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Research article

Research of juice-containing products with collagen additive "in vivo"

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Abstract

One of the most important tasks for food enterprises is the development of food rations and products with high biological value, enriched with essential substances. To expand the range of such products, non-traditional plant and animal raw materials are widely used. Protein raw materials are of particular interest, since they have structural and physiological significance for the human body. Collagen is one of the most popular and multifunctional bioactive supplements. Collagen is a major protein in human connective tissue: cartilage, cornea, arteries and skin. It affects their strength and energy, structure and elasticity, strengthens not only muscle fibers, but also smooth muscles, such as the heart. Collagen is constantly reproduced in the human body on its own, but after a person reaches 25 years of age, this process slows down and progressively weakens in old age. Collagen storage disorders can also be the body at levels over 90%, which is unattainable when collagen is taken in pill triggered in situations of constant overload (for example, in strength sports or in cases of intense physical activity). The liquid form of collagen for drinking is absorbed by or capsule form. We have developed recipes for a new range of juice products based on fruit and vegetable raw materials, enriched with vegetable and animal collagen, tomato, beef, pork and fish. The optimal amount of this biological additive in beverages has been determined, while ensuring the required quality of products, the preservation of its active properties has been studied. Also, studies have been carried out on juice products with a collagen supplement "in vivo".

Key words: juice products; animal and plant collagen; formulations; clinical research; blood; urine

Introduction

Today, collagen and its derivatives are increasingly considered as an active dietary supplement, which is included in the food product or taken orally by a person. Drinking collagen is actively used by athletes, third-party active lifestyles, as well as in various diets, it is recommended to be taken only for a limited time and in certain prescription use by other components. At the same time, there are food products such as juice drinks, nectars, which contain many useful components and the addition of collagen to them, which have improved their nutritional value. When using collagen in juices and drinks, it is necessary to take into account its and the active acidity of the product, which affects the process of its swelling, which will certainly affect the organoleptic characteristics of the drinks [1].

Our research was devoted to the development of juice-containing food drinks formulations using various types of animal and plant collagen. The main goal of the study was to ensure the minimization of the effect of collagen on the organoleptic and physicochemical characteristics of the developed recipe compositions of the finished product, while observing its useful biological active properties. The following research tasks were solved:

1. The assortment and organoleptic characteristics of different types of collagen of animal and vegetable tomato, beef, pork and fish origin have been investigated.

2. Studied the features of various types of collagen for their use in juice products. The mass fraction of collagen in ready-made juice-containing juice was determined, which is 5% and allows you to get a drink in terms of quality in accordance with the regulatory document.

3. The degree of biologically active protein supplements in the finished juice products of the supplement was investigated. It was determined that the most acceptable type of collagen for the enrichment of juice-containing products is collagen of animal origin - beef.

4. Formulations of juice-containing products with collagen have been developed - these are vegetables and berries, blended nectars and drinks with pulp.

5. The organoleptic and physicochemical indicators of the quality of ready-made juice drinks enriched with collagen and their compliance with the requirements of normative and technical documentation have been investigated [2, 3].

Clinical studies of blended fruit and berry enriched with collagen - nectars and juice products with pulp "in vivo" were also carried out.

Materials and Methods

In vivo studies of juice-containing products with collagen supplements were carried out on laboratory animals. As laboratory animals, nonlinear white rats were used, which were sexually mature animals at the age of 6-8 months and weighing 180-200 g. All animals were divided into 4 groups of 10 pieces each. The first 3 groups of animals were injected with nectar "Beet-mango-apple" with the addition of collagen of: beef, tomato, pork, respectively, made according to the recipe presented in table 1.

| Prescription component | The ratio of the components in the finished product,% | | | |
|-----------------------------|---|----------------------|----------------------|--|
| | Group No.1 | Group No.2 | Group No.3 | |
| Concentrated apple juice | 5,6 | 5,6 | 5,6 | |
| Concentrated beet juice | 15,5 | 15,5 | 15,5 | |
| Concentrated mango puree | 10,0 | 10,0 | 10,0 | |
| Sugar syrup (sugar) | 8,9 | 8,9 | 8,9 | |
| Citric acid | 0,07 | 0,07 | 0,07 | |
| Beef collagen | 5,0 | - | - | |
| Tomato collagen | - | 5,0 | - | |
| Pork collagen | - | - | 5,0 | |
| Water | brought up to 1000 l | brought up to 1000 l | brought up to 1000 l | |

Table 1: Recipe for nectar "Beet-Mango-Apple"

The group of animals No.4 was the control. This group of animals was injected with physiological saline. When administered, the animals in the control group experience the same stress as the animals in the research groups.

At the end of the ten-day period of the introduction of these solutions into the indicated groups of laboratory animals, the following analyzes were carried out to determine their functional state:

Clinical blood parameters (26 indicators)

- WBC (white blood cells) - the absolute content of leukocytes;

- RBC (red blood cells - red blood cells) - the absolute content of erythrocytes;

- HGB (Hb, hemoglobin) - the concentration of hemoglobin in whole blood;

- HCT (hematocrit) - hematocrit;

- PLT (platelets blood plates) the absolute content of platelets
- MCV average erythrocyte volume;

- MCH - the average content of hemoglobin in a single erythrocyte in absolute units;

- MCHC - the average concentration of hemoglobin in the erythrocyte mass;

- MPV (mean platelet volume) average platelet volume;
- PDW relative width of platelet distribution by volume;
- PCT (plate letcrit) thrombocyte;
- LYM (LY) (lymphocyte) the absolute content of lymphocytes;

- MXD (MID) - the absolute content of a mixture of monocytes, basophils and eosinophils;

- NEUT (NE) (neutrophils) the absolute content of neutrophils;
- MON (MO) (monocyte) the absolute content of monocytes;
- EOS absolute content of eosinophils;
- BAS the absolute content of basophils;
- IMM absolute content of immature granulocytes;
- ATL the absolute content of atypical lymphocytes;
- GR (GRAN) the absolute content of granulocytes;

- MCHC = HGB / HCT - the average concentration of hemoglobin in the erythrocyte;

- RDW – Red cell Distribution Width - coefficient of variation of the average volume of erythrocytes;

- RDW - CD - relative width of distribution of erythrocytes by volume, standard deviation;

- RDW - CV - relative width of distribution of erythrocytes by volume, coefficient of variation;

- P-LCR large platelet ratio;
- ESR (ESR) erythrocyte sedimentation rate.

Clinical examination of urine

The experiments were carried out using standard test-strips (Fig. 1 and Fig. 2), allowing to obtain the following specific values of clinical indicators of urine of animals, which show the effect of the injected solutions on their functional state.



Figure 1: Test-strips for determining protein content



Figure 2: Test-strips for determining the content of ketone bodies (acetone) and glucose

Results and discussion

beverages on blood counts

These indicators are:

- protein content;
- the content of ketone bodies (acetone);
- acidity (pH)
- glucose content.

The results of studies on the averaged values of blood parameters and the range of their change for group No.4, control animals, which were injected with saline instead of a juice-containing solution, are shown in table. 2.

Investigation of the effect of collagen supplementation in

| Blood parameter | Reference values | Experimental values |
|-----------------|------------------|---------------------|
| RBC | 6,49 -7,79 | 7,22 |
| HGB | 130-145 | 138 |
| НСТ | 0,386 - 0,419 | 0,40 |
| MCV | 54,1-59,8 | 56,82 |
| MCH | 18,1 - 20,1 | 19,15 |
| MCHC | 332-352 | 342,83 |
| RDW-CV | 8,9-10,3 | 9,25 |
| RDW-SD | 21-22,7 | 21,97 |
| PLT | 250 -909 | 557,33 |
| PCT | 0,0009 - 0,0049 | 0,0033 |
| MPV | 7,6 - 7,9 | 7,76 |
| PDW | 10,0 - 11,5 | 11,0 |

| P-LCC | 39-118 | 79,16 | |
|-------|-------------|-------|--|
| P-LCR | 13,0 - 15,7 | 14,41 | |
| WBC | 6,25 - 13,6 | 7,54 | |
| NEU | 0,23 - 7,37 | 2,66 | |
| LYM | 3,92 - 4,98 | 4,37 | |
| MON | 0,05 - 0,42 | 0,14 | |
| EOS | 0,15 - 0,75 | 0,33 | |
| BAS | 0,03 - 0,24 | 0,08 | |
| LIC | 0,01 - 0,16 | 0,05 | |

Table 2: Indicators of blood analysis of animals of the control group No.4

Since similar blood parameters of experimental animals were measured in other groups, their comparison will reveal the effect of collagen supplements in juice drinks on the physiological state of the studied animals.

| | U | Table 3, the fol | llowing comparison |
|-----------|------------|------------------|--------------------|
| Parameter | Group No.1 | Group No.2 | Group No.3 |
| RBC | 7,79 | 7,74 | 8,23 |
| HGB | 143,60 | 147,71 | 155,57 |
| HCT | 0,43 | 0,44 | 0,45 |
| MCV | 54,72 | 56,78 | 54,57 |
| MCH | 18,48 | 19,09 | 18,89 |
| MCHC | 337,2 | 336,29 | 345,71 |
| RDW-CV | 9,2 | 9,36 | 9,24 |
| RDW-SD | 21,66 | 22,30 | 21,69 |
| PLT | 617,20 | 556,43 | 617,86 |
| PCT | 0,004 | 0,004 | 0,005 |
| MPV | 7,36 | 7,29 | 7,71 |
| PDW | 10,04 | 9,60 | 10,99 |
| P-LCC | 68,60 | 58,14 | 84,71 |
| P-LCR | 11,22 | 10,74 | 13,89 |
| WBC | 6,71 | 5,81 | 6,61 |
| NEU | 1,80 | 1,60 | 2,49 |
| LYM | 4,64 | 3,92 | 3,88 |
| MON | 0,07 | 0,08 | 0,07 |
| EOS | 0,08 | 0,13 | 0,07 |
| BAS | 0,11 | 0,07 | 0,08 |
| LIC | 0,01 | 0,01 | 0,02 |

Table 3: Comparison of indicators of blood analysis of different groups of animals

An analysis of the comparison of the results obtained allows us to conclude that the introduction of both a juice drink of the basic recipe and a juice drink made according to recipes enriched with collagen of various origins to animals leads to changes in some blood parameters of experimental animals. Table 4 shows that the deviation of these single blood parameters is in different cases - from + 7.5% to -80.0%.

| Parameter | Norm (average baseline) | Group No.1 | Deviation from the norm, % | Group No.2 | Deviation from the norm, % | Group No.3 | Deviation from the norm, % |
|-----------|-------------------------------|---------------|----------------------------------|---------------|----------------------------------|---------------|----------------------------------|
| RBC | 7,22 | | | | | 8,23 | +13,9 |
| HGB | 138,0 | | | | | 155,57 | +17,6 |
| HCT | 0,40 | 0,43 | +7,5 | 0,44 | +10,0 | 0,45 | +12,5 |
| PDW | 11,0 | | | 9,60 | -12,7 | | |
| P-LCR | 14,42 | | | 10,74 | -25,5 | | |
| WBC | 7,55 | | | 5,81 | -23,1 | | |
| EOS | 0,33 | 0,08 | -75,8 | | | | |
| LIC | 0,05 | 0,01 | -80,0 | 0,01 | -80,0 | | |

Table 4: Deviation of single indicators of blood analysis of different groups of animals from the basic indicators

However, such deviations arise when comparing the corresponding blood parameters in the groups of animals with the corresponding average baseline in the control group of animals No.4.

Table 2 shows the range of changes in each indicator of the blood test in the control group of animals No.4. If we compare the isolated blood parameters in each group of experimental animals, taking into account their range of change in the control group, then the difference in almost all the noted indicators in all groups will not exceed from 2.6% to 7.4%.

This difference does not exceed the accuracy of the methods for determining all the parameters under consideration [4] and, therefore, the

data obtained on the blood composition of experimental animals after the introduction of a juice-containing drink without and with collagen additives of various origins practically do not differ from the control values. This is what is illustrated by the data given in table. 5.

| Parameter | Norm (average baseline) | Group No.1 | Deviation from the norm, % | Group No.2 | Deviation from the norm, % | Group No.3 | Deviation from the norm, % |
|-----------|-------------------------------|---------------|----------------------------------|---------------|----------------------------------|---------------|----------------------------------|
| RBC | 6,49 -7,79 | | , | | , | 8,23 | 5,6 |
| HGB | 130-145 | | | | | 155,57 | 7,29 |
| HCT | 0,386 - 0,419 | 0,43 | 2,60 | 0,44 | 5,0 | 0,45 | 7,40 |
| PDW | 10,0 - 11,5 | | | 9,60 | 4,0 | | |
| P-LCR | 13,0 - 15,7 | | | 10,74 | 17,0 | | |
| WBC | 6,25 - 13,6 | | | 5,81 | 7,0 | | |
| EOS | 0,15 - 0,75 | 0,08 | 6,70 | | | | |
| LIC | 0,01 - 0,16 | 0,01 | 0 | 0,01 | 0 | | |

 Table 5: Deviation of single indicators of blood analysis of different groups of animals from the basic indicators, taking into account their range of change

Only 2 of the given 8 indicators do not fall into the specified range of deviations. These are: P-LCR - coefficient of large platelets; EOS is the absolute content of eosinophils. Both indicators are very dangerous when the normative values are exceeded, since they may indicate significant problems associated with ischemic heart disease and the occurrence of blood clots [5], as well as chronic pathologies of the gastrointestinal tract and malignant diseases [6].

However, in our case, both indicators do not exceed the indicated limits, but, on the contrary, are somewhat lower. Such deviations are possible in the presence of stressful situations in experimental animals. The introduction of solutions to animals for ten days, of course, were stressful conditions and a decrease, rather than an increase in the P-LCR and EOS indices, indicates that the injected solutions do not affect the blood parameters [7].

Study of the Effect of Collagen Additives in Beverages on Urine Parameters

When conducting a study of urine of experimental animals, the following indicators were determined:

- The value of the pH value;
- Protein content;
- Glucose content;
- The content of ketone bodies acetone.

As in blood studies, four groups of experimental animals took part in the experiments. The analysis of the data obtained is given in table. 6, which compares the indicators of the control group of animals with the corresponding indicators of the other three groups. The structure of the table is similar to table. 1, however, due to the fact that urine indicators were determined using the color change of the test strips, the last column of the table contains not the average value of the parameter, but the number of test strips with its specific value.

| Parameter | Range | Number of test-strips |
|-------------------------------------|--------|-----------------------|
| pН | 6,0 | 10 |
| protein content, mg/100 ml | Neg ++ | Neg 3 |
| + (30) | | + - 4 |
| ++(100) | | ++ - 3 |
| glucose content, mg/100 ml | Neg. | 10 |
| content of ketone bodies, mg/100 ml | 2,5 | 10 |

Table 6: Indicators of urine analysis of animals in the control group

Table 7-9 show urine analyzes of animals in groups.

(Recipe for pork collagen drink)

| Parameter | Range | Number of test-strips |
|----------------------------------|----------|-----------------------|
| pH | 6,0 -7,5 | 6,0 - 4 |
| | | 6,5-2 |
| | | 7,5 - 6 |
| protein content, mg/100 ml | Neg +++ | Neg 3 |
| + (30) | | + - 5 |
| ++(100) | | ++ - 0 |
| +++(300) | | +++ -2 |
| glucose content, mg/100 ml | Neg. | 10 |
| content of ketone bodies, mg/100 | Neg. | 10 |
| ml | | |
| ml | 1105. | 10 |

Table 7: Indicators of urine analysis of animals in group No.3

(Recipe for beef collagen drink)

| Parameter | Range | Number of test-strips |
|-------------------------------------|----------|-----------------------|
| pH | 6,0 -8,0 | 6,0-2 |
| | | 7,0-5 |
| | | 8,0 - 3 |
| protein content, mg/100 ml | Neg ++ | Neg 6 |
| + (30) | | + - 5 |
| ++(100) | | ++ - 3 |
| | | |
| glucose content, mg/100 ml | Neg. | 10 |
| content of ketone bodies, mg/100 ml | Neg. | 10 |

Table 8: Indicators of urine analysis of animals in group No.1

(Recipe for a drink with tomato collagen)

| Parameter | Range | Number of test-strips | | |
|---|----------|-----------------------|--|--|
| pH | 6,0 -8,0 | 6,0 - 2 | | |
| | | 6,5-2 | | |
| | | 8,0 - 6 | | |
| protein content, mg/100 ml | Neg + + | Neg 2 | | |
| + (30) | | + - 6 | | |
| ++(100) | | ++ - 2 | | |
| glucose content, mg/100 ml | Neg. | 10 | | |
| content of ketone bodies, mg/100 ml | Neg. | 10 | | |
| Table 9: Indicators of urine analysis of animals in group No. 2 | | | | |

Conclusions

Analyzing the presented results, it can be concluded that an increase in the pH value of the urine of the studied animals does not lead to significant changes in its main indicators. Moreover, with the introduction of samples of juice-containing products, which are enriched with collagen of various origins, to animals, the quantitative protein content did not increase, but even decreased. At the same time, the content of ketone bodies and the content of glucose in the urine of animals did not change and was negative.

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