

# Sub-chronic administration of Jojoba oil meal (*Simmondsia chinensis*) induce thyrotoxicity in rabbits



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## Abstract

Investigations were carried out to study the effect of supplementation of jojoba meal (*Simmondsia chinensis*) on biochemical, haematological and thyroid hormone profile in broiler rabbits. A total of forty New Zealand white rabbits (4 week old) were randomly allotted to four dietary treatment groups, comprising ten rabbits in each group. The control group (T0) was fed with complete feed with no jojoba oil meal and test groups were fed complete feed with 5, 15 and 25 per cent jojoba oil meal in T1, T2 and T3 groups, respectively. Hematology and serum biochemical profile indicated reduction in haemoglobin (Hb), packed cell volume (PCV) and total erythrocyte count (TEC) in T2 and T3 groups compared to T0 and T1 groups on 30<sup>th</sup> day of study, while serum calcium, phosphorus, cholesterol and total protein were not affected. It was observed that serum tri-iodothyronine (T<sub>3</sub>) and thyroxin (T<sub>4</sub>) levels in jojoba meal treated rabbits were declined in dose-dependent manner due to thyrotoxicity. Toxicopathological lesions include infiltration of inflammatory cells (T1) or inter-follicular thyroiditis in rabbits fed jojoba meal @ either 15 (T2) or 25 % (T3).

**Key words:** Jojoba, rabbits, *simmondsia chinensis*, haematocrit, thyroid

## Introduction

Jojoba (*Simmondsia chinensis*) is a perennial woody shrub indigenous to arid land in the western United States and Mexico particularly Sonoran deserts of Arizona, California, and Mexico and is being advocated and developed as a potential cultivated crop for warm, arid regions of the world (Hogan, 1979). The primary economic product from jojoba is colorless odorless polyunsaturated wax that is structurally similar to sperm whale oil, commonly called as jojoba oil which constitutes 50% of the seed and the residue (meal) that remains after oil extraction contains 26-30% crude protein (Verbiscar and Banigan, 1978; Verbiscar *et al.*, 1980). Residue (meal) could be used as a feed ingredient in addition to leaves and twigs. Jojoba seeds contain simmondsin, cyanomethylene cyclohexyl glucoside which is toxic to rats (Booth *et al.*, 1974) and some animals and simmondsin 2'-ferulate and other structurally related glucosides (Elliger *et al.*, 1974; Verbiscar and Banigan, 1978) both of which rendered meal unsuitable as livestock feed. Animal production in many parts of the world is becoming less efficient due to insufficient availability

of green fodders especially in tropical country like India. Many attempts have been made in this direction. Very few animal experiments (Trei *et al.*, 1979; Cokelaere *et al.*, 1993) have been conducted to ascertain the unconventional feeding value as well characterization of toxic effects of jojoba meal. (Verbiscar *et al.*, 1981; Flo *et al.*, 1999). Keeping this in view the present study was conducted to investigate influence of jojoba meal on hematological and serum biochemical profiles including thyroid hormones in broiler rabbits.

## Material and methods

Jojoba meal was procured from Association of the Rajasthan Jojoba Plantation and Research project, Jaipur, Rajasthan. Forty weaned (4 weeks old) New Zealand white rabbits of comparable body weights were selected for the study. They were randomly allotted to four dietary treatment groups consisting of ten in each group namely, T0 (basal diet without jojoba), T1 (basal diet with 5% jojoba), T2 (basal diet with 15% jojoba) and T3 (basal diet with 25% Jojoba). Basal diet were prepared in the form of complete ration using

locally available ingredients such as maize, sunflower cake, groundnut cake, wheat bran, red gram pod husk, mineral mixture, vitamin mixture and salt. Experimental diets, T1 T2 and T3 were prepared by replacing 5, 15 and 25 per cent jojoba meal. The rabbits were kept in individual cages (15'' x 18'' x 11'') and maintained in a well ventilated laboratory animal house.

The blood was collected from each rabbit by puncturing ear vein on 0, 30, 60 and 90 day of feeding trial. Hematological parameters were estimated as per Schalm (1965). Blood serum was subjected to serum calcium (Corentz 1982), serum proteins (Vatzidis, 1977), serum phosphorus (Munoz, 1983), serum cholesterol (Tietz, 1998) and serum T<sub>3</sub> and T<sub>4</sub> levels (Blackmore *et al.*, 1978)

The data generated in the present study with regard to dry matter (DM) intake, digestibility of nutrients and growth rate were subjected to statistical analysis according to Snedecor and Cochran (1985).

## Results

The chemical composition of the jojoba meal is presented in Table 1. The jojoba oil meal used in the preparation of complete ration of experimental rabbits in the present study comprised 27.49% crude protein (CP), 11.59% ether extract (EE), 7.37% crude fiber (CF) and 3.05 % total ash (TA).

Table 1: Chemical composition of jojoba meal

Parameters	Per cent dry matter basis
Organic matter	96.95
Crude protein	27.49
Crude fat	11.59
Crude fiber	7.37
Total ash	3.05
Neutral detergent fiber	28.34
Acid detergent fiber	26.21
Mixed toxicant	5.83
Hexane extract	10.75

The serum profile of calcium, phosphorus, total protein and cholesterol is presented in Table 3. The mean serum calcium (mg/dl) and phosphorus (mg/dl) level in experimental groups T0, T1, T2 and T3 were 8.42 and 4.00; 7.8 and 3.87; 8.59 and 3.91; 8.50 and 3.91, respectively. The average serum protein (g/dl) concentration was ranging from 6.37 (T0) to 6.52 (T1). The serum biochemical parameters did not differ significantly except the serum cholesterol ( mg/dl ) level which got reduced significantly (  $P \leq 0.05$  ) on day 30<sup>th</sup> of collection in T1 ( 36.75), T2 (30.50 ) and T3 (25.12) as compared to T0 ( 41.78 ).

Table 2: Effect of jojoba oil meal on haematocrit of experimental rabbits

Days	T0	T1	T2	T3
<b>Haemoglobin (%)</b>				
0	12.60±0.32	11.5±0.33	13.1±0.42	11.2±0.25
30	13.78±0.43 <sup>a</sup>	14.5±0.61 <sup>b</sup>	11.15±0.10 <sup>c</sup>	11.7±0.48 <sup>d</sup>
60	12.90±0.17	-	-	-
90	12.10±0.26	-	-	-
<b>Mean</b>	<b>12.88±0.21</b>	<b>12.03±0.55</b>	<b>12.44±0.46</b>	<b>11.47±0.26</b>
<b>Packed cell volume (%)</b>				
0	37.00±0.79	35.25±0.96	37.75±0.73	35.75±1.08
30	41.5 <sup>a</sup> ±1.29	43.51 <sup>b</sup> ±1.78	33.51 <sup>c</sup> ±0.35	35.30 <sup>d</sup> ±1.44
60	39.00±0.50	38.38±0	-	-
90	36.50±0.83	-	-	-
<b>Mean</b>	<b>38.50±0.66</b>	<b>39.22±1.58</b>	<b>37.33±1.41</b>	<b>34.42±0.80</b>
<b>Total erythrocyte count (millions/cumm)</b>				
0	6.16±0.13	5.87±0.15	6.28±0.12	5.45±0.18
30	6.8±0.22 <sup>a</sup>	7.2±0.30 <sup>b</sup>	5.55±0.03 <sup>c</sup>	5.86±0.24 <sup>d</sup>
60	6.33±0.07	-	-	-
90	6.05±0.14	-	-	-
<b>Mean</b>	<b>6.40±0.11</b>	<b>6.53±0.26</b>	<b>6.04±0.16</b>	<b>5.92±0.14</b>
<b>Total leukocyte count ( x 10<sup>3</sup>)</b>				
0	6.31±0.99	6.55±0.27	6.62±0.12	7.37±0.42
30	7.18±0.14	6.75±0.32	6.90±0.35	8.86±0.39
60	7.10±0.11	-	-	-
90	7.20±0.23	-	-	-
<b>Mean</b>	<b>6.95±0.27</b>	<b>6.68±0.20</b>	<b>6.71±0.15</b>	<b>8.02±0.40</b>

The blood biochemical profile such as Hb, PCV, TEC, TLC and serum cholesterol did not differ at zero, 60<sup>th</sup> and 90<sup>th</sup> day among the groups however, the values were reduced on 30<sup>th</sup> days collection in T2 and T3 groups (Table 2). The MCV (cu  $\mu$ ) values were ranging between 60.10 to 60.23, MCH ( $\mu$ g) values ranging from 19.37 to 20.20 and MCHC(g/dl) values ranging from 32.44 to 32.2 between the experimental groups however there was no significant difference between the test groups irrespective of period of collection. The mean serum calcium (mg/dl) and phosphorus (mg/dl) level in experimental groups T0, T1, T2 and T3 were 8.42 and 4.00; 7.8 and 3.87; 8.59 and 3.91; 8.50 and 3.91, respectively and did not differ significantly.

The initial values of tri-iodothyroine T<sub>3</sub> (ng/dl) and Thyroxin T<sub>4</sub> ( $\mu$ g/dl) were 69.0  $\pm$  3.45 and 3.9 $\pm$ 0.19, respectively. After 90 days of feeding trial the values were found to be 54.0 $\pm$ 2.7, 2.7 $\pm$ 0.13; 24.0 $\pm$ 1.2, 2.7 $\pm$ 0.13; 21.5 $\pm$ 1.07, 1.9 $\pm$  0.09 and 19.0 $\pm$ 0.95, 1.3 $\pm$  0.06 in T0, T1, T2 and T3 groups respectively. It was observed that serum levels of tri-iodothyronine(T<sub>3</sub>) and thyroxin(T<sub>4</sub>) were declined in rabbits receiving jojoba oil meal in dose dependent fashion.

## Discussion

### *Effect of Jojoba meal on hematological parameters*

The blood hematology picture (Hb, PCV, TEC and TLC) did not show any significant difference between the experimental groups on zero, 60<sup>th</sup> and 90<sup>th</sup> day of study (Table 2). However, Hb, PCV and TEC were significantly reduced in T2 and T3 groups compared to T0 and T1 groups on 30<sup>th</sup>

day of study however, when compared to control, changes in haematocrit values observed in various treatment groups were well within the physiological consideration (Schalm, 1965) similar to the observation of Trei *et al.* (1979) and Cokelaere *et al.* (1993). Similarly, the mean haemogram values such as MCV, MCH and MCHC values of the experimental groups showed no significant difference between the test and control groups irrespective of period of collection. Similar findings were observed in rats by Trei *et al.*, 1979 and Cokelaere *et al.* (1993).

### *Effect of Jojoba on serum biochemical profile*

Serum calcium, phosphorus and total protein (Table 3) were not affected by diets containing jojoba oil meal in rabbits at zero, 30<sup>th</sup>, 60<sup>th</sup> and 90<sup>th</sup> day of collection. No earlier studies were available to validate the present findings and to compare the results. However, the values obtained in the study were well within the normal physiological range.

Similarly, serum cholesterol (Table 3) level was significantly low on day 30 in rabbits however its level were comparable to control on day 60 or 90 in T1, T2 and T3 groups compared to control group. Similar findings were observed in New Zealand white rabbits by Clarke *et al.*, (1981) wherein they have opined that jojoba oil get absorbed across the intestinal mucosa, contrary to the hypothesis that it is totally excreted and not metabolized, However, Flo *et al.*, (1999) observed an increase in plasma total cholesterol levels in obese and lean Zucker rats and phenomena was related to an increase in HDL-cholesterol levels.

Table 3: Effect of jojoba oil meal on serum biochemical profile in experimental rabbits

Days	T0	T1	T2	T3
<b>Calcium (%)</b>				
0	8.56 $\pm$ 0.24	8.53 $\pm$ 0.30	8.72 $\pm$ 0.05	8.74 $\pm$ 0.09
30	8.37 $\pm$ 0.78	8.25 $\pm$ 0.06	8.33 $\pm$ 0.02	8.21 $\pm$ 0.12
60	8.06 $\pm$ 0.05	8.12 $\pm$ 0	-	-
90	8.62 $\pm$ 0.19	-	-	-
Mean	8.42 $\pm$ 0.08	7.80 $\pm$ 0.54	8.59 $\pm$ 0.08	8.5 $\pm$ 0.12
<b>Phosphorus (g/dl)</b>				
0	4.01 $\pm$ 0.01	4.00 $\pm$ 0.01	3.91 $\pm$ 0.07	3.98 $\pm$ 0.03
30	3.98 $\pm$ 0.02	3.90 $\pm$ 0.05	3.91 $\pm$ 0.08	3.93 $\pm$ 0.05
60	4.01 $\pm$ 0.02	3.96 $\pm$ 0	-	-
90	4.01 $\pm$ 0.01	-	-	-
Mean	4.00 $\pm$ 0.01	3.87 $\pm$ 0.08	3.91 $\pm$ 0.05	3.91 $\pm$ 0.03
<b>Total protein (g/dl)</b>				
0	6.44 $\pm$ 0.23	6.58 $\pm$ 0.18	6.48 $\pm$ 0.12	6.64 $\pm$ 0.11
30	6.38 $\pm$ 0.16	6.54 $\pm$ 0.08	6.46 $\pm$ 0.37	6.49 $\pm$ 0.28
60	6.35 $\pm$ 0.06	6.27 $\pm$ 0	-	-
90	6.32 $\pm$ 0.05	-	-	-
Mean	6.37 $\pm$ 0.07	6.52 $\pm$ 0.09	6.47 $\pm$ 0.14	6.47 $\pm$ 0.14
<b>Cholesterol (mg/dl)</b>				
0	41.50 $\pm$ 0.38	41.91 $\pm$ 0.78	43.82 $\pm$ 1.36	44.55 $\pm$ 0.68
30	41.78 <sup>a</sup> $\pm$ 0.56	36.75 <sup>b</sup> $\pm$ 0.91	30.50 <sup>c</sup> $\pm$ 1.17	25.12 <sup>d</sup> $\pm$ 0.46
60	41.84 $\pm$ 0.032	38.66 $\pm$ 0	-	-
90	41.80 $\pm$ 0.33	-	-	-
Mean	41.73 $\pm$ 0.20	39.25 $\pm$ 0.97	39.31 $\pm$ 2.80	36.24 $\pm$ 3.65

Mean value bearing different superscripts in a row differ significantly (P $\leq$ 0.05)

## Toxicopathology of thyroid

The mean concentration of tri-iodothyronine  $T_3$  (ng/dl; Fig 1) and thyroxin  $T_4$  ( $\mu\text{g/dl}$ ; Fig 2) values were reduced in T1, T2 and T3 groups compared to control (T0). The decreased  $T_3$  and  $T_4$  value in fasting animals is a well known phenomenon (Kuhn *et al.*, 1985). The reason may be due to lower 5' monodeiodinase activity as observed in fasted growing chickens (Decuyper *et al.*, 1984).

Mean value bearing different superscripts in a row differ significantly ( $p \leq 0.05$ ) Fig. 1. Serum levels of tri-iodothyronine ( $T_3$ ) in rabbits receiving diets supplemented with 5% (T1); 15% (T2) or 25% (T3) and zero per cent (Control; T0) jojoba oil meal

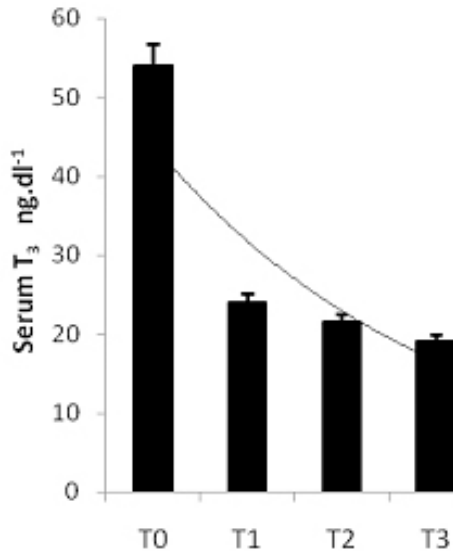
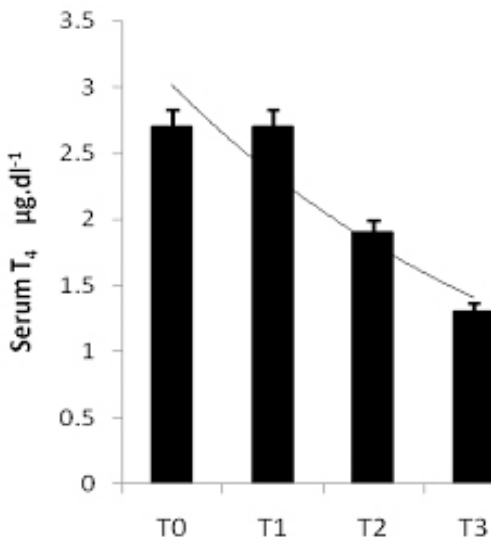
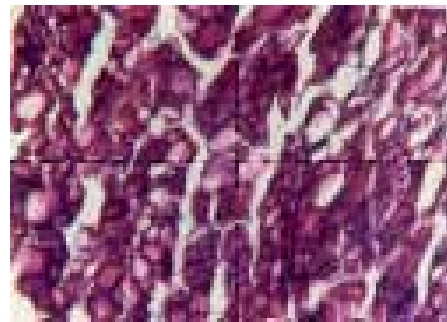


Fig. 2. Serum levels of thyroxin ( $T_4$ ) in rabbits receiving diets supplemented with 5% (T1); 15% (T2) or 25% (T3) and zero per cent (Control; T0) jojoba oil meal



Prolonged protein malnutrition in growing rats decelerates the decrease in  $T_3$  and  $T_4$  concentration compared to normally fed rats (Tulp *et al.*, 1979). The prolonged food intake reduction induced by simmondsin may also a strong reason for decrease in plasma  $T_3$  and  $T_4$  values observed in the present study and in concurrence with Cokelaere *et al.*, (1998). Observations made in this study were comparable with early findings in rats (Cokelaere *et al.*, 1993; Cokelaere *et al.*, 1998) and chicken (Arnunts *et al.*, 1993). Lesions in thyroid gland include focal infiltration of inflammatory cells in experimental rabbits receiving jojoba meal @ 5 per cent, inter-follicular thyroiditis with lymphocytic infiltration was observed in rabbits receiving jojoba meal @ either 15 (Fig.3) or 25 per cent. Booth *et al.*, (1974), Trei *et al.* (1979), Cokelaere *et al.*, (1993) and Cokelaere *et al.*, (2000) observed no histopathological changes in liver, kidney, thyroid, lungs, spleen caecum and ileum in rats and lambs fed various level of jojoba meal.

Fig. 3. Photomicrograph of thyroid of rabbit (T2 group) showing inter-follicular thyroiditis with lymphocytic infiltration (H & E; 100X)



To conclude, supplementation of jojoba oil meal for 90 days in the diets of rabbits did not significantly alter either serum biochemical profile or haemogram values. However, serum levels of tri-iodothyronine ( $T_3$ ) and thyroxin ( $T_4$ ) were reduced in dose dependent manner consequent to toxicopathological lesions induced by thyrotoxic factor(s) present in jojoba oil meal.

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