Research Article

Response of Onion Yield under Field Conditions to Humic Substances Derived from Different Sources

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Abstract | A field experiment was conducted at NARC, Islamabad to evaluate the effect of humic substances (HSs) derived from coal, sunflower, and maize waste materials on onion yield. Four HSs rates; 0, 20, 30 and 40 kg/ha were applied to onion (*variety Red Swat*) transplanted earlier to field (plot size=1.2×0.8 m^2). Results showed that maximum bulb yield of 25.1 and 24.5 t/ha were obtained with application of plant derived HSs; SFDHSs and MDHSs respectively, while 24.7 t/ha bulb were obtained with CDHA applied at the rate of 20 kg/ha. Almost same trend was observed with respect to the number of bulbs per plot. The highest number of 28.3 bulbs/plot were obtained in CDHA given at 20 kg/ha, followed by 25.5 and 24.6 bulbs/plot in SFDHSs and MDHSs both applied at 30 kg/respectively. On overall, results from this study implies it is optimum and economical to apply CDHA at 20 kg/ha whereas SFDHSs and MDHSs at 30-40 kg/ha for study area and vegetable.

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Introduction

Pakistan is primarily an agricultural country. Out of total 80 million ha (mha) geographical land mass, 22 mha is cultivated feeding over 180 million populations in the country. Most of land (18 mha) is irrigated while, remaining is cultivated under dry farming. The soils are dominantly alkaline calcareous whereby, availability of plant nutrients found to be the main hindrance in enhancing agricultural productivity. Coupled with macro and micronutrients deficiency, soil is characteristically low in organic matter (OM=<1%).

Having low soil fertility and soil OM, the low availability of nutrients due to poor recycling and absorption on exchangeable site thereof crops are applied with fertilizers such as urea and diammonium phosphate (DAP) for N and P respectively. On average threefold increase in food crop production in last 30 years mainly ascribed to thirteen fold increase in fertilizer use (FAO, 2004). Wheat, cotton and paddy are three main crops consuming major share of fertilizer usage such as 36%, 14% and 10% respectively. However due to decline fertilizer subsidies (N, P and K in 1986, 1995 and 1997 respectively) under structural adjustment & economic reform program coupled

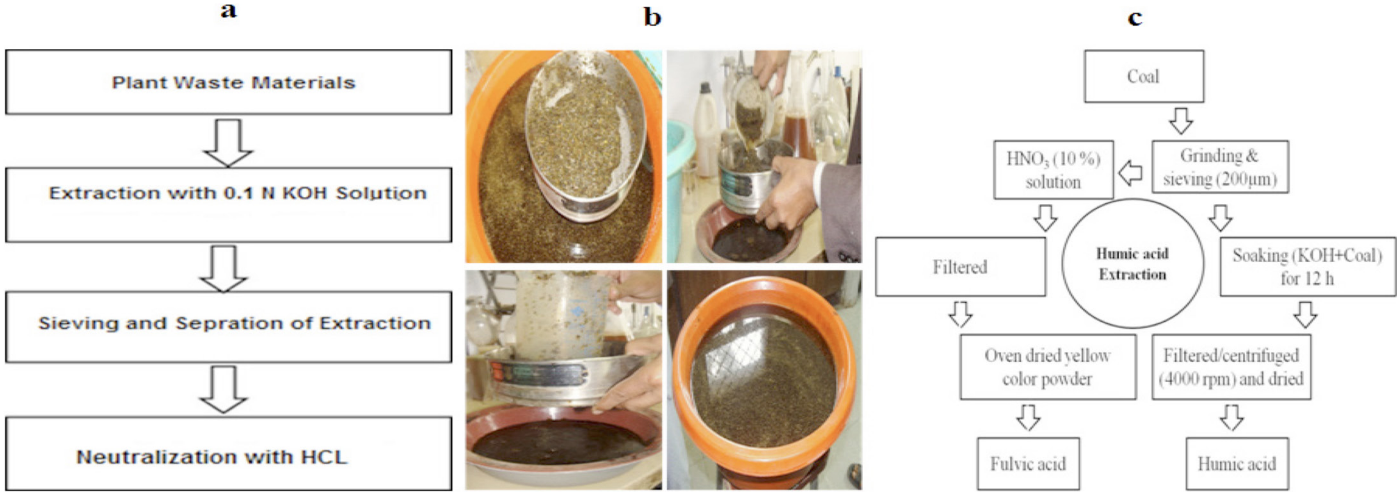

Figure 1: *Schematic diagram showing extraction of PDHSs (a) and CDHA (b)* (Khan et al., 2013b; Khan et al., 2014).

with price hike, the cost of production proportionally increased (FAO, 2006), and consequently production cost increased.

In order to keep production cost rational different attempts are being undertaken to incorporate fertilizer based on organic sources such as decomposed crop residues and humic substances (HSs). The HSs have both direct effect such as root hair formation and lateral root development (Canellas et al., 2002) as well as indirect effects on metabolism of soil microbes, availability of micronutrients (Fe, Zn and Cu), improve soil physical structure, biological membrane permeability, and acts as hormone like substances (Chen and Aviad, 1990; Clapp et al., 2001; Nardi et al., 2002; García-Mina et al., 2004).

Earlier research confirmed not only the beneficial effect of plant derived humic substances (PDHSs), and coal derived humic acid (CDHA) on vegetables as well as increases micronutrients solubility in soil (Khan et al., 2013a; 2013b; Khan, 2014). As component of OM, HSs are biogenic, heterogeneous, organic substances having yellow to black color, high molecular weight and refractory (McCarthy and Malcolm, 1990).

Onion (*Allium Cepa*. L), as integral part of food in kitchen is occupying an important role in our daily vegetable use. In Pakistan during 2012-2013, area under onion cultivation stands 126,000 ha with average production and yield of 1.6 mt and 13.4 t/ha respectively (Agric. Statistics of Pakistan, 2012-13). As a cool season crop it required high temperature and consequently long photoperiod is required for bulb formation. It gives good yield if grown on fertile and well-drained soil having acidic pH (5.8 to 6.5).

This study aimed at rationalizing the use of fertilizer with incorporation of HSs derived from different organic sources such as plant and coal for the improving production of onion in the study area.

Materials and Methods

Chemical analysis of soil

Soil pH and EC were determined in 1:1 soil paste, total organic C was measured by wet oxidation method (Walkley and Black, 1934), available P was determined as described by Olsen (1954) using 0.5 M $NaHCO_3$ at pH 8.5 (1:2 soil: extract ratio). Micronutrients (Zn, Cu, Fe and Mn) were determined with diethylene triamin pentaacetic acid (DTPA) test of Lindsay and Norvell (1978) using atomic absorption spectroscopy (Perkin Elmer, 800).

Extraction and quantification of humic substance

The ground plant materials of maize (cob parts) and sunflower (head parts) were soaked overnight into 0.1 N KOH solutions in 1:20 ratio (Figure 1a,b). The HSs was extracted from suspension by sieving (1.29 mm), and neutralizing through addition of dil. HCl soln. Concentration of HSs in extracted solution was quantified by spectrophotometer at 450 nm wavelength using calibration curve previously established through standard humic acid (HA) materials (Aldrich, Co. Germany). From coal samples, HA was also extracted from ground coal sample bypassing through 50 µm, soaked in 0.1 N KOH for 12 hours at 1:10 ratio, filtered (1.29 mm), centrifuged (4000 rpm for 5 min.)

Table 3: *Effect of humic substances on bulb number/plot, and bulb diameter.*

	Number bulb/plot			Bulb diameter (mm)		
HA rate (kg/ha)	CDHA	SFDHSs	MDHSs	CDHA	SFDHSs	MDHSs
0	$18.0^{c}\pm0.4$	$17.0^{d}\pm0.4$	$17.0^{c}\pm0.7$	$43.6^{c}\pm0.4$	$43.3^{b}\pm0.8$	$42.3^{c}\pm1.4$
20	$28.0^{a}\pm0.4$	$23.0^{b}\pm0.7$	$21.0^{b}\pm0.7$	$64.3^{a}\pm1.7$	$48.0^{b}\pm1.2$	$46.0^{b}\pm0.7$
30	$27.0^{ab}\pm1.4$	$25.0^{a}\pm0.4$	$24.0^{a}\pm0.4$	$62.3^{ab}\pm0.8$	$59.6^{a}\pm4.0$	$53.6^{a}\pm1.0$
40	$25.0^{b}\pm0.7$	$21.0^{c}\pm0.4$	$21.0^{b}\pm0.4$	$60.3^{b}\pm1.1$	$58.3^{a}\pm0.4$	$54.6^{a}\pm0.4$
LSD	2.24	1.33	1.53	3.02	5.72	2.66

Data are mean (n=3). Means followed by different letters are significantly different from each other at p≤ 0.05. Values are means ± standard error; **CDHA:** *Coal derived humic acid;* **SFDHSs:** *Sunflower derived humic substances;* **MDHSs:** *Maize derived humic substances.*

Table 1: *Selected chemical properties of soil.*

pH	EC	OM	P_{Olsen}	$K_{Exchan.}$	Zn	Cu	Fe	Mn
(1:1)	ds/m	%	mg/kg		(DTPA ext.) (mg/kg)			
8.0	0.24	1.2	6.5	84	0.38	0.92	9.8	9.6

OM: *Organic matter;* **EC:** *Electrical conductivity;* **DTPA:** *Diethylene triamine pentaacetic acid .*

Table 2: *Elemental analysis of humic substances.*

	N	C	H	S
HSs	%			
SFDHSs	1.25	53.48	3.22	0.77
MDHSs	1.61	51.78	3.12	0.66
CDHA	1.42	52.31	3.15	0.71

SFDHSs: *sunflower derived humic substances;* **MDHSs:** *Maize derived humic substances;* **CDHA:** *coal derived humic acid.*

and dried (Figure 1 c). Same as mentioned above the concentration of CDHA materials were determined by spectrophotometer at 450 nm wavelength.

Elemental analysis of HSs through CHNS elemental analyzer in National Centre of Excellence in Geology, Uni. Peshawar.

Testing of HSs

A field experiment was conducted in the experimental area of Land Resources Research Institute (LRRI) at NARC, Islamabad (33.7167°N and 73.0667°E) to assess the response of onion yield to PDHSs, and CDHA, Onion variety *Swat 1* previously grown in nurseries were transplanted to field in Jan. 2014 to field (plot size =1.2 × 0.8 m^2), arranged in randomized complete block (RCB) design. The PDHSs and CDHA were applied at 0 (no HA), 20, 30 and 40 kg/ha. Soil samples were collected before onset of experiment, and at the end just after crop harvest. Soil samples collected were processed for Physico-chemical analysis such as soil pH (1:1), EC (1:1), OM, macro and micro nutrients using standard analytical procedure listed in materials and methods section (Table 1). The experiment lasts for 180 days. Agronomic data such as number of bulbs per plot (0.96 m^2), bulb diameter, bulb weight per plot and biomass weight were determined. The plant tissues after harvest was collected and processed for macro and micronutrients analysis.

Statistical analysis

Statistical tests were performed using SAS (SAS 9.1.3, SAS Institute, Cary, NC). One way analysis of variance (ANOVA; PROC ANOVA) was used for analyses of (bulb number and diameter, bulb weight) to test the treatment effect. Fisher's least significant difference (*LSD*) tests were used to distinguish among means ($P \leq 0.05$).

Results and Discussion

Analysis of soil and HSs

Soil properties listed in Table 1 show that soil has pH 8 and EC <1.0 ds/m, and 1.2 % OM. As the soil has pH > 7.4, hence Olsen P (6.5 mg kg^{-1}) shows that soil is P deficient. Having slightly high pH, soil is also deficient in tested micronutrients (Zn, Cu, Fe and Mn). Elemental composition of CDHSs, PDHSs (sunflower derived as SFDHSs, and maize derived as MDHSs) presented in Table 2, showed that Carbon (C) constitute over 50% followed by Hydrogen (H) as ~3% and Nitrogen (N) as > 1% in all HSs derived from three sources.

Agronomic parameters

Number of bulbs per plot, bulb diameters and bulb weight per plant: Statistical analysis showed that humic acid application significantly ($p \leq 0.05$) increased both the number of bulbs per plot and bulb diameter (Table 3). With application of CDHA greater no. of bulb as much as >28 were found with application of CDHA at 20 kg/ha followed by 27 bulbs/plot with HSs at 30 kg/ha showing that HSs at 20 kg/ha was optimum dose in case of coal derived HA. While in case of PDHSs; SFDHSs and MDHSs at 30 kg/ha resulted maximum bulb numbers/plot. Bulb diameters showed a similar trend with CDHA at 20 kg/ha producing a maximum bulb diameter of 64.3 mm, while 59.6 mm and 54.6 mm when SFDHSs applied at 30 kg/ha, and MDHSs at 40 kg/ha respectively. Highest bulb weight per plant such as 117 g/plant was obtained with application of CDHA at 30 kg/ha while, 112.3 g/plant, and 100 g/plant were obtained with application of SFDHSs, and MDHSs given at 40 kg/ha. These results showed that HSs derived from different sources have different effect (Table 4).

Table 4: *Effect of Humic acid from different sources on bulb weight/ plant (g).*

	Bulb weight (g/plant)		
HA rate (kg/ha)	CDHA	SFDHSs	MDHSs
0	89.3^{b}±0.25	83.3^{c}±0.25	87.0^{c}±0.27
20	116.0^{a}±0.18	92.0^{b}±0.36	90.3^{c}±0.34
30	117.0^{a}±0.55	106.3^{a}±0.39	100.0^{b}±0.35
40	115.6^{a}±0.55	112.3^{a}±0.54	109.0^{a}±0.38
LSD	5.38	7.29	4.34

Data are mean (n=3). Means followed by different letters are significantly different from each other at p≤ 0.05. Values are means ± standard error, Means followed by different letters are significantly different from each other at P≤ 0.05; **CDHA:** *Coal derived humic acid;* **SFDHSs:** *Sunflower derived humic substances;* **MDHSs:** *Maize derived humic substances.*

Bulb yield (t/ha)

Statistical analysis showed that HA significantly ($p\leq$ 0.05) increased bulb yield (Table 4). Results show that the maximum yield (24.7 t/ha) was received with the application of CDHA at 20 kg/ha while with the application of SFDHSs, and MDHSs an increase of 25.1 and 24.5 t/ha were recorded respectively. It is clear from the result that 20 kg/ha of CDHA is the optimum dose for obtaining higher yield, and neither further increase nor decrease have any impact on bulb yield. In case of SFDHSs optimum dose is 30 kg/ha, and further increase decreases the yield to 22.6 t/ha. The MDHSs shows that with the increase in the dose (20-40 kg/ha) yield increased from 17.2 to 24.5 t/ha. Possible mechanism could be that HSs help in root elongation, lateral root development as reported by Canellas et al. (2002) (Table 5).

Table 5: *Effect of humic acid on onion bulb yield (t/ha).*

Yield (t/ha)			
HA rate (kg/ha)	CDHA	SFDHSs	MDHSs
0	18.3^{b}±0.25	17.1^{c}±0.25	13.0^{d}±0.55
20	24.7^{a}±0.18	17.8^{c} ±0.36	17.7^{b}±0.31
30	18.6^{b}±0.55	25.1^{a}±0.39	24.5^{a}±0.23
40	18.4^{b}±0.55	22.6^{b} ±0.54	15.5^{c} ±1.35
LSD	1.11	1.06	2.01

Data are mean of (n=3). Means followed by different letters are significantly different from each other at p≤ 0.05. Values are means ± standard error, Means followed by different letters are significantly different from each other at P≤ 0.05; **CDHA:** *Coal derived humic acid;* **SFDHSs:** *Sunflower derived humic substances;* **MDHSs:** *Maize derived humic substances.*

The role of HSs in increasing bulb yield is evident from this study which could be due to its pivotal role in soil fertility and plant nutrition. The stimulatory effect of HSs on plant growth could be either direct or indirect. Indirect effect ascribed to improving soil physical (Fortuna and Ortega, 1989), chemical (Hayes and Clapp, 2001), and biochemical characteristics (Lizarazo et al., 2005). Direct uptake of macromolecule of HSs trigger biochemical changes in plant, and consequently promote plant growth (Chen and Aviad, 1990; Chen et al., 2000; Cesco et al., 2002). Our earlier findings also show the increase of crop yield on two type soils viz clayey soil and sandy soil amended with humic substances (Khan et al., 2016 unpublished). The increase in tuber yield in current study could be attributed by the positive effect caused by addition of HSs. In production agriculture the excessive use of either water soluble nitrogenous fertilizer, or phosphorus fertilizer fixed in soil. Such fertilizer use in concurrent with HSs would make the former effective by the stimulatory effect of the latter.

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Authors Contribution

AK planned the reseach, designed experiments, collected and analysed data and wrote the manuscript. RUA analysed the results statistically and revised the draft. MZA helped in experimental work and writing og the manuscript. FH provided technical input in every stage of palnning and write up. MEA provided technical input and JD helped in writing up.

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