The Conjunctiva Plays an Important Role in Modulating Ocular Surface Tear

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Introduction

Manifestation of dry eye disease (DED) includes tear film (TF) instability and hyperosmolar condition, leading to immunoinflammatory and mechanical injuries to the ocular surface [1, 2]. Specifically, DED involves defects in aqueous, lipid and/or mucin layers of the TF [3, 4]. Etiologically DED is categorized into aqueous tear-deficient and evaporative dry eye [5]. In the United States, 3.23 million women and 1.68 million men 50 years and older suffer from varied severity of DED [6, 7]. Most importantly, DED largely impairs the activities of daily living, thus negatively impacting vision related quality of life [8]. Since ocular surface plays a significant role in modulating tear volume and composition, maintaining and/or restoring the normal function of ocular surface tissues could be a novel treatment strategy for DED.

The ocular surface is a collection of anatomically continuous epithelial and glandular tissues that are functionally linked to maintain the TF [10]. The main lacrimal gland (LG) has been considered the major source of tears [11]. However, there is strong evidence that ocular surface tissues are competent to maintain adequate tear secretion in the absence of the main LG [12]. In fact, contributions from accessory LGs, corneal and conjunctival epithelia to tear volume have been widely recognized [13, 14, 15, 16]. The conjunctiva, and to a very small extent the cornea, are involved in basal tear production [9]. The conjunctiva in particular has the ability to modify the TF by absorbing/secreting electrolytes and water and by secreting proteins such as mucin [17]. It has also been previously speculated that conjunctival fluid flow may play an important role in hydrating the mucus secreted by goblet cells [18]. Previous studies supported that rabbit conjunctival epithelium has the capacity to be the primary source of TF [19-21]. In rabbits, the basal conjunctival fluid secretion rate is 0.79 ml/min whereas the basal tear production rate is 0.72 ml/min [19] and conjunctival fluid secretion was reported to be 175% greater than tear turnover [20, 21]. The conjunctiva occupies 17 times more surface area than the cornea in human and about 9 times more in rabbits [22]. Given such a large surface area ratio, the role played by the conjunctiva in modulating the TF should not be underestimated [2, 22].

The conjunctival fluid flow can either be paracellular [23] or transcellular [24] and both pathways are driven by either active Cl− secretion or an osmotic gradient. The osmotic gradient (which regulates the direction of water flow across plasma membranes) is established by the net influx of Na+, K+ and Cl− into cells and efflux of H+ and HCO3− out of cells; both corneal and conjunctival epithelia have the ability to secrete Cl− and absorb Na+ [17]. In fact, in conjunctival epithelium, Cl− secretion (basolateral to apical movement) accounts for nearly 60% and Na+ reabsorption (apical to basolateral) accounts for about 40% of net ionic transport [17, 26]. This ionic transport determines the amount of tears collected in the conjunctiva sac which possibly affects the stability of TF [27]. It has been reported that uridine triphosphate analog such as Diquafosol (targeting surface epithelial P2Y2 receptors) stimulates Cl− and fluid secretion from the mucosal surface of isolated rabbit conjunctiva [19, 28] as well as increases mucin-like glycoprotein secretion from the ocular surface of rats and rabbits [29, 30]. Topical
ocular instillation of 3% diquafosol ophthalmic solution has been shown efficacious in human DED treatment [31, 32]. This again strongly supports the involvement of conjunctival epithelium in ocular fluid secretion.

The ionic gradient across ocular surface is regulated by ionic transporters across the conjunctival epithelium such as cystic fibrosis transmembrane conductance regulator (CFTR), sodium potassium chloride co-transporter, sodium potassium ATPase, and epithelial sodium channels (ENaC) [25]. The CFTR, a c-AMP activated Cl channel, functions as potential major pathways for Cl transport at the ocular surface [9]. The competence for CFTR facilitated Cl transport has been previously shown at the ocular surface in mouse [33] and subsequently in rat conjunctiva [34]. Similarly, the ENaC in mouse, [35] rabbit and human conjunctiva [36] mediates active Na+ re-absorption to maintain the electrolyte/water homeostasis [27].

Further, the transcellular flow is potentially mediated in parts by the membranous aquaporins (AQPs) type water selective channels in the ocular surface, [21] supporting their possible role in water/fluid transport and TF homeostasis [37]. The AQP-3, [38] AQP-4 [16] and AQP-5 [16, 39] have been detected in the conjunctiva. Our research demonstrated that tear secretion was not affected in the absence of the main LG, nictitating membrane and Harderian gland in rabbits [16, 40]. A spontaneous improvement of dry eye phenotypes and ocular surface inflammation (with no external intervention) were also observed in this mixed-mechanism rabbit dry eye model. Our findings strongly suggested the presence of a compensatory mechanism in the remaining ocular surface tissues. Our group demonstrated for the first time that AQP-4 is expressed by rabbit conjunctival epithelium and in addition, revealed the potential role of AQP-4 and AQP-5 in the tear fluid secretion by the conjunctival epithelium [16, 40]. Interestingly, it has been concluded that water transport facilitated by conjunctival encoded AQP-3 does not play any role in transconjunctival fluid movement [37]. Overall, across conjunctival epithelium, AQPs and CFTR have been identified as the principal molecular pathways for water and Cl transport, respectively, [2, 40] thus serving as attractive targets for drug development in the treatment of DED [40, 41]. A recent report demonstrated in mice the potential utility and efficacy of newly identified small-molecule CFTR activators as a novel prosecretory treatment for DED [2].

**Conclusion**

In conclusions, significant contributions from the conjunctival epithelium in tear secretion are being increasingly recognized. The conjunctiva possibly plays a pivotal role in the maintenance of ocular surface homeostasis. Thus, understanding the physiology of conjunctival epithelium in order to optimize its fluid and mucin secretion capacity is imperative for developing alternative DED treatments. Further research will be crucial to delineate specific mechanisms by which the conjunctival epithelium modulates tear quality and quantity, and to identify potential novel treatment strategies for DED.

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