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Bio Stimulant Activity of Protein Hydrolysate: Influence on Plant Growth and Yield

Research Article

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Abstract

Fertilization practices and growing techniques play a major role in influencing plant productivity. Fertilizers which have minimum impact on the environment are gaining popularity. In this regard, organic bio-stimulants such as protein hydrolysates, humates, etc., are becoming popular, as they reduce the use of chemical fertilizer by up to fifty percent. In this study, we focus on the influence of protein hydrolysate of animal origin on growth and productivity of different crop plants. We observed a positive effect of protein hydrolysate on plant growth and yield across crop species. Both soil and foliar treatments had a higher impact on the initial plant growth compared to control; however, soil application was more effective than foliar application. All parameters recorded including root and shoot length, leaf area, total chlorophyll content, photosynthetic rate, and yield were greater in treated plants. We report here that, protein hydrolysate when supplemented through soil, positively influences plant growth and metabolism; thereby, contributing towards higher crop yields. Hence, protein hydrolysates can be effectively used as organic fertilizers to improve productivity of the crops.

Key words: Bio-stimulant; Protein hydrolysate; Amino acids; Peptide; Growth; Yield

Introduction

Bio-stimulants are considered to be a group of substances known to have profound effect on the growth and development of plants. Zhang and Schmidt [1] first defined bio-stimulants as "materials that in minute quantities, promote plant growth". Many other definitions have evolved over time and each of them emphasize on different roles of these bio-stimulants. About 60 different substances have been grouped under the label of bio-stimulants [2]. A bibliographic analysis has been conducted by du Jardin [3] on eight categories of bio-stimulants such as humic substances, complex organic materials, beneficial chemical elements, inorganic salts including phosphite, sea weed extracts, chitin and chitosan derivatives, anti-transpirants, hormone containing products, amino acids, peptides, and other N-containing substances.

Bio-stimulants can be made available to plants through foliar sprays or through soil application. Foliar applied bio-stimulants were shown to reach mesophyll cells by absorption through cuticle and epidermal cells [4-6]. When supplied through soil, the absorption occurs through root epidermal cells and gets redistributed through xylem [7]. These formulations decrease the need of chemical fertilizers and have the capacity to satisfy the nutritional requirements of plants and also further result in higher yield [8].

Formulations containing amino acids, peptides, polyamines, and betaines were found to have plant growth stimulation activity. These compounds have come into scrutiny for their stimulatory effect which is distinct from the effect of additional nitrogen source [9,10]. Amino acids can easily be absorbed by plant roots and through foliage [11,12]. They have the capacity to enhance nitrogen use efficiency of the plants and further stimulate photosynthesis and plant growth [13]. Their effects on carbon and nitrogen metabolism, and plant primary and secondary metabolism have been reported by Schiavon et al. [14] and Maini [15]. Hormone-like-activity including Auxinlike-activity and Gibberellin-like-activity has also been attributed to amino acids and peptides [14].

Protein hydrolysates containing amino acids and peptides can be of either plant or animal origin. They are manufactured by chemical or enzymatic hydrolysis of plant residues or animal connective tissues [10,16-19]. The primary aim of this paper is to describe the physiological effect of amino acids and peptides, collectively called protein hydrolysate, on plant growth processes and crop yield.

Materials and Methods

Experiments were conducted to evaluate the role of an animal based protein hydrolysate, on early seedling growth in crop species such as Paddy, Finger millet, Cowpea and in Radish, where yield was also recorded. Protein hydrolysate used in this study was supplied by ISAGRO Asia Agro Chemicals Pvt. Ltd, Mumbai, India; with the trade name of Siapton 10L. The chemical composition of the concentrated formulation is given by Parrado et al. [13] and Mladenova et al. [20].

Seedling Growth in Paddy

CleriGel 1% (HiMedia Laboratories Pvt. Ltd, Mumbai, India) containing 0.1%, 0.2% and 0.5% (v/v) protein hydrolysate formulation was prepared and poured into test tubes. Rice seeds (CV: MTU 1010) were germinated on sterile water. Germinated seeds having two mm radical length were placed on CleriGel containing different concentrations of protein hydrolysate. The test tubes were incubated in dark for seven days. Root and shoot length were measured after seven days.

Seedling Growth in Finger millet

Seeds of Finger millet genotype GPU 28 were sown in nursery trays filled with red soil and Farm Yard Manure (FYM) in the ratio of 1:1. Nine days after sowing the plants were thinned to five seedlings per cup. Protein hydrolysate was supplied at two concentrations, 0.25% and 0.5%, either as a foliar treatment or via soil application. Foliar application was done by smearing protein hydrolysate solution to both upper and lower surfaces of the leaf using cotton swabs. For soil application, 5ml of the solution was applied per 100g of dry soil. Seven days after application root and shoot lengths were measured.

Seedling growth and photosynthesis in Cowpea

Seeds of Cowpea genotype C-152 were germinated on sterile water. Germinated seeds with two mm radical length were planted in pots containing 500g of soil and FYM mix. Nine days after germination, five plants were maintained in each pot and 20 ml of protein hydrolysate was supplied to each pot. Four concentrations of protein hydrolysate, viz 0.2%, 0.4%, 0.6%, and 0.8%, were used for the study. Influence of protein hydrolysate application on root and shoot growth, leaf area and total chlorophyll was studied seven days after soil application.

Cowpea plants raised in containers with soil and FYM mixture 1:1 (w/w) were treated with protein hydrolysate by applying 20 ml solution per pot. Seven days after soil application, observations on photosynthetic rate and stomatal conductance were recorded using a portable photosynthetic system (LICOR LI-6400).

Productivity in Radish

Radish seedlings were raised in pots containing 13 kg of soil and FYM mixture. Two plants were maintained per pot. Plants were irrigated every day to bring the water status to field capacity. When plants were 30 days old, 150 ml of 0.2%, 0.4%, and 0.6% protein hydrolysate was applied to the soil. Plants were harvested on 60th day of planting and fresh weight of Radish was recorded.

Results

Seedling growth in Paddy and Finger Millet

Paddy: Seedlings grown on CleriGel-protein hydrolysate medium showed increased root and shoot length. Presence of protein hydrolysate in the rooting medium resulted in significant increase in root and shoot length, both increasing with increasing concentration of protein hydrolysate. The effect observed was greater in root length, which showed an increase of 1.3 times over control. (Figures 1, 5a and 5b)

Finger millet: The root and shoot growth increased with increasing concentration of protein hydrolysate up to 0.5%. The maximum increase in shoot and root length was seen when protein hydrolysate was applied to soil. The increase in root and shoot length was to the extent of 126% and 79% at 0.5% soil application; and 30%

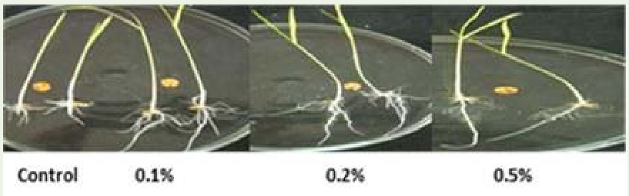

Figure 1: Root and Shoot growth of nine day old Rice seedlings grown in test tubes over CleriGel media incorporated with various concentrations Siapton 10L.

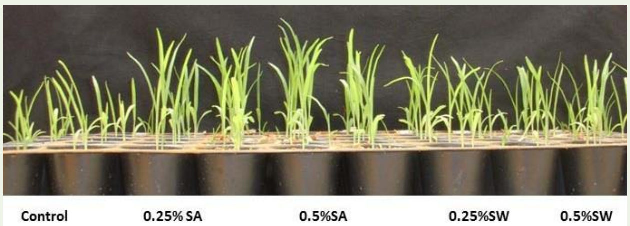

Figure 2: Response of nine day old Finger millet seedling to different concentrations of Siapton 10L (SA: Soil Application, SW: Foliar Application).

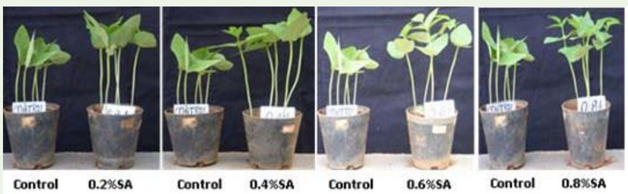

Figure 3: Response of Cowpea seedlings supplemented with 0.2% 0.4%, 0.6% and 0.8% Siapton 10L through soil application (SA). Observations were recorded seven days after treatment to nine day old seedlings.

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and 38% at 0.5% foliar application respectively when compared to control. (Figures 2, 6a and 6b)

Growth parameters in Cowpea

Protein hydrolysate treatment was effective in increasing root growth, shoot growth and leaf area in Cowpea. Root and shoot length of seedlings increased with increase in protein hydrolysate concentration up to 0.6% (Figure 7a & 7b). Leaf area of Cowpea seedlings increased linearly up to 0.4% protein hydrolysate concentration. (Figure 7c). Application of protein hydrolysate also resulted in early development of trifoliate leaves in the seedlings. (Figure 3)

The chlorophyll pigment concentration was determined in the cotyledonary leaves of Cowpea seedlings. The results show that chlorophyll content increased with increase in concentration of protein hydrolysate up to 0.8%. (Figure 7d).

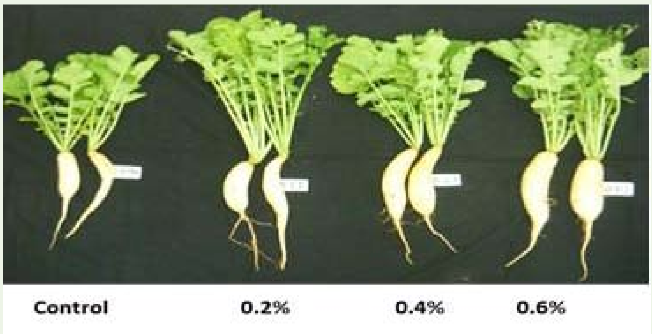

Figure 4: Yield of Radish plants treated with 0.2%, 0.4%, and 0.6% Siapton 10L thirty days after sowing.

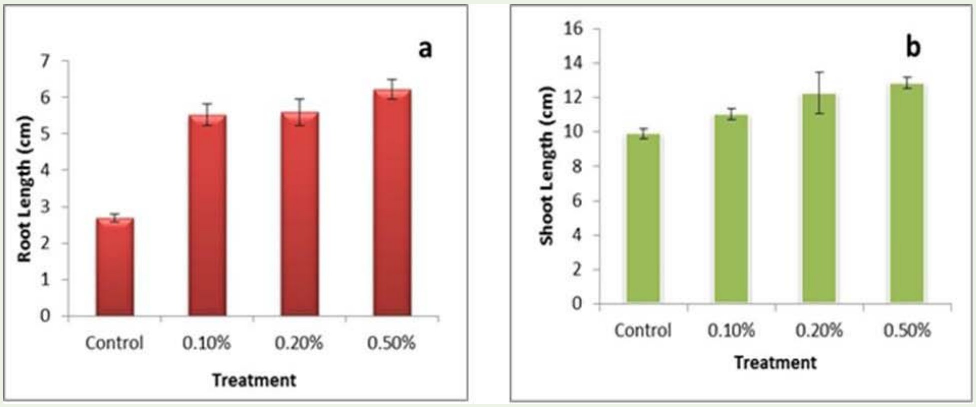

Figure 5: Root (a) and Shoot (b) length of nine day old Rice seedlings grown on 1% CleriGel incorporated with different concentrations of Siapton 10L.

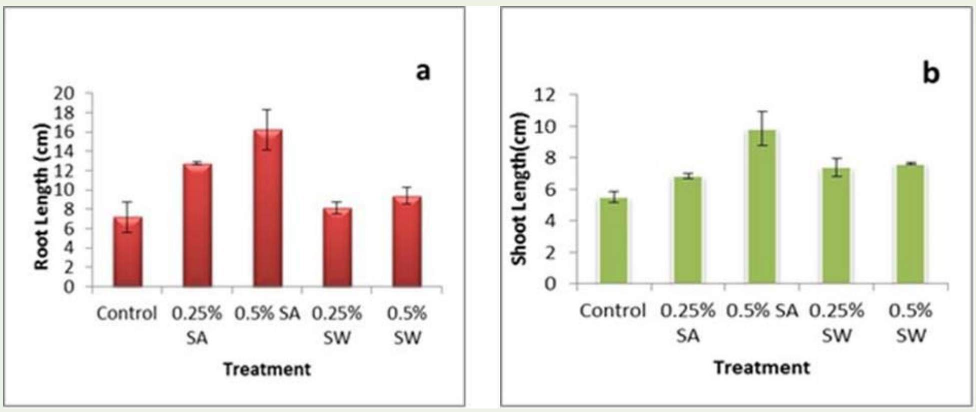

Figure 6: Root (a) and Shoot (b) length of Finger millet seedlings treated with Siapton 10L. Observations were recorded seven days after treatment to nine day old seedlings. (SA: Soil Application, SW: Foliar Application).

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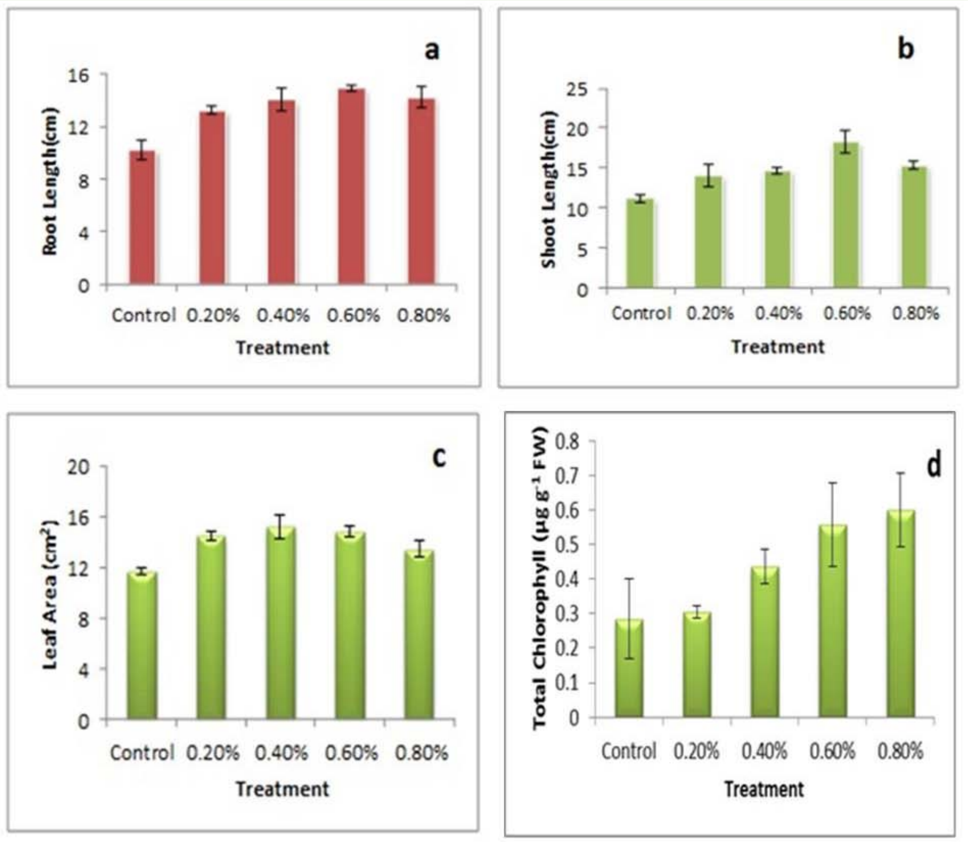

Figure 7: Response of Cowpea seedlings to Siapton 10L: Nine day old Cowpea seedlings were treated with 0.2%, 0.4%, 0.6%, and 0.8% Siapton 10L through soil application.Root length (a), shoot length (b), leaf area (c) and total chlorophyll content (d) were recorded seven days after treatment.

Photosynthetic Rate in Cowpea

Seedlings treated with protein hydrolysate showed significant increase in photosynthetic rate as well as stomatal conductance. Soil application of protein hydrolysate increased photosynthetic rate by 35.1% over control and stomatal conductance increased by 21.7% over control. (Figure 8a and 8b)

Productivity in Radish

Application of protein hydrolysate to the soil resulted in significant increase in fresh weight of Radish. The increase was to an extent of 26%, 46%, and 60% with 0.2%, 0.4%, and 0.6% protein hydrolysate treatments respectively when compared to control. (Figure 4 and 9)

Discussion

Protein hydrolysates containing amino acids and peptides are known to have bio-stimulant like activity. Karnoc [2] has listed amino acids and peptides to be among the constituents of bio-stimulant formulations. Peptides which are absorbed by plants are hydrolysed by various peptidases to the constituent amino acids which can be used as an alternative source of nitrogen and carbon or also used for protein synthesis [21,22]. Studies have indicated that amino acids and proteins may have a role in nitrogen acquisition [23-25]. Amino acids increase the plant efficiency by instigation of metabolism and metabolic processes [26].

The influence of protein hydrolysate on seedling growth was studied in crops such as Paddy, Finger millet, Cowpea and Radish. The results clearly show that protein hydrolysate application stimulates root and shoot growth in all the crops tested. The effect on root growth was substantially greater than that on the shoot growth. Studies conducted with Cowpea showed that soil application improves photosynthetic surface area to the extent of 30%. Increase in root length would influence higher uptake of water as well as nutrients from the soil leading to higher growth rate through higher photosynthetic surface area. Observations on gas exchange parameters also indicated a significant increase in photosynthetic rate as well as stomatal conductance. These observations clearly prove that increase in total canopy photosynthesis by virtue of increased leaf area, increased carbon assimilation rate per unit leaf area. Increased availability of photosynthates resulted in increased plant height and root length. As an extrapolation of work done in Cowpea, further experiments conducted with Radish in containers proved that 0.6% protein hydrolysate soil application one month after planting improved productivity by 59.56%. These results substantiate the improved photosynthetic activity and nutrient absorption by the roots which is reflected in terms of stimulated growth and increase in harvestable yield of Radish.

Since the protein hydrolysate used is known to contain free amino acids and low molecular weight short peptides, we assume that these

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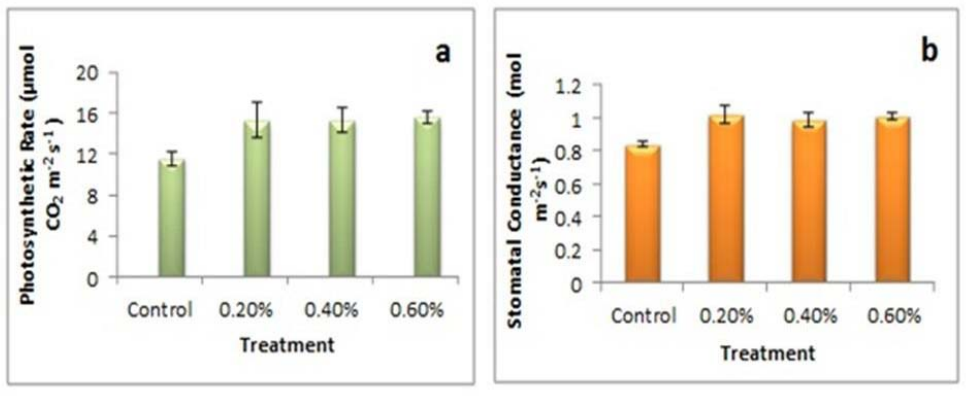

Figure 8: Photosynthetic rate (a) and stomatal conductance (b) of Cowpea seedlings in response to soil application of Siapton 10L.

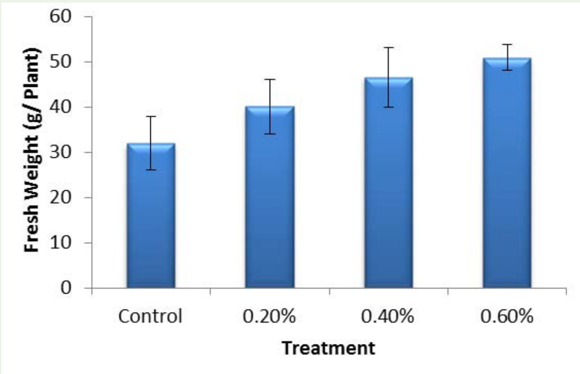

Figure 9: Radish yield in response to 0.2%, 0.4% and 0.6% Siapton 10L soil application.

amino acids serve as immediate organic nitrogen source for synthesis of other macromolecules as previously reported [27-30]. In addition to amino acids, short peptides serve as a source of nitrogen nutrition which immediately advances plant metabolism.

Our experiments demonstrate the importance of application of protein hydrolysate in improving plant growth. We conclude that protein hydrolysate formulations can be used as biostimulants or bio-fertilizers to achieve higher yields in crop plants. The protein hydrolysate used in this study was, Siapton 10L, a commercial formulation obtained from hydrolysis of skin and hair wastes [13]. It is known to contain various amino acids and short chain peptides [20].

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