

ELISA

Materials and methods

Immunological reactions were assessed by solid-phase non-competitive indirect enzyme immunoassay using a commercial Reagent Kit for semi-quantitative enzyme immunoassay for allergen-specific IgG antibodies (LLC NPO Immunoteks, Stavropol, Russia). This Kit uses monoclonal anti-IgG antibodies included in the peroxidase conjugate, capable of detecting antibodies of the immunoglobulin G class in human serum/plasma, which interact affinity with allergens sorbed on the surface of a polystyrene tablet. The unbound components of the analyzed sample and the excess of the conjugate are washed off, and the activity of peroxidase in the immune kits is determined using a chromogenic substrate (tetramethylbenzidine).

One Kit is designed to determine the content of specific IgG in eight test samples to 22 allergens (Table 1).

Table S3. Food allergens in the Reagent Kit for semi-quantitative enzyme immunoassay for allergen-specific IgG antibodies

No.	Allergen	No.	Allergen
1.	Milk concentrate protein (micellar casein)	12.	Beef protein hydrolyzate (beef protein)
2.	Dry egg white (egg protein)	13.	Whey Protein Concentrate (Whey Protein)
3.	Orange	14.	Corn grits
4.	Potato	15.	Baker's yeast
5.	Peanut	16.	Hazelnut
6.	Honey	17.	Strawberry
7.	Almond	18.	Pomegranate
8.	Oat groats	19.	Coconut
9.	A mixture of brown and wild rice	20.	Flax seed
10.	Soy protein isolate (soy protein)	21.	Wheat groats
11.	Barley groats	22.	Brewer's yeast

ELISA was carried out on a Multiskan FC microplate photometer (Thermo Scientific, USA) using the instructions for the Reagent Kit provided by the manufacturer.

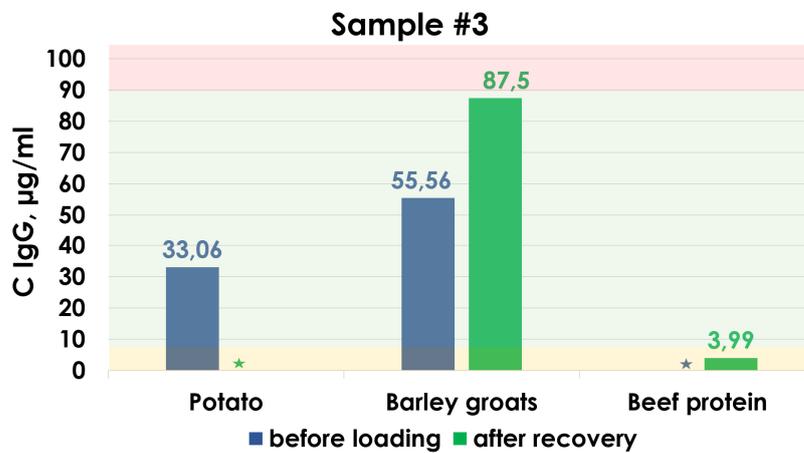
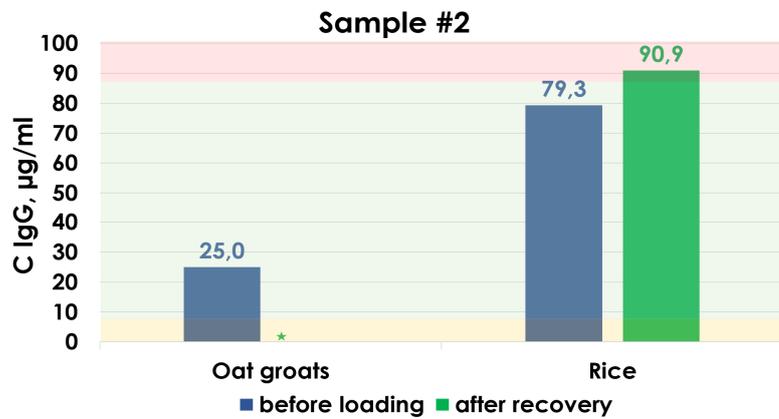
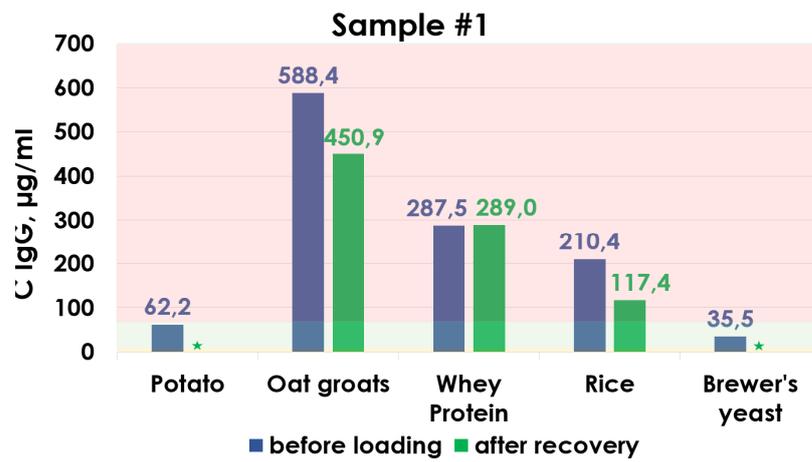
Interpret the results obtained according to the manufacturer's instructions (Table 2)

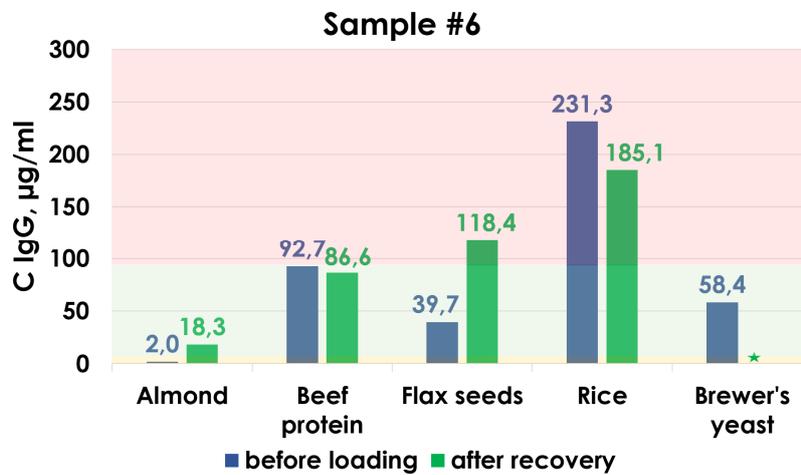
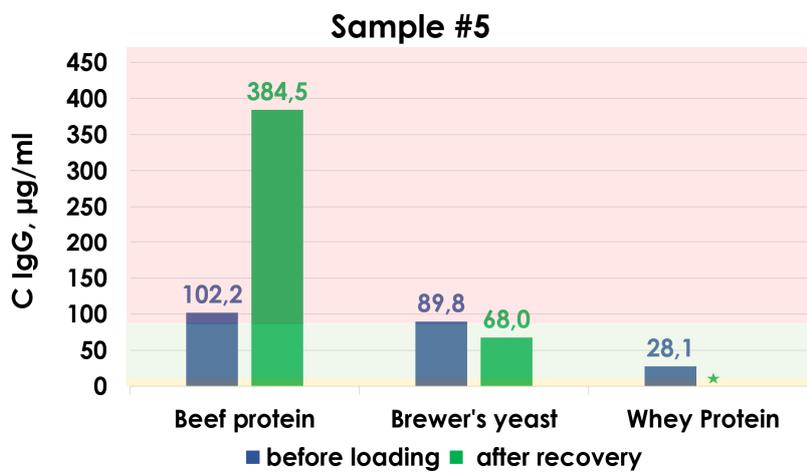
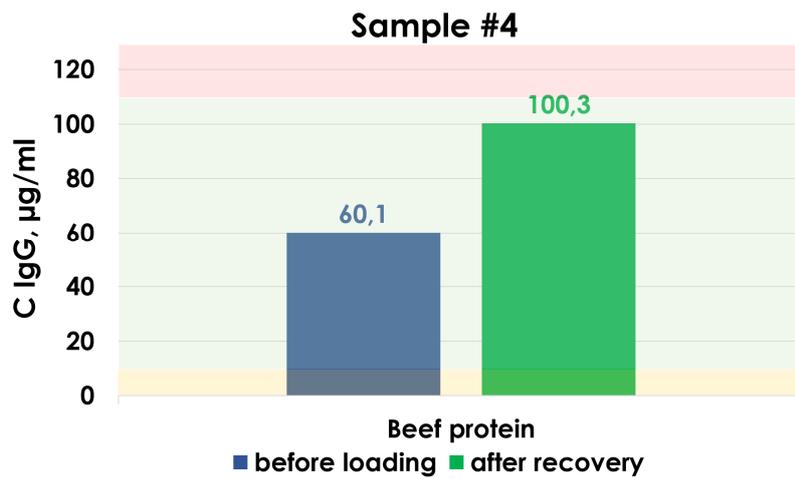
Table S4. Interpretation of ELISA results

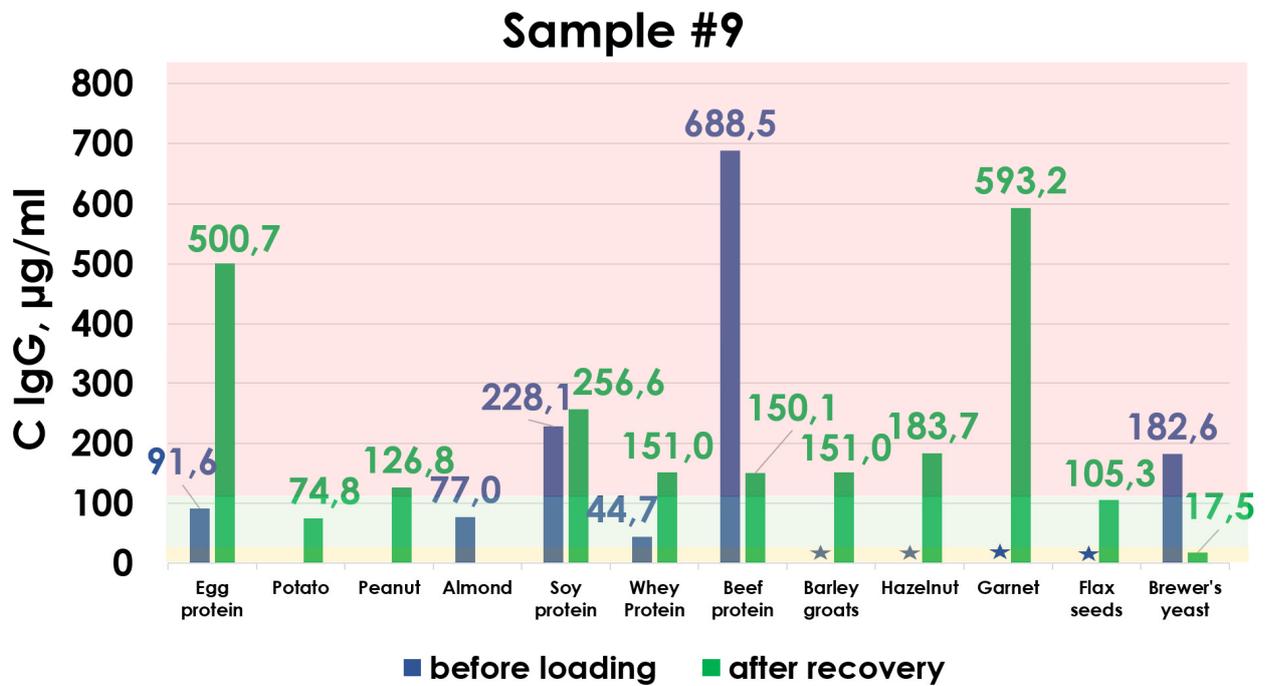
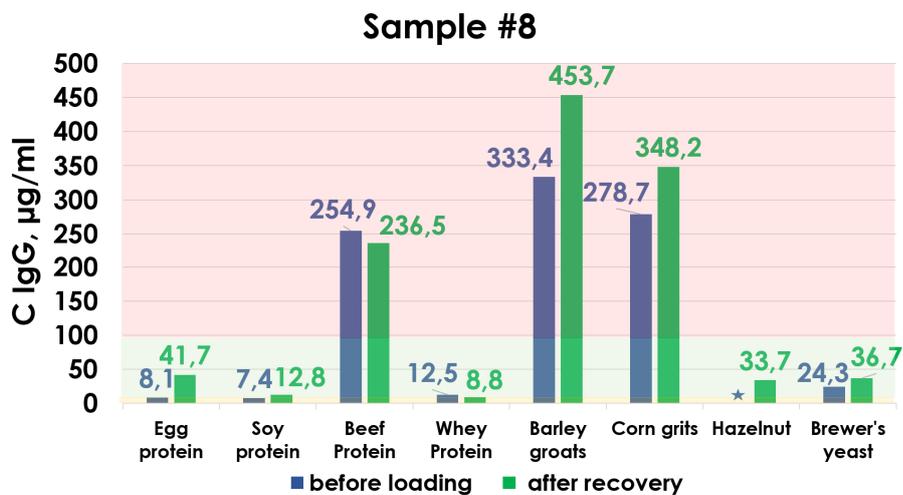
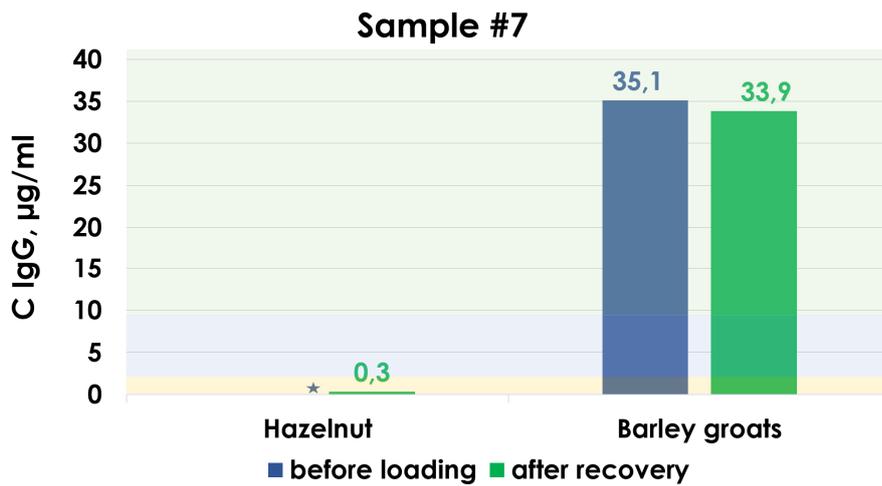
IgG concentration ($\mu\text{g/ml}$)	Result
0	Negative
0-0,88	Mild allergic reaction
0,88-8,8	Moderate allergic reaction
8,8-88	High allergic reaction
88-880	Very high allergic reaction

Results of determination of hyperreactive reaction to food allergens mediated by immune processes

As a result of a study of a group of athletes aimed at determining IgG-mediated immunological reactions to 22 food antigens in plasma samples, it was found that all participants in the study had an allergic reaction to food components (Figure 1).







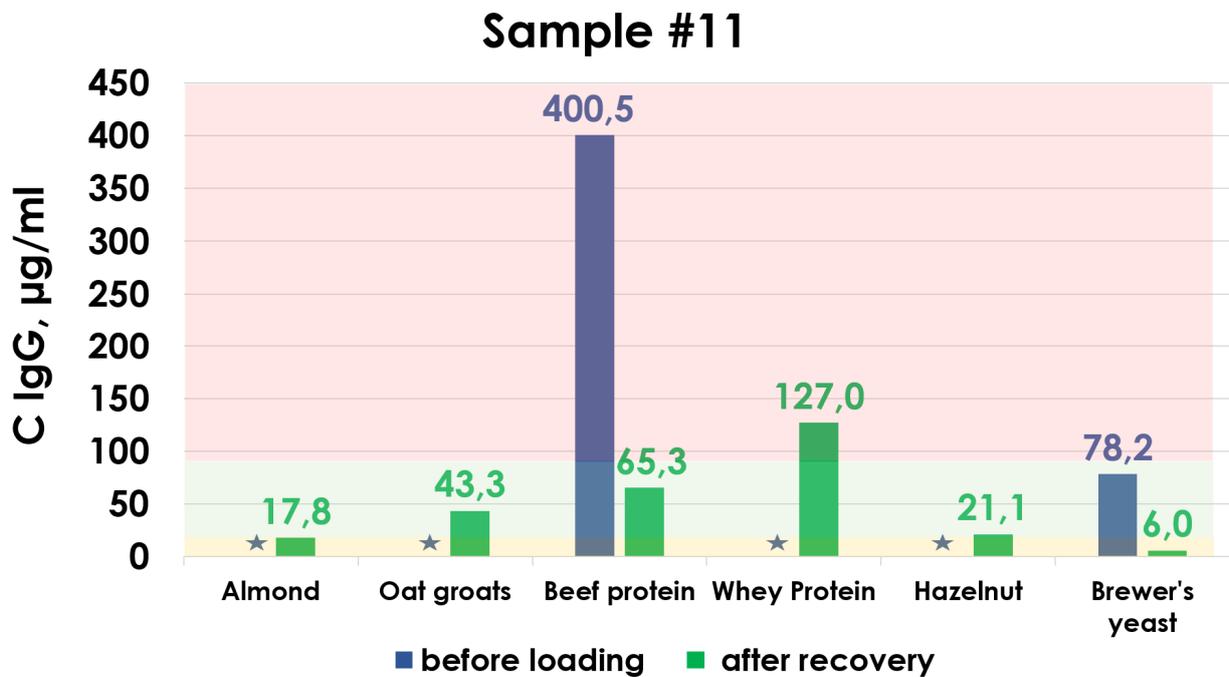
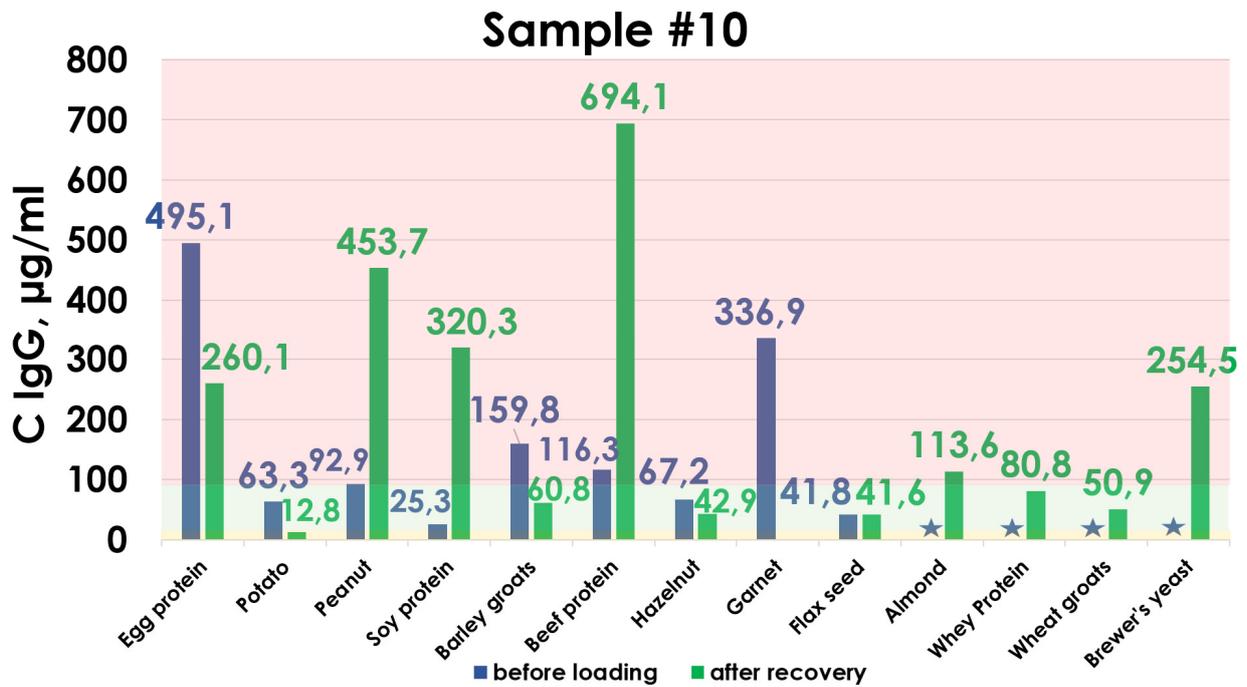
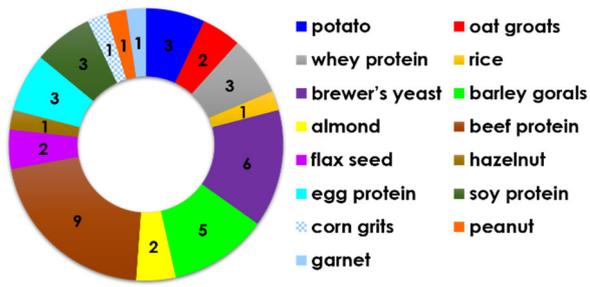


Figure S1. Results of quantitative determination of the content of IgG class antibodies in the blood plasma of athletes (the number of technical repetitions is 3). Allergens are presented for which the IgG concentration was greater than 0.88 µg/ml (hyperreactivity to food allergens). An asterisk indicates allergens for which the calculated IgG concentration was 0 µg/ml. The yellow and blue zones correspond to a mild and moderate negative allergic reaction, respectively. The green and pink zones are high and very high allergic reactions, respectively.

a



b

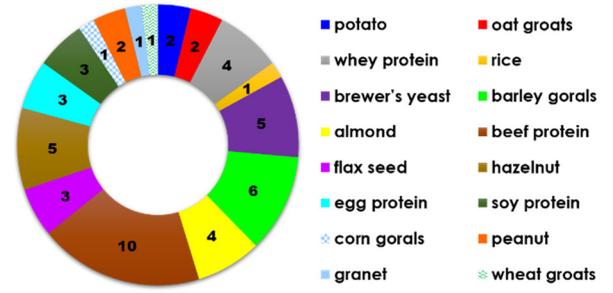


Figure S2. The occurrence of an allergic reaction to food components before exercise (a) and after a recovery period of 24 hours (b).