with thyrotropin receptor antibody levels. J Endocrinol Invest 2023;46:2309–17.

[18] Lytton SD, Ponto KA, Kanitz M, Matheis N, Kohn LD, Kahaly GJ. A novel thyroid stimulating immunoglobulin bioassay is a functional indicator of activity and severity of Graves' orbitopathy. J Clin Endocrinol Metab 2010;95:2123–31.

[19] Ponto KA, Diana T, Binder H, Matheis N, Pitz S, Pfeiffer N, et al. Thyroid-stimulating immunoglobulins indicate the onset of dysthyroid optic neuropathy. J Endocrinol Invest 2015;38:769–77.

[20] Sarić Matutinović M, Diana T, Nedeljković Beleslin B, Ćirić J, Žarković M, Kahaly GJ, et al. Clinical value of functional thyrotropin receptor antibodies in Serbian patients with Graves' orbitopathy. J Endocrinol Invest 2022;45:189–97.

[21] Kampmann E, Diana T, Kanitz M, Hoppe D, Kahaly GJ. Thyroid stimulating but not blocking autoantibodies are highly prevalent in severe and active thyroid-associated orbitopathy: a prospective study. Int J Endocrinol 2015;2015:678194.

[22] Diana T, Krause J, Olivo PD, König J, Kanitz M, Decallonne B, et al. Prevalence and clinical relevance of thyroid stimulating hormone receptor-blocking antibodies in autoimmune thyroid disease. Clin Exp Immunol 2017;189:304–9.

[23] Ponto KA, Binder H, Diana T, Matheis N, Otto AF, Pitz S, et al. Prevalence, phenotype, and psychosocial well-being in Euthyroid/hypothyroid thyroid-associated orbitopathy. Thyroid® 2015;25:942–8.

[24] Eckstein AK, Lösch C, Glowacka D, Schott M, Mann K, Esser J, et al. Euthyroid and primarily hypothyroid patients develop milder and significantly more asymmetrical graves ophthalmopathy. Br J Ophthalmol 2009;93:1052–6.

[25] Morshed SA, Latif R, Davies TF. Characterization of thyrotropin receptor antibody-induced signaling cascades. Endocrinology 2009;150:519–29.

[26] Davies TF, Andersen S, Latif R, Nagayama Y, Barbesino G, Brito M, et al. Graves' disease. Nat Rev Dis Primers 2020;6:52.

[27] Diana T, Kahaly GJ. Thyroid stimulating hormone receptor antibodies in thyroid eye disease—methodology and clinical applications. Ophthal Plast Reconstr Surg 2018;34:S13–9.

[28] Rapoport B, Chazenbalk GD, Jaume JC, McLachlan SM. The thyrotropin (TSH)-releasing hormone receptor: interaction with TSH and autoantibodies. Endocr Rev 1998;19:673–716.

[29] Kahaly GJ. The Thyrocyte-Fibrocyte link: closing the loop in the pathogenesis of Graves' disease? J Clin Endocrinol Metabol 2010;95:62–5.

[30] Cui X, Wang F, Liu C. A review of TSHR- and IGF-1R-related pathogenesis and treatment of Graves' orbitopathy. Front Immunol 2023;14.

[31] Krieger CC, Neumann S, Gershengorn MC. Is there evidence for IGF1R-stimulating abs in graves' orbitopathy pathogenesis? Int J Mol Sci 2020;21.

[32] Zhang L, Ji Q, Ruge F, Lane C, Morris D, Tee A, et al. Reversal of pathological features of Graves' orbitopathy by activation of forkhead transcription factors, FOXOs. J Clin Endocrinol 2016;101:114–22.

[33] Huang Y, Fang S, Li D, Zhou H, Li B, Fan X. The involvement of T cell pathogenesis in thyroid-associated ophthalmopathy. Eye 2019;33:176–82.

[34] Fang S, Lu Y, Huang Y, Zhou H, Fan X. Mechanisms that Underly T cell immunity in Graves' Orbitopathy. Front Endocrinol 2021;12.

[35] Grubeck-Loebenstein B, Trieb K, Sztankay A, Holter W, Anderl H, Wick G. Retrobulbar T cells from patients with Graves' ophthalmopathy are CD8+ and specifically recognize autologous fibroblasts. J Clin Invest 1994;93:2738–43.

[36] Pappa A, Calder V, Ajjan R, Fells P, Ludgate M, Weetman AP, et al. Analysis of extraocular muscle-infiltrating T cells in thyroid-associated ophthalmopathy (TAO). Clin Exp Immunol 1997;109:362–9.

[37] Förster G, Otto E, Hansen C, Ochs K, Kahaly G. Analysis of orbital T cells in thyroid-associated ophthalmopathy. Clin Exp Immunol 1998;112:427–34.

[38] Kahaly GJ, Shimony O, Gellman YN, Lytton SD, Eshkar-Sebban L, Rosenblum N, et al. Regulatory T-cells in Graves' orbitopathy: baseline findings and immunomodulation by anti-T lymphocyte globulin. J Clin Endocrinol Metab 2011;96:422–9.

[39] Xia N, Zhou S, Liang Y, Xiao C, Shen H, Pan H, et al. CD4+ T cells and the Th1/Th2 imbalance are implicated in the pathogenesis of Graves' ophthalmopathy. Int J Mol Med 2006;17:911–6.

[40] Aniszewski JP, Valyasevi RW, Bahn RS. Relationship between disease duration and predominant orbital T cell subset in Graves' ophthalmopathy. J Clin Endocrinol Metab 2000;85:776–80.

[41] Bahn RS. Graves' ophthalmopathy. N Engl J Med 2010;362:726–38.

[42] Huang Y, Fang S, Zhang S, Zhou H. Progress in the pathogenesis of thyroid-associated ophthalmopathy and new drug development. Taiwan J Ophthalmol 2020;10:174–80.

[43] Zheng J, Duan H, You S, Liang B, Chen Y, Huang H. Research progress on the pathogenesis of Graves' ophthalmopathy: based on immunity, noncoding RNA and exosomes. Front Immunol 2022;13.

[44] Łacheta D, Miśkiewicz P, Głuszko A, Nowicka G, Struga M, Kantor I, et al. Immunological aspects of graves' ophthalmopathy. Biomed Res Int 2019;2019: 7453260.

[45] Fang S, Huang Y, Wang S, Zhang Y, Luo X, Liu L, et al. IL-17A exacerbates fibrosis by promoting the proinflammatory and profibrotic function of orbital fibroblasts in TAO. J Clin Endocrinol Metabol 2016;101:2955–65.

[46] Fang S, Huang Y, Zhong S, Zhang Y, Liu X, Wang Y, et al. IL-17A promotes RANTES expression, but not IL-16, in orbital fibroblasts via CD40-CD40L combination in thyroid-associated ophthalmopathy. Invest Ophthalmol Vis Sci 2016;57:6123–33.

[47] Fang S, Huang Y, Zhong S, Li Y, Zhang Y, Li Y, et al. Regulation of orbital fibrosis and adipogenesis by pathogenic Th17 cells in graves orbitopathy. J Clin Endocrinol Metabol 2017;102:4273–83.

[48] Antonelli A, Ferrari SM, Frascerra S, Ruffilli I, Pupilli C, Bernini G, et al. β (CCL2) and α (CXCL10) chemokine modulations by cytokines and peroxisome proliferator-activated receptor-α agonists in Graves' ophthalmopathy. J Endocrinol 2012;213:183–91.

[49] Chen B, Tsui S, Smith TJ. IL-1β induces IL-6 expression in human orbital fibroblasts: identification of an anatomic-site specific phenotypic attribute relevant to thyroid-associated ophthalmopathy. J Immunol 2005;175:1310–9.

[50] Chung SA, Jeon BK, Choi YH, Back KO, Lee JB, Kook KH. Pirfenidone attenuates the IL-1β-induced hyaluronic acid increase in orbital fibroblasts from patients with thyroid-associated ophthalmopathy. Invest Ophthalmol Vis Sci 2014;55: 2276–83.

[51] Han R, Smith TJ. T helper type 1 and type 2 cytokines exert divergent influence on the induction of prostaglandin E2 and hyaluronan synthesis by interleukin-1beta in orbital fibroblasts: implications for the pathogenesis of thyroid-associated ophthalmopathy. Endocrinology 2006;147:13–9.

[52] Koumas L, Smith TJ, Feldon S, Blumberg N, Phipps RP. Thy-1 expression in human fibroblast subsets defines myofibroblastic or lipofibroblastic phenotypes. Am J Pathol 2003;163:1291–300.

[53] Han R, Smith TJ. Induction by IL-1 beta of tissue inhibitor of metalloproteinase-1 in human orbital fibroblasts: modulation of gene promoter activity by IL-4 and IFN-gamma. J Immunol 2005;174:3072–9.

[54] Jiang M, Zhao J, Wang P, Yan S, Wang Y. Research progress in Th17 cells and the relevant cytokines in Graves' ophthalmopathy. Zhong Nan Da Xue Xue Bao Yi Xue Ban 2022;47:1748–53.

[55] Fang S, Huang Y, Liu X, Zhong S, Wang N, Zhao B, et al. Interaction between CCR6+ Th17 cells and CD34+ fibrocytes promotes inflammation: implications in graves' orbitopathy in Chinese population. Invest Ophthalmol Vis Sci 2018;59: 2604–14.

[56] Fernando R, Atkins SJ, Smith TJ. Slit2 may underlie divergent induction by thyrotropin of IL-23 and IL-12 in human Fibrocytes. J Immunol 2020;204: 1724–35.

[57] Usuki J, Matsuda K, Azuma A, Kudoh S, Gemma A. Sequential analysis of myofibroblast differentiation and transforming growth factor-β1/Smad pathway activation in murine pulmonary fibrosis. J Nippon Med Sch 2012;79:46–59.

[58] Rajaii F, McCoy AN, Smith TJ. Cytokines are both villains and potential therapeutic targets in thyroid-associated ophthalmopathy: from bench to bedside. Expert Rev Ophthalmol 2014;9:227–34.

[59] Lee J, Aoki T, Thumkeo D, Siriwach R, Yao C, Narumiya S. T cell-intrinsic prostaglandin E(2)-EP2/EP4 signaling is critical in pathogenic T(H)17 cell-driven inflammation. J Allergy Clin Immunol 2019;143:631–43.

[60] Pan Y, Wang M, Chen X, Chen Y, Ai S, Wang M, et al. Elevated IL-38 inhibits IL-23R expression and IL-17A production in thyroid-associated ophthalmopathy. Int Immunopharmacol 2021;91:107300.

[61] Brusko TM, Putnam AL, Bluestone JA. Human regulatory T cells: role in autoimmune disease and therapeutic opportunities. Immunol Rev 2008;223: 371–90.

[62] Pawlowski P, Grubczak K, Kostecki J, Ilendo-Poskrobko E, Moniuszko M, Pawlowska M, et al. Decreased frequencies of peripheral blood CD4+CD25+ CD127-Foxp3+ in patients with graves' disease and graves' orbitopathy: enhancing effect of insulin growth Factor-1 on Treg cells. Horm Metab Res 2017; 49:185–91.

[63] Li C, Yuan J, Zhu YF, Yang XJ, Wang Q, Xu J, et al. Imbalance of Th17/Treg in different subtypes of autoimmune thyroid diseases. Cell Physiol Biochem 2016; 40:245–52.

[64] Shao S, Yu X, Shen L. Autoimmune thyroid diseases and Th17/Treg lymphocytes. Life Sci 2018;192:160–5.

[65] Rodríguez-Muñoz A, Vitales-Noyola M, Ramos-Levi A, Serrano-Somavilla A, González-Amaro R, Marazuela M. Levels of regulatory T cells CD69(+)NKG2D(+) IL-10(+) are increased in patients with autoimmune thyroid disorders. Endocrine 2016;51:478–89.

[66] Xu Q-Q, Zhang X-L, Luo B, Li T, Yuan G, Shao S-Y. Role of peripheral regulatory T lymphocytes in patients with thyroid associated ophthalmopathy during systemic

glucocorticoid treatment: a prospective observational study. Curr Med Sci 2023;43:130–8.

[67] Zhao S, Wang X, Huang F, Zhou Y, Meng D, Zhao D, et al. A role of NR4A2 in Graves' disease: regulation of Th17/Treg. Endocrine 2024;83:432–441.

[68] van Steensel L, Paridaens D, van Meurs M, van Hagen PM, van den Bosch WA, Kuijpers RW, et al. Orbit-infiltrating mast cells, monocytes, and macrophages produce PDGF isoforms that orchestrate orbital fibroblast activation in Graves' ophthalmopathy. J Clin Endocrinol Metab 2012;97:E400–8.

[69] Eckstein AK, Quadbeck B, Tews S, Mann K, Krüger C, Mohr CH, et al. Thyroid associated ophthalmopathy: evidence for CD4(+) gammadelta T cells; de novo differentiation of RFD7(+) macrophages, but not of RFD1(+) dendritic cells; and loss of gammadelta and alphabeta T cell receptor expression. Br J Ophthalmol 2004;88:803–8.

[70] Pawlowski P, Poplawska I, Mysliwiec J, Dik WA, Eckstein A, Berchner-Pfannschmidt U, et al. Search of reference biomarkers reflecting orbital tissue remodeling in the course of Graves' orbitopathy. Folia Histochem Cytobiol 2020;58:37–45.

[71] Chen MH, Chen MH, Liao SL, Chang TC, Chuang LM. Role of macrophage infiltration in the orbital fat of patients with Graves' ophthalmopathy. Clin Endocrinol (Oxf) 2008;69:332–7.

[72] Virakul S, Phetsuksiri T, van Holten-Neelen C, Schrijver B, van Steensel L, Dalm V, et al. Histamine induces NF-κB controlled cytokine secretion by orbital fibroblasts via histamine receptor type-1. Exp Eye Res 2016;147:85–93.

[73] Philipp S, Horstmann M, Hose M, Daser A, Görtz GE, Jesenek C, et al. An early wave of macrophage infiltration intertwined with antigen-specific proinflammatory T cells and browning of adipose tissue characterizes the onset of orbital inflammation in a mouse model of graves' orbitopathy. Thyroid 2022;32:283–93.

[74] Lu Y, Wang Y, Wang Y, Wu Y, Huang Y, Liu X, et al. M1-like macrophages modulate fibrosis and inflammation of orbital fibroblasts in graves' orbitopathy: potential relevance to soluble Interleukin-6 receptor. Thyroid 2023;33:338–50.

[75] Hai YP, Saeed MEM, Ponto KA, Elflein HM, Lee ACH, Fang S, et al. Single-blind, case-control, immunohistochemical study of orbital tissue in thyroid eye disease. Thyroid 2022;32:1547–58.

[76] Zhang P, Zhang X, Xu F, Xu W, Zhu H. Elevated expression of interleukin-27, IL-35, and decreased IL-12 in patients with thyroid-associated ophthalmopathy. Graefes Arch Clin Exp Ophthalmol 2023;261:1091–100.

[77] Harper ME, Seifert EL. Thyroid hormone effects on mitochondrial energetics. Thyroid 2008;18:145–56.

[78] Yehuda-Shnaidman E, Kalderon B, Bar-Tana J. Thyroid hormone, Thyromimetics, and metabolic efficiency. Endocr Rev 2014;35:35–58.

[79] Venditti P, Puca A, Di Meo S. Effects of thyroid state on H2O2 production by rat heart mitochondria: sites of production with complex I-and complex II-linked substrates. Horm Metab Res 2003;35:55–61.

[80] Weitzel JM, Iwen KA, Seitz HJ. Regulation of mitochondrial biogenesis by thyroid hormone. Exp Physiol 2003;88:121–8.

[81] Liemburg-Apers DC, Willems PH, Koopman WJ, Grefte S. Interactions between mitochondrial reactive oxygen species and cellular glucose metabolism. Arch Toxicol 2015;89:1209–26.

[82] Sinha RA, Singh BK, Zhou J, Wu Y, Farah BL, Ohba K, et al. Thyroid hormone induction of mitochondrial activity is coupled to mitophagy via ROS-AMPK-ULK1 signaling. Autophagy 2015;11:1341–57.

[83] Lesmana R, Sinha RA, Singh BK, Zhou J, Ohba K, Wu Y, et al. Thyroid hormone stimulation of autophagy is essential for mitochondrial biogenesis and activity in skeletal muscle. Endocrinology 2016;157:23–38.

[84] Palma FR, Gantner BN, Sakiyama MJ, Kayzuka C, Shukla S, Lacchini R, et al. ROS production by mitochondria: function or dysfunction? Oncogene 2024;43:295–303.

[85] Murphy MP. How mitochondria produce reactive oxygen species. Biochem J 2009;417:1–13.

[86] Gheorghiu ML, Badiu C. Selenium involvement in mitochondrial function in thyroid disorders. Hormones 2020;19:25–30.

[87] Mancini A, Di Segni C, Raimondo S, Olivieri G, Silvestrini A, Meucci E, et al. Thyroid hormones, oxidative stress, and inflammation. Mediators Inflamm 2016;2016.

[88] Wang D, Feng J-F, Zeng P, Yang Y-H, Luo J, Yang Y-W. Total oxidant/antioxidant status in sera of patients with thyroid cancers. Endocr Relat Cancer 2011;18:773–82.

[89] Piazera BKL, Gomes DV, Vigário P, Salerno VP, Vaisman M. Evaluation of redox profiles in exogenous subclinical hyperthyroidism at two different levels of TSH suppression. Arch Endocrinol Metabol 2018;62:545–51.

[90] Fahim YA, Sharaf NE, Hasani IW, Ragab EA, Abdelhakim HK. Assessment of thyroid function and oxidative stress state in foundry workers exposed to lead. J Health Pollut 2020;10:200903.

[91] Kochman J, Jakubczyk K, Bargiel P, Janda-Milczarek K. The influence of oxidative stress on thyroid diseases. Antioxidants (Basel) 2021;10.

[92] Chakrabarti SK, Ghosh S, Banerjee S, Mukherjee S, Chowdhury S. Oxidative stress in hypothyroid patients and the role of antioxidant supplementation, Indian. J Endocrinol Metab 2016;20:674–8.

[93] Riis KR, Larsen CB, Medici BR, Jensen CZ, Winther KH, Larsen EL, et al. Hypothyroid women have persistently higher oxidative stress compared to healthy controls. Eur Thyroid J 2023;12:e230167.

[94] Resch U, Helsel G, Tatzber F, Sinzinger H. Antioxidant Status in Thyroid Dysfunction. 2002.

[95] Staal J, Blanco LP, Perl A. Editorial: mitochondrial dysfunction in inflammation and autoimmunity. Front Immunol 2023;14:1304315.

[96] Jing W, Liu C, Su C, Liu L, Chen P, Li X, et al. Role of reactive oxygen species and mitochondrial damage in rheumatoid arthritis and targeted drugs. Front Immunol 2023;14.

[97] Yang Z, Shen Y, Oishi H, Matteson EL, Tian L, Goronzy JJ, et al. Restoring oxidant signaling suppresses proarthritogenic T cell effector functions in rheumatoid arthritis. Sci Transl Med 2016;8. 331ra38-331ra38.

[98] Doherty E, Oaks Z, Perl A. Increased mitochondrial electron transport chain activity at complex I is regulated by N-acetylcysteine in lymphocytes of patients with systemic lupus erythematosus. Antioxid Redox Signal 2014;21:56–65.

[99] Bartalena L, Piantanida E. Cigarette smoking: number one enemy for graves ophthalmopathy. Pol Arch Med Wewn 2016;126:725–6.

[100] Oeverhaus M, Winkler L, Stähr K, Daser A, Bechrakis N, Stöhr M, et al. Influence of biological sex, age and smoking on Graves' orbitopathy - a ten-year tertiary referral center analysis. Front Endocrinol (Lausanne) 2023;14:1160172.

[101] Bartalena L, Martino E, Marcocci C, Bogazzi F, Panicucci M, Velluzzi F, et al. More on smoking habits and Graves' ophthalmopathy. J Endocrinol Invest 1989;12:733–7.

[102] Thornton J, Kelly S, Harrison R, Edwards R. Cigarette smoking and thyroid eye disease: a systematic review. Eye 2007;21:1135–45.

[103] Lu R, Wang P, Wartofsky L, Sutton BD, Zweier JL, Bahn RS, et al. Oxygen free radicals in interleukin-lβ-induced glycosaminoglycan production by retro-ocular fibroblasts from normal subjects and graves' ophthalmopathy patients. Thyroid 1999;9:297–303.

[104] Lahooti H, Champion B, Wall JR. Relationship between smoking and serum levels of eye muscle and orbital connective tissue antibodies in patients with graves ophthalmopathy. Endocrine 2023. https://doi.org/10.1007/s12020-023-03335-5.

[105] Xing L, Ye L, Zhu W, Shen L, Huang F, Jiao Q, et al. Smoking was associated with poor response to intravenous steroids therapy in Graves' ophthalmopathy. Br J Ophthalmol 2015;99:1686–91.

[106] Görtz GE, Horstmann M, Aniol B, Reyes BD, Fandrey J, Eckstein A, et al. Hypoxia-dependent HIF-1 activation impacts on tissue remodeling in graves' ophthalmopathy-implications for smoking. J Clin Endocrinol Metab 2016;101:4834–42.

[107] Lanzolla G, Marcocci C, Marinò M. Antioxidant therapy in Graves' Orbitopathy. Front Endocrinol 2020;11.

[108] Diana T, Daiber A, Oelze M, Neumann S, Olivo PD, Kanitz M, et al. Stimulatory TSH-receptor antibodies and oxidative stress in graves disease. J Clin Endocrinol Metab 2018;103:3668–77.

[109] Hou T-Y, Wu S-B, Kau H-C, Tsai C-C. The role of oxidative stress and therapeutic potential of antioxidants in graves&rsquo. Ophthal Biomed 2021;9:1871.

[110] Bednarek J, Wysocki H, Sowiński J. Oxidative stress peripheral parameters in Graves' disease: the effect of methimazole treatment in patients with and without infiltrative ophthalmopathy. Clin Biochem 2005;38:13–8.

[111] Bednarek J, Wysocki H, Sowiński J. Peripheral parameters of oxidative stress in patients with infiltrative Graves' ophthalmopathy treated with corticosteroids. Immunol Lett 2004;93:227–32.

[112] Acibucu F, Öztürk DD, Kizildag C, Aslan MZ, Gulumsek E, Sumbul MS, et al. Proptosis is associated with thiol-disulfide in patients with Graves' ophthalmopathy. Arch Endocrinol Metab 2022;66:191–7.

[113] Choi W, Li Y, Ji YS, Yoon KC. Oxidative stress markers in tears of patients with Graves' orbitopathy and their correlation with clinical activity score. BMC Ophthalmol 2018;18:303.

[114] Tsai CC, Cheng CY, Liu CY, Kao SC, Kau HC, Hsu WM, et al. Oxidative stress in patients with Graves' ophthalmopathy: relationship between oxidative DNA damage and clinical evolution. Eye (Lond) 2009;23:1725–30.

[115] Tsai C-C, Kao S-C, Cheng C-Y, Kau H-C, Hsu W-M, Lee C-F, et al. Oxidative stress change by systemic corticosteroid treatment among patients having active graves ophthalmopathy. Arch Ophthalmol 2007;125:1652–6.

[116] Akarsu E, Buyukhatipoglu H, Aktaran S, Kurtul N. Effects of pulse methylprednisolone and oral methylprednisolone treatments on serum levels of oxidative stress markers in Graves' ophthalmopathy. Clin Endocrinol (Oxf) 2011;74:118–24.

[117] Londzin-Olesik M, Kos-Kudla B, Karpe J, Nowak A, Nowak M. The effect of immunosuppression on selected antioxidant parameters in patients with graves' disease with active thyroid-associated orbitopathy. Exp Clin Endocrinol Diabetes 2021;129:762–9.

[118] Tsai CC, Wu SB, Cheng CY, Kao SC, Kau HC, Chiou SH, et al. Increased oxidative DNA damage, lipid peroxidation, and reactive oxygen species in cultured orbital fibroblasts from patients with Graves' ophthalmopathy: evidence that oxidative stress has a role in this disorder. Eye (Lond) 2010;24:1520–5.

[119] Tsai CC, Wu SB, Cheng CY, Kao SC, Kau HC, Lee SM, et al. Increased response to oxidative stress challenge in Graves' ophthalmopathy orbital fibroblasts. Mol Vis 2011;17:2782–8.

[120] Hondur A, Konuk O, Dincel AS, Bilgihan A, Unal M, Hasanreisoglu B. Oxidative stress and antioxidant activity in orbital fibroadipose tissue in graves' ophthalmopathy. Curr Eye Res 2008;33:421–7.

[121] Heufelder AE, Wenzel BE, Bahn RS. Methimazole and propylthiouracil inhibit the oxygen free radical-induced expression of a 72 kilodalton heat shock protein in Graves' retroocular fibroblasts. J Clin Endocrinol Metabol 1992;74:737–42.

[122] Burch HB, Lahiri S, Bahn RS, Barnes S. Superoxide radical production stimulates retroocular fibroblast proliferation in Graves' ophthalmopathy. Exp Eye Res 1997;65:311–6.

[123] Lu RUI, Wang P, Wartofsky L, Sutton BD, Zweier JL, Bahn RS, et al. Oxygen free radicals in interleukin-lβ-induced glycosaminoglycan production by retro-ocular

fibroblasts from normal subjects and graves' ophthalmopathy patients. Thyroid® 1999;9:297–303.
[124] Bartalena L, Tanda ML, Piantanida E, Lai A. Oxidative stress and Graves' ophthalmopathy: in vitro studies and therapeutic implications. BioFactors 2003; 19:155–63.
[125] Van Regemorter E, Joris V, Van Regemorter V, Marique L, Behets C, Lengelé B, et al. Downregulation of caveolin-1 and upregulation of deiodinase 3, Associated with hypoxia-inducible factor-1α increase, are involved in the oxidative stress of graves' orbital adipocytes. Thyroid® 2020;31:627–37.
[126] Tsai CC, Wu SB, Kao SC, Kau HC, Lee FL, Wei YH. The protective effect of antioxidants on orbital fibroblasts from patients with Graves' ophthalmopathy in response to oxidative stress. Mol Vis 2013;19:927–34.
[127] Buonfiglio F, Böhm EW, Pfeiffer N, Gericke A. Oxidative stress: a suitable therapeutic target for optic nerve diseases? Antioxidants 2023;12:1465.
[128] Ko J, Kim JY, Kyoung Chae M, Jig Lee E, Sook Yoon J. PERK mediates oxidative stress and adipogenesis in Graves' orbitopathy pathogenesis. J Mol Endocrinol 2021;66:313–23.
[129] Wiersinga WM. Smoking and thyroid. Clin Endocrinol (Oxf) 2013;79:145–51.
[130] Yuksel N, Yaman D, Tugce Pasaoglu O, Pasaoglu H. The effect of smoking on mitochondrial biogenesis in patients with graves ophthalmopathy. Ophthal Plast Reconstr Surg 2020;36:172–7.
[131] Kau HC, Wu SB, Tsai CC, Liu CJ, Wei YH. Cigarette smoke extract-induced oxidative stress and fibrosis-related genes expression in orbital fibroblasts from patients with graves' ophthalmopathy. Oxid Med Cell Longev 2016;2016: 4676289.
[132] Cawood TJ, Moriarty P, O'Farrelly C, O'Shea D. Smoking and thyroid-associated ophthalmopathy: a novel explanation of the biological link. J Clin Endocrinol Metabol 2007;92:59–64.
[133] Yoon JS, Lee HJ, Chae MK, Lee SY, Lee EJ. Cigarette smoke extract-induced adipogenesis in Graves' orbital fibroblasts is inhibited by quercetin via reduction in oxidative stress. J Endocrinol 2013;216:145–56.
[134] Görtz GE, Philipp S, Bruderek K, Jesenek C, Horstmann M, Henning Y, et al. Macrophage-orbital fibroblast interaction and hypoxia promote inflammation and adipogenesis in graves' orbitopathy. Endocrinology 2022;164.
[135] Marique L, Senou M, Craps J, Delaigle A, Van Regemorter E, Wérion A, et al. Oxidative stress and upregulation of antioxidant proteins, including adiponectin, in extraocular muscular cells, orbital adipocytes, and thyrocytes in graves' disease associated with orbitopathy. Thyroid® 2015;25:1033–42.
[136] Craps J, Joris V, Baldeschi L, Daumerie C, Camboni A, Buemi A, et al. miR-199a downregulation as a driver of the NOX4/HIF-1α/VEGF-a pathway in thyroid and orbital adipose tissues from graves′ Patients. Int J Mol Sci 2022;23:153.
[137] Hikage F, Atkins S, Kahana A, Smith TJ, Chun T-H. HIF2A–LOX pathway promotes fibrotic tissue remodeling in thyroid-associated orbitopathy. Endocrinology 2018;160:20–35.
[138] Khong JJ, McNab AA, Ebeling PR, Craig JE, Selva D. Pathogenesis of thyroid eye disease: review and update on molecular mechanisms. Br J Ophthalmol 2016;100: 142–50.
[139] Akbarian S, Chaibakhsh S, Kashkouli MB, Karimi N, Abdolalizadeh P, Ghahvehchian H. A systematic review on the role of antioxidants in thyroid eye disease. J Curr Ophthalmol 2022;34:16–24.
[140] Duntas LH. Selenium and inflammation: underlying anti-inflammatory mechanisms. Horm Metab Res 2009;41:443–7.
[141] Rayman MP. The importance of selenium to human health. Lancet 2000;356: 233–41.
[142] Rotondo Dottore G, Leo M, Casini G, Latrofa F, Cestari L, Sellari-Franceschini S, et al. Antioxidant actions of selenium in orbital fibroblasts: a basis for the effects of selenium in graves' orbitopathy. Thyroid 2017;27:271–8.
[143] Kim BY, Jang SY, Choi DH, Jung CH, Mok JO, Kim CH. Anti-inflammatory and antioxidant effects of selenium on orbital fibroblasts of patients with graves ophthalmopathy. Ophthal Plast Reconstr Surg 2021;37:476–81.
[144] Marcocci C, Kahaly GJ, Krassas GE, Bartalena L, Prummel M, Stahl M, et al. Selenium and the course of mild Graves' orbitopathy. N Engl J Med 2011;364: 1920–31.
[145] Lanzolla G, Marinò M, Marcocci C. Selenium in the treatment of Graves' hyperthyroidism and eye disease. Front Endocrinol (Lausanne) 2020;11:608428.
[146] Hou TY, Wu SB, Kau HC, Tsai CC. The role of oxidative stress and therapeutic potential of antioxidants in Graves' Ophthalmopathy. Biomedicines 2021;9.
[147] Rotondo Dottore G, Ionni I, Menconi F, Casini G, Sellari-Franceschini S, Nardi M, et al. Action of three bioavailable antioxidants in orbital fibroblasts from patients with Graves' orbitopathy (GO): a new frontier for GO treatment? J Endocrinol Invest 2018;41:193–201.
[148] Mokhtari V, Afsharian P, Shahhoseini M, Kalantar SM, Moini A. A review on various uses of N-acetyl cysteine. Cell J (Yakhteh) 2017;19:11.
[149] Lai ZW, Hanczko R, Bonilla E, Caza TN, Clair B, Bartos A, et al. N-acetylcysteine reduces disease activity by blocking mammalian target of rapamycin in T cells from systemic lupus erythematosus patients: a randomized, double-blind, placebo-controlled trial. Arthritis Rheum 2012;64:2937–46.
[150] Winans KR, Lai ZW, Faraone S, Phillips PE, Banki K, Perl A. MTORC1 blockade with rapamycin and N-acetylcysteine reduces anti-phospholipid antibody levels in controlled clinical trials of patients with SLE. Arthritis Rheumatol 2017;69 (suppl 10). https://acrabstracts.org/abstract/mtorc1-blockade-with-rapamycin-and-n-acetylcysteine-reduces-anti-phospholipid-antibody-levels-in-controlled-clinical-trials-of-patients-with-sle/. Accessed March 15, 2024.
[151] Perl A. mTOR-dependent autophagy contributes to end-organ resistance and serves as target for treatment in autoimmune disease. eBioMedicine 2018;36: 12–3.
[152] Nasr S, Perl A. Principles behind SLE treatment with N-acetylcysteine. Immunometabolism 2022;4:e00010.
[153] Rotondo Dottore G, Ionni I, Menconi F, Casini G, Sellari-Franceschini S, Nardi M, et al. Antioxidant effects of β-carotene, but not of retinol and vitamin E, in orbital fibroblasts from patients with Graves' orbitopathy (GO). J Endocrinol Invest 2018;41:815–20.
[154] Xu D, Hu MJ, Wang YQ, Cui YL. Antioxidant activities of quercetin and its complexes for medicinal application. Molecules 2019;24.
[155] Yoon JS, Lee HJ, Choi SH, Chang EJ, Lee SY, Lee EJ. Quercetin inhibits IL-1β-induced inflammation, hyaluronan production and adipogenesis in orbital fibroblasts from Graves' orbitopathy. PloS One 2011;6:e26261.
[156] Lisi S, Botta R, Lemmi M, Sellari-Franceschini S, Altea M, Sisti E, et al. Quercetin decreases proliferation of orbital fibroblasts and their release of hyaluronic acid. J Endocrinol Invest 2011;34:521–7.
[157] Yoon JS, Chae MK, Lee SY, Lee EJ. Anti-inflammatory effect of quercetin in a whole orbital tissue culture of Graves' orbitopathy. Br J Ophthalmol 2012;96: 1117–21.
[158] Yoon JS, Chae MK, Jang SY, Lee SY, Lee EJ. Antifibrotic effects of quercetin in primary orbital fibroblasts and orbital fat tissue cultures of Graves' orbitopathy. Invest Ophthalmol Vis Sci 2012;53:5921–9.
[159] Hai YP, Lee ACH, Chen K, Kahaly GJ. Traditional Chinese medicine in thyroid-associated orbitopathy. J Endocrinol Invest 2023;46:1103–13.
[160] Lee JS, Kim J, Lee EJ, Yoon JS. Therapeutic effect of curcumin, a plant polyphenol extracted from *Curcuma longae*, in fibroblasts from patients with graves' orbitopathy. Invest Ophthalmol Vis Sci 2019;60:4129–40.
[161] Yu WK, Hwang WL, Wang YC, Tsai CC, Wei YH. Curcumin suppresses TGF-β1-induced myofibroblast differentiation and attenuates angiogenic activity of orbital fibroblasts. Int J Mol Sci 2021;22.
[162] Galiniak S, Aebisher D, Bartusik-Aebisher D. Health benefits of resveratrol administration. Acta Biochim Pol 2019;66:13–21.
[163] Borra MT, Smith BC, Denu JM. Mechanism of human SIRT1 activation by resveratrol*. J Biol Chem 2005;280:17187–95.
[164] Ciccone L, Piragine E, Brogi S, Camodeca C, Fucci R, Calderone V, et al. Resveratrol-like compounds as SIRT1 activators. Int J Mol Sci 2022;23.
[165] Kim CY, Lee HJ, Chae MK, Byun JW, Lee EJ, Yoon JS. Therapeutic effect of resveratrol on oxidative stress in graves' orbitopathy orbital fibroblasts. Invest Ophthalmol Vis Sci 2015;56:6352–61.
[166] Li H, Ma C, Liu W, He J, Li K. Gypenosides protect orbital fibroblasts in graves ophthalmopathy via anti-inflammation and anti-fibrosis effects. Invest Ophthalmol Vis Sci 2020;61(5):64.
[167] Ma C, Li H, Liu W, Lu S, Li X, Chen J, et al. Therapeutic effect of gypenosides on antioxidant stress injury in orbital fibroblasts of graves' orbitopathy. J Immunol Res 2022:4432584. https://doi.org/10.1155/2022/4432584.
[168] Imenshahidi M, Hosseinzadeh H. Chapter 14 - Berberine neuroprotection and antioxidant activity. In: Martin CR, Preedy VR, editors. Oxidative stress and dietary antioxidants in neurological diseases. Academic Press; 2020. p. 199–216.
[169] Diao J, Chen X, Mou P, Ma X, Wei R. Potential therapeutic activity of berberine in thyroid-associated ophthalmopathy: inhibitory effects on tissue remodeling in orbital fibroblasts. Invest Ophthalmol Vis Sci 2022;63:6.
[170] Li H, Zhang Y, Min J, Gao L, Zhang R, Yang Y. Astragaloside IV attenuates orbital inflammation in Graves' orbitopathy through suppression of autophagy. Inflamm Res 2018;67:117–27.
[171] Li H, Yuan Y, Zhang Y, Zhang X, Gao L, Xu R. Icariin inhibits AMPK-dependent autophagy and adipogenesis in adipocytes in vitro and in a model of Graves' orbitopathy in vivo. Front Physiol 2017;8:45.
[172] Li H, Min J, Chen Y, Li H, Zhang Y. Polydatin attenuates orbital oxidative stress in Graves' orbitopathy through the NRF2 pathway. Chem Biol Interact 2020;315: 108894.
[173] Rhiu S, Chae MK, Lee EJ, Lee JB, Yoon JS. Effect of tanshinone IIA in an in vitro model of Graves' orbitopathy. Invest Ophthalmol Vis Sci 2014;55:5900–10.
[174] Guo R, Li L, Su J, Li S, Duncan SE, Liu Z, et al. Pharmacological activity and mechanism of tanshinone IIA in related diseases. Drug Des Devel Ther 2020: 4735–48.
[175] Chandran G, Sirajudeen KN, Yusoff NS, Swamy M, Samarendra MS. Effect of the antihypertensive drug enalapril on oxidative stress markers and antioxidant enzymes in kidney of spontaneously hypertensive rat. Oxid Med Cell Longev 2014;2014:608512.
[176] de Cavanagh EM, Inserra F, Ferder L, Fraga CG. Enalapril and captopril enhance glutathione-dependent antioxidant defenses in mouse tissues. Am J Physiol Regul Integr Comp Physiol 2000;278:R572–7.
[177] Botta R, Lisi S, Marcocci C, Sellari-Franceschini S, Rocchi R, Latrofa F, et al. Enalapril reduces proliferation and hyaluronic acid release in orbital fibroblasts. Thyroid 2013;23:92–6.
[178] Ataabadi G, Dabbaghmanesh MH, Owji N, Bakhshayeshkaram M, Montazeri-Najafabady N. Clinical features of graves' ophthalmopathy and impact of enalapril on the course of mild graves' ophthalmopathy: a pilot study. Endocr Metab Immune Disord Drug Targets 2020;20:139–48.
[179] Waldeck-Weiermair M, Yadav S, Kaynert J, Thulabandu VR, Pandey AK, Spyropoulos F, et al. Differential endothelial hydrogen peroxide signaling via Nox isoforms: critical roles for Rac1 and modulation by statins. Redox Biol 2022;58: 102539.

[180] Nilsson A, Tsoumani K, Planck T. Statins decrease the risk of orbitopathy in newly diagnosed patients with graves disease. J Clin Endocrinol Metabol 2021;106:1325–32.

[181] Lanzolla G, Sabini E, Leo M, Menconi F, Rocchi R, Sframeli A, et al. Statins for Graves' orbitopathy (STAGO): a phase 2, open-label, adaptive, single Centre, randomised clinical trial. Lancet Diabetes Endocrinol 2021;9:733–42.

[182] Broderick C, Forster R, Abdel-Hadi M, Salhiyyah K. Pentoxifylline for intermittent claudication. Cochrane Database Syst Rev 2020;(Issue 10):CD005262. Art. No.: CD005262.

[183] Zajączkowski S, Ziółkowski W, Badtke P, Zajączkowski MA, Flis DJ, Figarski A, et al. Promising effects of xanthine oxidase inhibition by allopurinol on autonomic heart regulation estimated by heart rate variability (HRV) analysis in rats exposed to hypoxia and hyperoxia. PloS One 2018;13:e0192781.

[184] Bouzas EA, Karadimas P, Mastorakos G, Koutras DA. Antioxidant agents in the treatment of Graves' ophthalmopathy. Am J Ophthalmol 2000;129:618–22.

[185] Finamor F, Martins JRM, Nakanami D, Paiva ERD, Manso PG, Furlanetto RP. Pentoxifylline (PTX)-an alternative treatment in Graves' ophthalmopathy (inactive phase): assessment by a disease specific quality of life questionnaire and by exophthalmometry in a prospective randomized trial. Eur J Ophthalmol 2004;14:277–83.

[186] Balazs C, Kiss E, Vamos A, Molnar I, Farid N. Beneficial effect of pentoxifylline on thyroid associated ophthalmopathy (TAO): a pilot study. J Clin Endocrinol Metabol 1997;82:1999–2002.

[187] Balazs C, Kiss E, Farid N. Inhibitory effect of pentoxifylline on HLA-DR expression and glycosaminoglycan synthesis of retrobulbar fibroblasts induced by interferon gamma. Acta Microbiol Immunol Hung 1997;44:173–9.