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Determination of Total Vegetal Proteins of *Cucumus Sativus*

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Abstract: Vegetal proteins are part of the human diet, which contain amino acids and thus promote good health. *Cucumis sativus* is commonly known as a cucumber and is valued not only for its unique taste characteristics but also for its possible nutritive value. Thus, the objective of this study is to provide a quantitative and qualitative assessment of the vegetal proteins of *Cucumis sativus* in combinations of different genotypes with varying combinability. It has been about the use of samples from high and low combinative capacity genotypes, and quantification of water content and protein quantity by spectrophotometric methods. The investigation, therefore, indicates distinct variations in the genotypes in the proportion of water and the amount of protein. In genotypes of high combinative capacity, Hybrid 4 was found to have the highest protein content in terms of 10.02 mg/mL dry material in the hybrid form and 9.76 mg/mL in the maternal form. In low combinative capacity genotypes, Hybrid 6 revealed a slight protein content of 8.37 mg/mL as compared to the Maternal form, comprising 9.60 mg/mL. Hybrid 4 was found to contain a total average of 267.22 mg/mL of protein, which signified good nutritional value. On the other hand, Hybrid 6 was the lowest in average quantity of protein at 223.10 mg/mL. All the measurements and variations in this study were highly significant ($p > 0.05$), which implied reduced variability within each genotype group. These studies can be useful to explore the knowledge of proteomics in Cucumis sativus, and they suggest its general usability in human nutrition that can be further advanced to discover its health and dietary potentials.

Keywords: *Cucurbitaceae; Vegetal Proteins; Plants*

1. Introduction

The cucumber is a member of the Cucurbitaceae family. The cucumber is classified within the genus Cucumis, which has 20 to 25 species predominantly located in Asia and Africa [1]. Only two species, *Cucumis sativus* (cucumber) and *Cucumis melo* (melon, muskmelon, and Persian melon), hold commercial significance. It is a perennial climbing or trailing vine, typically exhibiting blooms of both sexes on a single plant (monoecious). Contemporary market hybrids are generated from genetically gynoecious lines, which exclusively produce female flowers [2]. Commercial seed batches contain up to 10% monoecious varieties to ensure adequate pollen for fruit set. European greenhouse cucumbers exhibit parthenocarpy, producing fruit without pollination [3]. The cucumber originates from northwest India and has been cultivated in that region for a minimum of 3000 years. The cucumber was recognized in France during the 9th century and became prevalent in England by 1327 [4].

Cucumbers are highly susceptible to low temperatures and can perish at 1°C. The minimum germination temperature is 16°C, the optimal range is 16°C to 35°C, the ideal temperature is 35°C, and the highest germination temperature is 40°C.

The soil temperature at planting must be at least 10°C for table cultivars and 13°C for gherkins.

cultivars. Pickling cucumber yields depend on the size of the fruit harvested. (15,000--17,000 kg per hectare). These products contain fair amounts of potassium, calcium, and folate and small amounts of other nutrients, including vitamin C, and are very low in calories [5].

Proteins are the main organic compounds in plants. As constructive compounds, proteins are contained in the cell wall, plasmatic membranes, cytoplasm, and cell organelles. In plants, the number of proteins varies between 0.2 and 6.5%, and in seeds, this percentage may exceed 21.3% [6].

The protein synthetic complex (nuclear, mitochondrial, plastid, and cytoplasmic) of a cell reflects the hereditary specificity of the genotype (race, line, or species). Quantitative and qualitative research on proteins elucidates the differential activity of genes and helps to establish the degree of gene expression regulation in cells and in plant tissues.

Vegetal proteins also play a functional role in human food intake and provide important amino acids as well as vitamins and minerals [7, 8]. The protein composition and identity of the cucumber have not been studied in detail. The evaluation of vegetal proteins is essential because the identification of their quantitative and qualitative characteristics is valuable for determining their health advantages and uses, as closely related to food science and nutrition.

Although a number of studies have focused on the overall nutritional quality of cucumbers, few a priori metrics of cucumber protein concentration have been systematically quantified or compared in terms of quality. This study intends to address this problem by presenting a comprehensive study on the identification and characterization of the vegetal protein *Cucumis sativus*, its mode of action, the health benefits associated with proteins, and how it may be useful in the diet.

2. Materials and methods

2.1 Sample collection and herbarium identification

Two cucumber families with parental traits and hybrids obtained after crossing were investigated. The parental genotypes of these two hybrids are characterized by HCC and LCC parental forms. The seeds were obtained from the Institute of Tiraspol and kindly provided by Dr.T. Homenco.

2.2 Protein determination

The amount of protein was calculated via Bradford's method [9]. The Bradford method is based on the noncovalent binding of the anionic form of the dye Coomassie Blue G-250 with protein [9]. The dye reacts chiefly with arginine residues, which have a positively charged side chain, and slight interactions have also been observed with basic residues (histidine and lysine) and aromatic residues (tyrosine, tryptophan, and phenylalanine). In the absence of protein, the dye reagent is pale red, and upon binding to protein, a blue color is generated with an absorbance maximum at 590 nm. It provides a nonlinear calibration curve of A590 against concentrations over the 0.2–20 mg range of total protein contained in a 20 mL sample volume (0.01–1 mg/mL). In the Bradford assay, curvature is due to depletion of free dye at high protein concentrations, and a better approximation of linearity can be achieved by plotting A595–A465 against concentration to take dye depletion into account. A 20 µl sample was mixed with 50 µl of 1 M NaOH. After this mixture was mixed, 1 ml of dye was added, and the mixture was incubated for 5 min. Then, the optical density was measured at 595 nm [10]. The amount of protein was calculated via the following formula:

$$C=(DO595\cdot Vp)/(0.060\cdot Vt)$$

C=The amount of protein. (mg/mL)

DO595= optical density under a wavelength of 595 nm.

Vp= volume of sample. (0.1 mg/ml) volume of sample

Vt =Volume of reaction medium. (3.0 mg/ml)

The protein concentrations in fresh and dry vegetal samples of genetically different HCC and LCC plants were subsequently calculated. To observe the differences between the given genotypes via polyacrylamide gel electrophoresis, electrophoresis was used. The protein extracts of the leaves were identified via polyacrylamide gel electrophoresis according to Laemmli (1970), who used 12% (w/v) polyacrylamide. Each sample (30 Fg protein) was loaded onto the slots. The gel was stained with Coomassie blue R250 and destained with aqueous acetic acid and methanol. Molecular weight markers ranging from 14 to 70 kDa were used to estimate the molecular weight of the purified protein. The gel was densitometrically scanned by using a ProAnalyzer scanner to measure the quantity of each band (10).

3. Results

3.1 Quantity of protein in fresh material

3.1.1 Genotypes with elevated Combinative Capacity

The data consisted of the absolute protein concentrations in micrograms per gram of fresh material for various genotypes, which were divided on the basis of their combined ability, high or low (Table 1). Hybrid 4 also presented greater protein amounts, with average values of 267.22 mg/mL, which varied from 245.18 to 292.01 mg/mL, respectively. This implies that Hybrid 4 can produce high amounts of protein; thus, in breeding programs, the crop would be ideal for increasing the nutritional value of the crop. The maternal form had a slightly lower average of 260.19 mg/mL, which also reflects the proper protein level but was still lower than that of Hybrid 4. The paternal form averages 260.65 mg/mL: the protein content in the paternal form deviates from the average while supporting the general theory of increased levels of protein in hybrids. Specifically, the protein content was within 260.65±60.15 mg/mL and 267.22±36.00 mg/mL for the HCC genotypes. The amount of protein in the HCC hybrid reached 292.01 μg/g, while that in its maternal form reached 282.28 mg/mL (Table 1).

3.2 Genotypes with low combinative capacity

Low Combinative Capacity Genotypes: Hybrid 6 has the lowest quantity of protein, with a mean quantity of 223.10 mg/mL. Compared with those of Hybrid 4, protein levels ranged from 215.22 mg/mL to 228.35 mg/mL, suggesting reduced protein manufacturing quality. Thus, the maternal form for Hybrid 6 averages 256.12 mg/mL, which is slightly greater than the average for Hybrid 6 but below the average for the high-combinative-capacity genotypes. As we already observed in the biochemical composition, the paternal form shows variability, although with a mean of 252.99 mg/mL, which is not close to the protein levels of the high-capacity hybrids. Low combinative capacity (LCC) genotypes presented relatively low protein levels; the scores obtained were 223.10 ± 16.45, 223.13 ± 15.53, 227.15 ± 15.83, and 252.99 ± 8.22 mg/mL, respectively. The LCC hybrid presented a maximum protein content of 223.10 mg/mL, and the maternal form presented a maximum protein content of 274.01 mg/mL (Table 1). From such a note stating $p > 0.05$, it may be concluded that the differences in the amount of proteins between these genotypes may not be statistically significant, indicating that a study with an increased sample size may be necessary to obtain more definitive results.

Table 1: Protein quantity in fresh material

Combinative capacity	Genotype	Protein quantity, mg/mL fresh material	
High	Hybrid 4	245,18	267,22±26,63
		292,01	
		264,46	
	Maternal form	252,78	260,19±7,91
		266,67	
		261,11	
	Paternal form	223,88	260,65±36,21
		275,80	
		282,28	
Low	Hybrid 6	215,22	223,10±6,94
		225,72	
		228,35	
	Maternal form	240,11	256,12±19,27
		254,24	
		274,01	
	Paternal form	258,97	252,99±17,00
		264,10	
		235,90	

Note: Values represent mean ± SD at n= 3, $p > 0.05$

3.3 Content of the water

The water content was relatively high, greater than 90%, for all the genotypes analyzed in the present study. This means that plants maintain many water reserves, which are essential for their metabolism, food movement, and overall well-being. Since water generally accumulates in plants under ideal conditions, an increase in water content could indicate good growth conditions.

3.4 Quantity of protein in dry material

3.4.1 Genotypes with elevated Combinative Capacity

The approximate average protein content in hybrid 4 was the highest among the genotypes; the values were 9.19, 10.95, and 9.92 mg/mL, respectively. The variation in measurements suggests that there could be changes in protein content even in relation to development at all stages or under all environmental conditions (Table 2). The average quantity of protein was 9.76 mg/mL dry material, and the protein content of the maternal form was slightly lower than that of the harvested HCC hybrid. This finding indicates that even though the maternal form is generative, over the years, hybrids have undergone improvement through breeding. The value of protein in the paternal form was also the lowest of all the HCC groups, which implies that this line might not be highly efficient in protein production. This may suggest the importance of choosing the right parent lines in the hope of achieving high protein yields in later breeding programmes.

3.5 Genotypes with low combinative capacity

This hybrid 6 has a lower protein level than the other HCC hybrids do, as would be expected from this kind of hybrid. The average Y value of the protein was 8.07 mg, 8.46 mg and 8.56 mg/mL, implying that although the HCC was consistent, some forms of protein were evident. Notably, the LCC material contained a greater protein content than did the hybrid material; the average protein content was 8.37 mg. This reasoning suggests that hybrids are usually better than pure types, but there are possibly

unique situations in which even a maternal line performs better than hybrids do. Like the HCC paternal form mentioned above, this line has the lowest protein level of all the corresponding forms. This finding conforms with the trend observed in HCC and underscores the need to choose better parental lines during breeding (Table 2). The variation in protein quantity between genotypes is not significant as confirmed by an ANOVA ($p > 0.05$). This means that, while trends can be established, these trends are not yet strong enough to state that one genotype is considerably superior to another in terms of protein yield.

Table 2: Water quantity and protein quantity in dry material

Combinative	Genotype	Water	Protein quantity, mg/mL dry material	
High	Hybrid 4	92	9,19	10,02±1,00
			10,95	
			9,92	
	Maternal form	92	9,48	9,76±0,30
			10,00	
			9,79	
	Paternal form	92	6,44	7,49±1,04
			7,93	
			8,12	
Low	Hybrid 6	92	8,07	8,37±0,29
			8,46	
			8,56	
	Maternal form	92	9,00	9,60±0,72
			9,53	
			10,28	
	Paternal form	90	7,77	7,08±0,51
			7,92	
			7,08	

Note: Values represent mean ± SD at $n= 3$, $p > 0.05$

A study of the data obtained revealed that the HCC hybrid contains the greatest amount of protein, with 292.01 mg/mL fresh mass and 10.95 mg/mL dry mass. Compared with the CWC hybrids, the LCC hybrids presented a much lower protein content across the genotypes used for analysis (Figure 1).

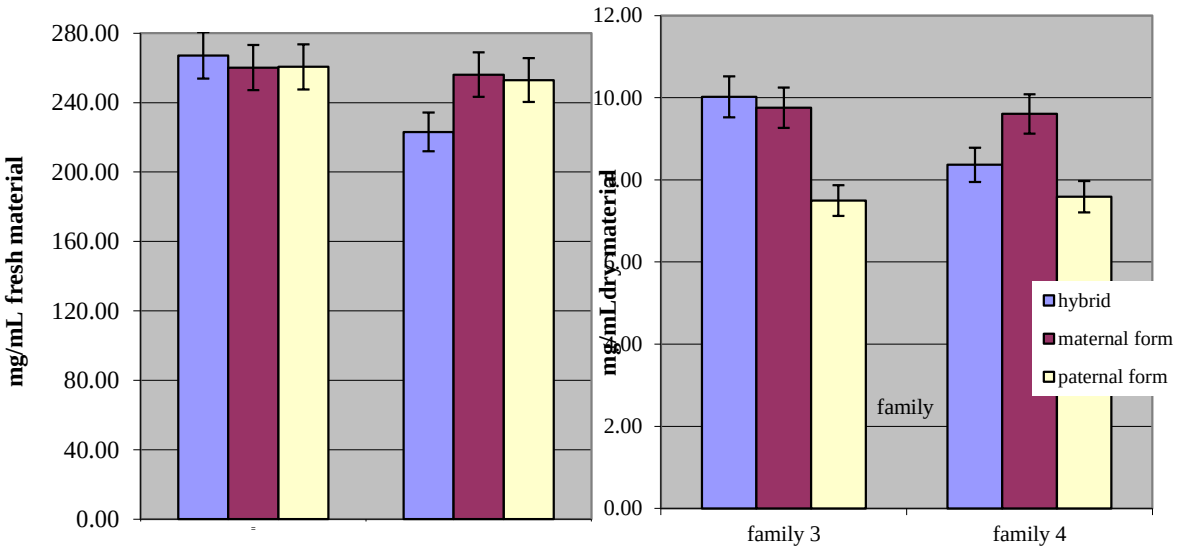

Figure 1: Protein quantity in fresh and dry material from different families.

4. Discussion

The global intake of plant protein has increased by 15% since the early 1960s, with health advantages associated with dietary fiber, vitamins, minerals, and phytochemicals from plant foods. The food industry is advancing plant-based proteins as substitutes for meat and animal-derived proteins to satisfy customer demand, which is influenced mostly by cultural, culinary, and religious preferences [11].

These findings reveal the presence of protein quantitative variations across the analyzed genotypes, together with a superior HCC showing pronounced protein levels compared with LCC hybrids. These findings raise the possibility of hybrid vigor, whereby hybrids excel, for instance, in protein synthesis over their parents. Ohindovschi, Cichna-Markl [12] reported that protein extraction from buffer solutions at various pH values indicated that optimal protein extraction occurs at neutral pH, whereas an increase in pH toward basic conditions results in a decrease in concentration. Suboptimal protein extraction was performed via acidic citrate buffer at pH 4.5.

High protein content is essential for the nutritional quality of plant-based foods, and these results are highly important for breeding programs aimed at the development of high protein varieties. Hybrid vigor is an important aspect of farming. The higher protein contents suggested that HCC hybrids might have potential for increased nutritional quality, as well as increased overall yield and better tolerance to biotic and abiotic stress. This characteristic is most imperative for various forms of food security, specifically in international food systems, where the need for healthy stock foods is increasing. Hence, when crossbred for hybrids that exhibit enhanced protein synthesis, breeders can help develop plants that can better feed the people in the population. In addition to quantity, the quality of plant proteins is crucial, contingent upon the composition and availability of key amino acids, amino acid imbalance, protein digestibility, and antinutritional factors, among others [13]. The increase in protein content of HCC hybrids not only increases nutritional value but also implies better resilience, rendering the hybrids good candidates for sustainable agriculture in unfavorable conditions. In addition, maximizing the amino acid composition and minimizing the antinutrient constituents of these hybrids would close the gap between the quality of plant and animal proteins and satisfy the global diet requirements better [14].

The results of protein advantage reported in HCC hybrids are in accordance with the new development of work in the area of crop genetic control of seed storage proteins. This has been followed recently by the identification of those transcription factors which induce the protein synthesis pathways in high-protein cultivars, hinting a potential markers of precision breeding programs [14]. This genetic

superiority can also have indirect advantages, since high protein concentrations have been found to be related to greater stress-adaptive processes in plants [15].

In this study, compared with LCC hybrids, HCC hybrids, especially Hybrid 4, presented greater protein synthesis ability. This finding only supports the notion of heterosis, where hybrids have greater performance than do the parental lines. Issues of protein disparity between maternal and paternal structures point to the role of parental selection in breeding performance. Unlike the total proteins, the protein levels of the maternal forms of both the HCC and LCC groups were comparable, indicating that both parental lines can contribute significantly to the protein content of the offspring.

Taken together, the findings of the present study provide promising supportive data for enhancing the protein capacity of HCC hybrids; however, further research is needed to elucidate the mechanisms of protein synthesis in different genotypes. To achieve a more profound understanding of the phenomena observed and search for new opportunities, the agricultural community needs to overcome the shortcomings of the current data streams. The findings elucidated the actuality of the ratio between various genotypes of plants with respect to protein content and identified the priorities of the respective hybrid selection, as well as the characteristics of the initial pairs in the framework of precisely oriented programs of plant breeding for obtaining improved nutritional value of grains. Future studies need to be carried out to investigate the synergistic role of phytochemicals in plant proteins in boosting the nutritional value. The developments in the realm of protein extraction have to respect the functional and bioactive properties with concern to efficiency. Explain the genetic and environmental antecedents of these traits.

5. Conclusions

The results revealed that high-combinative-capacity genotypes, especially Hybrid 4, produced more protein in both fresh (with an average of 245--292 mg/g fresh weight (FW)) and dry material (average of 9.19--10.95 mg/g dry weight (DW)) than did low-combinative-capacity hybrids, such as Hybrid 6 (with an average of only 215--228 mg/g FW in fresh material and 8.07--8.56 mg/g DW). In general, there was no significant variation in the protein content between the maternal and paternal forms ($p > 0.05$). Future studies should look at the genetic and environmental effects on protein yield and the replication of this study in a larger population to increase the protein content of these genotypes.

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