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MAIZE PRODUCTION AND NITROGEN USE EFFICIENCY IN RESPONSE TO NITROGEN APPLICATION WITH AND WITHOUT HUMIC ACID

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ABSTRACT

Humic acid (HA) extracted from poorly weathered Pakistani coal finds effective use in crop production and soil fertility management against low organic matter and soil nutrient depletion problems. An experiment with three HA levels (H0 = without HA, H1 = 2.5 L HA ha^{-1} and H2 = 5 L HA ha^{-1}), four N rates (N1 = 125 kg ha^{-1}, N2 = 150 kg ha^{-1}, N3 = 175 kg ha^{-1}, N4 = 200 kg ha^{-1}) and two controls N0P0K0 (No addition) and N0P1K1 (150 kg P_2O_5 ha^{-1} + 100 kg K_2O ha^{-1} without N) was conducted in 2010 at Research Farms, University of Agriculture Faisalabad, Pakistan using randomized complete block design and three replications to compare the effect of N use with and without HA on growth, yield, N use efficiency and economic feasibility of hybrid maize production. Results indicated that H1 rate could not provide promising results compared to urea only treatment (H0), while application of 5 L HA ha^{-1} provided maximum ($P < 0.05$) plant height (169.45 cm), leaf area index (3.71m^2 m^{-2}), total nitrogen uptake (183.17 kg ha^{-1}), grain protein concentration (9.99%), grain oil concentration (4.24%), N partial factor productivity (53.99 kg kg^{-1}), N-agronomic efficiency (40.15 kg kg^{-1}), N uptake efficiency (1.03 kg kg^{-1}), N-physiological efficiency (39.18 kg kg^{-1}) and N-internal utilization efficiency (37.37 kg kg^{-1}). The degree of synchrony between N demand and supply also increased with increase in HA application rate. When integrating with 5 L HA ha^{-1}, upper levels of N (175 & 200 kg ha^{-1}) were at par ($P > 0.05$) providing maximum grain yield (9.72 & 9.86 t ha^{-1}, respectively) and post-harvest total soil N concentration (1.53 & 1.57 g kg^{-1}). Upper N level of 200 kg ha^{-1} could not improve N use efficiency indices over application rate of 175 kg N ha^{-1}. The supreme value cost ratio (17.04 Rs. $Rs.^{-1}$) was also obtained with combination of 5 L HA and 175 kg N ha^{-1}. Against sole use of inorganic N, usual practice of common maize growers, results of this study recommend combined use of urea and HA for higher yield and better soil health.

Key words: Humic acid, Maize, N use efficiency, Synchrony between N demand and supply, Economic analysis.

INTRODUCTION

Maize (*Zea mays* L.) is the third most important cereal crop in the world after wheat and rice. It has pivotal position among the cash crops due to variety of uses of maize products and by products (Rizwan *et al*., 2003). During 2010-11, maize was grown on an area of 0.974 m ha with total annual production of 3.71 m tones (Govt. of Pakistan, 2011). There is wide gap between the average national yield and the progressive farmers` attained yield. The yield of this crop is also tremendously lower than other maize growing countries of the world (Rasheed *et al*., 2004). This may be due to the fact that most of the soils of Pakistan are poor in organic matter and hence in nitrogen being located in arid-semi arid climate.

Intensive use of inorganic fertilizers is associated with nutrient imbalance and decline in crop yield (Obi and Ebo, 1995) and especially if yield/economic return is not high enough, high cost of fertilizers render their use unprofitable (Tolessa and Friesen, 2001). Since crop response to applied fertilizer depends on soil organic matter, therefore, in scenario of less organic matter and continuous cropping systems, constant application of fertilizers could bring soil deprivation (Agboola and Omueti, 1982). Additionally, soils of Pakistan are alkaline and calcareous in nature with low organic matter and thus facing the problem of nutrient depletion. This has resulted in low fertilizer use efficiency of applied fertilizers (Ahmad *et al*., 2005). Therefore, there is a need of an alternative approach to increase crop production with minimum use of chemical fertilizers (Khaliq *et al*., 2004).

Humic acid (HA) is a naturally occurring organic compound polymeric in nature and is attributed to execute a broad diversity of functions (Schnitzer and Khan, 1972). It is formed by the decaying organic materials and is present in soils, lignites and peat (Lawson and Stewart, 1989). Complexing ability of HA and the properties regarding base exchange capacity could be significant with respect to transport of metal ions in soil via plant tissues and stabilization of soil organic matter in opposition to microbiological attack (Vaughan and MacDonald, 1976).

Humic acid carries an essential role in agriculture (Akinci *et al*., 2009) and application has already been established to show positive association with growth and yield of various crops, for example; tobacco roots (Mylonas and Mccants, 1980), peanut, clover plants and soybean (Tan and Tantiwiramanond, 1983), wheat (Malik and Azam,1985), olive plants (Tattini *et al*., 1991), chicory (Valdrighi *et al*., 1996), wheat, rice and radish (Hai and Mir, 1998), tomato (Adani *et al*., 1998; Dursun *et al*., 1999; Atiyeh *et al*., 2002), *Agrostisstolonifera* L. (Liu *et al*., 1998), eggplant (Dursun *et al*., 1999), ryegrass (Bidegain *et al*., 2000), cucumber (Atiyeh *et al*., 2002), *Helianthus annuus* L. (Kolsarıcı *et al*., 2005). According to Sarir and Durrani (2006), application of humic acid could enhance availability of plant nutrients and concomitant increase in yield and yield components of maize and could also make the soil environment more conducive for enzymatic activities which antagonize root diseases. Humic acids play an important role in the global nitrogen cycle by influencing the distribution, bioavailability and ultimate fate of organic nitrogen and there seems to be a synergistic relation between humic acid application and urea treatment (Dong *et al*., 2009).

Poorly weathered coal present in Pakistan consists of sizeable amount of humic acid which can be used effectively to boost up agricultural production (Hai and Mir, 1998). But research is limited on rate, timing and method of application of this indigenous HA in crop production. In this experiment, nitrogen fertilizer was integrated with commercial humic acid to observe whether chemical N fertilizer in combination with humic acid could improve yield and N use efficiency of maize over the application of chemical fertilizer alone? Also to test whether the additional cost incurred for the use of humic acid was justified by increased maize yield?

MATERIALS AND METHODS

Site Description: Field experiment was conducted at Research Farms (31°N: 73°E and 184.4 m above sea level), University of Agriculture Faisalabad, Pakistan during 2010. Soil was sandy clay loam with pH 7.7; Saturation 35%; EC_e 3.2 dS m^{-1}; CEC 6.1 $cmol_{(+)}$ kg^{-1}; Organic matter 0.61%; Total nitrogen 0.02%; Available P 6.9 mg kg^{-1} and Exchangeable K 129 mg kg^{-1}.

Experimentation: Seedbed was prepared by cultivating the soil for 2-3 times with tractor mounted cultivator followed by planking. Monsanto: DK-6525 maize hybrid was sown on 75 cm apart ridges within an experimental plot of size 10 m^2. Sowing was done on February 18, 2010 and manually with the help of dibbler by maintaining plant to plant distance of 20 cm and 2-3 seeds were placed per hole. After germination thinning was done to keep one plant and maintain plant to plant distance, bringing recommended density of 66666 maize plants ha^{-1}.

A 3 × 6 factorial experiment was conducted in randomized complete block (RCB) design using three replications.Three humic acid (HA) levels were used including H0 (without HA), H1 (2.5 L HA ha^{-1}) and H2 (5 L HA ha^{-1}). A total of six inorganic fertilizer application rates were tested; N0P0K0 (No addition), N0P1K1 (150 kg P_2O_5 ha^{-1} + 100 kg K_2O ha^{-1}), N1P1K1 (125 kg N ha^{-1} + 150 kg P_2O_5 ha^{-1} + 100 kg K2O ha^{-1}), N2P1K1 (150 kg N ha^{-1} + 150 kg P_2O_5 ha^{-1} + 100 kg K_2O ha^{-1}), N3P1K1 (175 kg N ha^{-1} + 150 kg P_2O_5 ha^{-1} + 100 kg K_2O ha^{-1}) and N4P1K1 (200 kg N ha^{-1} + 150 kg P_2O_5 ha^{-1} + 100 kg K_2O ha^{-1}). Whole of the P_2O_5 and K_2O was applied through Single Super Phosphate (SSP) and Sulphate of Potash (SOP), respectively, at sowing time. However, nitrogen was applied in two splits, 25% of rate at seed bed preparation and the rest at V9 (9 leaves stage) growth stage of maize plants. Liquid commercial humic acid (8%) was applied after germination by mixing with irrigation water. Up to crop physiological maturity, a total of 6 irrigations were applied using canal water. Granular insecticide Furadon (3% G) (FMC, Pakistan) was applied twice @ 20 kg ha^{-1} to control stem borer. Weeds were controlled manually. Harvesting and sample collection was done on May 28, 2010.

Growth and yield analysis: Ten plants were selected randomly from two central rows of each experimental unit and their height was measured in cm from ground surface to the top of plant. Then average plant height was determined. Leaves were counted and then average was worked out. Mean leaf area and leaf area per plant were calculated using the formulae given in author's previous paper (Niaz *et al*., 2014). Then Leaf Area Index was calculated using the following formulae; (Amanullah, 2004).

$$\text{Leaf Area Index } (m^2\ m^{-2}) = \text{Leaf area per plant x No of plants } m^{-2}$$

The average cob weight and the average grain yield per cob were calculated. Shelling percentage was recorded by multiplying the ratio of grain yield per cob (g) and the cob weight (g) with 100. Three samples, each of 1000 grains were taken randomly from the seed lot of each subplot, weighed and then averaged. Grain yield was recorded on subplot basis and then converted into tons per hectare (t ha^{-1}). After separating the cobs, the stalks were dried and weighed to record data on stover yield. Stover yield was recorded on subplot basis and then converted into tons per hectare (t ha^{-1}). Crop from each subplot was harvested manually, sun dried and weighed to determine the biological yield (grain, stover and pith yield) in kg per plot and then converted to t ha^{-1}. Harvest index was calculated by multiplying the ratio of grain yield (t ha^{-1}) and the biological yield (t ha^{-1}) with 100. The plant samples were dried at 70°C to a constant weight to determine the total above ground dry matter.

Weather data: Meteorological data was taken from nearly situated meteorology cell. During experiment, mean temperature, total rainfall, mean relative humidity, mean evapotranspiration, mean sun shine and mean wind speed were recorded to be 27.4 ºC, 21.3 mm, 44.2%, 4.7 mm, 8.7 hours and 4.8 km h^{-1}, respectively.

Chemical analysis: Total N in soil and the plant parts was estimated by Kjeldahl's distillation method after Jackson (1962). Soil particle size analysis and organic matter contents were determined by using methods described by Moodie *et al*. (1959). While other soil properties and extractable K from soil samples were analyzed using the methods described by U.S. Salinity Laboratory Staff (1954). Soil available P was recorded with method used by Watanabe and Olsen (1965). Oven dry weights of above ground plant parts were recorded before powdering to a particle size <0.2 mm and distillation which were later on used to estimate total N uptake in kg ha^{-1}. Grain protein concentration was determined by multiplying the grain N with 6.25 (Amanullah, 2004). Oil content of the representative seed sample was determined by Soxhlet method described by Low (1990).

N use efficiency and synchrony between N demand and supply: Agronomic indices used to measure nitrogen use efficiency included;
N-Partial Factor Productivity
PFP_N = kg grain yield per kg nitrogen applied
N-Agronomic Efficiency
AE_N = kg grain yield increase per kg nitrogen applied
N Uptake/Recovery Efficiency
RE_N = kg increase in N uptake per kg N applied
N-Physiological Efficiency
PEN = kg grain yield increase per kg increase in N uptake from fertilizer
N-Internal Utilization Efficiency
IE_N = kg yield per kg nitrogen uptake
Degree of Synchrony between N Demand and Supply
$F_N/(1-I_N/U_N)$
Where;

- F_N: The amount of applied N fertilizer (kg N ha^{-1})
- I_N: Crop N uptake in control plots without applied N (kg N ha^{-1})
- U_N: Crop N uptake in aboveground biomass at physiological maturity in N-fertilized plots (kg N ha^{-1})

More the value of this expression, lesser will be the synchrony between N supply and demand and vice versa.

Economic analysis: Local market rates were used for recording grain and stover value. Variable cost for each N application treatment was recorded comprising of urea-N rates according to global markets, additional charges for N application and the cost of humic acid. Finally, net return was calculated as value of the increased yield produced as a result of two inputs applied (N and humic acid) less the cost of those two variables (i.e. variable cost) while the value cost ratio was found as the ratio between the value of the additional crop yield and the variable cost.

Statistical Analysis: Data were subjected to analysis of variance (ANOVA) and least significant difference test (LSD) at $P \leq 0.05$ (Steel *et al.*, 1997) was used for significant differences between treatment means. Summary of ANOVA is given in Table I.

RESULTS

Growth parameters: Plant height increased with increase in humic acid application rates; maximum (169.45 cm) was recorded with H2 as compared to H1 (159.14 cm) and H0 (150.89 cm). The maximum leaf area index (3.71m^2 m^{-2}) was also recorded with H2 and it was statistically significant from lower humic acid levels. However, H1 (3.26 m^2 m^{-2}) could not bring significant increase over urea only treatment (H0) giving 3.09 m^2 m^{-2} leaf area index (Table II).

Regarding inorganic fertilizer rates, increase in N application rates recorded a consistent increase in plant height and leaf area index. Over all the N treatments, application rate of N4P1K1 provided statistically significant plant height (188.90 cm). Nevertheless, the application rate N3P1K1 was also at par to N4P1K1 for leaf area index (5.23 and 5.46 m^2 m^{-2}, respectively). The control treatment N0P0K0 came with the minimum values for plant height (115.14 cm) and leaf area index (0.94 m^2 m^{-2}), nevertheless, regarding the leaf area index, control treatment N0P1K1 (1.04 m^2 m^{-2}) was also at par to N0P0K0 treatment (Table II).

Yield parameters: Maximum cob weight (147.29 g) was recorded with higher humic acid level, H2. Here, intermediate level, H1, was also statistically different from urea only treatment, H0, i.e. giving significantly higher measurement of 129.76 g compared to 121.73 g, respectively (Table II). Biological yield and harvest index also increased statistically with increase in humic acid levels. The minimum measurements of 14.33 t biological yield ha^{-1} and 34.49% harvest index were recorded with the lowest rate of H0; H1 lifted these parameters to 14.97 t ha^{-1} and 35.40%, respectively; while the maximum values of biological yield (17.08 t ha^{-1}) and harvest index (38.96%) were taken by using the highest rate of H2 (Table II). Regarding to inorganic fertilizer rates, increase in N application rates recorded a consistent increase in cob weight. Over all the NPK treatments, application rate of N4P1K1 provided significant cob weight (190.98 g), biological yield (21.30 t ha^{-1}) and harvest index (40.82%). However, application rate of N3P1K1 was also at par to rate N4P1K1 providing 21.00 t ha^{-1} of biological yield.The N0P0K0 rate came with the minimum value of cob weight (51.76 g); biological yield (8.16 t ha^{-1}) and harvest index (28.25%)(Table II).

Other yield parameters, grain yield per cob, shelling percentage, 1000-grain weight, grain yield and stover yield were significantly affected by the interaction between humic acid and inorganic fertilizer rates (Table I). Significant grain yield per cob (196.96 g) was observed with treatment combination of H2N4P1K1. While for the minimum grain yield per cob; interactions H0N0P1K1 (19.51 g), H1N0P1K1 (19.47 g) and H0N0P0K0 (17.92 g) were found at par (Figure 1). The combinations of H2N3P1K1 and H2N4P1K1 were at par for maximum shelling percentage (94.38% and 96.32%, respectively) whereas H0N0P0K0 (42.49%), H0N0P1K1 (42.37%) and H1N0P0K0 (41.46%) recorded the minimum shelling percentage (Figure 1). The interaction of H2N4P1K1 increased 1000 grain weight (304.09 g) but non-significantly over combination of H2N3P1K1(298.74 g). Similarly, H1N0P0K0 (100.54 g) and H0N0P0K0 (98.67 g) were at par for minimum 1000 grain weight (Figure 1).

Integration of fertilizer N with intermediate level of humic acid application (H1, 2.5 L ha^{-1}) could not bring significant increase in grain yield over urea only treatment (H0, 0 L ha^{-1}), but when urea was applied in combination with 5 L HA ha^{-1} a significant increase was recorded (Figure 2). It is clear that the interaction of H2N4P1K1 increased grain yield (9.86 t ha^{-1}) but non-significantly over combination of H2N3P1K1 (9.72 t ha^{-1}). Regarding minimum grain yield, interactions H0N0P1K1 (2.18 t ha^{-1}), H1N0P0K0 (2.11 t ha^{-1}) and H0N0P0K0 (1.92 t ha^{-1}) were statistically at par (Figure 2). The maximum stover yield (10.02 t ha^{-1}) was recorded with treatment combinations of H2N3P1K1 and H2N4P1K1 (Figure 2) but some other interactions were also at par with these two including H1N3P1K1 (9.86 t ha^{-1}), H1N4P1K1 (9.79 t ha^{-1}), H0N3P1K1 (9.75 t ha^{-1}) and H0N4P1K1 (9.61 t ha^{-1}). The minimum stover yield (3.8 t ha^{-1}) was recorded with treatment combination of H0N0P0K0 but interactions H1N0P0K0 (3.98 t ha^{-1}), H0N0P1K1 (4.05 t ha^{-1}) and H1N0P1K1 (4.33 t ha^{-1}) were also at par providing the restricted stover yield (Figure 2).

N uptake parameters: Maximum grain nitrogen (1.60%) and total nitrogen uptake (183.17 kg ha^{-1}) were recorded with H2 compared to 1.46% grain nitrogen and the 157.02 kg total nitrogen uptake ha^{-1} obtained with H1. While, the lower humic acid rate, H0, yielded minimum values of grain nitrogen concentration (1.41%) and total nitrogen uptake (152.63 kg ha^{-1}) (Table III). Regarding inorganic fertilizer rates, two N rates, N3P1K1 and N4P1K1 were found at par for maximum grain nitrogen concentration (1.86 and 1.89%, respectively). Similarly, treatments N0P0K0 and N0P1K1 were at par for minimum grain nitrogen (0.90 and 0.96%, respectively). However, significantly maximum total nitrogen uptake (248.57 kg ha^{-1}) was recorded with higher N application rate, N4P1K1. While the control treatment N0P0K0 provided the minimum (63.25 kg ha^{-1}) N uptake (Table III). Stover N concentration was significantly affected by the interaction of humic acid and inorganic fertilizer application rates. Regarding maximum stover nitrogen concentration, combinations H2N4P1K1 (0.62%) and H2N3P1K1 (0.61%) were found at par to each other while significant from all others. The minimum stover nitrogen concentration (0.24%) was recorded with treatment combination of H0N0P0K0 but it was statistically at par with H0N0P1K1 (0.25%), H1N0P0K0 (0.25%), H1N0P1K1 (0.26%), H2N0P0K0 (0.26%) and H2N0P1K1 (0.26%) (Figure 3).

Grain quality parameters: The grain protein concentration increased statistically with increase in humic acid application rates; the maximum (9.99%) was recorded with H2 as compared to H1 (9.11%) and H0 (8.83%). The maximum grain oil concentration (4.24%) was recorded with H2. Rate of H1 (3.93%) could not provide significant increase in grain oil concentration over H0 which recorded grain oil concentration of 3.83% (Table III). The two higher N rates, N3P1K1 and N4P1K1 were found at par ($P>0.05$) for grain protein concentration (11.84% and 11.63%, respectively). The significantly maximum grain oil (5.41%) concentration was recorded with the highest rate of N4P1K1. The two control treatments N0P1K1 and N0P0K0 were found non-significantly different for minimum grain protein (5.98 and 5.64%, respectively) and grain oil (2.38 and 2.28%, respectively) concentrations (Table III).

N use efficiency parameters: N-partial factor productivity (PFP_N) and N-agronomic efficiency (AE_N) found increasing statistically with increase in humic acid application rates; upper rate H2 provided the maximum calculations for PFP_N (53.99 kg kg^{-1}) and AE_N (40.15 kg kg^{-1}) compared to H1 (43.92 kg kg^{-1} and 30.08 kg kg^{-1}, respectively) and H0 (41.75 kg kg^{-1} and 27.91 kg kg^{-1}). Similarly, the upper level, H2 provided the maximum N-uptake efficiency; RE_N (1.03 kg kg^{-1}), N-physiological efficiency; PE_N (39.18 kg kg^{-1}) and the N-internal utilization efficiency; IE_N (37.37 kg kg^{-1}). Nevertheless, for RE_N, PE_N and IE_N, medium humic acid level H1 (0.84 kg kg^{-1}, 35.90 kg $kg^{-1}$and 34.97 kg kg^{-1}) could not bring significant increase over H0 with measurements of 0.81 kg kg^{-1}, 34.67 kg kg^{-1} and 34.15 kg kg^{-1}, respectively (Table IV).

With respect to inorganic fertilizer N rates, lower three N rates; N1P1K1, N2P1K1 and N3P1K1 were at par ($P>0.05$) regarding PFP_N with measurements of 47.33 kg kg^{-1}, 47.23 kg kg^{-1} and 48.08 kg kg^{-1}, respectively. Further increment in nitrogen rate up to N4P1K1 curved the PFP_N to significantly minimum value of 43.58 kg kg^{-1}. Similarly, AE_N increased consistently with increasing N rates but up to N3P1K1 (35.61 kg kg^{-1}); further increase in N to N4P1K1 curved this increasing trend giving AE_N of 32.68 kg kg^{-1}. The minimum AE_N (29.88 kg kg^{-1}) was observed with lowest N application rate N1P1K1. The maximum RE_N of 1.00 kg kg^{-1} was recorded with treatment N3P1K1 that was significant from all other treatments. The other three N levels could be arranged in the decreasing order of RE_N as; N4P1K1 > N2P1K1 > N1P1K1 giving measurements of 0.91 kg kg^{-1}, 0.85 kg kg^{-1} and 0.80 kg kg^{-1}, respectively (Table IV).

Synchrony between N demand and supply: The maximum value of $\{F_N/(1-I_N/U_N)\}$ (244.94) was recorded with H0 followed by H1 (241.23) and H2 (226.76). Thus, the value of the expression $\{F_N/(1-I_N/U_N)\}$ decreased with increase in humic acid rate, i.e., the degree of synchrony between N demand and supply increased with increase in humic acid application from H0 to H2 (Table IV). Regarding N application rates, maximum value of $\{F_N/(1-I_N/U_N)\}$ (272.40) was observed with N4P1K1 that was statistically significant from other treatments while the minimum value (208.30) was observed with N1P1K1. Thus, the degree of synchrony between N demand and supply found significantly decreasing with increase in N application rate from N1 to N4 (Table IV).

Post-harvest soil total N concentration: The most significant influence of combined use of urea and humic acid compared to urea only treatment was recorded in case of post-harvest soil total N concentration (Figure 4). Without inorganic N, humic acid could not add to soil N reservoir over control treatments but integration of both N and HA significantly improved residual soil N which would better affect soil fertility status for the succeeding crops. The maximum post-harvest soil N concentration (1.57 g kg^{-1}) was observed in treatment combination of H2N4P1K1 that was statistically at par to H2N3P1K1 (1.53 g kg^{-1}). Therefore, use of upper HA level affected more promisingly to the residual soil N. On the other side, interactions H0N0P0K0 (0.14 g kg^{-1}), H0N0P1K1 (0.15g kg^{-1}), H1N0P0K0 (0.15g kg^{-1}), H1N0P1K1 (0.15g kg^{-1}), H2N0P0K0 (0.16g kg^{-1}) and H2N0P1K1 (0.17g kg^{-1}) were at par for minimum post-harvest soil N concentration (Figure 4).

Economic analysis: Data regarding economic analysis (Table V) indicate that maximum net returns of Rs. 170793 ha^{-1} attained in case of treatment combination of H2N4P1K1 followed by the combinations of H2N3P1K1, H2N2P1K1, H1N4P1K1 and H0N4P1K1 having net returns of Rs. 168804.2, 135083.2, 134978.2 and 128131.4 ha^{-1}, respectively. The lowest net return (Rs. 64596.2 ha^{-1}) was obtained with H0N1P1K1 treatment combination. Regarding value cost ratio (VCR), the maximum value of 17.04 was obtained with combination H2N3P1K1 and it was followed by H2N2P1K1 (15.68), H2N4P1K1 (15.41), H2N1P1K1 (14.53) and H1N3P1K1 (13.63). The least value of VCR was noted to be 10.13 and it was in combination of H0N1P 1K1. Integrated use of urea and humic acid thus proved to be economically feasible too, compared to urea only treatments (Table V).

Table 1. Summary of analysis of variance (ANOVA).

Sr. No.	Parameters	HA Rates (H)	N Rates (N)	HA Rates x N Rates (H x N)
1	Plant height	**	**	NS
2	Leaf area index	**	**	NS
3	Cob weight	**	**	NS
4	Grain yield per cob	**	**	**
5	Shelling percentage	**	**	**
6	1000 grain weight	**	**	**
7	Grain yield	**	**	**
8	Biological yield	**	**	NS
9	Harvest Index	**	**	NS
10	Stover yield	**	**	*
11	Grain N concentration	**	**	NS
12	Stover N concentration	**	**	**
13	Total N uptake	**	**	NS
14	Grain protein concentration	**	**	NS
15	Grain oil concentration	**	**	NS
16	N-partial factor productivity	**	**	NS
17	N-agronomic efficiency	**	**	NS
18	N-uptake efficiency	**	**	NS
19	N-physiological efficiency	**	NS	NS
20	N-internal utilization efficiency	**	NS	NS
21	Synchrony b/t N demand and supply	**	**	NS
22	Post-harvest total soil N concentration	**	**	**

NS = non-significant (P>0.05); * = significant (P<0.05); ** = highly significant (P<0.01)

Table 2. Plant height, leaf area index, cob weight, biological yield and harvest Index as influenced by humic acid and inorganic fertilizer rates.

	Plant height	Leaf area index	Cob weight	Biological yield	Harvest Index
	cm	$m^2\ m^{-2}$	g	t ha^{-1}	%
Humic Acid Rates					
H0	150.89 ± 7.60c	3.09 ± 0.42b	121.73 ± 13.69c	14.33 ± 1.28c	34.49 ± 1.25c
H1	159.14 ± 7.34b	3.26 ± 0.44b	129.76 ± 13.41b	14.97 ± 1.28b	35.40 ± 1.09b
H2	169.45 ± 6.84a	3.71 ± 0.49a	147.29 ± 12.93a	17.08 ± 1.28a	38.96 ± 1.24a
LSD ($P \leq 0.05$)	**4.68**	**0.19**	**2.80**	**0.27**	**0.60**
Inorganic Fertilizer Rates					
N0P0K0	115.14 ± 2.78e	0.94 ± 0.04d	51.76 ± 3.74f	8.16 ± 0.41e	28.25 ± 0.59e
N0P1K1	124.84 ± 4.31d	1.04 ± 0.04d	63.30 ± 5.40e	9.01 ± 0.44d	30.91 ± 0.88d
N1P1K1	167.26 ± 3.32c	3.18 ± 0.11c	151.50 ± 3.34d	15.29 ± 0.43c	38.53 ± 0.82c
N2P1K1	175.08 ± 4.65b	4.27 ± 0.16b	161.83 ± 4.44c	17.99 ± 0.47b	39.23 ± 0.71bc
N3P1K1	187.72 ± 2.18a	5.23 ± 0.24a	178.20 ± 3.67b	21.00 ± 0.44a	39.95 ± 0.76b
N4P1K1	188.90 ± 3.33a	5.46 ± 0.13a	190.98 ± 3.88a	21.30 ± 0.40a	40.82 ± 0.67a
LSD ($P \leq 0.05$)	**6.61**	**0.27**	**3.97**	**0.39**	**0.84**

Means (n = 3) sharing similar letters in a cell are statistically non-significant (P>0.05)
H0 = without Humic Acid, H1 = 2.5 L Humic Acid ha^{-1}, H2 = 5 L Humic Acid ha^{-1}, N0 = without N, N1 = 125 kg N ha^{-1}, N2 = 150 kg N ha^{-1}, N3 = 175 kg N ha^{-1}, N4 = 200 kg N ha^{-1}, P0 = without P, P1 = 150 kg P_2O_5 ha^{-1}, K0 = without K, K1 = 100 kg K_2O ha^{-1},

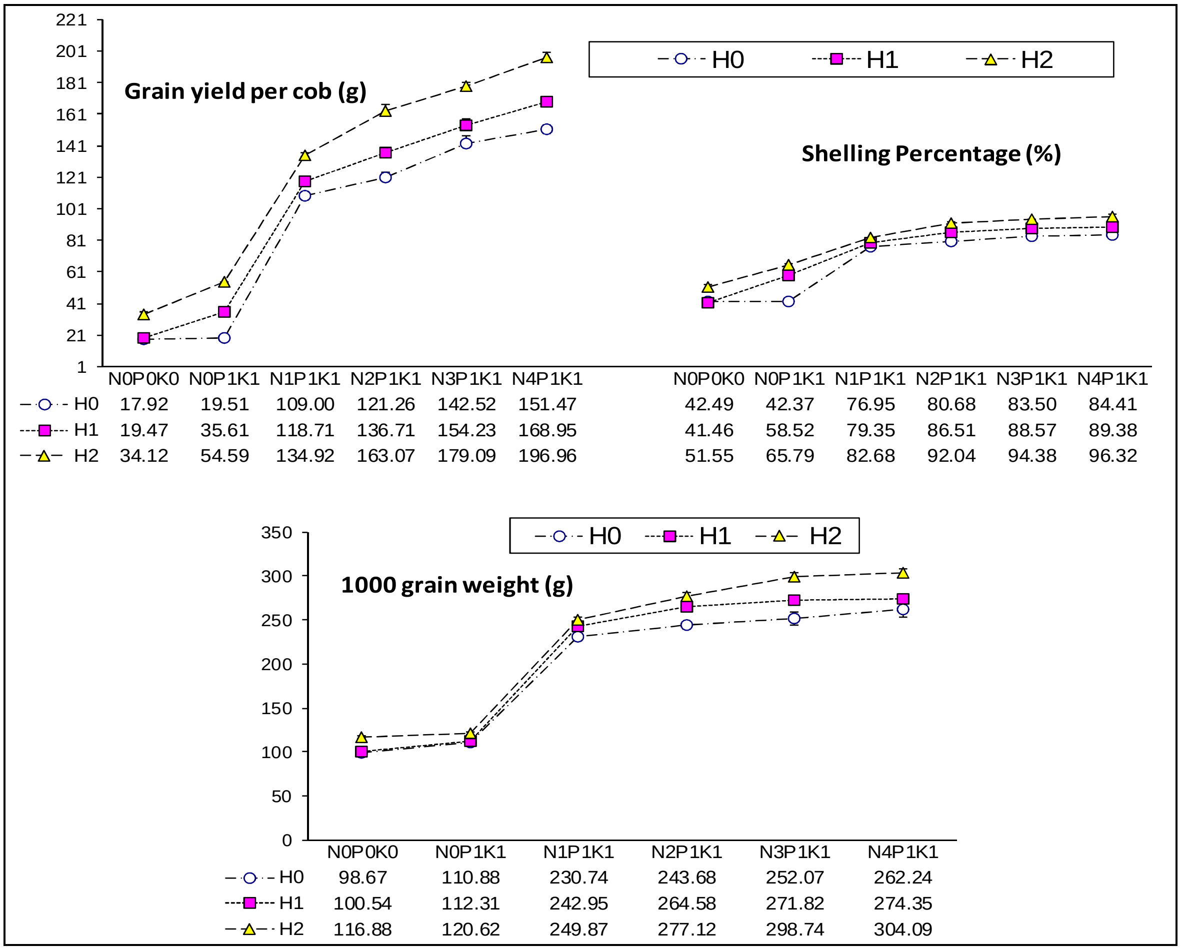

Figure 1. Grain yield per cob, shelling percentage and 1000 grain weight as influenced by the interaction of N and HA application rates: H0 = without Humic Acid, H1 = 2.5 L Humic Acid ha^{-1}, H2 = 5 L Humic Acid ha^{-1}, N0 = without N, N1 = 125 kg N ha^{-1}, N2 = 150 kg N ha^{-1}, N3 = 175 kg N ha^{-1}, N4 = 200 kg N ha^{-1}, P0 = without P, P1 = 150 kg P_2O_5 ha^{-1}, K0 = without K, K1 = 100 kg K_2O ha^{-1},

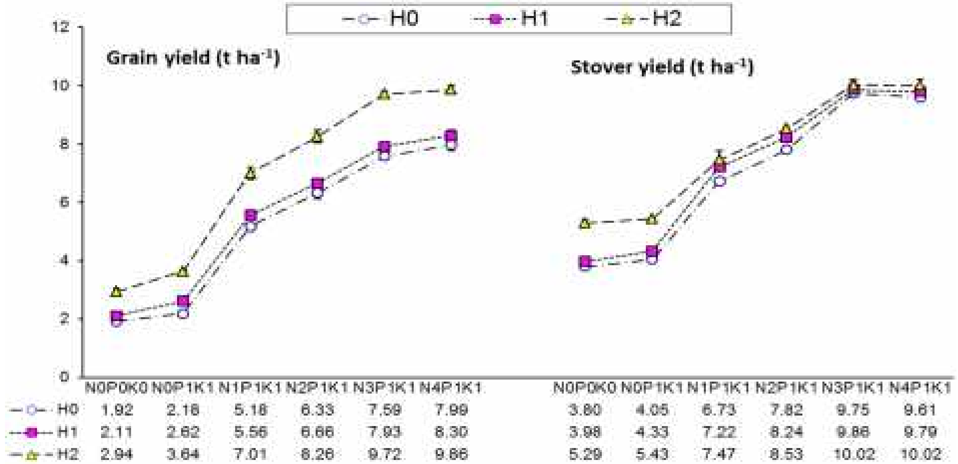

Figure 2. Grain and stover yield of maize as influenced by the interaction of N and HA application rates
H0 = without Humic Acid, H1 = 2.5 L Humic Acid ha^{-1}, H2 = 5 L Humic Acid ha^{-1}, N0 = without N, N1 = 125 kg N ha^{-1}, N2 = 150 kg N ha^{-1}, N3 = 175 kg N ha^{-1}, N4 = 200 kg N ha^{-1}, P0 = without P, P1 = 150 kg P_2O_5 ha^{-1}, K0 = without K, K1 = 100 kg K_2O ha^{-1},

Table 3. Grain N concentration, total N uptake, grain protein and oil concentrations as influenced by humic acid and inorganic fertilizer rates

	Grain N concentration	Total N uptake	Grain protein concentration	Grain oil concentration
	%	kg ha^{-1}	———%———	
Humic Acid Rates				
H0	1.41 ± 0.091c	152.63 ± 17.18b	8.83 ± 0.57c	3.83 ± 0.29b
H1	1.46 ± 0.098b	157.02 ± 17.33b	9.11 ± 0.62b	3.93 ± 0.29b
H2	1.60 ± 0.109a	183.17 ± 19.06a	9.99 ± 0.68a	4.24 ± 0.33a
LSD ($P \leq 0.05$)	**0.04**	**5.44**	**0.25**	**0.14**
Inorganic Fertilizer Rates				
N0P0K0	0.90 ± 0.013d	63.25 ± 3.24f	5.64 ± 0.08d	2.28 ± 0.05e
N0P1K1	0.96 ± 0.017d	73.52 ± 3.78e	5.98 ± 0.10d	2.38 ± 0.05e
N1P1K1	1.60 ± 0.031c	166.03 ± 5.03d	10.01 ± 0.20c	4.07 ± 0.10d
N2P1K1	1.72 ± 0.040b	193.86 ± 6.39c	10.77 ± 0.25b	4.73 ± 0.14c
N3P1K1	1.86 ± 0.052a	240.43 ± 7.39b	11.63 ± 0.33a	5.11 ± 0.12b
N4P1K1	1.89 ± 0.042a	248.57 ± 6.22a	11.84 ± 0.26a	5.41 ± 0.08a
LSD ($P \leq 0.05$)	**0.06**	**7.70**	**0.36**	**0.19**

Means (n = 3) sharing similar letters in a cell are statistically non-significant ($P>0.05$)
H0 = without Humic Acid, H1 = 2.5 L Humic Acid ha^{-1}, H2 = 5 L Humic Acid ha^{-1}, N0 = without N, N1 = 125 kg N ha^{-1}, N2 = 150 kg N ha^{-1}, N3 = 175 kg N ha^{-1}, N4 = 200 kg N ha^{-1}, P0 = without P, P1 = 150 kg P_2O_5 ha^{-1}, K0 = without K, K1 = 100 kg K_2O ha^{-1}

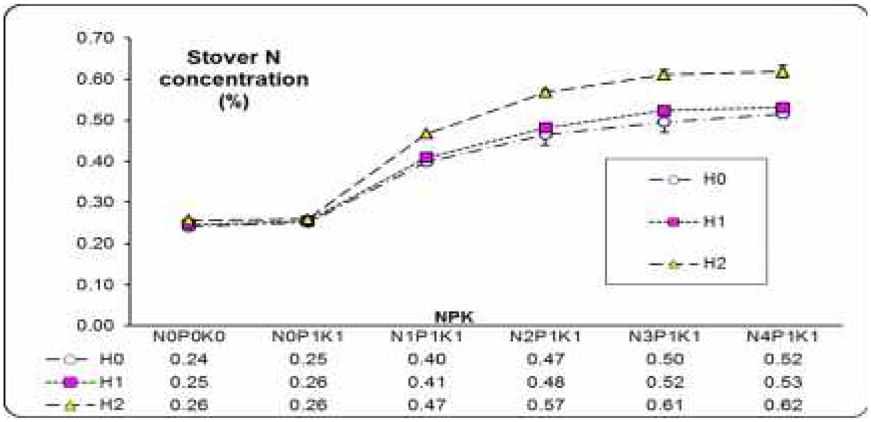

Figure 3. Stover N concentration as influenced by the interaction of N and HA application rates
H0 = without Humic Acid, H1 = 2.5 L Humic Acid ha^{-1}, H2 = 5 L Humic Acid ha^{-1}, N0 = without N, N1 = 125 kg N ha^{-1}, N2 = 150 kg N ha^{-1}, N3 = 175 kg N ha^{-1}, N4 = 200 kg N ha^{-1}, P0 = without P, P1 = 150 kg P_2O_5 ha^{-1}, K0 = without K, K1 = 100 kg K_2O ha^{-1},

Table IV. N-partial factor productivity, N-agronomic efficiency, N-uptake efficiency, N-internal utilization efficiency and expression for synchrony between nitrogen demand and supply {$F_N/(1-I_N/U_N)$} as influenced by humic acid and inorganic fertilizer rates

	N-Partial factor productivity (PFP_N)	N-Agronomic efficiency (AE_N)	N-Uptake efficiency (RE_N)	N-Physiological efficiency (PE_N)	N-Internal utilization efficiency (IE_N)	Degree of synchrony b/t N demand and supply {$F_N/(1-I_N/U_N)$}
	——kg kg^{-1}——					
			HA Rates			
H0	41.75±0.57^{c}	27.91±0.88c	0.81±0.03^{b}	34.67±0.62^{b}	34.15±0.39^{b}	244.94±6.70^{a}
H1	43.92±0.57^{b}	30.08±0.73^{b}	0.84±0.02^{b}	35.90±0.78^{b}	34.97±0.49^{b}	241.23±7.03^{a}
H2	53.99±0.96^{a}	40.15±0.75^{a}	1.03±0.03^{a}	39.18±0.91^{a}	37.37±0.59^{a}	226.76±7.76^{b}
LSD ($P \leq 0.05$)	**1.37**	**1.37**	**0.05**	**2.30**	**1.51**	**4.78**
			Inorganic Fertilizer Rates			
N1P1K1	47.33±2.31^{a}	29.88±2.31^{c}	0.80±0.04^{c}	36.94±1.44^{a}	35.52±0.90^{a}	208.30±3.98^{d}
N2P1K1	47.23±2.07^{a}	32.69±2.07^{b}	0.85±0.04^{c}	38.13±0.99^{a}	36.48±0.68^{a}	228.30±3.83^{c}
N3P1K1	48.08±1.91^{a}	35.61±1.91^{a}	1.00±0.04^{a}	35.56±0.69^{a}	34.94±0.51^{a}	241.58±2.57^{b}
N4P1K1	43.58±1.52^{b}	32.68±1.52^{b}	0.91±0.03^{b}	35.71±1.04^{a}	35.05±0.77^{a}	272.40±2.35^{a}
LSD ($P \leq 0.05$)	**1.58**	**1.58**	**0.06**	**2.65**	**1.74**	**5.52**

Means (n = 3) sharing similar letters in a cell are statistically non-significant ($P>0.05$)
H0 = without Humic Acid, H1 = 2.5 L Humic Acid ha^{-1}, H2 = 5 L Humic Acid ha^{-1}, N0 = without N, N1 = 125 kg N ha^{-1}, N2 = 150 kg N ha^{-1}, N3 = 175 kg N ha^{-1}, N4 = 200 kg N ha^{-1}, P1 = 150 kg P_2O_5 ha^{-1}, K1 = 100 kg K_2O ha^{-1},

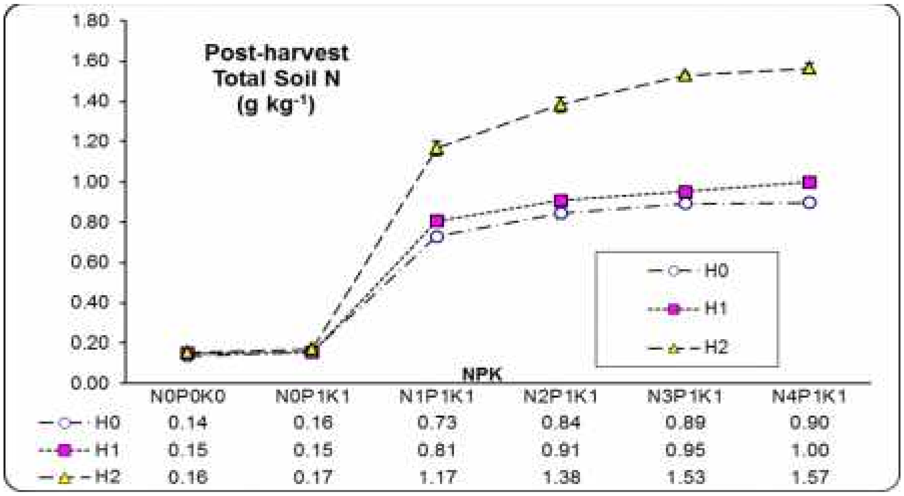

Figure 4. Post-harvest soil total N concentration as influenced by the interaction of N and HA application rates
H0 = without Humic Acid, H1 = 2.5 L Humic Acid ha^{-1}, H2 = 5 L Humic Acid ha^{-1}, N0 = without N, N1 = 125 kg N ha^{-1}, N2 = 150 kg N ha^{-1}, N3 = 175 kg N ha^{-1}, N4 = 200 kg N ha^{-1}, P0 = without P, P1 = 150 kg P_2O_5 ha^{-1}, K0 = without K, K1 = 100 kg K_2O ha^{-1},

Table V. Economic analysis for the various treatment combinations

	Grain Value @ Rs. 23/kg	Stover Value @ Rs. 1000/ton	Gross Returns	Gross Returns above Control	Variable Cost	Net Returns	Value Cost Ratio; VCR
	(A)	**(B)**	**C=(A+B)**	**(D)**	**(E)**	**(D-E)**	**(D/E)**
	——Rs. ha^{-1}——						**Rs. Rs.$^{-1}$**
H0N0P0K0	44192.59	3803.59	47996.18	xxxxx	xxxxx	xxxxx	xxxxx
H0N0P1K1	50157.18	4051.27	54208.45	xxxxx	xxxxx	xxxxx	xxxxx
H1N0P0K0	48430.33	3979.16	52409.49	-1798.96	xxxxx	xxxxx	xxxxx
H1N0P1K1	60224.73	4331.94	64556.67	10348.22	xxxxx	xxxxx	xxxxx
H2N0P0K0	67553.30	5293.04	72846.34	18637.89	xxxxx	xxxxx	xxxxx

H2N0P1K1	83705.43	5431.64	89137.07	34928.62	xxxxx	xxxxx	xxxxx
H0N1P1K1	119154.73	6724.91	125879.65	71671.19	7075	64596.19	10.13
H0N2P1K1	145650.40	7817.68	153468.07	99259.62	8400	90859.62	11.82
H0N3P1K1	174659.05	9745.71	184404.76	130196.30	9725	120471.30	13.39
H0N4P1K1	183783.79	9606.10	193389.89	139181.44	11050	128131.44	12.60
H1N1P1K1	127872.33	7223.47	135095.80	80887.35	7475	73412.35	10.82
H1N2P1K1	153172.33	8240.91	161413.25	107204.79	8800	98404.79	12.18
H1N3P1K1	182336.33	9859.55	192195.88	137987.43	10125	127862.43	13.63
H1N4P1K1	190846.33	9790.29	200636.62	146428.17	11450	134978.17	12.79
H2N1P1K1	161161.00	7470.84	168631.84	114423.38	7875	106548.38	14.53
H2N2P1K1	189957.00	8534.61	198491.61	144283.15	9200	135083.15	15.68
H2N3P1K1	223514.00	10023.68	233537.68	179329.22	10525	168804.22	17.04
H2N4P1K1	226833.67	10017.79	236851.45	182643.00	11850	170793.00	15.41

* Variable Cost carries; N cost @ Rs. 53 kg^{-1} + Rs. 450/- as top dressing charges of 2nd N split + cost of humic acid @ Rs. 160 L^{-1}.(85 Pakistani rupees = 1 US dollar)

In addition, Rs. 37000 ha^{-1} as uniform charges (seedbed preparation, land lease, hybrid seed, planting, P, K, irrigation, plant protection, and labors used for various operations plus application of first or basal dose of N at seed bed preparation) were also employed to all the experimental plots including controls.

H0 = without Humic Acid, H1 = 2.5 L Humic Acid ha^{-1}, H2 = 5 L Humic Acid ha^{-1}, N0 = without N, N1 = 125 kg N ha^{-1}, N2 = 150 kg N ha^{-1}, N3 = 175 kg N ha^{-1}, N4 = 200 kg N ha^{-1}, P0 = without P, P1 = 150 kg P_2O_5 ha^{-1}, K0 = without K, K1 = 100 kg K_2O ha^{-1},

DISCUSSION

Our results elucidated synergism between humic acid and urea. It is obvious from the data that addition of humic acid improved the effectiveness of urea increasing growth, yield and nitrogen uptake and use efficiency of maize crop. Synchrony between N demand and supply, post-harvest soil total N and economic feasibility of maize production also improved by humic acid addition compared to urea only treatment. The likely reasoning/hypothesis regarding the stimulatory action of humic substances could be copious, and the leading one which describes somewhat “direct” action of humic acid on plants is its hormonal nature. But there might be some other reasoning giving “indirect” actions, like; effects on uptake dynamics of soil nutrients, soil properties and soil microorganisms (Nardi *et al*., 1988; Casenave de Sanfilippo *et al*., 1990). In addition to these, improvement in cell permeability and increased metallic ions uptake could also be attributed to be the likely causes (Chen and Aviad, 1990).

The positive effect of humic acid on crop growth and yield could be mainly due to hormone-like activities through their association in photosynthesis, cell respiration, protein synthesis, oxidative phosphorylation, and various enzymatic reactions (Vaughan *et al*., 1985; Chen and Aviad, 1990). Nardi *et al*. (1988) also reported that humic substances exhibited auxin-, gibberellin-, and cytokinin-like activities.

Integrated use of urea with humic acid could also be suggested because independent urea application could suffer severe volatilization losses as ammonia not engaged by plants is quickly oxidized. This chemolithoautotrophic oxidation of ammonia to NO_2 is restricted by NH_3 availability (Laanbroek and Woldendorp, 1995). So, when ammonia oxidizers come in competition with heterotrophic bacteria and plants for ammonia, humic acid may have a role in controlling the soil NH_3 availability because of its adsorption characteristics (Mackowiak *et al*., 2001). Ammonia–N could be fixed abiotically to soil organic matter, lignin, peat or coal (Dong *et al*., 2009). Increased synchrony between N demand and supply and the post-harvest soil total N concentration might be ascribed to the fact that humic acid absorbs nutrients and especially ammonium in its structure and then releases slowly helping avoid losses through volatilization, leaching and/or fixation (Sibanda and Young, 1989).

Humic acid and urea, therefore, found to be in synergism effect with each other. Our results are supported by Zheng (1991) who also found that addition of humic acid could increase crop yields as compared to independent application of urea. Results are also in line with those of Ahmad and Tan (1991); Sarir (1998); Sharif *et al*. (2002) and Eyheraguibel *et al*. (2008).

Conclusion: Increase in application rate of humic acid increased economic yield and N use efficiency of hybrid maize. Compared to no humic acid, upper rate of humic acid and particularly when integrated with higher rates of nitrogen provided promising results. More research is invited to check humic acid potential whether capable to decrease the inorganic N requirement for economical maize production and also to study the residual effect of humic acid on the succeeding crop.

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