HISTOPATHOLOGICAL EFFECT OF SUNSET YELLOW IN ALBINO MICE TREATED WITH VITAMIN E

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Abstract

The objective of the present study was to evaluate the toxic effect of Sunset yellow on the histological state of internal organ and weighted change of mice. Forty adult Swiss Albino mice at the age of two months were divided into four groups. The 1st group (G1) was administrated orally with (30 mg/kg b.w) of Sunset yellow daily for 8 weeks, 2nd group (G2) was administrated vitamin E (15 I.U./kg) for 8 weeks. 3rd group (G3) was administrated of sunset yellow and vitamin E daily for 8 weeks. 4th group (G4) was considered as control negative group. At end of the experiments (8 weeks), all animals were sacrificed and Specimens were taken from internal organs like, kidney, spleen, heart, intestine and brain. The tissues were kept in 10% formaldehyde solution, for fixation and then processed routinely by using the histokinete. Tissue sections were embedded in paraffin blocks and sectioned by microtome and stained with hematoxylin and eosin stain, then examined by using light microscope. The pathological lesions showed that the animals exposed to toxic dose of Sunset yellow was characterized by inflammatory reaction, hemorrhage, congested blood vessels, necrosis, fibrosis and multiple granuloma lesions in internal organs and proliferation Astrocyte and gliosis in the brain while less lesions were recorded in the groups treated with vitamin E showed improvement against toxic effect of Sunset yellow.

Key words: Histopathology, kidney, Sunset yellow, Vit E.

Introduction

Food additives are defined as chemical substances deliberately added to foods, directly or indirectly in known quantities for purposes of assisting in the processing of foods; preservation of foods; or in improving the flavor, texture, or appearance of foods (Daniel, 2007).

Sunset yellow (molecular weight 452.36) is an azo dye, is orange yellow in color and is permitted food color in India. It is extensively used in almost every type of food preparation like sweets, jams and jellies, soft drinks, candies, ice cream, canned juice, sauces, pickles and so forth. In the past few years, use of some food dyes including sunset yellow was banned in United States and Japan owing to its mutagenicity which has been evidenced from several mammals bioassays (Ching et al., 2005; Feng et al., 2012).

Investigated the acute in vivo histological effects of Egg Yellow, which is a mixture containing Sunset Yellow FCF, by determining histopathological changes in some rat tissues. The colour was orally administered (dissolved in 5 ml distilled water) to rats at doses of 500, 1000, or 2000 mg/kg B.w/day for 3 days. Treated rats showed remarkable differences compared to controls. Gross examination of tissues revealed marked ulcerative lesions and haemorrhage on the antra of stomach of rats given the colourant at 2000 mg/kg B.w. Gross examination also revealed mild splenomegaly, hepatomegaly and enlarged pale kidneys in the rats administered the colourant at 1000 mg/kg B.w or 2000 mg/kg B.w Histopathological examination of sections from the kidney, spleen, stomach and ileum of rats treated with Sunset Yellow FCF revealed a variety of dose-related.

Materials and Methods

Forty adult male Swiss Albino mice and aged 8 weeks and weighed range (20-25g), supplied from animal house of the College of Vet. Med. University of Baghdad was used in present study. They were housed and maintained in a conventional animal facility, with controlled conditions of temperature (20 ± 5℃). The animals were fed on special formula of food pellets and given water ad libitum. Throughout the experiments, each group of mice was housed in plastic cage containing hard-wood chip as
bedding. The bedding was changed continuously to ensure a clean environment. The experiment is University of Baghdad College of Veterinary Medicine Department of pathology and poultry.

**Experimental Design**

Forty adult males Swiss Albino mice, aged 8 weeks, were divided into four groups. The 1<sup>st</sup> group (G1) was administrated orally with (30 mg / kg B.w) of Sunset yellow daily for 8 weeks, 2<sup>nd</sup> group (G2) was administrated vitamin E (150 I.U./kg) for 8 weeks. 3<sup>rd</sup> group (G3) was administrated of sunset yellow and vitamin E daily for 8 weeks. 4<sup>th</sup> group (G4) was considered as control negative group.

**Sunset yellow preparation**

Done by dissolving 300mg of sunset yellow in 100ml distal water to prepare concentration 3gm/ml to give at dose volume of 0.1ml/10g B.w and at dose of 30mg/KgB.w.

**Vitamin E preparation**

Optimal dose of Vitamin E α-tocopherole in mice (15 I.U./kg B.w.) (The Journal of Nutrition, 1982). 400 I.U. capsule Vitamin E completed to 40 ml with olive oil (stock solution) each 1ml of stock solution contain 10 I.U. of Vitamin E so 1.5 ml of stock solution contain 15 I.U. Vitamin E. This 1.5 ml completed to 10 ml by olive oil. Dosing volume 0.1 ml /10 gm B.W. mice including 0.15 I.U. Vitamin E.

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<th>Before</th>
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<tr>
<td>G1</td>
<td>33.60±0.72A</td>
<td>29.40±0.40B b</td>
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<tr>
<td>G2</td>
<td>33.30±0.67B</td>
<td>37.20±0.65A a</td>
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<tr>
<td>G3</td>
<td>34.40±0.76A</td>
<td>29.40±1.06B b</td>
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<tr>
<td>G4</td>
<td>32.50±0.65B</td>
<td>36.40±0.75A a</td>
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LSD=2.9; The different capital letters refer significant differences between times within one raw at (P<0.05); The different small letters indicate significant differences between the groups within one column at (P<0.05).

**Fig. 1:** Histopathological section in the kidney of animal treated with sunset yellow at 8 weeks post treatment shows haemorrhge, marked mononuclear cells aggregation in the interstitial tissue and degeneration of epithelial cells of renal tubules, hyaline droplets within the proximal and distal convoluted renal tubules. These structures appeared as deep eosinophilic round or oval in shape (H&E stain 40X).

**Fig. 2:** Histopathological section in the kidney of animal treated with sunset yellow at 8 weeks post treatment shows, marked mononuclear cells aggregation in the interstitial tissue and degeneration of epithelial cells of renal tubules, amyloid like substance deposition in congested red pulp (H&E stain 40X).

**Fig. 3:** Histopathological section in the lung of animal treated with sunset yellow at 8 weeks post treatment haemorrhge perivascular and peribronchiolar lymphocytic cuffing (H&E stain 10X).
Histopathology

Pieces of spleen, brain, heart, stomach, intestine and kidney were fixed in 10% normal buffer formalin for 72 hours for routine histopathological examination (Luna et al., 1968).

Result

Histopathological Changes Of Animals Treated With Sunset Yellow at 8 weeks Post treatment the main lesion in kidney characterized by haemorrhage, marked mononuclear cells aggregation in the interstitial tissue and large numbers of hyaline droplets within the proximal and distal convoluted renal tubules. These structures appeared as deep eosinophilic round or oval in shape show in fig. 1, as well as marked mononuclear cells aggregation in the

Fig. 4: Histopathological section in the spleen of animal treated with sunset yellow at 8 weeks post treatment shows severe haemorrhage and amyloid like substance deposition in congested red pulp (H&E stain 40X).

Fig. 5: Histopathological section in the brain of animal treated with sunset yellow at 8 weeks post treatment shows mononuclear cells aggregation severe dilation and congestion meningeal blood vessels (H&E stain 40X).

Fig. 6: Histopathological section in the myocardium of animal treated with sunset yellow at 8 weeks post treatment shows severe congestion of blood vessels with infiltration of inflammatory cells mainly neutrophils (H&E stain 40X).

Fig. 7: Histopathological section in the stomach of animal treated with sunset yellow at 8 weeks post treatment shows hyperplasia of mucosa and hyperkeratosis (H&E stain 40X).
interstitial tissue and degeneration of epithelial cells of renal tubules and amyloid like substance deposition in congested red pulp show in fig. 2. The main lesion in lung characterized by haemorrhge perivascular and peribronchiolar lymphocytic cuffing show in fig. 3. The main lesion in spleen characterized by severe haemorrhge and amyloid like substance deposition in congested red pulp show in fig. 4. The main lesion in brain characterized by mononuclear cells aggregation severe dilation and congestion of meningeal blood vessels Show in fig. 5. The main lesion in myocardium characterized by severe congestion of blood vessels with infiltration of inflammatory cells mainly neutrophils show in fig. 6. The main lesion in Stomach characterized by hyperplasia of mucosa and hyperkeratosis show in fig. 7. The main lesion in Intestine characterized by shows hemorrhage, severe fibrosis and mononuclear cells infiltrations of mucosa leading to damage of the mucosal glands show in fig. 8. Histopathological change in Of Animals Treated With vitamin E. The main lesion in kidney characterized by neutrophils and mononuclear cells aggregation around and in the lumen of congested blood with acute cellular degeneration in the epithelial lining cells of renal tubules

Fig. 8: Histopathological section in the intestine of animal treated with sunset yellow at 8 weeks post treatment shows haemorrhge, severe fibrosis and mononuclear cells infiltrations of mucosa leading to damage of the mucosal glands (H&E stain 40X).

Fig. 9: Histopathological section in the kidney of animal treated with vitamin E at 8 weeks post treatment shows neutrophils and mononuclear cells aggregation around and in the lumen of congested blood with acute cellular degeneration in the epithelial lining cells of renal tubules (H&E stain 40X).

Fig. 10: Histopathological section in the spleen of animal treated with vitamin E at 8 weeks post treatment shows moderated proliferation of lymphocytes and mononuclear cells in congested blood vessels and red pulp (H&E stain 40X).

Fig. 11: Histopathological section in the spleen of animal treated with vitamin E at 8 weeks post treatment shows proliferation of megakaryocytes and mononuclear cells in congested blood vessels and red pulp (H&E stain 40X).
show in fig. 9. The main lesion in spleen characterized by moderated proliferation of lymphocytes and mononuclear cells in congested blood vessels and red pulp show in fig. 10 proliferation of megakarocytes and mononuclear cells in congested blood vessels and red pulp show in fig. 11. The main lesion in lung characterized by mononuclear cells aggregation around blood vessels and peribronchiolar show in fig. 12.

Discussion

The study showed levels sunset yellow led to depress and showed decrease appetite for food consumption during the study period. The loss of body weight and anorexia significantly decrease (P<0.05) in G1 (sunset) and G3 (sun set and vit E) as compared to G2 vit E and control group. The loss of body weight and anorexia significantly decrease (P<0.05) in G1 (sunset) and G3 (sun set and vit E) as compared to G2 vit E and control group may be attributed to the effect of a synthetic azo dye “SY” on both true and pseudo-cholinesterases in rats, causing a significant decrease in these enzyme activities (Osman et al., 2004). This relationship between those enzymes (cholinesterases) and the behavioural symptom (apathy) were also indicated by (Mega et al., 2005), who published that the improvement in apathy was following administration of galantamine - a cholinesterase adjustment. vacuolization were observed in SY-treated kidney of the present study as a prominent symptom, may attributed to disturbance of ionic milieu of the cell with consequent retention of water and sodium leading to cellular swelling (Jaarsma et al., 2001; Wiedemann et al., 2002).

Thereupon the accumulation of ions and fluids in cytosol would rapidly pass through leaky membranes of cell vacuolated organelles and finally lead to cell lysis (Gores et al., 1990; Straus, 2001; Sarhan and Al-Sahaf, 2011). Interpreted such vacuolation as pathological changes of permeability, whereas phagosomes and lysosomes lost their normal location and fused to form large vacuoles or spheres in kidney and liver of peroxidase-treated rats in an azo dye medium. Furthermore, the clear cytoplasmic vacuoles contain predominantly electron-lucent material consistent with phospholipid in hepatic phospholipidosis of rats (Obert, 2007). The histopathological study by (Sharma et al., 2004), revealed severe degenerative changes in the liver, kidney and testes as a toxicity of another popular azo food dye blend ‘tomato red’ on male albino mice (Himri, 2011) showed tubular dilatation with thickened basement membrane, tubular degeneration and dilatation of the glomerular capillaries and atrophy of glomerulus. For both liver and kidney phenomena represented by hepatic impairment, edema, congestion and kidney apoptosis, with atrophy of renal corpuscles were observed. Degree and severity of histopathological aspects observed were directly proportional to the concentration of the administered dyes (Russ et al., 2000).

References


