The Egyptian Journal of Hospital Medicine Vol., 2 : 23 - 46 March 2001
I.S.S.N: 12084

Progressive effects of the interaction of Sodium nitrite and sunset yellow on different physiological parameters in albino rats

Eman. G.E. Helal
Al- Azhar University, Faculty of Science for girls, Zoology department.

Abstract:

It has been noticed that the Egyptian children usually eat and drink food containing both food preservative and food colourants at the same time . This has attracted the attention to study the interaction between one of food preservatives (sodium nitrite) and a food coloring agent (sunset yellow). The mixture of the two agents at the limited dose of each was found to be a lethal dose. So, $1/10^{th}$ of this dose was used daily for 30 days.

Animals were devided into three groups. The first group served as a control, while the second group was orally administered a mixture of 10mg sod. nitrite ($NaNO_3$)/kg and 0.5 mg/ kg /day sun set yellow (S.S.Y)/. The third group received selenium (5 mcg/kg) in addition to the above mentioned mixture. After 30 days of treatment, half of the animals from each group were decapitated. The other half of animals was left for another 15 days without treatment for recovery.

Ingestion of the mixture of (NaNO3 and S.S.Y) significantly decreased rat body weight, R B C and WBC counts, Hb %. , Hct %. Serum inorganic phosphorus, serum protein and serum albumin. Significant increases were observed in serum glucose, T3, T4, calcium, GGT, LDH, Cpk, ALK.ph and cholesterol. Also cholesterol of brain, liver and heart were significantly elevated. No changes were recorded for organ/ body weight, respiratory rate, heart beats, rectal temperature, AST and ALT activities of serum and tissues, acid phosphatase activity, total lipids of serum and tissues, cholesterol of muscle and kidney and serum triglycerides

A complete recovery from the abnormalities of most biochemical and haematological parameters was observed after 15 days recovery or when selenium was administered

This draws attention to the dangers of interactions of such preservatives and colorants. The present study showed that even the permitted doses of colourants and food preservatives may be harmful.

Introduction:

Food additives are substances intentionally added to food. They may be natural or synthetic (Harris, 1986). The principal classes of food additives are coloring agents, preservatives, flavours, emulsifiers and stabilizers (Lindsay, 1985). One of the principal preservatives are the nitrites which are used in the form of salts or free acids (HMSO, 1987). The use of sodium nitrite as a preservative is common in cooked meat, sausages milk used for some types by cheese and pizza . Because of the use of more than one type of such food ,the percentage of nitrite content of the daily food ration may be higher than the admissible level (Bilczuk *et al.*, 1991). Apparently very little nitrites are formed by endogenous synthesis and most, if not all are of dietary origin (Bartholomew and Hill, 1984).

Food colorants may often be considered simply cosmetic in nature, but its role is very significant. Both food quality and flavor are closely

Refree : Prof ; Ismail Sadek

associated with color. Consumers are conditioned to expect food of certain colors and to reject any deviation from their expectations (Amerine *et al.*, 1965).

Almost 25 years ago selenium was recognized as an essential element, lack of which resulted in the development of peroxidaton in membranes. It is required for the activity of glutathione peroxidases (Combs *et al.*, 1975) and is believed to act as an antioxidant (Rotruck *et al.*, 1973). Recently, selenium was used in treatment of free radical – associated disease such as diabetes (Kahler *et al.*, 1993).

It has been noticed that children often eat foods containing preservatives and at the sametime drink some drinks containg colorants. The question arises here whether the food preservative and colorants would interact with each other? The aim of this work is therefore, to test this possibility by mixing a limited dose of sodium nitrite (one of the most used preservatives) with limited dose of sun – set yellow (one of the most attractive colorants) and test a recombinant effect on rats. The interaction of both limited doses resulted in a recombinant lethal dose that led to the death of all rats used. So, this study was planned to achieve two goals: the first is: to follow up the biological effect of this mixture on young male albino rats; and the second: is to study the effect of selenium (one of the most potent antioxidant) in order to illustrate the possibility of ameliorating the expected hazards.

Material and Methods:

Thirty young male albino rats (weighing about 70-80g) were used in this study. Animals were housed in stainless steel cages , fed on rat chew and offered water *ad libidum* The animals were divided into three equal groups (10 rats each) as follows:

The first group : (control group). ,

The second group : orally administered 10mg $NaNO_3$/ kg and 0.5 mg sunset yellow (S.S.Y) daily for a month.

The third group: received the same doses of $NaNO_3$ and S.S.Y in addition to 50 mcg/ kg/ day of selenium administered orally to each rat, for one month.

Body weights, respiratory rate, heart beat and rectal temperature were recorded once a week throughout the experimental period. After 30 days of treatment, 5 animals of each group were weighted and then decapitated. While the other half was kept for two weeks, without any additional treatment for recovery.

From each of the decapitated rats, liver, kidney, hearts and testes were dissected out, cleaned from adherent tissues and weighed at once. Pieces of liver, skeletal muscle, kidney, heart and brain were incubated in an appropriate amount of 30% potassium hydroxide for total protein determination. Other pieces were immersed in concentrated sulfuric acid for total lipid determination or in saline solution and kept in the refrigerator at -20°C for determination of cholesterol, AST and ALT.

Blood samples were collected for hematological and biochemical parameters. EDTA, an anticoagulant, was added to collected blood for hemato -logical parameters, while the blood samples for biochemical parameters were centrifuged for 10 min. at 5000 rpm and supernatent sera were separ -ated for analysis without storage or delay. Hemoglobin concentration was determined according to Van–Kampen & Zulstra (1961). Red and white blood cells were counted , and haematocrit values (Hct) were estimated using the

technique of Rodak (1995). The bioche -mical analysis was carried out on the blood sera. Glucose determination was based on the enzymatic method described by Siest & Schielf (1981). Total proteins were estimated using the Biuret method as described by Doumas (1975). Total lipids were determined according to the method of knight *et al*., (1972). Albumin was evaluated according to the method of Webster (1977), while serum cholesterol was determined as mentioned by Fossati and Medici (1987). AST and ALT activities were accomplished using the method of Reitman and Frankel (1975). Gamma – glutamyltranspeptidase was estimated by the method of Meister and Groffith. (1981). While, lactic dehydrogenase (LDH) activity was determined acco -rding to Raabo (1963). CPK (creatinine - phospho - kinase)assay was perfo -rmed using sigma chemical company reagent kits (st. louis Mo).

Triglycerides were determined by the method of Rojkin *et al*. (1974). Alk -aline phosphatase activity was measu -red according to the method of Belfied and Goldberg (1971). And the activity of plasma acid phosphatase was determined according to the methods of Tietz (1986).

Inorganic phosphorus was determined according to the method of Fiske and Subbarow (1925) and serum calcium (Ca^{++}) concentration was esti -mated according to the method adopted by Ray Sarkar and Chauhan (1967). Thyroid hormores assay were determined by using the enzyme – linked immunosorbent assay (ELISA) (Withley *et al*., 1996).

Student t-test was used for comparison of data obtained for different parameters from different experimental animal groups. Significant differences between the means of control and treated groups were considered only at $p < 0.05$ (Sokal and Rohif, 1981).

Results:

Control young rats showed a net gain (19. 6 %. and 18. 9 %.) in body weight after the treatment and recovery periods respectively, while the gain of body weight in rats treated by the mixture of sodium nitrite , sunset yellow ($NaNO_3$ and S.S.Y) and selenium significantly was ($p<0.01$) lower (16.2% and 16.4%)as compared to control. On the other hand, there was significant ($p\leq .01$) weight loss in rats treated with a mixture ($NaNO_3$ and S.S.Y). After recovery period of 15 days, there was a gain in the body weight (10.6%). The organs/ body weight of male albino treated and control rats after both treated and recovery periods are presented in table (1). No significant detectable changes in the relative weight of the kidney, brain, heart, liver and testis were recorded in all treated groups throughout the experimental period.

All of the animals showed insign -ificant changes in respiratory rates, heart beats and rectal temperature, after both treatment and recovery periods (Table 2)

Investigation of the effect of oral administration of the mixture of sod. nitrite and sunset yellow on various haematological indices of rat red blood cells revealed highly significant decr -ease ($p<.01$) in all parameters. Supplem -entation with selenium, howe -ver ,resulted in values for hematological parameters which were not significant from control rats. After the recovery period, no significant change was recorded in all treated groups (Table 3). Investigation of other hematological indices revealed that treatment with the mixture caused significant reduction in

white blood cell count (Table 3).

The treated rats showed a highly significant increase (P<0.01)all param - eters. in glucose level compared to that in the control ones. While, selenium treated rats had an insignificant change in glucose level after both treatment and recovery periods in comparison with the control (Fig 1).

A statistically significant increase in the level of both T_3 and T_4 was observed in rats which received sod. nitrite and sun-set yellow compared to the control , after both treatment and recovery period. These levels reached the normal value in the group treated with both the additives and selenium (Fig 2 and 3).

Fig (4), showed the statistical analysis of the mean value of calcium in the different groups. A significant increase in serum concentration of calcium (P<0.01) in the group treated with ($NaNO_3$ + S.S.Y) after treatment and recovery periods. However, insignificant changes were recorded in the group treated with food additives and selenium throughout the experim - ental period. On the other hand, a significant decrease (P< 0.01) was recorded in serum phosphorus level in all treated groups after 30 days of treatment while, the group treated with the dual treatment {($NaNO_3$ + S.S.Y) and selenium)} showed insignificant change after the recovery period (Fig 5).

The present results indicate that, AST and ALT activities (of serum and tissues) were still within the normal values for all groups after the treated period (30 days) or the recovery period (15 days) in comparison with that of the control group (Table 4 & 5).

Administration of both $NaNO_3$ and S.S.Y increased (P<0.01) serum activities of GGT, Cpk and alkaline phosphatase compared to those of the control till the end of the experiment. (Figs 6, 8 and 9). No significant changes of these enzymes activities were observed in the group given selenium. Further, serum LDH activity exhibited a highly significant increase ($p < 0.01$) in food aditives tread group after treated period only as compared with the control ones. (Fig 7). However, no significant changes were recorded in serum acid phosphatase activity (Fig 10) in all groups. The administration of selenium showed insignificant changes of all the tested enzyme activities throughout the experiment.

Table (6) showsed total serum protein contents in control and in the two different treated groups. Total serum protein contents were signifi - cantly decreased ($P < .01$) due to the mixture treatment after the treated period only. Selenium treatment caused insignificant changes in total serum protein content as compared to control rats. Total protein of brain, liver, muscle, kidney and heart recorded insignificant changes in all treated groups throughout the experimental period (Table 6).

Concerning the effect of the mixture on serum albumin, the result revealed a significant decrease (P< .01) after the treatment period. While insignificant changes were recorded in case of selenium treated group and in the mixture treated group after recovery period. Whereas, the level of serum globulin recorded insignificant changes after the treatment period followed by a significant decrease after the recovery period in case of a mixture treated group. On the other hand, selenium treated group revealed a significant decrease in serum globulin ($P < .05$) which was more pronounced ($P < .01$) after the recovery period. A/G recorded insignificant changes in all groups except that treated with selenium where it recorded a significant increase (P <

.01) after the recovery period only (Table 6).

The data on serum and tissue total lipids, total cholesterol and serum triglycerides were shown in tables (7 and 8). No significant difference betw - een groups were observed in serum and tissue total lipids (Table 7), and serum triglycerides (Table 8) throughout the experimental period. On the other hand, serum cholesterol showed a significant increase ($P < .01$) in a mixture treated rats after a month of treatment. A highly significant elevation ($P < .01$) was detected in cholesterol of brain, liver and heart for a mixture treated rats but they were not affected by the adm - inistration of selenium. This elevation was not maintained after the recovery period (Table 8).

Discussion:

The present observations of body weight loss after the treatment with both food preservator and food colorant may be due to the reduction of food utilization (Grant and Butler, 1989). On the other hand, the reduction of mean body weight may be due to the increase in the level of both nitrite and sunset yellow leading to increased catabolic processes in the body. Greenblatt and Mirvish (1972), Maekawa *et al.* (1982) and Til *et al.* (1998) recorded reductions of body weight gain due to nitrite treatmeats. Many investigators recorded a reduction in body weight as a result of colorants supplementation (Brozelleca *et al.* 1989; Osman *et al.*, 1995 and Abu El – Zahab *et al.*, 1997). In the prese - neat findings, it was cleared that selen - ium ameliorate the catabolic effect of both $NaNO_3$ and S.S.Y.

The present results showed insignificant changes of the relative weight of the tested organs of the male rats treated with ($NaNO_3$ + S.S.Y) or with ($NaNO_3$ + S.S.Y. + selenium). Hirose *et al.* (1993) and yoshidae *et al.* (1994) reported an increase of absolute and relative liver and kidney weights of rats treated with sodium nitrite. These results are in contrary with those of the present work. This could be due to the lessions and other disturbances (Dini *et al.*, 1992, and Hirose *et al.*, 1993) leading to loss of nutrient and fluids or to inhibition of gastrointestinal mucosa Na^+ / K^+, ATP ase and alkaline phosphatase (Bruning-Fann and Kaneene, 1993).

The present results revealed that respiration rate , heart beats and bodi tenyerature. are almost the same in all groups even under the treatment conditions.

Administration of both sod. nitrite and S.S.Y for one month to rats induced a decrease of W.B.Cs, R. B.Cs, Hb% and Hct %. It is known that nitrites convert the ferrous ion of haemoglobin to ferric ion both in vivo and vitro (Ganong, 1997). This can explain the reduction of haemoglobin level. In other words, administration of both nitrite and S.S.Y leads to haematopoietic tissue hypoxia resulting on the long term (one month in the present study) to a decrease of red blood cell production and hence to reduction of blood heamoglobin level.

The decrease of haemoglobin due to nitrite treatment has been reported using different animals including rats (Abdel – Rahim *et al.*, 1988, Smith, 1991 and Reutov *et al.*, 1993), mice (Walker *et al.*, 1957), dogs (Harely and Robin, 1962), Swine and sheep (London *et al.*, 1967). Further, nitrites have been reported to induce a reduction of haemoglobin level in human (Heisler *et al.*, 1974 and white, 1975).

The present results are, in part, comparable to those obtained by Rastogi and Prasad (1983 a and b) where they found that feeding of albino

mice in the common food colour metanil yellow led to changes in hematological values. Total erythrocyte count and hemoglobin had decreased. Erythrocyte sedimentation rats (E.S.R), mean corpuscular volume (MCV) and mean corpuscular hemoglobin concentration (MCHC) had increased. These facts suggested the occurance of normochromic macrocytic anemia.

Differential leukocyte count showed marked increase in the number of lymphocytes and monocytes and decrease in the number of neutrophils and eosinophils (Rastogi and Prasad, 1983b). The present study was in accordance with Mackenzie *et al.*, (1992) who found a reduction in total white blood cell counts as a result of caramel treatment in rats. Also it was clear that dual treatment in the present study had ameliorated all haematological variation.

Treating the rats with both $NaNO_3$ and S.S.Y induced a gradual significant increase in the blood sugar level. However, one can expect that sodium nitrite produced hyperglycemia due to deficiency of insulin release. It is known that nitric oxide is formed from nitrites (and nitrate) at least by the vascular epithelial cells (Harriosn and Bates, 1993 and Katzung, 1995). Both nitric oxide and nitrites open potassium channel (Katzung, 1995) which through closing voltage gated Calcium channels decreases intracellular calcium. Calcium is known to trigger insulin secretion, (calcium channel blockers are known to produce hyperglycemia; (katzung, 1995).

The present results go in paralled with those reported on the hyperglycemic effect of sodium nitrite in rats (Abdel – Rahim *et al.*, 1988 and Shelpov *et al.*, 1991). They reported that in presence of nitrite ion, the activity of amylase increases beside an inhibition of adrenaline – induced activation of phosphorylase. This results in the liberation of glucose from glycogen, so blood glucose increases while liver glycogen decreases.

The elevation of level of serum glucose was also interpreted by the effect of sun-set yellow on enzyme system of the glycolytic pathway. It is not surprising to find an enhanced heperglycemia due to the new compouned resulted from the reaction of sodim nitrite and sun set – yellow treatment of the rats. The observed improvement shown in glucose in selenium treated group may be due to its action as antioxidant. These results are in contrast with Rasekh *et al.*, (1991) who observed hyperglycemic effect of selenium in rats.

The present study revealed that administration of sod. nitrite and sunset yellow to rats caused variable degree of stimulation of thyroid gland function after treated and recovery periods. This was proved by the significant increase in serum thyroid hormones T_3 and T_4. The interaction between sod. nitrite and sunset yellow may give a new chemical component, which has a stimulatory effect on thyroid gland. This effect could be attributed to its chemical structure that can compete with thyroxine – binding globulin leading to its deficiency and to hyperthyrodism by feed – back mechanism (Gold and Vladutin, 1994).

These changes in thyroid hormones could also be resulted from alteration in the pitutary – thyroid axis as a consequence of the stressing effect of this new component. This was in accordance with El – Saadaney (1991).

It is known that thyroid hormones enter the brain. Large doses of thyroid hormones cause irritability, restlessness and rapid mentation. Also thyriod hormones have marked effect on brain

development. (Ganong 1997, Guyton and Hall, 1998).

The present work concluded that, the studied food additives can markedly alter the endocrine function of thyroid gland leading to hyper function. This might play a role in children hyperactivity probably through affec - ting higher centers in the brain. Also it is clear that selenium ameliorate this effect due to its antioxidant property. This may be due to blocking the gene - ration and propagation of free radicals.

The result of this study demonstrated that the administration of ($NaNO_3$ + S.S.Y) caused a significant increase in serum calcium level. This observation was similarly recorded by Sharma (1989) who recorded higher values of minerals in rats treated with metanil yellow. On the other hand, the present results showed a significant decreare in serum phosphate after treatment with the mixture of $NaNO_3$ and S.S.Y. Also it was clear that selenium could not ameliorate the effect of this new compound ($NaNO_3$ + S.S.Y) on serum phosphote during the treated period, while after the recovery period it turned back to the normal value in selenium treated group. Helal *et al.* (2000) stated that S.S.Y did not affect serum level of both Calcium and Phosphate . So, the present results may be due to the new compound (resulted from the reaction between both $NaNo_3$ and S.S.Y).

Many enzymes like alkaline phosphatase (AP) and GGT tend to be released into plasma in large amounts following the hepatocellular damage (Whitby *et al.*, 1992). Gamma glutamyle transferase is considered to be more specific for liver function tests. Its activity is markedly increased in plasma in both primary and secondary carcinoma of liver (Whitby *et al.*, 1992).

In the present work, both GGT and AP were increased in the group of rats given ($NaNO_3$ + S.S.Y). However, the significant reduction in GGT and AP activities in selenium treated group may be due to the good effect of the selenium to improve the activity of liver cells or to stop the damage of liver cell membranes and hence the release of their enzymes. The insignificant changes in AST and ALT activities in all treated groups may be due to AST and ALT activities is less sensitive than GGT.

The effect of the mixture of $NaNO_3$ and S.S.Y on serum enzyme activities (LDH and CPK) in this study provides further evidence on the effect of this mixture on the liver and heart. This elevation could be attributed to a generalized increase in membrane activity . (Doran and Wilinson, 1975; Ebashi *et al.*, 1959).

The present study showed a significant increase in serum LDH activ -ity indicating cellular damage. Morlier *et al.* (1991) reported that increased lipid peroxidation is accompanied by the release of LDH reflecting membrane damage. Rybczynska *et al.* (1996) found that lipid peroxidation of cell membranes is associated with inactiva - tion of membrane bound enzymes. Based on these molecular events, it is possible to explain systemic and metabolic responses evidenced in the present study by elevated activity of serum LDH as well as increased contents of cholesterol, in addition to a drop in total protein content of a mixture ($NaNO_3$ + S.S.Y) treated rats, which are reduced to reach normal levels after the treatment with selenium.

Sato *et al.* (1979) indicated that sever deficiency of selenium develops myodystrophy in cattles. Therefore, it is possible to assume that selenium may exert a protective role against the

mixture toxicity, since it ameleorated all of the enzyme activities investigated. A significant decrease of the total serum protein was recorded after treatment with the mixture of $NaNO_3$ and S.S.Y for one month. The decrease of total serum protein due to the mixture treatment was reflected on serum albumin level where a remarkable decrease was recorded. It is suggested that this decrease of albumin resulted from liver function impairment induced by the nitrite. The globulin fraction, on the other hand, was not affected generally at the sametime, but it was affected after the recovery period. Selenium plus the mixture treatment lead to a decrease in globulin all over the experimental time and an increase A/G ratio after the recovery period.

The harmful effect of nitrite is reflected on the biosynthesis of protein as reported by Eremin and Yocharina (1981). They found that serum protein of rats are decreased due to the stimulatory effect of the nitrite on the thyroid and adrenal glands that leads to block of protein synthesis while fast breakdown occurs. This leads to an increase of free amino acids and to a decrease of protein turnover Yanni *et al*., 1991). Furthermore, sodium nitrites have been reported to produce retardation of growth (Atef *et al*., 1991), necrotic changes of the liver and deterioration of the liver function and renal tubules (Anthony *et al*., 1994; El-Bllal *et al*., 1994; Guler *et al*., 1994 and Rodriguez – Morona and Tarazona, 1994), reduction of myocardial GSH-PX (Yang and Wang, 1991), gastric function and alteration of gastric mucosal absorption and decreased food consumption (Burning – Fann and Kaneene, 1993 and Hirose *et al*., 1993).

However, it is clear that sodium nitrite decrease total serum proteins and albumin mainly through its effects on the liver either through inhibiting oxidative phosphorylation process and hence the availability of the energy source of protein synthesis (Anthony *et al*., 1994) and other metabolic processes or through the necrotic changes especially of the plasma membrane (Guler *et al*., 1994). At the same time, the nitrite effects on the process of reabsorption in the kidney tubules and impaired absorption of digested food material cannot be ignored. Rodriguez – Morona and Tarazona (1994) indicated that uronyl nitrate decreases proximal tubular reabsorption which results in the activation of glomelular feed – back and lowers nephron filtration rate. Also, these results find good support in the study carried out by Helal *et al*. (2000) who illustrated a marked decrease of serum protein in rats after treatment with sunset yellow or carmine.

Furthermore, it is conceivable to assume that selenium may exert a protective role against the mixture – induced toxicity. Evidence of this view in the present findings is that selenium treatment of the mixture – intoxicated rats resulted in an increase of total serum proteins. This effect probably reflects the ability of selenium to protect the protein manufacturing machinery from the mixture – induced cellular damage.

In the present investigation, significant changes occurred in the level of serum or tissues (examined) total lipids of rats treated with the mixture alone or the mixture and selenium for one month of treatment. Similar results obtained in case of serum triglycerides. Cholesterol is the most abundant steroid in the cells of higher animals. It is considered an essential structural component of cell membrane (Thorpe *et al*., 1964). In the present study, serum, brain, liver and heart cholesterol were increased significantly ($p < .01$) over

controls in the group treated with mixture ($NaNO_3$ + S.S.Y). While no significant changes were observed in rats treated with selenium in addition to the mixture. Similar rise in serum cholesterol level was encountered simultaneously with a reduction in serum selenium in rats fed on diet provided with 24mg nitrate/ kg diet for 159 days. When reducing agents such as selenium or ascorbic acid were added to the diet, serum cholesterol was fallen and serum selenium was raised (Angelis *et al*., 1996). The authors suggested that the adverse effects of nitrate diet may occur in relation to peroxidation. This suggestion may support the findings of Bruning – Fann and Kaneene (1993) where, they found that nitrate ingestion in monogastric animals has been linked to interference with the metabolism of other antioxidants than selenium such as ascorbic acid, vitamin A. and E. Hence, the increased level of serum cholesterol noted herein in rats exposed to the mixture could be attributed to the peroxidation of cell membrane lipids (Standberg, 1977). So, a current evidence indicated that nitrites act as cell membrane oxidants (Beaupre and Schiffman, 1994).

The present study draws attention to the danger of extra serum cholesterol as a result of the mixture intake. It can be stored as a component of pathologic deposits in the arteries representing a potential risk for the development of a-therosclerosis, thus increasing the possibility of suffering from cardiova -scular diseases and hypertension. Furthermore, hyperfunction of the thyroid gland manifested by higher levels of serum T3 and T4 can lead to thyrotoxicosis.

From the present results, it can be pointed out that most of the metabolic and pathological criteria in the tested rats altered by administration of this mixture regressed after recovery of many to normal status in days after stoppage of adminstration of mixture or as a result of administration of selenium.

The continuing world wide interest in toxicity/ carcinogenicity studies on the various edible food additives available will eventually yield unlimited information aiming at the improvement of adverse effects of subs -tance as certain behaviours affecting mankind.

However, one should put into consideration the difficulty of extrapolation from animal studies to humans, moreover the difficulties in estimating human exposure to food additives.

Nevertheless, it is advisable to minimize the use of food additives in food and avoid using them as much as possible in infants and children foods.

References:

1. Abdel – Rahim, E.A.; El – Desoky, G.E.; Shaban, O.A and Afify, A.s. (1988): Studies on the effect of sodium nitrite on hemoglobin fractions and growth rate of albino rats. Bull. Fac. of Agric. Univ. of Cairo, 39 (4): 1503 – 1561.
2. Abou El – Zahab, H.S.H.; El – Khyat, Z.A.; Awadallah, R. and Mahy, K.A. (1997): Physiologlcal effects of some synthetic food coloring additives on rats. Boll. Chim. Farm., 136 (10): 615 – 627.
3. Amerire, M. A.; Pangborn, R.M.; Rossler, E.B. (1965): Principles of sensory Evaluation of Food. academic Press, New York.
4. Angelis, R. C – DE; Terra, I.C.M; Scialfa, J.H. and Klemps, F.I. (1996): Dietary nitrite and scavenger antioxidants trace

5. elements. Inter. J. Food sci. Nutr., 47: 23 – 26.
6. Anthony, M.L.; Gartland, K.P.; Beddell, C.R.; Lindon, J.K. (1994): Studies on the biochmical toxiology of uranyl nitrate in the rat. Arch. Toxicol., 68 (1): 43 – 53.
7. Atef, M.; Abo – norage, M.A.; Hanafy, M.s. and Agag, A.E. (1991): Pharmacotoxicological aspects of nitrate and nitrite in domestic fowls. Br. Poult. Sci., 32 (2): 399 – 404.
8. Bartholomew, B. and Hill, M.J. (1984); The pharmacology of dietary nitrite and the origin of urinary nitrate. Fd. Chem. Toxic., 22: 789.
9. Beaupre, S.R. and Schiffman, F.J. (1994): Rush hemolysis, a bile cell hemolytic anemia associated with volatile liquid nitrite use. Arch. Fam. Med. 3: 545 –548.
10. Bellfield, A. and Goldberg, D.M. (1971): Hydrolysis of aadenosine – monophosphate by acid phosphatase as measured by continuous spectrophotometric assay. Enzyme, 12: 561.
11. Bilczuk, L., Gowin, A.; Ebertowska, Z. and Mach, H. (1991): Nitrate and nitrite levels in daily food rations of children from the rural pulowy regions. Rocz. Panstw. Zaki. Hig. 42 (2): 139 – 147.
12. Brozelleca, J.F.; Olson, J.W. and Reno, F.E. (1989): Life time toxicity carcinogenicity study of FD & C Red no. 40 (Allura Red) in Sprague – Dawley rats. Fd. Chem. Toxic., 27 (11): 701 – 706.
13. Bruning – Fann, C.S. and Kaneene, J.B. (1993): The effect of nitrate, nitrite and N – nitroso compounds on animal health. Vet. Hum. Toxicol., 35 (3): 237 –253.
14. Combs, G.F.; Noguchi, T. and scott, M.L. (1975): Mechanism of action of selenium and vitamin E in protection of biological membranes. Federation proceedings., 14 (11): 2090 – 2095.
15. Dini, L., Bernardini, R.; Resti, M. and viercei, A. (1992): Unusual reaction to food additives. Pediatr. Med. Chir., 14 (1): 39 – 42.
16. Doran, G.R. and Wilinson, J.H. (1975): The origin of the elevated activities of creatin kinase and other enzymes in the sera of patients with myxoedema. Clin. Chem. Acta., 62: 203.
17. Doumas, B.T. (1975): standards for total serum protein assays. A collaborative study. Clin Chem., 21 (8): 1159 – 1161.
18. Ebashi, S.; Toykura, Y.; Momoi, H. and sugita, H. (1959): High creatine phosphokinase activity of serum of patients with progressive muscular dystrophy. J. Biochem. (Tokyo), 46: 103.
19. El – ballal, S.S.; Ezzo, O.H.; Shalaby, S.I. A. and Fawzy, Y. (1994): Sodium nitrite toxicity in Barki sheep. Egypt. J. of comparative pathology and Clinical pathology. 7 (2): 353 – 361.
20. El – Saadany, S.S. (1991): Biochemical effect of chocolate colouring and flavouring like substances on thyroid function and protein biosynthesis. Die Nahrung, 4: 335 – 43.
21. Eremin, Y.N and Yochorina , M.G. (1981) : Effect of nitrites on the thyroid gland of rats in response to different diets of iodine deficiency .Vopr .Pitan .5 : 60-62.
22. Fisk, C.H. and subbarow, M. (1925): The colorimetric determination of phosphorus. J. Biol. Chem. 66: 375 – 383.

23. Fossati, p - and Medici, R. (1987): Abstract book: International symposium on cholesterol control and radiovascular diseases: prevention and Therapy. Milan. Italy.
24. Fujitani, T. (1993): Short – term effect of sodium benzoate in F – 344 rat and B6 C3 F1 mice. Toxicol. Lett., 69 (2): 171 – 179.
25. Ganong, W.F. (1997): Review of Medical Physilogy. 8th ed. Libraure du liban, Appelton of Longe, lebanon, California, 296 – 311.
26. Gold, E. and Vladutin, A. (1994): Latrogenic hyperthyrodism of long duration in an individual with thyroxin – binding globulin deficiency. Clin. Chem, 40 (12): 2323 – 2324.
27. Grant, D. and Bulter, W.H. (1989): Chronic toxicity of sodium nitrite in male F – 344 rat. Fd. Chem. Toxic., 27 (9): 565 – 571.
28. Greenblatt, M. and Mirvish, S.S. (1972): Dose – respon studies with concurrent administration of piperazine and sodium nitrite to strain a mice. J. Nat. Concer Int., 49 : 119 – 124.
29. Guler, A.H.; Sapan, N.; Ediz, B.; Genc, Z. and Ozkan, K. (1994): Effect of copper on liver and bone metabolism in malnutrition. Turkish, J. Pediat., 36 (3): 205 – 213.
30. Guyton, A.C. and Hall, J.E. (1998): Text book of medical physiology. 9th ed. WB. Saunders company, Philadelphia, London, Toronto, Sydney, Tokyo, 945 – 956.
31. Harley, J.D. and Robin, H. (1962): The effect of the nitrite ion on intact human erythrocytes. Blood, 20 (6): 710 – 713.
32. Harper, F.A. (1977): Review of physiological chemistry. Long medical publication, less Altes, California, 15th ed. Pp: 326 – 379.
33. Harris, J.B. (1986): Natural toxins. Animal, plant and microbial. Cited in food and additives in tolerance in childhood (1994). P. 179. Black well scientific London – Boston.
34. Harrison, D.G. and Bates, J.N. (1993): The nitronassodilators: new ideas about old drugs. Circulation, 87: 1461.
35. Heisler, E.G.; Siciliano, J.; Krulick, S.; Feinberg, J. and Schwartz, J.H. (1974): Changes in nitrate and nitrite content and search for nitrosamines in storage, Abused spinach and beets. J. Agric. Food Chem., 22 (6): 1029 – 1032.
36. Helal, E.G.E.; Zahkouk, S.A, M and Mekawy, H.A. (2000): Effect of some food colours (synthetic and natural products) on liver and kidney functions of young albino rats. The Egyptian Journal of hospital medicine. 1:
37. Hirose, M.; Tanaka, H.; Takahashi, S.; Futakuchi, M.; Fukushima, S. and Ito, N. (1993): Effects of sodium nitrite and catechol in a rat multiorgan model. Cancer Res. 53 (1): 32 – 37.
38. H M S O. (Her majesty's stationary office, London). (1987): Food additives. The balanced approach. Cited in food additives intolerance in childhood (1994) p. 180. Black well scientific. London – Boston.
39. Kahler, W.; kuklinski, B.; Ruhlmann, C. and plotz, C. (1993): Diabetes mellitus. A free radical – associated disease. Results of adjuvant antioxidant supplementation. Z. Gesamte. Inn. Med., 18 (5): 223 – 232.
40. Katzung, BG. (1995): Basic and clinical pharmacology. A long

medical book, 6th ed., less Altes, California, p.p: 199 – 203.
41. Kelloff, G. J.; Boone, C.U.; steele, V.E.; Fay, J.R.; Lubet, R.A.; crowell, J.A. and sigman, c.c. (1991): Mechaniestic considerations in chemopreventive drug development. J. Cell. Biochem. Suppl., 20: 1-21.
42. Knight, J.A.; Anderson, S. and Rawle, J.M. (1972): chemical basis of the sulfo – phosphovanillin reaction for estimation total serum lipids. Clin. Chem., 18 (3): 199 – 202.
43. Lindsay , R.C (1985) :Food additives in fennema .cited in :Food additives intolerance in child hood .P.179.Ed .David , T.J. Blackwell scientific. London-Boston.
44. London, W.T.; Henderson, W. and Cross, R. F. (1967): An attempt to produce chronic nitrite toxicosis in swine. J. A. V. M. A., 150 (4): 398 – 402.
45. Mackenzie , K.M, Boysea, B.G, Field, W.E, Petseel, S.A.W, chappel,C.I, Enerson,J.L AND STAALEY ,J (1992): Toxicity studies of caramel colour 111 and 2-acetiyl –4 (5)-tetrahydroxybutylimidazole in F344 rats. Food and chemical toxicology. 30(5) : 417-425.
46. Maekawa, A.; Ogin, T.; Onodera, H; Furuta, K.; Matsuoka, C.; Ohho, Y. and Odoshima, S. (1982): Carcinogenicity studies of sodium nitrite and sodium nitrate in F – 344 rats. Fd. Chem. Toxic., 20: 25 – 33.
47. Meister, A.; S.S. and Groffith, O.W. (1981): Gammaglutamyl transpeptidase. Method Enzymol., 77: 237 – 253.
48. Morliere, P.; Moysan, A.; Santus, R.; Huppe; J.C. and Dubertret, L. (1991): UVA – induced lipid peroxidation in cultured human fibroblasts. Biochem. Biophy. Acta. 1084: 261 – 268.
49. Osman, M.A.; Afifi.; A.; Hussien, R.M.; Kamilia, B.; Abdel – Aziz and Salah, S.H. (1995): Long – term biochemical and genotoxicity studies of four synthetic food and drug colorants in mice. Bull. Fac. Pharm. (Cairo Univ.), 33 (1): 13 – 12.
50. Raabo, E. (1963): Determination of serum lactic dehydrogenas by the tetrazolium salt method. Scard. J. Clin. And Lab. Invest., 15: 233.
51. Rasekh, H.R.; Potmis, R.A.; Nonavinakere, V.K.; Early, J.L. and Iszard, M.B. (1991): Effect of selenium on plasma glucose of rats: role of insulin and glucocorticoides. Toxicol. Lett., 58 (2): 199 – 207.
52. Rastogi, P.B. and Prasad, O.M. (1983a): Haematological changes induced by Feeding a common food colour metanil yellow in albino mice. Toxicol. Lett., 16: 103 – 108.
53. Rastogi, P.B and Prasad, O.M. (1983b): Haematological abnormalities induced by prefeeding a common food colour metanil yellow in mice. Proc. Acad. Sci. India Sec. B., 53 (1): 1 – 10.
54. Ray Sarkar, B.C. and Chauhan, U.P.S. (1967): A new method for determining microquantities of calcium in biological materials. Anal. Biochem., 20: 155 – 166.
55. Reitman, S. and Frankel, S. (1975): A colourimetric method for the determination of serum glutamic oxaloacetic and glutamic pyrovic transaminases. Am. J. Clin. Path., 28: 56.
56. Reutov, V.P.; Sorokina, E.G; Pinelis, V.G; Korshunova, T. S.; Rodionov, A.A; Koshelev, V.B; Strukova, S.M; Kaiushin, L.P; Braquet, P. and Komissarova, L.K.H. (1993): The compensatory –

adaptive mechanisms in nitrite – induced hypoxia in rats. Biull. Eksp. Biol. Med., 116 (11): 506 – 508.
57. Rodak, F.P. (1995): Routin laboratory evaluation of blood cells and bone marrrow. In: Diagnostic hematology, pp.: 125 – 129, W. B. sounders Comp. Phild. London, Toronto, Montreal, Sydney, Tokyo.
58. Rodriguez – morona, P.A. and Tarazona, J.N. (1994): Nitrite – induced methemoglobin formation and recovery in rainbow trout at high chloride concentrations. Bull. Environ. Contam. Toxicol. 53 (1): 113 – 139.
59. Rojkin, M.L.; Olguin, D.E.; Mariani, M.C.; Drappo, G.A.y. and sosa, C.P. (1974): Proteinastotales del sureo: causes mas Frecuentes de error en la roaccion del Biuret Nuevo reactivo cupraol calino estable. Biog. Del. Atlantico, V. 1163 – 1193.
60. Rotruck, J. T.; pope, A.L.; Ganther, H.E.; Swanson, A.B.; Hofeman, D.G. and Hoekstra, W.G. (1973): Selenium: biochemical role as a compenent of glutathione peroxidase. Sience, 179; 588 – 590.
61. Rybczynska, M.; Hoffmann, S. and Goslar, J. (1996): Molecular changes in erythrocyte membranes induced by nitroimidazoles and radiation. Pol. J. Pharmacol., 48: 269 – 280.
62. Sato, T.y.; Ose; Ishikawa, T. and Sakai, T. (1979): Toxicolgical effect of selenium on fish. Colloq. On aquatic environment in pacific region Taipei (china): 184 – 192.
63. Sharma, S.D. (1989): Renal gross biochemistry of albino rat influen - ced by a common food color, Metanil yellow. J. Adv. Zool, 10 (2): 95 – 98.
64. Shelpov, V.; Chekulaev, V. and pasha – Zade, G. (1991): Factors within the body determining the glycogen reserves in the tissues of rats. Biomed. Sci., 2 (2): 111 – 120.
65. Shen, H.M., Shi, C.Y.; Lee, H.P. and Ong, C.N. (1991): Aflatoxin B1 – induced lipid peroxidation in rat liver. Toxicol. Appl. Pharmacol., 127 (1): 145 – 150.
66. Siest, G. and Schielf, M.J. (1981): Interpretation Des Examens De laboratoire, Karger ed., pp.: 206 – 223.
67. Smith, R.P. (1991): Chemicals reacting with various forms of hemoglobin: biological significance, mechenisms and determination. J. Forensic Sci., 36 (3): 662 – 672.
68. Sokal, R.R. and Rohif, F.J. (1981): Biometry: The principles and practice of statistics in biological research. 2nd ed. Freeman, W.H. Company San. Francisco.
69. Standberg, A.S. (1977): Nitrate and nitrite supply and metabolism in man. (Abstract) Nutr. Abs. Revs. Ser (A), 47: 1119.
70. Thorpe, W.V; Bary, H.G. and James, S.P. (1964): "Biochemistry for medical students" 8th Ed. J. and A. churchill L.T.D, London, P. 80.
71. Tietz, N.W. (1986): Text book of clinical chemistry, W.B. Saunders Co., London, Philadelphia.
72. Til, P.; Falke, H.E.; Kuper, C.F. and william, M. I. (1998): Evaluation of the oral toxicity of potasium nitrite in a 13 – week drinking water study in rats. Fd. Chem. Toxic., 26 (10): 851 – 859.
73. Van kampen, E. J. & Zulstra, W.G. (1961): standerdization of hemoglobinometr: The hemoglobin cyanide method. Clin. Chem. Acta.: 538 – 540.

74. Walker, B. S; Boyd, W.C. and Asimov, I. (1957): Biochemistry and human metabolism. Williams & Wilkins – Baltimore, New York.
75. Webster, D. (1977): Albumin stardards and measurement of serum albumin with bromochresol green. Clin. Chem., 23: 663 – 666.
76. Whitby, L.G.; Smith, A.F.; Beckell, G.J and Waker, S.W (1992): Liver diseases "In Lecture Notes on clinical Biochemistry". Blackwell scientific publication. 5th edition.
77. White, J.W. (1975): Relative significance of dietary sources of nitrate and nitrites. J. Agric. Food Chem., 23 (5): 886 – 891.
78. Whitley, R.J; Meikle, A.W; Watts, N.B. (1996): Endocrinology. In: Tietz N.W. Fundamantals of clinical chemistry. W.B. Sawnders Company, philadelphia, London, Tronto, 673 – 704.
79. Yang, A and Wang, F. (1991):Effect of sodium nitrite on myocardial glutathione peroxidase and protective action of vitamin E and Selenium. Biomed. Environ. Sci. 4 (4): 373 – 375.
80. Yanni, M.; Abdel – Dayem, S.M. and Abdel – Azim, B.H. (1991): Biochemical and Histological changes due to preservatives in rats. Egypt. J. Histol.; 14 (2): 431 – 440.
81. Yoshida, Y.; Hirose, M.; Takaba, K.; Dinura, J. and Ito,N. (1994): Induction and promotion of forestomach tumors by sodium nitrite in combination with ascorbic acid or sodium ascorbate in rats with or without N – methyl – N – nitro – N – nitrosoguanidine pretreatment. Int. J. Cancer, 56: 124 – 128.

Table (1): The change in body weight gain and in the ratio of organ weight/ body weight of control, treated with NaNO3 + S.S.Y and treated with NaNO3 + S.S.Y + selenium after experimental and recovery periods in albino rats.

Parameter		Treated period (1 month)			Recovery period (15 days)		
		Control	$NaNo_3$ and sun – set yellow	$NaNo_3$ + (s.s.y) + selenium	Control	$NaNo_3$ + sunset yellow (s.s.y)	$NaNO_3$ + (S.S.Y) + selenium
% of body weight	**X** **S.E** **P**	**19.6** **.26**	**11.9** **0.69** **<.01**	**16.2** **.5** **<.01**	**18.94** **.4**	**10.6** **.4** **<0.01**	**16.4** **0.6** **<.01**
Kidney/b.wt	**X** **S.E** **P**	**.7** **.02** —	**.5** **.02** —	**.6** **.02** —	**.6** **.03**	**.5** **.02** —	**.6** **.04** —
Brain b.wt	**X** **S.E** **P**	**.86** **.03** —	**.86** **.03** —	**.89** **.03** —	**.9** **.02**	**.85** **.04** —	**.9** **.02** —
Cardio – somatic index	**X** **S.E** **P**	**0.6** **.02**	**.4** **.02** —	**.5** **.03** —	**.62** **.04**	**.45** **.02** —	**.5** **.03** —
Hepato – somatic index	**X** **S.E** **P**	**3.2** **.09**	**2.5** **0.1** —	**2.5** **.09** —	**3** **.1**	**2.9** **.1** —	**3.8** **.09** —

Gonado – somatic index	**X** **S.E** **P**	**1.07** **.02**	**.9** **.05** —	**.8** **.04** —	**1.1** **.02**	**.8** **.05** —	**.9** **.03** —

Table (2): The effect of sodium nitrite and sun – set yellow (10 mg $NaNO_3$ and 0.5 mg S.S.Y / kg) and selenium (50mcg/kg) on rectal temperature, respiratory rate and heart beats of albino rats after experimental and recovery periods.

Parameter		Treated period (1 month)			Recovery period (15 days)		
		Control	$NaNO_3$ + S.S.Y	$NaNO_3$ + (S.S.Y) + selenium	Control	$NaNO_3$ + (S.S.Y)	$NaNO_3$ + (S.S.Y) + selenium
Respiratory rate (breath/ min)	**X** **S.E** **P**	**49** **1.8**	**51** **.5** —	**48** **1** —	**48.2** **1.2**	**48.6** **.9** —	**46.4** **.4** —
Heart beat/ min	**X** **S.E** **P**	**136** **1.8**	**139.6** **.7** —	**135** **1.5** —	**134** **1.8**	**133.6** **1.1** —	**129** **2.4** —
Rectal temperatures °C	**X** **S.E** **P**	**34.72** **.09**	**34.52** **.7** —	**34.84** **.3** —	**34.64** **.15** —	**33.94** **.4** —	**34.24** **.2** —

Table (3): The effect of sodium nitrite(10mg/kg) and sun – set yellow (0.5 mg/kg) and selenium (50mcg/kg) on red blood cells (RBCs), while blood cells (WBCs), hemoglobin concentration (HB%) and hematocrit value (Hct %) of albino rats after experimental and recovery periods.

Parameter		Treated period (30 days)			Recovery period (15 days)		
		Control	$NaNO_3$ + S.S.Y	$NaNO_3$ + (S.S.Y) + selenium	Control	$NaNO_3$ + (S.S.Y)	$NaNO_3$ + (S.S.Y) + selenium
W.B.Cs X 10^3	**X** **S.E** **P**	**8.5** **.2**	**6.9** **.1** **<0.1**	**8.22** **.1** —	**8.82** **.05**	**7.16** **.1** **<.01**	**8.7** **.1** —
R.B.Cs X10^6	**X** **S.E** **P**	**5.88** **.05**	**4.9** **.05** **<.01**	**5.82** **.06** —	**5.9** **.07**	**5.58** **.16** —	**5.8** **.08** —
H b%	**X** **S.E** **P**	**16** **.1**	**13.78** **.1** **<.01**	**15.64** **.19** —	**15.82** **.1**	**14.2** **.2** **<.01**	**15.72** **.2** —

Hct %	X S.E P	41.4 .9	36.8 .5 <.01	40.6 .6 ___	42 .9	37.2 .5 <.01	40.8 .3 ___

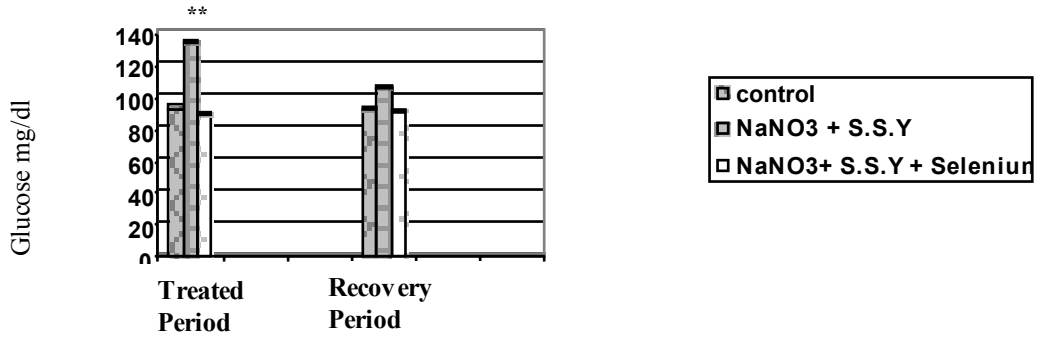

Fig (1): Glucose concentration (mg/ dl) in control, mixture treated and (mixture treated and selenium administration) rats after treatralt and recovery periods.

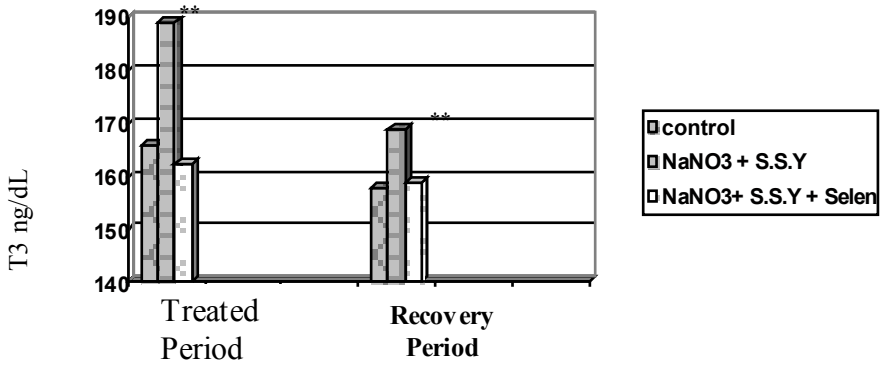

Fig (2): Serum T3 ng/dl treated with a mixture, a mixture and selenium and their control after treated and recovery peroids.

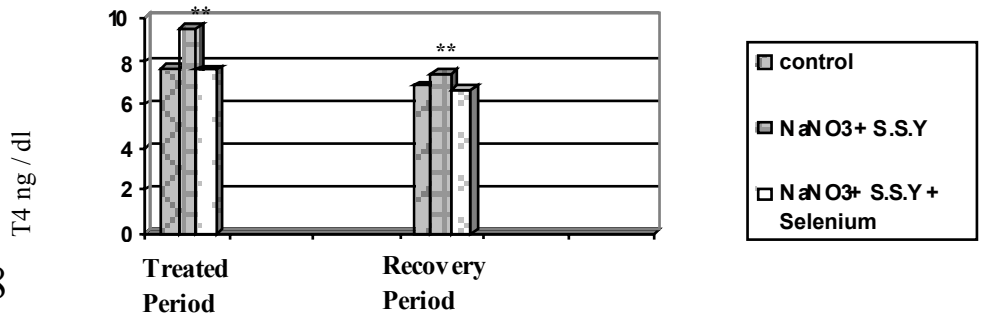

Fig (3): Serum T4 ng/dl in rats treated with a mixture, and selenium and their control after treated and recovery periods.

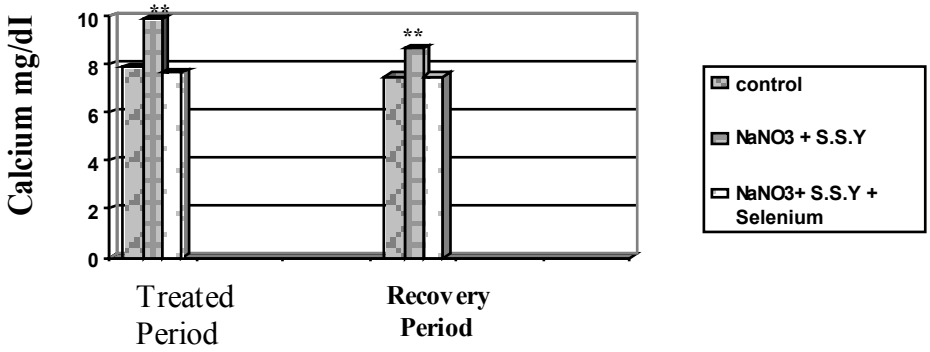

Fig (4): Serum calcium (mg/dl) in rats treated with a mixture, a mixture and selenium and their control after treated and recovery periods.

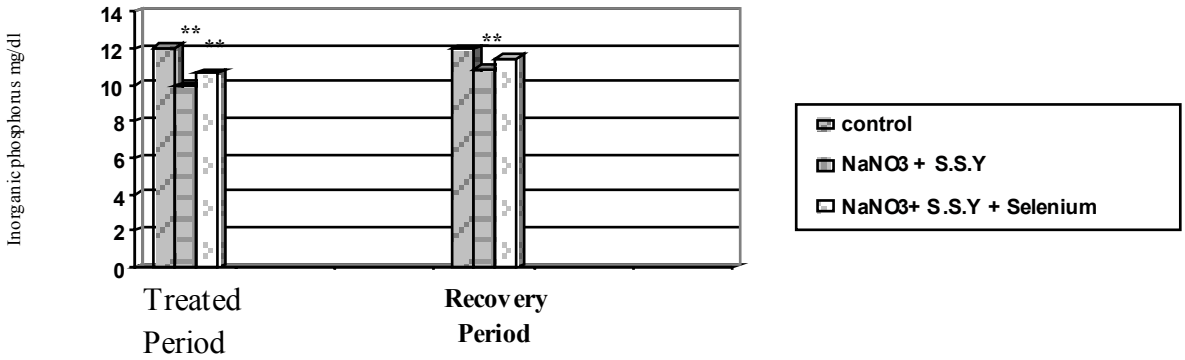

Fig (5): Serum inorganic phosphorus (mg/dl) in rats treated with a mixture, a mixture and selenium and their control after treated and recovery periods.

Table (4): AST activity in serum (U/L) and tissues (brain, liver, muscle, kidney and heart (U/ gm) in control, a mixture treated and a mixture and selenium treated rats after experimteal and recovery poiods.

Parameter		Treated period (30 days)			Recovery period (15 days)		
		Control	$NaNO_3$ + S.S.Y	$NaNo_3$ + (S.S.Y) + selenium	Control	$NaNO_3$ + (S.S.Y)	$NaNO_3$ + (S.S.Y) + selenium
Serum AST (U/L)	X S.E P	23.8 .7	37 1.2 normal - value	22.8 1.1 normal - value	21.2 1	27.4 1 normal - value	20.6 .9 normal - value
Brain AST U/g	X S.E P	15 .4	24.4 .9 normal - value	15.4 .5 normal - value	14.0 .7	21.6 .5 normal - value	14.0 .07 normal - value
Liver AST U/g	X S.E P	18.0 1.1	26.2 .5 normal - value	17.8 .7 normal - value	17.2 .8	17.2 1.1 normal - value	15.8 .9 normal - value
Muscle AST U/g	X S.E P	16.6 1.4	21.0 1.1 normal - value	16 1.4 normal - value	16.8 1.7	14.8 .9 normal - value	13.5 .6 normal - value
Kidney AST U/g	X S.E P	18.8 1.2	20.3 1.5 normal - value	16.0 .3 normal - value	17.0 .7	16.6 1.6 normal - value	15.2 0.9 normal - value
Heart AST U/g	X S.E P	13.4 .5	20.6 1.4 normal – value	14 .2 normal - value	13.8 0.9	11.4 .7 normal - value	14.6 0.9 normal - value

Table (5): ALT activity in serum, brain, liver, muscle, kidney and heart in a mixture treatment, mixture and selenium treatment and their control after treated and recovery periods.

Parameter		Treated period (1 month)			Recovery period (15 days)		
		Control	$NaNO_3$ + S.S.Y	$NaNo_3$ + (S.S.Y) + selenium	Control	$NaNO_3$ + (S.S.Y)	$NaNO_3$ + (S.S.Y) + selenium
Serum ALT U/L	**X S.E P**	**22.2 1**	**28 1**	**23.8 .7 ———**	**17.8 .5**	**28.4 1.1 ———**	**18.6 .4 ———**
Brain ALT U/g	**X S.E P**	**14 .7**	**23.2 2 ———**	**14.8 .4 ———**	**14.2 .5**	**19 .4 ———**	**14.2 1.4 ———**
Liver ALT U/g	**X S.E P**	**16.4 .5**	**24.2 2 ———**	**17 .7 ———**	**15.2 .6**	**21.2 .6 ———**	**17.8 .6 ——**
Muscle ALT U/g	**X S.E P**	**18.6 .2**	**24.2 1.1 ———**	**16 .7 ———**	**16.4 1.2**	**21.4 .4 ———**	**14 .7 ———**

Kidney ALT U/g	X S.E P	16 .7	20.6 .4 ---	14.8 .9 ---	15.2 .9	17.4 .9 ---	16.6 1.2 ---
Heart ALT U/g	X S.E P	18.8 .6	21.4 .8 ---	15 .3 ---	16.4 1.2 ---	18.6 .5 ---	13.2 1.7 ---

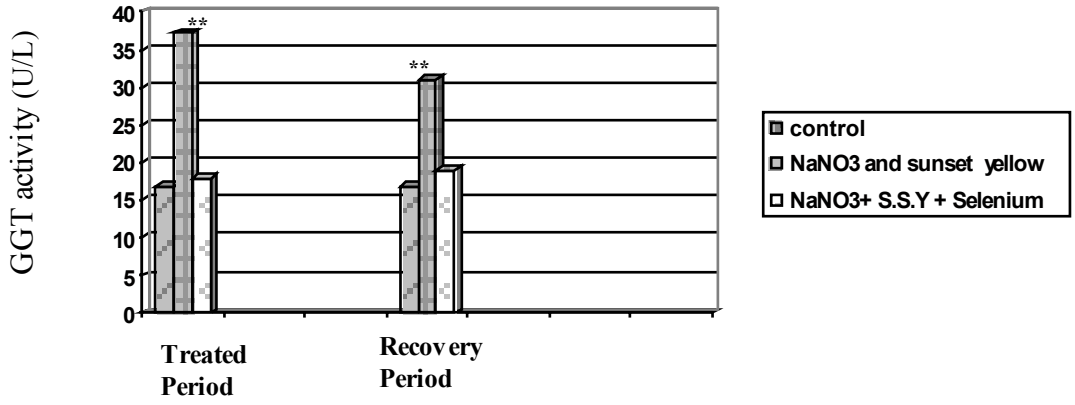

Fig (6): Serum α - GT activity (U/L) in control, a mixture treated and a mixture and
selenium treated rats after treated and recs very periods.

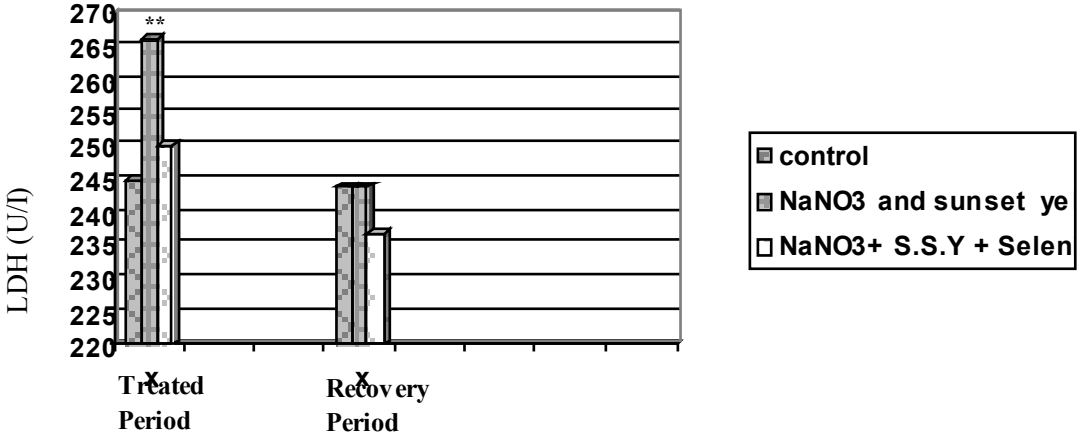

Fig (7): Serum LDH activity (U/l)in rats treated with a mixture, a mixture and seleavum and their control after treated and recovery periods.

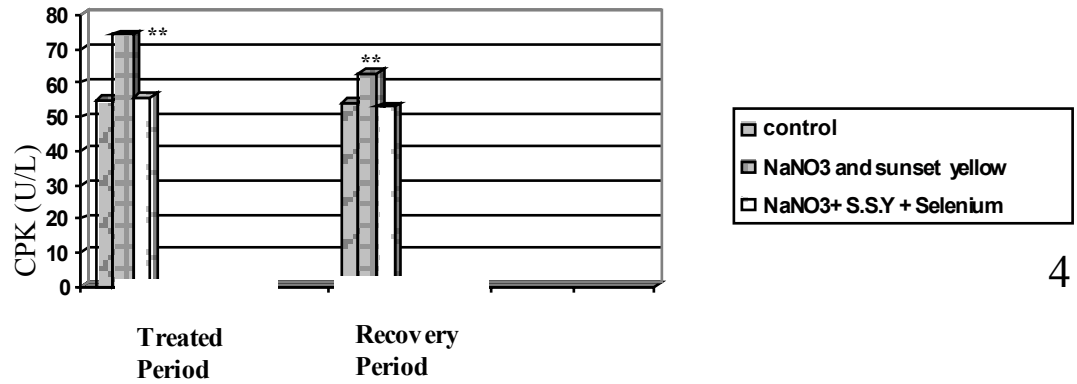

Fig (8): Serum CPK activity (U/L) in rats treated with a mixture, a mixture and selenium and their control after treated and recovery periods.

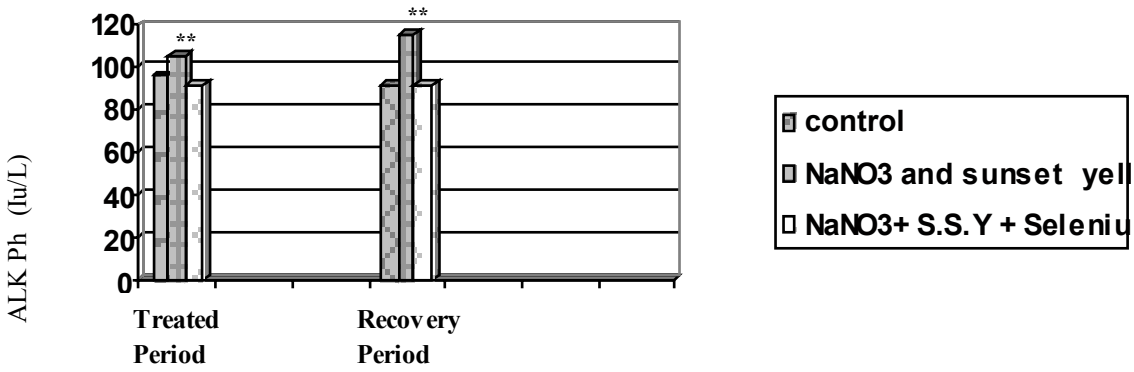

Fig (9): Serum MK. Ph activity (Iu/L) in rats treated with a mixture, amixture and selenium and their control after treated and recovery periods.

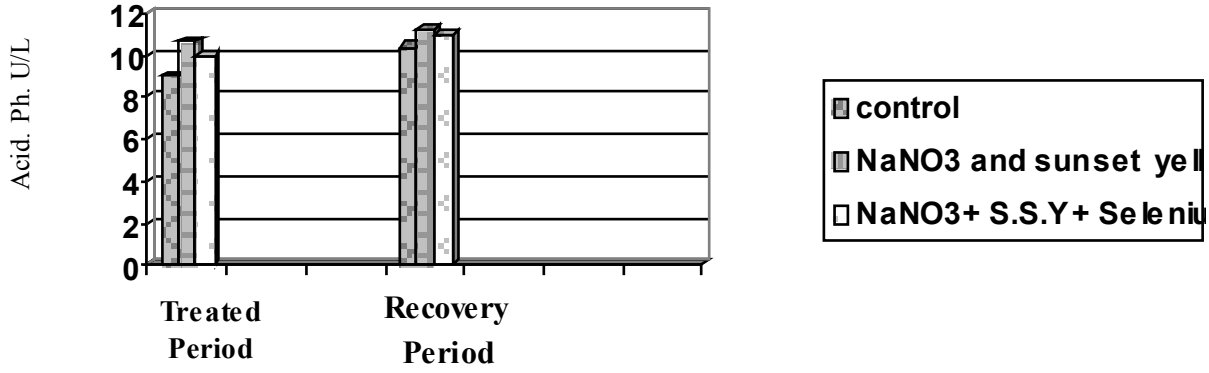

Fig (10): Serum Acid. Ph. Activity (U/L) in rats treated with a mixture, a mixture and selenium and their control after treated and recovery periods.

Table (6): Total proteins (serum, brain, liver, muscle, kidney and heart), serum (albumin and globulin) an A/G in control, a mixture treated and a mixture and selenium treated rats after treated and recovery periods.

Parameter		Treated period (30 days)			Recovery period (15 days)		
		Control	$NaNO_3$ + S.S.Y	$NaNO_3$ + (S.S.Y) + selenium	Control	$NaNO_3$ + (S.S.Y)	$NaNO_3$ + (S.S.Y) + selenium
Serum protein g/dl	**X** **S.E** **P**	**8.34** **.3**	**6.48** **.03** **<.01**	**7.7** **.2** ___	**7.9** **.16**	**7.78** **.19** ___	**7.4** **.2** ___
Brain protein mg/g	**X** **S.E** **P**	**110.4** **.6**	**110** **.7**	**112.8** **1.1** ____	**110** **.7**	**110.5** **.8** ___	**110** **.7** ___
Liver protein mg/g	**X** **S.E** **P**	**56** **1.8**	**55** **2.2** ___	**53** **1** ___	**50** **3.5**	**53.6** **2.4** ___	**52.6** **3.5** ___
Muscle protein mg/g	**X** **S.E** **P**	**92** **1.2**	**90.8** **.9** ___	**92.2** **.1** ___	**92.4** **.4**	**91.4** **.9** ___	**92.4** **1.2** ___
Kidney protein mg/g	**X** **S.E** **P**	**68.4** **.9**	**64.6** **2.1** ___	**68** **.7** ___	**67.6** **1**	**64.2** **1.5** ___	**68** **.7** ___
Heart protein mg/g	**X** **S.E** **P**	**88** **.9**	**85.2** **1.5** ___	**86.2** **.5** ___	**87** **1.1**	**88.6** **.4** ___	**89.2** **.4** ___

Serum albunin g/dl	**X** **S.E** **P**	**5.54** **.1**	**4.22** **.1** **<.01**	**5.7** **.17** ___	**4.9** **.14**	**4.84** **.09** ___	**5.2** **.2** ___
Serum globulin g/dl	**X** **S.E** **P**	**2.71** **.13**	**2.14** **.17** ___	**2.2** **.17** **<.05**	**3.1** **.1**	**2.6** **.18** **<.05**	**2.2** **.2** **<.01**
A/G	**X** **S.E** **P**	**2.04** **.1** ___	**1.98** **.18** ___	**2.68** **.25** ___	**1.58** **.1**	**1.76** **.22** ___	**2.66** **.1** **<.01**

Table (7): Total lipids (serum, brain, liver, muscle, kidney and heart), in control, a mixture treated, a mixture and selenium treated rats after experimental and recovery periods.

Parameter		Treated period (30 days)			Recovery period (15 days)		
		Control	$NaNO_3$ + S.S.Y	$NaNO_3$ + (S.S.Y) + selenium	Control	$NaNO_3$ + (S.S.Y)	$NaNO_3$ + (S.S.Y) + selenium
Serum total lipids mg/g	**X** **S.E** **P**	**388** **4.8**	**390** **5.1** ___	**398** **3.7** ___	**382** **7.3**	**388** **7.3** ___	**378** **5.8** ___
Brain total lipids (mg/g)	**X** **S.E** **P**	**49** **1.8**	**58** **4.1** ___	**56** **3** ___	**51.6** **2.6**	**52** **2.5** ___	**49.6** **1.6** ___
Liver total lipids (mg/g)	**X** **S.E** **P**	**92.6** **2.8**	**99** **2.4** ___	**96** **1.6** ___	**92** **2.5**	**96** **1.4** ___	**91** **1.1** ___
Muscle total lipids (mg/g)	**X** **S.E** **P**	**46.4** **2.7**	**56.2** **4.1** ___	**51.4** **4.6** ___	**45** **2.8**	**47** **1.2** ___	**44.6** **1.6** ___
Kidney total lipids (mg/g)	**X** **S.E** **P**	**44.4** **1.5**	**53.2** **3.8** ___	**46.6** **1.2** ___	**45** **1.4**	**48.2** **1.3** ___	**43.4** **1.8** ___

Heart total lipids (mg/g)	**X**	**53.6**	**55.8**	**49.5**	**49.6**	**44.8**	**43.6**
	S.E	**1.8**	**2.3**	**1**	**2.2**	**5.2**	**2**
	P		**___**	**___**		**___**	**___**

Table (8): Total cholesterol (serum, brain, liver, muscle, kidney and heart),and serum triglycerides in control, a mixture treated, and mixture and selenium treated rats after experimental and recovery periods.

Parameter		Treated period (30 days)			Recovery period (15 days)		
		Control	$NaNO_3$ + S.S.Y	$NaNO_3$ + (S.S.Y) + selenium	Control	$NaNO_3$ + (S.S.Y)	$NaNO_3$ + (S.S.Y) + selenium
Serum total cholesterol mg/dl	**X**	**131**	**150**	**122**	**129**	**136**	**129**
	S.E	**4.5**	**4.0**	**8**	**7.8**	**4**	**4**
	P		**<.05**	**___**		**___**	**___**
Brain cholesterol mg/g	**X**	**34.4**	**46**	**38.4**	**33.8**	**37.4**	**37.2**
	S.E	**1.6**	**2.1**	**1.4**	**1.2**	**2.2**	**1.1**
	P		**<.01**	**___**		**___**	**___**
Liver cholesterol mg/g	**X**	**28**	**39.2**	**30**	**30.8**	**26.4**	**27.6**
	S.E	**.9**	**1.6**	**.9**	**1.8**	**1.5**	**1.8**
	P		**<.01**	**___**		**___**	**____**
Muscle cholesterol mg/g	**X**	**17.2**	**21.8**	**18**	**14.2**	**15.2**	**16.2**
	S.E	**.9**	**2.5**	**.7**	**.9**	**1.1**	**1.2**
	P		**___**	**___**		**___**	**___**
Kidney cholesterol mg/g	**X**	**14.4**	**16.4**	**16.2**	**13**	**17.6**	**15**
	S.E	**.7**	**.7**	**.9**	**.9**	**2.1**	**.8**
	P		**___**	**___**		**___**	**___**

Heart cholesterol mg/g	X S.E P	18.4 .8	23.8 1.2 <.01	17.6 1.3 ___	17 .9	21 1.8 ___	16.6 1.8 ___
Serum triglycerides mg/dl	X S.E P	98 3.7	118 5.8 ___	104 4 ___	90.8 1.7	99 3.5 ___	95 1.8 ___

التأثيرات المتطورة للتفاعل بين نترات الصوديوم و صن ست الأصفر على مختلف المعايير الفسيولوجية فى الفئران البيضاء

د/إيمان جمال الدين عزت هلال
قسم علم الحيوان - كلية العلوم للبنات – جامعة الأزهر

لقد لوحظ ان الأطفال المصريين عادة يأكلون ويشربون مأكولات تحتوى على مواد حافظة و مـواد ملونة في نفس الوقت .ولقد جذب هذا السلوك الآنتباه لدراسة التفاعل بين أحد المـواد الحافظـة (نتـرات الصوديوم) واحد المواد الملونة (صن ست الأصفر) ولقد وجد ان خلط النسبة المسموحة لكلا من هـاتين المادتين قد ادى الى جرعة مميتة لذلك تم استخدام 10/1 من هذه الجرعة على الفئران لمدة 30 يوم .

قسمت الحيوانات إلى ثلاث مجموعات. استخدمت المجموعة الأولى كمجموعة ضابطة بينما جرعت المجموعة الثانية خليط من (10 مجم نترات الصوديوم و 0.5 مجم /كجم /يوم صن ست الأصفر) اما المجموعة الثالثة فلقة تناولت (5 مجم /كجم) من السيلينيوم بألاضافة للخليط السابق. واستمرت المعاملة لمدة 30 يوم ثم ذبحت نصف الجميع وترك النصف الاخر بدون اى معاملة للاستشفاء فترة 15 يوم وقد ادى تجريع الخلـيط (نترات الصوديوم وصن ست الاصفر) الى نقص معنوى فى وزن الجسم ،وعدد كرات الدم الحمراء و البيضاء ونسبه الهيموجلوبين و الهيماتوكريت و الفوسفورالغير عضوى وبروتين والبيومين المصل بينما لوحظت زيادة معنوية فى جلوكوز المصل ،وهرمونات الغدة الدرقية (T4 ، T3) والكالسيوم و GGT، LDH، CPK , وانزيم الفوسفاتيز القاعدى و الكولسيترول. كذلك ارتفع مستوى الكولستيرول فى المخ والكبد والقلب بينما لم يتأثر نسبة وزن الاعضاء لوزن الجسم ومعدل التنفس وضربات القلب ودرجة حرارة الجسـم ونشـاط

انزيمىAST، ALT فى المصل و الأنسجة ونشاط انزيم الفوسفاتيز الحامضى و الدهون الكلية فى المصـل و الأنسجة والكولستيرول فى العضلات و الكلى وكذلك الدهون الثلاثية فى المصل .

ولقد تم شفاء تام من اغلب هذه التغيرات فى معايير الدم و القياسات البيوكيميائية بعد فترة 15 يـوم مـن الاستشفاء او بعد تناول عنصر السيلينيوم .

وهذه الدراسة تجذب الانتباه للتفاعل الذى يحدث بين المواد الحافظة و الملونة عند تناولهم فى نفس الوقت كما توضح ان تناول النسب المسموحة من هذه الاضافات عندما تؤخذ سويا او بكميات زائدة تؤدى الى اضرار خطيرة .