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Original Article

Serological Changes Induced by Blend of Sunset Yellow, Metanil Yellow and Tartrazine in Swiss Albino Rat, *Rattus Norvegicus*

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ABSTRACT

Objective: The present study was carried out to evaluate the toxic effect of blend of some food colors on Swiss albino rats. **Materials and Methods:** A blend (1:1:1) of sunset yellow, metanil yellow and tartrazine showed additive effects on serological parameters which indicate that addition of these dye together in food stuff may give rise to more toxic effects than are produced by each dye individually. Animals were divided into four groups (I, II, III, and IV). First group was treated as control and respective group of animals received 25, 50 and 75 mg/kg body weight blend of food colors by gavaging up to 30 days. **Results:** The serological study showed a decrease in total protein and albumin and an increase in alkaline phosphatase, SGPT and total bilirubin. The results revealed that oral administration of these blend did not affect the body weight gain. **Conclusion:** The prolonged consumption of the blend may cause adverse effect on human health.

Key words: Metanil yellow, sunset yellow, Swiss rat and blend, tartrazine

INTRODUCTION

The human beings have been using color additive for a very long time to hide poor quality of spoiled food products and to provide attractive appearance to them. Many natural and synthetic food colors are used to enhance the appearance of food products, however 95% of those used today are synthetic because they are produced easily, cheaper and provide better coloration. According to food adulteration act (FDA) eight food colors in India are permitted to be used in the eatables in a limited quantity.[1] Out of which tartrazine and sunset yellow are the most widely used food colorant in cold drinks, sweets, bakery, ice creams, snack food, cereals, packaged soup etc., Metanil yellow is a non-permitted synthetic food color and being used extensively for coating turmeric and arhar dal. As the survey in urban and rural areas of Bareilly region had been done regarding the food habits among individual to know the extent of consumption of these synthetic color during normal meal, so the present study is planned to evaluate the toxicity of blend of these food colors on Swiss albino rat.

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MATERIALS AND METHODS

Experimental animal

Adult Swiss albino rats, 8-9 week old, weighing 90 ± 5 g were used for the present study. They were maintained under standard laboratory conditions. Animals were housed in the polypropylene cages and maintained under standard conditions i.e. 12 h light and dark cycle, 25 ± 3°C temperature, 40-60 relative humidity and were fed on low protein diet procured from Aashirwad food Ltd, Chandigarh, India. The water was given *ad libitum* to experimental animals.

Chemical used

All the chemicals, sunset yellow (C. I. 15985), tartrazine (C.

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I. 19140) and metanil yellow (C. I. 13065) were purchased from Otto Chemical Company Pvt. Ltd. Mumbai (India) for experiments. The blend was formed by mixing them in equal ratio and orally administered to experimental animals.

Experimental design

Animals were divided into four groups having six animals in each group. The animal of I group was treated as control and received only low protein diet during experiment. The animals of II, III and IV groups were fed on low protein diet mixed with 25, 50 and 75 mg/kg body weight/day dose of blend for 30 days. These groups served as experimental groups.

Parameter studied

Body weight

The percentage of body weight gain was calculated as follows.

$$\frac{\text{Mean final weight - mean initial weight}}{\text{Mean initial weight}} \times 100$$

Serum analysis

By decapitation of each animal, blood was collected in clean centrifuged tube placed at 4°C for 2-3 h and then centrifuged at 3000 rpm for 15 minutes and serum was separated and kept at –20°C till serological analysis was done.

Serum alkaline phosphatase was determined by the method of Kind and King.[2] Serum total bilirubin was measured by the method of Doumas *et al*.[3] Serum total protein was determined according to the method given by Lowry *et al*.[4]

RESULTS

Sub-chronic toxicity studies were conducted on Swiss albino rat. Feeding on the blend of food color at different doses i.e. 25, 50 and 75 mg/kg body weight for 30 days did not show mortality of rats.

Body weight

The mean body weight in control and experimental group is shown in Table 1. The blend of above-mentioned food colors caused an increase in the body weight with the different doses (Group II, III and IV). In group II, the animal gained more body weight i.e. 13.64% in comparison with the animal of groups III (11.32%) and IV (10.78%). In the control group, the percent gain in body weight was 14.01%.

Serological examination

Serological results after the treatment period revealed significant changes in the experimental group when compared with the control group. The total protein and albumin were decreased in all experimental groups (II, III and IV) when compared to control [Figures 1 and 2, Table 2].

The level of alkaline phosphatase was significantly increased in all experimental groups i.e. 160.2, 182, 201 U/L with respective doses when compared with control, i.e. 150.5 U/L [Figure 3].

The levels of total bilirubin and SGPT were increased significantly in the treated group with 50 and 75 mg/kg body weight when compared to control. The blend caused a decrease in serum bilirubin at low dose (Group II) while no

Table 1: Changes in body weight of albino rats fed with blend of sunset yellow, metanil yellow and tartrazine

Group	Body weight (grams) (mean±S.E.M.)		% Gain in body weight
	Initial	Final	
I	92±0.46	107±0.37	14.01
II	95±0.32	110±0.55	13.64
III	94±0.46	106±0.41	11.32
IV	91±0.42	102±0.53	10.78

S.E.M. = Standard error of mean

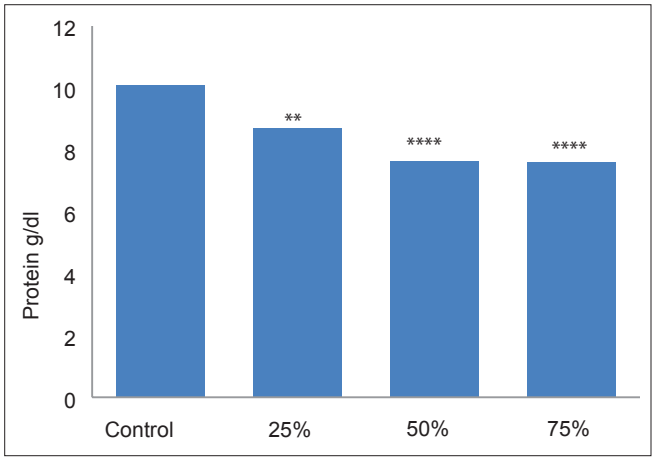


Figure 1: Effect of blend of food color on total protein

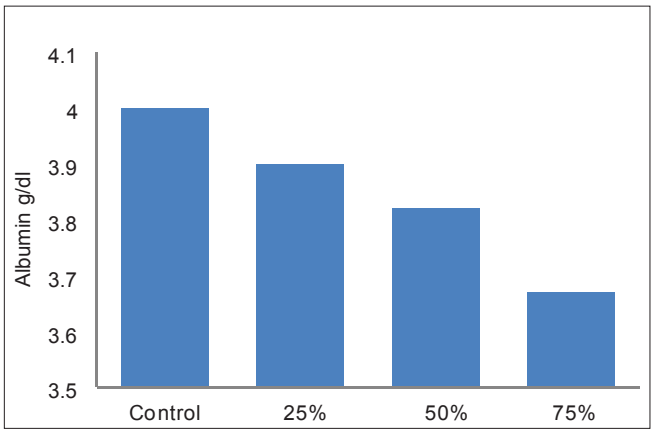


Figure 2: Effect of blend of food color on albumin

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Table 2: Serological changes in albino rats fed with blend of sunset yellow, metanil yellow and tartrazine

Group	Dose	Proteing/dl	Albuming/dl	Alkaline phosphatase U/L	Total bilirubin mg %	SGPT U/L
I	Distilled water	10.08±0.16	4.00±1.12	150±1.26	0.87±0.27	21±1.21
II	25 mg/kg body weight	8.7±0.27**	3.90±1.17	168±0.97	0.73±0.18	21±0.89
III	50 mg/kg body weight	7.62±0.22****	3.82±0.52	182±1.07*	0.93±0.32	27±0.72**
IV	75 mg/kg body weight	7.59±0.18****	3.67±0.37	201±1.62**	1.03±0.41	42.7±0.98****

Value represent the mean±SEM, *P<0.05 significantly different from control, **P<0.05 very significantly different from control, **** P<0.05extremely significantly different from control, SGPT = Serum glutamate pyruvate transaminase

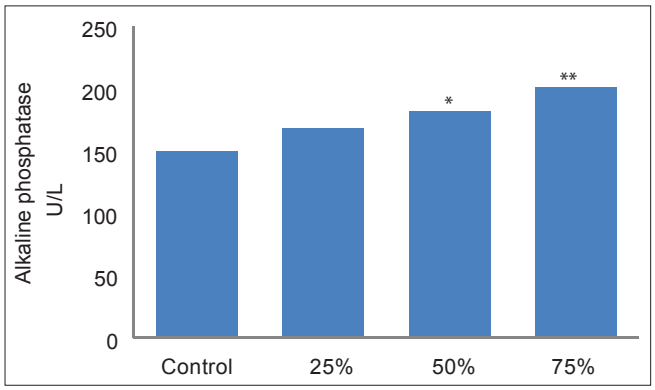


Figure 3: Effect of blend of food color on alkaline phosphatase

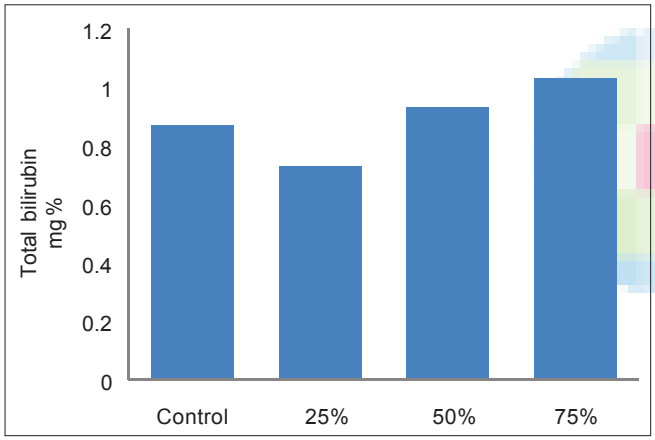


Figure 4: Effect of blend of food color on total bilirubin

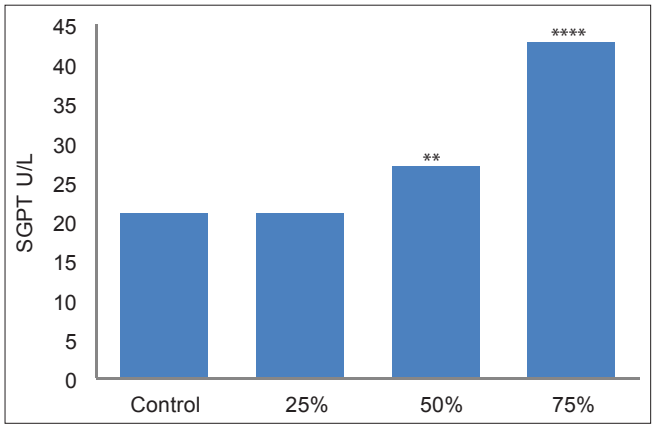


Figure 5: Effect of blend of food color on SGPT

change in SGPT at the same dose (Group II) in comparison to control [Figures 4 and 5].

DISCUSSION

The present study showed that dose feeding animals showed an increase in body weight. A similar finding was also reported by Gautam *et al.*[5] He studied the toxic impact of tartrazine on Swiss albino mice and also found an increase in body weight in both experimental groups i.e. for low dose (0.2 g/kg body weight) and high dose (0.4 g/kg body weight) groups. Hasan[6] also showed weight gain in experimental animals treated with 7.5 mg/kg body weight tartrazine and 0.15 and 0.3 mg/kg chocolate brown. Chatterjee and Shinde[7] also reported an increase in the body weight over 20% above the mean body weight. Similar results have also been reported by Osman *et al.*[8] in mice fed with synthetic food colorant; Sharma *et al.*[9] in mice fed with orange red; Sharma *et al.*[10] in mice fed with apple green and Chakravarty *et al.*[11] in mice fed with lead chromate.

However, Sharma *et al.*[1] studied the hemotoxic effect of chocolate brown on Swiss albino mice and observed no significant change in body weight of experimental mice when compared with control. On the contrary, Sharma *et al.*[12] showed a highly significant decrease in the body weight of experimental animal when fed with tartrazine; Abdel-Aziz *et al.*[13] in rat fed with some synthetic food colorant; Helal *et al.*[14] in rat fed with some synthetic and natural food colorant.

The present study revealed a significant increase in alkaline phosphatase in rats which consumed different doses of blend of food color. These results are in accordance with Singh[15] Helal[16] Mahmoud[17] Sharma *et al.*[1] and Sharma *et al.*[18] Singh[15] also showed an increase in alkaline phosphatase in rats fed with metanil yellow and maintained on low protein diet. Helal[16] studied the effect of oral administration of the mixture of sodium nitrate and sunset yellow given daily for 30 days to albino rat and observed a significant increase in alkaline phosphatase. Sharma *et al.*[1] observed the effect of feeding different dose of blend (Chocolate brown) on experimental mice and serological studies revealed an increase in the alkaline phosphatase. Mahmoud [17] observed an increase in alkaline phosphtase in rat fed with synthetic dye brilliant blue. Sharma *et al.*[18] observed an increase in alkaline phosphatase in female Swiss albino mice fed with kesari powder (a blend of tatrazine and sunset yellow).

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In the present study, a decrease in total protein and albumin was reported [Figures 1 and 2]. Similar result has also been reported by Sharma *et al.*[18] in Swiss albino mice fed with kesari powder. He reported a significant decrease in total serum protein at both dose levels of kesari powder. The present observations are also in accordance with Ashour and Abdel-aziz[19] and Chakravarty *et al.*[20] Latha and Jeevaratanm[21] observed a 13-week oral toxicity study of carotenoid pigment in rat and reported a slight decrease in total protein and albumin in treated groups in comparison to the control group. On the contrary, Sharma *et al*.[12] reported a significant increase in serum protein at low dose but the increase was found to be non-significant at high dose in Swiss albino mice fed with tartrazine. Sharma *et al*.[1] also reported an increase in serum protein in Swiss albino mice fed with chocolate brown. Himri *et al.*[22]studied the 90 day oral toxicity study of tartrazine and evaluated that the total protein significantly increases in a group treated with 7.5 and 10 mg/kg body weight but decreases in a group treated with 5 mg/kg body weight.

The present study showed a slight increase in total bilirubin and SGPT. Latha and Jeevaratnm[21] also observed a slight increase in SGPT in treated male and female rats in comparison to the control group. Mahmoud[17] also reported a significant increase in total bilirubin in rats after the administration of brilliant blue supplemented diet.

CONCLUSION

The present subchronic study of blend of food color indicates serological changes in experimental animal and the prolonged consumption of the blend may cause adverse effect on human health.

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