Investigation into the phospholipids of porcine adipose tissue

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Through triglycerides are quantitatively the main components of the adipose tissue, other tissue components play - nevertheless an important role in the changes occuring during storage and processing. The study of these components may contribute to the better understanding and forecast of the biochemical changes in the post mortem state and of the chemical changes during heat treatment.

In this research work adipose tissues of different sorts of pigs of different age and feeding were studied for some years. The phospholipid content varied between 7 - 3o p.p.m. Phosphatidylcholine, phosphatidylethanolamine, lysophosphatidylethanolamine and phosphatidylserine were the main phospholipid components. During storage the decomposition of the phospholipids, principally that of the phosphatidylethanolamines was observed.

On the basis of these investigations no direct significant correspondence could be found between the composition of the forage and the phospholipid content. In the discolouration by the heat treatment presumably not the quantity of the phospholipids or their decomposition products but other components of the complex colour forming reaction are determinant.

Untersuchung der Phospholipide der Fettgewebe von Schweinen

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Obwohl Triglyceride mengenmässig die wichtigsten Komponenten der Fettgewebe sind, spielen immerhin die anderen Fettgewebekomponenten auch eine bedeutende Rolle in den Veränderungen, die während der Lagerung und der Verarbeitung eintreten. Deshalb kann ihr gründliches Studium zur besseren Kenntnis bzw. Vorverkündigung der im post mortem Stadium sich abspielenden biochemischen oder der während der Wärmebehandlung stattfinden den chemischen Veränderungen beitragen.

Im Rahmen dieser Forschungsarbeit wurden durch mehrere Jahre die Fettgewebe von zahlreichen Schweinen von unterschiedlicher Art, Lebensalter und Fütterung untersucht. Der durch Mikromethode bestimmte Gesamtphospholipidgehalt schwankte zwischen 7 - 3o p.p.m. Cholinphosphatide, Äthanolaminphosphatide, Lysoäthanolaminphosphatide und Serinphosphatide waren die wichtigsten komponenten. Die Zersetzung der Phospholipide - in erster Linie die der Äthanolaminphosphatide - war während der Lagerung wahrnehmbar.

Aufgrund der bisherigen Untersuchungen war es nicht möglich einen direkten signifikanten Zusammenhang zwischen Futterzusammensetzung und Phospholipidgehalt zu finden. In der durch Wärmebehandlung hervorgerufenen Verfärbung ist vermutlich nicht die Menge der Phospholipide bzw. ihrer Spaltprodukte der bestimmende Faktor, sondern andere Komponenten der komplexen Farbbildungsreaktion.

9.2

Examen des phospholipides des tissus adipeux du porc

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Quoique les triglycerides sont quantitativement les composant les plus importants des tissus adipeux, pourtant les autres composants jouent un rôle appréciable dans les changements pendant le stockage et les traitements technologiques. E'est pourquoi leur examen approfondi peut contribuer à une connaissance et une prévision mieux changement biochimiques pendant l'état post mortem et des changement chimiques pendant le traitement thermique.

Dans le cadre de ce travail de recherche tissus adipeux des porcs de différentes spèces, âge et fourragement étaient examinés durant des années. La teneur en phospholipides totales variait entre 7 - 3o p.p.m. Les phosphatidyl-cholines, les phosphatidyl-éthanolamines, les phosphatidyl-sérines étaient des composants les plus importants des phospholipides. Pendant le stickage la décomposition des phospholipides - en premiére ligne celle des phosphatidyl-éthanolamines était observée.

Sur la base des examèn jusqu' à présent il n'a pas réussi à trouver une corrélation directe de signification entre la composition du fourrage et la teneur en phospholipides. Dans le changement de la couleur pendant le traitement thermique présomptivement ce n'est pas la quantité des phospholipides ou de leur produits de décomposition qui est le facteur définiant mais celle des autres composants de la réaction complexe de la formation de la couleur.

Исследование фосфолипидов жировой ткани свиней.

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Триглийериды являются главными компонентами жирных тканей всеже и другие компоненты играют значительную роль в изменениях происходящих при хранении и переработке. Исследование этих компонентов может помагать в достижении более глубакого понятия процессов происходящих после убоя и при тепловой обработке.

В рамках этой научной работы были исследовани жировые ткани свиней различного происхождения, века, веса и откорма в течение выше лет. Содержание фосфолипидов изменялось между 7 - 3o ppm. Лецитины, этаноламинфосфатиды, лизоэтанольаминфосфатиды и серинфосфатиды являлись главными фосфолипидными компонентами. Во время хранения начинается расщепление фосфатидов в первом ряде этанольаминфосфатидов. На основе исследований не обнаружена прямая коррелация между составом корма и фосфолипидного состава жировых тканей. В процессов цветообразования при тепловой обработке продукты распада фосфолипидов не являются лимитирующим фактором не вероятно другие компоненты комплексной реакции цветообразования.

9.2

Investigation into the phospholipids of porcine adipose tissues

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Introduction

Through triglycerides are quantitatively the main components of the adipose tissues, other tissue components play - nevertheless, an important role in the changes during occuring storage and processing. The study of these components may contribute to the better understanding and forecast of the biochemical and chemical changes in the post mortem state and during heat treatment. The different phospholipids are standing components of the adipose tissue. During the storage a hydrolitic cleavage of the phospholipids may occur. There are also indications that some products of the enzymatic cleavage of the formation of lipofuscin like pigments (1,2,3,4). The basic components of phospholipids such as par example ethanolamine play also an important role by forming colouring matters during heat treatment of fats. The possible reactions are well summarized by Pokorny et al. (5,6,7).

The main purposes of the investigations published in this paper are the following ones: determination of the quantity of phospholipids in different porcine adipose tissues, characterization of the main phospholipid components, changes of the phospholipids during storage, investigation of the possible role of the phospholipids in some colour forming processes.

Materials and Methods

The adipose tissues of pigs were investigated for some years. The effect of feeding, of location and storage was investigated. The samples of porcine adipose tissues were selected after slaughtering and stored by a temperature of 8-10°C. The feeding experiments were organized by Hungarian Research Institute of Animal Production.

Extraction of phospholipids

After homogenizing the adipose tissues were extracted with a threefold quantity of petroleumether at room temperature during 4 hours. The extraction was twice repeated and the extracts collected. After evaporation about 90% of solvent the concentrate was extracted by equal quantity of methanol and the extract concentrated by evaporation (Solution I.). The remaining material after methanol extraction was extracted by a chloroform-methanol (2:1) mixture. The extract was also concentrated by evaporation (Solution II.).

Determination of total phosphorus content (8)

An aliquot part of the solutions I. and II. were evaporated to dry. The solvent free remaining material was treated by 2 ml of 70%-ic prchloric acid. The dark solution was heated in a sandbath until the total discoloration. 1 ml of this solution was mixed with 1 ml 2 n sulfuric acid and 3 ml of ammoniummolibdenate solution. After 20 minutes the absorbance was measured at 658 nm by a Spektromom 240 spectrophotometer. The phosphorus content was calculated on the basis of calibration curve prepared by investigation of KH_2PO_4 standard solutions. For the calculation of phospholipid content a factor of 22,6 was used according the literature (9).

TLC of phospholipids (10)

Kieselgel-G layer was used after 1 hour activation at 105°C and defatting by an acetone-petroleumether (1:3) mixture. Development was made by a chloroform-methanol-28%-ic ammoniumhydroxide (65:25:5) mixture (11). For detection a perchloric-acid-ammoniummolibdenate solution resp. 45%-ic sulfuric acid was used (12). For qualitative evaluation (identification) phospholipid standards of Serva were used. The quantitative determination was made by a Videodensitometer (Chinoin, Hungary). The following phospholipids were determined quantitatively: phosphatidyl-ethanolamine (PE), phosphatidylcholine (PC), lysophosphatidyl-ethanolamine (LPE), lysophosphatidyl-choline (LPC), phosphatidyl-inosite (PI), and phosphatidyl-serine (PS).

Other data

The phosphatidyl-choline : phosphatidyl-ethanolamine ratio (E/C), and the phosphatide : lysophosphatide ratio (P/LP) was calculated on the basis of data of quantitative TLC determinations.

Results and discussion

Some data representing the total phospholipid content of adipose tissue from pigs of different age and weight (1oo-11o kg, and 16o-18o kg) are summarized in Table 1. As it is seen from the table the characteristic phospholipid content lies between 15-25 ppm. The main phospholipid components are phosphatidyl ethanolamin, phosphatidyl-choline and lysophosphatidyl-ethanolamine (See Table 2.).

The distribution of the phospholipids depending on the location of adipose tissues was also investigated. It was stated that, the longitudinal distribution is quite homogeneous. In cross section the adipose tissue layers located nearer to the body surface of animal had in most cases a significantly higher phospholipid content. The ratio of different phospholipid components is practically independent from the location.

During feeding experiments the fat and protein content of feed was changed by addition of different quantities of fish meal. On the basis of this investigations no direct significant correlation could be found between the composition of forage and the phospholipid content.

The characteristic changes of phospholipids during the storage at 8-1o^{o}C are represented by the data collected in Tables 3. and 4.

On the basis of results it can be stated that the relative quantity of phosphatidyl-ethanolamine and phosphatidyl-choline decreased during the storage. Parallely and increase in relative lysophosphatidyl-ethanolamine and lysophosphatidyl-choline content is observed. These changes may be well expressed by using P/LP ratio which is decreased during storage (See Table 5).

Total phospholipid content of porcine adipose tissues from animals of different weight (age)

Table 1

No of sample	Weight of the animals	Phospholipid content (ppm)
1		14,8
2		2o,5
3	1oo-11o kg	14,9
4		14,4
5		18,2
6		17,1
7		13,8
8	16o-18o kg	13,5
9		22,4
lo		21,9
11		23,5
12		19,2
13		11,7
14		21,9
15		34,4

The data summarized above show that the decomposition of phospholipids in the adipose porcine tissue is relatively fast.

This fact support the views that the products of phospholipid cleavage may occur in the tissues and may take part in different interactions with other components of adipose tissue. On the basis of our experiments it may be stated that in the discolouration by the heat treatment presumably not the quantity of the phospholipids or their decomposition products but other components of the complex colour forming reaction are determinant.

The ratio of different phopholipid components in the adipose porcine tissues (% of the total phospholipid content)

Table 2

Number of sample	1	2	3	4	5	6	7	8
PE	24,4	21,3	15,6	25,3	22,1	21,3	21,6	26,4
PC	41,2	44,o	48,5	43,4	35,6	39,4	43,5	36,2
LPE	21,1	24,9	25,4	18,3	29,3	23,7	22,5	26,4
LPC	8,1	8,o	6,4	9,4	9,3	1o,3	8,1	7,4
PI + PS	5,2	1,8	4,1	3,6	3,7	5,3	4.2	3.6
P/LP	2,24	1,99	2,o1	2,48	1,49	1,78	2,13	1,7o
E/C	o,92	o,89	o,75	o,83	1,14	o,91	o,85	1,21

The changes of phospholipids during storage at 8.1o°C (Ratio of components in %)

Table 3

Number of sample	1			2			3			4		
Storage time (days)	O	6o	9o	O	6o	9o	O	6o	9o	O	6o	9o
PE	26,2	14,5	1o,1	29,3	15,o	13,7	38,7	11,9	8,7	21,6	6.3	5.4
PC	42,2	38,4	35,3	43,3	27,5	2o,1	22,5	12,7	8,9	48,7	45,o	43,7
LPE	26,6	36,1	4o,7	15,5	2o,o	23,4	19,5	38,1	4o,2	1o,8	18,9	2o,9
LPC	3,1	9,5	12,4	6,9	35,3	4o,9	11,3	35,7	39,4	8,1	23,3	27,9
PI + PS	1,9	1,6	1,5	5,o	2,2	1,9	8,o	1,6	1,9	1o,8	6,3	2,1
P/LP	2,3	1,2	o,9	3,2	o,8	o,6	2,o	o,3	o,2	3,5	1,2	1,o
E/C	1,2	1,1	1,1	o,9	o,6	o,6	1,7	1,o	1,o	o,6	o,4	o,4

The changes of phospholipids during storage at 8-1o°C (Ratio of components in %)

Table 4

Number of sample	5		6		7		8	
Storage time (days)	O	6o	O	6o	O	6o	O	6o
PE	2o,7	9,2	25,3	11,1	18,4	8,9	27,2	13,4
PC	47,1	29,4	41,3	34,6	43,4	21,5	38,9	2o,9
LPE	16,9	27,3	14,1	24,2	27,9	4o,2	18,3	29,1
LPC	11,7	32,o	9,3	26,4	6,4	26,9	9,4	34,3
PI + PS	3,6	2,1	1o,o	3,7	3,9	2,o	6,2	2,3
P/LP	2,4	o,8	2,8	o,9	1,8	o,5	2,3	o,6
E/C	o,8	o,6	o,8	o,6	o,9	o,9	o,9	o,8

The changes of total phospholipid content in adipose porcini tissues during storage at 8-1o°C

Table 5

N°of sample	Storage time (days)			
	O	3o	6o	9o
	Phospholipid content ppm			
1	24,5	1o,4	4,8	3,2
2	2o,o	16,3	13,8	13,o
3	35,8	12,9	8,9	6,8
4	27,9	7,6	5,6	3,8
5	29,4	15,4	5,8	-
6	2o,1	12,2	3,5	-
7	24,2	13,8	4,o	-
8	24,3	14,5	4,6	-

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