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Studies on the Effect of Cyanobacteria on Tobacco Pasted Albino Rat (*Rattus norvegicus*)

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ABSTRACT

The albino rat treated with tobacco decreases body weight than the control rat fed with normal diet then treated with Synechocysis pevalekii showed increased body weight the control as well as the tobacco pasted rat. Regarding enzyme status, alkaline phosphate and acid phosphatase enzyme showed very good promise in the experimental rat treated with Cyanobacteria diet. The heamatological study carried on the blood control, tobacco pasted and cyanobacteria treated rat, display no remarkable change except for slight rise in WBC, RBC, platelet, albumin, glucose, protein and haemoglobin. The present investigation confirmed that marine blue green algae can easily be digested and assimilated and therefore can serve as a source of diet for an albino rat and good remedy for affected tissues.

Key words: Nictiona tobacum, blue green algae, testis, liver, epididymis.

INTRODUCTION

Tobacco is the name given to the plant and leaves of species of Nicotiana. The tobacco of commerce is derived almost entirely from Nictiona tobacum (Encyclopedia Britanica, 2008).The pharmacological activity of tobacco is due almost entirely to its nicotine content, an alkaloid which is a powerful and rapidly acting toxic chemical. The betel chewing habit as there appears to be relationship between the geographical distribution of buccal carcinoma and betel chewing (Avsan Maruthit et al., 2004).The blue green algae are recognized as a primary source of some exciting molecules found in marine invertabrates (Pietra, 1997).Blue green algae in general contain a significant amount of carotenoids, namely, beta carotene, lycopene and lutein providing it with good antioxidant properties. An early study on blue green algae's cancer-preventive properties in human was performed on tobacco induced oral leukoplakia. Mathew et

al., (1995) found that oral supplementation with Spirulina fusiformis resulted in complete regression of 57 percent of subjects with homogenous leukoplakia. After discontinuation of Spirulina supplementation, almost half of the complete respondents developed recurrent lesions. Blue-green alga has also been reported to modulate other systems and improve metabolism (Schaeffer and Krylov, 2000). Hence the experiments were designed to study the effect of tobacco on the oral mucosa of rat and also its effect on the organs like testis, liver, epididymis. Since the carcinogenic effects of tobacco were known to be reduced by cyanobacteria.

MATERIALS AND METHODS

The cyanobacteria of Synechocystis pevalekii filament forming a thallus with more or less constant sheata marine cyanobacterium was obtained from the culture. Culture media of the, Tom isolate and maintain the marine cyanobacteria,appropriate media are used and congenial environmental conditions are maintained phosphate, nitrate, magnesium and calcium are the macronutrients, generally required by them. The essential micronutrients are iron, zinc, manganese, copper and molybdenum. Since they are mostly adopted to diffused light, good growth is obtained at 1,400 lux,intensity the optimal temperate required for their growth is 27-35^{0}C.A number of culture media for growing marine cyanobacteria are available, the most commonly used are ASN-III,ASP-2 and enriched sea water medium.

RESULTS AND DISCUSSION

Biochemical composition

For the analysis of biochemical composition the marine water cyanobacteria species algae such as Synechocysis pevalekii involved in the present investigation, the Synechocysis pevalekii showed maximum concentration of CHO, Protein, Lipid (Table1).

Body weight and Food intake

The weight of the control albino rat increased from 165+0.5 to 190-+0.3 g at the same time tobacco pasted albino rat, weight of the body was decreased 6 percent after 15 days, the affected rat treated with cyanobacterial sample Synechocysis pavalekii for 15 days. After that weight slightly increases (66%) was recorded (Table 2).In the treatment with albino rats body weight increased considerably, compared to the control although food intake increased suggesting that this strain enhanced food consumption (Table2).

Enzymes status of albino rat

The effect of cyanobacteria mixture diet on alkaline phosphates and acid phosphatase in the blood of albino rat has been presented vividly (Table3).The alkaline phosphatase of the control rat showing a 286 IU/L compared to initial day 16.26 percent were increased. Tobacco pasted albino rat only 15 percent increased at the same time significant increase was recorded 37.98 percent in the cyanobacteria treated albino rat (Table3).

In another experiment an increase level of acid phosphatase

Enzyme 10-+0.001 experimental control rat, 27-+ with has been noted in cyanobcteria treated albino rat. In tobacco pasted albino rat acid phosphatase decrease. The enzyme status of albino rats after 30 days of feeding with control and experimental diet was clearly depicted (Table4).

Among the both, alkaline and acid phosphatase were conveniently presented in a higher concentration in an experimental rat fed with cyanobacteria diet than the normal control rat and tobacco pasted rat. The enzyme strategy in an albino rat critically focusing the fact that the two enzymes invariably and actively involved on the metabolism of the experimental albino rats. The reason for the decline in the concentration of acid phosphatase and alkaline phosphate in the metabolism of the blood of the target groups. The study critically assayed the molecular activity of the enzymes can be focused through the growth of the control and experimental rats. An increasing concentration of enzymes in an experimental rat can be directly correlated with an increasing weight of the experimental rats. This has been clearly presented in (Table4), where the experimental rat with an increasing weight than the cyanobacteria fed rat has some stimulatory effect on the growth and metabolism of an albino rat Ramakrishnan et al., 2006. It is also due to the fact that the nutrient strategy of cyanobacteria, normally having more concentration of protein (Table4) might have some influence on the muscular protein inturn increase the weight of the experimental subject.

Composition of diet

The present study intensively formulated two types of diet. A control diet has been prepared with feed ingredients of groundnut oilcake (43%), corn flour (5%), ragi flour (43%), rice flour (7%) in our laboratory and this has been given daily 3 dose to the control group of 4 albino rats with the water and libitum. An experimental diet was prepared carefully with the formulation of ground nut oil cake (25%), corn flour (5%), ragi flour (25%), rice flour (5%) and cyanobacteria (35%).This has been prepared in the form at pellets and fed to an experimental group of the number of albino rats in our laboratory at normal room temperature (32^{o}C)

Heamatological characters

Haemoglobin

The haemoglobin content of blood in control albino rats with a mean of 18.6+0.001 g/dl and tobacco pasted albino rat 12+0.002 was recorded, where cyanobacteria treated albino rats showing a mean of 21.6+0.02 g/dl of blood. The rat treated with Synechocysis pevalekii resulted in significant in blood constituents (Table5).The recover of haemoglobin in 15 day 53.19 percent was recorded.

RBC counts

RBC counts of control albino rats having 4.74+0.1 per μl blood and tobacco pasted albino rat 2.91+0.1.Whereas in treated albino rats it was deviated 3.94+0.1μl was recovered. This has been clearly indicated (Table6).

WBC total count

WBC counts of control tats were showing 11.4+0.1μl of blood and tobacco pasted albino rat 12.9+0.2, whereas treated albino rats were showing 14.8+0.1μl of blood. The respective WBC counts has been presented (Table7).This data were clearly indicated the recover the WBC (32.142%).

Platelet counts

The result of platelet counts of control albino rats showing 5.2+ 0.02 and tobacco pasted albino rat 6.06 percent was decreased, while the cyanobacteria treated albino rats having 4.9+0.12 per μl

of blood platelets cells (Table8).This result showed the cyanobacteria treated albino rat 58.06 percent were increased.

Glucose

The serum glucose concentration of experimental rat showed an alteration of 81.0+0.3 mg/dl and it was drastically decrease in tobacco pasted rats,54+0.2 mg/dl and it was depicted in (Table9)compared to control rat(64+0.11),tobacco pasted rat clearly increased the drastical decrease.

Albumin

Similarly, albumin showed decrease level of 1.67+0.02 in the experimental rat and a increasing level of 1.9+0.01 were estimated in control rat 1.4+0.01 (Table10).

Total proteins

The amount of total serum protein in the experimental animals showed significant fluctuation for mistance an higher percentage of protein 4.0+0.02 in experimental rats and lower level of protein 2.0+0.01 in control rats were noted in our investigation(Table11).

Histological study

The histological changes observed in the control as well as tobacco pasted and cyanobacteria diet groups are presented in the form of photomicrographs.

Oral cavity

In the oral cavity, the tongue and the cheek mucosa were studied for any incidence of neoplastic growth.

Tongue H and E

The anterior part of the tongue showed no significant change from the control plate. The posterior part of the tongue showed alterations in tobacco pasted albino rat H and E section studied from tobacco induced tongue show squamous epithelial hyperplasia focally with few areas of dysplastic epithelium in the lower one third and no evidence of frank malignancy.

Cyanobacteria induced show reversal of hyperplastic and dysplastic changes.

Check mucosa

Significant changes were observed in both the experimental groups. Epithelium showing focal areas of thickening and atrophy was observed in both the experimental groups after 30 days, Acanthis, Hyperkeratoses and epidermal bullar was observed in tobacco pasted animals after this treated cyanobactria are regained the structure tobacco paste. Cheek tobacco induced and cyanobacteria induced are similar changes so that of tongue, no evidence of malignancy. In 15 days tobacco induced severe changes in the testis and liver when compared with tongue and cheek.

Testis

A significant decrease in the spermatogenic stages of the testis was observed in both the experimental groups. Testis H and E sections studied from tobacco induced testis show areas of normal spermatogenesis admixed with tubules exhibiting germ cells with nuclear irregularity

suggestive of intra tubular germ cell neoplasia (ITGCN). Cyanobacteria induced testis show normal spermatogenesis and ITGCN changes are not seen (Sapori et al., 1998).

Liver

Liver with tobacco induction & E sections studied from the liver show effacement of normal architecture of the liver extensive areas of venous congestion. Hepatocystes exhibit granular ground glass cytoplasm and vesicular nuclei with prominent nucleoli. No evidence of cirrhosis and malignant transformation.Cyanobacteria H and E sections studied show hepatocystes regained their normal architecture with trabecular pattern and extensive areas of regeneration exhibited as cells with intences eosinophilic cytoplasm and double nucleated cells are seen. No evidence of necrosis.

Table1.Biochemical composition (%) of cyanobacterial trial species

S.No	Cyanobacteria	Protein	Lipid	Carbohydrate
1.	Synechocystis pevalekii	70.4+0.2	0.01+0.01	37.8+2

Table2.Increasing growth and weight of albino rat (g) fed with control and experimental diet after 30 days.

S.No	Experiment	Initial	Final after 30 d	% of grain/loss
1.	Control	165+0.5	190+0.3	15.5
2.	Tobacco paste	145+0.2	135+0.3	-6.896
3.	Tobacco cyanobacteria treated	130+0.3	127+0.2	-2.307 (66.66%)

Table3.Alkaline phosphatase (IU/L) status of albino rats fed with cyanobacteria diet

S.No	Experiment	Initial	Final after 30 d	% of grain/loss
1.	Control	246+0.001	286+0.01	16.260
2.	Tobacco paste	237+0.001	273+0.01	15.189
3.	Tobacco cyanobacteria treated	258+0.01	256+0.02	37.98

Table4.Acid phosphatase (IU/L) status of albino rats fed with cyanobacteria diet

S.No	Experiment	Initial	Final after 30 d	% of grain/loss
1.	Control	9.7+0.1	10+0.001	3.092
2.	Tobacco paste	11.3+0.1	19+0.003	68.14
3.	Tobacco cyanobacteria treated	10.2+0.2	27+0.002	164.70

Table5.Haemoglobin (gms) determination of an albino rats fed with cyanobacteria diet

S.No	Experiment	Initial	Final after 30 d	% of grain/loss
1.	Control	24.6+0.001	28.6+0.01	16.260
2.	Tobacco paste	23.7+0.001	27.3+0.01	15.189
3.	Tobacco cyanobacteria treated	25.8+0.001	25.6+0.02	37.98

Table6.RBCs Erythrocytes counts (mil/cu.mm) status of an albino rats fed with cyanobacteria diet

S.No	Experiment	Initial	Final after 30 d	% of grain/loss
1.	Control	4.62+0.1	4.74+0.1	2.597
2.	Tobacco paste	3.20+0.2	2.91+0.1	-9.06
3.	Tobacco cyanobacteria treated	2.84+0.2	3.94+0.1	37.32

Table7.WBCs (Leucocytes) counts (mil/cu.mm) status of an albino rats fed with cyanobacteria diet

S.No	Experiment	Initial	Final after 30 d	% of grain/loss
1.	Control	8.4+0.1	11.4+0.1	35.7
2.	Tobacco paste	14.1+0.1	12.9+0.2	-8.510
3.	Tobacco cyanobacteria treated	11.2+0.1	14.8+0.1	32.14

Table8. Platelet counts of an albino rats fed with cyanobacteria diet

S.No	Experiment	Initial	Final after 30 d	% of grain/loss
1.	Control	3.0+0.01	3.2+0.02	6.66
2.	Tobacco paste	3.3+0.02	3.1+0.15	-6.06
3.	Tobacco cyanobacteria treated	3.1+0.02	4.9+0.12	58.06

Table9. Glucose of an albino rats fed with cyanobacteria diet

S.No	Experiment	Initial	Final after 30 d	% of grain/loss
1.	Control	58+0.3	64+0.11	10.34
2.	Tobacco paste	59+0.2	54+0.2	-8.47
3.	Tobacco cyanobacteria treated	57+0.4	81+0.3	42.10

Table 10.Albumin of an albino rats fed with cyanobacteria diet

S.No	Experiment	Initial	Final after 30 d	% of grain/loss
1.	Control	1.3+0.2	1.4+0.001	7.69
2.	Tobacco paste	1.8+0.1	1.6+0.02	-11.11
3.	Tobacco cyanobacteria treated	1.3+0.01	1.9+0.01	46.15

Table11. Total proteins of an albino rats fed with cyanobacteria diet

S.No	Experiment	Initial	Final after 30 d	% of grain/loss
1.	Control	1.7+0.1	2.0+0.1	17.64
2.	Tobacco paste	2.0+0.2	2.8+0.3	40
3.	Tobacco cyanobacteria treated	2.1+0.3	4.0+0.2	90.47

Fifteen days experimental albino rats treated with tobacco paste. After that the albino rat treated with Synechocysis pevalekii, the body weight increased whereas with control diet, the body weight of rat decreased with advancing days. The food intake of the control rats ranged from 5 to 10g per animals with the minor fluctuation. In the treatment with cyanobacteria diet, the body weight increased more than the control, suggesting that these strains Synechocysis pevalekii species contributed to better conversion efficiency since there is a better metabolic phosphorylation.12different species of cyanobacteria so as to find the possibility of using them as nutritional supplements (Sundararaman et al., 2007).The present investigation the albino rat treated with tobacco decrease body weight than the control rat fed with normal diet then treated with Synechocysis pevalekii showed increased body weight than the control as well as the tobacco albino rat. Regarding enzyme status alkaline phosphatase and acid phosphatase enzyme showed very good progress in the experimental rat treated with cyanobacteria diet. This changed blood picture suggests that definitely there may be some a biochemical alteration happens in the target tissues such as liver and muscles. The present result are in conformity to earlier reports of (Venkadesh et al., 1976; Amitabha Ghosh et al., 1985; Zhang et al., 2001; Sindhu and Abraham, 2006) suggested that variation in enzyme activity may be related to protein structure and duration

of feed retention in digestive tract which in turn dependent on the fibre content and physical consistency of the rat. The haematological studies carried out with the blood of control, tobacco pasted and cyanobacteria treated animals, displayed no remarkable expert for slight rise in WBC,RBC,platelet,albumin,glucose,protein and haemoglobin.Albino rats treated with cyanobacteria diet increase the RBC counts when compared with control rats and tobacco pasted rats.RBCs counts when compared with control rats and tobacco pasted rats.RBCs are a part of homeostatic system and their number in circulation and constant under normal circumstances (Thompson,1975).Any drastic change in the RBC count, reflect a pathological state(Inwood and Thomson,1976).Though at the outset, increase in RBC counts in the present study proves that in view at the constancy in the density of the blood leads the blood homeosporis.

Haemoglobin

In the present study the haemoglobin content of the cyanobacteria treated diet growing an increasing trend in an albino rat than the control rats and tobacco pasted rats. The normal range of blood haemoglobin concentration in main is 14.5 to 17g/100ml (Oser, 1965) and in rats steady concentrations of 17.4to 19.3g/100 ml have been observed in the present study.

TC (Leucocytes)

The leucocytes of experimental rats showed a considerable increase at Rs.800cells/cu/mm while it was slightly lesser 8.400cells/cu/ml in the control rats and tobacco pasted animal slightly increased observed in the present study (Cheyne, 1964).

CONCLUSION

In the present work the reported increased levels of transaminases, which is attributed to the catabolism of protein and amino acids is suggestive of the degeneration of the testis, due to the toxic effects of tobacco treatment. These findings suggest that oral tobacco paste painting on mice significantly alters the histology of the cheek mucosa, tongue, epididymis and testes and definitely infilters on the normal functioning of the testes, seminal vesicle and liver. It has also been shown that the 15 days effect of oral tobacco painting than the cyanobacteria treated experiment was good result was recorded.

REFERENCES

[1] Nobel laureates, *Britannica-Ultimate*-**2008,** pp 417-845.
[2] Avsan Maruthit Y, Rao RS, Palivela H, Thakre S, *J Environ Sci Eng*, **2004**, 268-273.
[3] Pietra F, *Nat. Prod. Reports*, **1997**, 453-464.
[4] Mathew B, Sankaranarayanan R, Nair PP,Varghese C, Somanathan T, Amma AB, Amma NS, Nair MK, *Nutr. Cancer*, **1995,** 197-202.
[5]Schaeffer DJ, Krylov VS, *Ecotoxicology and Environmental Safety*, **2000**, 45:208-227.
[6]Ramakrishnan G, Raghavendran HR, Vinodhkumar R, *Chem Biol Interact*, **2006**, 104–14.
[7] Sapori ML, Kozak W, Savage SM, Geng Y, Keuger MJ, *Exp med boil*, **1998**, 279-289.
[8] Sundaraman M, Boobathi T, and Gopinath S, *Algae and Cyanobacteria in Extreme Environments*, **2007**, 209-224.

[9] Venkatesh B, Mukherji AP, Mukhopadhyay PK, and Dehadrai PV, *Indian Acad.Sci*, **1976**, 457-462.
[10] Amitabha Ghosh, Suniti Misra K, Ashis Dutta A, Choudhury, *Phytochemistry*, **1985**, 1725-1727.
[11] Zhang BH, Feng R, Fang L, and Qinglian W, *Bot. Bull. Acad. Sin*, **2001,** 9–16.
[12] Sindhu M, and Abraham TE, *Food Chem. Toxicol*, **2006**, 198-206.
[13] Thompson RB, *The English Language Book Society and Pitman Medical*, Oxford, **1975**, pp 8-16.
[14] Inwood MJ, and Thomson S, *Lynch's Medical Laboratory Technology*, London, **1976**, pp1064-1322.
[15] Oser B L, *Hawk's Physiological Chemistry*, McGraw-Hill, New York, **1965**, pp 1472.
[16] Cheyne GA, *Blackwell Scientific Publications*, Oxford, **1964**, pp. 397.