

# A novel source of biofertilizer from feather biomass for banana cultivation

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**Abstract** Feather waste is a promising protein biomass available as by-product from poultry processing was found to be rich in peptides, amino acids, and minerals like nitrogen, phosphorus, potassium, calcium, magnesium, iron, manganese, zinc, and copper. Soil and foliar application of these products, besides representing a sustainable solution to the problem of feather disposal, may also represent an effective strategy to tackle the environmental effluence. As a consequence, they were also found to be very attractive in elevating the protein, amino acids, reducing sugar, total chlorophyll, and proline content of plants. On the other side, fertilizing effect enhanced the antioxidant potential of banana fruit which was assessed using 2, 2-diphenyl-1-picrylhydrazyl, ferric reducing/antioxidant power, and *N*, *N*-dimethyl-*p*-phenylenediamine. This was associated with considerably higher antioxidant contents like total phenolics and flavonoids. Therefore, the application of this organic amendment could promote and improve the agro-ecosystem, human health; soil biological activities, and at the same time enhance the production of plant or products rich in bioactive substances.

**Keywords** Amino acids · Antioxidants · Banana · Bioenhancer · *Chryseobacterium* sp. RBT · Feather waste

## Introduction

The feathers are the complex structural protein rich in disulphide bonds, hydrogen bonding, and hydrophobic interaction making it resistant to degradation by most plant, animal, and microbes (Coward-Kelly et al. 2006). Poultry feather

contributes about 10 % to the broiler's live body weight, while dried feather contains 85–99 % proteins (Papadopoulos 1985). Agrahari and Wadhwa (2010) estimated that about 24 billion chickens are killed annually worldwide through which 8.5 billion tons of poultry feather are produced, among which India alone contributes about 350 million tons annually. Currently, the feather waste is treated by using chemical (acid and alkali) and physical (cooking or burning) treatments which are expensive, hazardous, and also leads to the contamination of air, water, and soil (Gupta and Ramnani 2006). The microbial bioremediation offers great advantages over conventional methods. Much of the previous work has been focused only on degradation, enzyme purification, and analysis of the degradation products, but little is known about its effects and further applications. As feathers are rich source of organic nitrogen, they suggest an opportunity for the development of new nitrogen-rich organic amendments that will serve the dual purpose of improving plant growth and stimulating microbial activity in the soil, beneficial to human health and environment.

The feather degrading bacterium *Chryseobacterium* sp. RBT used in the present study was capable to hydrolyze the native chicken feathers within 30 h. According to our previous observation feather hydrolysate was rich source of amino acids like leucine, valine, methionine, glycine, cysteine, serine, phenylalanine, tryptophan, lysine, and tyrosine (Gurav and Jadhav 2012). Amino acids are well known as biostimulants as it promotes the vegetative growth, nutrient uptake in plants, abiotic and biotic stresses tolerance resulting in increased crop yield (Vernieri et al. 2006).

## Material and methods

### Chemicals and poultry feathers

The solvents, chemical, and reagents used in this study were of analytical grade obtained from local suppliers. Chicken

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feathers were collected as a waste by-product generated through poultry processing unit in Kolhapur, India. Raw feathers were washed under tap water, sun dried, and stored.

#### Biodegradation of native chicken feathers using *Chryseobacterium* sp. RBT

The feather-degrading bacterium previously isolated and identified as *Chryseobacterium* sp. RBT (accession number GU481093) was used in this study. The feather degradation was carried out in flask containing feather and basal salt medium (in grams per liter)  $\text{KH}_2\text{PO}_4$  (4),  $\text{Na}_2\text{HPO}_4$  (6),  $\text{NaCl}$  (5),  $\text{MgSO}_4$  (0.1), and native chicken feathers (10). The 2-ml seed culture of *Chryseobacterium* sp. RBT (1.12 O.D. at 660 nm) was transferred to feather basal salt medium and agitated on rotary shaker (140 rpm) at 37 °C for 30 h (Gurav and Jadhav 2012). The degraded broth was pasteurized and used for further studies.

#### Analysis of the feather degradation end products

The amount of total soluble protein was determined by Lowry method (Lowry et al. 1951) and amino acids using ninhydrin (Moore and Stein 1957). The total nitrogen (N) (Kjeldahl method), phosphorus (P), and potassium (K) (flame photometry) were determined. Also, the presence of metal-like copper (Cu), iron (Fe), calcium (Ca), manganese (Mn), magnesium (Mg), and zinc (Zn) were examined by means of atomic absorption spectroscopy (Perkin-Elmer analyst, USA), (APHA 1998).

#### Degraded feather waste as biofertilizer

##### *Plantation and dose application*

The tissue cultured banana plantlets (*Musa* sp. known as Grand niene or G9) were generous gift from Callus Biotech Pvt. Ltd, Kolhapur, India. Plantlets obtained were of uniform height and age. The 30×30 m land was selected near Akluj, India, which was divided into three sets each having 10×10 m area. Plantation was done on 5 September 2011 in the selected plot with spacing of 2×2 m between two plants and rows consisting 36 plantlets for each treatment. Each set was amended with FDP as 20 % root dose (RD) and 5 % shoot dose (SD) through fertigation and foliar spray at the interval of 15 days. The control plants without treatment were maintained. All the plants were irrigated regularly through drip irrigation and the experiment was carried out in triplicate. Regular practices required for the cultivation like weeding and earthing up were done routinely.

#### *Chemical analysis of banana plant and fruit*

**Chlorophyll estimation** The fresh leaves of treated and control plants were collected randomly after 4 months of plantation; washed thoroughly and blotted. Then, 1 g of leaf tissue was crushed in acetone (80 % v/v) and the final volume was adjusted to 20 ml and stored overnight under refrigeration. This extract was then filtered through glass wool followed by centrifugation. The absorbance of the supernatant was recorded at 663 and 645 nm using spectrophotometer (Shimadzu UV-1800, Japan) and the total chlorophyll content was determined as the mean of all readings (Arnon 1949).

##### *Quantification of free protein, amino acids and reducing sugars*

The fresh fruit pulp/vegetative tissue (1 g) was crushed in 100 mM phosphate buffer (pH 7; for protein) and in 80 % ethanol (for amino acids); this extracts were centrifuged and the aliquots were pooled for the estimation of free protein (Lowry et al. 1951) and amino acids (Moore and Stein 1957). The fruit pulp (1 g) was homogenized in 90 % ethanol and then centrifuged at 10,000 rpm. The residue was again extracted and subjected for evaporation and dissolved in distilled water and reducing sugar was measured by glucose standard curve (Miller 1959).

##### *Estimation of free proline*

The assessment of the free proline content in vegetative tissue was determined using method of Bates et al. (1973). Briefly, fresh tissue (1 g) was homogenized in 3 % (w/v) aqueous 5-sulfosalicylic acid, centrifuged, and the supernatant was reacted with equal amount of acid ninhydrin and glacial acetic acid. After 1 h of incubation, the reaction mixture was extracted with toluene. The chromophore containing toluene was aspirated and optical density was measured at 520 nm (UV-1800, Shimadzu, Japan) using toluene as a blank. The amount of proline was determined using standard L-proline and expressed as milligram of proline per gram fresh weight (FW).

##### *Preparation of extract*

The peeled pulp/vegetative tissue were crushed in food processor to produce uniform slurries. In extraction process, about 2 g of slurry was homogenized in different solvents (methanol, ethanol, and water) in the ratio 1:10. Extraction was carried out on orbital shaker for 24 h followed by centrifugation at 15,000 rpm and the supernatant was used for the analysis of antioxidants, phenolics, and flavonoids.

##### *Analysis of total phenolics*

The total phenolics of treated and control plants were determined using Folin–Ciocalteu's reagent (Singleton and Rossi 1965). An aliquot (500  $\mu\text{l}$ ) of methanolic extract (ME), ethanolic extract (EE), and

aqueous extract (AE) of vegetative tissue/fruit was added to 500  $\mu\text{l}$  of distilled water and 1,800  $\mu\text{l}$  of Folin–Ciocalteu reagent, followed by the addition of 1200  $\mu\text{l}$  15 % (w/v) sodium carbonate. After 90 min incubation in the dark, the absorbance was recorded at 765 nm against blank. Total phenolic content was expressed as milligrams of gallic acid equivalents (GAE) per gram of fresh weight using calibration curve.

**Determination of total flavonoids** A total flavonoid content of ME, EE, and AE of the fruit and vegetative tissue was measured using colorimetric method (Chang et al. 2002). In brief, 500  $\mu\text{l}$  of plant extract was mixed with distilled water (2,000  $\mu\text{l}$ ) followed by 10 % (w/v) aluminum chloride (100  $\mu\text{l}$ ), 1 M potassium acetate (100  $\mu\text{l}$ ), and distilled water (2,800  $\mu\text{l}$ ). After 30 min incubation, the absorbance was recorded at 415 nm (Shimadzu UV-1800, Japan). The total flavonoids content was quantified according to the standard curve and was reported as milligrams of rutin equivalents per gram fresh weight.

#### Evaluation of antioxidant activity

**DPPH radical scavenging activity** The 2, 2-diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging capacity was determined according to Thaipong et al. (2006). In its radical form, DPPH absorbs at 517 nm, but upon reduction with an antioxidant, its absorption decreases due to the formation of its nonradical form, DPPH-H. The ME, EE, and AE (200  $\mu\text{l}$ ) of the treated and control fruits was reacted with DPPH solution (2.8 ml) in the final reaction volume of 3 ml. The decrease in absorbance of resulting solution was then measured at 517 nm using spectrophotometer (UV 1800, Shimadzu, Japan) against methanol. The results were expressed as percent radical scavenging activity (%RSA) and were calculated using the following equation.

$$\% \text{RSA} = (A_{\text{control}} - A_{\text{sample}}/A_{\text{control}}) \times 100$$

Where  $A$ =absorbance at 517 nm.

**FRAP assay** The ferric reducing/antioxidant power (FRAP) assay was performed as described by Benzie and Strain (1996). Briefly, the working FRAP reagent was prepared by mixing 300 mM acetate buffer (pH3.6), 10 mM 2, 4, 6-tripyridyl-s-triazine in 40 mM HCl and 20 mM  $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$  in 10:1:1 ratio prior to use. The ME, EE, and AE (200  $\mu\text{l}$ ) of fruit were react with 3 ml of the FRAP reagent. The final reaction volume mixture was made up to 4 ml. After incubation (30 min), the absorbance was measured at 593 nm using (UV 1800, Shimadzu,

Japan). The results were recorded as absorbance where higher absorbance indicates higher reducing power.

**DMPD scavenging capacity** The *N, N*-dimethyl-*p*-phenylendiamine (DMPD) assay was performed according to the method described by Fogliano et al. (1999) with some modifications. Briefly, 100 mM DMPD solution was prepared by dissolving 209 mg DMPD in 10 ml distilled water. Of this solution, 1 ml was transferred to 100 ml 0.1 M acetate buffer (pH5.2) followed by addition of 0.2 ml of a 0.05 M ferric chloride solution which resulted in the colored radical cation DMPD<sup>+</sup>. The 200  $\mu\text{l}$  of ME, EE, and AE were added to 2 ml of DMPD<sup>+</sup> solution and the total volume was adjusted to 3 ml. After 10 min, the absorbance was recorded at 505 nm by UV-visible spectrophotometer using buffer as blank. The DMPD<sup>+</sup> scavenging capacity was calculated by following equation (Jagtap et al. 2011).

$$\text{DMPD}^+ \text{ scavenging capacity}(\%) = (A_0 - A_1/A_0) \times 100$$

Where,  $A_0$  is the absorbance of the initial concentration of DMPD and  $A_1$  is the absorbance of the remaining concentration of DMPD.

#### Soil and plant analysis

The total N (Kjeldahl method), P and K (flame photometry) content of the soil was determined. Similarly, soil and plant samples were subjected to wet digested in nitric acid/perchloric acid (3:1, v/v). After dilution, samples were filtered and analyzed for the presence of Cu, Fe, Ca, Mn, Mg, and Zn using atomic absorption spectrophotometer (Perkin-Elmer analyst, USA). All the analytical techniques were performed according to APHA (1998).

## Results and discussion

#### Flowering, harvesting, and yield

The flowering in the RD-treated plant was observed after 249.6 days, SD of 256.2 days, and control of 274.3 days of plantation. The harvesting of bunch of plants treated as RD and SD was done on 27 August 2012. Whereas, the untreated plants harvesting was delayed by 28.3 days. The data regarding hands per bunch, fingers per bunch, and bunch weight of the treated and control plants has shown in Table 1.

#### Evaluation of the banana plant and fruit

**Photosynthetic pigment** The chlorophylls are the most essential pigments of photosynthesis. The banana plants treated with the FDP as RD showed higher total chlorophyll

**Table 1** Number of hands per bunch, fingers per hand, and bunch weight of treated and control plants. Total chlorophyll content of the leaves; proline in vegetative tissue, and reducing sugars content of the ripe banana fruits. Data shown as mean±SEM,  $n=10$ 

	Hands/bunch	Fingers/hand	Bunch weight (kg)	Total chlorophyll (mg g <sup>-1</sup> FW)	Proline (mg g <sup>-1</sup> FW)	Reducing sugars (mg g <sup>-1</sup> FW)
Root dose (20 %)	10.09	24.89	27.44	1.435±0.08	0.111±0.003	6.752±0.12
Shoot dose (5 %)	10.04	22.53	26.19	1.213±0.01	0.123±0.002	6.261±0.09
Control	9.16	19.14	23.12	0.896±0.05	0.078±0.002	5.812±0.11

Plantation was done on 5 September 2011 and harvested on 27 August 2012. The harvesting of untreated plants was delayed by 28.3 days.

FW fresh weight

content (1.435±0.08 mg g<sup>-1</sup>FW) followed by SD (1.213±0.01 mg g<sup>-1</sup>FW). The control plants showed less total chlorophyll content (0.896±0.05 mg g<sup>-1</sup>FW). The data is shown in Table 1.

According to Neals (1956), formation of chlorophyll molecule is dependent on nutrients like N, Mg, S, Ca, Fe, Mn, as well as Zn available to the plants. These metals were present in good amount in feather hydrolysate were applied to the plants as RD and SD may have enhanced the chlorophyll content. Previously, it is well stated that higher chlorophyll content is the indication of physiologically active healthy plants and those which are deprived in their nutritional condition showed poor growth and chlorosis (Zarco-Tejada et al. 2004).

**Estimation of free proline** The results revealed highest proline accumulation in banana plants supplemented as SD 0.123±0.002 mg g<sup>-1</sup> FW, whereas the RD showed 0.111±0.003 mg g<sup>-1</sup> FW and control 0.078±0.002 mg g<sup>-1</sup> FW proline content. The results are represented in Table 1.

Previously, it has been reported that under stress conditions, many plant species accumulate proline as an adaptive response to the adverse conditions (Verbruggen and Hermans 2008). However, there are also some circumstantial evidences suggesting that proline accumulation may occur in physiological nonstressed conditions for the development purposes. The proline accumulation may play an important role in flowering and developmental both as a metabolite and as a signal molecule or in the increase demand of protein synthesis (Mattioli et al. 2009).

**Evaluation of the reducing sugar** The reducing sugar contents of the treated and control fruit was evaluated. The substantial increase in reducing sugar content of ripe banana fruits was higher in RD-treated plants (6.752±0.12 mg g<sup>-1</sup> FW), followed by SD (6.261±0.09 mg g<sup>-1</sup>FW) and control (5.812±0.11 mg g<sup>-1</sup>FW). All the results are quoted in Table 1.

Banana fruit is a cost-effective energy source and used by endurance athletes because of the perception that they are a good source of energy. The elevated sugar content may be

due to higher concentrations of the photosynthetic pigments which favored the synthesis of carbohydrates on application with FDP. During the ripening process, this starch were converted into sugars through enzymatic breakdown process which showed accumulation of reducing sugars as reported by Beaudry et al. (1989).

**Peptides and amino acid as bioenhancer** The feathers are rich source of protein (90 %) which was solublized into peptides and amino acids using microbial keratinase. Banana plants, supplemented with this mixture showed growth promoting activity by increasing protein and amino acid content of the plant. It was found that RD was most effective for elevating the protein content of ripe fruit (16.13±0.63 mg g<sup>-1</sup>FW) and vegetative part (6.48±0.19 mg g<sup>-1</sup>FW). Furthermore, RD was also effective in enhancing the level of amino acids in fruit (2.96±0.11 mg g<sup>-1</sup>FW) and vegetative tissue (1.31±0.02 mg g<sup>-1</sup>FW). Similarly, the SD was also effective for enhancing the protein and amino acid content compared to the control plants. The result of the protein and amino acid contents are quoted in Table 2.

Currently, most of the hydrolyzed protein fertilizers available in the market are obtained by strong chemical hydrolytic process resulting in high contents of unwanted D-amino acids (Cavani et al. 2003). The biodegraded keratin waste contained amino acids which may have chelating action on micronutrients (Vesela and Friedrich 2009). According to Cao et al. (2012), peptide and amino acid mixture is the ideal fertilizer for growth and development of plants. Similarly, Endres and Mercier (2003) have also stated that plants can absorb and assimilate amino acids not only from the soil but also directly through leaves so we have also tried FDP as a foliar spray which showed interesting results. The synthetic growth regulators used in agriculture system have detrimental effects on living beings and environment (Abou Dahab and Abd El-Aziz 2006). Thus, it was clear that good characterisation of such organic residues is obliging to enhance the growth, particularly in terms of yield and nutritional quality of fruits is important in being able to understand and predict the value of FDP as biofertilizer.



**Table 2** Total phenolic and total flavonoid content of the banana fruit and vegetative tissue in various solvent extracts. The protein, amino acid and sugar content of ripe banana fruit and vegetative tissue. Data shown as mean±SEM,  $n=10$ 

		Total phenolics (mg GAE g <sup>-1</sup> FW)			Total flavonoids (mg RE g <sup>-1</sup> FW)			Protein (mg g <sup>-1</sup> FW)	Amino acid (mg g <sup>-1</sup> FW)
		AE	ME	EE	AE	ME	EE		
Ripe banana fruit	RD	0.786±0.006	1.072±0.009	0.919±0.007	0.388±0.003	0.612±0.005	0.519±0.004	16.13±0.63	2.96±0.11
	SD	0.705±0.006	0.982±0.004	0.821±0.005	0.305±0.001	0.535±0.004	0.463±0.003	15.60±0.30	2.44±0.09
	C	0.653±0.004	0.823±0.007	0.756±0.006	0.261±0.001	0.472±0.002	0.413±0.003	15.11±0.82	2.03±0.13
Vegetative tissue	RD	2.040±0.030	2.343±0.050	2.166±0.040	1.184±0.003	1.910±0.020	1.419±0.010	6.480±0.19	1.31±0.02
	SD	1.872±0.008	2.130±0.020	1.888±0.080	1.031±0.007	1.593±0.006	1.256±0.009	5.940±0.18	1.11±0.02
	C	1.341±0.060	1.687±0.009	1.478±0.060	0.712±0.009	1.222±0.010	0.891±0.006	4.020±0.10	0.89±0.01

AE aqueous extract, ME methanolic extract, EE ethanolic extract, GAE gallic acid equivalent, RE rutin equivalent, FW fresh weight, RD root dose (20 %), SD shoot dose (5 %), C control plant

**Total phenolics and flavonoids content** Polyphenolics are broadly distributed in the plant kingdom and considered as the most potent natural antioxidants (Karaman et al. 2010). The banana plants supplemented with FDP showed increase in their content of total phenolics and flavonoids. Root dose was most effective in enhancing the phenolic contents in ME of fruit (1.072±0.009 mgGAEg<sup>-1</sup>FW) and vegetative part (2.343±0.050 mgGAEg<sup>-1</sup>FW). Similar trend was observed in total flavonoids for root dose; the ME of treated plants exhibited maximum readings, i.e., (0.612±0.005 mg REg<sup>-1</sup>FW) and (1.910±0.020 mgREg<sup>-1</sup>FW) in fruit and vegetative extract, respectively. The data regarding total phenolic and flavonoid contents of ME, EE, and AE of the treated and control plants has been noted in Table 2.

In plants, polyphenols are generally involved in defense mechanism against biotic and abiotic stress as well as contributing to plant colors. They are ubiquitous in all plant organs and therefore a vital part of the human diet including fruits which are considered as a supplementary dietary source of various antioxidant phytochemicals. The antioxidant activities of polyphenols may be related to their redox properties, which allow them to act as a reducing agents or hydrogen/electron donors, scavenge free radicals, and terminate radical chain reactions (Le et al. 2007). Antioxidants promotes possible beneficial roles in human health, such as reducing the risk of cancer, cardiovascular disease, may prevent or repair cell damage caused by reactive oxygen species (Tepe et al. 2006). Currently, the natural polyphenol antioxidant from plant sources has gained enormous attention as their protective effects against different degenerative diseases and also due to toxicity and carcinogenicity of the synthetic antioxidants. Previously it has been also reported that increase in the polyphenols in different crops amended with organic wastes, suggested that organic waste may cause change that favors the accumulation of antioxidants (McGrath et al. 1994). Likewise, the plants treated with the FDP enhanced the contents of the polyphenolic antioxidants. Ren et

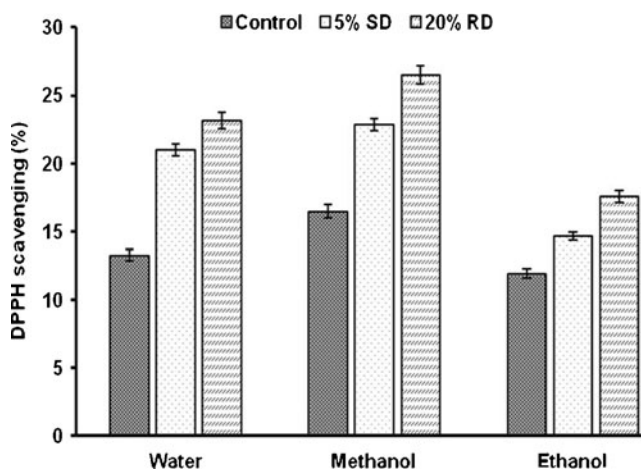
al. (2001) has also demonstrated that organically grown spinach contained 120 % higher antioxidant activity and Chinese cabbage showed about 20–50 % higher antioxidant activity compared to their conventionally grown plants.

#### Antioxidant potential of banana fruits

Antioxidants are defined as compounds that can delay, inhibit, or prevent the oxidation of enzymes, proteins, DNA, and lipids by scavenging free radicals and diminishing oxidative stress caused due to reactive oxygen and/or nitrogen species, superoxide anion, hydrogen peroxide, hydroxyl radical, and peroxy nitrite (Ames et al. 1993). DPPH is a free radical assay often used to measure the radical scavenging activity of natural compounds. The DPPH radical scavenging activity (in percent) of different fruit extracts (ME, EE, and AE) of treated and control plants were evaluated (Fig. 1). The RD was most effective which showed higher %RSA in ME of fruit (26.47 %). On the other hand plants supplemented with SD also demonstrated %RSA in ME (22.85 %). The control plants showed less %RSA of ME (16.45 %).

FRAP assay is widely used to determine the efficiency of antioxidant compounds to compete with the FRAP reagent and reduce the ferric to ferrous. This assay is one of the most simple, rapid, inexpensive direct tests of total antioxidant power of a sample. Antioxidant compounds that are able to function in this approach are categorized as secondary antioxidants where they suppress the radical formation and prevent oxidative damage. The antioxidant capacity of ripe banana fruit using FRAP assay is shown in Fig. 2. The reducing power in ME of fruit was found to be higher in plants treated as RD (0.424±0.001) followed by SD (0.386±0.002). Control plants did not showed any significant results (ME is 0.334±0.005).

The DMPD assay has some advantages like high stability end point, quick reaction time, cost effectiveness, and is less cumbersome. At acidic pH in presence of a suitable oxidant



**Fig. 1** DPPH radical scavenging activity (in percent) of banana fruit in different solvent extracts. Control, SD=5 % (shoot dose), RD=20 % (root dose), data expressed in mean±SEM, n=5

solution (FeCl<sub>3</sub>), DMPD can form a stable red radical cation (DMPD<sup>+</sup>). Antioxidant compounds, which transfer a hydrogen atom to DMPD<sup>+</sup>, quench the red color, and produce a bleaching of the solution proportional to their amounts (Fogliano et al. 1999). As shown in Fig. 3, it was demonstrated that DMPD radical scavenging activity (in percent) of the plant extract treated as RD showed higher level of scavenged DMPD<sup>+</sup> radicals in ME (68.96 %). Whereas, SD showed 63.40 % RSA in the methanolic extract. The control plants showed less RSA, i.e., 54.25 % in ME.

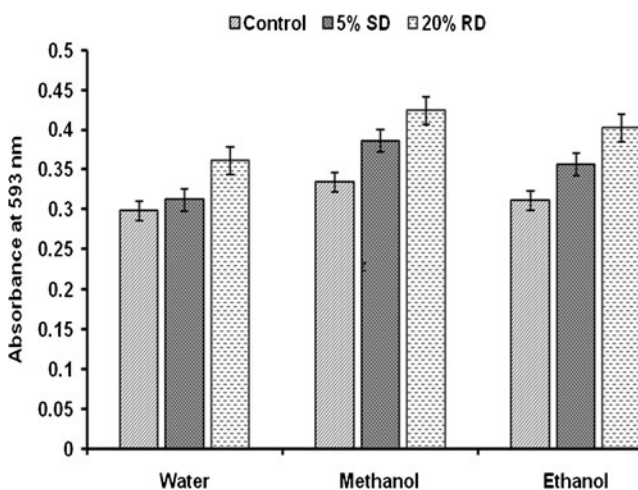
The highest antioxidant activity was observed in methanol revealed that methanol soluble factor was most likely responsible for reducing potential of the fruit extract. The difference in management of soil fertility affects the soil dynamics and plant metabolism, which may result in

differences in plant composition (Martins et al. 2005). In fact, several authors reported a similar increase in the antioxidant potential of different crops amended with organic wastes, suggesting that organic matter causes changes that favor the accumulation of antioxidants (Martins et al. 2005). So it is well stated here that the FDP application was helpful for the enhancement of antioxidant potential of the fruit.

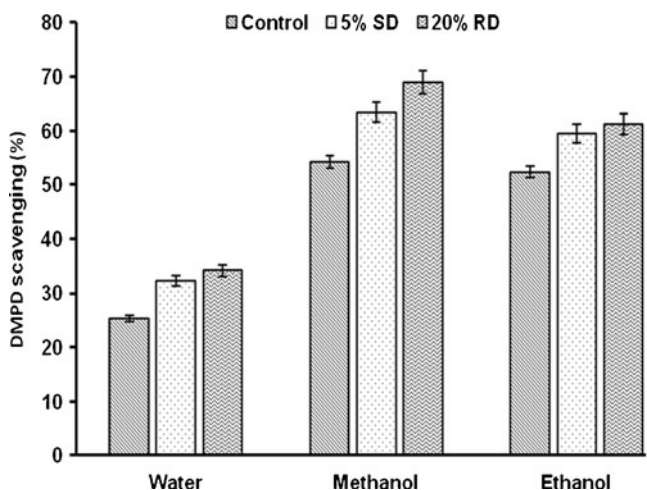
Macronutrients and micronutrients of soil and plants

Organic wastes are utilized in agriculture mainly for improving the soil physical and chemical properties and as well as nutrient source for growing crops. The potential of degraded chicken feather products as renewable source of fertilizer was investigated. It was found that feather hydrolysate showed presence of the macro (N, P, K, Ca, and Mg) and micro (Fe, Mn, Zn, and Cu) nutrients as reported in Table 3. The fertilizing with FDP in the form of RD may have increased both soil and plant mineral contents. The SD and control plants showed less metals accumulation in vegetative part and as well as in the soil.

Previous literature reported that feather waste by-product of poultry slaughter house can be directly used as slow-release nitrogen fertilizer for organic farming (Hadas and Kautsky 1994). Nevertheless, the disadvantage of this process was very low bioavailability of protein nitrogen to plants, because the feathers are not degraded by most common proteolytic enzymes. So we have exploited the potentiality of *Chryseobacterium* sp. RBT that can hydrolyse the chicken feathers into peptide and amino acids which played remarkable role as ideal crop bioenhancer. Beside the potent application as root dose, the plants also responded well for foliar fertilization which involves the application of a high nitrogen-content material to leaves at the time of active



**Fig. 2** FRAP of banana fruit in different solvent extracts. Control, SD=5 % (shoot dose), RD=20 % (root dose), data expressed in mean±SEM, n=5



**Fig. 3** DMPD<sup>+</sup> radical scavenging activity (in percent) of banana fruit in different solvent extracts. Control, SD=5 % (shoot dose), RD=20 % (root dose), data expressed in mean±SEM, n=5

**Table 3** Metal analysis of the FDP, soil, and vegetative tissue treated with FDP. Data shown as mean±SEM,  $n=5$ 

	Feather degradation products	Soil			Banana vegetative tissue		
		RD	SD	C	RD	SD	C
Nitrogen	9.22±0.687 <sup>a</sup>	1.254±0.06 <sup>a</sup>	0.912±0.04 <sup>a</sup>	0.875±0.02 <sup>a</sup>	3.122±0.09 <sup>a</sup>	2.926±0.11 <sup>a</sup>	2.711±0.05 <sup>a</sup>
Phosphorus	0.490±0.124 <sup>a</sup>	0.434±0.02 <sup>a</sup>	0.364±0.04 <sup>a</sup>	0.298±0.05 <sup>a</sup>	0.250±0.001 <sup>a</sup>	0.199±0.003 <sup>a</sup>	0.174±0.002 <sup>a</sup>
Potassium	0.630±0.321 <sup>a</sup>	1.986±0.08 <sup>a</sup>	1.890±0.11 <sup>a</sup>	1.765±0.04 <sup>a</sup>	3.508±0.084 <sup>a</sup>	3.243±0.126 <sup>a</sup>	3.010±0.07 <sup>a</sup>
Copper	6.45±0.154 <sup>b</sup>	98.61±0.555 <sup>b</sup>	90.41±0.564 <sup>b</sup>	89.75±0.430 <sup>b</sup>	9.966±0.354 <sup>b</sup>	6.723±0.312 <sup>b</sup>	7.364±0.235 <sup>b</sup>
Magnesium	1,711±0.231 <sup>b</sup>	81.78±0.341 <sup>b</sup>	78.00±0.356 <sup>b</sup>	83.75±0.412 <sup>b</sup>	163.68±0.514 <sup>b</sup>	159.12±0.445 <sup>b</sup>	153.84±0.400 <sup>b</sup>
Manganese	87.82±0.247 <sup>b</sup>	809.24±0.621 <sup>b</sup>	691.34±0.845 <sup>b</sup>	353.78±0.454 <sup>b</sup>	162.54±0.777 <sup>b</sup>	193.36±0.856 <sup>b</sup>	107.52±0.731 <sup>b</sup>
Calcium	2,212±1.012 <sup>b</sup>	22.05±0.189 <sup>b</sup>	17.75±0.185 <sup>b</sup>	23.62±0.211 <sup>b</sup>	2,029±3.542 <sup>b</sup>	1,829±2.244 <sup>b</sup>	1,617±1.542 <sup>b</sup>
Zinc	132.89±0.780 <sup>b</sup>	134.75±0.354 <sup>b</sup>	125.23±0.587 <sup>b</sup>	26.03±0.220 <sup>b</sup>	37.44±0.398 <sup>b</sup>	39.48±0.311 <sup>b</sup>	33.06±0.454 <sup>b</sup>
Iron	153.02±0.378 <sup>b</sup>	6,418±2.141 <sup>b</sup>	5,398±1.540 <sup>b</sup>	3,902±1.323 <sup>b</sup>	104.2±0.454 <sup>b</sup>	93.56±0.241 <sup>b</sup>	83.2±0.548 <sup>b</sup>

RD root dose (20 %), SD shoot dose (5 %), C control plant

<sup>a</sup> Dry weight (in percent)

<sup>b</sup> Milligrams per kilogram dry weight

growing season, thereby inducing rapid vegetative growth and crop yield.

The growing poultry industries are producing about millions of tons of noxious organic solid by-products like feathers. Although chicken feather contains high nitrogen content, they cannot be used directly as fertilizer due to the non-available form of nitrogen (Górecki et al. 2006). Therefore, recycling of this burdensome feather wastes into renewable source of nutrients is the subject of interest among animal breeders (Bertsch and Coello 2005). So there was a need of appropriate treatment to make them bioavailable to crops as a source of the organic nutrients. The biodegradation of chicken feather was performed by *Chryseobacterium* sp. RBT within 30 h, and the degraded product contains soluble proteins (3.28 mgml<sup>-1</sup>) and amino acids (4.83 mgml<sup>-1</sup>).

Banana is the most important commercial fruit crop grown all over the tropical regions of world, hence was selected as model crop for dose application. This crop requires a substantial amount of fertilizers for commercial cultivation, which is costly and may be hazardous when used excessively. Thus, there was a need of substitute like feather hydrolysate rich in active biochemicals, organic nitrogen, as well as other important macro- and microelements as source of organic nutrients. Previously, Cao et al. (2012) has reported the foliar application of the feather degradation products to Chinese cabbage and observed only the morphological changes in plants. In the present study we have amended these degraded products in horticulture practices for fertigation and foliar spray and assessment of plant biochemicals related to the growth, nutritional quality of fruit and soil metals was performed.

To the best of our knowledge and literature survey, this is the first report highlighting field studies and nutritional

quality enhancement using low-cost feather-based medium using biological method as a first step on the way to the industrial scaling strategy.

## Conclusions

The present study highlighted a successful process for the application of feather degradation products as a source of organic nutrient to banana plants. These feather degradation products constituted a promising approach in sustaining soil properties and promoted the production of plant sources rich in health beneficial compounds like antioxidants. The results indicated that this method will play the dual role as a bioenhancer to reduce use of chemical addition to agriculture and to eliminate the poultry waste treatments or disposal problem. Such cost effective practices could reduce resistant waste and obtain a valuable product that can open several avenues for further research and large-scale commercial production.

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