The effect of isotretinoin administration and withdrawal on submandibular salivary gland of albino rats
(Histological and Immunohistochemical study)

Thesis

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﴿ قَالَ لَهُ مُوسَى هَلْ أَتَّبِعُكَ عَلَى أَنْ تُعَلِّمَنِ مِمَّا عُلِّمْتَ رُشْدًا ﴾

صدق الله العظيم

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Dedication

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List of Abbreviations

Abbreviation	Meaning
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
BV	Blood vessel
Cl	Chloride
CT	Connective tissue
dl	Deciliter
ED	Excretory ducts
EM	Electron microscope
GCT	Granular convoluted tubules
gm	Gram
H&E	Hematoxylin and Eosin
H1	Histamine 1 receptor
H_2O_2	Hydrogen peroxide
HCl	Hydrochloric acid
HCO_3	Bicarbonate
ID	Intercalated ducts
Ig A	Immunoglobulin A
K	Potassium
kg	Kilogram
Lab	Laboratory
LM	Light microscope
mag	Magnification
mg	Milligram
Na	Sodium
^oC	Celcius
Orig.	Original
pH	Potential Hydrogen
RBCs	Red blood cells
ROS	Reactive oxygen species

St.D	Striated ducts
SD	Standard deviation
SG	Salivary gland
SMG	Submandibular salivary gland
T3	Triiodothyronine
T4	Thyroxine
TEM	Transmission electron microscope
TNF	Tumor necrosis factor
TRAIL	TNF related apoptosis inducing ligand
TSH	Thyroid stimulating hormone

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Abstract

Background: Isotretinoin is the most common drug used in acne treatment worldwide, and despite its wide range of dermatological indications, it has many side effects on different body tissues.

Aim of the study: To investigate the effect of Isotretinoin on rat submandibular salivary gland histologically and immunohistochemically.

Materials and methods: Forty-two adult male albino rats were used, divided into control and experimental groups. The control group (group I) was further subdivided into 3 subgroups: subgroup IA, IB and IC. Referring to the experimental group (group II) was also further subdivided into three subgroups: subgroup IIA received 1.5 mg/kg/day Isotretinoin orally for 28 days, subgroup IIB received same dose as subgroup IIA and duration but were left for recovery for one week and subgroup IIC received same dose and duration but were left for recovery for 2 weeks.

Results: Histological examination of the subgroup IIA (Isotretinoin group) showed that acinar cells demonstrated some histological alterations including pyknotic, hyperchromatic and crescent shaped nuclei with variable sized cytoplasmic vacuolations. The ducts showed some signs of degeneration with loss of their normal cellular outlines. However, there was some improvement in subgroup IIB that escalated in subgroup IIC that was almost close to the control group or the normal state.

Apoptotic changes expressed by anti-active caspase 3 were highest in subgroup IIA and statistical results showed statistical significance between studied groups.

Conclusion: The use of Isotretinoin leads to histological alterations in submandibular salivary glands of rats by inducing apoptotic effect on their cells.

Introduction

Saliva is a most valuable oral fluid that is critical to the preservation and maintenance of oral health, it is often taken for granted and receives little attention until quantity or quality is diminished. Saliva is a clear, slightly acidic mucoserous secretion obtained from the major and minor salivary glands. The submandibular salivary gland is one of the major salivary glands, it is a compound tubuloalveolar exocrine gland that contributes to approximately two thirds of salivary output **(De Almeida et al., 2008)**.

When the amount of saliva that bathes the oral cavity is reduced, a condition known as Xerostomia develops commonly known as dry mouth. The use of xerostomic medications is the most frequently reported cause of xerostomia **(Guggenheimer and Moore, 2003).**

Systemic retinoids are natural or synthetic derivatives of vitamin A, one of which is isotretinoin (13 cis retinoic acid) an effective and increasingly popular treatment for acne vulgaris. Isotretinoin is approved as treatment of severe acne resistant to standard therapy. It has been prescribed to many patients worldwide since it was first introduced in 1982 **(Borovaya et al., 2014; Li et al., 2012)**.

Isotretinoin is used for an extensive array of other dermatological conditions including photoaging, rosacea, seborrheic dermatitis, folliculitis, granuloma annulare, periorificial dermatitis, a variety of disorders of keratinization, and sarcoidosis **(Charakida et al., 2004)**.

Although isotretinoin is a potent anti-acne drug and has many beneficial uses, it has a multitude of side effects, some of them are well known and predictable as xerosis, xerostomia and cheilitis **(Brzezinski et al., 2017)**. Also, it is a known teratogenic medication and its prohibited in pregnancy. Besides some less common side effects as depressive and suicidal issues **(Oliveira et al., 2018)**. Thus studying its effect on the submandibular salivary gland was the main scope of this study.

Review of Literature

Salivary glands are fundamental organs that produce and secrete saliva to the oral cavity, they are essential for maintaining the oral cavity homeostasis. Saliva is a multifunctional fluid, it provides the mucosal lubrication, salivary electrolytes, antibacterial components and different enzymes to protect and maintain the oral mucosa and teeth surface integrity **(Feller et al., 2013; Carpenter, 2013)** Therefore, the secretory function of salivary glands is extremely important for maintaining the oral health and its partial or total loss due to gland loss or degeneration is very critical **(Vissink et al., 2010; Pringle et al., 2013; Nelson et al., 2013; Holmberg and Hoffman, 2014)**

One of the main functions of salivary glands is providing the continuous flow of saliva into the oral cavity to keep the oral mucosa in a moist state so making it less susceptible to abrasion and to remove micro-organisms, desquamated epithelial cells, leucocytes and food debris during swallowing process. **(Dawes, 2012).**

Although saliva is about 99% water and 1% organic and inorganic molecules, a very important difference between them as two fluids is that saliva contains mucins “heavily glycosylated glycoproteins that are produced by the submandibular and