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ARTICULO DE INVESTIGACION

Interplay among oxidative stress, genetic and epigenetic events in human biological processes: A possible bases of medical ozone mechanism of action

Interacción entre el estrés oxidativo, los eventos genéticos y epigenéticos en los procesos biológicos humanos: posibles bases del mecanismo de acción del ozono médico.

Trabajo Presentado en el "Taller Ozone Frontier IV"

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ABSTRACT

Reactive oxygen species (ROS) can act as damaging molecules, but also represent central hubs in cellular signalling networks. Increasing evidence indicates that ROS increase perform an important role in the pathogenesis of diverse diseases degenerative and pathogen mediated although the underlying mechanisms and consequences of pathophysiology elevated ROS in most of them are still not completely resolved. The search was carried in the LILACS, PAHO, SciELO, EMBASE, PubMed and Infomed databases, by Google search engines and Google Scholar where the key words were placed and permit identification of original articles, theses, other articles of bibliographic review and high citation index journals published from 1950 to 2019, in Spanish or English. Two hundred thirty documents were identified, of which 58 were selected. The most important oxidants and reactive oxygen species produced by various physiological pathways, chemical and biological factors, including genetic and pathological conditions have been revised. More recently, alterations of the epigenetic landscape, which can affect DNA methylation, post-translational histone modifications, ATP-dependent alterations to chromatin and non-coding RNA transcripts, have been considered to be of increasing importance in the pathogenesis of most of diseases and aging. Different authors showed that epigenetic changes are highly dynamic and reversible. Thus, they might provide an important link between the actions of ROS, epigenetic and diseases. Ozonetherapy used by diverse application ways supports its pharmacological effect in different oxidative stress-associated diseases. Some authors recognized ozone interaction with diverse components in biological environment mediate formation of peroxides and aldehydes that activate different pathways as Nrf2 and NF-kB axes influencing also epigenetic signatures. As result, dose concept in ozone therapy and it hormetic response has a crucial role to manage the equilibrium inflammation / pro-inflammation responds. This review will provide an overview of the role of ROS in modulating the epigenetic landscape in the context of physiology but also link of redox state and disease and ozonetherapy biological effects.

Keywords: Platelet Rich Plasma; Inflammation; Ozone; HIV; Lipodystrophy.

RESUMEN

Las especies reactivas de oxígeno (ROS) pueden actuar como moléculas dañinas, pero también representan centros centrales en las redes de señalización celular. La evidencia creciente indica que el aumento de ROS desempeña un papel importante en la patogénesis de diversas enfermedades degenerativas y mediadas por

patógenos, aunque los mecanismos subyacentes y las consecuencias de la fisiopatología elevaron el ROS en la mayoría de ellos todavía no se resuelven por completo. La búsqueda se realizó en las bases de datos de LILACS, OPS, SciELO, EMBASE, PubMed e Infomed, por los motores de búsqueda de Google y Google Scholar, donde se colocaron las palabras clave y permitieron la identificación de artículos originales, tesis, otros artículos de revisión bibliográfica e índice de citas alto. Revistas publicadas de 1950 a 2019, en español o inglés. Se identificaron doscientos treinta documentos, de los cuales se seleccionaron 58. Se han revisado los oxidantes más importantes y las especies reactivas de oxígeno producidas por diversas vías fisiológicas, factores químicos y biológicos, incluidas las condiciones genéticas y patológicas. Más recientemente, las alteraciones del paisaje epigenético, que pueden afectar la metilación del ADN, las modificaciones de histonas postraduccionales, las alteraciones dependientes de ATP a la cromatina y las transcripciones de ARN no codificantes, se han considerado cada vez más importantes en la patogénesis de la mayoría de las enfermedades y envejecimiento. Diferentes autores mostraron que los cambios epigenéticos son altamente dinámicos y reversibles. Por lo tanto, podrían proporcionar un vínculo importante entre las acciones de ROS, epigenética y enfermedades. La ozonoterapia utilizada por diversas formas de aplicación respalda su efecto farmacológico en diferentes enfermedades asociadas al estrés oxidativo. Algunos autores reconocieron la interacción del ozono con diversos componentes en el medio biológico que median la formación de peróxidos y aldehídos que activan diferentes vías como los ejes Nrf2 y NF-kB que influyen también en las firmas epigenéticas. Como resultado, el concepto de dosis en la terapia de ozono y su respuesta hormética tiene un papel crucial para controlar el equilibrio que responde a la inflamación / proinflamación. Esta revisión proporcionará una visión general del papel de ROS en la modulación del paisaje epigenético en el contexto de la fisiología, pero también el vínculo entre el estado redox y la enfermedad y los efectos biológicos de la ozonoterapia.

Palabras clave: Plasma Rico en Plaquetas; Inflamación; Ozono; VIH; Lipodistrofia.

INTRODUCTION

In aerobic and anaerobic biological systems in general a countless number of oxidation-reduction (redox) processes take place in physiological conditions. Homeostasis of cellular redox state consist in maintaining the balance between formation and elimination of oxidant mediators and their products in the cell (Alfadda et al., 2012; Halliwell, 2007).

Oxygen is essential for life of organisms that use it. This element develops an important function as deposit of electrons during cellular respiration and in the synthesis of ATP molecules: adenosine triphosphate acid, essential molecule for the majority of cellular processes. More than 95% of the oxygen consumed by aerobic organisms is completely reduced to H2O during mitochondrial respiration and a small percentage (< 5 %) is converted to semi-reduced species known as reactive oxygen species (ROS) potentially damaging which can be or not of a radical nature, constituting the starting point of cellular oxidative injury (Halliwell et al., 2003; Jasmin et al., 2000).

Free radicals are chemical species capable to exist individually. They have one or more unpaired electrons in their most external orbital, thus enabling them to react by oxidation-reduction with a great number of molecules. Other enzymes in different cellular organelles also produce ROS such as xanthine oxidase, NADPH oxidase, cytochrome P450, and cyclooxygenase, among others (Folsome, 1989; Gospodaryov et al., 2012).

ROS are highly reactive having very short medium life cycle times. Despite this, in biologic systems, and as result of evolution, there is a complex system of biomolecules with antioxidant properties which interact with ROS, preventing formation of others, oxidation of structural and/or transient biomolecules or removing oxidation and recycling basic molecular building blocks. This system, including antioxidants of primary, secondary and tertiary character, is not completely efficient and, therefore, there is a basal level of reaction with biologic macromolecules like lipids, proteins, carbohydrates and nucleic acids, which can bring about a change of their structures and functions. olecules are oxidized with important

effects for the cell and tissue environment since, generally, they are related by signaling circuits to the modulation of processes such as transcription, apoptosis plus others (Alfadda & Sallam, 2012; Folsome, 1989; Halliwell, 2007). Though ROS are vital in the physiologic processes, at certain circumstances, increase in their generation and in the related oxidized products, it is associated themselves to the cause as well as the consequence of a number of pathologies. Oxidative stress (OS) is a cellular state in which homeostasis of oxidation-reduction mechanisms is altered, that is, the balance between pro-oxidants and antioxidants. This imbalance is produced at short- or long-term provoking a disruption of the signaling mechanisms and cellular control as consequence of improving the pro-oxidation processes and hindering the antioxidant mechanisms(Gitler et al., 2003; Halliwell, 2007).

Current scientific evidences show that OS is associated with many different diseases of high morbility/mortality or other chronic pathologies such as atherosclerosis and cancer, the two main causes of death in developing countries. Just the same is for complications of other pathologic conditions like arthritis, diabetes, nephropathy, viral infections and dementia, and the biologic process for aging, which accelerates in relation to the magnitude of OS (D'Autréaux et al., 2007; Harman, 1956; Liguori et al., 2018).

For modulating these processes, in order to benefit human health, various alternatives have been experimented in preclinical and clinical practices such as dietetic adequacy, antioxidant supplementation with vitamins or natural products and ozone therapy, among others. They all have action grounds in the bio-oxidative process with demonstrations of efficacy limited to research and/or application conditions. An update revision of these topics is the goal of the present review.

MATERIALS AND METHODS

Reactive oxygen species and oxidative stress in redox signaling in pathogenesis and physiopathology of a number of diseases

A great number of pathologies have been associated with OS. Among them are the two main causes of death in developing countries: atherosclerosis and cancer. Other pathologic conditions are also related such as cardiovascular diseases, cataracts, rheumatoid arthritis, diabetes, malaria, Parkinson´s disease, Alzheimer´s disease and other dementias, autoimmune diseases, chronic inflammation, among others. Biologic process of aging is accelerated according to the OS magnitude to which very different species organisms are subject to. Magnitude or extension of the oxidative damage increases while an organism ages and has been postulated as main cause of aging.

Results from a study carried out in seven countries indicated that at the level of the population, total serum cholesterol was closely linked to coronary heart disease incidence. Likewise, a solid correlation between ingestion of saturated fats and serum cholesterol was also observed indicating that variation the concentration of serum cholesterol in populations could greatly obey to differences in ingestion of saturated fats (Keys et al., 1984).

Damage by ROS and modulation of different processes implying cellular function alteration has been recognized to contribute to aging and is involved in pathogenesis of infectious and non-infectious diseases. Association to diseases is controverted for in many cases it is questioned whether OS is involved in the consequence or cause. But, undoubtedly, it is related to progression of symptomatology for diverse pathological conditions.

Current evidences of biomedical research and clinical practice in ozone therapy

Ozone, a gas mainly found in the atmosphere is originated in nature from oxygen and the energy generated by electric storms. This gas is precisely known due to its key role in the atmosphere as a filter of UV radiations. Its medical applications are more recent and are basically based on using its high oxidizing capacity coping with biomolecules while administered as a gas, ozonated water, hemotherapy or ozonated oils. In this way, in the biological media, a controlled stress is generated, which activates endogenous antioxidant responses. At least, so far, a receptor has not been described. Due to the aforementioned scientific proof of its biologic activity turns out to be difficult(WFOT, 2015).

The hypothesis that an oxidizing agent such as ozone can induce an antioxidant effect represented a great challenge for researchers in this issue. In 1998 was the first experimental work reporting elucidation of the called oxidative conditioning. Then, effects of O3 on neuromodulation were evaluated. It was found that this gas is able to inhibit the release of neuromediators by the effect probably related to modulation of cytosolic calcium concentrations the presynaptic level(Borrelli et al., 2011).

At present, ozone therapy (OOT) is applied to Infections, Inflammation, Surgery, pain, Ischemia, Vascular pathologies, Ophthalmic pathologies, Nervous system, Ulcers, Diabetes, and Infarction as well as others. OOT is a complementary medical practice of great use in patients who do not respond to conventional therapies or reject them(Aoron et al., 2015).

OOT also uses diverse routes of administration such as the intramuscular, sub-cutaneous, sauna, rectal insufflation, vaginal, among others, depending on the pathology and always following validated protocols.

In a particular way, OOT application in rheumatoid artritis, diabetes, retinitis pigmentosa and alcohol withdrawal has been evaluated and demonstrated its effectiveness even with protective effects in several organs (Díaz et al., 2012; León et al., 2016; Sánchez et al., 2005). As such, this method is practiced today in hospital infrastructures at several levels in Cuba (Gregorio Martínez, 2014). Ozone therapy´s efficacy in this case is equivalent to conventional therapies and, additionally, produces a significant drop in health expenses(Viebahn-Hänsler et al., 2016).

OOT is not a panacea, it has precise indications with which achieves a great therapeutic success. Other indications have led to a mid-success and others have not been useful. Precisely, one of the key recommendations of experimented researchers in preclinical research in different models and pathologies is to determine the cellular redox state toward recognizing the antioxidant capacity of the system and to adequate the doses to use for achieving and inductor effect of the endogenous antioxidant activity(Viebahn-Hänsler et al., 2016).

With this evidence and after the therapy, control of doses and the number of ozone therapy cycles for a specific patient comes to be more precise.

Doctor must respect all criteria related to good clinical practices recommended in OOT. This also refers to the staff, facilities, etc(Borrelli & Bocci, 2011).

It is necessary to precise that it is administered as mixture of oxygen-ozone, the latter between 0.21 µmol/mL (10µg/mL of gas per mL of blood) up to 1.68 µmol/mL (80 µg/mL of gas) which mean low doses according to the patient´s characteristics and with a number of

controlled treatments. It is considered a pro pharmaceutical since when acting with endogenous systems rapidly produces hydroperoxides, ozonides, aldehydes, alkenals and other reactive mediators which are the ones that produce the biological effect(Viebahn-Hänsler et al., 2016).

Its main effects demonstrated (Bocci et al., 2015; Borrelli & Bocci, 2011; Shanta et al., 2017)are:

-Regulating and restituting the antioxidant-pro-oxidant balance.

-Regulating the glucose levels in blood.

-Having a certain thrombolytic effect.

-Regulating the nitric oxide levels and maintains the vascular tone.

-Preserving the adenosine levels and the a1 receptors.

-Reducing inflammatory processes and pain.

-Having an antimicrobial action and accelerating healing.

One of the most affected topic by scientific specialization is the dose-response relationship many biomedical disciplines have shown great interest for the nature of this phenomenon in the zone with the lowest doses.

In the past, certain types of dose-response slopes were described as two-phase slopes, bell type curve, type U, type J, non-monotonic, bitonic, bimodal, bidirectional, Law of Arndt-Schulz, Hueppe rule, adaptive response, preconditioning, functional antagonism, hormologosis, hormesis, overcompensation, rebound effect, Law of Yerkes-Dodson, among others. Hormesis phenomenon was initially enunciated in the last century by two researchers, Arndt and Shulz. Hormesis (etymologically: to excite) explains the fact that many substances, in their interaction with living organisms can exert opposite effects: a stimulating effect at low doses and an inhibitor effect at high doses(Bocci & Borrelli, 2015; Bocci et al., 2011; Shanta & Amruta, 2017).

While graphic relationship between a conventional pharmaceutical and its biological effect can be represented as a sigmoid curve, the pharmacological response of ozone has the form of an inverted U. Response a conventional pharmaceutical, generally, can be explained by its interaction with receptors: at a higher dose the effect is greater until all receptors are occupied which is when the known maximum response is attained (Bocci et al., 2011).

In contrast with ozone mechanism, according current information available seems to be more complex for which its pharmacologic use is reduced to a very concrete interval of concentrations. Exposed biological systems respond in a compensatory way, repairing damages and reestablishing homeostasis. This phenomenon appears, just the same, in chemical interactions such as synergism and addiction (Bocci et al., 2011).

These findings indicate that hormesis is generalizable to the wide biological spectrum. Ozone therapy is related to these variations. For example, one of the mechanisms proposed to explain its effects by increase of concentrations of lipoperoxides is induction of the synthesis of antioxidant enzymes, among which is glutathione peroxidase. Synthesis of these enzymes

requires, among other things, media concentration of transition metals like Cu, Zn, Mn, Se, Fe, as well as others. Patients with nutrition deficiencies must wait a less biologic response(Shanta & Amruta, 2017).

Stimulation of production of 2,3 Diphosphoglycerate (DFG) by ozone is one of the mechanisms on which its tissue oxygenating actions are based. This stimulation in the production of DFG seems to depend on different factors such as the dose and therapy repetition(Bocci et al., 2011).

Ozone acts by different mechanisms of action. Its effects make the blood to flow more and modulate the immunologic system. Also key effects as germicidal, analgesic and anti-inflammatory actions have been demonstrated. Due to its effects on the vascular endothelium, allows more blood flow to tissues and thus, more oxygen. Also, its revitalizing effect on tissues and its capacity to remove toxins has been described. It is known that ozone´s effect occur due to two general mechanisms.

The first is related to its germicidal effects at high concentrations. A second mechanism, which occurs at low doses, is related to stimulation of endogenous responses and synthesis of mediators at long-term as described.

A great number of preclinical researches (model of Ischemia/reperfusion or hepatocellular injury with CCl4 in Sprague-Dawley or Winstar rats, respectively) have evidenced that during application of ozone 50 mg/mL by rectal route, the redox antioxidant indexes (superoxide dismutase activity, catalase activity, gluthatione concentration) were significantly modified in the group with ozone application showing beneficial effects as well as protective of the injury of the organ under study (Ajamieh et al., 2003; Ajamieh et al., 2002; León et al., 1998).

Other studies have compared the effects in models with rats of ozone application´s effects in post and preconditioning conditions with similar effects in stimulation of antioxidant protection and reduction of oxidative damage to lipids (Kadiiska et al., 2013). Findings back up the impact of a low oxidant concentration, stimulant of enzymes and antioxidant substrates with protection against damage.

Other researchers have conducted assays in humans employing OOT in diabetic foot and asthma with evaluation of redox state indices proving that the biologic grounds for ozone´s effect are in the redox hemodynamic system associated to reestablishing or cellular and tissue protection (Hernández et al., 2005; Sánchez et al., 2005).

Other studies identify the analgesic effect of OOT through adenosine and its receptors A1 and relation to the oxidant/pro-oxidant balance of system (Peralta et al., 2000).

Other broad arsenal of evidences have been obtained in the course of time with the use of ozonated oils, and ozonated water in preclinical research as well as in clinical practice that point out to the antioxidant induction mechanism through NrF2 pathways(Galiè et al., 2019).

It is interesting to highlight that there are reports about ozone generated *in vivo* in activated neutrophils. This discovery is of significant impact for demonstrates that this substance has a physiologic role, not only as bactericidal agent but otherwise it could form part of physiologic mechanisms of inflammation widening and activation of associated gene. Ozone is formed from singlet oxygen, a reaction probably catalyzed by antibodies (Wentworth et al., 2002).

The epigenetic machinery

Waddington first introduced the concept of epigenetics in 1939, "the actual interactions between genes and their products to phenotype into being" (Waddington, 1939). Then, Holliday, in 1987, redefined the term epigenetics as heritable changes in gene expression that are not due to alterations in the DNA sequence (Deans et al., 2015; Holliday, 1987). Therefore, these heritable changes, regulated by different systems include DNA methylation (DNAm), noncoding RNAs, histone modifications, and variants (Egger et al., 2004).

These mechanisms have been shown to be indispensable in the regulation of tissue gene expression, X-chromatin inactivation, and genomic imprinting. All of these modifications put together create "the epigenetic landscape," allowing the genome to display unique properties and distribution patterns in different cell types for its cellular identity (Sharma et al., 2010).

DNA methylation was the first epigenetic modification discovered, and it is the best and most mechanistically understood.

The enzymes that shape the DNA methylation patterns are the DNA methyltransferases (DNMTs), which are introduced onto the C5 position of cytosine residue, a methyl group (5mC) deriving from S-adenosylmethionine (SAM).In mammals, there are three types of enzymes, DNMT1,DNMT3a, and DNMT3b, which modify cytosine followed by a guanine residue, known as CpG dinucleotide. Even though DNA methylation is a stable epigenetic mark, it canbe removed as a consequence of passive or active demethylation processes. Passive loss of methylation can be achieved through successive cycles of DNA replication in the absence of functional enzymes, such as DNMT1/UHRF1 (Wu et al., 2014), down regulation of the DNMT enzymes (Oda et al., 2013), DNMT cytosolic localization (Jurkowska et al., 2011), and impairment of DNMT recruitment on DNA (Bostick et al., 2007). The active removal of 5mC has been shown to be through the formation of 5-hyroxymethylcytosine (5hmC), oxidized by the ten-eleven translocation (TET) enzymes. The oxidized products can then be processed directly by the TDG (thymine DNA glycosidase) generating a site that can be repaired by the BER machinery (Kohli et al., 2013) or deaminated by activation-induced cytidine (AID) deaminase deaminases, generating 5-hydroxymethyluracil (5hmU),which can also be excised by the TDG (Nabel et al., 2012).

Eukaryotic DNA is packaged into chromatin, consistingof nucleosome units wrapping 147 bp of DNA around an octamer of four core histones (H2A, H2B, H3, and H4).The DNA bridging of two adjacent nucleosomes is the linker histone H1, termed linker DNA. Historically, chromatin has been classified as either euchromatin or heterochromatin, according to its compaction state, even though there is a spectrum of chromatin states, suggesting it to be a highly flexible macromolecule. Chromatin structure can be modified by writer, reader, and eraser chromatin enzyme complexes that can remodel the nucleosomes or modify the histones through posttranslational modifications (histone acetylation, phosphorylation, glycosylation, ubiquitylation, and SUMOylation), establishing different chromatin transcriptional states (Ernst et al., 2011). Lastly, noncoding RNAs (ncRNAs) can exert their regulatory function by acting as epigenetic regulators of gene expression and chromatin remodeling. Detailed mechanisms are still at a very early stage, but they are known to recruit different histone-modifying enzymes that recognize (read), add (write), remove (erase), and replace chromatin modifications. A bona fide example is the long noncoding RNA (lncRNA), XIST, the X-inactive specific transcript, that coats one of the X chromosome by recruiting the polycomb repressive complex 2 (PRC2), triggering the heterochromatinization and transcriptional repression of the entire X chromosome (Lee et al.,

1999; Zhao et al., 2008). Therefore, understanding how the dynamics and the regulation of different epigenetic modifications are involved in the process of aging is of great interest and also in pathophysiology of diverse diseases.

Epigenetic and redox biology

Oxidative stress as a consequence of ROS accumulation; this phenomenon increases with age, and it is accompanied by a decline in the cell repair machinery, which will eventually cause a wide range of DNA lesions leading to mutations as well as a disruption in the epigenetic state ofthe cell. Herein are some examples of how this tight interconnection interplays between the effect of oxidative stress and the epigenetic landscape. For example, ROS can influence the methylome through the formation of oxidized DNA lesions formed by hydroxylation of pyrimidines and 5mC, which can interfere due to structural similarities with epigenetic signals related to 5-hmC (Lewandowska et al., 2011).

ROS also affects DNA demethylation by DNA oxidation and TET-mediated hydroxymethylation (Chia et al., 2011). ROS can indirectly modulate the activity of the epigenetic machinery since histone-modifying enzymes depend on intracellular levels of essential metabolites, such as Acetyl-CoA, Fe, ketoglutarate, NAD+, and S-adenosylmethionine, indicating that epigenetic changes are tightly linked to global cellular metabolism and energy levels of the cell (Simpson et al., 2012). Therefore, oxidative stress can globally influence the cell on multiple levels, from DNA and histones to histone modifiers, which will directly affect the epigenetic landscape of the cell. Epigenetics could help to explain the mechanisms non-dependent of gene sequence by which antioxidants or oxidants from surrounding biological environment contributes to modulate gene expression and also disease development. The main epigenetic mechanisms are covalent modifications of histones, DNA and non-coding RNAs, such as miRNAs.

These covalent modifications could include acetylation, hydroxylation and methylation (Cencioni et al., 2013).

ROS can influence the methylome through the formation of oxidized DNA lesions formed by hydroxylation of pyrimidines and 5mC, which can interfere, due to structural similarities, with epigenetic signals related to 5-hmC. ROS also affects DNA demethylation by DNA oxidation and TET-mediated hydroxymethylation. ROS can indirectly modulate the activity of the epigenetic machinery since histone-modifying enzymes depend on intracellular levels of essential metabolites such as Acetyl-CoA, Fe, ketoglutarate, NAD+, and S-adenosylmethionine, thus indicating that epigenetic changes are tightly linked to global cellular metabolism and energy levels of the cell (Arita et al., 2014).

Therefore, OS can globally influence the cell on multiple levels, from DNA and histones to histone modifiers, which will directly affect the epigenetic landscape of the cell (Guillaumet-Adkins et al., 2017).

ROS-induced abnormal DNA covalent pattern alterations are implied in different conditions such as malignant transformation and progression of numerous tumors. Also, it is important to consider the influence of OS and epigenetic signature in infertility and aging process (Cencioni et al., 2013).

The epigenetic changes induced by the environmental factors appear to be important in adulthood. One of the most interesting characteristics of the epigenetic signatures is that they may be partially reversible (Egger et al., 2004).

Based on a body of emerging evidences, it seems that epigenetic mechanism modulates gene expression concomitant to NF-kB activation, in many cases as consequence of the beneficial effects of low doses of phytochemicals and also related to bioxidative therapies including ozone therapy(Schladweiler et al., 2016).

They can also be inherited associated to some antioxidants enzymes polymorphism which could determine oxidative behavior persistence and regulate covalent modifications of histones and DNA (Guillaumet-Adkins et al., 2017).

Among the nutritional factors (one source of both antioxidant and oxidants) that have been linked to epigenetic modifications we found methyl donors, diets with excessive or deficient amount of proteins or calories, some fatty acids, minerals and vitamins, as well as plant compounds such as polyphenols, isothiocyanates, isoflavones and catechins. Also the use of different drugs could impact on oxidative tension and resound on epigenetic signatures (Bocci & Borrelli, 2015; Owona et al., 2019).

In short, the use of ozone in human therapeutics and its success depends on continuing deepening in the mechanisms of action, the effect in pathophysiological conditions, the routes of administration and demonstration of its molecular and biologic effect in correspondence with the designs of the studies employed.

CONCLUSIONS

Herein, we review how the interlinked effects of oxidative stress and epigenetic changes affect the diseases and also the process of aging. These findings open new horizons in the comprehension of the molecular basis of biological process related to the production of ROS and its effects on the epigenetic machinery. OOT applications in different clinical condition demonstrate induction of antioxidant system with amelioration of disease evolution and biomolecules protection of oxidation. Therefore, it will only be the complete understanding of this molecular process of event-associated diseases and ozone effect that will help us tackle comorbidities and influence on quality of life.

CONFLICTS OF INTEREST

The authors declare that there is no conflict of interest regarding the publication of this paper.

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