### Leather industry waste used to obtain biostimulant protein gels

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Leather industry generates significant amounts of solid and liquid waste representing a great threat to environment. Previous research reported that the animal tissue by-products might be recovered into valuable products due to their rich content of protein and fat, carbohydrate, minerals, vitamins as well as other nutrients (Chojnacka *et al.*, 2021). Hydrolysis, extraction and enzymatic hydrolysis are mainly used to valorize leather solid waste through obtaining protein hydrolysates which are included in the plant biostimulants category. Increased interest in protein hydrolysates was noted due to their beneficial effects on crop performances (Du Jardin, 2015). The hydrolysates contained mainly glycine, alanine and proline, which are involved in stimulating plant growth by supporting chlorophyll synthesis and chelating micronutrients (Mikula *et al.*, 2023), or improving the resilience of plants to abiotic stress.

Good results have been obtained in terms of yield and quality by using biostimulants in tomato crops. Tomato (*Lycopersicon esculentum* Mill.) is a vegetable with important role in the human diet, representing a source of healthy compounds such as vitamins, minerals, lycopene,  $\beta$ -carotene, and anticancer agents (Dias, 2012).

The research performed in this study was focused on testing a protein gel based on bovine gelatin and keratin hydrolysate on tomato crop and their effects on the seedlings were assessed. Positive effects of the treatment applied were recorded regarding the biometric parameters and the content of assimilatory pigments, dry matter and total soluble sugars of the tomato seedlings leaves.

### Materials and methods

Bovine hide and sheep wool were supplied by a slaughterhouse and a sheep farmer from the Constanta County, Romania. The gelatine was obtained by acid hydrolysis from bovine delimed hide at high temperature. The keratine hydrolysate was obtained from sheep wool by alkaline hydrolysis. A combination of bovine gelatin with keratin mixed in a 1:1 ratio (labeled GBK) were used for the treatment of the tomato plants by application on the root plants. The protein gel was physico-chemically characterized according to the standards in force regarding the dry substance, total ash, total nitrogen, free amino acids and protein content, pH, bloom test, and viscosity.

Tomato seeds (*Lycopersicon esculentum* Mill.) of the BPK 16021 hybrid supplied by Marcoser SRL (Galati County, Romania) were selected for the research. The experiment consisting of two variants (untreated control and seedlings treated with the protein gel GBK) was installed in the experimental greenhouses from USAMV Bucharest. Three treatments with stimulant gel were applied on the tomato seedlings by watering the soil once every 10 days with diluted solutions of gel (30 g in 150 mL of water). The control group received only equal amount of water.

The seedlings were measured regarding biometric parameters (plant height and number of leaves) and content in total chlorophyll, carotenoids, dry matter and soluble sugars. Methods used for the measurement of these parameters were according to the standards in force or in house methods.

#### **Results and discussions**

Table 1 presents the physico-chemical characterization of the tested protein gel (GBK). Good contents of protein and amino acids are noted, which are important for the development of tomato plants, promoting the rapid biosynthesis of different types of proteins.

Table 1.1 hysico-chemical characteristics of the used protein get					
Dry substance (%)	31.32	Total free amino-acids (mg/g)	1.50		
Total ash (%)	9.36	pH	5.59		
Total nitrogen (%)	15	Bloom test (g)	130		
Protein content (%)	84.26	Viscosity (mPa*s)	2		

Table 1. Physico-chemical characteristics of the used protein gel

During the experiment the tomato seedlings cultivated in the greenhouse (Figure 1) were monitored and measured.



Figure 1. Experimental variants of tomato seedlings in the greenhouse

Biometric measurements of the seedlings showed higher values of the average number of leaves (by 37.5%) and an increase of plants height (by 27.8%) after the last treatment compared to the untreated plants (Figure 2).



Figure 2. Variability of the biometric parameters of the tomato seedlings

Biochemical analyses indicated higher amounts of assimilatory pigments (3.25 times more chlorophyll and 2.25 times more carotenoids respectively) in the leaves of the treated plants relative to the control plants (Table 2). This increase positively influenced the photosynthetic activity and consequently the achievement of higher content of dry matter and total sugars in the seedlings leaves (Table 2).

Table 2. Variability of some biochemical parameters of the tomato seedlings					
Experimental	Total	Carotenoids	Dry matter	Total soluble	
variants	chlorophyll	(mg/g)	(%)	sugars	
	(mg/g)			(mg/100 g)	
Control	$0.72{\pm}0.02$	$0.028 \pm 0.0014$	$14.58 \pm 0.49$	26.73±2.84	
GBK	$2.34{\pm}0.09$	$0.063 \pm 0.0054$	26.75±1.07	74.12±2.78	

# Conclusions

Beneficial effects of the protein gel treatment were noticed on the analyzed parameters in tomato seedling leaves. The biostimulant activity of the tested product was attributed to the amino acids content, promoting the chlorophyll biosynthesis in leaves and thus a better accumulation of primary metabolites (soluble sugars).

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