Keratolytic and Keratostatic Activity of Selenium Sulfide (SeS2)in Modifying Meibomian Gland Disease

Abstract # 3533505

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BACKGROUND

Meibomian gland dysfunction (MGD) is the leading cause of dry eye disease, clinical signs of obstructive MGD – characterized by reduced secretion and alterations in the physical properties of meibum – are present in 86% of people with DED. [1]. Research over the past decade has pointed to hyperkeratinization, or the build-up and shedding of the keratin protein into the meibomian gland ducts, as the underline cause of obstructive MGD. The shedding of keratinized material into the meibomian gland ducts results in obstruction of the gland orifice, degenerative dilatation of the duct and acinar cells, gland atrophy, changes in the viscosity and appearance of the meibum, and reduced meibum output [2,3,4] . The goal of MGD treatment is to restore the normal flow of meibomian gland secretions. It is hypothesized that removing or softening the keratin obstruction will enable restoration of normal mebium secretion. Meibomian glands and sebaceous glands share the same embryological origin, hence adopting dermatologic treatments using keratolytic agents to treat skin conditions (e.g., dandruff) and applying them to lid margin diseases such as MGD is likely to succeed. Selenium sulfide (SeS2), a potent keratolytic and keratostatic agent, breaks down aberrant disulfide bonds and prevents access keratin deposition, thus it is a potential treatment of MGD.

OBJECTIVES

Determine whether SeS2 has an effect on the following:

- Keratolytic activity determined by the reduction of disulfide bonds and formation of free thiols
- Keratostatic activity affecting cellular differentiation & proliferation of keratocytes determined by evaluation of tissue and cellular turn over

METHODS

Keratolytic activity: evaluated using ex-Vivo human tissue; Human skin organ culture was obtained from a healthy patient undergoing plastic surgery (female; abdominal). The study was initiated at the day of surgery. The skin was cleaned, washed with PBS and then the epidermis was peeled.

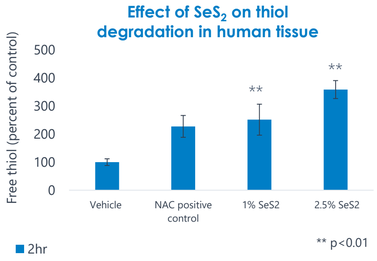
Thiol activity: the epidermis samples were incubated with 0.005% trypsin (in PBS) at 37°C overnight to obtain the stratum corneum. Stratum corneum was then incubate with different test articles or controls. Ellman's reagent was used for quantification of free thiols (S-H groups), formed from keratin reduction of disulfide bonds.

Keratostatic tissue turn over: Test Items and vehicles were applied topically and incubate for 2hr, followed by epidermis separation from the dermis and staining with BrdU.

Keratostatic in-vitro: effect evaluated using human epidermal keratinocytes (NHEK) Cell proliferation using MTT assay, Hoechst staining and BrdU incorporation assay.

Cell turnover using FACS propidium iodide (PI)

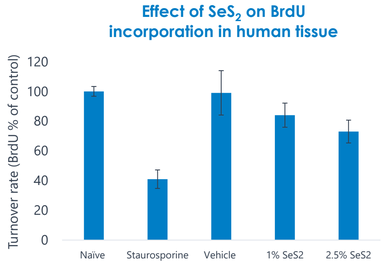
RESULTS: KERTOLYTIC EFFECT HUMAN TISSUE



Thiol degradation

SeS_2 caused significant keratolytic activity in human skin with 251% and 358% increase in free thiols following 2hr incubation with 1% and 2.5% SeS_2 formulation, respectively.

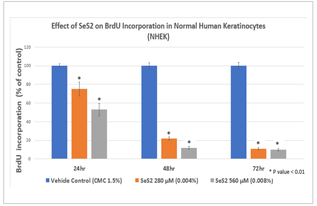
RESULTS: KERTOLYTIC EFFECT HUMAN TISSUE



BrdU incorporation

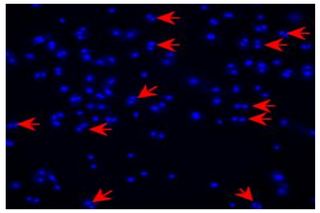
SeS_2 caused concentration dependent reduction of cell turnover ($p<0.05$). Staurosporine, a known cell cycle arrest agent, used as positive control.

RESULTS: KERTOSTATIC EFFECT NHEK CELL LINE



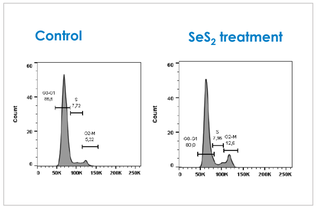
BrdU incorporation

Concentration- and time-dependent reduction of keratinocyte proliferation observed at 24, 48 and 72hr ($p<0.01$) following treatment with SeS_2.



Hoechst staining

Numerous paired nuclei, suggestive of a cell division slowing down or blockade



FACS – cell cycle

Increased accumulation of cells in the cell cycle & mitosis phase (G2/M phase), indicate decrease in cell division

CONCLUSIONS

The pathogenesis of MGD is a result of accumulation of excess keratin in the gland's canal and within the lipid. The results suggest that selenium sulfide works in multiple pathways that are involved in MGD including reduced cell turnover and activity of keratinocytes and softening of keratin through the reduction of disulfide bonds. Hence both in vitro and ex vivo models suggest that selenium sulfide may be a promising candidate for the treatment of MGD

DISCLOSURES

Yair Alster- Azura Ophthalmics; I, E, P&S
Hila Epstein-Barash – Azura Ophthalmics; I, E&P
Hadas Rapaport - Azura Ophthalmics; I, E&P
Omer Rafaeli - Azura Ophthalmics; I, E&P

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