Medical Hypotheses 92 (2016) 3–6

Contents lists available at ScienceDirect

Medical Hypotheses

journal homepage: www.elsevier.com/locate/mehy

Possible role of antioxidants and nitric oxide inhibitors against carbon monoxide poisoning: Having a clear conscience because of their potential benefits

Sumeyya Akyol [a,*], Sevda Yuksel [b], Sultan Pehlivan [c], Haci Kemal Erdemli [d], Mehmet Akif Gulec [e], Bahattin Adam [f], Omer Akyol [g]

[a] Department of Medical Biology, Faculty of Medicine, Turgut Ozal University, Ankara, Turkey
[b] Department of Child Development, School of Health, Turgut Ozal University, Ankara, Turkey
[c] Ankara Regional Office of Council of Forensic Medicine, Morgue Department, Ankara, Turkey
[d] Department of Biochemistry Laboratory, Corum Training and Research Hospital, Corum, Turkey
[e] Department of Medical Biochemistry, Faculty of Medicine, Turgut Ozal University, Ankara, Turkey
[f] Department of Biochemistry & Molecular Medicine, University of California, Davis, California, United States
[g] Department of Medical Biochemistry, Faculty of Medicine, Hacettepe University, Ankara, Turkey

ARTICLE INFO

Article history:
Received 8 January 2016
Revised 19 March 2016
Accepted 8 April 2016

Keywords:
Carbon monoxide poisoning
Anoxia
Injury
Reactive oxygen species
Nitric oxide
Antioxidant therapy

ABSTRACT

Carbon monoxide poisoning is one of the important emergency situations manifested by primarily acute and chronic anoxic central nervous system (CNS) injuries and other organ damages. Current descriptions and therapeutic approaches have been focused on the anoxic pathophysiology. However, this point of view incompletely explains some of the outcomes and needs to be investigated extensively. Considering this, we propose that reactive oxygen species (ROS) including especially nitric oxide (NO) are likely to be a key concept to understand the emergency related to CO poisoning and to discover new therapeutic modalities in CO toxicity. If we consider the hypothesis that ROS is involved greatly in acute and chronic toxic effects of CO on CNS and some other vital organs such as heart, it follows that the antioxidant and anti-NO therapies might give the clinicians more opportunities to prevent deep CNS injury. In support of this, we review the subject in essence and summarize clinical and experimental studies that support a key role of ROS in the explanation of pathophysiology of CO toxicity as well as new treatment modalities after CO poisoning.

Background

The mechanism of brain injury after acute carbon monoxide (CO) intoxication has been investigated extensively in both *in vitro* and *in vivo* experimental studies as well as forensic cases [1]. Although pathological changes after CO poisoning are very well known in terms of gross and microscopical level for several organs including central nervous system (CNS), the pathophysiological changes at tissue level remain largely unknown [2]. Oxidative stress is accused to be one of the main determiners for CO-related neuronal injuries [3,4]. Late changes associated with forensic CO poisoning have been known to be comparable to post-ischemic reperfusion injuries [5]. Upon normobaric and/or hyperbaric oxygen therapy after CO-induced tissue hypoxia may be followed by reoxygenation injury in the CNS. Oxygen therapy might be resulted in increased production of reactive oxygen species (ROS), which in turn oxidizes proteins, lipids, and other cellular elements and components, whereby resulting in reperfusion injury as seen in any kind of ischemia/reperfusion (I/R) injuries [6]. A major source of ROS in biological systems is dioxygen (O_2), known as molecular oxygen. One of the most important enzymatic source of superoxide anion radical ($O_2^{\cdot-}$), is xanthine oxidase (XO). This enzyme catalyzes the conversion of hypoxanthine and xanthine to uric acid, the rate-limiting step in purine nucleotide catabolism. Current interest in XO is originated from its proposed role in any kind of I/R injuries. XO enzyme increases through the proteolytic conversion of xanthine dehydrogenase and produces enormous amount of $O_2^{\cdot-}$ [7].

Lately, a study [8] drew attention to the role of inducible nitric oxide synthase (iNOS) expression in CO-induced myocardial

* Corresponding author at: Department of Medical Biology, Turgut Ozal University Medical Faculty, Etlik, Ankara, Turkey.
E-mail address: sumeyyaak@gmail.com (S. Akyol).

http://dx.doi.org/10.1016/j.mehy.2016.04.015
0306-9877/

damage during I/R in rat heart. The concern related with local changes in NOS expression was noteworthy. It has been pointed out that CO exposure caused to increase in neuronal and iNOS expression, increase in cardiac NO production by iNOS, and decrease in myocardial hypersentisitivity to I/R by a specific inhibitor of iNOS in CO-exposed rat hearts. Overall, it has been revealed the implication of iNOS in the higher sensitivity of myocardial tissue to ischemic attack after prolonged daily exposure to CO at levels relevant to urban environment such as secondhand smoking or urban pollution. The major result was that iNOS mediates this damaging effect of chronic CO exposure via a NO/ROS-dependent pathway that can be avoided with a specific blocker. We would like to add novel aspects by introducing a major route through which NO could trigger/prevent I/R injury inducing by CO, which is not emphasized adequately up to now.

The hypothesis/theory

CO toxicity has been known to be connected with the proteins/mechanisms functioning oxygen carrier/user such as hemoglobin, myoglobin, cytochrome oxidase, and cytochrome P450 enzymes [9]. It is a poisonous gas, which is readily absorbed and transferred into the bloodstream, which in turn reaches, to the all the vital organs primarily CNS. Besides direct toxic effect to the brain mainly in the areas such as cerebral cortex, globus pallidus, caudate putamen, hippocampus, and striatum [10], CO results in some acute and chronic neuropsychological manifestations such as delirium, amnesia, urine and fecal incontinence, depression, anxiety, Parkinson-like symptoms, dyspnea, nausea, and weakness [11]. Other most affected organ after CNS is heart which might be manifested by premature ventricular contractions, atrial fibrillations, arrhythmias, heart blocks, ischemic influence, elevations in cardiac biomarkers, echocardiogram variations, and changes in electrocardiogram outputs [12].

The intracellular events and toxicological mechanisms after CO poisoning remains unclear. Cumulative evidences from *in vitro* and *in vivo* studies points out ROS as causative factors and secondary actors within cells from various organs [13]. We put forward the idea previously that ROS is the main element in CO-induced CNS injury leaving the therapeutic approaches not being extrapolated in details [5]. We hypothesized here that specific/selective/general antioxidant therapy modalities could be more convenient in acute and chronic treatment of CO poisoning cases to accelerate the healing process and/or to convert vital functions of organs faster in addition to the classical normo/hyper oxygen therapies.

Evidences supporting hypothesis

ROS, which include $O_2^{\cdot-}$, hydroxyl radical ($^{\cdot}OH$), hydrogen peroxide (H_2O_2), singlet oxygen ($^1O_2$), and NO, may lead to cellular injury when they are generated extremely or when the antioxidant defense systems are disrupted. The roles of different types of NOS on I/R are very different. A sudden burst of NO, which is typical of iNOS activity during reperfusion, may promote cell damage via cellular lipid peroxidation [14] (Fig. 1). Superoxide dismutase (SOD) is a potent protective enzyme that can selectively scavenge $O_2^{\cdot-}$ by catalyzing its dismutation to H_2O_2 and O_2. Other antioxidant enzymes, glutathione peroxidase (GSH-Px) and catalase (CAT) catalyze the conversion of H_2O_2 to water by using different pathways including reduced glutathione (GSH) and reduced nicotinamide adenine dinucleotide phosphate (NADPH) as cofactors. A balance between the production of ROS and the activities of antioxidant enzymes as well as the level of antioxidants is essential for the normal functioning of the CNS, heart and other organs [15]. Some of these enzymes have been applied in different I/R injuries of animals as remedial agents [16–18].

NO production has been suggested to have both detrimental and beneficial effects after I/R injury [19]. NO could react with $O_2^{\cdot-}$ to produce a potentially harmful oxidant, peroxynitrite [20] (Fig. 1). In the light of the recent publications, it has been shown that NO may have additive effect on the myocardial injury. It then aggravates and triggers further damaging events. On the other hand, NO could inactivate pro-inflammatory inducible enzymes [21] that may induce some forms of heart injury. On these grounds, it is intriguing to wonder about the exact role and distribution of NOS in rat heart and also in human heart after the realization of experimental findings to the projection of human heart. NO plays a major role in controlling some of the cellular and organ functions in the human body [22]. It is the only endogenous molecule that functions as a hormone, ROS, neurotransmitter, constitutive mediator, inducible mediator, cytoprotective molecule and cytotoxic molecule. Current data have revealed the existence of possible associations between NO-derived cardiotoxicity and chronic CO exposure. Basic mechanism of CO toxicity has been classified as hemoglobin, myoglobin, cytochrome oxidase, and cytochrome P450-dependent mechanisms. All of them can be explained with tissue hypoxia. In addition to that, oxidative stress has been attributed to play a certain role in CO toxicity, which we reviewed in depth [5]. NO itself was found to be increased 9-fold immediately after CO poisoning [23]. Conversely, Qu et al. found that NO level in blood was reduced after severe and moderate CO poisoning compared to control group [24]. From this point of view, they suggested to use low NO concentration inhalation in treatment of acute CO poisoning. Obviously, it can be alleged that mitochondrial dysfunction, oxidative stress including NO increase, and lipid peroxidation are all expected after CO poisoning as a vicious cycle.

Consequences of the hypothesis and discussion

Thus, we believe that supplementation of nonenzymatic antioxidants, including vitamin E, C, and beta-carotenes plus one of anti-NO agents in addition to the common CO toxicity therapy may protect membranous structures from lipid peroxidation by ROS and NO species (Fig. 1). Other alternatives are iron chelator deferoxamine, Mn(III) tetrakis (4-benzoic acid) porphyrin (MnTBAP), glutathione, acetyl cysteine, tempol, H_2S [25], N-acetyl cysteine, melatonin, seleno-methionine, and allopurinol [5]. Our hypothesis is that caffeic acid phenethyl ester (CAPE) (Fig. 2) may be a promising, specific and effective treatment option of acute CO poisoning. CAPE has been isolated and characterized several decades ago by Grunberger et al. [26] but there is no data on the toxicity *in vivo* and no clinical usage up to now. This novel hypothesis is based on the following facts: CAPE completely blocks production of ROS in human neutrophils and in xanthine/XO system at 10 μM concentration showing its competent antioxidant capacity [27]. It is a potent inhibitor of NFκB and has lipophilic chemical characteristics allowing it to reach CNS easily [28]. Although future researches are needed to look into possible therapeutic strategies to improve the functioning of brain and heart in hypoxic conditions after CO poisoning, experimental studies so far have reported beneficial effects of CAPE in ameliorating the functioning of brain and heart in hypoxic conditions and oxidative stress-related damages [29–32] after hyperbaric oxygen therapy. CAPE has been proposed to prevent renal I/R injury via NO metabolism. Pretreatment of the rats with CAPE before renal I/R diminished tissue NO levels in some extent but it was not statistically significant [33].

The effect of CO poisoning on the NO system in the striatum of free-moving rats by means of *in vivo* brain microdialysis was investigated [34]. It was found in this study that CO poisoning