

Structural and functional changes in the internal rat's organs under the influence of a food additive complex

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Abstract: The following article brings to attention the use of various Sodium based food additives; their biochemical, morphological and in part physiological impacts on the human body (brain, cardiovascular system, kidneys), especially emphasizing the direct and indirect effects on the adrenal glands. The use of food additives as preservatives, taste enhancers and coloring agents like MSG (monosodium glutamate), sodium nitrite and Ponceau 4R (tri-sodium salt) are now studied for their detrimental health effects on humans. Most commonly faced problems with use of additives are elevated blood pressure, obesity (by interruption of the leptin-mediated signalling pathway of the hypothalamus and increasing appetite), impaired glucose tolerance and insulin resistance to peripheral glucose uptake leading to increased levels in blood consequently affecting the brain (diabetic neuropathy, vascular dementia). In the brain, prolonged elevated levels of glutamate (excitatory neurotransmitter) can overexcite metabotropic and ionotropic receptors resulting in death of nerve cells. Ponceau 4R or E124 is banned in countries like USA and Canada for its carcinogenic effects. The issue arises in the level of usage i.e. The quantity of these additives, which can cause clinical, morphological and physiological impacts on the human body, especially adrenal glands. As the endogenous nitrites are cytoprotective but excess of nitrites can lead to oxidative stress; thus leading to impaired action of the hypothalamus-pituitary-adrenal axis (in the mammalian experiment), showing stunted growth, slow or non-functional neonatal brain development; these similarities are comparable with those in humans as observed in PTSD (post-traumatic stress disorder).

Index Terms: monosodium glutamate, sodium nitrite, Ponceau 4R, food additives, adrenal glands.

Introduction

In Asian countries, food additives are used ubiquitously, primarily MSG, also known as “Chinese salt” and Ponceau 4R. They became popular due to their characteristic “umami” or savory taste [1] and the bright red color it imparted to Asian curries, increasing palatability. China is the most significant producer and consumer of these additives [2]. Sodium nitrite is a widely used preservative in cured or processed meats, which turn into nitrosamines, a carcinogen associated with gastric cancer, breast cancer and prostate cancer (E Chazelas et al.2021) [3]. Its common signs are cyanosis, hypoxia, bradycardia and hypotension resulting from methemoglobinemia. Hypoxia induces catecholamine secretion from the carotid body (glomus cells) and brain; however, no significant action by adrenal chromaffin cells are produced [4]. Ingestion of MSG orally did not alter pituitary hormones other than ACTH and prolactin [5]. However, in neonates and under prolonged hypoxic conditions, It directly acts on chromaffin cells to release catecholamines even before complete formation of splanchnic nerves to cope with stress. Some researchers found that glutamate from the outside brain cannot cross the blood-brain barrier and have no profound effect unless given in supra-physiologic doses [6]. Circumventricular organs are the area with no blood-brain barrier thus is susceptible to increased glutamate levels. MSG is responsible for many clinical manifestations like headache and nausea via peripheral N-methyl-D-aspartate receptors’ activation [7], reproductive toxicity [8], decreased activity of anti-oxidants as well as increased lipid peroxidation can cause renal toxicity in case of chronic consumption of MSG, may produce reactive oxygen species (ROS) resulting in oxidative stress [9].

Neuro-endocrine mechanism of stress control

The pituitary-adrenal axis is activated during stress [10] stimulates the release of corticotrophin-releasing hormone (CRH) from the hypothalamus, further stimulating the anterior pituitary to release ACTH and posterior pituitary to release vasopressin; these act on adrenal cortex and medulla respectively; releasing cortisol (ADH action mediated by V1a and V1b receptors present on adrenal cortex and medulla respectively) [11, 12].

Theoretically, an excessive amount of glutamate must have a hindering impact on brain cells, however in practicality (in vivo), bodily mechanisms inhibit this in three possible ways; preventing dietary glutamate from entering circulation from GIT (gastrointestinal tract), blood-brain barrier prevents entering of glutamate from the circulation into the brain, glutamate transporters on synaptic membranes keeps it balanced. Hence, no concrete evidence could be produced, succumbing to neurodegenerative disease-causing effects of MSG. But, does that eliminate the possibility of glutamate causing neurodegenerative disease? Chronically elevated glutamate levels, though moderated, tend to prompt neurodegenerative diseases [15].

Pathology

In an experiment on rats, MSG causes metabolic changes characterized by an increase in the ability of adipocytes to transport glucose and synthesize lipids, which leads to increased sensitivity to insulin [16]. Monosodium glutamate has been shown to cause damage to the nuclei of the medulla oblongata [17]. MSG causes degenerative and atrophic changes in the fundal part of the stomach

and Brunner's glands of the duodenum. Inflammation and necrosis of hepatocytes were observed in the liver. In addition, hypertrophy of epithelial cells lining bronchioles, deformation of type I pneumocytes, and partial loss of cytoplasmic organelles in type II pneumocytes were recorded.

Sodium nitrite (E250) is used as an antioxidant. Beneficial and antibacterial effect of E250: it prevents the growth of Clostridium botulinum bacteria, the causative agent of botulism. In the process of adding sodium nitrite, interacting with myoglobin, it forms nitrosomyoglobin, which gives meat products their characteristic "meat" color. However, in large quantities, E250 is very poisonous [19].

Food additive E250, when taken in a significant amount - about several grams may cause serious poisoning (forms methemoglobin), and lead to paralysis of the vascular centre and death. In addition, E250 can bind to blood cells and thus prevent oxygen delivery. Products with sodium nitrite are especially not recommended for children since the child's hemoglobin is the most susceptible.

Sodium nitrite is not a carcinogen but a very toxic substance capable of reacting with nitrosation to form nitrosamines and nitrosamides, many of which have carcinogenic properties. Eating foods with the E250 additive can cause severe thirst. It is not recommended to strongly heat products with preservative E250 because in the process of heating during the reaction of sodium nitrite with amino acids, carcinogens nitrosamines are formed - derivatives of ammonia, which act as substances that can lead to oncological diseases. This indicates the potential for cancerous changes when consuming products that have undergone heat treatment in the presence of sodium nitrite. Using E250 can lead to a sharp decrease in muscle tone and blood pressure. Irritation of the skin and mucous membranes occurs [19]. Penetration of this substance through intact skin is possible. Causes vomiting, loss of consciousness, prolonged contact with sodium nitrite causes skin irritation, swelling of the upper and lower extremities, reduced vision, bluing of the tips of the fingers, toes, end of the nose, and changes in the central nervous system, blood system, and liver. In addition, there is a connection between the use of sodium nitrite and the occurrence of migraine.

Ponceau 4R or E124 is a dye of synthetic origin with a red color. Food additive E124 is widely used in the confectionery industry for the attractiveness of various candies, cookies and cakes. E124 is also used in the production of different meat products. However, published scientific studies have shown a connection between this substance and the occurrence of tumors in laboratory animals. In addition, the E124 is an allergen and can cause harm by causing anaphylactic shock or an asthma attack in asthmatics and people with poor aspirin tolerance. In addition, Ponceau 4R is included in the list of dyes that provoke hyperactivity in children [20].

Rats injected with a large amount of sodium glutamate have lost the parallel arrangement of the cells of the fascicular zone. Most cells had darkly stained nuclei; their cytoplasm was excessive and pale with numerous lipid inclusions. Dilation and overflow of capillaries, a large number of macrophages lining the sinusoids were observed. Some cells had compressed apoptotic nuclei; their cytoplasm contained mitochondria and lipid inclusions. Macrophages contained many lysosomes. Statistically, the analysis of morphometric results showed a significant reduction in cortical thickness compared to the control group. In addition, it was confirmed that lipid peroxidation after oxidative stress leads to loss of membrane integrity and cell degeneration. Analysis of the oxidative stress of spermatozoa contributes to changes in protein properties. Thus, it affects receptor function, enzymes, antibodies, and transport proteins and causes changes in DNA.

E621 has been reported to have a direct toxic effect on cell function, caused primarily by an imbalance in the homeostasis of cysteine, a glutathione precursor, leading to depletion of intracellular glutathione levels and a decreased ability of cells to protect themselves from oxidative reactions and damage. Thus, oxidative stress and the accumulation of free radicals cause monosodium glutamate toxicity.

It was noted that the use of monosodium glutamate caused endocrine disorders, changes in immune reactions, and dysfunction of lymphocytes and macrophages in particular.

Histomorphometry, performed by calibration with an ocular micrometer on the kidney tissue of the control and experimental groups, revealed significant differences in glomeruli with increased length and Bowman's capsule size with increased Bowman's space.

This study showed that the toxic effect of MSG on renal glomeruli was focal and segmental. Glomerular compression, increased cell proliferation, exudation of capillary contents with obliteration of their lumen and possible hyalinization were noted [21]. Renal tubules showed patchy turbid edema, necrotic cells with nuclei of karyolysis and cellular debris seen in the lumen. The experimental group's morphometric measurements (increased glomerular dimensions, Bowman's capsule and space) were also significantly different from the control group. Renal proximal convoluted tubules were also more affected than other tubules with inflammatory infiltration and focal hemorrhagic areas [22].

Wide vacuolization of the cells of the fascicular zone in combination with expanded and congested sinusoids also indicates hyperactivity of the cells. In addition, the ratio of the width of the fascicular zone to the glomerular zone was comparatively increased in rats treated with monosodium glutamate.

Rats treated with E621 had characteristic features such as growth retardation, obesity, and in many cases tail amputation [23].

In an experiment on newborn rats from mothers who used sodium nitrite during pregnancy, a chaotic arrangement of tubules was observed in the kidneys, and in some cases exfoliated cells were present [24]. The intake of NaNO₂ by females affects the dynamics of NO-synthetase activity in the kidneys, at first, it exceeds the control data, and then quickly decreases. Experimental animals have more stroma and visually fewer glomeruli.

The fur of animals that used the Ponceau 4R supplement was dyed pink and the gastrointestinal tract pink. In addition, such changes as the liver and thymus in punctate inclusions, kidney stones, and enlarged cecum were sometimes observed.

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