

Influence of dietary fats on plasma cholesterol and body weight in Indian desert gerbils (*Meriones hurrianae* Jerdon)

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Introduction

The Indian desert gerbil (*Meriones hurrianae* Jerdon) is a destructive rodent found in the desert ecosystem and lives on desert vegetation making afforestation difficult (Muthanna 1974, Prakash 1981). This species has been successfully bred for the first time in our laboratory under semi-natural conditions with carefully monitored feeding and husbandry conditions (Saibaba *et al.* 1988). Growth curves and baseline data have been generated and published (growth, reproduction, haematological and biochemical parameters) with the view to explore this species as a new animal model for nutritional, toxicological and safety evaluation studies (Saibaba *et al.* 1992). Our colony of Indian desert gerbils has been listed in the "International index of laboratory animals" (Festing 1993) and reproductive performance has been scrutinized (Saibaba 1987). The information available on hypercholesteremia in the Indian desert gerbil and the response to various fats on cholesterol metabolism is scanty. In addition, rats do not serve as good model to study hypercholesteremia. In some cases, rabbits have been used and they seem to respond well hypocholesteremic studies.

Early studies on the Mongolian gerbils (*Meriones unguiculatus*) and larger species such as *Meriones libycus* indicated that these animals respond well to dietary protein, soya protein and dietary fat (Forsythe III 1986, Chu & Hegsted 1976). Besides a few studies have also been reported on the effect of cholesterol, phytosterol and essential fatty acid requirements (Garden *et al.* 1959, Chu & Hegsted 1981). Although these studies reported the gerbils response of dietary fats similar

to that of humans and rats, no exhaustive studies have been done in Indian desert gerbils to evaluate the effect of unsaturated oils on cholesterol levels, in particular, Rice bran oil (RBO). In this paper we report the results of studies on the Indian desert gerbil (*Meriones hurrianae*; Jerdon) which indicate that this species could be used for studying the effect of unsaturated fats on serum cholesterol, growth and various other parameters.

Materials and methods

Male and female gerbils (*Meriones hurrianae*; Jerdon) about 4 weeks old were used in these experiments. Before the experiment, three adults per sex were housed in polypropylene cages (Sixe L 410 × W 280 × H 150 mm) with stainless steel top grill having facilities for keeping pelleted feed and water in glass bottles. Clean paddy husk and sand were used as bedding materials and changed twice a week.

The room temperature was 24–26°C, relative humidity was 50–60 % and the lighting was maintained at 12 h light/12 dark cycle. Until the start of the experiment the animals were fed a commercial and autoclaved "Gold Mohur" brand pelleted rat feed manufactured by M/S Lipton India Ltd., Bangalore - 560 052 (a subsidiary of "Unilever" of England), for a period of 4 weeks and had protected municipal tap water ad libitum. After 4 weeks the animals were divided in 4 groups, each of 8 animals. The animals were housed individually in polypropylene cages of dimensions L 290 × B 220 × H 140 mm. The animals were fed different diets containing 5 % Peanut oil (PNO) and 5 % RBO. The diets had the following composition: both

Table 1. Chemical and fatty acid composition of the vegetable oil based diets.

	PNO	RBO
Chemical composition (%)		
Protein	16.8	17.2
Fat	5.0	5.0
Carbohydrate	77.2	77.8
Fatty acid composition (%)		
C 12:0	0.10	0.59
C 14:0	0.40	1.62
C 16:0	15.38	40.42
C 18:0	0.21	0.15
C 18:1	45.38	40.42
C 18:2	34.58	36.58
C 18:3	0.12	0.80
SFA	22.58	25.20
MUFA	45.38	40.42
PUFA	34.57	37.46
P/S ratio	1.53	1.48
18:1/18:2	1.22	1.10

For salt mixture see Ref. 15 and for Vitamin mixture see Ref. 16. SFA: saturated fatty acid include 12:0, 14:0, 16:0 and 18:0.

MUFA: Monounsaturated fatty acids include 18:1.

PUFA: Polyunsaturated fatty acids include 18:2 and 18:3.

P/S ratio: Polyunsaturated/saturated fatty acid ratio.

diets had Wheat 26.66 % Ragi 31.66 % Bengal gram dhal 26.67 %, Casein 6.67 % and PNO (Postman brand, Manufactured by Ahmed mills, Faruk Anwar Co., Raichur, India), or RBO (Harvest oil Company, Tiruchirapalli, India) at a level of 5 %. The diets contained appropriate amounts of salt mixture (Chu & Hegsted 1976) and vitamin mixture (Hegsted *et al.* 1974). The chemical and fatty acid composition of the diets are shown in Table 1. The diets were mixed and pelleted before feeding to the animals. Before the beginning of experiment the animals were bled by ear vein puncture and the blood was collected over heparin. The plasma was then separated and was used for initial cholesterol values. The body weight and feed intake were recorded weekly for 10 weeks. At the end of 10 weeks, the animals were anesthetized with ether after an overnight fast. Blood samples were collected by cardiac puncture and the

serum was separated and stored at -4°C until analysis. Livers were removed and weighed.

Lipid analysis: Lipids from the diets were extracted by the method of Folch *et al.* (1967), and the total cholesterol in the serum was estimated by the method of Scarcy & Bengquist (1960).

Fatty acid analysis: The lipid extract was evaporated using a stream of nitrogen and methylated with methanolic HCL as described by Kates (1972). Methyl esters of fatty acids were dissolved in chloroform and analysed by gas chromatography (Packard) with a flame ionisation detector and a stainless steel column (2 m \times 0.2 cm I.D) packed with 10 % DEGS on gas chrom Q with a nitrogen flow rate of 20 ml/min. Column temperature was maintained at 180°C isothermally with the injector and detector at 230°C and 240°C respectively. Peak areas of chromatograms were measured by Shimadzu 2A integrator and identified by comparison with standard fatty acid methyl esters (Ackman 1969). Results are reported as percent fatty acids. All the reagents were of analytical grade and standard fatty acids were obtained from Sigma Chemical Co., St. Louis, Missouri, USA.

Statistical analysis of data: Statistical analysis of the data was performed using analysis of variance (ANOVA) and students "t" test. The difference was considered significant when $p < 0.05$.

Results

Following the 10 weeks study all animals appeared healthy, and the food intake, body weight gain and feed efficiency ratio (FER) are given in Table 2. Between the RBO male and female gerbils. Groups II and IV there was a significant difference in body weight at the end of the study ($p < 0.01$). Similar observations were made among PNO fed groups of males and females (group I and III). However, when the groups I and II were compared there was no significant growth difference between the males. On the other hand when the groups III and IV (PNO and RBO fed

Table 2. Body weight, feed intake and FER of gerbils on diet with PNO or RBO.

Group	No. of animals	Initial weight (g + SD)	Final weight (g + SD)	Body weight (g)	Feed intake /day gerbil (g)	FER
I 5 % PNO (male)	8	26.66 + 1.80	144.66 + 5.08**	88.0	3.92	0.35
II 5 % RBO (male)	8	28.00 + 2.09	113.83 + 2.76**	86.0	3.90	0.32
III 5 % PNO (female)	8	26.66 + 1.47	104.66 + 4.37*	79.0	3.22	0.32
IV 5 % RBO (female)	8	24.83 + 1.35	95.16 + 5.39	71.0	3.08	0.28

* $p < 0.05$ group III compared to group IV.

** $p < 0.01$ group I compared to group III and group III and group II compared to group IV.

females) were compared there was a significant growth difference ($p < 0.01$). The feed efficiency ratio (FER) did not indicate any unusual consumption of food in the four groups. The results of cholesterol and liver weight are given in Table 3. The initial cholesterol values were similar in both males and females.

Five percent PNO or RBO in the diet increased serum cholesterol in all groups. The hypercholesterolemic response was stronger in males than in females ($p < 0.01$). The serum cholesterol was lower in gerbils on RBO diet than in gerbils on PNO diet. However, this difference was significant ($p < 0.05$) for females only. There was no significant difference between groups in liver weight.

Discussion

The main objective of the present study was to examine if the Indian desert gerbil (*Meriones hurrianae*, Jerdon) can serve as an animal model to study the blood cholesterol responses in dietary studies to obtain information on possible differences in hypocholesterolemic responses. Studies have suggested that gerbils respond to even small changes in dietary lipids although there were some disadvantages in using this animal model. Such as the small size, low growth rate and the amount of blood obtainable (Zeman 1967, Gordon & Cekleniak 1961). Rats which have been used extensively for evaluating the effect of dietary lipids on cholesterol metabolism, show relatively little change in blood lipids

Table 3. Liver weight and serum cholesterol concentration of gerbils on diet with peanut oil or rice bran oil.

	5 % PNO	5 % RBO
Serum cholesterol (mg/100 ml + SD)		
Day 28 (males)	24.60 + 8.4	20.00 + 4.6
Day 100 (males)	135.14 + 11.15**	120.84 + 7.54**
Day 28 (females)	23.80 + 5.5	20.60 + 5.8
Day 100 (females)	109.39 + 9.26d*	85.59 + 4.83
Absolute liver weight (g + SD)		
Day 100 (males)	3.67 + 0.53	3.40 + 0.41
Day 100 (females)	3.20 + 0.16	3.18 + 0.24
Relative liver weight		
(male)	2.54	2.96
(female)	3.06	3.34

* $p < 0.05$ females on PNO diet compared to females on RBO diet.

** $p < 0.01$ males compared to females receiving the same diet.

even on cholesterol feeding. Thus the gerbil appears to be a better model than the rat to study lipid metabolism. The cholesterol lowering effect to polyunsaturated fats have been well documented in different species including gerbils (Avigan & Steinberg 1958, Albers & Gordon 1969).

The fatty acid composition of PNO and RBO is not very different with respect to unsaturated fats, polyunsaturated/saturated ratio, oleic/linoleic acid ratio and other fatty acids. The growth of male gerbils in the PNO groups and RBO groups is higher than that of the female gerbils, probably due to the higher food intake when compared to females. RBO has been reported to have a high unsaponifiable matter (Sharma & Rukmini 1987) although the composition *per se* is similar to PNO. The present study demonstrates that the response of male gerbils is different from that of females with respect to growth and serum cholesterol. Similar observations were made by other workers on Mongolian gerbils (*Meriones unguiculatus* and *Meriones libycus*) where the feed intake and dietary response to lipids were similar (Chu & Hegsted 1980, Nicolosi *et al.* 1976). However, in both sexes of gerbil RBO was less hypercholesterolemic than the PNO. Absolute and relative liver weight was no different in gerbils on PNO or RBO diet. Therefore, the differences observed in the serum cholesterol concentrations are apparently not due to the polyunsaturated fatty acids alone but probably due to various other factors present in the RBO and perhaps the difference in species also. However, studies on rats indicate that several factors seem to contribute to the cholesterol lowering effect. PNO contains linoleic acid (18:2) which has cholesterol lowering effect. Recent studies of RBO indicate the presence of oleic acid (18:1) and linoleic acid (18:2) besides other cholesterol lowering factors such as 24-methyl-cycloartenol and plant sterol which are known to reduce serum cholesterol levels (Grundy *et al.* 1991, Kiribuchi 1983). In addition, RBO contains oryzenol which is a mixture of ferulic acid esters and

triterpenols which also contribute to the cholesterol lowering effect (Gardon *et al.* 1959, Nakayama *et al.* 1987). Thus, these factors might contribute synergistically to the cholesterol lowering effect of RBO. In conclusion, the present study shows that the cholesterol lowering effect of PNO on gerbils did not parallel to cholesterol lowering effect of RBO.

The Indian desert gerbil seems to be well suited for many studies of nutrition and may in some cases be an attractive alternative to the rat.

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Summary

Two groups of Indian desert gerbils (*Meriones hurrianae*; Jerdon) were fed diets containing different fats of plant origin for a period of 10 weeks. The control gerbils had 5% peanut oil (PNO) whereas the experimental groups were fed with 5% Rice bran oil (RBO). After feeding for 10 weeks, the growth of the female gerbils fed RBO was significantly lower ($p < 0.01$) than that of RNO fed female gerbils. The growth difference between the male and female gerbils with respect to PNO was not however significant. There was no significant difference between the male and female groups with respect to liver weight. The RBO fed gerbils seemed to have low cholesterol level in the serum with significantly different levels between the males and females ($p < 0.01$). Thus, the present studies suggest that Indian desert gerbils are sensitive to a cholesterol lowering effect of vegetable oils and that these animals could be used as an experimental animal model instead of rats for evaluating the effect of various dietary fats.

Yhteenveto / K. Pelkonen

Kahdelle ryhmälle intialaisia aavikkogerbiilejä (*Meriones hurrianae*) syötettiin 10 viikon ajan kahta erilaista kasvirasvoja sisältävää ruokaa. Kontrolliryhmän ruuassa oli 5% maapähkinäöljyä ja koeryhmän ruuassa 5% riisiöljyä. Riisiöljyä saavien naasgerbiilien kasvu oli vähäisempää kuin kontrolliryhmän naaraiden ($p < 0.01$) ja sama suuntaus näkyi uroksissa. Maksapainoissa ei ollut eroja. Riisiöljyä saaneiden gerbiilien seerumin kolesterolitaso oli matala ja urosten ja naaraiden välillä oli merkittävä ero ($p < 0.01$). Johtopää-

tös on, että aavikkogerbiili on herkkä kasviöljyjen kolesterolia laskevalle vaikutuksella ja näitä eläimiä voitaisiin käyttää rotan sijasta eläinkoemallina arvioitaessa erilaisia ravinnon rasvoja.

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