HISTOLOGICAL STUDY ON THE BENEFICIAL EFFECT OF GLUTATHIONE FOR CENTRAL NERVES SYSTEM FROM DAMAGE INDUCED BY D-GALACTOSE

Sadiq Safaa Mahdi¹, Wifaq Albazi², Muna Hussain Al-Aameli³

¹ Department of Physiology & Pharmacology / College of Veterinary Medicine, University of Kerbala, Iraq.
² Department of Physiology & Pharmacology / College of Veterinary Medicine, University of Kerbala, Iraq.
³ Department of anatomy & histology / College of Veterinary Medicine, University of Kerbala, Iraq.

ABSTRACT

Twenty-four female white albino rats were used in this experiment. Animals were divided to four groups each group contain six (6) rats. They were treated daily for six weeks with intraperitoneal injections of D-Galactose, glutathione (GSH), D-Galactose and GSH. The first group (G1) was intraperitoneal injected only normal saline about the same volume that use in the other groups and it was set as control group. While 100mg/kg b.w of D-galactose was injected the second group (G2) and 100mg/kg b.w of glutathione was injected the third group (G3). Whilst the fourth group (G4) was injected with both D-galactose and glutathione both with (100mg/kg b.w). Through the current study, the results of the cerebrum and cerebellum sections stained with Congo red were revealed accumulation of beta amyloid in these tissues which appeared red area in the beta amyloid site accumulation of rats group (G2) which supplementation D-gal and it was same area less in the combination rats group (G4) which are treated with (GSH and D-gal).

Keyword: D-galactose, Glutathione, beta amyloid, inflammation, Congo red

I. INTRODUCTION

D-galactose is a monosaccharide sugar with the same chemical formula as glucose (C6H12O6). It has a structure that is very similar to glucose, with the exception of one hydroxyl group that is at a different location. {1}. D-galactose delivery to animals can cause elements of brain aging that are analogous to human brain aging in many respects. {2}. Memory loss, neuronal degeneration and apoptosis, increased oxidative stress, reduced ATP production, increased mitochondrial DNA mutation, poor mitochondrial structure, and aberrant gene expression in the brain are only some of the symptoms. {3,4,5}. Glutathione is a tripeptide consisting of -L-glutamyl-L-cysteinylglycine that is found in all mammalian organs at values of 1–10 mM. {6}. GSH is an important regulator of redox signaling, plays a role in xenobiotic detoxification, and influences cell proliferation, apoptosis, immunological function, and fibrogenesis. {7}. Because amyloid plaques are a hallmark of Alzheimer's disease, researchers are looking at medicines that inhibit beta-amyloid aggregation as a possible therapy. According to some studies, the toxic effects of beta-amyloid begin before plaques and oligomers develop, therefore researchers have been looking for strategies to avoid the harmful interactions between beta-amyloid and nerve cells before plaques and oligomers form. {8}. The brain's detoxification of reactive oxygen species is crucial, and the antioxidant glutathione plays a significant role. A role in these activities. The availability of glutathione precursors has a big impact on the glutathione concentration of brain cells. Different extracellular glutathione precursors are preferred by different kinds of brain cells. {9}.

II. MATERIALS AND PROCEDURES.

Twenty-four (24) female white albino rat were used in this experiment and their ages between (12-14) weeks and they were (180-210) gram b.w and the animals were placed in good condition in special plastic cages and provided the animals with the appropriate conditions. In terms of temperature around (30 ±5 C°) and ventilation and the light system was 12 hrs per day. The animals were divided to four groups each group contain 6 rats and they were treated daily for six weeks with intraperitoneal injections of D-galactose, GSH, D-galactose and GSH.
The first group (G1) was injected intraperitoneally with only normal saline about the same volume that use in the other groups and it was set as control group. The second group (G2) was injected intraperitoneally of D-galactose (100mg/kg b.w) \cite{10}. The third group (G3) was injected intraperitoneally with glutathione (100mg/kg b.w)  \cite{11}. The fourth group (G4) was injected with both D-galactose and glutathione both with (100mg/kg b.w).

Tissue samples (cerebrum and cerebellum) were applied with 10% formalin solution for 2 days. The samples then dehydrated in ascending grades of alcohol and used xylene to clearing, and used paraffin wax to bury. The blocks were carefully oriented to have the cross-sections to be cut (6) \( \mu \)m thickness serial sections. The sections were deparaffinized and hydrated for special stain Congo-red staining.

### III. RESULTS

Results of stained sections with Congo red stain taken from the central nerves system (cerebrum and cerebellum) of D-gal (G2) group and the combine group (G4) showed accumulation of beta amyloid plaques as red area in the tissue of the central nerves system but in group (G4) they were less in number and smaller in size, while they were absent in both control group (G1) and in the GSH group (G3).

![Image of cross section in Rat cerebrum of D-gal (100mg/kg) group (G2) showing accumulation of beta amyloid plaque in the tissue (X100).](image)

Figure 1: Cross section in Rat cerebrum of D-gal (100mg/kg) group (G2) show accumulation of beta amyloid plaque in the tissue (X100).
Figure 2: Cross section in Rat cerebrum of the combination group (D-gal+GSH) (G4) show accumulation of beta amyloid plaque in the tissue (X100)
Figure 3: Cross section in Rat cerebellum of the D-gal group (G2) stain with Congo red stain show accumulation of beta amyloid plaque in the tissue (X100).

Figure 4: Cross section in Rat cerebellum of the combination group (D-gal+GSH) (G4) stain with Congo red stain show accumulation of beta amyloid plaque in the tissue (X100).

Table (1) The protective role of GSH on the beta amyloid concentration in serum.
The capital letters in the vertical direction indicate significant differences (P <0.01).

The Table showed that use of glutathione as antioxidant reduced the constriction of beta amyloid in the body when compared to the group that was injected only with D-galactose at level of (p-value <0.01), The main value of beta amyloid was (12.7±3.33, 26.78±4.47, 8.55±1.37, 22.31±1.95) for groups Control, d-gal GSH and D-gal+GSH respectively.

### IV. DISCUSSIONS

Our outcomes study showed that there is a decrease in the beta amyloid accumulation in the treated group when compared to the group of d-galactose and the size of beta amyloid plaques was also decrease.

The beta-amyloid precursor protein is found on many types of cell membranes, and this precursor protein generally releases -amyloids at a high pace into the plasma and cerebrospinal fluid. Internalized β-amyloids fold into β -folded or β -pleated shape and stack on each other to create mass of fibrils and aggregates known as plaques. {12}

Because Congo red adheres to amyloid fibrils in several proteins, it can be used as a diagnostic test for amyloid. And when it bind it will show as red color {13}

D-galactose is a reducing monosaccharide found in tiny amounts in all living cells. Increased generation of advanced glycation end products and reactive oxygen species have been linked to oxidative damage in a variety of tissues and organs after an excessive dosage of D-galactose is given. {14}

The over dose of D-galactose case oxidative stress on CNS and substantial evidence that oxidative damage to the brain is an early event in Alzheimer's disease. {15}

The oxidized to reduced glutathione ratio is used to determine the severity of the effects of oxidative stress, Antioxidants have long been touted as a way to delay the course of Alzheimer's disease. {16}

In this current study there is significant increase in beta amyloid in d-gal group because that d-gal is increase the oxidative stress of nerve cell and lead to increase in beta amyloid{17} Extracellular senile plaques and the accumulation of peptides present intracellularly in the brain, such as the protein tau consisting largely of amylooidal material are used to diagnose Alzheimer’s disease.. GSH levels drop as people get older.. Which rise to an increment in the production of amyloid peptides. {18}

### V. CONCLUSION

At the conclusion of the study, it was found that glutathione has decrease the harmful effect of D-gal by decrease Beta-amyloid plaques accumulation.

**REFERENCES**