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Scurvy induced changes and ascorbate defense in albino rats, Rattus norvegicus

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Abstract: An attempt has been made to find out effectiveness of scorbutogenic diet in albino rats, *Rattus norvegicus*. These animals received 50 and 80 gm cakes of scorbutogenic diet for 15 and 30 days respectively. No scurvy like symptoms were observed rather boils got appeared on their hind limbs, below ear and neck region. Steroid producing glands indicated enhanced steroidogenesis as evidenced by decreased concentrations of adrenal ascorbate and cholesterol. The results suggested that scorbutogenic diet was ineffective to cause scurvy in this species.

Keywords: Scurvy, Ascorbate, Steroidogenesis, Mammal

INTRODUCTION

Many mammalian species are able to synthesize ascorbate (vitamin-C) in their liver and kidneys except primates (including humans) guinea pigs and bats because they devoid of hepatic enzyme L-gulanolactone oxidase which is responsible for the conversion of glucose to ascorbate (Kipp et al., 1996; Gerardo et al., 2000). They must receive ascorbate from an exogenous source otherwise signs of scurvy like bleeding gums, weak skeleton, loss of teeth, loss of appetite and hairs, fatigue, joint weakness, fragile bones, loose muscles and ageing would appear and the animals are unable to synthesize intercellular substances like proteins, lipids and collagen etc (Barnes et al., 1973; Jones et al., 1973; Brake, 1988). Ascorbate function as anti-scurvy agent and also helpful in preventing arterial stiffness, diabetes etc (Wilkinson et al., 1999; Zollinger et al., 1999). Ascorbate can prevent the formation of free radicals and hence act as antioxidant (Simon and Hudes, 2000; Nockles, 1990). The albino rat (Rattus norvegicus) a mammalian rodent can synthesize ascorbate (Jackel et al., 1950; Train and Trounson, 1993). Scorbutogenic diet is composition of the substances capable in producing disease scurvy (Roy et al., 1973). According to Bjorkhem et al. (1978) scurvy stress might increase the rate of synthesis of corticoids to such a level that it can not be further stimulated by ACTH (ardenocorticotropin hormone). Lack in ACTH activity is due to lack of ascorbate in the gland at that time (Clayman et al., 1970). Ascorbate deficiency has negative effect on reproduction in both males and females (Odumosu and Wilson, 1973; Clow et al., 1975; Sherman et al., 1980; Chinoy et al., 1986; Carson et al., 1988 and Fazleabas et al., 1997). Ascorbate and collagen increases embryo implantation in rodents (Train and Trounson, 1993).

Previous studies on scurvy were performed in those species that can not synthesize their own ascorbate (Banerjee and Ganguli, 1962; Debora *et al.*, 1998 and Gerardo *et al.*, 2000). Hence in present investigation an attempt has been made in albino rats to find out effectiveness of scorbutogenic diet in correlation with liver, adrenals and gonads.

MATERIALS AND METHODS

Thirty male albino rats weighing 70-100 gm were divided into three groups comprising of 10 rats in each group. The albino rats were reared in the animal house under normal environmental conditions and were kept in plastic cages of size 18"X12"X12 covered by wire net. Controls (group I) were fed on usual diet of soaked gram, bread etc and experimental albino rats received scorbutogenic diet cakes. Water was provided *ad libitum*.

Group I: Normal untreated control; Group II: Scorbutogenic diet 50 gm/animal/day for 15 days. Group III: Scorbutogenic diet 80 gm/animal/day for 30 days All albino rats were sacrificed on the last day of experiment and the concerned tissues were preserved in their respective media for biochemical analysis. The contents of ascorbate (adrenal and testes) and cholesterol (liver, adrenal and testes) were estimated via spectrophotometric methods of Dabrowaski *et al* (1988) and Washburn and Nix (1974) respectively.

RESULTS

Morphological changes: The albino rats of group III developed boils on their hind limbs and neck region. There were no changes seen in the food habits, normal routine and behavior of the treated animals as compared with controls.

Body weight changes: There was increase in body weights

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Table 1. Effect of Scorbutogenic diet on body weights (gm) of albino rats.

Groups	Treatment and doses	Initial body weight	Body weight after 14 days	% Change	Initial body weight	Body weight after 30 days	% Change
Group I	Untreated control	80.50 <u>+</u> 0.96	80.00 ± 0.71	0.62	80.50 <u>+</u> 0.96	81.00 <u>+</u> 0.84	0.62
Group II	Scorbutogenic diet for 14 days	83.50 <u>+</u> 1.92	84.50 <u>+</u> 1.34	1.20	-	-	-
Group III	Scorbutogenic diet for 30 days	84.00 <u>+</u> 1.58	-	1.04	-	87.50 <u>+</u> 1.00	4.17

Mean + SE of 10 animals

by 1.20% and 4.17% in the animals of group II and III respectively (Table 1).

Biochemical changes: There was significant decrease in the ascorbate and cholesterol contents of adrenal in albino rats of group III. However, there was no noticeable change in the albino rats of group II (Table 2 and 3).

DISCUSSION

In the present investigation scorbutogenic diet failed to create metabolic imbalances and hence scurvy like symptoms in the rats. It is evidenced as there were no

Table 2. Effect of Scorbutogenic diet on ascorbate content (mg/g) of liver and adrenals of albino rats.

Groups	Treatment and Doses	Liver	Adrenals
Group I	Untreated control	3.48 + 0.07	3.32+0.08
Group II	Scorbutogenic diet for 14 days	3.46 <u>+</u> 0.06	3.35 ± 0.02
Group III	Scorbutogenic diet for 30 days	3.47 + 0.07	*3.24 + 0.03

Group III and II were compared with group I (n=10). Values are represented as mean \pm SE; *P<0.05; ANOVA

consistent with the studies of Bjorkhem et al. (1978) and Chinoy et al. (1986). Further ascorbate also helpful in preventing oxidative damages or in preventing formation of free radicals (Valkonen and Kuusi, 2000 and Kharb, 2000). It may be assumed that scorbutogenic diet was stressful and hence some content of ascorbate got consumed against this stress. As stated by Nockle (1990) stress causes the animal to increase its metabolic rate which through normal oxidative metabolic pathway reduces increased free radicals. This was another reason that the antioxidants get consumed and their requirement in the diet will increase during stress. An increase in the body weights of these animals might be due to swelling, odema and Boils (Elster, 1950). It may be assumed that some substance of scorbutogenic diet was allergic that causing boils in these animals.

Conclusion

The present study concluded that administration of scorbutogenic diet failed to create scurvy in albino rats as they can synthesize ascorbate in liver. Further only a dose of 80 gm scorbutogenic diet for one month was effective to stimulate adrenals for enhancing synthesis of corticoids.

Table 3. Effect of Scorbutogenic diet on cholesterol content (mg/gm) of liver, adrenal and testes of albino rats.

Groups	Treatment and Doses	Liver	Adrenals	Testes	
Group I	Untreated control	26.35 <u>+</u> 0.88	49.39 <u>+</u> 1.05	25.63 ± 2.22	
Group II	Scorbutogenic diet for 14 days	25.92 <u>+</u> 0.72	49.44 <u>+</u> 1.25	26.43 ± 0.24	
Group III	Scorbutogenic diet for 30 days	26.11 <u>+</u> 0.35	* 41.14 <u>+</u> 1.79	27.45 ± 0.51	

Group III and II were compared with group I; (n=10). Values are represented as mean + SE; *P< 0.05; ANOVA.

any changes in the level of ascorbate and cholesterol from liver after scorbutogenic diet administration. A dose of 80 gm scorbutogenic diet for 30 days (group III) was sufficient to suppress the level of cholesterol and ascorbate only from adrenals. After three weeks this dose exerted severe stress that enhanced the synthesis of corticoids in adrenals. Ascorbate found in the adrenal cortex is necessary for the conversion of cholesterol to pregnanolone; first step of steroidogenesis. Hence decreased concentration of ascorbate and cholesterol from adrenals is indicative of their consumption during enhanced synthesis of corticoids (Tables 2 and 3). It is

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