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Proximate, amino acid and mineral composition of Turkey-hen muscle and skin

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ABSTRACT

The muscle and skin were analysed for proximate, amino acid and mineral in matured female turkey (turkey-hen) (*Meleagris gallopavo* L.). The proximate values were high (g/100g dry weight): in organic matter, 99.16-99.22; protein, 53.6-73.5; energy, 1329.8–1374.2kJ/100g. Fatty acid and crude fat were better concentrated in the skin. The range of gross energy contribution by protein was 66.3-94.0% whereas it was 5.9-32.6% by fat and 1.1-1.2% by carbohydrate. Utilizable energy due to protein was 39.8-56.4% showing turkey-hen would prevent protein – energy malnutrition. The samples were good sources of Na, K, Fe and Mg, but all better in the muscle than the skin. The samples were very good sources of essential amino acids having the required levels for all ages from pre-school children (2-5years): in both muscle and skin, the amino acids in that category were: Ile, Met+Cys; in addition the muscle would satisfy such ages in Lys and Thr. Try was not determined. The lowest essential amino acid score was 0.85(Fle) in muscle and 0.60 (Thr) in the skin. The F test results showed that only the total non-essential amino acids/total essential amino acids were significantly different at $p<0.05$ in the muscle as well as in the skin.

Key words: Turkey-hen, proximate, amino acid, mineral, composition.

INTRODUCTION

The domesticated turkey is a large poultry bird raised for food. The modern domesticated turkey descends from the wild turkey (*Meleagris gallopavo*), one of the two species of turkey (genus *Meleagris*); however, in the past the ocellated turkey (*Meleagris ocellata*) was also domesticated. Despite the name, turkeys have no relation to the country of Turkey and are instead native to North America.

The turkey is reared throughout the temperate parts of the World, and is a popular form of poultry, partially because industrialized farming has made it very cheap for the amount of meat it produces. The female domesticated turkey is referred to as a *hen* and the chick as *poult*. In the United State, the male is referred to as a *tom*, whilst in Europe, the male is a *stag*[1]. Turkey are traditionally eaten at Christmas in Britain and Thanksgiving in the United State. In Nigeria, turkey meat is becoming a delicacy particularly at Christmas.

Broad-breasted varieties are prized for their white meat, fast growth and excellent feed-conversion ratios[1]. Raw turkey skin colour is off white to cream-coloured. Under the skin the colour ranges from a pink to a lavender blue depending on the amount of fat just under the skin[2].

There are no reports on the chemical composition of female turkey meat. Due to the emphasis placed on the nutritive value of food by consumers a great need exists for information on nutritional composition of turkey meat. The present study was therefore undertaken in attempt to gain some information on the proximate, amino acid and mineral composition of the muscle and skin which are generally used in the turkey meat industry. The skin was being analysed separately to know its nutritive value because it is often discarded in order to avoid the fat from turkey meat. The turkey sample used was the white plumage turkey-hen bird with a bare, wattled head and neck.

MATERIAL AND METHODS

Collection and treatment of samples

The turkey-hen used was a matured bird. Prior to butchering, food was withheld for a day to help ensure the digestive system was empty. Head was held on the stump and the turkeys head removed with an axe. At the end of bleeding, the turkey was plucked. When all the feathers were removed, the turkey's anus was rinsed to remove any residue, then a sharp knife was inserted just below the hip bone without puncturing any of the internal organs. The turkey was then removed, both skin and muscle sliced, rinsed and dried in the oven. Before drying, samples for mineral determination had been cut apart. Muscle and skin used were those from the breast part. The dried samples were ground, sieved and kept in freezer in McCartney bottles pending analysis.

Proximate analysis

Moisture, ash, crude fat and crude fibre were determined by the AOAC methods[3], while nitrogen was determined by the micro-Kjeldahl method[4] and the percentage of nitrogen was converted to crude protein by multiplying by 6.25. Both carbohydrate and organic matter were determined by difference.

The crude fat values were used to calculate the total fatty acids by multiplying with a conversion factor of 0.945 (for poultry)[5]. The calorific values in kilojoules were calculated by multiplying the crude fat, protein and carbohydrate by Atwater factor of 37, 17 and 17 respectively. Determinations were in duplicate.

Mineral analysis

The tissue samples were prepared for mineral analysis by the method of wet digestion using a combination of nitric acid and perchloric acid (70% solution). 1.0g minced tissue plus 5ml HNO_3 and 2ml $HClO_4$, digested in fume cupboard, heating to a volume of 3-5ml, 10ml distilled water added and filtered into 50ml flask. The filtered solutions were made up to volume with deionized water[6]. Na, K, Cu, Ca, Fe, Mn, Zn, Co and Mg were determined by means of atomic absorption spectrophotometer (Buck Scientific Model-200A/210) with typical variations of ±20%. All chemicals used were of British Drug House (BDH) analytical grade.

Amino acid analysis

The method of amino acid analysis was by ion-exchange chromatograph (IEC)[7] using the Technicon Sequential Multisample Amino Acid Analysis (TSM) (Technicon Instruments Corporation, New York). Details of procedure had been given earlier[8].

Estimation of qualify of dietary protein

The amino acid score was done by the following formula[9]: Amino acid score = Amount of amino acid per test protein [mg/g]/Amount of amino acid per protein in reference pattern [mg/g].

Determination of the total essential amino acid (TEAA) to the total amino acid (TAA), i.e. (TEAA/TAA); total sulphur amino acid (TSAA); percentage cystine in TSAA (%Cys/TSAA); total aromatic amino acid (TArAA) etc, while the predicted protein efficiency ratio was determined using one of the equations developed by Alsmeyer *et al*[10]; i.e.: P-PER = -0.468+0.454 (Leu)-0.105 (Tyr). Theoretical estimation of isoelectric point (pI)

pI determination can be carried out by the equation of the form[11]:

$$IPm = \sum_{i=1}^{n} IpiXi$$

Where IPm is the isoelectric point of the i^{th} amino acid in the mixture and Xi is the mass or mole fraction of the i^{th} amino acid in the mixture[12].

Statistical analysis

Calculations made were the grand mean standard deviation, coefficients of variation in percent and F test setting the confidence level at $p<0.05$[13]. Other calculations were the energy values contributed by protein, fat and carbohydrate as well as utilizable energy due to protein.

RESULTS AND DISCUSSION

Proximate composition

The proximate values of turkey-hen muscle and skin are shown in Table-1. The organic matter

(OM) of 99.16 – 99.27/100g were higher than 98.97g/100g in Ostrich muscles[14], 91.07g/100g in trunk fish, and the values reported for four fresh water fishes of *Mormyrops delicious* (86.4g/100g), *Bagrus bayad* (75.0g/100g), *Synodontis budgetti* (84.0g/100g) and *Hemichronis fasciatus* (76.0g/100g)[16]. The protein content for the muscle was higher than 72.89g/100g reported for trunk fish as well as another report on trunk fish (74.5g/100g)[17]; 76.2g/100g (lean pork) and 72.5g/100g (lean meat of sheep)[18]; 89.0g/100g (ostrich)[14]; 73.7g/100g (beef)[19] and 87.2g/100g (chicken)[20]. The crude fat levels had high disparity between the muscle and the skin with a high level of coefficient of variation percent (CV%) of 70.2. From literature, while the value of fat in the muscle (2.12g/100g) was claose to the value in ostrich (2.74g/100g)[14], the skin value of 12.1g/100g was close to the level of 12.6% in chicken [20], but both were far lower than the level in beef (22.3g/100g)[19]. The ash levels in the turkey-hen samples were lower than the levels in ostrich, beef and chicken with consequent likely lower levels of minerals in the turkey-hen than those three literature samples. The energy levels in our sample (1.33-1.37MJ) was much lower than the levels in sheep lean meat (2.06MJ)[18] and lean pork (2.29MJ)[18]. The energy levels of 1.33-1.37MJ were close to 1.3-1.6MJ/100g from cereals[21] showing turkey-hen meat to be a reasonable source of energy. Although the level of carbohydrate was low, 0.07-092g/100g, it was the most varied with CV% of 85.9. the most concentrated sample was the OM (99.16-99.22g/100g) and it was the least varied (0.06%). Table 7 showed that the F test result was not significant in proximate levels at $p<0.05$.

Table- 2 shows the various energy values as contributed by protein, fat and carbohydrate. The daily energy requirement for an adult is between 2500-3000 Kcal (10455-12548kJ) depending on his physiological state while that of infants is 740 Kcal (3094.68kJ)[22,23]. This implies that while an adult man would require between 786.20-943.59g (muscle) and 760.81-913.11g (skin) to meet his minimum requirement, infants would require 232.72g (muscle) and 225.20g (skin). The utilizable energy due to protein (UEDP%) for turkey-hen (assuming 60% utilization) was 56.41 (muscle) and 39.77 (skin). Both values are far greater than the recommended safe level of 8% for an adult man who requires about 55g protein per day with 60% utilization. This shows that the protein concentration in turkey-hen in terms of energy would be more than enough to prevent energy malnutrition in children and adult fed solely on turkey-hen as a main source of protein. The PEF% (Table 2) was low in muscle (5.90) but within the recommended level of 30%[24] and 35%[25] in skin (32.6%) for total fat energy intake, this is useful for

Table 1: Proximate composition (g/100g edible portion) of turkey-hen muscle and skin (dry weight)

Component[a]	Muscle	Skin	Mean	SD[b]	CV%[c]
Total ash	0.73	0.84	0.79	0.06	7.01
Moisture	23.54	32.56	28.05	4.51	16.08
Protein (Nx6.25)	73.54	53.58	63.56	9.98	15.70
Crude fat	2.12	12.10	7.11	4.99	70.18
Crude fibre	ND[d]	ND	-[e]	-	-
Carbohydrate	0.07	0.92	0.50	0.43	85.86
Organic matter	99.27	99.16	99.22	0.06	0.06
Fatty acid	2.00	11.43	6.72	4.72	70.22
Energy (kJ/100g)	1329.81	1374.2	1352.01	22.20	1.64

[a]Determinations were in duplicate.
[b]SD = standard deviation
[c]CV% = Coefficient of variation percent
[d]ND = not detected
[e]Not applicable

closely followed by aspartic acid (8.79-10.02g/100 cp). This was the exact trend in the AA profile of ostrich, beef and chicken. Lysine and leucine were also highly concentrated as in the case of chicken, beef and ostrich meat. Our histidine level (2.47-2.60g/100g cp) were close to the levels in ostrich (2.03), beef (3.20) and chicken (3.04). also phenylalanine level was 3.80g/100g cp in the skin but the value of 4.79g/100g cp in the muscle was favourably comparable to the values in ostrich (4.84), beef (4.48) and chicken (4.48). Our values in Ile, Val and Met were intermediate with regard to corresponding values in ostrich, beef and chicken. The remaining non-essential AA were very close to the corresponding levels in ostrich, beef and chicken. On comparison with pork[18] and mutton[18], the following AA were better concentrated in pork and mutton than turkey-hen muscle: Leu, Ile, Lys, Cys, Thr, Val, His and total essential amino acids; however the reverse was the case in Met, Tyr and Phe. The FAO standards for pre-school children (2-5years) were (g/100 protein)[28]: Leu (6.6), Ile (2.8), Lys (5.8), Met+Cys (2.5), His (1.9) and total (33.9). based on this information, the muscle would provide enough or even more than enough of Ile, Lys. Thr, Met+Cys, Phe+Tyr, val, His and total 90.91% level of Leu, Try was not determined; the skin would provide enough or more of Ile, Met+Cys, Phe+Try, Val, His, Lys and total requirement but would provide 70.29% level of Thr while Try was not determined. The following values would show the position of the quality of the turkey-hen muscle and skin protein; the essential amino acid (EAA) requirements across board are (values with His) (g/100g protein): infant (46.0), pre-school (2-5years) (33.9), school child (10-12years) 24.1) and adult (12.7) and without His: infant (43.4), pre-school (32.0) school child (22.2) and adult (11.1)[28]; from our own results based on this standards, we have : 35.5g/100g (with His) and 32.9 (no his) in muscle; 35.4g/100g (with His) and 32.9 (no His) in skin. Our results were also close to the levels of the total EAA in egg: 51.2 (with His) and 49.0 (no His); cow's milk: 50.4 (with His) and 47.7 (no His); beef: 47.9 (with His) and 44.5 (no His)[28].

Table 4: The amino acid composition (g/100g crude protein edible portion) of turkey-hen samples (dry weight)

Amino acid	Muscle	Skin	Mean	SD	CV%
Lysine (Lys)*	7.01	5.94	6.48	0.54	8.26
Histidine (His)*	2.60	2.47	2.54	0.07	2.56
Arginine (Arg)*	7.10	5.49	6.30	0.81	12.79
Aspartic acid (Asp)*	10.02	8.79	9.41	0.62	6.54
Threonine (Thr)*	3.44	2.39	2.92	0.53	18.01
Serine (Ser)	4.02	3.07	3.55	0.48	13.40
Glutamic acid (Glu)	13.71	12.22	12.97	0.75	5.75
Proline (Pro)	3.05	2.86	2.96	0.10	3.21
Glycine (Gly)	5.07	5.00	5.04	0.04	0.70
Alanine (Ala)	4.18	3.64	3.91	0.27	6.91
Cystine (Cys)	0.92	0.95	0.94	0.02	1.60
Valine (Val)*	4.35	3.94	4.15	0.21	4.95
Methnione (Met)*	2.55	2.70	2.63	0.08	2.86
Isoleucine (Ile)*	3.41	3.52	3.47	0.6	1.59
Leucine (Leu)*	6.06	6.85	6.46	0.40	6.12
Tyrosine (Try)	3.67	2.79	3.23	0.44	13.62
Phenylalanine (Phe)*	4.79	3.80	4.30	0.50	11.53
Tryptophan (Try)*	-	-	-	-	-
Protein (g/100g)*	84.19	79.25	81.72	2.47	3.02

*Essential amino acid. [a]Dry weight and fat free basis.

The various categories of the AA are shown in Table – 5. The current total EAA (TEAA) is comparable to some literature values (g/100g cp): 35.1 in *Zonocerus variegatus*[29]; 35.0 in *Macrotermes bellicosus*[30]; 42.8 in *Limicolaria* sp., 36.1 in *Archatina archatina and* 45.0 in *Archachatina marginata*[31]. The predicted protein efficiency ration (P-PER) was 1.93 (muscle) and 2.27 (skin). The P-PER values were better than 1.21 (cowpea), 1.82 (pigeon pea)[32]; 1.62 (millet *ogi*) and 0.27 (sorghum *ogi*)[33] but lower than 4.06 (corn *ogi*) and reference casein with PER of 2.50[33]. The Leu/Ile ration was low hence no concentration antagonism might be experienced in the turkey-hen meat. In the results of the isoelectric points, there was a shift from 4.41 in skin to 5.01 in the muscle. (In an unreported work by Adeyeye E.I., 2007, it was found experimentally that the pl of the turkey-hen muscle was 5.0.) The calculation of pl from amino acids would assist in the production of the protein isolate of an organic product.

Most animal proteins are low in Cys, for examples: 36.3% in *M. bellicosus*[30]; 25.6% in *Z. variegatus*[29]; 35.3% in *A. marginata*, 38.8% in *A. archatina* and 21.0% in *Limicolaria* sp. respectively[31]. The present results corroborated these literature observations with values of 26.03-26.51%. In contrast, many vegetable proteins, contain substantially more Cys than Met, example 62.9% in coconut endosperm[8]. Thus for animal protein diets, or mixed diets containing animal protein, Cys is unlikely to contribute up to 50% of the TSAA[7]. The percentage of Cys in TSAA had been set at 50% in rat, chick and Pig diets[7]. Cys has positive

Table 5: Essential, non-essential, acidic, neutral, sulphur, aromatic (g/100g crude protein edible portion) of turkey-hen (dry weight)

Amino acid	Muscle	Skin	Mean	SD	CV%
TAA	81.16	76.42	78.79	2.37	3.01
TNEAA	39.85	39.32	39.59	0.27	0.67
TEAA -with His	41.31	37.10	39.21	2.11	5.37
-no His	38.71	34.63	36.67	2.04	5.56
%TNEAA	49.10	51.45	50.28	1.18	2.34
%TEAA-with His	50.90	48.55	49.73	1.18	3.36
-no His	47.70	45.32	46.51	1.15	2.56
TNAA	40.72	41.51	41.12	0.40	0.96
%TNAA	50.17	54.32	52.25	2.08	3.97
TAAA	23.73	21.01	22.37	1.36	6.08
%TAAA	29.24	27.49	28.37	0.88	3.08
TBAA	16.71	13.90	15.31	1.41	9.18
%TBAA	20.59	18.19	19.39	1.20	6.19
TSAA	3.47	3.65	3.56	0.09	2.53
%TSAA	4.28	4.78	4.53	0.25	5.52
%Cys/TSAA	26.51	26.03	26.27	0.24	0.91
TArAA	8.46	6.59	7.53	0.94	12.43
%TArAA	10.42	8.62	9.52	0.90	9.45
P-PER*	1.93	2.27	2.10	0.17	8.10
Leu/Ile	2.65	3.33	2.99	0.34	11.37
%Leu-Ile	43.73	48.61	46.17	2.44	5.28
pl[a]	5.01	4.41	4.71	0.30	6.37

*P-PER = predicted protein efficiency ratio

[a]pl = isoelectric point.

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effects on mineral absorption, particularly zinc[34, 35]. Table – 6 shows that Ile had the lowest essential amino acid score (EAAS) with a value of 0.85 (85%) in muscle and Thr with 0.60 (60%) in the skin. However, the EAA most often acting in a limiting capacity are Lys, Met+Cys, Thr and Try[36], hence, Thr would act as the limiting amino acid in both samples of the turkey-hen. Therefore, in order to fulfil the day's needs for the EAA in turkey-hen, 100/86 or 1.16 times as much muscle protein would have to be eaten when it is the sole protein in the diet; in skin, it would be 100/60 or 1.67 times protein level.

Table 6: Essential amino acid scores of turkey-hen

Amino acid	Muscle	Skin	Mean	SD	CV%
Ile	0.85	0.88	0.87	0.02	1.73
Leu	0.87	0.98	0.93	0.06	5.95
Lys	1.27	1.08	1.18	0.10	8.09
Met +Cys	0.99	1.04	1.02	0.03	2.46
Phe +Tyr	1.41	1.10	1.26	0.16	12.35
Thr	0.86	0.60	0.73	0.13	17.81
Try	-	-	-	-	-
Val	0.87	0.79	0.83	0.04	4.82
Total	0.94	0.94	0.94	0.0	-

The summary of the various F test calculations are depicted in Table – 7. Only the intra values of essential amino acids and non-essential amino acid were significantly different at $p<0.05$ in both skin and muscle of turkey-hen.

In summary, *Meleagris gallapavo* L. hen (turkey-hen) muscle and skin would serve as good sources of protein, metabolizable energy, Na, K, Mg and Fe as well as high quality essential amino acids when it is consumed as a meat source or as a supplement/fortifier in lower quality protein foods.

Table 7: F test calculation results (in summary) of turkey-hen

Parameter	F-calculated	Degrees of freedom	F-test	Remark
Proximate*	1.07	7/7	3.79	NS[c]
Energy contribution*	1.07	4/4	6.39	NS
Mineral*	4.02	3/4	9.12	NS
Amino acid*	1.25	16/16	2.40	NS
Essential amino acid*	1.22	8/8	3.73	NS
Non-essential amino acid*	1.25	7/7	3.79	NS
TNEAA/TEAA (Muscle)[b]	5.42	7/8	3.73	*
TNEAA/TEAA (skin)[b]	5.30	7/8	3.73	*
Essential amino acid scores[a]	1.72	7/7	3.79	NS

[a]F test in muscle/skin.
[b]TNEAA/TEAA = total non-essential amino acid/total essential amino acid.
[c]NS = not significant at $p< 0.05$.
* = significant at $p<0.05$.

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REFERENCES

1. http: /en. wikipedia. org/wiki/Turkey-(domesticated)
2. http: www. teacher vision. fen. Com/ thanksgiving/birds/2418.html.
3. AOAC, *Official Methods of Analysis*, 15th edn., Association of Offical Analytical Chemist, Washington DC (1990).
4. Pearson D, *Chemical Analysis of Foods*, 7th edn., Churchill, London, 7-11 (1976).
5. Greenfield, H, Southgate, DAT, *Food Composition Data Production, Mnaagement and use*, 2nd edn., FAO, Rome, 223 (2003).
6. Walsh, LM., (Ed.), *Instrumental Methods for Analysis of Soils and Plant Tissue*, Soil Science Society of America, Inc., Mafison, Wisconsin, USA, 27 (1971).
7. FAO/WHO, *Protein Quality Evaluation*, Report of Joint FAO/WHO Expert Consultation. FAO Food and Nutrition Paper 51, FAO/WHO, Rome (1991).
8. Adeyeye, E.I, *Orient. J. Chem.*, **20** (3): 47/ (2004).
9. FAO/WHO, *Energy and Protein Requirements*, Technical Report Series No. 522. WHO, Geneva, Switzerland, 1-118 (1973).
10. Alsmeyer, RH., Cunningham AE, Happich ML, *Food Technology*, **28**: 34 (1974).
11. Olaofe O, Akintayo ET, *The Journal of Technoscience*, **4**: 49 (2000).
12. Finar IL., *Organic chemistry*, 2, 5th edn., ELBS and Longman Group Ltd., London, 651-652 (1975).
13. Christain, GD., *Analytical Chemistry*, 2nd edn., John Wiley and Sons Inc., New York, 69 (1977).
14. Sales, J., Hayes J.P., *Food Chemistry*, **56** (2): 167 (1996).
15. Adeyeye, E.I., Adamu, AS., *Biosci., Biotech. Res. Asia*, 3(2), 265(2005).
16. Abdullahi, SA., Abolude, DS., *Academy Journal of Science and Engineering*, **2**(1): 18 (2002).
17. Adeyeye, EI., Akpambang , VOE., Adebomojo, IA., *Pak J Sci Ind Res*, **46**(6): 270 (2003).
18. Fornias OV, *Edible By-products of Slaugher Animals*, FAO Animal Production and Health Paper 123, FAO, Rome, 9(1996).
19. USDA, *Composition of Foods: Poultry Products*, Agriculture Handbook No. 8-5, United States Department of Agriculture, Washington DC (1986).
20. USDA, *Composition of Foods: Poultry Products*, Agriculture Handbook No. 8-5, United States Department of Agriculture, Washington DC (1979).
21. Paul, AA., Southgate DAT, *McCance and Widdowson's The Composition of Foods*, 4th end., HMSO, London (1978)
22. Bingham, S., *Nutrition: A consumer's guide to good eating*, Transworld Publishers, London, 123-127 (1978)
23. Edem, DO., Amugo, (I, Eka OU, *Tropical Science*, **30**(1): 59 (1990)
24. National Advisory Committee on Nutrition Education (NACNE), *Proposal for nutritional guidelines for healthy education in Britain*, Health Education Council, London (1983)
25. Committee on Medical Aspects (COMA) of Food Policy, *Diet and cardiovascular disease*, HMSO, London (1984)
26. Harris SD, Morris CA, May SG, Jackson TC, Lucia LM, Hale DS, Miler RK, Keeton JT, Savell JW, Acuff GR, *Ostrich Meat Industry Development. A and M Reports to the AOA*, Final Report to American Ostrich Association, Texas Station, TX, USA (1994).
27. Monson ER, Hallberg L, Layrisses M, Hegsted DM, Cook JD, Mertz W, Finch CA, *Am J Clin Nutr*, **31**:134 (1978).
28. FAO/WHO/UNU, *Energy and Protein Requirements*, WHO Tech Report Ser. No. 724, Geneva, 205 (1985).
29. Adeyeye, E I, *Tropical Science*, **45**(3): 124 (2005).
30. Adeyeye, EI., *Journal of Chemical Society of Nigeria*, **30** (2): 145 (2005).
31. Adeyeye, EI., Afolabi EO, *Food chemistry*, **85**: 535 (2004).
32. Salunkhe, Dk., Kadam, SS., *Handbook of world food legumes, nutritional chemistry, processing technology and utilization*, CRC Press, Boca Raton, FL (1989).
33. Oyarekua, MA., Eleyinmi, AF., *Food Agriclture and Environment*, **2**(2): 94 (2004).
34. Mendoza C, *Intern J Food Sci Tech*, **37**: 759 (2002).
35. Sandstrom, B., Almgren A, Kivisto B, Cederblad A, *Jour of Nutr*, **119**: 48 (1989).
36. Bingham, S., *Dictionary of Nutrition*, Barrie and Jenkins Ltd., London, 76-281 (1977).