Peptides from anchovy waste for foliar spray application in microgreens

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Bioactive peptides have gained prominence in agricultural applications on account of their potential to increase the germination, productivity and quality of a wide range of horticultural and agronomic crops (Colla et al., 2015). Peptides derived from industrial discards are promising options for effectively upgrading waste to valuable commodities besides addressing environmental pollution issues.

Presently, consumers are questing for novel, fresh, healthy products having culinary attributes. The specialty crop growers and researchers are updating with the possibilities to tap niche products. A recent high demanded specialty crops are Microgreens, also called 'vegetable confetti'. They are defined as tender immature greens produced from the seeds of vegetables, herbs, or grains (Kyriacou et al., 2016). They have delicate textures, characteristic flavors and are loaded with health promoting nutrients than their mature counterparts. Microgreens are generally harvested upon appearance of the first pair of true leaves, generally within 7-21 days from seed germination depending on the species and growing conditions. Foliar spraying is generally adopted in microgreens as it offers specific advantages over soilapplied fertilizers, with the nutrients applied being directly taken-up by their target organs, providing specific and rapid response.

The present study focused on the optimized extraction of peptides from anchovy head waste collected from processing industry for its application as a crop stimulant in microgreens from green grams. Response Surface Methodology was employed for deriving optimized peptides using enzyme papain. The effect of hydrolytic conditions viz.,enzyme substrate ratio (1.0 - 5.0 %) and hydrolysis time (2 - 24 hours) under optimized temperature (60°C) and pH (6.5), on the protein recovery, and yield were considered. An enzyme-substrate ratio (E/S) of 5% and 23.48 hours of hydrolysis time with a desirability of 0.942 was found to be the optimized protocol for deriving peptides with maximum yield (8.26%) and protein recovery (73.63%).

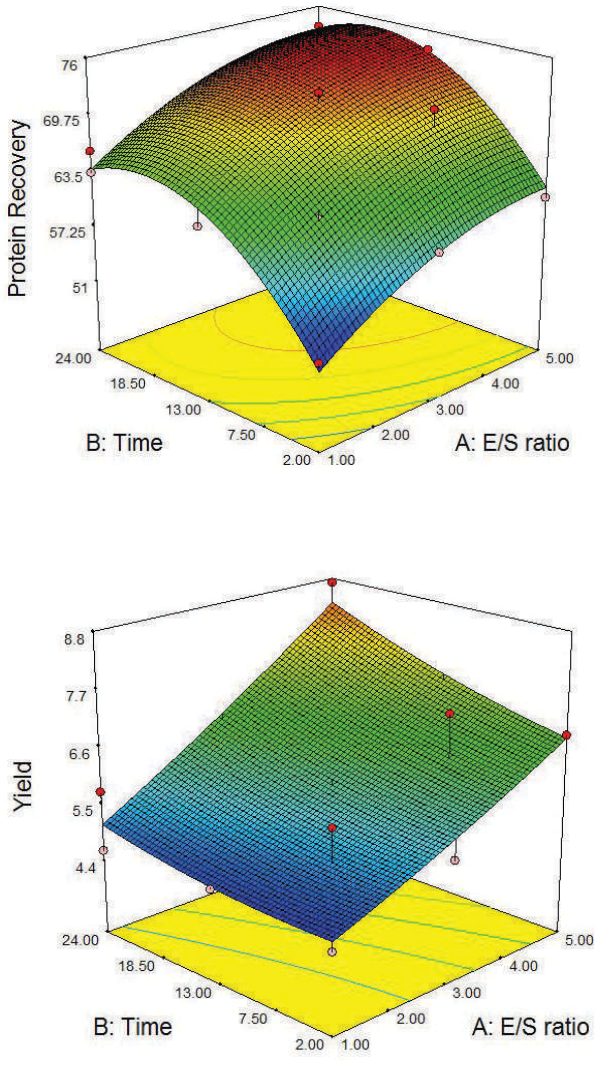

***Fig. 1** Variations in protein recovery (%) and yield a. in response to enzyme-substrate ratio and hydrolysis time*

The optimized peptides indicated a degree of hydrolysis (formol method) of 25.2% and proteolytic activity of 1.316 µmoles tyrosine liberated/mg protein. The peptides were applied as foliar spray periodically for a duration of one week in green gram microgreens, at concentrations of 0.5% ($\mathbf{P_{0.5}}$) and 1.0% ($\mathbf{P_{1.0}}$), keeping a control (C) for comparison. The germination rate as well as biomass was comparatively higher for microgreens subjected to 0.5% peptide foliar application ($\mathbf{P_{0.5}}$) (Table 1). Chlorophyll index also indicated superiority for $\mathbf{P_{0.5}}$ and it was observed that higher treatment of 1.0 % negatively impacted the chlorophyll content of the samples. Water retention in the microgreens were comparatively higher in treated samples. Overall acceptability of the sample indicated a higher sensorial preference for untreated samples (control) than treated ones. The acceptability of microgreens treated with 1.0 % ($\mathbf{P_{1.0}}$) peptides was affected, as the higher peptide concentration influenced the flavour, colour and taste of the microgreens.

The study suggested selection of optimized hydrolytic conditions for deriving specific hydrolysates for foliar spray formulation to incorporate into plants for encouraging their growth. A peptide application of 0.5% from anchovy waste was found optimum for microgreens from green gram. Further studies are recommended to completely explore the beneficial properties and mechanisms of these peptides as well as to determine different product formulations and application methods under a range of agro-ecological conditions.

Table 1. *Quality characteristics of microgreens from green gram*

Parameters	Control	$P_{0.5}$	$P_{1.0}$
Germination rate (%)	78.7	87.3	69.3
Biomass (g)			
Leaf	3.82	4.72	5.16
Root	2.61	3.37	3.14
Shoot	4.42	6.15	4.52
Chlorophyll a (µg/ml)	25.44	26.17	19.42
Chlorophyll b	9.15	10.17	6.11
Carotene	7.06	6.96	6.08
Water retention capacity (%)	63.64	68.38	70.9
Sensory attributes	9.0	8.0	5.0

References:

Colla, G., Nardi, S., Cardarelli, M., Ertani, A., Lucini, L., Canaguier, R., & Rouphael, Y. (2015). Protein hydrolysates as biostimulants in horticulture. Scientia Horticulturae, 196, 28-38.

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