Comparative Study on Main Biochemical Indexes of Black Chicken and Domestic Chicken

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Abstract: Taking the serum of domestic chicken and black chicken as the research object, the content of cholesterol and the activity of glutamic pyruvic transaminase in serum were determined and compared, and the contents of albumin and γ-globulin in serum were separated and identified by cellulose acetate membrane electrophoresis. The results showed that the cholesterol content of domestic chicken was higher than that of black chicken, which was 1.3 times higher than that of black chicken. The glutamic pyruvic transaminase activity of black chicken was 1.35 times higher than that of domestic chicken. On the one hand, the study compares the nutritional value of the two kinds of chicken, which provides a new basis for the purchase of consumers and the development of related products; on the other hand, the level of serum ALT has important clinical significance in the early diagnosis, evaluation and prognosis of liver cirrhosis.

1. Introduction

Serum is a yellowish transparent liquid precipitated by blood coagulation. It can provide basic nutrients, hormones and various growth factors, binding proteins, contact-promoting and stretching factors, and can protect cells from mechanical damage. It contains a variety of plasma proteins (α, β, γ, albumin), polypeptides, cholesterol, glutamic pyruvic transaminase and so on [1]. Through the analysis of the composition of the serum, we can sense some changes in the body. The detection of glutamic pyruvic transaminase in serum can detect liver injury, so it can be used as a basis for the diagnosis of liver disease [2]. Modern medicine shows that the detection of the level of serum ALT has important clinical significance for the early diagnosis, evaluation and prognosis of liver cirrhosis. Because of the importance of serum, the determination of total cholesterol in serum is a routine item in blood lipid analysis. It is of certain significance to understand the serum components and the methods of analysis and identification of each component [3].

2. Materials and Methods

2.1 Materials and Reagents

Domestic chicken and black chicken serum and chicken liver (refrigerator)

Table 1 the Name of the Experiment and Its Experimental Reagents

Experimental project	Experimental reagent
Determination of serum total cholesterol by o-phthalaldehyde method	O-phthalaldehyde reagent, 90% acetic acid, mixed acid (90% acetic acid plus the same volume of concentrated sulfuric acid), standard cholesterol storage solution (1mg/ml), standard cholesterol application solution (0.1mg/ml)
Isolation and identification of serum albumin and γ-globulin	Gel chromatography of serum, PBS, (NH4) 2SO4 solution, biuret reagent, casein, sucrose, $BaCl_2$ solution. Sephandex G-25 gel, distilled water, potassium dichromate, blue glucan

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2.2 Methods

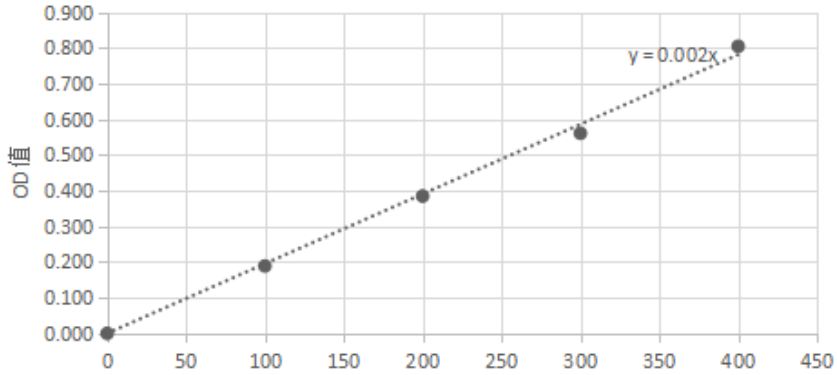
2.2.1 Determination of Serum Total Cholesterol by o-Phthalaldehyde Method

(1) Drawing of standard curve; Take 5 clean and dry test tubes, number them, and add reagents according to Table 2.

Table 2 Determination of Serum Total Cholesterol by o-Phthalaldehyde Method-- Drawing of Standard Curve

Serial number / reagents	0	1	2	3	4
Standard cholesterol application solution/ml	0	0.1	0.2	0.3	0.4
Acetic acid/ml	0.4	0.3	0.2	0.1	0
Phthalaldehyde reagent/ml	0.2	0.2	0.2	0.2	0.2
Distilled water/ml	0.01	0.01	0.01	0.01	0.01
Mixed acid/ml	4.0	4.0	4.0	4.0	4.0
The content of total cholesterol in 100mL/mg	0	100	200	300	400

(2) After adding, mixing and standing for 10 minutes, the standard curve was made with 550nm wavelength colorimetric determination, absorbance value as ordinate and cholesterol content as Abscissa. The standard curve is shown in figure 1.



Total cholesterol content / mg in 100mL serum

Fig.1 Standard Curve of Serum Total Cholesterol

(3) Determination of samples.

Take 2 clean and dry test tubes, number them, and add the reagent according to Table 3.

Table 3 Determination of Serum Total Cholesterol by o-Phthalaldehyde Method-Determination of Samples

Serial number / reagents	Control	Sample
Acetic acid/ml	0.4	0.4
Serum/ml	0.01	0.01
Phthalaldehyde reagent/ml	0	0.2
Absolute ethanol/ml	0.2	0
Mixed acid/ml*	4.0	4.0

Add, mix, stand for 10 minutes, colorimetric at 550nm wavelength, calibrate the zero point with the control tube, and the total cholesterol content (mg) in the 100mL sample can be obtained by the standard curve.

2.2.2 Isolation and Identification of Serum Albumin and Γ-Globulin

(1) Salting-out of serum albumin and γ-globulin.

Take a centrifuge tube, add 2mL serum, add 2mLPBS (diluted serum), shake well, add pH=7.2 (NH4) 2SO4 solution 2ml drop by drop, shake for 30min, centrifuge 20min (v=3000rpm). The supernatant (mainly containing albumin) was obtained by adding 3.132g (NH4) 2SO4 to make the

supernatant saturated and resting for 30 minutes. The precipitate obtained by centrifugation 20min (v=3000rpm) was dissolved with 1mLPBS, and the albumin was concentrated with sucrose after 1 hour of running water dialysis. Precipitation (mainly containing γ-globulin)-dissolved by adding 1mLPBS, adding saturated (NH4) 2SO4 solution 0.5mL (equivalent to 33% saturated (NH4) 2SO4 solution) for 30 minutes, centrifuging 20min (v=3000rpm) to precipitate the precipitate, then dialyzing for 1 hour with running water and concentrating with sucrose to obtain γ-globulin.

(2) Gel chromatography.

Pour the gel into the gel column, stir the water layer gently with a glass rod before each addition, continue to add the glue to the 10cm off the pipe mouth, place the water flat with the glue surface, add samples along the pipe wall, add some eluent, and finally elute with distilled water to collect the glue again.

(3) Identification of serum albumin and γ-globulin (cellulose acetate film electrophoresis).

The cellulose acetate film was soaked in buffer solution for 20 minutes, and the recognition surface and flax surface were removed. Absorb the excess liquid with filter paper, draw a thin line at 2cm at one end of the thin film hemp noodle with a pencil, dip the sample on one side of the slide, and sample at the thin line. Hang the film face down on the filter paper bridge of the electrophoretic cell bracket, and one end of the sample is close to the negative electrode; after electrification, make the U fan 80V, wait for the 10min, and make the U fan 120V electrophoresis 40min. After the film was taken out, the film was stained with amino black 10B dye and stained with 10min. Rinse and get the results.

3. Result

(1) Determination of serum total cholesterol by o-phthalaldehyde method.

The cholesterol content of domestic chicken was 30.5 mg per 100ml, and that of black chicken was 23.5mg.

(2) Isolation and identification of serum albumin and γ-globulin.

The main results are as follows: the experimental results of gel chromatography;

Identification of serum albumin and γ-globulin: (separation and identification of serum albumin by cellulose acetate film electrophoresis). From top to bottom, they are whole serum electrophoretic map, γ-globulin electrophoretic map and serum albumin electrophoretic map.

Fig.2 Separation of Serum Albumin by Cellulose Acetate Film Electrophoresis

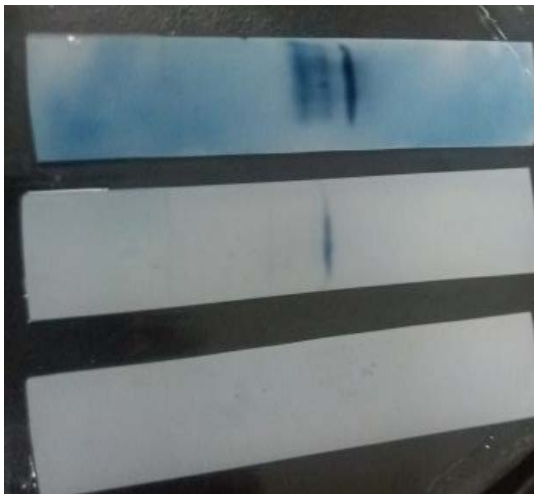


Fig.3 Identification of Serum Albumin by Cellulose Acetate Film Electrophoresis

(3) When determining the content of serum cholesterol, the method used is the o-phthalaldehyde method, and the measured data is relatively small, which may be due to the error of the reagent during the experimental operation, which leads to the change of the experimental results.

(4) Separation and identification of serum albumin and γ-globulin: A.When the mixed solution of potassium dichromate and blue glucan is added to the column, do not touch the gel surface and avoid the damage of the gel surface. When the sample passes through the chromatographic column, the material with higher molecular weight cannot enter the gel particles through the mesh, but flows along the gap between the gel particles, so the process is shorter and flows out of the chromatographic column first. B.The proteins on different bands can be analyzed by electrophoresis, and there are only four bands on chicken serum by cellulose acetate film electrophoresis. the four bands from negative to positive are γ globulin, β protein, α protein and albumin respectively. In this experiment, the separation and identification of different proteins can be mastered, and the proteins can be concentrated with sucrose at the same time. In the sampling process to be uniform, using a small number of methods.

4. Conclusion

(1) In this experiment, the content of serum total cholesterol was determined quantitatively by o-phthalaldehyde method, protein was separated and purified by salting-out and dialysis, and serum albumin and gamma globulin were separated and identified by cellulose acetate membrane electrophoresis [4].

(2) The experiment of cholesterol content determination shows that the cholesterol content of black chicken is lower than that of domestic chicken, so we can choose black chicken to reduce cholesterol intake and fat production.

(3) The activity of glutamic pyruvic transaminase in black chicken was higher than that in domestic chicken, which indicated that the protein metabolism in black chicken was exuberant.

(4) In this experiment, through the determination of cholesterol, protein and glutamic pyruvic transaminase activity in the serum of many chickens and black chickens, we know that there are some differences in species between domestic chickens and black chickens, and we can choose black chickens to get more nutrition [5].

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