

## Cholesterol Content in Meat of Some Poultry and Fish Species As Influenced by Live Weight and Total Lipid Content

T. KOMPRDA,\* J. ZELENKA, E. FAJMONOVÁ, P. BAKAJ, AND P. PECHOVÁ

Department of Food Technology, Mendel University Brno, Zemědělská 1,  
CZ-61300 Brno, Czech Republic

Total cholesterol content in 24 fillets (F) of males and females of common carp, 35 samples of male chicken breast meat (BM) and thigh meat (TM), and 48 samples of male turkey BM and TM, respectively, was determined by high-performance liquid chromatography after total lipid (TL) extraction using *n*-hexane/2-propanol mixture. Cholesterol content in male carp fillets (77.6 mg/100 g) was higher ( $P < 0.001$ ) in comparison with females (69.4 mg/100 g). Irrespective of the sex differences, cholesterol content increased ( $P < 0.01$ ) in the sequence chicken BM (53.0 mg/100 g) = turkey BM (53.0) < turkey TM (61.5) < carp F (73.5) < chicken TM (82.9 mg/100 g). Cholesterol content in chicken TM decreased ( $P < 0.05$ ) with increasing live weight reached at the age of 43 days, but did not change ( $P > 0.05$ ) in other tested tissues. Cholesterol concentration in TL of all five tested tissues within three animal species decreased sharply ( $P < 0.001$ ) with increasing TL content reached in a given tissue at the fixed age. It follows from the results of the study that a two hundred gram portion of carp F and chicken TM without skin represents 49 and 55% of the upper limit of daily cholesterol intake, respectively.

**KEYWORDS:** Cholesterol; common carp; chicken; turkey; healthy nutrition

### INTRODUCTION

As an indispensable constituent of the cell membranes and brain tissue, cholesterol is a very important substance in the human organism. However, the opinions regarding relationships of dietary cholesterol intake and the process of atherosclerosis are ambiguous. A correlation between serum cholesterol level and mortality rate on the cardiovascular diseases in man was proved in many studies (1). Lower consumption of foods with high cholesterol content was the consequence of this fact.

On the other hand, the endogenous cholesterol synthesis in liver is three times higher in comparison with usually consumed amounts, which led to weakening of the importance of dietary cholesterol and increasing of an interest in total dietary energy intake, saturated, monounsaturated and polyunsaturated (PUFA) fatty acid intake, and PUFA n-6/PUFA n-3 ratio in foods (2).

However, the changes in plasmatic cholesterol level depends significantly on dietary cholesterol. Hypercholesterolaemia is possible to induce in experiments on primates, only using the diets containing cholesterol (3).

The daily intake of cholesterol is currently recommended not to exceed 300 mg (4). Therefore, the knowledge about cholesterol content in foods is important, especially in poultry and fish meat, because consumption of these foods is currently increasing based on the recommendations of healthy nutrition.

The data regarding cholesterol content in turkey meat are rather scarce. Prusa (5) reported only the relationship of

cholesterol content with other quality characteristics without giving absolute figures. Wong and Sampugna (6) determined cholesterol in unspecified retail turkey meat. Cholesterol content in chicken (7, 8) or carp meat (9–11) was reported more often, but in the case of carp, mostly without specifying method of cholesterol determination.

It is known that cholesterol content in animal tissues can be influenced by dietary treatment (8), despite the regulatory mechanisms on the level of synthesis and absorption, which supposedly maintain cholesterol concentration in these tissues (12).

Cholesterol in meat is possible to determine using colorimetric (13), enzymatic (14), and various chromatographic methods, gas chromatography (15), or high-performance liquid chromatography (HPLC) (16). However, the values measured by different techniques are difficult to compare, which was demonstrated by Arneith and Al-Ahmad (16), who reported the differences in cholesterol content in pork higher than 1 order (30–450 mg/100 g).

The objective of the present study was to compare the cholesterol content in meat of poultry and fish species most commonly consumed in central Europe. In an array of data regarding cholesterol content in foods, the new contribution of the study can be seen in the following aspects:

Cholesterol content of the animal tissues can be influenced by the composition of the feed mixtures, especially by the ratio of polyunsaturated fatty acids (17). Many previous experiments were carried out with the retail food samples (6) and were not

\* Corresponding author. E-mail komprda@mendelu.cz.

**Table 1.** Nutrient and Quantitatively Important Fatty Acid Content in the Finishing Feed for Chickens, Turkeys, and Carps Used in the Experiment

	finishing feed for		
	chickens	turkeys	carps <sup>a</sup>
Nutrient Content (g/100 g)			
crude protein	17.9	20.1	18.4
crude fat	4.9	4.1	5.0
crude fiber	2.1	3.6	2.4
Fatty Acid Content (g/100 g)			
palmitic acid	0.38	0.60	0.76
oleic acid	1.14	1.04	0.67
linoleic acid	1.15	1.27	0.94
$\alpha$ -linolenic acid	0.13	0.06	0.49
EPA + DHA <sup>b</sup>	0.01	0.01	0.13

<sup>a</sup> The figures are based on the assumption that carps consumed feed at the harvest time (autumn) in an approximate ratio 20% plankton + benthos and 80% supplementary wheat (an exact ratio was not possible to measure). <sup>b</sup> Eicosapentaenoic + docosahexaenoic acid.

controlled from this viewpoint. In the present experiment, only defined material from the own feeding trial (with known fatty acid composition) was used.

The data regarding various foods were often obtained using different analytical methods of uneven accuracy (16), sometimes even the method of cholesterol determination is not mentioned (10). The present experiment provides data of cholesterol content in meat of three different species using a single method of determination, which belongs to the most reliable (HPLC).

An assessment of possible effects of live weight and fat content reached at the fixed (that is slaughter) age of the animals on cholesterol content in the tissues was an integral part of the experiment. The above-mentioned relationships regarding the different tissues of the different animal species were compared using the linear regressions equality test, which provides information not found in the available literature.

## MATERIALS AND METHODS

The most commonly consumed fish and bird species in the Czech Republic, common carp, and chickens and turkeys were evaluated in the experiment. Because composition of the diet, especially  $\alpha$ -linolenic acid ratio, can influence cholesterol content in the product (17), the experiment was controlled from this viewpoint. Selected data of the composition of the finishing feed mixtures regarding nutrient and important fatty acid content are presented in **Table 1**. Crude protein, crude fat, and crude fiber were determined according to ref 18. Fatty acids were separated after diethyl ether lipid extraction and combined basic (methanolic solution of CH<sub>3</sub>ONa) and acidic (BF<sub>3</sub> in CH<sub>3</sub>OH) derivatization, using HP 4890 chromatograph and capillary column Omega wax TM250 30 m  $\times$  0.25 mm  $\times$  0.25  $\mu$ m according to ref 17.

**Fish Samples.** Six hundred carps (*Cyprinus carpio*) were stocked in an experimental pond in the spring. Natural feed was supplemented by wheat, according to the usual husbandry practice. After three years, 120 fish of the same age were taken within the autumn harvest, weighed individually, and slaughtered, and 24 carps of each sex were selected within the live weight span as wide as possible. Fish were skinned, filleted, weighed, and stored in the dark glass powder bottles at  $-20$  °C until the successive analyses.

Twelve fillets within each sex were thawed at the room temperature, homogenized in the Moulinex blender (Moulinex, France), and analyzed. Another twelve fillets were put into the preserve jars, a digital thermometer was inserted into the muscle, and jars were placed in the thermostat oven preheated to 200 °C. Fillets were stewed until the internal temperature of 80 °C was reached. Then, the samples were sharply cooled, homogenized as mentioned above, and analyzed.

**Chickens.** Cockerels (hybrid ROSS 208) were fed the commercial feed mixtures pertinent to a given age period. At the age of 43 days, 35 birds, selected based on the live weight within the range as wide as possible, were slaughtered, then the breast meat and thigh meat (without skin and rid of the visible external fat) were separated and homogenized in the Moulinex blender and stored as mentioned above.

**Turkey Samples.** Forty-eight male turkeys (BUT Big 6 hybrid), fed the usual commercial feed mixtures pertinent to the corresponding age categories, were selected from the flock at the age of 140 days, based on the above live weight criterion (live weight span as wide as possible). *Musculus pectoralis profundus* (designated as breast meat) and *m. biceps femoris* + *m. semitendinosus* + *m. semimembranosus* (thigh meat) were separated, rid of skin and visible external fat, and processed as mentioned above.

**Extraction of Total Lipid from the Tissues.** The particular tissue was thawed at room temperature and added to a 500-mL Erlenmeyer flask according to the presumed total lipid content: 70 g of the carp fillet and poultry breast meat and 50 g of chicken and turkey thigh meat. The muscle tissue was spiked with 5 mL of the internal standard solution for cholesterol determination: 1 g of stigmaterol (Merck, Germany) dissolved in 250 mL of *n*-hexane/2-propanol 3:2, v/v mixture (HIP 1).

The sample was then extracted 1 min with 180 mL HIP 1 using the DIAX 900 homogenizator (Heidolph, Germany). The mixture was filtered through the Büchner funnel. Aqueous solution of Na<sub>2</sub>SO<sub>4</sub> (120 mL, 1 g of anhydrous salt per 15 mL of water) was added. After shaking and separation of the layers in the separation funnel, the *n*-hexane layer was filtered through anhydrous Na<sub>2</sub>SO<sub>4</sub> to a 250-mL volumetric flask. Water layer was re-extracted with 50 mL of HIP (7:2, v/v; HIP 2).

The *n*-hexane layer after re-extraction and drying with anhydrous Na<sub>2</sub>SO<sub>4</sub> was added to the 250-mL volumetric flask and filled up to the mark with *n*-hexane. Combined extracts were transferred to the 500 mL round-bottom flask and the content was evaporated on a rotary vacuum evaporator (RV 05-ST IP-B model; IKA Labortechnik, Germany) at 40 °C. Evaporation was finished under nitrogen, total lipids were determined gravimetrically.

**Releasing of Cholesterol from Ester Bonds and Cholesterol Determination.** The solid residue after the HIP extraction was redissolved in 30 mL HIP 1. One mL of the dissolved sample was mixed with 10 mL of sodium methoxide (20 g of NaOH, 300 mL of CH<sub>3</sub>OH, 200 mL of diethyl ether, and 0.075 g of phenolphthalein in 500 mL of solution) in the separation funnel. The mixture was left to stand at room temperature for 30 min and then neutralized with the methanolic HCl (150 mL of concentrated HCl and 675 mL of methanol). The sample was then shaken with 30 mL of *n*-hexane (p.a., Merck, Germany), the separated *n*-hexane layer was passed through anhydrous Na<sub>2</sub>SO<sub>4</sub> and evaporated to dryness.

The solid residue was dissolved in 2 mL of acetonitrile:2-propanol (8:2, v/v) mixture and analyzed by HPLC on Eclipse XDB-C18 column (150  $\times$  2.1 mm, 5  $\mu$ m particle size; Agilent Technologies, USA).

HPLC set (Ecom, Czech Republic) consisted of LCP 4100 gradient pump, LCD 2083 UV detector, LCI injector and LCO 101 column oven. A mobile phase was acetonitrile/2-propanol (95:5, v/v; Merck, Germany), flow rate, 0.5 mL/min; injection volume, 20  $\mu$ L. Cholesterol was detected at 210 nm.

Apart from cholesterol determination, dry matter content was determined according to ref 19.

**Statistical Evaluation.** The differences between tissues (carp fillets, F, chicken or turkey breast meat, BM and thigh meat, TM) were assessed using one-way classification of the variance-ratio test, including the multiple-range Duncan test. Significance of the linear and quadratic term was tested by the polynomial regression analysis when the dependence on live weight at the fixed age was evaluated. The Unistat package, version 4.53 (Unistat Ltd., London, England) was used.

The differences between the courses of corresponding regressions (which were statistically significant) were tested by means of *F*-criterion using linear regressions equality test according to (20):  $F = (n_1y_1^2 + n_2y_2^2 - ny^2 + b_1^2s_{1x} + b_2^2s_{2x} - b^2(s_{1x} + s_{2x} + s_{3x}))/2s^2$ , where  $n = n_1 + n_2$ ,  $x = (n_1x_1 + n_2x_2)/n$ ,  $y = (n_1y_1 + n_2y_2)/n$ ,  $b = (s_{1x}b_1 + s_{2x}b_2 + s_{3x}b_3)/(s_{1x} + s_{2x} + s_{3x})$ ,  $s_{3x} = n_1x_1^2 + n_2x_2^2 - nx^2$ ,  $b_3 = (n_1x_1y_1 + n_2x_2y_2 - nxy)/s_{3x}$ ,  $s^2 = ((n_1 - 2)s_{1y}^2 + (n_2 - 2)s_{2y}^2)/(n_1 + n_2 - 4)$ ,

$s_{1x} = s_{1x}^2(n_1 - 1)$ ,  $s_{2x} = s_{2x}^2(n_2 - 1)$ ,  $y_1$  and  $y_2$  are the means of the content of a given dependent variable, and  $s_{1y}^2$  and  $s_{2y}^2$  are their variances,  $x_1$  and  $x_2$  are the means of particular independent variables (live weight and total lipid content, respectively),  $s_{1x}$  and  $s_{2x}$  are their standard deviations, and  $b_1$  and  $b_2$  are corresponding coefficients of the linear term of the regressions, which were compared.

## RESULTS AND DISCUSSION

### Total Lipid Extraction and Cholesterol Determination.

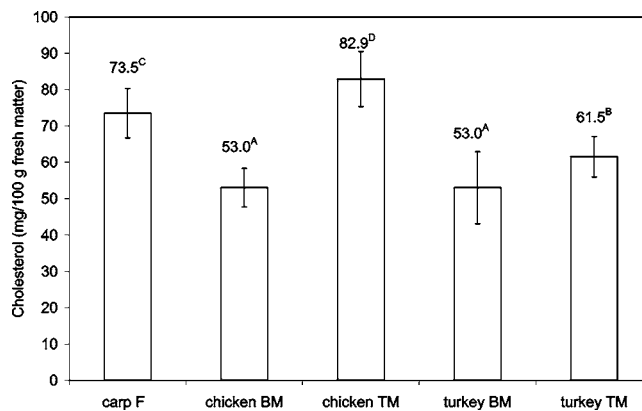
Total lipid is usually extracted from animal tissues by chloroform/methanol mixture in similar experiments (21). However, Hara and Radin (22) reported the following advantages of hexane/2-propanol mixture used in the present study: less of nonlipid coextractives and less toxic solvents. Moreover, according to (16), the procedure with HIP has higher reproducibility of the results.

Cholesterol content determined in various animal tissues depends strongly on the analytical method. Blanch et al. (23) reported an overestimation of the cholesterol content determined by the colorimetric method in comparison with the gas chromatography. Also, according to ref 16, chromatographic determinations are superior to colorimetric or enzymatic ones. However, cholesterol content found in chicken BM in the present experiment (53 mg/100 g) is similar to the results of ref 8, which are based on the enzymatic determination (55 mg/100 g), but lower in comparison with the gas chromatographic data in ref 7: 63–67 mg/100 g.

**Carp Fillets – Effect of Sex and Heat Treatment.** The live weight of fish was in the range of 1172–3196 g in the present experiment, the mean  $\pm$  standard deviation being  $2044 \pm 66.9$  g. Females tended ( $P > 0.05$ ) to be slightly heavier than males,  $2164 \pm 96.9$  and  $1923 \pm 87.3$ , respectively.

Total lipid content (HIP extract) in raw meat was not different ( $P > 0.05$ ) between females ( $8.1 \pm 2.0$  g/100 g of fresh tissue) and males ( $8.0 \pm 1.6$  g/100 g). However, cholesterol content in the raw fillets prepared from males was substantially higher ( $P < 0.001$ ) in comparison with females:  $77.6 \pm 7.0$  and  $69.4 \pm 7.8$  mg/100 g of the fresh tissue, respectively. Higher cholesterol content in males in comparison with females was also found in pork cuts (81 vs 74 mg/100 g) (24), lamb carcasses (3.26 vs 1.74 mg/g of the carcass fat) (25), and turkey TM (78 vs 71 mg/100 g of the fresh tissue at the 90th day of age; birds fed the commercial feed mixture) (26). However, these data cannot be generalized: in comparison with the male counterparts, reported (27) higher cholesterol content in female nutria (36 vs 29 mg/100 g), and (17) in female turkey BM (52 vs 41 mg/100 g; birds fed the diet supplemented with linseed oil).

An explanation of the sex differences is not given in any of the quoted papers. One possible factor could be the level of the sex hormones in serum (28). Sex hormones were not measured in the present experiment, but we do not suppose their higher levels, because carps were not slaughtered in a reproductive age. The above differences are discussed in the literature mostly from the viewpoint of serum cholesterol in man, and the results are often contradictory: Weggemans et al. (28) reported higher total serum cholesterol in women (5.12 vs 4.88 mmol/L), Hergenc and coworkers (29) in men (189–214 vs 186–203 mg/dL). It is also interesting that according to Hergenc and coworkers (29), correlation between cholesterol and testosterone was insignificant in man, but significant in women ( $P < 0.01$ ). Moreover, it is known from animal experiments (30), that the correlation between serum and tissue (meat) cholesterol is poor. Therefore further experiments are needed to elucidate the sex



**Figure 1.** Comparison of cholesterol content in carp fillets ( $F$ ,  $n = 24$ ), chicken ( $n = 35$ ) and turkey ( $n = 48$ ) breast (BM), and thigh meat (TM).

differences in cholesterol content in the muscle tissue of fish (and other animals).

On the basis that carp (fish meat in general) is consumed without considering the sex, an effect of heat treatment was evaluated, and the comparison of carp meat and poultry meat was performed using the overall set of fish (both males and females;  $n = 24$ ). Cholesterol content within this set was  $73.5 \pm 6.8$  mg/100 g of the fresh tissue.

As regards the heat treatment, cholesterol content in raw meat ( $73.5 \pm 6.8$  mg/100 g of fresh matter) did not differ ( $P > 0.05$ ) from stewed fillets ( $76.6 \pm 10.2$  mg/100 g). Due to the different dry matter content in raw and stewed meat (in stewed meat 2% higher,  $P < 0.001$ ), the above comparison was also expressed on dry matter basis. The values were also not different ( $P > 0.05$ ):  $281.5 \pm 33.1$  and  $274.1 \pm 40.8$  mg/100 g of dry matter for raw and stewed fillets, respectively.

**Comparison of Total Lipid and Cholesterol Content in Carp, Chicken, and Turkey Meat.** Total lipid (TL) content was the lowest in turkey breast meat ( $1.5 \pm 0.3$  g/100 g of fresh tissue), but this value was not different ( $P > 0.05$ ) from chicken BM ( $1.9 \pm 0.7$  g/100 g). TL in turkey thigh meat was higher than both above values ( $2.4 \pm 0.5$  g/100 g), but lower ( $P < 0.01$ ) in comparison with chicken TM ( $7.2 \pm 1.9$  g/100 g). The highest ( $P < 0.01$ ) TL content was found in carp:  $8.1$  g/100 g of the fresh tissue (the mean of males and females).

Comparison of cholesterol content of the five tissues within the three species is presented in **Figure 1**. Cholesterol content increased ( $P < 0.01$ ) in the sequence chicken BM ( $53.0 \pm 5.3$ ) = turkey BM ( $53.0 \pm 9.9$ ) < turkey TM ( $61.5 \pm 5.6$ ) < carp fillets ( $73.5 \pm 6.8$ ) < chicken TM ( $82.9 \pm 7.6$  mg/100 g fresh matter). The sequence is likely determined (among other things) by the interspecies differences, differences in cholesterol content between various tissues within a given species, total lipid content in a given tissue, cholesterol concentration in TL of a given tissue, and fatty acid composition of the feed mixtures. Higher cholesterol content in TM of both poultry species in comparison with BM is likely a consequence of substantially higher total lipid content in thigh meat. Despite the insignificant relationship between cholesterol content and total lipid content found in most of the analyzed tissues (discussed in a more detail hereinafter), cholesterol content (mg/100 g of the fresh tissue,  $Y$ ) increased with increasing TL content in the tissue (% ,  $X$ ), when the regression was calculated over the range of all three species and all three tissues:  $Y = 59.5 + 2.26X$  ( $P < 0.001$ ,  $R^2 = 0.34$ ).

Second highest cholesterol content in carp fillets (**Figure 1**) is in agreement with the data of (31), who reported slightly higher cholesterol content in most analyzed fish species (49–92 mg/100 g) in comparison with meats (45–84 mg/100 g).

The fact that cholesterol content in carp fillets, despite higher total lipid content in this tissue, was lower in comparison with chicken TM in the present experiment can be a consequence of a significant decline of cholesterol concentration (% of TL) with increasing total lipid content in the tissue (Figure 4). Moreover, due to a much higher content of  $\alpha$ -linolenic acid in the carp diet in comparison with both poultry species (Table 1), the ability of this dietary fatty acid to decrease cholesterol content in meat, proved in our previous experiment in turkeys (17), could manifest itself.

According to Geri et al. (10), cholesterol content in skinned right fillet of the common carp did not differ statistically either due to the age (12–18 months, 90–87 mg/100 g) or due to the different environmental rearing conditions (warm and natural, 91 and 88 mg/100 g, respectively). The direct comparison with our data is not possible, because the quoted authors did not mention method of cholesterol determination.

Cholesterol content in the sample from different edible parts of the common carp was 55 mg/100 g in the experiment of (11) based on gas chromatographic determination. Bieniarz et al. (32) found (using colorimetric determination) cholesterol content in muscle (including skin) of related species crucian carp (*Carassius carassius*) and grass carp (*Ctenopharyngodon idella*) reared in storage pond 68.4–68.9 mg/100 g of muscle + skin, and in wintering pond 63.4–80.7 mg/100 g of muscle + skin. The differences between above quoted experiments and the present study may be among other things due to the different method of cholesterol determination, which would be a confirmation of the data of ref 16.

As regards turkey, Wong and Sampugna (6) found cholesterol content in various raw ground retail samples between 73 and 91 mg/100 g. These are higher values in comparison with our results, and the reason can again be the different method of extraction and determination (gas chromatography without prior lipid extraction in the quoted paper) and/or analyzing of different tissues. These tissues were not further specified in the study of ref 6, but total lipid content was 5.9–10.8 g/100 g, which is several times higher in comparison with our results.

Cholesterol content in chicken BM is comparable with the data of ref 8, but is lower in comparison with the results of ref 7 (see above). As regards chicken thigh meat, our results (82.9 mg/100 g) agree in this case with the data of ref 7 (79–86 mg/100 g), but are lower in comparison with enzymatically determined cholesterol in the study of (8). On the other hand, cholesterol content in both chicken tissues in the present study was higher in comparison with the results of our previous experiment (33), likely due to the modifications in the method of cholesterol determination resulting in higher recoveries within the present experiment.

Comparison of the results of the present study and of related experiments indicates great differences in cholesterol content in similar materials due to the analytical method used. Therefore, cholesterol content in a given food should be referred to together with the method of determination.

Moreover, it follows from the results of the present experiment and of the similar experiments that cholesterol content in such a recommendable food as fish or lean poultry meat can be as high as or higher than cholesterol content in fatty pork. A two hundred gram portion of carp fillet and chicken thigh meat without skin represents 49 and 55% of the upper limit of daily cholesterol intake, respectively. On the other hand, it is only 35% in the case of chicken or turkey breast meat (without skin).

**Effect of Live Weight on Total Lipid and Cholesterol Content in Meat.** Total lipid content increased linearly and

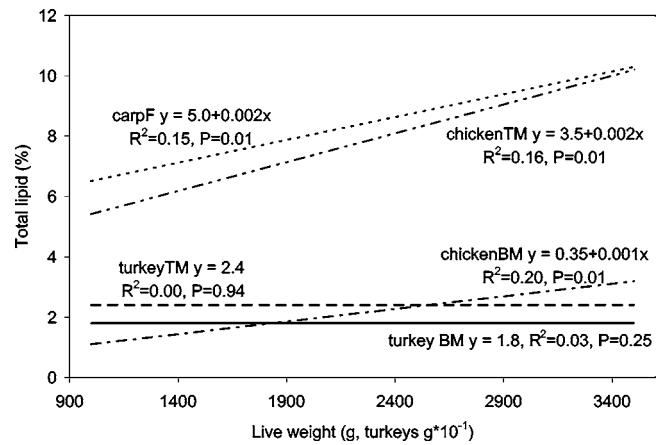


Figure 2. Dependence of total lipid content in carp fillets ( $F$ ,  $n = 24$ ), chicken ( $n = 35$ ) and turkey ( $n = 48$ ) breast (BM), and thigh meat (TM) on live weight reached at a slaughter age.

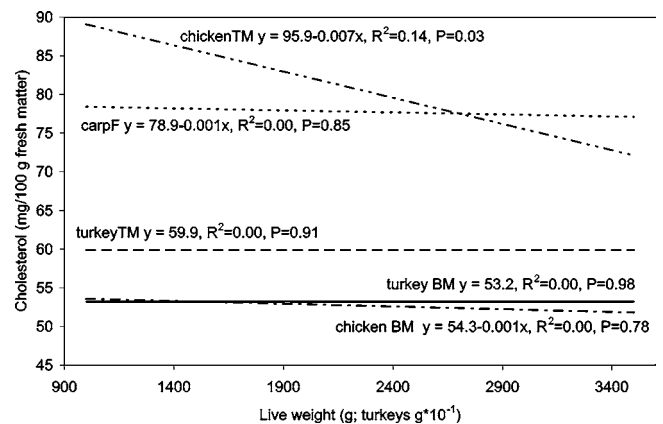
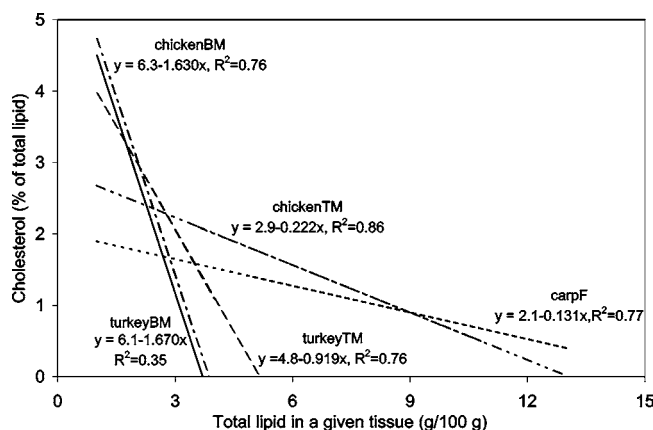


Figure 3. Dependence of cholesterol content in carp fillets ( $F$ ,  $n = 24$ ), chicken ( $n = 35$ ) and turkey ( $n = 48$ ) breast (BM), and thigh meat (TM) on live weight reached at a slaughter age.

significantly ( $P = 0.01$ ) with the increasing live weight at the given age in carp fillets and in both chicken tissues in the present experiment (Figure 2). Inclusion of the quadratic term was not significant in any case. The course of the regressions was different between chicken BM and TM ( $F$ -value, 154.2;  $P > 0.05$ ), but did not differ between chicken thigh meat and carp fillets ( $F$ -value, 2.67;  $P < 0.01$ ), based on the linear regressions equality test. On the other hand, TL content in both turkey breast meat and turkey thigh meat was independent of live weight ( $P > 0.05$ ).

The reason of the latter fact is difficult to explain. Turkeys were taken (similarly to carps and chickens) within the range of live weight as wide as possible (12–24 kg) to obtain sufficient variability of total lipid and consequently of cholesterol content due to the live weight differences. However, the variability of TL values (expressed as the standard deviation) was even lower in turkey tissues (0.3 and 0.5 in BM and TM, respectively) in comparison with both chickens (0.7 and 1.9) and carps (2.0 and 1.6, respectively).

Cholesterol content significantly decreased in chicken TM ( $P < 0.05$ ), but did not change ( $P > 0.05$ ) in four other tested tissues with the increasing live weight (Figure 3). In our above-mentioned previous experiment (33), only a tendency ( $P > 0.05$ ) of cholesterol content to decrease in both BM and TM of chickens was found. Geri et al. (9) reported a decreasing trend of cholesterol content in meat of mirror carp as body weight increased in the range of 40–2800 g. Similarly, Mathew et al. (34) found slightly higher cholesterol levels in smaller specimens



**Figure 4.** Dependence of cholesterol concentration in total lipid in carp fillets (*F*,  $n = 24$ ), chicken ( $n = 35$ ) and turkey ( $n = 48$ ) breast (BM), and thigh meat (TM) on total lipid content in these tissues.

of the same species than in the bigger ones, when comparing 97 fish species.

As regards turkey tissues in the present experiment, the relationship between cholesterol content and live weight was similar to that between TL and live weight (that is insignificant,  $P > 0.05$ ).

An organism maintains cholesterol homeostasis because this sterol is an integral and indispensable constituent of the cell membranes. The relatively constant cholesterol content in muscle tissue of other animal species (cattle and pigs) was reported previously (12, 30), which was confirmed in the present experiment in turkey or fish meat. The decrease of cholesterol content in chicken TM in the present experiment is rather atypical from this viewpoint. However, the latter dependence can be explained by the sharp (and highly significant,  $P < 0.001$ ) decrease of cholesterol concentration in total lipid of the muscle tissue (in % of TL) with increasing TL content in the tissue (this relationship is discussed in more detail in the following text, see also **Figure 4**). Because the intramuscular fat content was very high in chicken TM (7.2%), the above trend of cholesterol "dilution" likely outweighed the more general trend of cholesterol homeostasis maintenance. This conclusion is corroborated by the fact that significant ( $P < 0.05$ ) negative correlation between cholesterol content (mg/100 g of the tissue) and TL content (% of the tissue) was found in chicken TM.

The situation was probably different in carp fillets, where the total lipid content was even higher than in chicken TM (8.1%), but the coefficient of the linear term of the regression of cholesterol concentration on total lipid content (**Figure 4**) was twice as low in comparison with the chicken TM (it was the lowest among all measured tissues).

It follows from the above-mentioned facts that the generally accepted notion of constant cholesterol level in animal muscle tissues requires further investigations, based on the results of the present experiment and similar studies, which suggest decreasing trend in cholesterol content with increasing live weight in some species. This trend is presumably in relationship with highly significant decrease of cholesterol concentration in total muscle lipids as total lipid content in the muscle tissue increases.

**Effect of Total Lipid on Cholesterol Content.** Cholesterol content tended to decrease with increasing TL content ( $P > 0.05$ ) in carp fillets, but tended to increase in both turkey tissues and in chicken BM ( $P > 0.05$ ). However, cholesterol content in chicken thigh meat (mg/100 g of the fresh matter,  $Y$ ) decreased significantly with increasing total lipid content

(g/100 g,  $X$ ) according to the equation  $Y = 93.5 - 1.49X$  ( $R^2 = 0.15$ ,  $P = 0.02$ ). Inclusion of the quadratic term was not significant. Contrary to our results regarding carp fillets, Osman et al. (35) reported that cholesterol levels in selected marine fish species increased with an increase in fat content, although the relationship was not significant.

Interesting results were found regarding dependence of cholesterol concentration in total lipid (percent of TL) on total lipid content (g/100 g of the fresh tissue) in the present experiment. Highly significant decrease ( $P < 0.001$ ) of cholesterol concentration in total lipid on TL content in each of five tested tissues within the three species is apparent from **Figure 4**. On the basis of the linear regressions equality test, the decrease of cholesterol concentration did not differ between carp fillets and chicken TM ( $F$ -value 4.86,  $P < 0.05$ ; similar result as in the case of total lipid content). The course of the above dependences was also the same in chicken BM and turkey BM ( $F$ -value 1.35,  $P < 0.01$ ).

The above cholesterol concentration decrease is possible to explain by the subcellular distribution of cholesterol in muscle tissue reported by ref 36. As the amount (per g) of storage material in intramuscular fat increased, membrane material (per g) decreased. This caused a change in cholesterol distribution in a sense of a dilution of the membranous component of the muscle fibers. Because the storage lipid component of muscle fibers contains only smaller part of total cholesterol in comparison with the membrane fraction, concentration of total cholesterol decreased.

We did not extract total lipid separately from the muscle fiber storage fraction and membrane fraction (it can be done using repeated centrifugation prior to the lipid extraction). Therefore, we were not able to evaluate the relationship between cholesterol and lipid when the above dilution of the membranous component by the increase of storage lipid is eliminated. It is not possible to disregard cholesterol in storage lipid, because this fraction still contains about 30% of total cholesterol (membranous fraction 60–80%). When the influence of the total lipid fraction was eliminated, that is when the dependence of cholesterol on the fat free matter of the given tissue (chicken and turkey BM and TM and carp fillet, respectively) was calculated, neither relationship was significant ( $P$  in the range 0.2 – 0.7,  $R^2$  in all cases lower than 0.03). In other words, cholesterol content was constant with increasing weight of the fat free matter of the tissue.

As far as the relationship between cholesterol content ( $Y$ , mg/100 g of the given tissue) and total lipid content ( $X$ , %) is concerned, the only significant regression (negative) was in the case of chicken TM:  $Y = 93.2 - 1.43X$  ( $P = 0.026$ ,  $R^2 = 0.14$ ). In carp fillets, cholesterol content only tended to decrease, whereas in all other cases (chicken BM, turkey TM, and BM, respectively) it tended to increase with increasing TL content in the tissue ( $P$  in the range 0.2–0.5,  $R^2$  nearing to zero). The latter findings are in agreement with the data of ref 31 or 37 who reported poor relationship between cholesterol content and fat content in various animal and fish species and different types of meat.

#### LITERATURE CITED

- (1) Griffin, B. A. Lipoprotein atherogenicity: an overview of current mechanisms. *Proc. Nutr. Soc.* **1999**, 58, 163–169.
- (2) Okuyama, H.; Kobayashi, T.; Watanabe, S. Dietary fatty acids – the n-6/n-3 balance and chronic elderly diseases. Excess linoleic acid and relative n-3 deficiency syndrome seen in Japan. *Prog. Lipid Res.* **1997**, 35, 409–457.

- (3) Ruddel, L. L.; Parks, L. S.; Hedrick, C. C.; Thomas, M.; Williford, K. Lipoprotein and cholesterol metabolism in diet-induced coronary artery atherosclerosis in primates. Role of cholesterol and fatty acids. *Prog. Lipid Res.* **1998**, *37*, 353–370.
- (4) James, W. P. T.; Ralph, A. Policy and a prudent diet. In *Human Nutrition and Dietetics*, 10th edition; Garrow, J. S.; James, W. T. P.; Ralph, A., Eds.; Churchill Livingstone: Edinburgh, Great Britain, 2000; p 837–845.
- (5) Prusa, K. J. Relationship of selected quality characteristics and the cholesterol and sodium content of turkey. *Poultry Sci.* **1986**, *65*, 1208–1210.
- (6) Wong, M. K.; Sampugna, J. Moisture, total lipid, fatty acids, and cholesterol in raw ground turkey. *J. Agric. Food Chem.* **1993**, *41*, 1229–1231.
- (7) Ajuyah, A. O.; Lee, K. H.; Hardin, R. T.; Sim, J. S. Influence of dietary full-fat seeds and oils on total lipid, cholesterol and fatty acid composition of broiler meats. *Can. J. Anim. Sci.* **1991**, *71*, 1011–1019.
- (8) Konjufca, V. H.; Pesti, G. M.; Bakalli, R. L. Modulation of cholesterol levels in broiler meat by dietary garlic and copper. *Poultry Sci.* **1997**, *76*, 1264–1271.
- (9) Geri, G.; Lupi, P.; Parisi, G.; Dell'Agnello, M.; Martini, A.; Ponzetta, M. P. Morphological characteristics and chemical composition of muscle in the mirror carp (*Cyprinus carpio* var. *specularis*) as influenced by body weight. *Aquaculture* **1995**, *129*, 323–327.
- (10) Geri, P.; Poli, B. M.; Gualtieri, M.; Lupi, P.; Parisi, G. Body traits and chemical composition of muscle in the common carp (*Cyprinus carpio* L.) as influenced by age and rearing environment. *Aquaculture* **1995**, *129*, 329–333.
- (11) Vácha, F.; Tvrzická, E. Content of polyunsaturated fatty acids and cholesterol in muscle tissue of tench (*Tinca tinca*), common carp (*Cyprinus carpio*) and hybrid of bighead carp (*Aristichthys nobilis*) with silver carp (*Hypophthalmichthys molitrix*). *Polish Arch. Hydrobiol.* **1995**, *42*, 151–157.
- (12) Harris, K. B.; Cross, H. R.; Pond, W. G.; Mersmann H. J. Effect of dietary fat and cholesterol level on tissue cholesterol concentrations of growing pigs selected for high or low serum cholesterol. *J. Anim. Sci.* **1993**, *71*, 807–810.
- (13) Bohac, C. E.; Rhee, H. R.; Cross, H. R.; Ono, K. Assessment of methodologies for colorimetric cholesterol assay of meats. *J. Food Sci.* **1988**, *53*, 1642–1644.
- (14) Krug, A.; Suleiman, A. A.; Guilbault, G. G.; Kellner, R. Colorimetric determination of free and total cholesterol by flow injection analysis with a fiber optic detector. *Enzyme Microbiol. Technol.* **1992**, *14*, 313–316.
- (15) Klatt, L. V.; Mitchell, B.; Smith R. L. Cholesterol analysis in foods by direct saponification – gas chromatographic method: Collaborative study. *J. AOAC Int.* **1995**, *78*, 75–79.
- (16) Arneth, W.; Al-Ahmad, H. Cholesterol. Bestimmung in Muskel- und Fettgewebe sowie in Innereien mittels HPLC. *Fleischwirtschaft* **1995**, *75*, 185–187.
- (17) Komprda, T.; Zelenka, J.; Bakaj, P.; Kladroba, D.; Blažková, E.; Fajmonová, E. Cholesterol and fatty acid content in meat of turkeys fed diets with sunflower, linseed or fish oil. *Arch. Geflügelk.* **2003**, *67*, 65–75.
- (18) AOAC. *The Official Methods of Analysis of AOAC International*. 17th edition; Horwitz, W., Ed.; AOAC Inc.: Arlington, Va, 2001; Vol. 2, pp 2200+.
- (19) CSN 570185. Testing methods for meat and meat products; Prague, 1985; 48 pp.
- (20) Rod, J.; Vondráček, J. *Polní pokusnictví*, 1st edition; State technical Publishing House: Prague, Czech Republic, 1973, 230 pp.
- (21) Phillips, K. M.; Tarragó-Trani, M. T.; Grove, T. M.; Grün, I.; Lugogo, R.; Harris, R. F.; Steward, K. K. Simplified gravimetric determination of total fat in food composites after chloroform-methanol extraction. *JAOCs* **1997**, *74*, 137–142.
- (22) Hara, A.; Radin, N. S. Lipid extraction of tissues with a low-toxicity solvent. *Ann. Biochem.* **1978**, *90*, 420–426.
- (23) Blanch, A.; López-Ferrer, E.; Barroeta, A. C.; Grashorn, M. A. Effect of different dietary fat sources on cholesterol content in tissues of broiler chickens. Comparison of two methodologies. In *Proceedings of the 12th European Symposium on the Quality of Poultry Meat*, Zaragoza, Spain, 1995; pp 453–459.
- (24) Dorado, M.; Martín-Gómez, E. M. M.; Jiménez-Colmenero, F.; Masoud, T. A. Cholesterol and fat contents of Spanish commercial pork cuts. *Meat Sci.* **1999**, *51*, 321–323.
- (25) Arsenos, G.; Zygojannis, D.; Kufidis, D.; Katsaounis, N.; Stamataris, C. The effect of breed slaughter weight and nutritional management on cholesterol content of lamb carcasses. *Small Ruminant Res.* **2000**, *36*, 275–283.
- (26) Komprda, T.; Šarmanová, I.; Zelenka, J.; Bakaj, P.; Fialová, M.; Fajmonová, E. Effect of sex and age on cholesterol and fatty acid content in turkey meat. *Arch. Geflügelk.* **2002**, *66*, 263–237.
- (27) Tulley, R. T.; Malekian, F. M.; Rood, J. C.; Lamb, M. B.; Champagne, C. M.; Redmann, S. M.; Patrick, R.; Kinler, N.; Raby, C. T. Analysis of the nutritional content of *Myocastor coypus*. *J. Food Comp. Anal.* **2000**, *13*, 117–125.
- (28) Weggemans, R. M.; Zock, P. L.; Urgert, R.; Katan, M. B. Differences between men and women in the response of serum cholesterol to dietary changes. *Eur. J. Clin. Invest.* **1999**, *29*, 827–834.
- (29) Hergenc, G.; Schulte, H.; Assmann, G.; vonEckardstein, A. Associations of obesity markers, insulin, and sex hormones with HDL-cholesterol levels in Turkish and German individuals. *Atherosclerosis* **1999**, *145*, 147–156.
- (30) Wheeler, T.L.; Davis, G. W.; Stoecker, B. J.; Harmon, C. J. Cholesterol concentration of *longissimus* muscle, subcutaneous fat and serum of two beef cattle breed types. *J. Anim. Sci.* **1987**, *65*, 1531–1537.
- (31) Piironen, V.; Toivo, J.; Lampi, A.-M. New data for cholesterol contents in meat, fish, milk, eggs, and their products consumed in Finland. *J. Food Comp. Anal.* **2002**, *15*, 705–713.
- (32) Bieniarz, K.; Koldras, M.; Kaminski, J.; Mejza, T. Fatty acids and cholesterol in some freshwater fish species in Poland. *Folia Univ. Agric. Stetin.* **2000**, *27*, 21–44.
- (33) Komprda, T.; Zelenka, J.; Tieffová, P.; Štohandlová, M.; Foltýn, J. Effect of the growth intensity on cholesterol and fatty acids content in broiler chicken tissues. *Arch. Geflügelk.* **1999**, *63*, 36–43.
- (34) Mathew, S.; Ammu, K.; Viswanathan Nair, P. G.; Devadasan, K. Cholesterol content of Indian fish and shellfish. *Food Chem.* **1999**, *66*, 455–461.
- (35) Osman, H.; Suriah, A. R.; Law, E. C. Fatty acid composition and cholesterol content of selected marine fish in Malaysian waters. *Food Chem.* **2001**, *73*, 55–60.
- (36) Hoelscher, L. M.; Savell, J. W.; Smith, S. B.; Cross, H. R. Subcellular distribution of cholesterol within muscle and adipose tissues of beef loin steaks. *J. Food Sci.* **1988**, *53*, 718–722.
- (37) Chizzolini, R.; Zanardi, E.; Dorigoni, V.; Ghidini, S. Calorific value and cholesterol content of normal and low-fat meat and meat products. *Trends Food Sci. Technol.* **1999**, *10*, 119–128.

Received for review May 20, 2003. Revised manuscript received September 25, 2003. Accepted September 29, 2003. The experiments were carried out with the financial support of the Mendel University Brno (project No. MSM 432100001) and the Grant Agency of the Czech Republic (project No. 525/01/P078).