ANTIOXIDANT PROPERTIES OF SOME NANOPARTICLE MAY ENHANCE WOUND HEALING IN T2DM PATIENT

Ghulam Mohammad1*, Vijendra K.Mishra2, H.P.Pandey1
1Department of Biochemistry, 2. School of Biotechnology, Banaras Hindu University Varanasi
221 005 INDIA

The delayed wound healing is a well known problem with Type 2 diabetic patient (T2DM). Understanding wound healing today involves much more than simply stating that there are three phases: “inflammation, proliferation, and maturation.” Wound healing is a complex series of reactions and interactions among cells and “mediators.” Each year, new mediators are discovered and our understanding of inflammatory mediators and cellular interactions grows. However, the lack of understanding of the molecular mechanisms and pathogenesis of impaired healing in chronic ulcer is a serious health care problem that contributes to excessive limb amputation and mortality. Oxidative stress is a major contributes for delayed wound healing and produce tremendous amount of ROS this is very harmful for surrounding tissue or cells. Recently, it was shown in some literature that few metal oxide nanoparticle like Al₂O₃, CeO₂, Y₂O₃, act as a ROS scavenger. The essentials of nanoparticle in ROS scavenging system have been presented in this article, which has portrayed current thinking that speculates the use of nanoparticle is another likely to cure of impaired healing of diabetic foot wounds. Thus nanoparticle mediated diabetic wound healing could also be clinically significant.

(Received July 30, 2008; accepted August 4, 2008)

Keywords: Y₂O₃; Al₂O₃; Free Radicals; ROS; Antioxidant; Apoptosis; Diabetic wound.

1. T2DM wound healing

Delayed wound healing is a well known problem with T2DM patients. Wound healing is a complex programmed sequence of cellular and molecular processes, including inflammation, cell migration, angiogenesis, provisional matrix synthesis, collagen deposition, and re-epithelization [1]. The healing process requires a sophisticated interaction among inflammatory cells, biochemical mediators, extracellular matrix molecules and micro environmental cell population. All of these events are stimulated by a number of mitogens and chemotactic factors. Now a day wound of T2DM patient is most common disease and still challenging its complete cure. Cell death, as a comprehensive consequence of non healing wound abnormalities, Healing impairment is characterized by delayed cellular infiltration and granulation tissue formation, reduced angiogenesis, decreased collagen, and its organization [2-5]. The mechanism of this alteration is thought to result from production of high level of reactive oxygen species and increased level of apoptosis, which in turn impairs keratinocyte endothelial cells, fibroblasts, and collagen metabolism [6].

2. Reactive oxygen species (ROS)

Reactive oxygen species include hydroxyl radical (OH⁻), superoxide radical (O₂⁻) and hydrogen peroxide (H₂O₂). ROS, particularly OH⁻, interact with lipids, proteins and nucleic acids resulting in loss of membrane integrity, structural or functional changes in protein and genetic mutations, respectively [7]. To neutralize, the toxic effect of ROS, the body utilizes several antioxidant defense systems, including both enzymatic such as superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase, GSH-Px and

* Corresponding author: gmkbiochembhu@gmail.com
glutathione-S-transferase (GST) and nonenzymatic such as glutathione, vitamin E components [8, 9]. Moreover, mitochondria are a rich source of generating $O_2^-$ is converted into $H_2O_2$ by SOD. $H_2O_2$, in the presence of $O_2$ and iron forms OH group, a more reactive form, which is further converted into lipid peroxide. Furthermore, catalase is present in peroxisomes in eukaryotic cells and it can transform $H_2O_2$ into $H_2O$ and $O_2$. On the other hand GSH-Px reduces the lipidic and non-lipidic hydroperoxides as well as $H_2O_2$ while oxidizing glutathione [8, 10]. GSH is also a potential co-oxidation substrate of COX-2 that can be oxidized in one electron fashion to GS which in turn, reacts with another GSH molecule to form GSSG, a reducing radical readily and effectively donating electron to molecular oxygen to generate superoxide [11]. GST catalyzes the conjugation of glutathione with toxic metabolites and xenobiatics compounds, which subsequently results in detoxification of toxic metabolites [12]. The process of lipid peroxidation involves oxidative conversion of polyunsaturated fatty acids into malondialdehyde (MDA), which is a marker of lipid peroxidation and is usually measured as thiobarbituric acid reactive substances (TBARS) or lipid peroxides [8]. COX and PGE2 have shown to be more important in the early stages of wound healing by promoting fibrosis and decreasing the accumulation of macrophages within the wound site [13]. PGE2 has been reported to be involved in proliferation of fibroblasts and the promotion of collagen synthesis at earlier time points during wound healing [14].

3. Apoptosis and reactive oxygen species

Mitochondria play an important role in apoptosis under a variety of proapoptotic conditions, such as oxidative stress [15]. Oxidative stress is believed to play an important role in the development of diabetic complications [16]. ROS may be involved in all stages of the wound healing process such as migration, adhesion, proliferation, neovascularization, remodeling, and apoptosis are main processes in wound healing regulated, or at least modulated, by ROS. Due to the underlying signaling and damage pathways, oxidative stress could result in disturbed wound healing. Enhanced ROS concentrations in chronic wounds are thought to drive a deleterious sequence of events eventually resulting in the nonhealing state. In chronic wounds, there are numerous sources of ROS. Prolonged inflammation with neutrophiles migration into the damaged tissue generating superoxide anion radicals in the oxidative burst reaction, hypoxia, and ischemia reperfusion are important mechanisms resulting in oxidative stress. Disruption of the redox balance and alterations to the level or activity of reducing enzymes that help to maintain the normal redox state could also contribute to poor healing and may even lead to damage to DNA within cells involved in the normal healing process. Consequently, increased oxidative stress, together with elevated levels of ROS, could result in damage and strand breakage of cellular DNA [17-19], in such cells in diabetes, as has been found to be the case [20-23]. Increased alteration and damage to DNA within the cells cause to change in membrane potential of mitochondria that leads to mitochondrial cytochrome c release is a key event in the activation of caspase-3, a downstream pivotal step to initiate apoptosis [24]. A correlation between ROS generation and the pathogenesis of various diabetic complications has been demonstrated [25].

4. Nanoparticles as ROS Scavenger

Major problem with T2DM patients is delayed healing of wound. Several literatures show this is because of increase in oxidative stress and decreased antioxidants level content leads to increased inflammation, which results huge amount of ROS production that leads to premature apoptosis of inflammatory cells. Recently, nanotechnology and nanoparticle field is a fast-growing research area. Research has already led to significant breakthrough and several products are available commercially. Recently, role of silver nanoparticle in diabetic wound healing therapy has also been considered. However, the complete mechanisms of silver nanoparticle in bacteriostatic phenomena in wound healing remain unknown. It has been reported a novel Ag+-loaded zirconium phosphate nanoparticle plays crucial role in diabetic wound healing [26]. Some of the nanoparticle now a day are preparing in a manner that act as free radical scavenger [27]. Moreover, reports reveals Cerium oxide (CeO₂, ceria) plays major active role because of its excellent free radical scavenging potentials [28]. This metal oxide is monodisperse particles with single crystals and few twin boundaries [29] as well as expanded lattice parameter [30]. Moreover, CeO₂ tends to be a nonstoichiometric compound. Cerium atom characterized by both +4 and +3 oxidation states. Literatures shows X-ray photoelectron spectroscopy and X-ray absorption near edge spectroscopy suggests that the concentration of Ce³⁺ relative to Ce⁴⁺ increases as
particle size decreases with a conservative $[\text{Ce}^{3+}]$ minimum of 6% in 6 nm nanoparticles and 1% in 10 nm particles [31]. This dual oxidation state means that these nanoparticles have oxygen vacancies [32]. The loss of oxygen and the reduction of $\text{Ce}^{4+}$ to $\text{Ce}^{3+}$ is accompanied by creation of an oxygen vacancy. This property is responsible for the interesting redox chemistry exhibited by $\text{CeO}_2$ nanoparticles and makes them attractive for wound healing process followed by scavenging properties.

Nanoparticles made of other metal oxides were also considered for its potential scavenger behavior. These included particles aluminum oxide ($\text{Al}_2\text{O}_3$), commonly known as alumina. That differs in crystal structure and yttrium oxide ($\text{Y}_2\text{O}_3$, yttria). Moreover, Alumina is also thermodynamically stable at all temperatures and has a corundum like structures with oxygen atoms adopting hexagonal close-packing and $\text{Al}^{3+}$ ions, filling 2/3 of the octahedral sites in the lattice. However, yttria has a cubical structure [33]. Yttrium oxide is now a day considered most significant due to its highest free energy of oxide formation from elemental yttrium among known metal oxides [34]. It is characterized by only small changes from stoichiometry under normal conditions of temperature and pressure [35] and by atmospheric absorption of $\text{H}_2\text{O}$ and $\text{CO}_2$ [36]. The particular polymorph of it is the monoclinic B form, which is closely related to its A form having hexagonal close-packing (hcp). Unlike the A form which has only sevenfold coordination states, it has six fold [35]. The B form structure is slightly less dense than the A form variant and has the yttrium cations in nonequivalent sites in the crystal [37]. These groups of nanoparticles are relatively nontoxic to neutrophiles and macrophages, and that $\text{CeO}_2$ and $\text{Y}_2\text{O}_3$ particles protect cells from death due to oxidative stress. This protection is due to the direct antioxidant properties of the nanoparticles.

Cerium and yttrium oxide nanoparticles are able to rescue cells from oxidative stress-induced cell death in a manner that appears to be dependent upon the structure of the particle but independent of its size within the range of 6–1000 nm. This might be useful for the diabetic wound healing. Furthermore, three alternative explanations were demonstrated that the cerium oxide and yttrium oxide particles protect from oxidative stress. They may act as direct antioxidants, block ROS production, which inhibit programmed cell death pathway and they may directly cause a low level of ROS production, which rapidly induces a ROS defense system before the glutamate-induced cell death program is complete. The latter is a form of preconditioning that could be caused by the exposure of cells to particulate material known to induce low levels of ROS [38].

Therefore it could be suggested that $\text{CeO}_2$, alumina and yttrium may be useful for diabetic wound healing mediated by ROS scavenging potentials of above nanoparticles and have diabetic wound healing therapy.

### 5. Conclusions

The aim of this article was to review the process of nanoparticle with a view toward examining it in the context of diabetic wound healing. These groups of metal oxide nanoparticle are relatively nontoxic to neutrophiles and macrophages and that $\text{CeO}_2$ and $\text{Y}_2\text{O}_3$ particles protect cells from death due to oxidative stress. This protection is due to the direct antioxidant properties of the nanoparticle. The essentials of nanoparticle in ROS scavenging system have been presented in this article, which has portrayed current thinking that speculates that use of nanoparticle as antioxidant is another likely to cure of impaired healing of diabetic foot wounds.

### References