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Measurement of *Malondialdehyde (MDA)* as a good Indicator of *Lipid Peroxidation*

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ABSTRACT

Redox homeostasis disruptions play a role in pathological conditions. Many compounds such as hydrogen peroxide are mediators and stimulate oxidative stress. Lipids as the main component of the cellular membrane play a role in maintaining cell structure integrity. The main target lipids of ROS attacks such as oxygen free radicals and lipid oxidation are associated with various pathological conditions. *Malondialdehyde (MDA)* is the result of lipid *peroxidation*, which is produced from damage to polyunsaturated fatty acids. MDA is a good indicator of lipid *peroxidation*.

INTRODUCTION

In the biological system there is a balance that is; *reducing*, oxidative and reducing reactions. Disruption of *redox* homeostasis results in either accumulation of *oxidizing* molecules due to over production or loss of cellular reduction ability. The accumulation of oxidizing agents can oxidize DNA, lipid proteins and which can change their structure, activity and physical properties [1].

ROS is a common form of oxidant in cells. ROS are formed from a partial reduction of oxygen molecules to *superoxide* ($O_2^{\cdot-}$), *hydrogen peroxide* (H_2O_2), hydroxyl ($\cdot OH$) [1]. Reactive oxygen species (ROS) are produced during the process of using oxygen (O_2), with a very reactive form. The existence of one or more electrons that are not paired in the outer orbit of the atom causes the

compound to be very reactive. *Stress Oxidative (OS)* is caused by an imbalance between pro-oxidants and antioxidants. This ratio can be changed due to increased levels of *reactive oxygen species (ROS)* [2, 3]. ROS can react with lipids, carbohydrates, proteins, nucleic acids, and macromolecules that can interfere with cell function. ROS reactions with lipids are known as *lipid peroxidation*. [4].

Essential component lipids are from the cell membrane that maintains cell structure and function. Lipids are the main target of ROS attacks such as oxygen free radicals and lipid oxidation is associated with various pathologies [5]. Lipids are classically divided into two groups: *polar* and *apolar*. Triglycerides (*apolar*), stored in various cells, but especially in tissues (fat), are usually a

form of energy storage in mammals. Polar lipids are a component of the cell membrane structure, participating in the formation of barrier cells and cellular organisms in the form of lipid liners. The main type of *lipid bilayer*, in almost all membranes is *phospho* glycerol-based lipids. The importance of lipid membranes can control the physiological state of membranes by modifying their bio-physical aspects, such as polarity and permeability. Lipids also have a key role in biology as molecular signaling [6].

Mechanism of autoxidation (peroxidation)

Lipids are an important component of transacting that maintains structure and control cell function. The main target lipids from ROS attacks such as oxygen free radicals and lipid oxidation are associated with various pathological conditions. Oxidation by free radicals in organic compounds has historically been referred to by organic and physical chemists as *autoxidation*. Lipids and biologically important compounds (proteins, carbohydrates, nucleic acids) undergo a reaction of oxidation by free radicals called *peroxidation*. *Peroxidation* is involved in a number of human diseases such as atherosclerosis, cancer, diabetes, recent diseases, as well as disorders and neurosurgery, Alzheimer's and Parkinson's. Lipid *peroxidation* is involved as a mediator of various pathologies including inflammation, cancer. In addition, lipid *peroxidation* acts as a regulator of non apoptotic cell death. [1,5]. MDA is the result of lipid *peroxidation*, which is produced from damage to polyunsaturated fatty acids. MDA is a good indicator of lipid *peroxidation* and as a biomarker of oxidative stress [7,8].

Fatty peroxy radicals are early PUFA *autoxidation* intermediates, and *fatty hydroperoxides* are the first primary oxidation products. *Fatty hydroperoxides*, labile species and easily enter radical reactions that lead to molecular transformation and decomposition. Although MDA is generally recognized as a volatile end product from heat, acid, or metal-catalysts, the composition of the *autoxidized* PUFA composition underlies the lipid transformation. One of the first mechanisms of MDA formation from PUFA containing three or more double bonds show that *fatty peroxyradicals* are produced during *autoxidation* and cyclize to *oxybridged* radicals. Subsequent decomposition of monocyclic peroxides through the *intramolecular*

process produces MDA (and other fragmentation products). Pryor et al. Formulated an alternative mechanism where *lipid peroxy radicals cyclize via oxybridged radicals, five-membered ring to allylic endoperoxide radicals*, which separate into prostaglandin-like *endoperoxides*. Intra-molecular process in the *endoperoxide* ring to produce MDA. (and *hydroperoxide*) [9].

Reduction of hydroperoxides, by bioactive agent reducing

The most important type of enzyme catalyst for *hydroperoxide* reduction so that hydroxide is formed is glutathione *peroxidase* (GPx). Lipid *hydroperoxides* are reduced in reactions involving *selenocysteine* residues GPx. As a result, lipid hydroxide and oxidized glutathione (9) are produced. Selenium is a micronutrient integral part of the enzyme glutathione *peroxidase* (GPX) [10]. GPX, an enzyme that depends on micronutrient selenium (Se), plays an important role in the reduction of hydroxide lipids. Compared to *hydroperoxides*, hydroxides have a significantly lower chemical reactivity character because they are considered stable and relative oxidation products are non-toxic [11].

Vitamin E is not an agent to reduce peroxides bonds, but as a radical scavenging agent. During the process of lipid peroxides, vitamin E acts as a donor of an intermediate electron *peroxy radical* (9). Cell plasma has non enzymatic scavenger free radicals such as ascorbic acid, *α-tocopherol* (vitamins C and E) [11].

Measuring malondialdehyde (MDA)

Malondialdehyde (MDA) is a product of lipid *peroxidation* that appears to be produced in a relatively constant proportion of the breakdown of polyunsaturated fatty acids. The MDA count is a good indicator of lipid *peroxidation* especially in vitro [11]. The chemical analysis of MDA began by measuring a component called *thiobarbituric acid-reactive substances* (TBARS) to estimate lipid *peroxidation* with *spectrophotometry*. Under acidic conditions (eq glacial acetic acid or sulfuric acid) and increasing temperature (eq 95°C), extended reaction time (eq 60 minutes), one MDA molecule reacts with two TBA molecules forming a red-colored, strongly visible light-absorbing (λ_{max} , 532 nm) and fluorescent (λ_{ex} , 515 nm); λ_{em} , 553

nm) derivate or condensation TBA-MDA adduct [12].

CONCLUSION

In this review, we summarize ROS can react with lipids, carbohydrates, proteins, nucleic acids, and macromolecules that can interfere with cell function. The main target lipids of ROS attacks such as oxygen free radicals and lipid oxidation are associated with various pathological conditions. The physiological and *pathophysiological* role of lipid peroxide, when lipids are the target of

oxidants, the lipid *peroxidation* process begins, a chain reaction produces MDA. Fatty *peroxyl* radicals are early PUFA *autoxidation* intermediates, and fatty *hydroperoxides* are the first primary oxidation products. Enzymes that reduce *hydroperoxide* to form hydroxides are glutathione *peroxidase* (GPx). Cell plasma has non enzymatic scavenger free radicals such as ascorbic acid, α -*tocopherol* (vitamins C and E). MDA analysis measures the components of *thiobarbituric* acid-reactive substances (TBARS) to estimate lipid *peroxidation* using *spectrophotometry*.

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