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NUTRITIONAL ASPECTS OF RAPESEED OIL: DIGESTIBLLITY, PROCESSING AND INFLUENCE OF ERUCIC ACID ON TISSUE LIPIDS

By B. L. Walker, S. P. Lall, S. J. Stinger and H. S. Bayley
Department of Nutrition
University of Guelph
Guelph, Ontarjo

### INTRODUCTION

Early work of Boer et al. (1947) indicated that rats grew more slowly on rapeseed oit than on butter fat. Duel et al. (1948) found that growth of rats was reduced appreciably by inclusion of 10% rapeseed oil (RSO) in the diet. Since RSO contains a high level of crucic acid (Cis-13-docosenoic) as compared to other vegetable oils, some workers have reported that most adverse effects of feeding rapeseed oil are associated with the crucic acid (Beare et al. 1959; Roine and Uksila 1959; Thomasson 1955). In contrast to these reports, several workers have found that crucic acid is not the sole cause for the poor performance of rats on rapeseed oil, but that an imbalance of saturated ammonounsaturated fatty acids (Hopkins et al. 1955; Murray et al. 1958) or the relatively low content of palmitic acid (Beare et al. 1963; Craig et al. 1963a, 1963b) in rapeseed oil could equally affect the growth. Recently, Rocquelin and Potteau (1968) have reviewed the relevant literature concerning the nutritive value of rapeseed oil.

Quantitative data on the utilization of RSO by poultry is quite limited. Kondra et al. (1968) added 15% rapeseed oil to a standard laying ration and observed a decrease in rate of egg production, egg weight and yolk weight. Joshi and Sell (1964) showed that inclusion of 5 or 10 percent RSO in the diet of turkey poults caused a depression in weight gain and feed consumption, with the magnitude of depression being in direct proportion to the RSO content of the diet. The metabolizable energy (M.E.) values indicated that the energy supplied by RSO was as available to poults as energy from soybean oil (SBO), sumflower oil or animal tailow. Salmon (1969a) reported that 9% RSO depressed the growth, feed conversion and dietary M.E. of growing turkeys as compared to 9% SBO; mixtures of RSO with 1/3rd beef tallow gave equal performance to 9% SBO. However, Blakely et al. (1965) found that 6 or 10% RSO was not growth depressing for 20 - 24 week old finishing turkeys.

In contrast to the growth depressing effects of RSO reported for rats and poults, Sell and Hodgson (1962) and Tsang et al. (1962) found no such detrimental effects with the chick. Sell and Hodgson (1962) observed that 4 or 8% RSO was as effective as comparable level of SBO in improving weight gain and efficiency of feed utilization of chicks. However, Salmon (1969b) reported that substitution of different levels of RSO for SBO in diets containing 10% total added oil depressed the growth of chicks but the M.E. content of the diets were approximately the same.

The following studies were undertaken to determine the M.E. content of RSO and rapeseed oil foots on chicks, turkey poults, rats and hens. Because of the deleterious effects reported for erucic acid, the utilization of canbra (zero erucic acid RSO) was also studied. In view of the findings of Sibbald et al. (1961, 1962) and Artman (1964) that mixing SBO and tallow resulted in a greater M.E. than predicted from the arithmetic mean of the two fats fed singly, the present studies were also designed to test the synergistic relationship between tallow and rapeseed oil. Studies were also made to compare the nutritional value of crude and refined RSO and to determine the response in egg size, hatchability and egg production.

Numerous nutritional studies have been carried out with diets high in erucic acid. For the most part, however, comparative studies on the deposition of this acid in different tissues and its effects on the fatty acid profile of such tissues have not been undertaken, although data are available for individual tissues (Carroll, 1962). The object of the present study was to rectify this situation.

METABOLIZABLE ENERGY AND PERFORMANCE OF REGULAR RAPESEED OILS VS. CANBRA OIL AND EFFECT OF REFINING RAPESEED OILS ON NUTRITIONAL VALUE

#### EXPERIMENT 1

The first experiment was designed to determine the M.E. content of degummed RSO, undegummed RSO, zero erucic RSO, RSO foots (acidulated soap-stocks from RSO), prime tallow and the 50/50 mixture of tallow and RSO or RSO foots. Because a sufficient amount of zero erucic RSO was not available at the time the experiment was conducted, the M.E. value for the mixture of tallow and zero erucic RSO was not determined.

Male, one day old White Rock chicks were housed in electrically

heated, wire-floored, battery brooders and fed a commercial starting ration until they reached the age of 2 weeks. Thereafter the birds were weighed, divided into groups having a weight spread of 5 g and distributed into pens until each pen contained 10 chicks. The experiment was of a randomized complete block design with each of the 12 experimental diets being allotted to 4 replicate pens. The birds were fed a practical corn-soybean meal diet or this diet containing 20% of prime tallow, degummed RSO, undegummed RSO, zero erucic RSO, RSO foots or 20% of a 50/50 mixture of the tallow with degummed RSO, undegummed RSO or RSO foots. The fats replaced the diet as a whole. The chicks received the experimental diets ad libitum from 2-4 weeks of age. During the last six days of the experiment, excreta samples were collected from each pen, pooled, frozen, lyophilized, ground and together with feed samples, analyzed to permit the determination of M.E. values according to the procedure outlined by Sibbald and Slinger (1963).

TABLE II

FAT UTILIZATION BY THE CHICKEN (1)

| Supplement          | Average<br>wt. gain | Average<br>feed cons. | Feed/Gain | Metaboliz                  | Metabolizable energy             |  |  |
|---------------------|---------------------|-----------------------|-----------|----------------------------|----------------------------------|--|--|
|                     |                     |                       |           | Derived                    | Calculated<br>from<br>components |  |  |
|                     | (g)                 | (g)                   |           | (Kcal/g)                   | (Kcal/g)                         |  |  |
| Basal               | 323e                | 693 <sup>a</sup>      | 2.15      | -                          | _                                |  |  |
| Deg. RSO            | 387 bcd             | 658 <sup>be</sup>     | 1.70      | 7 <b>.</b> 89 <sup>d</sup> | -                                |  |  |
| Undeg. RSO          | 374d                | 638°                  | 1.71      | 7.99 <sup>d</sup>          | <del>-</del>                     |  |  |
| ZRS0 <sup>(2)</sup> | 411ab               | 673 <sup>ab</sup>     | 1.64      | 8.710                      | _                                |  |  |
| Prime tallow        | 390bcd              | 651bc                 | 1.67      | 7.12 <sup>e</sup>          | ~                                |  |  |
| + Deg. RSO          | 409ab               | 671 <sup>ab</sup>     | 1.64      | 9.01 <sup>b</sup>          | 7.50                             |  |  |
| + Undeg. RSO        | 427a                | 657 <sup>be</sup>     | 1.54      | 9.18ab                     | 7.56                             |  |  |

<sup>(1)</sup> Treatments followed by the same letter are not significantly different (P<0.05)

<sup>(2)</sup>Crude zero erucic rapeseed oil

The metabolizable energy values derived for the fats and fat mixtures on chicks, together with the weight gain, feed consumption and feed: gain data, are presented as means in Table II. When added to the ration at a 20% level, the M.E. value of undegummed RSO was significantly (P<0.05) higher than that of degummed RSO. However, weight gain and feed consumption were lower for undegummed RSO than degummed RSO or tallow. It would appear that undegummed RSO may contain some factor(s) which reduces feed consumption but that the gums enhance energy utilization.

The M.E. content of zero erucic RSO was significantly higher than that of any other fat fed singly. Also, weight gain and feed efficiency for chicks fed zero erucic RSO were superior to the other oils. Although RSO foots gave inferior performance, the M.E. value was not significantly different from the remaining fats. The M.E. value data indicate that mixtures of fat contained more available energy than the mean of the components. Both fat mixtures showed approximately a 20% increase in the M.E. over the values calculated from the components. The weight gain and feed consumption data also provided evidence that synergism occurred within the fat mixtures.

## EXPERIMENT 2

The second experiment was designed to determine the M.E. content of the same samples of RSO and RSO by-product used in the previous trial but using turkey poults.

The experimental design and the methods were essentially the same as for experiment 1, except that an additional treatment of a 50/50 mixture of tallow and zero erucic RSO was included in this trial. One day old female Large White turkey poults obtained from a commercial hatchery were allotted on a weight basis into equal lots of 8 birds each. Four lots of poults were fed each experimental diet for 3 weeks. The basal diet was composed of practical type ingredients. During the last week of the experiment excreta samples were collected and subjected to the various analyses indicated previously for M.E. determination.

The M.E. value (Table III) determined on turkey poults suggest that this species made better use of all the fats than did chicks. Some of the oils were almot 100% utilized; the undegummed RSO showed an M.E. value of 9.57 vs. a gross energy of 9.45 thus suggesting that the product enhanced the energy utilization of the fat in the basal diet. However, the poults receiving the diets containing RSO consumed significantly less food and gained less weight than the poults fed the basal ration. This would

TABLE III

FAT UTILIZATION BY THE TURKEY (1)

|              |                  |                       |           | Metaboliz<br>Derived | able energy<br>  Calculated |
|--------------|------------------|-----------------------|-----------|----------------------|-----------------------------|
| Supplement   | Average wt. gain | Average<br>feed cons. | Feed/Gain | Derived              | from components             |
|              | (g)              | (g)                   |           | (kcal/g)             | (Kcal/g)                    |
| Basal        | 377a             | 550 <sup>a</sup>      | 1.46      | -                    | -                           |
| Deg. RSO     | 226c             | 305¢                  | 1.35      | 8.44b                | _                           |
| Undeg. RSO   | 212c             | 293°                  | 1.38      | 9.57a                | -                           |
| ZRSO(2)      | 254bc            | 318 <sup>b</sup>      | 1.26      | 9.32 <sup>a</sup>    | -                           |
| Prime tallow | 264b             | 370 <sup>b</sup>      | 1.40      | 8.04°                | -                           |
| + Deg. RSO   | 244bc            | 326 <sup>b</sup>      | 1.34      | 9.49a                | 8.22                        |
| + Undeg. RSO | 224c             | 322 <sup>b</sup>      | 1.44      | 9.53ª                | 8.80                        |
| + ZRSO       | 222c             | 293 <sup>c</sup>      | 1.32      | 9.25 <sup>a</sup>    | 8.68                        |
| Sx           | 12.8             | 18.4                  |           | 0.12                 |                             |

<sup>(1)</sup> $_{\rm Treatments}$  followed by the same letter are not significantly different (P<0.05)

indicate that some factor(s) in rapeseed oil reduces feed intake and weight gain in turkey poults. The lower feed intake and growth with RSO may be related to the higher M.E. content of these fats as compared with tallow. The fact that tallow resulted in lowered feed intake and growth in relation to the basal diet suggests that the diets to which fat was added contained a surplus of energy in relation to protein and the higher M.E. of RSO may have exaggerated this imbalance.

Although the mixture of fats contained more M.E. than the mean of the components, the degree of improvement was not as great as with the chicks except for degummed RSO. As with the chicks, the zero erucic RSO gave superior performance compared to the the remaining rapeseed products when fed singly. The lower level of synergism as compared with chicks is possibly due to the fact that the M.E. values for these fats were already very high and

<sup>(2)</sup>Crude zero erucic rapeseed oil

thus the mixing of the fat products with tallow would not be expected to enhance the utilization of the RSO products to any large extent. The fatty acid mixture needed for optimum absorption in turkey poults is obviously different from that required in the chick.

### EXPERIMENT 3

The purpose of this experiment was to determine the digestible energy (D.E.) content of the same samples of RSO and RSO foots described in experiment 2 but using rats.

TABLE IV

FAT UTILIZATION BY THE RAT(1)

|                            |                  |                   |                   | Digestible energy          |  |  |
|----------------------------|------------------|-------------------|-------------------|----------------------------|--|--|
| Supplement                 | wt. gain         | feed cons.        | Derived           | Calculated from components |  |  |
|                            | (g)              | (g)               | (Kcal/g)          | (Kcal/g)                   |  |  |
| Basal                      | 117 <sup>b</sup> | 473 <sup>a</sup>  | _                 | _                          |  |  |
| Deg. RSO                   | 142 <sup>a</sup> | 374 <sup>bc</sup> | 8.70°             | _                          |  |  |
| Undeg. RSO                 | 123 <sup>b</sup> | 342 <sup>c</sup>  | 9.03 <sup>b</sup> | _                          |  |  |
| ZRS0(2)                    | 138ª             | 381 <sup>bc</sup> | 9.44 <sup>a</sup> | _                          |  |  |
| Prime tallow               | 165 <sup>a</sup> | 419 <sup>b</sup>  | 8.70°             | _                          |  |  |
| + Deg. RSO                 | 157 <sup>a</sup> | 397 <sup>b</sup>  | 9.06 <sup>b</sup> | 8.70                       |  |  |
| + Undeg. RSO               | 134 <sup>b</sup> | 372 <sup>bc</sup> | 9.13 <sup>b</sup> | 8.92                       |  |  |
| + ZRSO                     | 147 <sup>a</sup> | 387 <sup>bc</sup> | 8.75 <sup>c</sup> | 9.07                       |  |  |
| $\mathbf{S}\mathbf{ar{x}}$ | 12.4             | 14.8              | 0.08              |                            |  |  |

<sup>(1)</sup> Treatments followed by the same letter are not significantly different (P<0.05)

Male Wistar strain weanling rats were randomly allotted into equal lots of 6 rats each and housed in individual, screen-bottomed cages. The rats were fed a practical corn-soybean meal basal diet containing 20% of the fats mentioned in

<sup>(2)</sup>Crude zero erucic rapeseed oil

Experiment 2 for  $^{\prime}$ 1 weeks. Food and water were supplied  $\underline{ad}^{^{c}}$  libitum. During the last week of the experiment excreta samples were collected and subjected to various analyses as indicated previously for M.E. determination.

The digestible energy values derived for the fats and fat mixtures are presented in the form of means in Table IV. Although the D.E. value of tallow and degummed RSO were equal (8.7 kcal/g), the values for the mixed fats were markedly higher than the mean of the component fats. Similarly, a synergistic response in D.E. was observed for the undegummed RSO and tallow mixture. The D.E. value of zero crucic was significantly higher than that of any other fat fed singly; however, no synergistic response was noted by mixing it with tallow.

The results indicate that the rat utilizes the energy in tallow to better advantage than the chick or turkey poult as compared to their utilization of rapeseed oils.

### EXPERIMENT 4

This experiment was designed to determine the reason for the synergism between RSO and tallow in the chick. Because of the indication that RSO is deficient in palmitic acid, the effect of mixing RSO with mixtures of saturated fatty acid was determined. Mixture A contained 43.7% palmitic acid, 48.9% stearic acid, 3.3% myristic acid and 4% others, while Mixture B contained 12% palmitic acid, 80% stearic acid, 2.7% arachidic acid, 2.4% behenic acid, 1.4% lignoceric acid and 1.5% others.

Male White Rock x Vantress chicks that had been reared on commercial starting diets for one week were used. The composition of the basal diet, the housing, randomization and parameters measured were similar to those outlined in Experiment 1. Two levels (2.5 and 5%) of fatty acid mixtures were added to the diets containing RSO and fed to the chicks from 1 to  $^{4}$  weeks of age.

The metabolizable energy values of the degummed RSO as affected by mixing with either tallow or saturated fats are presented in Table V. The M.E. values of mixtures A and B were determined in a separate experiment using the experimental design used here and the values obtained were 0.63 and 0.03 kcal/g respectively. A synergistic response Was noted when RSO was mixed with tallow. Supplementation of degummed RSO with the saturated fatty acid mixtures resulted in an increase in the M.E. content of the fats. The proportions of palmitic and stearic acids did not markedly alter the response. The increase in M.E. was much greater with

TABLE V

SATURATED ACIDS AND FAT UTILIZATION BY THE CHICKEN(1)

|                         |                     |                       |           | Metaboliz         | Metabolizable energy             |
|-------------------------|---------------------|-----------------------|-----------|-------------------|----------------------------------|
| Supplement              | Average<br>wt. gain | Average<br>Feed cons. | Feed/Gain | Derived           | Calculated<br>from<br>components |
|                         | (g)                 | (g)                   |           | (Kcal/g)          | (Kca1/g)                         |
| Basal                   | 51.1b               | 859a                  | 1.68      | l                 | 1                                |
| Prime tallow            | 503b                | 804pc                 | 1.60      | 7.20°             | ı                                |
| Rapeseed oil (Degummed) | 543b                | 753°                  | 1.40      | 7.42b             | ı                                |
| + Prime tallow          | 562a                | 806bc                 | 1.43      | 8.39a             | 7.31                             |
| + Mixture A (15:5)      | 538 <sup>b</sup>    | $841^{\mathrm{b}}$    | 1.56      | p590°2            | 5.72                             |
| + Mixture A (17.5:2.5)  | q905                | 821b                  | 1.62      | 7.48 <sup>b</sup> | 25.9                             |
| + Mixture B (15:5)      | 238b                | $821^{\mathrm{b}}$    | 1.53      | 6.70 <sup>d</sup> | 5.57                             |
| + Mixture B (17.5:2.5)  | 244b                | 824p                  | 1.52      | 7.42 <sup>b</sup> | 6.50                             |
| SX                      | 14.6                | 16.4                  |           | 0.13              |                                  |
|                         |                     |                       |           |                   |                                  |

(1)Treatments followed by the same letter are not significantly different  $(P\langle 0.05)$ 

the higher levels as compared with the lower levels of both mixtures A and B. These results indicate that a part of the synergistic effect found by mixing tallow and RSO is attributable to the presence in the tallow of the saturated fatty acids, palmitic and stearic.

### EXPERIMENT 5

This experiment was designed to compare the utilization of crude and refined RSO. Two different species of RSO were used in this experiment, Brassica napus (B. nap.) which contained 32% erucic acid and Brassica campestris (B. camp.) which contained 23% erucic acid (Table 1).

TABLE 1

FATTY ACID COMPOSITION OF SOME SUPPLEMENTARY FATS

| Fatty<br>acid | RSO<br>foots | Prime<br>tallow | Degummed<br>RSO | Zero erucic<br>RSO | Brassica<br>campestris | Brassica<br>napus |
|---------------|--------------|-----------------|-----------------|--------------------|------------------------|-------------------|
|               | %            | %               | %               | %                  | %                      | %                 |
| 16:0          | 5.5          | 27.0            | 3.2             | 4.0                | 3.2                    | 4.1               |
| 16:1          | 0.7          | 4.2             | 0.2             | 0.3                | 0.4                    | 0.5               |
| 18:0          | 1.8          | 20.9            | 1.5             | 2.2                | 1.2                    | 1.9               |
| 18:1          | 16.6         | 41.4            | 27.4            | 61.3               | 31.6                   | 23.7              |
| 18:2          | 15.6         | 4.8             | 18.3            | 20.0               | 19.3                   | 16.0              |
| 18:3          | 8.7          | 1.5             | 8.1             | 9.7                | 10.1                   | 6.9               |
| 20:1          | 8.2          | -               | 11.9            | 1.1                | 11.2                   | 14.9              |
| 22:1          | 42.9         | -               | 29.3            | 1.2                | 23.0                   | 32.2              |

The findings in previous experiments indicated that rapeseed gums may influence the M.E. content of RSO, therefore different levels of rapeseed gums were added to determine its influence on the utilization of RSO as a source of energy.

The experimental procedures were similar to those employed in the prevous experiment. Day old, male, White Leghorn chicks were used in this trial. Five different levels, 0.1, 0.2, 0.4, 0.8, 1.6%, of rapeseed gums were added, at the expense of the

basal, to diets containing 20% refined B. camp. RSO. The birds received the experimental diets from 2-4 weeks of age.

The data presented in Table VI indicate that refining significantly decreased the M.E. content of both B. camp. and B. nap. RSO, with the magnitude of depression being greater for B. camp. RSO. The results indicate that the M.E. content of zero erucic RSO was lower than that of crude B. nap. or crude B. camp. RSO. However, it should be emphasized that crude zero erucic RSO was used in the previous trial while refined zero erucic RSO was used in the present experiment. The depression in M.E. was not attributable to the oxidation of the oil samples during refining since the determined peroxide values for the crude and refined oils were essentially the same. It appears that some other factor(s) in the refining process affected the nutritional value of rapeseed oil.

TABLE VI

FAT UTILIZATION BY THE CHICKEN (1)

|  |                                      |                                      |                                      | Metaboliz   | able energy                      |
|--|--------------------------------------|--------------------------------------|--------------------------------------|---|----------------------------------|
| Supplement   | Average<br>wt.gain                   | Average<br>feed<br>cons.             | Feed/Gain                            | Derived   | Calculated<br>from<br>components |
|  | (g)                                  | (g)                                  |                                      | (Kcal/g)  | (Kcal/g)                         |
| Basal<br>Crude B. camp RSO<br>Refined B. camp RSO<br>Crude B. nap. RSO<br>Refined B. nap. RSO<br>Refined Zero erucic | 170 <sup>a</sup><br>160 <sup>a</sup> | 401a<br>308b<br>318b<br>321b<br>335b | 2.37<br>1.88<br>2.28<br>1.87<br>2.10 | 9.10 <sup>a</sup> 6.74 <sup>d</sup> 8.96 <sup>a</sup> 6.97 <sup>b</sup> 7.09 <sup>d</sup> | -<br>-<br>-<br>-                 |
| RSO<br>Tallow  | 165 <sup>a</sup><br>164a             | 347°<br>331b                         | 2.11<br>2.02                         | 6.30e   | _                                |
| + Refined B. camp.  RSO + Refined B. nap.  | 158a                                 | 308b                                 | 1.94                                 | 8.10bc  | 6.52                             |
| RSO  | 165 <sup>a</sup>                     | 319 <sup>b</sup>                     | 1.93                                 | 8.33 <sup>b</sup>   | 6.64                             |
| + Refined zero<br>erucic RSO   | 173 <sup>a</sup>                     | 325 <sup>b</sup>                     | 1.87                                 | 7.90 <sup>e</sup>   | 6.70                             |
| Sx   | 5.4                                  | 12.8                                 |                                      | 0.16  |                                  |

<sup>(1)</sup> Treatment followed by the same letters are not significantly different (P $\langle 0.05 \rangle$ )

The results show that refined B. camp., B. nap. and zero erucic RSO, when mixed with tallow, exerted a beneficial effect on M.E. Though a significant increase in M.E. was noted for the mixture of tallow and refined B. nap. RSO, and tallow and refined B. camp. RSO, the M.E. values of the respective crude oils were still higher than the mixtures. The results also indicate that refining reduced weight gain and feed efficiency with both B. camp. and B. nap. RSO.

The incorporation of rapeseed gums into diets containing RSO significantly increased the M.E. content of refined B. camp. RSO (Table VII). On the other hand, weight gain and feed consumption were not significantly affected by the levels of gums employed. The maximum increase in M.E. was noted with the highest level of rapeseed gums (1.6%); however, there was no significant difference in M.E. between the other levels of rapeseed gums. In calculating the increase in M.E. due to the addition of gums, the gums were considered to have an M.E. equivalent to the gross energy of this product. Thus, the response to the gums is at least as great as the response shown here. In spite of the increase in M.E. value of refined B. camp. RSO by the supplementation with gums, the M.E. value of crude B. camp. RSO was much greater than refined oil containing the highest level of gums. Therefore, the removal of gums from the rapeseed oil was not the major factor in the poor utilization of refined RSO.

TABLE VII

INFLUENCE OF GUM ON UTILIZATION OF RAPESEED OIL (1)

| Supplement  | Average<br>wt.gain                          | 1   | Feed/Gain  | M.E. of<br>RSO  | Increase in M.E.         |
|---|---|---|--|---|--------------------------|
| Basal Crude B. Camp. RSO Refined B. Camp. RSO + 0.1% rapeseed gum + 0.2% rapeseed gum + 0.4% rapeseed gum + 0.8% rapeseed gum + 1.6% rapeseed gum | (g) 170a 162a 139b 146b 142b 141b 145b 145b | (g) 401a 308b 318b 322b 318b 309b 316b 303b | 2.34<br>1.88<br>2.28<br>2.21<br>2.24<br>2.20<br>2.17<br>2.31 | (Kcal/g)  - 9.10a 6.74c 7.20bc 7.17bc 7.12bc 7.12bc 7.42b | (%) 6.9 6.4 5.7 5.6 10.0 |
| $S\overline{x}$   | 5.4   | 12.8  |  | 0.16  |                          |

<sup>(1)</sup> Treatment followed by the same letter are not significantly different (P<0.05)

### EXPERIMENT 6

This experiment was carried out in order to compare the reproductive performance of laying hens when fed B. camp. and B. nap. RSO (crude and refined), zero erucic RSO, RSO foots, corn oil and tallow. The M.E. content of the oils was also determined with the laying hens fed 20% of the fats.

White Leghorn hens of approximately 26 weeks of age were used in the study. The birds were fed a commercial ration for four weeks before being placed on the experimental diets. Fifteen groups of 8 hens each were equalized in egg production and randomly assigned to the various dietary treatments. The birds were fed a practical corn-soybean meal diet or this diet containing 10 or 20% of B. camp. or B. nap. (crude and refined), RSO foots, tallow and corn oil. Because a sufficient amount of zero erucic RSO was not available, this oil was tested only at the 10% level. The hens were maintained on the diets for 12 weeks during which individual bird egg production and egg weight records were kept. During the last four weeks of the experiments pullets were artificially inseminated with the semen from cockrels fed a commercial diet and the eggs incubated.

TABLE VIII

INFLUENCE OF DIETARY FAT ON EGG WEIGHT AND EGG PRODUCTION

|  | Av. eg  | g wt.   | Av.egg produ   | uction(HDB)  |
|--|---|---|--|--|
| Supplement   | 10% oil   | 20% oil   | 10% oil  | 20% oil  |
|  | %   | %   | %  | %  |
| None Crude B. Camp. RSO Refined B. Camp. RSO Crude B. Nap. RSO Refined B. Nap. RSO RSO foots Refined zero crucic RSO Corn oil Tallow | 54.7±0.4 <sup>(1)</sup> 51.8±1.0 50.7±0.6 47.2±0.4 49.1±0.7 52.5±0.9 54.5±0.6 55.5±0.4 54.5±1.1 | 54.7±0.4<br>46.1±1.4<br>43.6±1.5<br>45.5±0.2<br>44.1±0.4<br>48.3±1.0<br>-<br>55.2±0.6<br>53.3±1.0 | 86.8±3.2<br>79.2±5.5<br>73.4±3.5<br>76.3±3.4<br>76.2±3.7<br>78.9±4.3<br>83.1±4.3<br>85.6±6.0<br>79.5±3.8 | 86.8±3.2<br>72.2±3.0<br>57.3±6.7<br>63.3±3.0<br>50.4±5.5<br>69.2±2.9 |

<sup>(1)</sup>Mean ± standard error of the mean

The results (Table VIII) indicate that the inclusion of 10 or 20% B. camp. or B. nap. RSO decreased the rate of egg production. At the 20% level both of the refined oils showed a decrease in egg production as compared with crude RSO; however, at the 10% level the decrease in egg production was observed only with the B. camp. RSO. The diets containing RSO foots also showed lower egg production as compared with the no-fat control. Birds fed zero erucic RSO showed significantly higher egg production than those fed B. camp. or B. nap. It would appear that the decrease in egg production with RSO was due to erucic acid.

Egg weights showed essentially the same trend as did egg production. After 12 weeks on the experimental diets, the hens receiving corn oil maintained slightly higher egg weights than those receiving the no-fat basal ration. Eggs from the hens fed either 10 or 20% B. camp. or B. nap. RSO or RSO foots were significantly lower in weight than eggs from hens fed the remaining diets. Average egg weight was greater for diets containing 10% zero erucic RSO than for the corresponding diets with regular rapeseed oits. It would appear that erucic acid is responsible for the major part of the egg weight reduction obtained with B. camp. RSO, B. nap. RSO and RSO foots. It is of interest that egg weight declined within a few days of the hens being placed on regular rapeseed oils or RSO foots.

TABLE IX

INFLUENCE OF DIETARY FAT ON HATCHABILITY OF TOTAL EGGS AND FERTILE EGGS

| Supplement   | Hatchability of total eggs 10% oil 20% oil  |   | Hatchab<br>of ferti<br>10% oil  |   |
|--|---|---|---|---|
| None Crude B. Camp. RSO Refined B. Camp. RSO Crude B. nap. RSO Refined B. nap. RSO RSO foots Refined Zero erucic RSO Corn oil Tallow | (%) 71.6±3.3 <sup>(1)</sup> 65.7±4.0 59.6±2.7 63.5±4.9 76.9±1.9 72.5±1.9 80.3±1.8 76.3±3.2 70.4±5.2 | (%) 71.6±3.3 66.9±3.6 26.4±2.2 31.2±4.0 56.1±4.1 44.6±5.4 | (%) 95.5±1.8 83.6±2.3 78.4±2.2 86.0±4.5 93.4±2.8 86.7±1.6  92.6±2.0 93.7±1.2 86.8±3.2 | (%) 95.5±1.8 86.8±4.2 70.4±4.6 76.3±4.8 79.0±1.0 68.3±4.3 |

<sup>(1)</sup> Mean ± Standard error of the mean

Hatchability of fertile eggs as well as of total eggs is presented in Table IX. A significant decrease in the hatchability of both fertile and total eggs was noted with hens fed crude and refined B. camp RSO, crude B. nap RSO and RSO foots. However, the hatchability of eggs from hens fed refined B. nap. RSO was not affected. A significant difference in hatchability between the levels of RSO was observed. All the rapeseed oils and RSO foots showed a lower hatchability at the 20% than at the 10% level. The highest hatchability of total eggs was observed for zero erucic RSO. In general, the results indicate that hatchability was higher for control diets as compared with diets supplemented with erucic acid containing RSO and RSO foots.

The M.E. values (Table X) indicate that rapeseed oils and RSO foots were reasonably well utilized by laying hens. However, as in the case of chicks, the refining process reduced the M.E. values particularly that of the B. Camp. RSO.

TABLE X

FAT UTILIZATION BY THE LAYING HEN(1)

| Supplement           | Metabolizable<br>energy(2) |  |  |
|----------------------|----------------------------|--|--|
| Crude B. camp. RSO   | 8.53a                      |  |  |
| Refined B. camp. RSO | 7•73 <sup>b</sup>          |  |  |
| Crude B. nap. RSO    | 8.88a                      |  |  |
| Refined B. nap. RSO  | 8.143a                     |  |  |
| RSO foots            | 7.70 <sup>b</sup>          |  |  |
| Corn oil             | 9.00a                      |  |  |
| Tallow               | 7.89b                      |  |  |
|                      |                            |  |  |

<sup>(1)</sup> Treatment followed by the same letter are not significantly different (P<0.05)

<sup>(2)</sup> The standard error of each of these values, which are means of 4 determinations, is 0.24 kcal/g

### SUMMARY AND CONCLUSIONS

When fed to chicks, turkey poults and rats, zero erucic acid RSO gave superior growth and feed efficiency and was a superior source of energy as compared with regular rapeseed oil. When included in the diets of Laying hens, B. napus and B. campestris rapeseed oils exerted a depressing effect on egg production, egg weight and hatchability. This suppression of reproductive performance was completely overcome when zero erucic acid rapeseed oil replaced the regular B. napus and B. campestris oils, suggesting that erucic acid is the detrimental factor involved in reproduction.

White rapeseed oils and rapeseed oil foots were reasonably well utilized as source of energy by chicks, poults and rats, there was a synergistic effect in energy utilization when rapeseed oils were mixed with tallow. Thus the energy content of the mixture of rapeseed oil and tallow was greater than that anticipated from the fats fed singly. Chicks fed mixtures of palmitic and stearie acids along with rapeseed oil, demonstrated similar synergism in fat utilization to those fed tallow with rapeseed oil. These results confirm and extend previous findings indicating that rapeseed oil is imbalanced in fatty acid make-up for maximum utilization, being too low in the long chain saturated fatty acids, palmitic and stearic. While synergism in energy utilization was also demonstrated when zero erucic acid oil was mixed with tallow, the effect was less than with regular B. napus and B. campestris oils. This is probably explained by the fact that zero erucic acid rapeseed oil was higher in energy value when fed singly than erucic acid-containing oils.

When fed to chicks, refined regular B. napus and B. campestris oils were markedly lower in M.E. content and gave poorer growth and feed efficiency than the corresponding crude oils. Similarly with laying hens, refining of the oils produced detrimental effects on energy utilization and reproductive performance. The evidence indicates that the removal of the gums present in the crude oils is partially responsible for the reduction in the M.E. value of the oils. However, this is not the major factor and experiments are now in progress in our laboratory to determine the effect of each step in the refining process on the nutritional value of rapeseed and other oils.

# DEPOSITION OF ERUCIC ACID IN THE TISSUES OF YOUNG ANIMALS

In the initial experiment of this study, male weanling rats were maintained for 12 weeks on semisynthetic diets containing either 2% corn oil or 2% corn oil plus 10% ethyl erucate, the erucate

being substituted for carbohydrate (dextrose) on a weight basis. No attempt was made to render the diets isocaloric. The object of this experiment was to determine the extent to which erucic acid was deposited in the tissues and corn oil was added to the diet as a source of essential fatty acids.

At the termination of the experiment, the rats were anesthetized with ether and exsanguinated. The tissues examined were the adrenals, brain, heart, kidney, liver, plasma, prostate, red cells and testes. Tissue lipids were extracted with chloroform-methanol and transesterified with boron fluoride-methanol. The esters were purified by thin-layer chromatography and subjected to gasliquid chromatography on a 200 x 0.2 cm column of 15% ethyleneglycol adipate operated isothermally at 210°C. In this experiment, tissues from three animals from each dietary group were analysed and the results were expressed as percent total fatty acid

| Acid  | Corn<br>oil                        | Corn oil-<br>ethyl<br>erucate <sup>(2)</sup>  | Olive<br>oil                               | Rapeseed<br>oil<br>(B.campestris)                 | Zero-erucic<br>rapeseed<br>oil (Canbra)   |
|---|------------------------------------|---|--|---|---|
| 16:0 <sup>(3)</sup> 18:0 18:1 18:2 18:3 20:1 22:1 | 10.2<br>2.0<br>25.7<br>59.4<br>1.7 | 2.3<br>0.4<br>5.6<br>10.4<br>0.3<br>-<br>81.0 | 11.1<br>2.7<br>76.6<br>7.9<br>0.5<br>Trace | 2.8<br>1.1<br>33.4<br>18.8<br>8.6<br>10.6<br>24.6 | 4.3<br>1.9<br>61.2<br>19.8<br>11.0<br>0.7 |

- (1)% Total fatty acids. Minor constituents omitted from the table
- (2) Erucic acid kindly donated by Canada Packers Ltd., Toronto, Ontario.
- (3)Abbreviation for fatty acids: X:Y Z, where X and Y represent the number of carbon atoms and double bonds respectively, and Z represents the number of carbon atoms between the terminal double bond and the methyl end of the molecule.

The fatty acid compositions of the two dietary fats are presented in Table XI. The diets contained 1.2% by weight (approximately 2.4% of total calories) of linoleic acid, more than enough to meet the essential fatty acid requirements of the young rat (1%

of total calories). The corn oil-erucate diet contained about 19% of the total calories as ethyl erucate.

The tissues examined can be divided into three groups according to the degree of deposition of erucic acid in them. Those containing over 5% erucic acid were the adrenals, plasma and heart. Erythrocytes, kidney and liver contained between 1 and 5% erucic acid. Less than 1% of this acid was detected in the brain, prostate and testes lipids. The overall fatty acid compositions of these tissues are presented in Tables XII - XIV.

TABLE XII

FATTY ACID COMPOSITIONS (1) OF ADRENAL, PLASMA AND HEART LIPIDS (EXPERIMENT 1)

|  | Adren         | als             | Plas          | ma              | Hea           | Heart                                       |  |
|--|---------------|-----------------|---------------|-----------------|---------------|---|--|
|  | Corn oil      | Corn-<br>erucic | Corn oil      | Corn-<br>erucic | Corn oil      | Corn-<br>erucic                             |  |
| 16:0   | 15.5          | 15.1            | 17.1          | 16.9            | 14.5          | 12.0  |  |
| 16:1<br>18:0                                   | 5.0<br>15.5   | 2.8<br>9.5      | 5.0<br>9.6    | 2.3<br>9.5      | 2.9<br>16.3   | 1.1<br>17.2                                 |  |
| 18:1   | 23.9          | 23.3            | 21.0          | 22.7            | 19.9          | 19.1<br>16.5                                |  |
| 18:2<br>20:1                                   | 3.2<br>Trace  | 3.7<br>5.5      | 12.0<br>Trace | 11.6            | 17.0<br>Trace | 1.9   |  |
| 20:4 <b>6</b> 6<br>22:1                        | 22.4<br>Trace | 10.5            | 25.7<br>Trace | 22.9<br>9.7     | 18.4<br>Trace | $\begin{bmatrix} 21.0 \\ 8.1 \end{bmatrix}$ |  |
| 22 <b>:</b> /ιω6                               | 6.9           | 11.11           | 0.3           | Trace           | 1.1           | -   |  |
| 22 <b>: 5.6</b> 6<br>22 <b>:</b> 6 <b>.</b> 33 | 3.6           | 0.9<br>Trace    | 6.0<br>0.5    | 0.2             | 4.9<br>1.4    | 0.2   |  |
| 24:1   | Trace         | 1.7             |               | -               | _             | _   |  |

<sup>(1)</sup> See footnotes Table X1.

High levels of erucic acid were found in the adrenals of rats receiving dietary erucate (Table XII). This acid accounted for over 20% of the total fatty acids in adrenal lipids. Moreover, these lipids also contained over 5% of eicosenoic acid, presumably derived by \$\beta\$-oxidation of erucic acid. This data is consistent with that of Carroll (1962) who found that dietary erucic acid accumulated in the adrenal cholesterol esters. As might be expected, significant levels of erucic acid (9.7%) were found in plasma. Again this is consistent with previous work (Carroll 1962) where this acid was found to be relatively abundant in plasma triglycerides, a reflection, no doubt, of the transport of absorbed crucic acid from the intestine to the liver and other

tissues. The heart was the only other tissue in which erucic acid constituted more than 5% of the total fatty acids.

TABLE XIII

FATTY ACID COMPOSITIONS (1) OF ERYTHROCYTES, KIDNEY AND LIVER LIPIDS (EXPERIMENT 1)

|                 | Erythroc    | ytes        | Kidn        | ey          | Liv         | er          |
|-----------------|-------------|-------------|-------------|-------------|-------------|-------------|
|                 | Corn oil    | Corn-       | Corn oil    | Corn-       | Corn oil    | Corn-       |
|                 |             | erucic      |             | erucic      |             | erucic      |
| 16:0            | 22.1        | 02.0        | 20.0        | 01.0        | 01.0        | 01.0        |
| 16:1            | 22:1<br>2.3 | 23.0<br>0.6 | 20.0<br>3.7 | 21.9<br>3.0 | 21.0<br>5.4 | 21.0<br>2.8 |
| 18:0            | 14.4        | 14.7        | 15.2        | 14.6        | 13.8        | 15.0        |
| 18:1            | 14.0        | 13.1        | 23.0        | 21.6        | 23.1        | 26.6        |
| 18:2            | 5.7         | 5.7         | 6.5         | 8.9         | 8.4         | 6.6         |
| 20:1            | Trace       | 1.2         | Trace       | 1.5         | Trace       | 1.2         |
| 20:4 <b>0</b> 6 | 23.8        | 22.3        | 24.1        | 22.8        | 18.6        | 17.7        |
| 22:1            | Trace       | 4.7         | Trace       | 3.7         | Trace       | 3.3         |
| 22:4 <b>ω</b> 6 | 2.5         | 2.8         | 0.4         | Trace       | 0.5         | Trace       |
| 22:5 <b>ω</b> 6 | 8.0         | 5.7         | 2.7         | Trace       | 5.0         | 2.5         |
| 22:6w3          | 0.3         | Trace       | 0.5         | -           | 1.6         | 1.1         |
| 24:1            | 0.4         | 1.5         | -           | -           | -           | -           |

(1) See footnotes Table XI.

Erythrocytes (4.7%), kidney (3.7%) and liver (3.3%) exhibited a moderate tendency to incorporate dietary erucic acid (Table XIII). This was somewhat surprising in the case of the erythrocytes since they generally will not incorporate significant amounts of non-physiological fatty acids.

The final group of tissues, brain, prostate and testes, failed to incorporate more than 1% of dietary erucic acid (Table XIV). This is not unexpected in the case of the brain lipids, since they are established in the pre-natal and immediate post-natal period and show very little susceptibility to dietary modification. However, the lack of erucic acid in the testes was somewhat unexpected. There have been reports in the literature (Noble and Carroll 1961) of dietary erucic acid causing testicular damage and sterility in rats and it was for this reason that these tissues were examined. It is possible that the length of the present experiment was too short for deleterious effects to be manifest. Moreover, microscopic examination of the epididymal fat pads revealed the presence of live spermatazoa, indicating that the animals were probably fertile.

FATTY ACID COMPOSITIONS (1) OF PROSTATE, TESTES AND BRAIN LIPIDS (EXPERIMENT 1)

|                                    | Prost         | ate             | Test         | es              | Bra           | in              |
|------------------------------------|---------------|-----------------|--------------|-----------------|---------------|-----------------|
|                                    | Corn oil      | Corn-<br>erucic | Corn oil     | Corn-<br>erucic | Corn oil      | Corn-<br>erucic |
| 16:0<br>16:1                       | 21.4<br>4.2   | 25.0<br>2.7     | 25.6<br>9.0  | 31.9            | 13.6<br>0.8   | 18.2            |
| 18:0<br>18:1                       | 8.3<br>28.8   | 9.4<br>33.L     | 4.6          | 6.3<br>15.8     | 19.1<br>21.0  | 21.1<br>23.6    |
| 18:2<br>20:1                       | 6.1<br>Trace  | 4.9<br>1.6      | 9.3<br>Trace | 3.2<br>0.3      | 1.0<br>Trace  | 0.2<br>2.8      |
| 20 : 4 <b>ω</b> 6<br>22 : 1        | 12.6<br>Trace | 12.6            | 8.2<br>Trace | 14.2<br>0.6     | 10.8<br>Trace | 10.5            |
| 22:4 <b>0</b> 6<br>22:5 <b>0</b> 6 | 0.9<br>2.8    | 1.3             | 1.1          | 21.5            | 3.3<br>6.9    | 2.9             |
| 22:6 <b>-</b> 3<br>24:1            | 0.1           | Trace<br>-      | 0.4          | 0.9             | 0.4           | 10.9            |

(1) See footnotes, Table XI.

In general, erucic acid was deposited in the tissues at the expense of the polyunsaturated acids derived from linoleic acid. Both arachidonic and 22:4 6, the major polyunsaturated acids, were decreased in the adrenals by feeding crucic acid. There was also a decrease in the 22:546 and 22:643 acids. However, this pattern was not followed exactly by the other tissues examined. In general, deposition of crucic acid in a tissue had very little effect on the arachidonic acid content of the tissue lipids. major effect was the replacement of the higher polyunsaturated fatty acids by erucic acid. Thus, decreases were usually noted in the 22:46 and 22:56 acids. A rather unusual effect was من noted in the testicular lipids, where both arachidonic and 22:5ه acids were increased when erucic acid was fed. The reason for this is not readily apparent, although it is possible that the decrease in these acids in these other tissues as a result of erucic acid deposition might result in their ready availability for incorporation in the testicular lipids where very little erucic acid was deposited. These results may also be due to the age of the animals employed. At three months, they would have just reached sexual maturity. It is known that the polyunsaturated fatty acid content of the testes does change as the animal achieves sexual maturity (Davis et al. 1966).

In a second experiment, male weanling rats were maintained on semi-synthetic diets containing 12% fat. Four different fats were employed, olive oil, corn oil-ethyl erucate (1:5), rapeseed oil (Brassica campestris) and zero-erucic acid rapeseed (Canbra). The duration of the experiment was 18 weeks and the animals were sacrificed and the tissues extracted as in the first experiment. In this experiment the prostate was omitted and the spleen was included in the study. Tissues from 5 animals from each dietary group were examined.

The object of the second experiment was to determine to what extent erucic acid was deposited in rat tissues when present as a constituent of rapeseed oil. In addition, the general effects of rapeseed oils (campestris and Canbra) on tissue lipids were investigated. Finally, by including olive oil and the corn oilethyl erucate diets, the comparative effects of oleic and erucic acids could be studied.

The fatty acid compositions of the four dietary fats are included in Table XI. Olive oil contained more saturated acids than the corn oil-erucic mixture, was slightly lower in linoleic acid and the oleic acid content was slightly lower than the erucic content of the other fat. However, it was considered to be a suitable control for the corn oil-erucic acid mixture and provided a means of comparing the dietary effects of oleic and erucic acid. The major differences between the two rapeseed oils were the lower levels of erucic and eicosenoic acids in the Canbra oil, and the higher level of oleic acid. Both fats were characterized by relatively low linoleic-linolenic acid ratios.

In general, when the corn oil-erucic acid diet was fed, the levels of erucic acid were slightly lower in rat tissues in the second experiment than in the initial experiment. The major exception was noted in the testes lipids. After 18 weeks on this diet, almost 5% of this acid had accumulated as compared with the 0.6% found in this tissue after 12 weeks. A slightly higher concentration of erucic acid was found in brain lipids in the second experiment but the level in this tissue was still less than 1%.

The deposition of erucic acid in the tissues followed the same pattern in the second experiment as it did in the first. The highest level of erucic acid was noted in the adrenals of rats fed the corn oil-ethyl erucate mixture and over 6% of this acid was found in the plasma and heart (Table XV). Spleen, testes and kidney contained 3 - 5% of this acid (Table XVI), red cells 3%, liver 2% and brain less than 1% (Table XVII). Low levels of erucic acid were noted in both the adrenals and brain of rats receiving the olive oil diet but this only amounted to about

TABLE XV

FATTY ACID COMPOSITIONS (1) OF ADRENAL, PLASMA AND HEART LIPIDS (EXPERIMENT 2)

|                 |       | Adrenals        | 8 . 5 |      |              | Plasma          | sma   |       |              | Heart           | ır t |       |
|-----------------|-------|-----------------|-------|------|--------------|-----------------|-------|-------|--------------|-----------------|------|-------|
|                 | Olive | Corn-<br>erucic | RSO   | ZRSO | Olive<br>oil | Corn-<br>erucic | RSO   | ZRSO  | Olive<br>oil | Corn-<br>erucic | RSO  | ZRSO  |
| 16:0            | 10.7  | 12.1            | 13.1  | 11.4 | 16.9         | <b>2.7</b>      | 16.1  | 15.0  | 13.2         | 14.2            | 10.5 | 11.4  |
| 16:1            | 1.5   | 2.6             | 2.3   | 2.0  | 3.4          | 2.51            | 2.8   | 2.0   | L.3          | 2.]             | L.3  | 1.6   |
| 18:0            | 13.5  | 9.5             | 11.0  | 17.8 | 6.2          | 11.1            | 9.1   | 7.8   | 16.2         | 13.6            | 15.2 | 15.4  |
| 18:1            | 31.6  | 20.8            | 28.3  | 28.7 | 42.5         | 17.8            | 27.8  | 37.0  | 30.2         | 20.9            | 21.8 | 29.4  |
| 18:2            | 3.4   | 5.3             | 5.5   | 2.2  | 8.1          | 12.6            | 13.3  | 15.5  | 8.8          | 16.5            | 16.5 | 15.7  |
| 18:3            | Trace | '               | 1.5   | 3.8  | Trace        | 0.1             | 1.6   | 3.0   | 0.1          | Trace           | 1.2  | 1.2   |
| 20:1            | 2.0   | 5.0             | 9.9   | 2.1  | ŧ₁0          | 0.8             | 3.3   | 0.8   | 0.7          | 2.0             | 3.6  | 9.0   |
| 20:4 <b>0</b> 6 | 9.91  | 13.1            | 6.2   | 16.2 | 15.9         | 24.5            | 18.4  | 12.7  | 5.71         | 1.71            | 17.7 | 12.2  |
| 22:1            | 1.1   | 15.1            | 11.7  | 1.2  | 1            | 7.8             | 2.9   | 6.0   | ı            | 4.9             | 3.1  | 0.1   |
| 24:1            | 1     | 3.2             | 2.8   | 9.0  | I            | 1               | 1     | ·—    | !            | ı               | ı    | ı     |
| 22:46           | 10.2  | 6.9             | 4.7   | 5.6  | 0.3          | 6.0             | 0.3   | Trace | 1.2          | 0.8             | 9.0  | 9.0   |
| 22:5 <b>u</b> 6 | 2.2   | 2.3             | Trace | 0.5  | 0.8          | 1.9             | Trace | Trace | 1.8          | 3.0             | 4.0  | Trace |
| 22:503          | 0.8   | Trace           | 1.2   | 2.3  | ı            | ı               | 0.3   | 9.0   | ı            | 0.5             | 1.0  | 1.6   |
| 22:6 <b>u</b> 3 | 1.3   | 0.8             | 1.2   | 2.3  | 1.7          | 1.4             | 1,6   | 2.4   | 6.4          | 1.4             | 4.9  | 6.2   |
|                 |       |                 |       |      |              |                 |       |       |              |                 |      |       |

(1) See footnotes Table XI

TABLE XVI

FATTY ACID COMPOSITIONS<sup>(1)</sup> OF SPLEEN, KIDNEY AND LIVER LIPIDS (EXPERIMENT 2)

|                 |              | Spleen          | en    |       |              | Kidney          | 3y   |       |              | Liver           | ı     |       |
|-----------------|--------------|-----------------|-------|-------|--------------|-----------------|------|-------|--------------|-----------------|-------|-------|
|                 | Olive<br>oil | Corn-<br>erucic | RSO   | ZRSO  | Olive<br>oil | Corn-<br>erucic | RSO  | ZRSO  | Olive<br>oil | Corn-<br>erucic | RSO   | ZRSO  |
| 16:0            | 18.7         | 23.7            | 17.6  | 9•91  | 4.81         | 22.2            | 18.7 | 18.3  | 18.2         | 23.1            | 17.9  | 18.5  |
| 16:1            | 2.6          | 5.3             | 2.8   | 2.4   | 2.8          | 9.4             | 3.4  | 3.4   | 3.1          | 3.5             | 3.0   | 2.4   |
| 18:0            | 4.6          | 8.3             | 6-2   | 8.9   | 11.4         | 11.4            | 10.3 | 4-7   | 12.4         | 14.6            | 12.5  | 12.6  |
| 18:1            | 46.3         | 27.8            | 31.8  | 6.04  | 40.8         | 25.7            | 29.8 | 44.8  | 37.4         | 24.8            | 28.6  | 30.5  |
| 18:2            | 5.1          | 7.5             | 10.2  | 11.9  | 6.2          | 4.6             | 10.7 | 12.3  | 5.6          | 2.2             | 9.6   | 11.6  |
| 18:3            | 0.1          | Trace           | 2.0   | 2.5   | Trace        | Trace           | 1.4  | 2.3   | ı            | 0.1             | 1.3   | 1.3   |
| 20:1            | 2.0          | 2.7             | 2.9   | 1.2   | 0.2          | 1.7             | 3.6  | 2.0   | 4.0          | 1.1             | 2.4   | 9.0   |
| 20:4 <b>~</b> 6 | 12.2         | 12.3            | 10.8  | 9.5   | 17.0         | 17.1            | 14.9 | 7.3   | 15.6         | 16.2            | 1,4.4 | 12.5  |
| 22:1            | 1            | 5.4             | 9.4   | 2.0   | ı            | 3.7             | 2.3  | 0.3   | ı            | 2.5             | 1,3   | 0.2   |
| $2^{h} : 1$     | 1            | J               | ı     | 1     | 1            | ı               | 1    | ı     | 1            | 1               | ı     | ŧ     |
| 22:4~6          | 1.6          | 2.4             | 1.4   | 1.1   | 0.5          | 9.0             | 0.5  | 0.3   | 0.2          | 9.0             | 0.5   | 0.2   |
| 22:5%           | 9.0          | 1.8             | Trace | Trace | 9.0          | 1.3             | 0.2  | Trace | 1.4          | 3.1             | 0.5   | Trace |
| 22:563          | ı            | ı               | 2.0   | 1.0   | ı            | J               | 4.0  | 0.1   | 1            | ı               | 6.0   | 1.1   |
| 22:663          | 1.1          | 1.4             | 1.0   | 1.1   | H•H          | 0.5             | 1.4  | 6.0   | 3.3          | 1.1             | 9.4   | 9.6   |
|                 |              | _               | _     |       |              |                 | _    |       | _            |                 | _     |       |

(1) See footnotes Table XI.

TABLE XVII

FATTY ACID COMPOSITIONS<sup>(1)</sup> OF RED CELL, TESTES AND BRAIN LIPIUS (EXPERIMENT 2)

|                 |              | RI              | RBC   |       |              | Testes          | e<br>S    |      |              | Bra             | Brain     |          |
|-----------------|--------------|-----------------|-------|-------|--------------|-----------------|-----------|------|--------------|-----------------|-----------|----------|
|                 | Olive<br>oil | Corn-<br>erucic | RSO   | ZRSO  | Olive<br>oil | Corn-<br>erucic | RSO ZRSO  | ZRSO | Olive<br>oil | Corn-<br>erucic |           | RSO ZRSO |
| 16:0            | 23.6         | 24.4            | 21.7  | 24.7  | 21.1         | 22.7            | 20.0 21.3 | 21.3 | 18.7         | 18.7            | 16.2      | 18.7     |
| 16:1            | 1.2          | 1.4             | 1.9   | 0.8   | 3.8          | 7.4             | 4.4       | 3.0  | 9.0          | 1.3             | 0.1       | 0.5      |
| 18:0            | 15.3         | 14.3            | 13.9  | 15.9  | 3.4          | 3.1             | 3.4       | 4.1  | 20.1         | 19.0            | 19.6      | 21.5     |
| 18:1            | 19.3         | 13.3            | 16.7  | 19.4  | 43.2         | 31.8            | 30.7      | 34.1 | 25.8         | 24.1            | 25.0      | 26.3     |
| 18:2            | ਦ•€          | 6.5             | 6.2   | 8.3   | 4.4          | 7.9             | 9.2       | 9.3  | 0.3          | 2.0             | 0.8       | 5.       |
| 18:3            | 1            | ı               | 0.5   | 2.0   | 0.1          | Trace           | 2.1       | 1.5  | 0.3          | 0.2             | 0.2       | 0.1      |
| 20:1            | 0.4          | 0.8             | 2.4   | 4.0   | 0.3          | 3.4             | 3.5       | 4.0  | 3.7          | 2.7             | 3.0       | 3.3      |
| 20:4 <b>w</b> 6 | 25.5         | 26.2            | 22.9  | 21.5  | 7.2          | 7.5             | 7.5       | 8.1  | 9.8          | 12.2            | 10.2      | 6.6      |
| 22:1            | 1            | 3.3             | 2.2   | 9.0   | ı            | 4.9             | 2.1       | 0.2  | 0.5          | 6.0             | 0.7       | 0.2      |
| 24:1            | ì            | ı               | ı     | (     | ı            | ı               | ı         | ı    | ı            | ı               | ı         | 1        |
| 22:406          | 2.5          | 3.2             | 1.8   | 1.0   | 1.3          | 0.1             | 1.6       | 1.3  | 3.4          | 3.8             | 3.4       | 3.0      |
| 22:5₩6          | 1.1          | 2.3             | Trace | Trace | 6.6          | 8.2             | 9.5       | 10.1 | 1.6          | 1.1             | 0.5       | Trace    |
| 22:503          | 6.0          | Trace           | 1.4   | 1.9   | ı            | J               | 1         | ı    | ı            | 1               | 2.0       | 0.1      |
| 22:603          | 2.2          | 1.4             | 2.6   | 2.5   | 0.8          | 1,.0            | 1.1       | 1.1  | 13.5         | 10.3            | 15.4 12.8 | 12.8     |
|                 | _            | _               | _     | _     | _            |                 |           |      |              |                 |           |          |

(1) See footnotes Table XI.

0.5 - 1% of the total fatty acids. Less erucic acid accumulated when campestris oil was fed to the rats. Except for the adrenals which contained 12% of this acid, the tissue lipids accumulated less than 5% erucic acid. Less than 1% erucic acid was found in the tissues of rats maintained on the Canbra oil, and in most instances less than half of this amount was found. The deposition of erucic was thus found to parallel the dietary level of this acid.

The inclusion of erucic acid in the diet was not only manifest by the deposition of this acid in the tissues but also resulted in the appearance of its metabolic products in the tissue lipids. The initial product of  $oldsymbol{eta} ext{-} ext{oxidation}$  of erucate is gadoleic acid (20:1) and this appeared in many tissues of animals receiving dietary erucate (Tables XV - XVII). Somewhat surprisingly, the 20:1 contents of rat tissues did not parallel the erucic acid concentrations when the corn oil-erucic acid diet was fed. concentration of gadoleic acid in the tissues might reflect the activity of the \$\beta\$-oxidation system in addition to the level of the precursor in each tissue. Care must be taken in interpreting this data since 20:1 may also result by elongation of oleic acid. That this was the case in certain tissues was evident from the relatively high levels of 20:1 in the brain (3.7%) and adrenals (2%) of rats fed olive oil. There was also a tendency for adrenals to elongate 22:1 to 24:1 (Table XV). As might be expected, the diet containing 20:1 (campestris oil) resulted in the greatest accumulation of this acid by most tissues. spleen and adrenals were most active in accumulating gadoleic acid when this oil was fed, while the brain, heart, kidney, plasma and testes exhibit somewhat similar properties in this respect.

The diets which were high in oleic acid (olive oil, Canbra oil) resulted in the highest levels of tissue oleic acid. Similarly, the two diets containing the rapeseed oils usually resulted in higher tissue linoleic acid concentrations than the other two diets. The effects of the four diets on the tissue polyunsaturated acids depended on the tissue under consideration. In the adrenals, the two diets containing erucic acid resulted in lowered levels of arachidonic acid relative to olive oil and the Canbra oil. However, the effect in the case of the campestris oil was not simply due to the replacement of arachidonic by erucic acid, but appeared to involve a complex interaction which included the inhibitory effects of linolenic acid on the elongation-desaturation reactions of linoleic acid and its metabolites. This latter effect was evident when the higher polysunsaturated acids were considered. Although the adrenals from the rats fed olive oil and zero-erucic rapeseed oil contained the same levels of 20:406,

the zero-crucic group contained much lower levels of  $22:\hbar\omega 6$  and  $22:5\omega 6$  than the olive oil group.

In most of the remaining tissues studied, arachidonic acid was in the same range as that observed in animals fed olive oil, and for the most part differences between the two groups were small. Arachidonic acid was slightly lower in the tissues from animals fed the zero-crucic rapesced oil. The difference between the olive oil and Cambra oil groups was most notable in the case of the kidney and heart. A possible explanation for the difference between the campestris and Cambra groups is that both linotenic and oleic acids compete with linoteic acid for the elongation-desaturation enzymes and the Cambra oil contains more oleic than the campestris. The crucic acid in the campestris interferes with the incorporation of higher polyunsaturated acids (cf. Experiment 1).

The two rapeseed oil diets resulted in slightly lower levels of 22:466 and particularly of 22:566 in the tissues. These acids were usually present in low concentrations in most instances and presumably the 63-metabolites (22:503 and 22:603) were able to compete on more equal terms with these 66 acids. In the testes, however, relatively high levels of 22:506 are present in normal animals and the linolenic acid-containing rapeseed oils did not result in any significant depression of this acid (Table XVII).

Preliminary effects on the physiological significance of the changes in tissue fatty acids have been conducted. In spite of the deposition of large quantities of crucic acid in the adrenal lipids, and incorporation which occurred primarily in the cholesterol ester fraction at the expense of polyunsaturated acids (Carroll 1962), no significant differences were noted in the ability of rats to produce corticosterone under stress (immersion in ice-cold water for 30 minutes). Similarly, no differences were noted in the reproductive capacity of the male rats on the four diets. However, the ability to sire young was somewhat lower than expected in all four dietary groups and this aspect is still under investigation. One characteristic of the testes lipids in all four groups was the wide range of values observed for 22:56, an acid previously associated with reproductive capacity in the rat. The ranges are summarized in Table XVIII together with values obtained for animals fed 10% corn oil or 10% hydrogenated coconut oil in other experiments. As may be seen, the possibility of essential fatty acid deficiency in individual animals cannot be ruled out. The arachidonic acid also exhibited this tendency to vary widely in concentration in testes lipids of rats in all four dictary groups, usually paralleling the values noted for 22: 56.

TABLE XVIII

RANGE OF 22:506 CONCENTRATIONS(1) IN RAT TESTES LIPIDS

|   | Range                    | Mean        |
|---|--------------------------|-------------|
| Olive Oil Corn Oil-Ethyl Erucate              | 7.0 - 13.7<br>5.8 - 12.9 | 9.9<br>8.2  |
| Rapeseed Oil Zero-Erucic Rapeseed Oil         | 4.2 - 17.8<br>6.0 - 14.4 | 9.5<br>10.1 |
| Corn Oil (10%) Hydrogenated Coconut Oil (10%) | -                        | 18<br>7     |

<sup>(1)</sup> See footnotes Table XI.

It is apparent that rat tissues exhibit a wide variation in their ability to incorporate erucic acid. The degree of incorporation reflects the dietary level of this acid and is practically insignificant when Canbra oil is fed. Erucic acid appeared to compete with higher polyunsaturated acids for incorporation in tissue lipids. There was also evidence for competition between linolenic and linoleic acid metabolites when the rapeseed oils were administered.

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