# Effects of various oils and of starvation on the lipid metabolism in brain

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#### Summary

In this study the effects of the dietary fat sources, viz., Dalda, mustard oil and groundnut oil in the brain lipid metabolism during starvation has been investigated. To find out this, these oils were fed to albino rats for 8 weeks followed by 3 and 5 days of starvation. Total as well as galactolipids of brain were not affected by dietary oils and starvation, whereas phospholipids of brain were significantly increased by fasting irrespective of the dietary fats. On the other hand, cholesterol was found to be increased in the groups fed with Dalda and groundnut oil and to be significantly decreased during fasting experiments.

Incorporation of (1-14C)acetate into lipid of the brain slices of rats fed different experimental diets than followed by starvation revealed that the uptake of radio-activity was higher in the group fed with groundnut oil, followed by Dalda. Low uptake was observed in the group fed with mustard oil.

### Zusammenfassung

Der Einfluß verschiedener Nahrungsfette auf den Lipidstoffwechsel des Gehirns während Hunger wurde untersucht. Albinoratten wurden 8 Tage lang mit Dalda, Senföl oder Erdnußöl gefüttert. Einige Tiere blieben anschließend 3 und 5 Tage lang ohne Nahrung. Die Gesamtlipide und die Galaktolipide wurden durch Nahrungsfette und durch Hunger nicht beeinflußt, während die Phospholipide unabhängig von der Art des Nahrungsfetts signifikant zunahmen. Cholesterin nahm in den mit Dalda und mit Erdnußöl gefütterten Gruppen zu und nahm ab während des Hungerns. In der Hungerperiode im Anschluß an die Fettzufuhr war der Einbau von 1-14C-Acetat in Lipide von Gehirnschnitten nach Erdnußöl höher als nach Dalda. Nach Senföl war der Einbau nur gering.

Key words: starvation, brain, lipid metabolism, dietary lipids

## Introduction

Lipogenesis (1) including fatty acid synthesis and elongation is reduced by long term fasting and is reversed on refeeding for a similar period both in the mammals and the birds (2). It has also been reported that the loss of body weight was greater after the feeding of high fat diet than that after the high carbohydrate diet (3). Brain weight is independent of the nature of dietary fats at all ages (4) but the postnatal under-nutrition significantly decreases the weight of brain (5). However, detailed changes in the lipid metabolism in the brain are not well defined. In the present study comparative effects of feeding different oils (mustard oil, *Dalda* and groundnut oil) on lipid changes in brain before and after starvation have been studied in rats.

#### Materials and methods

Mustard oil (rich in unsaturated acids and erucic acid), groundnut oil (rich in unsaturated fatty acids and arachidonic acid) and partially hydrogenated *Dalda\** oil (rich in saturated fatty acids) diets (Table 1 for fatty acid analysis) were prepared by adding one or the other fat at 20% level toskim milk powder (20%), starch (51%), crushed gram (6%), mineral mixture with common salt (0.5% each) and yeast powder (2%). The fat free diet was prepared by adding additional 20% starch in place of oils. The feeds were supplemented with vitamin A and vitamin D. Forty male albino rats weighing 50–70g each were produced from the Haryana Agricultural University, Hissar and acclimatised for one week before giving experimental diets. Groups of ten rats each were given the experiment diet *ad lib*.

After 8 weeks, four rats from each group were killed to collect brain tissue for analysis. Total lipids were extracted (6). Phospholipids (7) and total cholesterol (8) were estimated. The fatty acid analysis was done after conversion to methyl esters (9), by gas liquid chromatography. The other four animals were starved for 3 and 5 days and their brain tissue was analysed as above. *In vitro* synthesis of various lipid fractions has been studied using (1-14C)acetate (10) in brain tissue slices.

Fatty acids	Hydrogenated fat (Dalda)	Mustard oil	Groundnut oil
C <sub>12:0</sub>	1.2	_	
C <sub>14:0</sub>	0.5	0.6	Traces
C <sub>16:0</sub>	19.8	4.3	8.3
C <sub>16:1</sub>	_	0.4	4.9
C <sub>18:0</sub>	60.3	1.5	2.4
C <sub>18:1</sub>	18.2	10.2	44.7
C <sub>18:2</sub>	_	19.0	32.1
C <sub>18:3</sub>	_	14.0	3.0
$C_{20:1}$	_	2.5	_
C <sub>20:4</sub>	_	_	4.5
$C_{22:1}^{20:1}$	-	47.5	-
Total fatty acid	ls.		
Saturated	81.8	6.3	10.7

Table 1. Fatty acid composition (relative %) of dietary oils.

93.7 (33.0)

89.3 (39.6)

18.2(-)

Unsaturated

<sup>-=</sup> absent; the figures in parentheses represent the per cent essential fatty acids component of the total fatty acids.

<sup>\*)</sup> Dalda = Partially hydrogenated groundnut oil

#### Results and discussion

Feeding of fat free diet or diets containing different fats for 8 weeks did not affect the total lipid content of brain tissue. Similar results were obtained under fasting conditions (Table 2). However, the incorporation of (1-14C)acetate into total lipids of brain indicates that their biosynthesis is

Table 2. Effect of dietary fats on lipid contents (mean  $\pm$  S.D.) of brain tissue under normal and starvation conditions.

Diet	Total lipids (mg/g)	Cholesterol (mg/g)	Phospholipids (mg/g)	$\frac{\mathrm{PL}^*}{\mathrm{Ch}}$
Fat free				
Fed for 8 weeks		$14.3 \pm 2.1$	$54.0 \pm 3.4$	3.8
3 days starvation		$13.6 \pm 1.4$	$75.0 \pm 6.0$	5.5
5 days starvation	$106.6 \pm 8.2$	$12.9 \pm 3.3$	$84.0 \pm 3.5$	6.5
Dalda (Hydrogena	ated oil)			
Fed for 8 weeks	$103.7 \pm 7.7$	$23.1 \pm 6.4$	$65.9 \pm 6.0$	2.9
3 days starvation	$114.6 \pm 6.3$	$20.6 \pm 3.7$	$75.0 \pm 5.2$	3.6
5 days starvation	$101.2 \pm 5.6$	$14.7\pm3.4$	$78.1 \pm 5.4$	5.3
Mustard oil				
Fed for 8 weeks	$99.0 \pm 7.6$	$19.0\pm2.1$	$69.1 \pm 8.41$	3.7
3 days starvation	$110.9 \pm 5.2$	$17.9 \pm 2.72$	$72.1 \pm 6.2$	4.0
5 days starvation	$105.0\pm8.0$	$18.8 \pm 1.0$	$81.6 \pm 8.1$	4.3
Groundnut oil				
Fed for 8 weeks	$102.7 \pm 4.7$	$24.5 \pm 5.4$	$52.3 \pm 1.4$	2.2
3 days starvation	$112.6 \pm 6.6$	$18.8 \pm 1.0$	$60.2 \pm 3.4$	3.3
5 days starvation	$114.7\pm8.2$	$15.4 \pm 1.4$	$87.8 \pm 7.2$	5.7

<sup>\*</sup> PL = Phospholipids

Ch = Cholesterol

Table 3. Effect of feeding different fats on the *in vitro* incorporation of  $(1^{-14}C)$  acetate into brain lipids of rats during normal and starvation conditions.

Oil sources in	Incorporation (nmoles per g wet tissue)			
experimental diets	Unstarved <sup>+</sup>	Starvation perio	od	
		3 days	5 days	
Fat free	$0.378 \pm 0.017$	$0.188 \pm 0.001$	$0.144 \pm 0.009$ $0.171 \pm 0.010$	
Dalda	$0.318 \pm 0.018$	$0.303 \pm 0.001$		
Mustard oil	$0.291 \pm 0.018$	$0.114 \pm 0.002$	$0.078 \pm 0.004$	
Groundnut oil	$0.397 \pm 0.004$	$0.347 \pm 0.011$	$0.147 \pm 0.002$	

<sup>&</sup>lt;sup>+</sup> The values represent the acetate incorporation after 8 weeks of feeding experiment

(The incubation media contained 5 uCi( $1^{-14}$ C)sodium acetate, KRB buffer, ph 7.4 and 200–300 mg brain slices in final volume of 5 ml. The incubations were carried out at 37 °C for 2 hrs with continuous shaking under aerobic conditions. The values are average of three replications with two observations in each case.)

Table 4. Distribution of (1-14C) sodium acetate (in vitro) into various lipid fractions of brain lipids of rats fed different oils.

Oil sources				Relative percentage of radioactivity	age of radio	activity		
diets	Choles- terol	Phosphatidyl serine	Sphingo- myelon	Phosphatidyl choline	Sulpha- tide	Phosphatidyl ethanolamine	Cerebro- sides	Total polar lipids
Fat free*	30.1	6.10	8.01	10.25	15.09	15.35	15.08	6.69
3 days fasting	24.1	7.30	8.60	10.95	17.08	13.87	18.08	75.9
5 days fasting	19.2	8.30	9.00	11.89	21.01	11.05	19.01	80.8
Dalda*	33.0	7.30	7.30	10.50	16.27	10.79	14.51	67.0
3 days fasting	26.2	8.30	8.60	11.65	17.21	9.18	18.01	73.8
5 days fasting	21.6	8.70	8.90	12.60	18.09	8.50	21.59	78.4
Mustard oil*	27.5	10.34	7.05	8.94	10.47	16.34	16.68	72.5
3 days fasting	22.0	10.00	11.40	10.50	11.40	15.70	18.80	78.8
5 days fasting	20.5	10.01	10.10	13.20	12.00	10.20	22.80	79.5
Groundnut oil*	30.0	9.62	11.68	10.65	8.59	8.43	19.58	70.0
3 days fasting	27.9	10.00	11.98	11.65	10.30	6.30	21.58	72.1
5 days fasting	20.2	10.05	12.98	12.50	13.30	4.39	26.38	79.8

\* Indicate the radioactivity of groups before starvation that were earlier fed for 8 weeks on respective diets. The values represent the mean of three replications having two observations each case.

Table 5. Effect of feeding different oils followed by starvation on the fatty acid composition of brain tissue (relative %).

Diets	C <sub>12:0</sub>	C <sub>14:0</sub>	C <sub>16:0</sub>	C <sub>16:1</sub>	C <sub>18:0</sub>	C <sub>18:1</sub>	C <sub>18:2</sub>	C <sub>18:3</sub>	C <sub>20:4</sub>	C <sub>44:1</sub>
Fat free diet (8 weeks) 3 days fasting 5 days fasting	8.46 16.12 1.66	111	23.95 22.20 19.16	111	25.52 26.02 24.05	26.60 26.20 32.55	5.28 - 0.50	_ _ 0.56	- 5.13	10.15 10.54 17.44
Dalda diet (8 weeks) 3 days fasting 5 days fasting	7.30 6.33 2.56	1 1 1	6.16 25.79 23.01	1 1 1	25.57 25.33 25.26	26.82 31.62 25.51	9.28	9.28 - 4.58	3.76	11.52 10.85 12.75
Mustard oil diet (8 weeks) 3 days fasting 5 days fasting	3.43 6.33 3.79	1 1 1	18.90 25.79 20.10	1 1 1	19.60 25.33 20.56	26.18 31.67 32.63	17.18 - 1.39	3.92 - 4.66	_ _ 3.84	$10.63 \\ 10.85 \\ 11.07$
Groundnut oil diet <sup>(a)</sup> (8 weeks) 3 days fasting <sup>(b)</sup> 5 days fasting	21.40 8.57 3.75	15.60 6.99	17.70 24.60 25.56	6.70	8.70 19.29 21.22	5.80 24.71 26.11	1 1 1	1 ! 1	4.35	8.27 6.43 19.03

The values represent the mean duplicate of pooled samples  $^{(a)}$  Present  $C_6=5.12$  and  $C_8=4.93,\,C_{10}=5.7$   $^{(b)}$  Present  $C_8=9.32$ 

- = absent

inhibited during starvation under all previous feeding regimes (Table 3) but the incorporation into phospholipids is enhanced (Table 4) indicating the importance of maintenance of cell membrane structure.

Irrespective of the experimental diets, the phospholipid content significantly increased during 3 and 5 days of fasting. Maximum increase was observed in rats on groundnut diet (Table 2), and fat free diet after 5 days of starvation. Similar was the effect on the ratio of phospholipids to cholesterol (Table 2). Increase in the ratio of phospholipid to cholesterol makes the cell membranes more fluid and leaky. The increased levels of phospholipid is probably due to their preferential synthesis to maintain the integrity of the cell structure and is evident from the incorporation of (1-14C)acetate into phospholipid species (Table 4). The lower levels of cholesterol with mustard oil may be due to the presence of unusual fatty acid, erucic acid and fat free diet in the former or due to deficiency of desirable fatty acids in both (11). After starvation the decrease in the cholesterol content is due to its decreased biosynthesis as evident from the incorporation of (1-14C)acetate in cholesterol (Table 4).

Fat free diet followed by starvation for 5 days resulted in a decrease in saturated fatty acid with corresponding increase in unsaturated fatty acid mainly oleic acid. Similar was the case with groundnut oil diet. These results indicate that during starvation unsaturated fatty acids especially oleic acid are preferentially retained. With *Dalda ghee*, initially providing mainly saturated fatty acids, reverse was the case (Table 5) whereas the starvation after mustard oil diet did not produce much change in the nature of fatty acids.

The results indicate that during starvation saturated fatty acids are utilized for providing energy and unsaturated fatty acids are spared for maintenance of remaining cell membrane structures. This is amply demonstrated by the incorporation of (1-14C)acetate into phospholipids and sterols (Tables 4 and 5).

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