RESEARCH ARTICLE OPEN ACCESS

The Effect of X-Ray Radiation to IL-10 Levels in Secretome Mesenchymal Stem Cells Cosmeceutical Product

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Received 03 April 2023
Accepted 23 April 2023
Available online on 30 August 2023

ABSTRACT

Background: X-ray radiation has been widely used in the pharmaceutical industry because of its ability to regenerate and repair damaged tissues. **Objective:** In this study, we evaluate the effect of X-ray radiation on the secretome cosmeutical product. **Methods:** We conducted interleukin 10 (IL-10) analysis by ELISA in each product sample after exposure to X-ray radiation. **Results:** The levels of IL-10 in each sample were significantly lower than those in the control samples. Moreover, the level of IL-10 in the product samples was significantly higher than that of the control sample. **Conclusion:** In conclusion, exposure to radiation during shipping or storage of skin care products can potentially damage the proteins in the products by inducing the production of reactive oxygen species (ROS) and decreasing the treatment effectiveness.

Keywords : Secretome,IL-10, X-ray radiation, Cosmeceutical Product.

INTRODUCTION

The body's largest organ, the skin, acts as the body's first line of defense against a variety of diseases and outside factors that can increase a person's vulnerability to infection and inflammation. Skin issues of all kinds, from acute to chronic, are frequently discovered. They might affect the structure and function of the skin or cause different skin reactions. In chronic skin disorders such infection, inflammation, and ultraviolet (UV) exposure, it is difficult to restore the structure and function of the skin, so more effective therapy is needed [1-4]. Recently, secretome is a bioactive compound used as skin disorders treatment and can improve the condition of skin disorder. Therefore, maintaining the secretome level in cosmetic products is very necessary.

Stem cell-based therapies are still being developed today, particularly for the treatment of skin diseases. Mesenchymal stem cells (MSCs), in particular, can release trophic substances that promote tissue regeneration and repair tissue injury. A bioactive substance called secretome is secreted by MSCs in conditioned conditions [5,6]. It has many growth factors, cytokines, different macromolecules, extracellular vesicles, including microvesicles and exosomes, and these substances can stimulate a variety of biological processes, particularly in the modulation of the production of diverse new tissues. The secretome is one of the options for treating a variety of skin diseases that have been widely reported in numerous researches because of its capacity to regenerate and repair damaged tissues. As a result, we shall examine the application of secretome in the skin [7-9].

Many cytokines, including interleukin-10 (IL-10), interleukin-6 (IL-6), and interleukin-1 (IL-1), as well as growth factors, including transforming growth factor-beta (TGF-β), VEGF, platet-derived growth factor (PDGF), and hepatocyte growth factor (HGF) are found in secretome hypoxia mesenchymal stem cells (S-MSCs), which can prevent UVB hyperpigmentation by decreasing

melanin. It has been demonstrated that S-MSCs with soluble molecules, such as IL-10 and TGF- β, can lower ROS levels, limit the production of melanin, and soothe inflammatory conditions of the skin [10,11]. IL-10 mediates the anti-inflammatory response by controlling the the degree and duration of inflammation, and also regulating another cytokines and growth factors at homeostatics mechanism [12,13]. The benefits of delivering S-MSCs over other substances include the fact that S-MSCs are active biomolecules without immunogenicity or side effects, and their small molecular size makes it simple for them to pass through the skin barrier and into the dermis. S-MSCs have the potential to be used as an alternative treatment to stop skin darkening brought on by UVB exposure[14].

Product quality testing is necessary to determine the stability of secretome products against various physical treatments, including changes in temperature, humidity, and exposure to radiation in the form of X-rays[15]. After being produced, cosmetic products will be distributed to stores before reaching consumers. In the distribution process, it is unavoidable that skincare products go through an X-ray machine inspection process. We are aware that the soluble chemicals in the secretome, particularly the protein IL-10, are sensitive to radiation and changes in the environment.

The daily use of X-ray inspection in the pharmaceutical industry, however, is producing mounting proof that inspected medications continue to function as they should, without endangering patient safety. However, if pharmaceutical firms still have worries in this area, their X-ray supplier should work fast to allay them. X-ray inspection may possess several benefits and enables pharmaceutical firms to execute fill level checks, physical contamination checks, presence and integrity checks, and other tasks that would otherwise be challenging or time-consuming. The energy dosage levels used for these quality assurance procedures are within those that research and experience indicate are safe and have no detrimental effects on the product. Thus, by this study we will evaluate the effect of x-ray inspection on secretome cosmeutical product.

METHODS

Cosmeutical Product-containing Secretome Hypoxia-MSC Preparation

MSCs cultured in serum-free complete medium were incubated under hypoxia conditions in the hypoxic chamber maintaining a gas mixture composed of 5% O_2 and balanced N2 at 37 °C for 12 h. MSCs conditioned medium was then collected after 12-hour incubation. The collected MSCs conditioned medium was centrifuged at 2000 rpm for 5 minutes to remove cell debris and passed through a 0.22-μm filter membrane (Corning, NY, USA) to remove the remaining cell debris. The conditioned medium collected then filtered using tangential flow filtration (TFF) using 5 and 300 kDa filters. The S-HMSCs were kept at 2-8℃ temperature for next usage. 5% of secretome added in serum formulation and then packed in glass bottle.

Xray Scanning

X-ray Machine Reading In general, how the X-ray machine works is: the serum product to be examined enters the inspection tunnel (tunnel system) via a conveyor belt. Next, the serum will be inspected and will be detected by a number of light barriers and sensors that will send a signal to the control unit to activate X-rays. X-rays will penetrate goods on the conveyor belt with a fan-shaped light piece by piece as part of the inspection process for goods to be examined will absorb the light emitted by the generator (X-ray generator) and pass the X-rays to the detectors on the two sides of the tunnel. The image signal received by the detectors will then be collected part by part and will form an image on the monitor screen. The basic principle of X-rays is that they penetrate materials, so

different materials will absorb X-rays at different rates. Based on the absorption of X-rays, the colors that appear on the X-ray machine monitor screen are divided into 3, namely:

1. Orange Color This color indicates that the material is organic, such as clothing, leather, paper and others.

2. Green Color This color indicates that the material is inorganic, such as: aluminum, plastic, circuit board and others.

3. Dark Blue This color indicates that the item is metal, such as: iron, steel and others.

In this study, the object used for analysis was serum product stored in glass packaging, because the storage bottle is an inorganic material, based on the absorption of X-rays, the display that appears on the X-ray machine monitor screen will appear green. The display on the X-ray machine monitor is not clear, but if the officers can carry out operations carefully and are supported by expertise and technical knowledge related to the field of beauty products, of course the officers can predict the type of fishery product that appears on the X-ray machine monitor screen.

IL-10 Concentration Analysis

The concentration of IL-10 pre and post X-Ray irradiation was determined with Enzyme-Linked Immunoabsorbance Assay (Bioenzy Human IL-10 ELISA, BZ-08122010-EB) based on manufacture protocol at 450 nm wavelength.

RESULT

X-ray radiation decrease IL-10 level in S-MSCs cosmeceutical product

In this study, the results of IL-10 level in each product were vary after the treatment of X-ray. It is known that the highest IL-10 level was found in the serum brightening at 93,82 %, while the lowest IL-10 level was found in the hair serum at 49,92 %. Il-10 levels in each product samples were decreased dramatically after exposure to X-ray radiation. It is known that the highest decrease was in brightening serum products, which was around 48.12%, meanwhile the lowest decrease was in face cream products, which was 2.68%.

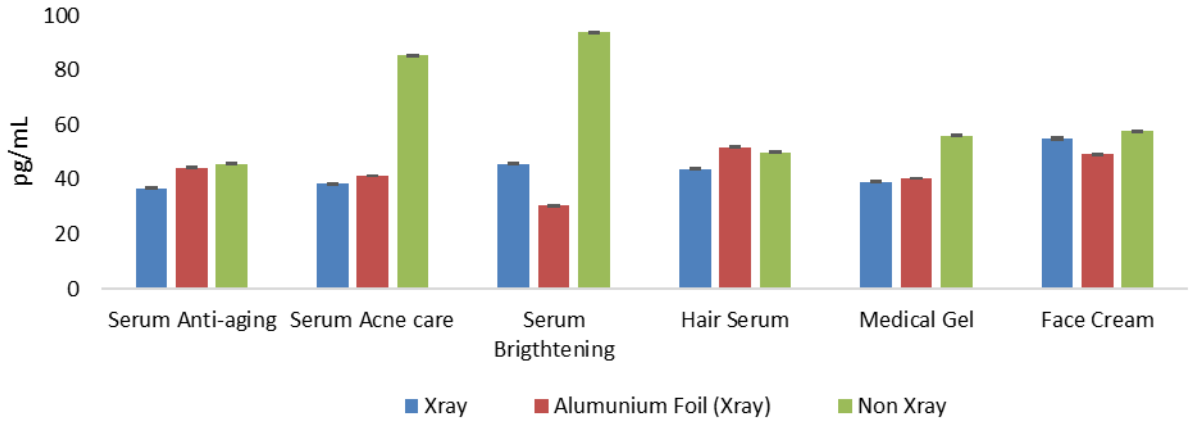

Figure. 1. Enzyme Linked Immunosorbent Assay of IL-10 in sample of the; (A) Anti-Aging Serum (B) Acne Care Serum, (C) Brightening Serum, (D) Hair Growth Serum, (E) Medicated Gel, (F) Face Cream**.** The data represent mean ± standard deviation.

DISCUSSION

Interleukin 10 (IL-10) is an anti-inflammatory cytokine that plays a key role in regulating immune responses. It is produced by various types of immune cells and has been shown to have protective effects in a range of diseases and conditions. However, exposure to ionizing radiation, such as X-rays, can lead to damage to molecules and cellular structures, including cytokines such as IL-10 [16-18]. Furthermore, X-ray radiation can also indirectly damage IL-10 molecules by affecting the production and activity of enzymes involved in their synthesis and degradation. Radiation-induced oxidative stress can lead to the production of reactive oxygen species (ROS), which can inhibit the activity of enzymes that help to synthesize and maintain the stability of IL-10 molecules. This can result in a decrease in the amount of IL-10 available in the sample and impair its ability to carry out its immune-regulatory functions [19,20].

The mechanism by which X-ray radiation can damage the structure of interleukin 10 (IL-10) protein involves the formation of reactive oxygen species (ROS), which are highly reactive molecules that can react with and modify proteins and other cellular structures. Radiation damage manifests as irreversible aggregation, unfolding, or fragmentation of protein. X-ray can directly break covalent bonds. Aqueous solvent in protein sample contributes as the source of radiation damage. When X-ray radiation penetrates a biological sample, it can generate ROS by ionizing water molecules and other cellular components, free hydroxyl (OH), hydroperoxyl (HO_2) radicals, and solvated electrons are produced from water photolysis [15,21], which then react with IL-10 molecules. The ROS can cause various modifications to the IL-10 protein structure, including the formation of cross-links, the introduction of oxidation products, and the breakage of peptide bonds [22,23]. The previous study held by Jeffries et al. showed that solution additives, such as DTT, ascorbate, and glycerol significantly reduce radiation damage of protein samples [21].

These modifications can impair the function of IL-10 by altering its structure and stability, affecting its ability to bind to its receptor and carry out its anti-inflammatory functions. For example, the formation of cross-links between IL-10 molecules can cause the protein to become insoluble and aggregate, which can prevent it from properly interacting with its receptor and impair its biological activity [24].

X-ray machines are not typically used in skin care product manufacturing, so there is generally no direct effect of X-ray radiation on the proteins in skin care products. However, exposure to X-ray radiation during shipping or storage of skin care products can potentially damage the proteins or other active ingredients in the product [25]. The effect of X-ray radiation on the proteins in skin care products depends on the dose and duration of exposure. X-ray radiation can cause damage to the proteins in skin care products by inducing the production of reactive oxygen species (ROS), similar to the mechanism by which radiation damages proteins in biological tissues. ROS can cause various modifications to the protein structure, such as the formation of cross-links, the introduction of oxidation products, and the breakage of peptide bonds. These modifications can impair the activity of the protein, affecting its efficacy and stability [26,27].

In addition, X-ray radiation can also cause changes in the pH, temperature, and other physical conditions of the skin care product, which can further affect the activity of the proteins in the product. To prevent damage to skin care products during shipping or storage, it is important to use appropriate packaging and storage conditions. For example, products should be stored in a cool, dry place away from direct sunlight, and packaging should be designed to protect the product from physical and environmental damage[28-30].

Potential damage to skin care products during distribution can be minimalized by:

- Choose the right packaging: Use packaging that is designed to protect the product from damage during travel. Consider using airtight containers or sealed bags to protect the product from exposure to air, moisture, and other environmental factors.
- Avoid excessive heat or cold: Exposure to extreme temperatures can damage skin care products and alter their texture, consistency, and efficacy. Avoid leaving products in hot cars or exposing them to freezing temperatures during travel.
- Keep products separate: Try to keep skin care products separate from other items in your luggage to avoid physical damage, such as being crushed or jostled.
- Consider a travel-sized version: If you are concerned about the safety of your full-size skin care products during travel, consider purchasing travel-sized versions of your favorite products. These smaller sizes are often designed to be more portable and less likely to break or leak during travel. In general, the risk of significant damage to skin care products during X-ray screening at airports is low. However, taking these steps can help to ensure that your products arrive at your destination intact and ready to use[30-32].

To minimize the risk of damage to skin care products during X-ray inspection, it is recommended to take the steps mentioned in the previous answer, such as using appropriate packaging and keeping products separate from other items in your luggage. If you are concerned about the safety of your skin care products, you may also consider contacting the manufacturer of the product for guidance on how to transport and store the product safely.

CONCLUSSION

In summary, X-ray radiation can potentially damage the proteins especially in this case IL-10 in skin care products during shipping or storage by inducing the production of ROS and causing changes in the physical conditions of the product. Proper packaging and storage conditions can help to minimize the risk of damage to skin care products during transport and storage.

ACKNOWLEDGEMENT

This work was supported by PT SCCR Indonesia – Semarang.

CONFLICT OF INTEREST

The authors have declared no conflict of interest.

AUTHOR CONTRIBUTION

ADA: writing-review and editing. SSG: writing original draft. SP: writing and editing. EH: performing ELISA assay. RA: sample preparation and data analysis.

FUNDING

None

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