Sodium Nitrite Effects on Some Blood and Biochemical Parameters in Glutathione Treated Male Rats

Huda K. Khassaf
Department of physiology, pharmacology and biochemistry; College of Veterinary Medicine; University of Basrah, Basrah, Iraq.

Corresponding author E-mail: Huda.khassaf@uobasrah.edu.iq

Orcid ID: https://orcid.org/my-orcid?orcid=0000-0002-8251-2776

Abstract

In industrial food production, food preservatives like sodium nitrite are used more frequently. The use of some food preservatives has been linked to teratogenic and carcinogenic effects. In order to investigate the effects of food additives and natural antioxidant substances on rats forty adult male albino rats, approximately one month and a half were divided into four groups as follows, Group 1 serves as control group supplement with (Nacl 0.9% normal saline).Group 2 received only sodium nitrite supplement (30 mg/kg BW), group 3 supplemented initially with sodium nitrite and glutathione and group 4 supplemented with glutathione alone (6.42 mg/kg BW). A variety of parameters were measured, including the number of red and white blood cells (RBCs & WBCs), hemoglobin (Hb) level, hematocrit (Hct) value, glucose level, and serum alanine transaminase and aspartate transaminase (ALT & AST) activity analysis. The results clearly showed that (RBCs), (Hb) concentration, and (Hct) value of rats treated with sodium nitrite for two months exhibit significant decrease when compared to the control and treated groups, rats given with either glutathione alone or glutathione plus sodium nitrite showed significant decrease in their serum glucose levels and there was discernible elevation in the serum activity of AST and ALT in rats treated with glutathione alone. Rats treated with sodium nitrite showed a substantial decrease ($P< 0.05$) in AST activity after 2months when compared with the control group, whereas significant increase ($P< 0.05$) in ALT activity were detected after 2months . These results indicate the harmful effects of using artificial supplements and colorings and also prove the improving effect of glutathione. Due to the potentially harmful effects of food additives, it is advised that the use of S. N. as an additive be kept to a minimum.

Key Words: Sodium nitrite, Food additive, Glutathione, Rats, carcinogenic.

Introduction:

Elements that are purposefully added to food as food additives either manufactured or natural, one of these main preservatives are nitrites, which can be found as free acids or salts. Sodium nitrite is frequently used as a preservative in cooked meat products like sausages (1). The proportion of nitrite content daily food consumption may be higher than the allowable amount due to the usage of multiple types of such foods (2). As food additives for humans, nitrites are mostly employed to produce a particular flavor and preserve meat products. Many organic nitrites and nitrates have been employed in clinical settings, but sodium nitrite is the only inorganic nitrite that has been shown to be therapeutic (3).

Environmental contaminants found in food and water such as nitrites and nitrates may play a role in the development of liver and renal disorders as well as immune system issues in domestic poultry (4) to prevent oxidative damage, glutathione (GSH) may function as a free radical acceptor. Leichtweis and Ji showed oral GSH administration raises plasma GSH levels and that dietary GSH can be absorbed intact from the intestinal lumen (5). Because GSH is a substrate for glutathione peroxidase, it helps shield cells from harmful substances and oxidative stress (3). It was mentioned by (6) a sensitive biochemical biomarker of chemical contamination is the glutathione system. Our study's objective was to elucidate the deleterious impacts of sodium nitrate on specific blood images and physiological metrics, as well as the prophylactic role of glutathione**.**

Material and Methods:

In this investigation, forty adult male albino rats, approximately one month and a half old were used. Rats range in weight from 100 to 150 g. The cages housing the animals were hygienic. Water and food were added freely.

Experimental animals were divided into four groups (10 for each/group) as follows:

Group 1 serves as control group and supplements with Nacl 0.9% normal saline.

Group 2 received a sodium nitrite supplement 30 mg/kg bw.

Group 3 supplemented initially with sodium nitrite and glutathione.

Group 4 supplemented with glutathione 6.42 mg/kg bw.

For two months, these therapies were administered orally through a stomach tube. Animals were decapitated at the end of the experiment, and blood samples were collected in sterile, dry centrifuge tubes. Before analysis, the serum was separated and stored at -20 °C. Heparinized capillary tubes were used to collect blood samples for hematological examination at the same time.

Measured Parameters:

A number of variables were recorded. The count of white and red blood cells, Hemoglobin, Hematocrit, glucose value and serum levels of aspartate transaminase and alanine transaminase (ALT & AST) activity was measured (7).

Results and Discussion:

The red blood corpuscles (RBCs), hemoglobin (Hb) concentration, and hematocrit (Hct) value of rats treated with sodium nitrite for two months exhibit significant decrease when compared to the control and treated groups according to data in table (1). Additionally; table (1) showed that there was a noteworthy reduction in the quantity of white blood cells across all three groups when compared to the control group, with the glutathione-treated group exhibiting the greatest significant rise.

Table 1: The effect of sodium nitrite and glutathione alone and their combination on blood parameters in male rats.

Parameters\group	RBC(n×106/mm³)	Hb(g\dl)	PCV%	WBC(n×103/mm³)
Group(1)	**7.846a ±0.032**	**15.151 b ±0.024**	**47.071 c ± 0.358**	**11.518 a ± 0.034**
Group(2)	**5.388 d ±0.031**	**13.110 d ± 0.041**	**38.357 d ±0.048**	**7.432 d ±0.023**
Group(3)	**7.519 c ±0.016**	**16.110 a ± 0.040**	**48.421 b ±0.013**	**9.632 C ±0.023**
Group(4)	**7.629b ±0.025**	**14.569 c ±0.040**	**50.722 a ±0.516**	**10.174 b ±0.037**
LSD	0.11	0.58	1.35	0.54

*Small different letters denote significant differences between groups (p≤0.05).

Table 2's data showed that, after two months, rats given with either glutathione alone or glutathione plus sodium nitrite showed significant decrease in their serum glucose levels. After two months, there was discernible elevation in the serum activity of AST and ALT in rats treated with glutathione alone. On the other hand, rats treated with sodium nitrite showed a substantial decrease (P< 0.05) in AST activity after 2months when compared with the control group, whereas significant increase (P< 0.05) in ALT activity were detected after 2months.

Table 2: The effect of sodium nitrite and glutathione alone and their combination on biochemical parameters glucose, ALT and AST on male rats

Parameters\group	Glucose (mg\dl)	AST(IU\L)	ALT(IU\L)
Group(1)	**99.334 a ±0.025**	**27.920 c ±0.014**	**24.522 c ±0.013**
Group(2)	**82.833 d ±1.964**	**55.400 a ±0.253**	**38.331 a ±0.017**
Group(3)	**95.522 c ±0.035**	**31.925 b ±0.015**	**26.721 b ±0.012**
Group(4)	**96.433 b ±0.042**	**25.628 d ±0.016**	**21.132 d ±0.017**
LSD	**0.91**	**2.29**	**2.19**

*Small different letters denote to significant differences between groups (p≤0.05).

The RBC count, Hb concentration, and Hct level in the sodium nitrite-treated groups decreased significantly, according to the study's data due to the potentially harmful effects of sodium nitrite on the liver, spleen, and bone marrow, the decline may be explained by microcytic and/or hypochromic anemia (8). The authors (8 and 9) report that methmoglobin (met Hb) levels increased noticeably along with this decline, indicating that treatment with sodium nitrite may have been the cause (9).

There was a notable decline in met hemoglobin (met Hb) levels concurrent with this decrease suggesting that sodium nitrite treatment may have caused it (9).

It is well known that the nitrite changes the ferrous to ferric ions in hemoglobin. Sodium nitrite rise met Hb, but had no effect on RBC hemolysis (10 & 11).

When there is toxicity, the liver performs an essential physiological role by getting rid of the toxicants once they have digested and have dropped dramatically in just two months. Our results are in agreement with (12). The current results showed a minimal decline in serum glucose levels in rats treated with sodium nitrite. The liver is an essential physiological organ in cases of poisoning because it gets rid of the toxicants when they are broken down and digested. This leads to a breach in the integrity of the cell membrane, which increases the amount of enzymes in the blood (12).

According to the current findings, rats given sodium nitrite had significantly higher serum AST and ALT activity. Increased blood transfer activity has been used as a marker for tissue injury. However, additional elements are taken into account for this process, such as changes in the cell membrane's permeability, an increase in the synthesis of the enzyme, or a decrease in the pace at which the enzyme degrades (13).

Correspondingly to recent researches the breakdown and necrosis of liver cells, along with cell wall destruction and cytolysis, are the causes of the elevation in serum AST and release a significant amount of these mitochondrial enzymes into the bloodstream. On the other hand, oxidation of significant iron-containing enzymes, such as cytochromes, which are involved in cellular respiration and other oxidation-reduction processes, may be the cause of nitrite's harmful effects on the liver. Rats given sodium nitrite treatment showed a substantial rise in blood AST and ALT activity, according to (14).

Also other researchers hypothesized that the observed enhancement of ALT activity was due to sodium nitrite contact with the enzyme molecule rather than with the tissues (15). As highly sensitive liver indicators, the activities of ALT and AST are generally markedly elevated after the administration of numerous hepatotoxic drugs, resulting in acute hepatocellular destruction or extrahepatic blockages Petra *et al.,* linked increased ALT and AST activities to liver cell injury, while (16) claim that coloring chemicals cause damage to liver tissue and significantly increase ALT, AST, and alkaline phosphatase.

Statistical analysis

All normally distributed data were reported as means ± SEM and were analyzed using the independent sample t-test and Pearson correlations, respectively, (SPSS, version 20.0, Chicago, IL, USA), to examine the differences in physiological and biochemical values for correlation analysis.When $P < 0.05$, the differences were deemed significant in (17).

Conclusion:

Food additives such as sodium nitrite can have dangerous effects; thus, it is best to utilize toxic compounds sparingly and offset any harmful effects with antioxidants like glutathione.

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تأثير نتريت الصوديوم على بعض المعايير الدموية والكيميائية الحيوية في ذكور الجرذان المعالجة بالجلوتاثيون

هدى كامل خصاف
فرع الفسلجة والادوية والكيمياء الحياتية. كلية الطب البيطري؛ جامعة البصرة، البصرة، العراق.

الخلاصة:
في إنتاج الأغذية الصناعية يتم استخدام المواد الحافظة الغذائية مثل نتريت الصوديوم بشكل متكرر. تم ربط استخدام بعض هذه المواد الحافظة الغذائية بالتأثيرات المسخية والمسرطنة. من أجل التحقق من تأثير المضافات الغذائية والمواد الطبيعية المضادة للأكسدة على الجرذان الأربعين من ذكور الجرذان البيضاء البالغة، تم تقسيمهم لمدة شهر ونصف تقريباً إلى أربع مجموعات على النحو التالي، المجموعة (١) هي مجموعة السيطرة اعطيت المحلول الملحي الاعتيادي بنسبة ٩و٠% تلقت المجموعة (٢)نتريت الصوديوم لوحده (٣٠ مجم / كجم من وزن الجسم) والمجموعة (٣) اعطيت نتريت الصوديوم (٣٠ مجم / كجم من وزن الجسم) والجلوتاثيون(٦٤٢ مجم / كجم من وزن الجسم)، والمجموعة (٤) اعطيت الجلوتاثيون لوحده (٦٤٢ مجم / كجم من وزن الجسم). تم قياس مجموعة متنوعة من المعاملات، بما في ذلك عدد خلايا الدم الحمراء والبيضاء ومستوى خضاب الدم والهيماتوكريت، ومستوى الجلوكوز وتحليل نشاط ناقلات أمين الألانين والاسبارتات. أظهرت النتائج بوضوح أن عدد كرات الدم الحمراء وتركيز الهيموجلوبين ومستوى الهيماتوكريت في الجرذان المعاملة بنتريت الصوديوم لمدة شهرين اظهرت انخفاضا معنويا عند مقارنتها بمجموعتي السيطرة والمعاملة، الجرذان التي أعطيت إما الجلوتاثيون وحده أو الجلوتاثيون زائد نتريت الصوديوم اظهرت انخفاضًا معنويا في مستويات الجلوكوز في الدم وكان هناك ارتفاع ملحوظ في نشاط ALT &AST.الجرذان المعاملة بنتريت الصوديوم اظهرت انخفاضا واضحا في نشاط ناقلات الاسبارتات بعد شهرين من المعاملة عند مقارنتها مع مجموعة السيطرة، بينما كان هنالك زيادة معنوية(< ٠.٠٥) في نشاط ناقلات الالانين التي تمت ملاحظتها بعد شهرين من المعالجة. تشير هذه النتائج إلى الآثار الضارة لاستخدام المكملات الغذائية والملونات الصناعية وتثبت أيضًا التأثير المحسن للجلوتاثيون، ونظرًا للآثار الضارة المحتملة للمضافات الغذائية يُنصح بتقليل استخدام نتريت الصوديوم كمضافات غذائية الى الحد الادنى.

الكلمات المفتاحية:، نتريت الصوديوم،المضافات الغذائية، الجلوتاثيون، الجرذان ، مسرطن.