FRONT FACE FLUORESCENCE SPECTROSCOPY AS A TOOL FOR COMPARISON OF AROMATIC AMINO ACIDS, NUCLEIC ACIDS, VITAMIN A AND FMRP CONTAINING IN DIFFERENT EGG TYPES

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Abstract
The objective of this study is to compare amino acids and nucleic acids, food Maillard reaction products and vitamin A containing in different egg types. Chicken eggs are the most commonly eaten eggs. They supply all essential amino acids for humans. Five fresh brown and white eggs come directly from the producer and available in the laboratory after about one week from lay. The egg whites and yolks from four different avian species (hen, goose, duck and quail) were studied. Normalized Fluorescence spectrum of amino acids and nucleic acids differs from each other by A1 and A2 components according to wavelengths of 320 nm, 340-360 nm and 400 nm. The method of investigation is based on front face fluorescence spectroscopy. For egg white samples the fluorescence emission spectra of amino acids and nucleic acids (280-450 nm) and food Maillard reaction products (380-580 nm) were recorded with the excitation wavelength set at 250 and 360 nm respectively. For egg yolk samples excitation spectra of vitamin A (270-350 nm) were recorded with the emission wavelength set at 410 nm.
In Kyrgyzstan there is no established standard on eggs and egg control isn't made (The Kyrgyz news agency, 2010). Therefore, this study is aimed to carry through.

Keywords: fluorescence, amino acids, nucleic acids, vitamin A, FMRP, eggs
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1. INTRODUCTION

Fluorescence spectroscopy is a rapid, sensitive, and non-destructive analytical technique providing in a few seconds spectral signatures that can be used as fingerprints of the food products (dairy products, fishes, edible oils, eggs, etc.). (Sбdeckб, Tythovб, 2007)
Eggs play an important role in our everyday food. In 2007 total egg production in the world was ca. 62.6 million tons; the European Union produced 9.9 million tons, representing 170 billion eggs (FAO, 2009). In one year in Kyrgyzstan are produced more than 294.4 million eggs. (NISM KR, 2008).

2. REVİEW OF THE LİTERATURE

Chicken eggs are the most commonly eaten eggs. They supply all essential amino acids for humans, (Montagne, 2001).) and provide several vitamins and minerals, including vitamin A, riboflavin (vitamin B2), folic acid (vitamin B9), vitamin B6, vitamin B12, choline, iron, calcium, phosphorus and potassium. They are also a single-food source of protein.
All of the egg's vitamin A, D, and E are in the egg yolk. The egg is one of the few foods that naturally contain vitamin D.
Due to their multifunctional properties (e.g., foaming, gelling and emulsifying), eggs are also widely used in the food industry and they are very good potential sources of raw materials for the pharmaceutical and cosmetic industries. (Huopalahti *et al*., 2007)
Eggs are highly versatile food containing many essential nutrients. Eggs of most species of birds may have similarities in nutritional composition and potential food usage, however, information on egg quality characteristics and utilization of egg for food and other purposes have been limited mostly to chicken eggs. Egg quality is composed of

those characteristics of an egg that affects its acceptability to consumers such as cleanliness, freshness, egg weight, shell quality; yolk index, albumen index, Haugh unit and chemical composition. (Song, Choi, Oh, 2000; Stadelman, 1977).

Chicken egg has been very well studied for its external and internal qualities as well as for its composition, however such information are not so abundantly documented in other poultry species.

Knowledge of the chemical composition and physicochemical properties of native egg white and yolk is necessary to interpret the functional and biological properties attributed to specific egg components.

However, it is also known that the cosmetic effect of quail egg is superior to that of hen egg since the former has higher egg yolk, dry matter and fat contents.

Especially the calcium, phosphorus, molybdenum, vitamin A and phospholipid /chiefly lecithin/ contents are high. (Closa *et al.* 1999) In recent years, it has been observed in the poultry breeding that quails were benefited as much as hens, both for their meat and eggs, therefore, commercial quail breeding have become widespread. (Altinel *et al.*, 1996)

At present, the consumption of quail eggs is limited in Poland mainly because of the lack of proper advertisements. Moreover, quail eggs are less appreciated due to lower level of consumer's awareness. In future, quail products could become more popular as a conventional food – source of vitamins, minerals, proteins and fatty acids, in the human diet, due to wider availability on the market and consumer needs for changing eating habits.

Hen eggs, most frequently used for human consumption, have already been thoroughly studied but there is still not enough information on the profile of fatty acids in eggs, especially from quail eggs. (Małgorzata Kaźmierska)

3. MATERIALS AND METHODS

3.1 Egg samples

Five fresh brown and white eggs come directly from the producer and available in the laboratory after about one week from lay. Each egg was weighed (±0.01 g) and then, the white was separated from the yolk.

3.2 Front face fluorescence measurements

Fluorescence spectra egg white and egg yolk was placed in 3 ml quartz cuvette and spectra was recorded at a room temperature (22-25ºC). Considering egg white samples, the emission spectra of amino acids and nucleic acids (280-450 nm) and food Maillard reaction products (380-580 nm) were recorded with the excitation wavelength set at 250 and 360 nm respectively. For egg yolk samples excitation spectra of vitamin A (270-350 nm) were recorded with the emission wavelength set at 410 nm. For each egg white and yolk, six spectra were recorded.

3.3 Mathematical treatment of data

The information obtained from spectrum analysis was treated by the method of SAISIR (Statistique Apliquee al'Explotation der Spectres Infrarouge) in MATLAB program. To see the difference of spectra all the spectra were normalized, to minimalize variations PCA (Principal Components Analysis) was used.

The PCA transforms the original variables into new axes, or principal components (PCs), which are orthogonal, so that the data set presented on these axes are uncorrelated with each others. Therefore, PCA expresses as much as possible the total variation in the data set in only a few PCs (Karoui *et al*, 2007).

To bear some relation to principal components regression PLS-DA was applied.

PLS is a regression method that finds the relationship between predictor variables (X) and dependent variable Y. PLS-DA looks for directions that are able to discriminate the classes (Davide Ballabio *et al*). In PLS-DA variables were treated according to 5th component.

Table 1: Comparison between chemical compositions of Sudanese white cheese produced in New Halfa

Egg type	Egg taken date	Mass of egg, g	egg taken day temperature,ºC	storage days	Storage temperature,ºC
Chicken white egg	09.03.11	42.8	+19	7	+5
Chicken brown egg	09.03.11	43.4	+19	7	+5
Duck egg	11.03.11	81.6	+6	5	+5
Goose egg	11.03.11	124.2	+6	5	+5
Qiail egg	12.03.11	13.2	+4	4	+22

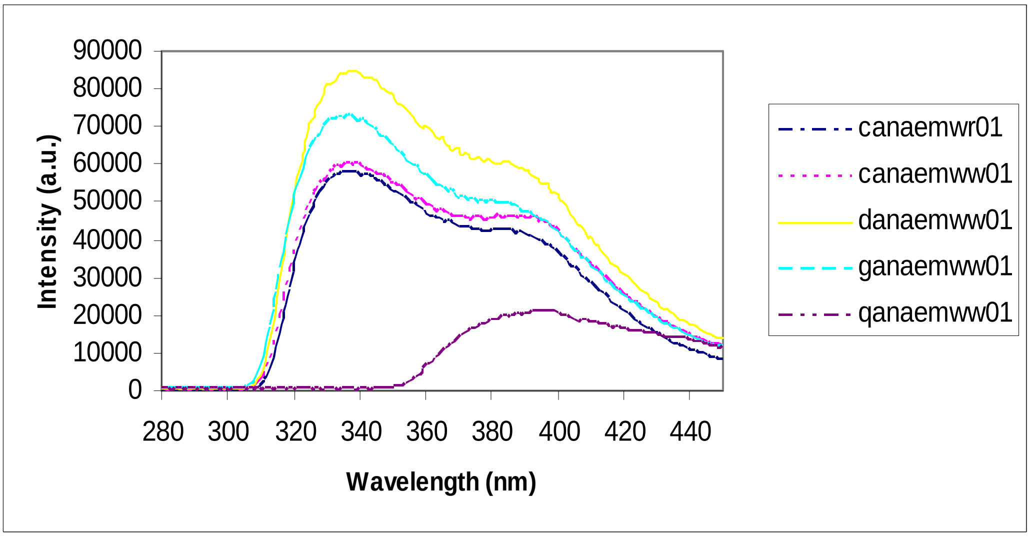

Fig.1. Normalized fluorescence spectrum of aromatic amino acids and nucleic acids of egg whites

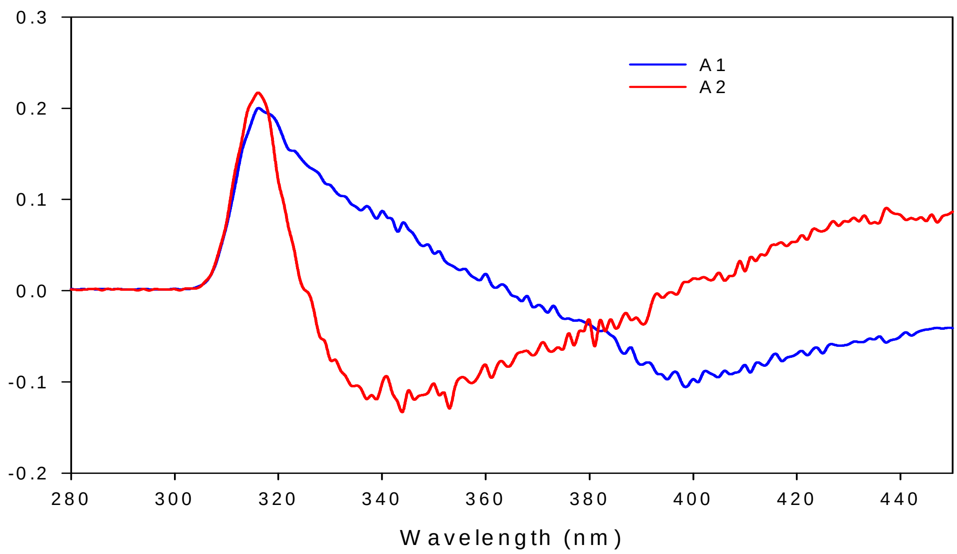

Fig.2. The differences of normalized fluorescence spectra of aromatic amino acids and nucleic acids according to the wave length

4. RESULTS AND DİSCUSSİONS

4.1 Aromatic amino acids and nucleic acids spectrum graphics

From Fig.1, it appeared that there are differences between the spectra of the different egg types. İn the quail egg the intensity of amino acids is lower than other egg types; duck egg's amino acid's intensity is higher. Spectrum of chicken white and brown egg and goose egg are between duck and quail egg's spectrum and has the same intensity. Normalized Fluorescence spectrum of amino acids and nucleic acids differs from each other by A1 and A2 components according to wavelengths of 320 nm, 340-360 nm and 400 nm (fig.2).

In this PCA analysis the graphic of A1 component against to A2 component gives the possibility to divide the eggs of different avian types (fig.3). The eggs of different avian types collecting in definite regions made groups.

According to A1 component quail egg collected in positive region, other egg types collected in negative region.

4.2 Maillard reaction products spectrum graphics

Maillard reaction products in egg whites were considered as powerful tool for the evaluation of egg freshness [12

The highest intensity of FMRP produced in egg whites was at chicken brown egg (fig.4); other eggs' intensity was at the middle.

Normalized Fluorescence spectrum of FMRP differences from each other by A1 and A2 components according to wavelengths of 425 nm, 450-480 nm (fig.5). This graphic gives the possibility to compare the eggs of different egg types by FMRP (fig.6). The eggs of different avian types collecting in definite regions made groups. Chicken white and brown egg and quail egg according to A3 component grouped at positive region, duck and goose at neutral region, according to A1 component chicken white, brown and goose egg moves towards neutral side. Quail egg according to A1 and A3 components is at positive side, duck egg according to A1 and A3 components is at negative side.

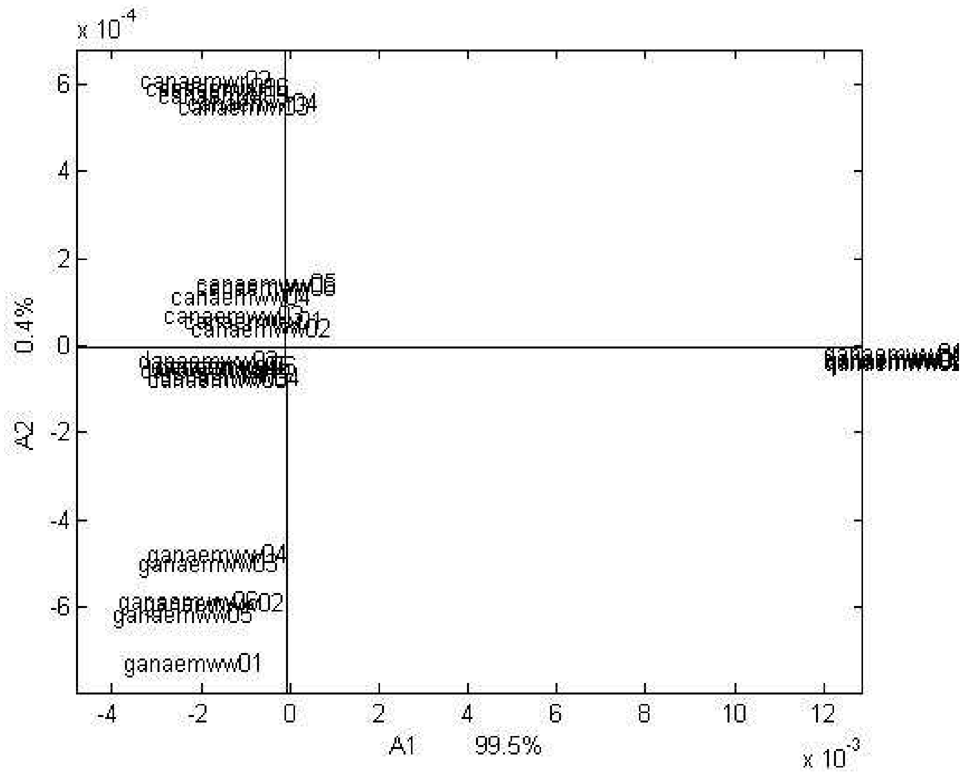

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Fig.3. Normalized spectra of egg whites according to A1 and A2 components of PCA. canaemww- chicken white egg, canaemwr- chicken brown egg, danaemww-duck egg, ganaemww- goose egg, qanaemww- quail egg.

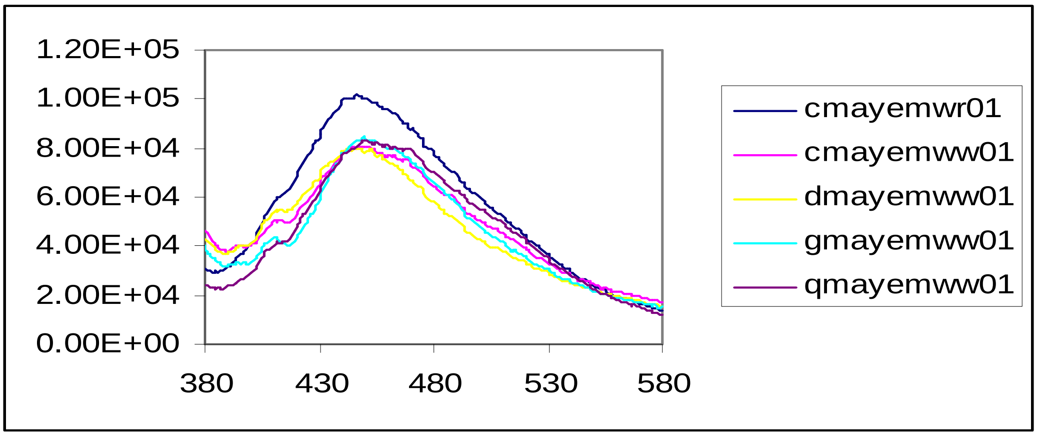

Fig.4. Normalized fluorescent Maillard reaction products fluorescence spectra recorded on egg whites.

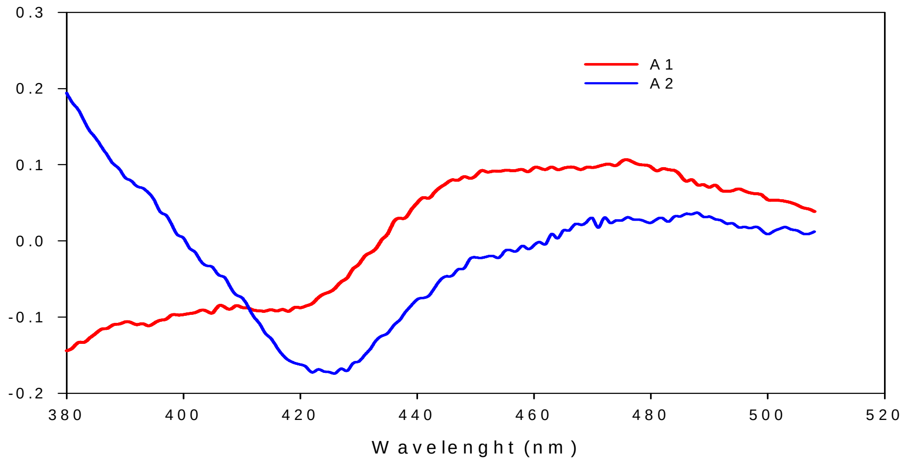

Fig.5. The differences of normalized fluorescence spectra of FMRP according to the wave length

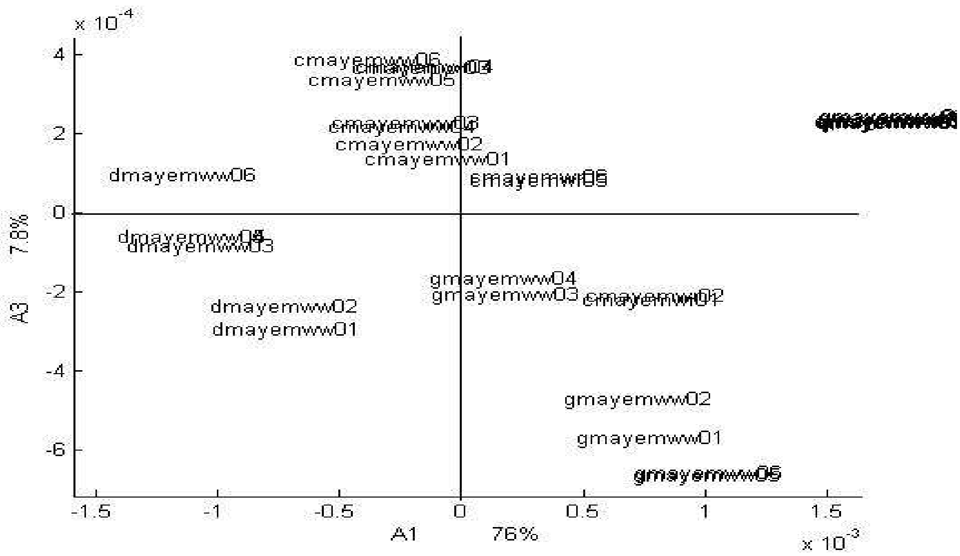

Fig.6. Normalized spectra of egg whites according to A1 and A3 components of PCA. cmayemww- chicken white egg, cmayemwr- chicken brown egg, dmayemww-duck egg, gmayemww- goose egg, qmayemww- quail egg.

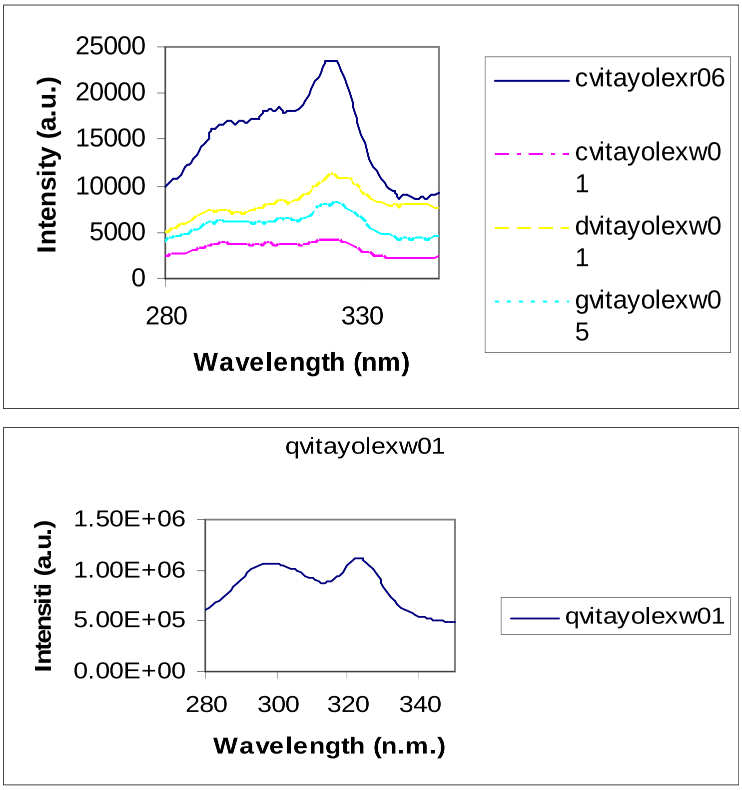

Fig.7. Normalized fluorescent vitamin A fluorescence spectra recorded on egg yolks

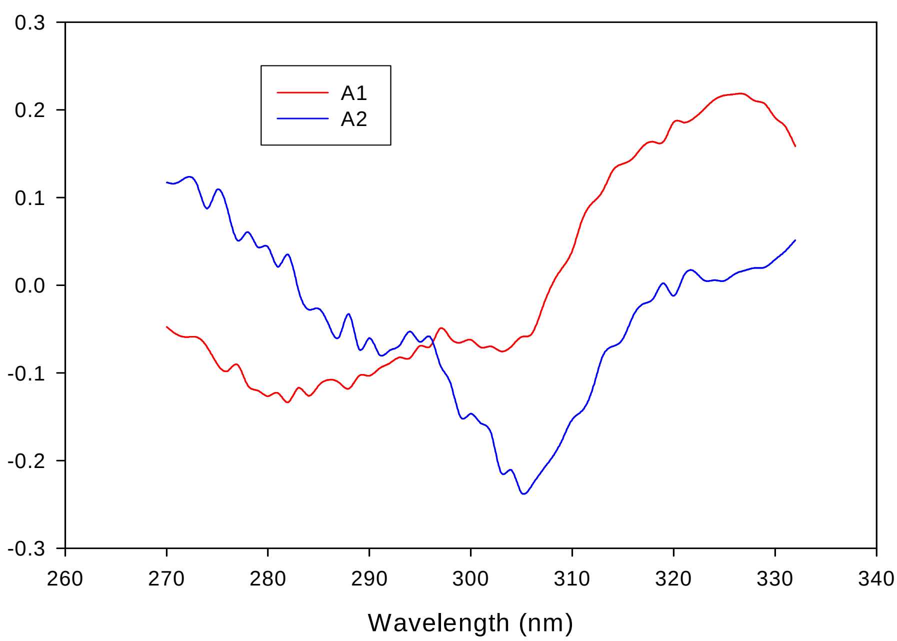

Fig.8. The differences of normalized fluorescence spectra of vitamin A in egg yolks according to the wave length

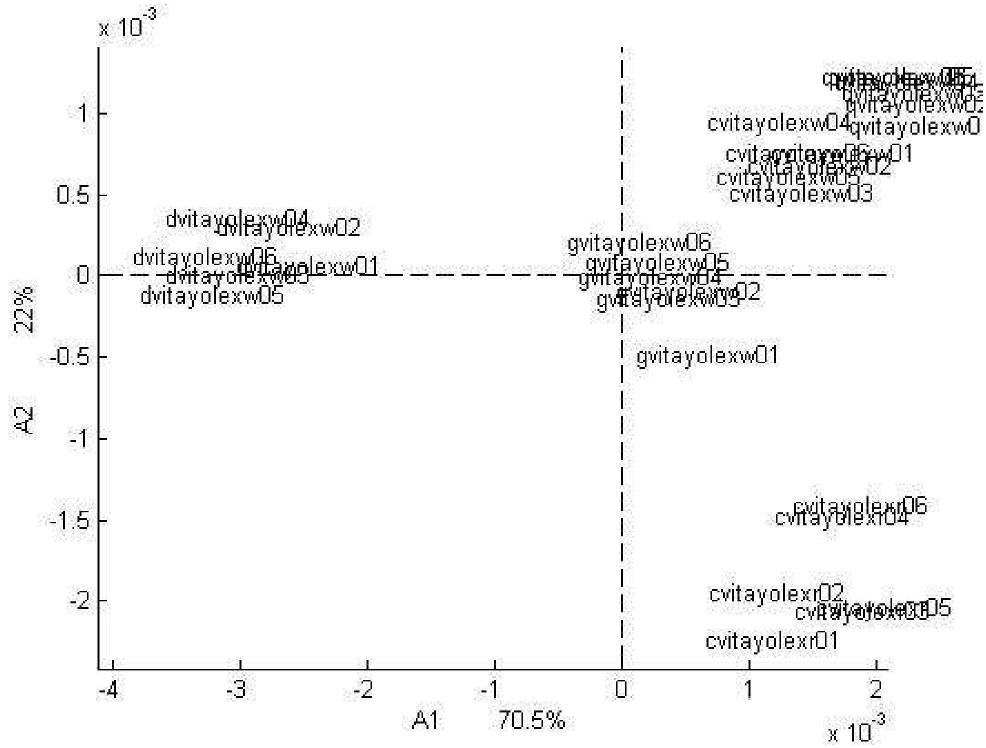

Fig.9. Normalized spectra of vitamin A in egg yolks according to A1 and A2 components of PCA. cmayemww- chicken white egg, cmayemwr- chicken brown egg, dmayemww-duck egg, gmayemww- goose egg, qmayemww- quail egg

4.3. Vitamin A in egg yolks spectrum graphics

According to intensity of vitamin A it is much higher in quail egg; the lowest intensity is at chicken white eggs (fig.7). Goose and duck egg are between chicken white and brown eggs.

Normalized Fluorescence spectrum of vitamin A differs from each other by A1 and A2 components according to wavelengths of 280-290 nm and 305 nm (fig.8).

This graphic gives the possibility to compare the eggs of different egg types by vitamin A (fig.9). The eggs of different avian types collecting in definite regions made groups. According to A1 component goose egg collected in neutral, duck egg is at negative region. Quail and chicken brown egg according to A1 and A2 component is at the positive region. Chicken white egg according to A1 component is at positive side, according to A2 component is at negative side.

4.4 PLS-DA results

Amino acids and nucleic acids				
	c	d	g	q
c	12			
d		6		
g			6	
q				6
FMR products				
	c	d	g	q
c	12			
d		6		
g			6	
q				6
Vitamin A				
	c	d	g	Q
c	12			
d		6		
g			6	
q				6

Fig.10. PLS-DA results

From fig.10 it is seen that each group of egg samples discriminates that type of eggs at 100 %/

5. CONCLUSIONS

After mathematical treatment of spectra analysis we can compare the eggs of different egg types.

In chicken brown egg the pick of intensity of vitamin A, aromatic amino acids and nucleic acids is high, according to FMRP it is the same with goose's egg. Chicken white egg gave the lowest intensity according to these parameters. Maillard reaction occurred between free amino acid groups in amino acid, peptide or protein structures and reduced sugars, as a result black and brown nitride polymers (melanoides) have been produced. İt is good if this reaction hasn't occurred, in the chicken's brown egg and goose's egg this index is high.

It means that chicken brown egg takes sunlight inside itself, because of that Maillard reaction is catalyzed. Reflecting of direct sunlight by white color can be the cause of low level of MRP in white eggs. FMRP spectrum in brown eggs may effect on the spectra of aromatic amino acids, nucleic acids and vitamin A. Therefore the pick of intensity in brown eggs must be high.

This method has given good results according to qualitative analysis in comparing eggs of different avian types by the parameters of aromatic amino acids, nucleic acids, vitamin A and FMRP.

Although PLSDA has given excellent results according to 5th component.

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