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Determination of fatty acid and triacylglycerol composition of human adipose tissue

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ABSTRACT

The fatty acid composition of adipose tissue was studied in a population in western Andalusia with a diet in which the fat contribution is mainly from olive oil. The lipid composition of adipose tissue, including the fatty acid composition of triacylglycerols, was examined by capillary gas chromatography. Thirty-five peaks were resolved, ranging in chain length from 12 to 24 carbon atoms, including geometric and positional isomers. The major triacylglycerol was POO, followed by PLO and OOO.

INTRODUCTION

Mediterranean populations have low incidence rates of cardiovascular disease and hypertension. This may be partly due to dietary factors, particularly a relatively high intake of monounsaturated fat as olive oil. It is known that in these and other populations, the diet [1–4], the presence of free radicals [5,6] and certain surgical operations [7,8] may modify the lipid composition of tissues.

Adipose tissue (AT) consists of adipose cells, fibroblasts, blood vessels and nerve fibres. It is almost totally fatty, basically as triacylglycerols (TG), whose fatty acid (FA) composition varies from individual to individual. Human AT is made up mainly of neutral lipids (<95%), the major ones being the triacylglycerols.

Traditionally, determinations of free and total fatty acids have been performed by gas chromatography (GC) on packed columns [9,10]. The use of capillary GC (cGC) with flame ionization detection has allowed us to study the composition of AT more systematically. Thus we have been able to study the major and minor FA, including their positional and geometric isomers.

Different studies of the FA composition of AT have demonstrated that in most mammals (including man, but excluding the marine mammals), the AT is made up of saturated or monounsaturated medium-chain FA, especially palmitic, stearic and oleic acids [11–13]. These acids are present in considerable amounts, allowing their detection by routine chromatography.

Although there have been numerous studies on FA composition using packed columns, we have been able to find only one study of the FA composition of AT using polar cGC. That study was carried out on the AT composition of a popula-

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tion in Newcastle (UK) [14], whose lipid contribution was based on saturated fats. Our study on the FA composition of AT was carried out on a population in western Andalusia (Spain), with a purely Mediterranean diet in which the fat contribution is mainly from olive oil, whose fundamental fatty acid is oleic acid. This study adds a new dimension to the study of AT composition, *viz.*, the FA composition of its TG.

Of all the studies carried out on human AT, we did not find any in which the integral composition of its TG was considered. Such a study would give an overall view of the constitution of human AT, and would be useful in clinical research on this tissue with regard to alimentation and the detection of cardiovascular diseases [15,16].

This study was carried out on two experimental groups of women and men whose typically Mediterranean diet (2.252 kcal) has olive oil as its fat base.

EXPERIMENTAL

The study was carried out on twenty subjects (ten men and ten women) with body mass index (BMI) 25.40 ± 3.0 . They had no known metabolic disease, and did not exhibit hyperglycaemia or dyslipaemia at the time of taking the sample. Written informed consent for a non-urgent surgical operation at abdominal level was obtained from each subject after fully explaining the procedure. The protocol was that approved by the Institutional Committee on Investigation in Humans.

A sample of AT (*ca.* 0.5 g) was extracted from the anterior abdominal wall of each subject immediately after opening the abdominal region. The tissue fragments were placed in physiological serum and immediately preserved in liquid nitrogen at -70°C until processed.

Standards

Fatty acid methyl ester (FAME) standards were obtained from Larodan Fine Chemicals (Malmo, Sweden). The internal standard solutions were prepared by dissolving 200 mg of

methyl tricosanoate (23:0) in 100 ml of hexane. Calibration solutions were prepared by dissolving known amounts of FAME standards in hexane containing 2,6-*tert.*-butyl-*p*-cresol (butylated hydroxytoluene, BHT), obtained from Sigma (Poole, UK).

Apparatus

For FA analysis, a Hewlett-Packard Series 5890a gas chromatograph with a flame ionization detector and heated injection ports was used. A Supelcowax 10 fused-silica capillary column (60 m \times 0.25 mm I.D., film thickness 0.25 μm) was obtained from Supelco (Bellefonte, PA, USA).

Mass spectral data were obtained with an automated gas chromatographic–mass spectrometric (GC–MS) system, consisting of an HP-5890 gas chromatograph interfaced directly to an AEI MS 30 VG/70 updated mass spectrometer and a VG11/250 data system (VG Analytical, Manchester, UK).

Triglyceride analysis was carried out using a Chrompack CP 9000 gas chromatograph (Chrompack International, Middelburg, Netherlands), fitted with a split injector and a flame ionization detector.

Biochemical measurements

Plasma levels of glucose, uric acid, insulin, total cholesterol, triglycerides, phospholipids, high-density lipoprotein HDL₂, HDL₃ and low-density lipoprotein (LDL)–cholesterol, HDL–cholesterol, A-I and B apolipoproteins, total proteins, sodium, potassium, urea and creatinine were determined in fasting venous blood obtained on the day of the examination with a Hitachi Model 747 automatic analyser.

Lipid extraction

Quantitative extraction of total lipids from tissue was carried out following the method of Folch *et al.* [17] in the presence of BHT as antioxidant. Tissue dissociation was achieved by homogenization in ice-cold chloroform–methanol (2:1, v/v) containing 0.01% BHT using an Ultra-Turax Type TP-18-1.

Separation and determination of lipids

Neutral lipids from the adipose tissue were separated by thin-layer chromatography (TLC) on silica gel 60 plates using hexane–diethyl ether–acetic acid (80:20:1) as solvent system. After development of the plate, the solvent was allowed to evaporate and the bands were detected with iodine vapour. This system separates phospholipids, cholesterol, triglycerides and cholesterol esters in increasing order of R_F values. Individual lipid zones were scraped from the TLC plates and eluted from the silica gel with either diethyl ether or chloroform–methanol according to the individual lipids. The adipose tissue lipid classes were quantified following separation on thin silica-coated quartz rods (Chromarod S) using an Iatroscan (Technical Marketing Assoc., Japan) equipped with a flame ionization detector (hydrogen flow-rate 175 ml/min, air flow-rate 1850 ml/min), a scanner (scanning speed 0.47 cm/s), and an integrator and recorder (sensitivity 10 mV, chart speed 0.42 cm). The Chromarods (Type S9) were successively developed using the following solvents: first chloroform–methanol–water–acetic acid (67:28:3:2) and second hexane–diethyl ether–formic acid (90:10:3).

Preparation of fatty acid methyl esters

Lipids were transmethylated according to the method of Morrison and Smith [18]. The lipid bands on silica gel 60 plates were sprayed lightly with a solution of 0.1% (w/v) BHT in methanol prior to detection. Neutral lipids were eluted with two 15-ml portions of chloroform–methanol (2:1, v/v). The solvent was evaporated in a stream of nitrogen and 10 μg of tricosanoic acid (23:0; internal standard) were added immediately, together with 200 μl of boron trifluoride–methanol complex. The sample was flushed with nitrogen, sealed in a vial fitted with a PTFE-lined cap and heated at 120°C for 1 h. After the sample had cooled, the FAME were extracted with 500 μl of hexane.

Analysis of FAME

The sample (a 1- μl injection of test material was made) was injected into the gas chromatograph.

Following injection, the oven temperature was maintained at 200°C for 10 min, then programmed at 2°C/min to a final temperature of 26°C, which was held for 30 min. The helium flow-rate was 2 ml/min, column head pressure 250 kPa, splitting ratio 1:25, detector and injector temperatures 275°C and detector auxiliary flow-rate 25 ml/min. For MS analysis, a Supelco-wax-10 fused-silica column (60 m \times 0.25 mm I.D., film thickness 0.25 μm) was used with helium as carrier gas. The column temperature was programmed from 130°C at 2°C/min to 200°C (to a total of 100 min) at 4°C/min; the injector temperature was 250°C. The MS conditions were as follows: electron-impact ionization, 70 eV; accelerating voltage, 4 kV; emission current, 100 μA ; and ion source temperature, 200°C. The data were processed with a VG 11/250 data system. Each FAME present in the extract was identified by comparison of its retention time and mass spectrum with those of authentic compounds.

Analysis of triglycerides

Samples (1 μl) of triglycerides in hexane (0.1%) were injected into the gas chromatograph equipped with a 25 mm \times 0.25 mm I.D. 400 65 HT aluminium-clad silica capillary column coated with 65% phenylmethylsilicone (Quadrex, New Haven, CT, USA) and operated under the following conditions: injector and detector temperatures, 360°C; initial column temperature, 350°C maintained for 1 min, then increased at 0.5°C/min to 360°C, which was held for 6 min. Helium was used as the carrier gas at a column head pressure of 130 kPa, the splitting ratio was 60:1 and the detector auxiliary flow-rate was 30 ml/min. The TG were identified by their elution times, because retention is affected not only by the number of carbon atoms but also by the number of double bonds [19].

RESULTS AND DISCUSSION

The study was carried out on normal subjects without known metabolic disorders and who showed no hyperglycaemia at the time of taking the sample. Table I shows the general data and

blood parameters of the subjects. It can be seen that the population had an average age of 38.57 ± 6.7 years and a mean BMI of 25.40 ± 3.00 . The mean systolic blood pressure (SBP), diastolic blood pressure (DBP), glycidic metabolism and lipid profile were in the normal ranges.

Table II compares the AT lipids studied. It can be seen that most of the lipids were TG, followed by phospholipids, free cholesterol and esterified cholesterol. It should be noted that in these samples we found virtually no diglycerides, mono-

glycerides or free fatty acids, and no significant differences between men and women.

The results for healthy subjects showed gender differences in fatty acid composition of adipose tissue (FACAT) (Table III and Fig. 1). The total amount of monounsaturated fat was higher and the amount of polyunsaturated fat was lower in males than in females. The main difference was in the relatively high amounts of oleic acid in the subjects, due mainly to the Mediterranean diet, in which olive oil is the most important contributor

TABLE I

GENERAL DATA, BLOOD PARAMETERS AND PLASMA LIPID LEVELS OF THE POPULATION STUDIED

Values are means \pm S.D. Non statistically significant differences were found.

| | All (n = 20) | Males (M) (n = 10) | Females (F) (n = 10) |
|---|--------------------|-----------------------|-------------------------|
| Age (years) | 40.66 \pm 18.5 | 38.00 \pm 18.47 | 43.33 \pm 18.53 |
| Quetelet index (BMI) (kg/m ²) | 25.40 \pm 3.00 | 24.37 \pm 4.29 | 26.44 \pm 1.72 |
| SBP (mmHg) | 127.08 \pm 17.37 | 130.00 \pm 17.88 | 124.17 \pm 17.86 |
| DBP (mmHg) | 72.50 \pm 9.71 | 75.00 \pm 10.49 | 70.00 \pm 8.94 |
| MBP (mmHg) | 90.69 \pm 11.69 | 93.33 \pm 12.82 | 88.50 \pm 10.56 |
| Fasting blood glucose (mg/dl) | 89.24 \pm 17.62 | 93.83 \pm 21.87 | 84.66 \pm 13.38 |
| Fasting blood insulin (mI.U./l) | 22.54 \pm 15.56 | 29.06 \pm 24.88 | 16.03 \pm 6.24 |
| Total cholesterol (mg/dl) | 190.07 \pm 30.51 | 153.75 \pm 20.69 | 226.40 \pm 40.33 |
| LDL-cholesterol (mg/dl) | 125.37 \pm 18.60 | 95.00 \pm 8.91 | 155.75 \pm 28.30 |
| HDL-cholesterol (mg/dl) | 27.30 \pm 12.95 | 36.00 \pm 5.10 | 38.60 \pm 5.80 |
| HDL ₂ -cholesterol (mg/dl) | 9.00 \pm 3.97 | 8.25 \pm 2.75 | 9.75 \pm 5.19 |
| HDL ₃ -cholesterol (mg/dl) | 28.62 \pm 7.01 | 27.75 \pm 4.27 | 29.5 \pm 9.75 |
| Triglycerides (mg/dl) | 139.87 \pm 23.98 | 124.00 \pm 19.66 | 155.75 \pm 28.30 |
| Phospholipids (mg/dl) | 200.75 \pm 24.96 | 179.75 \pm 8.22 | 221.75 \pm 41.71 |
| Apo A-I (mg/dl) | 110.80 \pm 11.51 | 92.35 \pm 7.35 | 129.25 \pm 15.67 |
| Apo B (mg/dl) | 117.55 \pm 22.97 | 95.85 \pm 14.45 | 139.25 \pm 31.49 |

TABLE II

LIPID CLASS COMPOSITION OF ADIPOSE TISSUE

| Lipid | Lipid composition (Mean \pm S.D., n = 10) (%) | |
|------------------------|---|------------------|
| | Men | Women |
| Neutral lipid (TG) | 97.00 \pm 1.01 | 97.01 \pm 1.56 |
| Phospholipid | 2.63 \pm 0.02 | 2.64 \pm 0.32 |
| Free cholesterol | 0.03 \pm 0.01 | 0.29 \pm 0.02 |
| Esterified cholesterol | 0.05 \pm 0.02 | 0.05 \pm 0.02 |

TABLE III

FATTY ACID COMPOSITION OF TOTAL LIPIDS AND NEUTRAL LIPIDS OF ADIPOSE TISSUE

Results are given as means \pm S.D. for ten separate determinations.

| Fatty acid ^a | Composition (% w/w) | | | |
|-----------------------------|---------------------|------------------|------------------|------------------|
| | Total lipids | | Triacylglycerols | |
| | Males | Females | Males | Females |
| 12:0 | 0.54 \pm 0.23 | 0.47 \pm 0.23 | 0.31 \pm 0.21 | 0.80 \pm 0.35 |
| 12:0(n - 9) | 0.18 \pm 0.08 | 0.11 \pm 0.08 | - | - |
| 14:0 | 2.54 \pm 0.45 | 2.76 \pm 0.29 | 2.65 \pm 0.45 | 2.69 \pm 0.09 |
| 14:1(n - 7) | 0.11 \pm 0.03 | 0.21 \pm 0.01 | - | - |
| 15:0 | 0.21 \pm 0.09 | 0.18 \pm 0.05 | 0.19 \pm 0.06 | 0.19 \pm 0.07 |
| 15:0ai | 0.09 \pm 0.02 | 0.10 \pm 0.02 | 0.06 \pm 0.01 | 0.05 \pm 0.04 |
| 16:0i | 0.23 \pm 0.13 | 0.18 \pm 0.14 | - | - |
| 16:0 | 0.06 \pm 0.02 | 0.12 \pm 0.03 | 0.10 \pm 0.07 | 0.08 \pm 0.03 |
| 16:0 | 20.89 \pm 3.34 | 20.00 \pm 1.94 | 21.01 \pm 1.56 | 20.71 \pm 4.75 |
| 16:1(n - 7) | 0.77 \pm 0.08 | 0.78 \pm 0.21 | 0.71 \pm 0.11 | 0.61 \pm 0.12 |
| 16:1(n - 9) | 2.77 \pm 0.87 | 3.44 \pm 1.51 | 3.50 \pm 1.10 | 4.10 \pm 1.80 |
| 17:0ai | 0.14 \pm 0.03 | 0.17 \pm 0.06 | 0.26 \pm 0.07 | 0.12 \pm 0.06 |
| 17:0 | 0.25 \pm 0.05 | 0.25 \pm 0.04 | 0.28 \pm 0.04 | 0.24 \pm 0.07 |
| 17:1(n - 8) | 0.23 \pm 0.03 | 0.31 \pm 0.07 | 0.27 \pm 0.08 | 0.24 \pm 0.02 |
| 16:4(n - 3) | 0.12 \pm 0.05 | 0.10 \pm 0.03 | 0.20 \pm 0.09 | 0.17 \pm 0.10 |
| 18:0 | 4.43 \pm 1.10 | 4.53 \pm 0.60 | 4.66 \pm 0.57 | 4.83 \pm 1.83 |
| 18:1(n - 9) | 47.39 \pm 5.82 | 46.24 \pm 4.18 | 46.42 \pm 5.72 | 49.31 \pm 6.97 |
| 18:1(n - 7) | 2.00 \pm 0.27 | 1.87 \pm 0.32 | 1.94 \pm 0.23 | 1.93 \pm 0.21 |
| 18:1t | 0.11 \pm 0.03 | 0.17 \pm 0.01 | - | - |
| 18:2(n - 6)t | 0.08 \pm 0.01 | 0.05 \pm 0.03 | - | - |
| 18:2(n - 6)c | 13.89 \pm 1.24 | 14.88 \pm 5.21 | 14.73 \pm 4.51 | 11.12 \pm 2.08 |
| 18:3(n - 6) | 0.12 \pm 0.03 | 0.17 \pm 0.12 | 0.18 \pm 0.15 | 0.09 \pm 0.01 |
| 18:3(n - 3) | 0.42 \pm 0.06 | 0.38 \pm 0.04 | 0.36 \pm 0.02 | 0.33 \pm 0.07 |
| 18:3t | 0.14 \pm 0.04 | 0.13 \pm 0.03 | - | - |
| 20:0 | 0.19 \pm 0.03 | 0.17 \pm 0.05 | 0.11 \pm 0.03 | 0.12 \pm 0.03 |
| 20:1(n - 9) | 0.70 \pm 0.16 | 0.75 \pm 0.05 | 0.11 \pm 0.03 | 0.12 \pm 0.03 |
| 20:1(n - 7) | 0.11 \pm 0.03 | 0.10 \pm 0.03 | 0.15 \pm 0.07 | 0.18 \pm 0.06 |
| 20:3(n - 9) | 0.20 \pm 0.08 | 0.26 \pm 0.10 | 0.28 \pm 0.07 | 0.26 \pm 0.12 |
| 20:3(n - 6) | 0.16 \pm 0.05 | 0.21 \pm 0.10 | 0.28 \pm 0.08 | 0.27 \pm 0.12 |
| 20:4(n - 6) | 0.34 \pm 0.09 | 0.32 \pm 0.05 | 0.26 \pm 0.07 | 0.31 \pm 0.12 |
| 20:3(n - 3) | 0.14 \pm 0.01 | 0.13 \pm 0.01 | - | - |
| 22:0 | 0.04 \pm 0.01 | 0.05 \pm 0.01 | - | - |
| 22:4(n - 6) | 0.17 \pm 0.02 | 0.14 \pm 0.05 | - | - |
| 22:5(n - 3) | 0.15 \pm 0.11 | 0.15 \pm 0.09 | 0.16 \pm 0.04 | 0.20 \pm 0.04 |
| 24:0 | 0.11 \pm 0.01 | 0.11 \pm 0.01 | - | - |
| Σ Saturated | 29.72 | 29.09 | 29.63 | 29.83 |
| Σ Monounsaturated | 54.19 | 53.87 | 53.79 | 57.23 |
| Σ Polyunsaturated | 16.12 | 17.03 | 16.59 | 12.92 |
| $\Sigma n - 6$ | 14.76 | 15.77 | 15.45 | 11.79 |
| $\Sigma n - 3$ | 0.83 | 0.76 | 0.72 | 0.70 |
| $\Sigma n - 9$ | 51.24 | 50.80 | 50.31 | 53.79 |
| $\Sigma n - 7$ | 2.99 | 2.96 | 2.80 | 2.72 |
| $\Sigma n - 6/\Sigma n - 3$ | 17.78 | 20.75 | 21.46 | 16.84 |
| $\Sigma n - 9/\Sigma n - 3$ | 61.73 | 66.84 | 69.87 | 76.84 |

^a ai = Antiiso; i = iso; t = trans; c = cis.

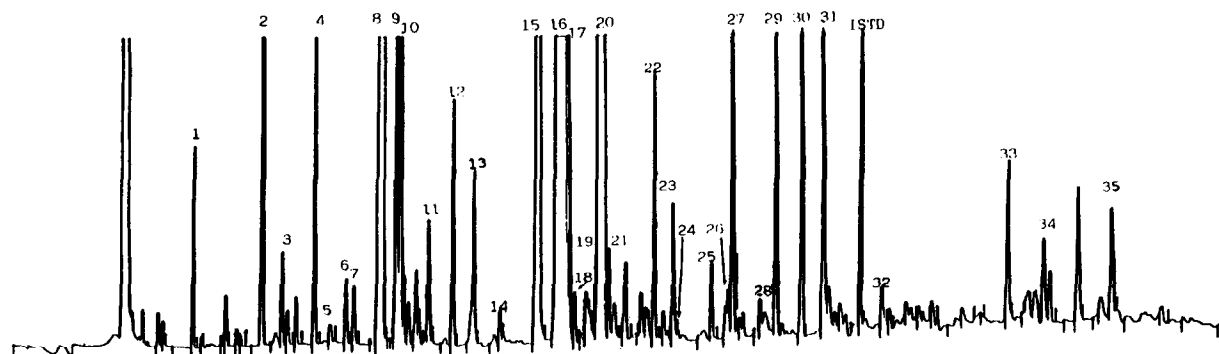


Fig. 1. Gas chromatogram of fatty acid methyl esters of human adipose tissue. Peaks: 1 = 12:0; 2 = 14:0; 3 = 14:1(*n* - 7); 4 = 15:0; 5 = 15:0ai; 6 = 16:0ai; 7 = 16:0i; 8 = 16:0; 9 = 16:1(*n* - 7); 10 = 16:1(*n* - 9); 11 = 17:0ai; 12 = 17:0; 13 = 17:1(*n* - 8); 14 = 16:4(*n* - 3); 15 = 18:0; 16 = 18:1(*n* - 9); 17 = 18:1(*n* - 7); 18 = 18:1*t*; 19 = 18:2(*n* - 9); 20 = 18:2(*n* - 6)*c*; 21 = 18:2(*n* - 6)*t*; 22 = 18:3(*n* - 3); 23 = 18:3(*n* - 6); 24 = 18:3*t*; 25 = 20:0; 26 = 20:1(*n* - 7); 27 = 20:1(*n* - 9); 28 = 22:0; 29 = 20:3(*n* - 9); 30 = 20:3(*n* - 6); 31 = 20:4(*n* - 6); 32 = 20:4(*n* - 3); 33 = 22:4(*n* - 6); 34 = 22:5(*n* - 3); 35 = 24:0. ISTD = internal standard. ai = Antiiso; i = iso; c = *cis*; t = *trans*.

of fatty acids. We expected this concentration of oleic acid to be much higher, as a comparison of our results with those of Cassidy *et al.* [14] and those of Van Staveren *et al.* [20] (from a Dutch population) showed similar concentrations of monounsaturated acids. There is an important difference between our study and previous studies using packed columns [12,13]. In all the latter, only seven fatty acids (lauric, myristic, palmitic, palmitoleic, stearic, oleic and linoleic) were measured, compared with 34 in this work. Positional and geometric isomers, among which the iso, antiiso, *n* - 7 and *trans* isomers stand out, were included. It is remarkable that 18:1(*n* - 7) could be separated from the large peak of 18:1(*n* - 9), as this separation is usually difficult under other conditions. The relatively high polarity of the column used in this study strongly suggests that the three 18:1 isomers observed in our chromatogram are due to 18:1(*n* - 9), 18:1(*n* - 7) and 18:1*t*. This last includes 9*t*, 10*t* and 11*t*. The total *trans* value is lower than reported by Hudgins *et al.* [21] in a study of a population of adult Caucasian males. These differences are probably due to the type of diet of the population in our study, as the population studied by Hudgins *et al.* were consumers of hydrogenated fats from margarines and shortenings.

We found that the amounts of palmitoleic

acid, 16:1(*n* - 9), were lower than those found by other workers. The levels of linoleic acid found in the present and other recent studies [22] were higher than in earlier studies [15]. This possibly reflects recent trends towards a higher dietary intake of linoleate from the increased consumption of sunflower oil and a decrease in that of olive oil (normally for economic reasons).

We did not find differences in composition due solely to sex differences, although Field *et al.* [22] reported that the dietary polyunsaturated/saturated (P/S) ratio was significantly related to the saturated and polyunsaturated content of stored lipids. However, an earlier study [23] indicated that diet did not account for the sex differences in depot fat composition reported.

The study of the fatty acid composition of the TG shows that as with total lipids, oleic acid was the major fatty acid, reaching *ca.* 50%. Palmitic acid followed with 20% and linoleic acid (18:2) with 12.16%. The amounts of arachidonic, dihomono- γ -linoleic, docosopentanoic and docosahexaenoic acids were negligible (<1%).

The most novel results of this work were obtained in the study of the TG as such (Table IV). On the capillary column, TG are separated according to chain length (*CN* separation). Moreover, each *CN* number is split up owing to the polarity differences in the TG. Polarity increases

TABLE IV
AVERAGE TRIACYLGLYCEROL COMPOSITION OF ADIPOSE TISSUE

| Triacylglycerol ^a | Composition (% w/w) | | |
|------------------------------|---------------------|----------------|------------------|
| | Total | Males (n = 10) | Females (n = 10) |
| MMP | 0.14 ± 0.03 | 0.14 ± 0.03 | 0.13 ± 0.02 |
| MMPo | 0.20 ± 0.06 | 0.40 ± 0.01 | 0.17 ± 0.05 |
| MPP | 0.40 ± 0.10 | 0.39 ± 0.16 | 0.49 ± 0.32 |
| MOM | 0.45 ± 0.12 | 0.47 ± 0.01 | 0.44 ± 0.01 |
| MLM | 0.44 ± 0.12 | 0.54 ± 0.01 | 0.41 ± 0.13 |
| PPP | 3.64 ± 2.41 | 3.66 ± 2.29 | 3.75 ± 1.82 |
| MOP | 1.72 ± 0.27 | 1.90 ± 1.06 | 1.63 ± 1.10 |
| MLP | 0.30 ± 0.20 | 0.43 ± 0.23 | 0.11 ± 0.03 |
| MLPo | 0.22 ± 0.02 | 0.21 ± 0.03 | 0.23 ± 0.01 |
| PPS | 0.14 ± 0.02 | 0.14 ± 0.02 | 0.13 ± 0.02 |
| POP | 5.88 ± 1.75 | 6.07 ± 1.63 | 5.93 ± 2.28 |
| PLP + PPoO | 8.86 ± 2.63 | 9.04 ± 2.49 | 8.96 ± 2.74 |
| PLPo + MLO | 2.13 ± 1.24 | 2.19 ± 1.87 | 2.05 ± 0.93 |
| MLL | 0.29 ± 0.19 | 0.25 ± 0.01 | 0.61 ± 0.44 |
| POS | 1.75 ± 0.17 | 2.01 ± 0.17 | 1.48 ± 0.66 |
| POO | 32.17 ± 3.29 | 32.78 ± 6.46 | 32.34 ± 1.92 |
| PLO | 16.28 ± 1.32 | 14.54 ± 1.14 | 16.58 ± 0.97 |
| PLL | 1.50 ± 0.50 | 1.68 ± 0.49 | 1.06 ± 0.51 |
| SOS | 0.25 ± 0.04 | 0.23 ± 0.03 | 0.33 ± 0.04 |
| SOO | 3.20 ± 0.38 | 3.75 ± 0.36 | 2.40 ± 0.31 |
| OOO | 11.80 ± 1.69 | 11.67 ± 1.53 | 12.36 ± 1.32 |
| SOL | 2.34 ± 0.62 | 2.35 ± 0.51 | 2.49 ± 0.53 |
| OLO | 4.68 ± 0.65 | 3.88 ± 0.52 | 4.98 ± 0.73 |
| OLL | 1.05 ± 0.23 | 1.08 ± 0.21 | 1.05 ± 0.22 |
| LLL | 0.20 ± 0.01 | 0.19 ± 0.01 | 0.21 ± 0.01 |

^a Nomenclature. Fatty acids: M = myristic acid, tetradecenoic acid, 14:0; P = palmitic acid, hexadecenoic acid, 16:0; S = stearic acid, octadecenoic acid, 18:0; Po = palmitoleic acid, hexadecenoic acid, 16:1; O = oleic acid, *cis*-9-octadecenoic acid, 18:1; L = linoleic acid, *cis,cis*-9,12 octadecenoic acid, 18:2. Glycerides: PPP = triglyceride *sn*-glycerol-tripalmitate; MLP = triglyceride *sn*-glycerol-myristate-linoleate-palmitate; PLO = triglyceride *sn*-glycerol-palmitate-linoleate-oleate, etc.

with increasing degree of unsaturation in fatty acids (Ln > L > O > S) and with the total number of double bonds in the TG (OOO > SOO > SOS > SSS). Accordingly, retention is highest for the TG containing most of the highest unsaturated fatty acids. Roughly, it can be stated that *CN*-non-unsaturated fatty acid (NUFA) mirror image separation is obtained but with much more refinement. Fig. 2 shows the analysis of TG AT on this capillary column.

Table IV shows the average TG content. The major triacylglycerol is seen to be POO (*sn*-glycerol-palmitate-oleate-oleate) followed by PLO (*sn*-glycerol-palmitate-linoleate-oleate) and

OOO (*sn*-glycerol-trioleate). These three TG together make up 60% of the total. The major TG contain oleic acid in their molecules, and this acid is normally esterified in the *sn*-2-position of the TG. The study of the position of fatty acids in the TG showed that in fact oleic acid was esterified in the *sn*-2-position in about 50% of cases, followed by linoleic, which was found in the *sn*-2-position in about 20% of cases. The *sn*-positions of the TG are normally occupied by saturated acids, in this instance palmitic and stearic acids.

Our results of TG composition cannot be compared with those of any other workers, as we have not found any previous work in which the TG composition of human AT was studied.

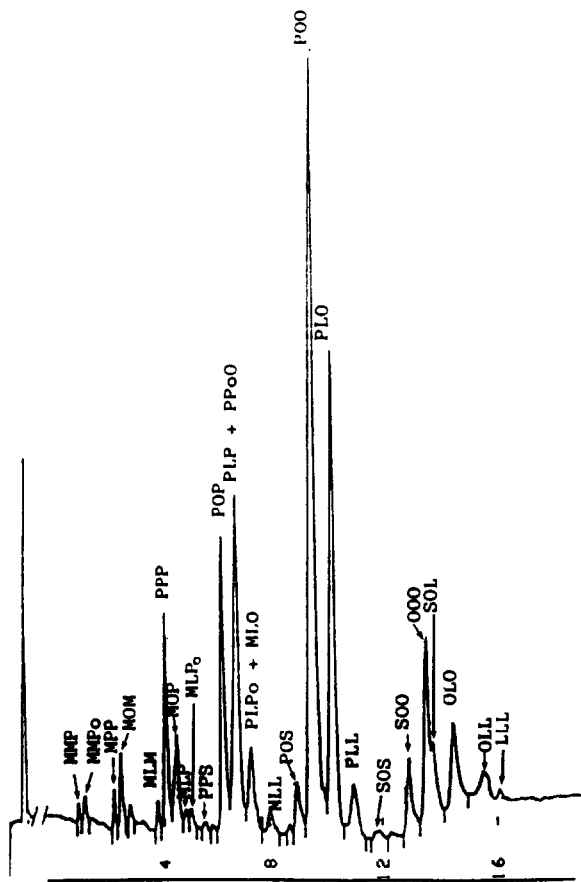


Fig. 2. Polar cGC of human adipose tissue fat triacylglycerols. The triacylglycerols are identified by the combination of the component fatty acids with regard to positional location. GC conditions and instrumentation as in the text. Peak identification as in Table IV.

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