

ORIGINAL RESEARCH ARTICLE

Humic acid as a growth booster: Evaluating its synergistic influence on three red radish varieties

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ABSTRACT

The study focused on investigating the effects of varying levels of HA (HA₁ = 0, HA₂ = 25, HA₃ = 50, HA₄ = 75, and HA₅ = 100) on Red Dragon, Red Prince, and Red Meat varieties of red radish. This analysis aimed to unravel the relationship between different levels of HA and their impact on the growth and productivity of red radish genotypes. The findings revealed that the Red Prince genotype attained the utmost plant height of 24.00 cm, an average of 7.50 leaves per plant, a leaf area of 23.11 cm², a canopy cover of 26.76%, a leaf chlorophyll content of 54.60%, a leaf fresh weight of 41.16 g, a leaf dry weight of 8.20 g, a root length measuring 9.73 cm, a root diameter of 3.19 mm, a root fresh weight of 27.60 g, a root dry weight of 6.75 g, and a remarkable total yield of 17.93 tons per hectare. The implications of this study are poised to benefit farmers within the Dera Ismail Khan Region, specifically in the plain areas of Pakistan, by promoting the cultivation of the Red Prince variety.

Keywords: Humic Acid; Red Dragon; Red Prince; Red Meat; Growth; Yield

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1. Introduction

Red radish, scientifically known as *Raphanus raphanistrum* and belonging to the esteemed family *Brassicaceae*, exhibits a captivating array of dense roots that showcase a remarkable diversity in scale, shape, and color^[1]. Renowned for its delectable flavor and tantalizing pungency, the red radish stands as a distinguished member of the radicular group. This extraordinary crop thrives in a multitude of climatic conditions, ranging from tropical to subtropical, and even extending its resilience to temperate regions. Such adaptability allows red radish to flourish across diverse environments, imparting it with a distinguished reputation^[2]. Cultivated primarily for its tender tuberous roots, which find their way into raw salad preparations as well as cooked vegetable dishes, red radish presents an alluring gastronomic experience^[3]. The youthful foliage of this plant also boasts culinary utility, serving as a delectable vegetable component. When considering the color aspect, it is noteworthy that pink-skinned radishes often exhibit a higher vitamin C content in comparison to their white-skinned counterparts. However, the presence of light ailment significantly impacts the vitamin C content within the roots of radishes^[4,5]. Beyond its culinary prowess, red radish finds its place in salad recipes, constituting a valuable reservoir

of antioxidants, vitamin C, and health-enhancing elements. Among these constituents, glucose isolates and phenolic compounds play an influential role, conferring the esteemed crop with its esteemed beneficial properties. It is within the realm of polyphenolic compounds that red radish's remarkable attributes find their foundation, bestowing upon it an inherent capacity to promote wellness and vitality^[6].

The auspicious red radish, *Raphanus raphanistrum*, manifests an extraordinary prowess in impeding the proliferation of cancerous cells, thereby delivering a significant blow to their malignant growth^[7]. Moreover, this prodigious botanical entity boasts a plethora of enzymatic agents intricately involved in the intricate process of digestion. Furthermore, its remarkable medicinal properties find application in the amelioration of gastric ailments, offering respite and healing to afflicted individuals^[8]. The red radish flourishes optimally under specific environmental conditions, exhibiting a predilection for sandy loam soils boasting a pH range of 6.5 to 7.0, while concurrently basking in the plentiful radiance of copious sunlight. These vital elements harmoniously converge to nurture the growth and development of this distinguished plant. In the realm of Chinese radishes, a distinctive classification system consisting of five categories delineates their remarkable diversity^[9]. These categories include red-skinned variants with red flesh, red-skinned counterparts with white flesh, red-skinned variations displaying pink-hued flesh, green-skinned specimens adorned with pink-colored flesh, as well as white-skinned variants accentuated by their pristine white flesh. Such intricate categorization attests to the multifaceted nature of this botanical marvel, showcasing its diverse expressions in terms of skin pigmentation and internal tissue composition^[10].

Chinese radishes stand as custodians of the world's most abundant and diverse genetic resources, thereby securing a prominent position in the realm of botanical diversity^[11]. Their distinguished stature extends beyond mere genetic distinctions, encompassing a remarkable range of attributes including robust and plump root structures,

varying forms, and an impressive array of leaf colors^[12]. Within the domain of agricultural science, safeguarding the sustenance of burgeoning global populations while preserving the invaluable natural resources for future generations assumes paramount importance. This imperative gains particular significance when contrasted against the backdrop of the green revolution witnessed in the 20th century. The exponential expansion of agricultural practices has reached disconcerting levels, resulting in dire consequences such as irreversible alterations in the microclimate on a global scale and the degradation of numerous ecological services. Thus, the pressing need of the hour entails the implementation of innovative methodologies that foster sustainable growth and development. Such novel techniques are indispensable to mitigate the adverse effects inflicted upon the delicate equilibrium of our biosphere, ensuring the harmonious coexistence of humanity and the natural world for generations to come.

Harnessing the potential of humic substance-based plant bio-stimulants emerges as a promising avenue to address the aforementioned challenges^[13]. The term "HA" serves as a marketable nomenclature frequently employed to denote the amalgamation of humic acid and fulvic acid, naturally occurring components found within these deposits. Humic acid encompasses a diverse assemblage of molecules that exhibit solubility in alkaline water while resisting dissolution in acidic conditions^[14-16]. Its composition typically comprises a blend of feeble aliphatic and aromatic organic acids, engendering a myriad of effects on both plant growth and soil characteristics^[17]. Remarkably, humic acid serves as a formidable agent in curtailing soil cracking, elevating the capacity of water retention, mitigating the availability of toxic substances, facilitating the accessibility of macro and micro nutrients to plants, and preserving optimal soil pH levels^[18,19]. Furthermore, humic acid exerts profound influence on the physical and biological transformations occurring within plant shoots and roots, encompassing critical aspects such as their morphological development, nutrient

uptake and assimilation, as well as nutrient supply to support robust growth and productivity^[20].

The motivation of this study is to assess the influence of varying levels of humic acid (HA) on three genotypes of Red Radish. The study aims to investigate the effects of HA on various growth parameters and productivity of the Red Radish genotypes. The aim of this study is to unravel the relationship between different levels of HA and their impact on the growth and productivity of Red Radish genotypes. The study aims to determine the optimal level of HA for improving growth, yield, and other parameters of Red Radish. The findings have implications for promoting the cultivation of specific Red Radish varieties to benefit farmers and enhance the local economy.

2. Materials and methods

During the Rabi season of 2021–2022, a pot experimental trial was conducted at the Department of Horticulture, Faculty of Agriculture, Gomal University Dera Ismail Khan to assess the impact of diverse humic acid (HA) applications on the growth and yield of three distinct red radish genotypes. Radishes are chosen to be cultivated in the Rabi season in Pakistan. Rabi season in Pakistan, with its cool temperatures and ample water availability, is ideal for radish cultivation. Radish is essentially a cold-season crop and can be cultivated throughout the year but October to January is the best period. The high temperature of summer causes the plant to develop small tops, and roots rapidly become pithy and strongly pungent after reaching maturity. For this reason, it is difficult to produce quality radish during midsummer.

The meticulous research endeavor employed a Factorial Completely Randomized Design (CRD), incorporating two pivotal factors. Factor-A encompassed the analysis of five distinct humic acid applications, while Factor-B focused on the examination of three unique red radish genotypes. Each treatment configuration was diligently replicated four times, ensuring robustness and reliability in the experimental setup. The comprehensive details of the experimental treatments can be found in **Table 1**.

Table 1. Treatment details

Factor-A	(Humic acid (HA)) levels kg ha⁻¹
HA ₁	0
HA ₂	25
HA ₃	50
HA ₄	75
HA ₅	100
Factor-B	(Red radish genotypes)
V ₁	Red Dragon
V ₂	Red Prince
V ₃	Red Meat

Pots filled with sun-dried sandy soil served as the experimental medium, meticulously prepared for the rigorous investigation.

The soil was carefully mixed, incorporating predetermined levels of humic acid containing 40% organic matter. In addition, the recommended NPK doses (120:65:100 kg ha⁻¹ NPK) were added and intimately integrated into the soil matrix. Notably, the NPK sources included Urea, which had a nitrogen content of 46%, SSP with a phosphorus pentoxide (P₂O₅) concentration of 20%, and SOP providing a potassium oxide (K₂O) content of 50%. In the waning days of November 2022, seeds of the meticulously selected red radish genotypes were sown in meticulously prepared pots, each of 20 liters in capacity. The seeds were carefully distributed in the pots, ensuring uniform spacing between each seed. Promptly after sowing, the pots received immediate irrigation, drenching the soil with moisture. Meticulous attention was paid to maintain the soil within the pots at optimal moisture levels, fostering an environment conducive to the germination of the seeds. Furthermore, a rigorous regime of cultural practices ensued, encompassing timely weeding, diligent irrigation, periodic spraying, and judicious application of insecticides, executed as necessitated by prevailing conditions. Upon emergence of the seedlings, a process of equitable thinning was undertaken, ensuring that a uniform distance of separation was maintained, resulting in the retention of four seedlings per pot.

3. Results and discussion

3.1 Plant height (PH)

3.1.1 Results

A remarkable divergence in humic acid levels engendered substantial variations in PH (expressed

in cm), spanning a range of 26.58 cm to 20.27 cm. Notably, the pinnacle of PH attainment, reaching an impressive 26.58 cm, was observed in the treatment involving HA₄ (with a humic acid application rate of 75 kg ha⁻¹). In stark contrast, the minimal plant height data of 20.27 cm was recorded in the treatment receiving HA₁ (devoid of humic acid application at 0 kg ha⁻¹). The relationship between PH and escalating levels of humic acid indicated an upward trend in PH until a specific threshold (at 75 kg ha⁻¹), beyond which the subsequent increase in humic acid had a detrimental effect, leading to a decline in PH.

3.1.2 Discussion

Table 2 illustrates the notable impact observed on PH, expressed in cm, when examining the combined influence of humic acid levels and red radish genotypes^[21,22]. The data exhibited a range of 31.39 cm to 17.83 cm. Significant disparities were observed among the treatments HA₄V₂, HA₅V₂ and HA₃V₃ in terms of PH, with HA₄V₂ reaching a towering height of 31.39 cm, followed by HA₅V₂ at 28.39 cm and HA₃V₃ at 26.18 cm. These treatments exhibited notable differences when compared to each other. Conversely, the lowest PH response (17.83 cm) was noted in the HA₂V₂ and HA₁V₁ treatments, which displayed a similar and significantly diminished performance. Intermediate results for PH were observed in the remaining treatments^[23,24].

Table 2. Plant height (cm) as influenced by various HA levels and red radish genotypes

HA (kg ha ⁻¹)	Red radish genotypes			Mean
	(V ₁) Red Dragon	(V ₂) Red Prince	(V ₃) Red Meat	
HA ₁ = 0	19.07 jk	21.70 gh	20.05 ij	20.27 e
HA ₂ = 25	23.97 de	17.83 k	24.88 cd	22.23 d
HA ₃ = 50	22.19 fg	20.67 hi	26.18 c	23.01 c
HA ₄ = 75	23.49 def	31.39 a	24.86 cde	26.58 a
HA ₅ = 100	23.45 ef	28.39 b	21.17 ghi	24.34 b
Mean	22.43 c	24.00 a	23.43 b	–

Different letters indicated considerable variation at $P \leq 0.05$, LSD ($P \leq 0.05$) HA = 0.65, LSD ($P \leq 0.05$) Genotype = 0.43, LSD ($P \leq 0.05$) HA × Genotype = 1.42.

3.2 Canopy cover percentage

3.2.1 Results

The percentage of canopy cover exhibited a range of values from 24.76% to 19.62%. Significantly maximum canopy cover percentages of 24.76% and 23.67% were observed in the treatments receiving HA₄ (with a humic acid application rate of 75 kg ha⁻¹) and HA₅ (with a humic acid application rate of 100 kg ha⁻¹), respectively. These findings highlight the notable influence of humic acid application rates on achieving higher canopy cover percentages. Remarkably, these two treatments displayed similar outcomes when subjected to mathematical comparison. In contrast, the lowest findings for canopy cover (19.62%) were recorded in the treatment receiving HA₁ (without any humic acid application at 0 kg ha⁻¹). The findings indicate a linearly favorable influence of incremental increases in humic acid levels on vegetative growth, as demonstrated by the percentage of plant

canopy cover. However, this influence reaches a specific threshold at 75 kg ha⁻¹, beyond which further increases in humic acid levels do not lead to significant improvements in canopy cover. Statistically, it was observed that beyond the threshold of 75 kg ha⁻¹, there was a decline in canopy cover. This decline indicates that 75 kg ha⁻¹ represents an optimum humic acid level for achieving enhanced canopy cover in red radish. The results pertaining to canopy cover percentage are visually presented in **Table 3**. Similarly, a significantly higher plant canopy cover percentage of 26.76% was recorded in the Red Prince genotype (V₂), followed by the Red Meat genotype (V₃) and the Red Dragon genotype (V₁), displaying canopy covers of 22.30% and 18.22%, respectively. These three treatments exhibited statistically distinct outcomes when compared to each other. The data exhibited a range of values from 28.87% to 13.57%.

3.2.2 Discussion

The treatments HA₄V₂, HA₅V₂, HA₁V₂, and HA₃V₂ exhibited statistically extreme canopy cover percentages, with values of 28.87%, 28.57%, 26.09%, and 25.50% as shown in **Table 3**. Notably, when subjected to statistical comparison, these treatments exhibited non-significant differences among them. Similarly, statistically similar outcomes were observed in the HA₂V₁, HA₅V₁ and HA₁V₃ treatments, with canopy cover percentages of 18.51%, 19.47% and 19.09%, respectively. The lowest response in terms of canopy cover percentage was recorded in the HA₂V₁ treatment (18.51%) and the HA₁V₁ treatment (13.57%), which exhibited statistically equivalent outcomes. All other treatments yielded intermediate outcomes with regard to canopy cover percentage^[25,26].

3.3 No of leaves/plant

3.3.1 Results

Table 3. Canopy cover % as influenced by various HA levels and red radish genotypes

HA (kg ha ⁻¹)	Red radish genotypes			Mean
	(V ₁) Red Dragon	(V ₂) Red Prince	(V ₃) Red Meat	
HA ₁ = 0	13.57 f	26.09 ab	19.09 de	19.62 d
HA ₂ = 25	18.51 e	24.76 bc	22.55 bcd	21.94 c
HA ₃ = 50	18.56 e	25.50 abc	22.34 cd	22.13 bc
HA ₄ = 75	20.90 de	28.87 a	24.80 bc	24.76 a
HA ₅ = 100	19.47 de	28.57 a	22.67 bcd	23.67 ab
Mean	18.22 c	26.76 a	22.30 b	—

Different letters indicated considerable variation at $P \leq 0.05$, LSD ($P \leq 0.05$) HA = 1.70, LSD ($P \leq 0.05$) Genotype = 1.12, LSD ($P \leq 0.05$) HA × Genotype = 3.72.

However, this positive effect reaches a threshold at 75 kg ha⁻¹. Beyond this threshold, further increases in humic acid levels have an adverse effect, leading to a decline in the leaf count. Among the red radish genotypes, the Red Prince genotype (V₂) exhibited the highest number of leaves per plant (7.50), followed by the Red Meat genotype (V₃) with 7.00 leaves per plant, and the Red Dragon genotype (V₁) with 6.62 leaves per plant. Numerically significant differences were observed between genotypes. When examining the interactive effect of humic acid levels and red radish genotypes, the treatment HA₄V₁ recorded the highest statistically significant number of leaves per plant (9.84). Following the HA₄V₁ treatment, the

subsequent treatments of HA₅V₂, HA₄V₂, and HA₄V₃ displayed average leaf counts of 8.60, 8.22 and 7.88 leaves per plant, respectively.

Significant impacts on the No of leaves/plant were observed due to the application of different humic acid levels. The treatment with the highest statistically significant number of leaves per plant (8.65) was observed in HA₄, which had a humic acid application rate of 75 kg ha⁻¹. This was followed by HA₅, with a humic acid application rate of 100 kg ha⁻¹, showing 7.27 leaves per plant. These two treatments exhibited a significant difference between them. Similarly, HA₃ and HA₂ treatments displayed statistically similar results with 6.78 and 6.61 No of leaves/plant, respectively. On the other hand, the minimum number of leaves (5.89) was recorded in the HA₁ treatment (without any humic acid application at 0 kg ha⁻¹). The findings suggest that there is an initial increase in the number of leaves per plant with an increase in humic acid levels.

subsequent treatments of HA₅V₂, HA₄V₂, and HA₄V₃ displayed average leaf counts of 8.60, 8.22 and 7.88 leaves per plant, respectively.

3.3.2 Discussion

By subjecting to statistical analysis, these three treatments exhibited similar behavior. Additionally, numerically similar results were observed in the treatments of HA₃V₃, HA₂V₃ and HA₃V₂, with 7.52, 7.30 and 7.23 leaves per plant, respectively. **Table 4** depicts the lowest response in terms of the number of leaves per plant (5.54, 5.56, 5.63 and 5.63) observed in the treatments HA₂V₁, HA₃V₁, HA₁V₁ and HA₁V₃, respectively^[22,27,28].

Table 4. Number of leaves plant⁻¹ as influenced by various HA levels and red radish genotypes

HA (kg ha ⁻¹)	Red radish genotypes			Mean
	(V ₁) Red Dragon	(V ₂) Red Prince	(V ₃) Red Meat	
HA ₁ = 0	5.63 i	6.43 h	5.63 i	5.89 d
HA ₂ = 25	5.54 i	7.00 efgh	7.30 def	6.61 c
HA ₃ = 50	5.56 i	7.23 defg	7.52 cde	6.78 c
HA ₄ = 75	9.84 a	8.22 bc	7.88 bcd	8.65 a
HA ₅ = 100	6.51 gh	8.60 b	6.70 fgh	7.27 b
Mean	6.62 c	7.50 a	7.00 b	–

Different letters indicated considerable variation at $P \leq 0.05$, LSD ($P \leq 0.05$) HA = 0.35, LSD ($P \leq 0.05$) Genotype = 0.23, LSD ($P \leq 0.05$) HA × Genotype = 0.78.

3.4 Leaf area (LA)

3.4.1 Results

The application of different humic acid levels had a significant impact on the LA of red radish plants. The treatment with the highest LA (28.70 cm²) was HA₄, which had a humic acid application rate of 75 kg ha⁻¹. This was followed by HA₅ (with a humic acid application rate of 100 kg ha⁻¹) and HA₃ (with a humic acid application rate of 50 kg ha⁻¹), with LA of 24.28 cm² and 21.01 cm², respectively. These three treatments showed substantial differences in their responses. On the other hand, the lowest outcomes (18.39 cm² and 17.28 cm²) were observed in the HA₂ treatment (with a humic acid application rate of 25 kg ha⁻¹) and the HA₁ treatment (without any humic acid application at 0 kg ha⁻¹), respectively. These two treatments did not show a significant difference between them. The findings once again indicated that the vegetative growth of red radish was positively influenced in a linear manner by an increase in humic acid levels, reaching a maximum effect at the HA₄ threshold of 75 kg ha⁻¹. However, beyond this threshold, an adverse impact on vegetative growth was observed. In terms of red radish genotypes, the Red Prince genotype (V₂) exhibited the largest leaf area,

measuring 23.11 cm², followed by the Red Dragon genotype (V₁) with a LA of 22.25 cm², and the Red Meat genotype (V₃) with a leaf area of 20.77 cm². These genotypes showed numerical differences in their LA.

3.4.2 Discussion

Table 5 demonstrates the interactive effect of humic acid levels and red radish genotypes on LA. Among the treatments, the HA₄V₂ treatment displayed the highest LA, measuring 30.76 cm². Following closely were the HA₄V₃ and HA₄V₁ treatments, with LA of 27.73 cm² and 27.70 cm², respectively. Interestingly, these two treatments exhibited similar behaviors and were not significantly different from each other. HA₅V₁ and HA₅V₂ also produced statistically similar results, with a LA of 25.34 cm² each. Similarly, HA₃V₂ and HA₃V₁ exhibited statistically similar LA of 21.58 cm² and 21.43 cm², respectively. The lowest response in terms of LA was noted in HA₂V₂ (17.54 cm²) and HA₁V₁ (17.77 cm²) individually, and these two interactions showed mathematically similar results. Among the treatments, the HA₁V₃ treatment showed the lowest average LA, measuring 15.26 cm². In contrast, the remaining treatments exhibited intermediate results for LA^[25,29,30].

Table 5. Leaf area (cm²) as influenced by various HA levels and red radish genotypes

HA (kg ha ⁻¹)	Red radish genotypes			Mean
	(V ₁) Red Dragon	(V ₂) Red Prince	(V ₃) Red Meat	
HA ₁ = 0	17.77 i	20.43 efg	15.26 j	17.82 d
HA ₂ = 25	18.98 ghi	17.54 i	18.64 hi	18.39 d
HA ₃ = 50	21.43 def	21.58 de	20.01 fgh	21.01 c
HA ₄ = 75	27.70 b	30.67 a	27.73 b	28.70 a
HA ₅ = 100	25.34 c	25.34 c	22.15 d	24.28 b
Mean	22.25 b	23.11 a	20.77 c	–

Different letters indicated considerable variation at $P \leq 0.05$, LSD ($P \leq 0.05$) HA = 0.69, LSD ($P \leq 0.05$) Genotype = 0.45, LSD ($P \leq 0.05$) HA × Genotype = 1.50.

3.5 Leaf chlorophyll content %

3.5.1 Results

The leaf chlorophyll content of the red radish plants varied within a range of 55.00% to 49.16%. The HA₄ treatment, which involved applying humic acid at a rate of 75 kg ha⁻¹, displayed the highest recorded values for leaf chlorophyll content. Similarly, the HA₅ treatment, where a humic acid application rate of 100 kg ha⁻¹ was used, also exhibited significant chlorophyll contents of 53.68% and 55.00%, respectively. These two levels showed a significant difference from each other. Conversely, the lowest leaf chlorophyll content (49.16%) was recorded in the HA₁ treatment (without any humic acid application at 0 kg ha⁻¹). The results demonstrated that applying humic acid at a rate of 75 kg ha⁻¹ led to an improvement in chlorophyll content when compared to other treatments. However, it was observed that higher application rates of humic acid had a diminishing impact on chlorophyll content. The Red Prince genotype (V₂) exhibited the highest recorded leaf chlorophyll content among the different genotypes, reaching 54.60%. It was closely followed by the Red Meat genotype (V₃) with a chlorophyll content of 53.52%, while the Red Dragon genotype (V₁) displayed a chlorophyll content of 48.58%. All of these genotypes exhibited statistically significant differences in their results.

When considering the combined effect of different humic acid levels and genotypes, it was found that the HA₅V₂ treatment displayed the highest leaf chlorophyll content at 59.55%. Additionally, the HA₄V₃ treatment exhibited a significant chlorophyll content of 57.62%. These two treatments showed similar behaviors and were not significantly different from each other. Statistical analysis revealed that the chlorophyll content observed in the HA₄V₃ treatment was not significantly different from that of the HA₄V₂ and HA₅V₃ treatments, which recorded chlorophyll contents of 56.60% and 56.22%, respectively.

3.5.2 Discussion

Likewise, the treatments HA₂V₃, HA₃V₂, HA₃V₃, HA₂V₂, HA₁V₁, HA₃V₁ and HA₄V₁ yielded statistically similar results, showcasing chlorophyll contents of 53.53%, 53.43%, 52.59%, 51.72%, 51.69%, 51.40% and 50.80%, respectively. All of these interactions showed non-significant differences from each other, as shown in **Table 6**. The treatments HA₅V₁, HA₂V₁ and HA₁V₃ exhibited the lowest values for leaf chlorophyll content, measuring 45.25%, 47.29% and 47.68%, respectively. Notably, all of these treatments were statistically indistinguishable from each other. On the other hand, the remaining treatments demonstrated intermediate results in terms of leaf chlorophyll content^[25,31–33].

Table 6. Leaf chlorophyll content % as influenced by various HA levels and red radish genotypes

HA (kg ha ⁻¹)	Red radish genotypes			Mean
	(V ₁) Red Dragon	(V ₂) Red Prince	(V ₃) Red Meat	
HA ₁ = 0	48.13 ef	51.69 d	47.68 f	49.16 d
HA ₂ = 25	47.29 f	51.72 d	53.53 cd	50.84 c
HA ₃ = 50	51.40 d	53.43 cd	52.59 d	52.48 b
HA ₄ = 75	50.80 de	56.60 b	57.62 ab	55.00 a
HA ₅ = 100	45.25 f	59.55 a	56.22 bc	53.68 ab
Mean	48.58 c	54.60 a	53.52 b	–

Different letters indicated considerable variation at $P \leq 0.05$, LSD ($P \leq 0.05$) HA = 1.33, LSD ($P \leq 0.05$) Genotype = 0.89, LSD ($P \leq 0.05$) HA × Genotype = 2.92.

3.6 Leaf fresh weight (LFW)

3.6.1 Results

The highest recorded value for LFW, measuring 39.86 grams, was observed in the HA₄ treatment. This was followed by the HA₅, HA₃ and HA₂ treatments, with LFW of 32.17, 31.06 and 29.16 g,

respectively. All of these treatments exhibited statistically significant differences from each other. Conversely, the lowest response in terms of LFW (23.77 g) was recorded in the HA₁ treatment. The relationship between increasing levels of humic acid and total LFW revealed a positive response, showing an increase up to a certain threshold (75

kg ha⁻¹). However, beyond this point, a declining trend was observed, negatively impacting the overall LFW. Significant variations in LFW were observed when assessing the impact of different genotypes. The Red Prince genotype (V₂) displayed the highest LFW at 41.16 grams, followed by the Red Dragon genotype (V₁) and the Red Meat genotype (V₃) with weights of 28.45 g and 24.00 g, respectively. Each of these genotypes showed significant differences from each other.

When considering the combined effects of humic acid levels and genotypes, the treatment HA₄V₂ demonstrated the significantly highest LFW, measuring 58.25 g. Subsequently, the HA₃V₂ and HA₅V₂ treatments followed with weights of 42.64 g and 37.50 g, respectively.

Table 7. Leaf fresh weight (g) as influenced by various HA levels and red radish genotypes

HA (kg ha ⁻¹)	Red radish genotypes			Mean
	(V ₁) Red Dragon	(V ₂) Red Prince	(V ₃) Red Meat	
HA ₁ = 0	21.57 i	35.23 cd	14.51 j	23.77 d
HA ₂ = 25	29.91 ef	32.17 def	25.39 ghi	29.16 c
HA ₃ = 50	24.73 hi	42.64 b	25.81 gh	31.06 b
HA ₄ = 75	32.31 def	58.25 a	29.04 fg	39.86 a
HA ₅ = 100	33.76 cde	37.50 c	25.24 ghi	32.17 b
Mean	28.45 b	41.16 a	24.00 c	—

Different letters indicated considerable variation at $P \leq 0.05$, LSD ($P \leq 0.05$) HA = 1.84, LSD ($P \leq 0.05$) Genotype = 1.21, LSD ($P \leq 0.05$) HA × Genotype = 4.01.

3.7 Leaf dry weight (LDW)

3.7.1 Results

Likewise, the maximum numerical value for LDW was observed in the HA₄ treatment, measuring 9.30 grams. The treatments HA₁, HA₅, HA₃ and HA₂ yielded progressively lower LDW, with values of 6.02 g, 5.42 g, 4.44 g and 4.04 g, respectively. It is important to note that all of these treatments showed statistically significant differences from each other. In contrast, the HA₁ treatment recorded the minimum data for LDW, measuring 2.75 g. Once again, our findings revealed a consistent pattern of a linear rise in LDW as the levels of humic acid increased, up until a certain threshold of HA₄ (75 kg ha⁻¹). However, beyond this point, we observed a declining trend and reduction in LDW. In terms of various genotypes, the LDW exhibited a range between 8.20 and 3.68 g. Statistically, the Red Prince genotype (V₂) displayed the highest leaf dry weight of 8.20 grams, followed by the Red

3.6.2 Discussion

Table 7 clearly illustrates significant differences among all of these treatments. The treatments HA₁V₂, HA₄V₁ and HA₂V₂ displayed numerically similar results in terms of LFW, with values of 35.23 g, 32.31 g and 32.17 g, respectively. These three treatments did not demonstrate significant interactions with each other. On the other hand, intermediate results were observed in the HA₃V₃, HA₂V₃, and HA₅V₃ treatments, recording LFW of 25.81 g, 25.39 g and 25.24 g, respectively. There were no significant differences observed among the interactions mentioned. However, the HA₁V₃ treatment displayed the LFW weight, measuring 14.51 g. The remaining treatments resulted in intermediate outcomes in terms of LFW^[34-36].

Meat genotype (V₃) at 3.70 g and the Red Dragon genotype (V₁) at 3.68 g. These two treatments were numerically similar. When considering the combination of humic acid levels and genotypes, the treatment HA₄V₂ exhibited the highest LDW, measuring 16.06 g. Following closely was the HA₂V₂ treatment, which recorded a LDW of 8.29 g.

3.7.2 Discussion

Non-significant behaviors were observed in the HA₅V₂, HA₅V₃, HA₄V₁, HA₄V₃, HA₃V₂ and HA₁V₂ treatments, with LDW of 6.18 g, 6.09 g, 6.03 g, 5.82 g, 5.47 g and 5.00 g, respectively, as shown in **Table 8**. In contrast, the HA₁V₃ and HA₂V₃ treatments demonstrated the lowest response in terms of LDW, yielding 1.06 grams and 1.88 grams, respectively. None of these interactions exhibited significant differences from one another, indicating similar outcomes. As for the

Table 8. Leaf dry weight (g) as influenced by various HA levels and red radish genotypes

HA (kg ha ⁻¹)	Red radish genotypes			Mean
	(V ₁) Red Dragon	(V ₂) Red Prince	(V ₃) Red Meat	
HA ₁ = 0	2.19 ghi	5.00 cdef	1.06 i	2.75 d
HA ₂ = 25	3.15 fgh	8.29 b	1.88 hi	4.04 c
HA ₃ = 50	3.00 gh	5.47 cde	3.64 efgh	4.44 c
HA ₄ = 75	6.03 c	16.06 a	5.82 cd	9.30 a
HA ₅ = 100	4.01 defg	6.18 c	6.09 c	5.42 b
Mean	3.68 b	8.20 a	3.70 b	–

Different letters indicated considerable variation at $P \leq 0.05$, LSD ($P \leq 0.05$) HA = 0.85, LSD ($P \leq 0.05$) Genotype = 0.56, LSD ($P \leq 0.05$) HA × Genotype = 1.86.

remaining treatments, they produced intermediate results in terms of LDW^[37–39].

3.8 Root length (RL)

3.8.1 Results

In the HA₄ treatment (750 HA kg ha⁻¹), the highest recorded RL reached 11.80 cm. Subsequently, the HA₅ (100 kg ha⁻¹) and HA₃ (50 kg ha⁻¹) treatments exhibited RL of 10.55 cm and 9.90 cm, respectively. Statistically significant differences were observed in both of these treatments. Furthermore, HA₃ displayed similarity to HA₂, with both treatments producing roots measuring 9.26 cm in length. On the other hand, the HA₁ treatment (0 kg ha⁻¹) recorded the minimum RL data of 8.55 cm.

3.8.2 Discussion

The findings suggest that as the application of humic acid increased up to 75 kg ha⁻¹, there was a corresponding increase in RL. However, beyond this threshold, the RL started to decline. When evaluating different genotypes, the highest RL (11.60 cm) was recorded in the Red Meat genotype (V₃), followed by the Red Prince genotype (V₂) and

the Red Dragon genotype (V₁), with lengths of 9.73 and 8.71 cm, respectively. All of these treatments exhibited significant differences from each other, as shown in **Table 9**.

The HA₄V₃ treatment exhibited the highest RL of 13.79 cm, which was a significant finding. It was closely followed by the HA₃V₃, HA₂V₃, HA₅V₃ and HA₄V₂ treatments, which displayed RL of 12.31 cm, 11.33 cm, 11.27 cm and 11.15 cm, respectively. There were no significant differences observed among the four treatments mentioned. Similarly, the HA₅V₂ and HA₄V₁ treatments exhibited comparable outcomes in terms of RL, with measurements of 10.70 cm and 10.50 cm, respectively. In addition, the HA₁V₃, HA₁V₂ and HA₂V₂ treatments showed intermediate results, with average RL of 9.30 cm, 8.75 cm and 8.43 cm, respectively. Conversely, the HA₁V₁ and HA₃V₁ treatments exhibited the minimum RL, measuring 7.60 and 7.78 cm, respectively. The two interactions did not show any significant differences when compared to each other. In contrast, all the remaining treatments produced intermediate outcomes in terms of RL, measured in cm^[29,40–42].

Table 9. Root length (cm) as influenced by various HA levels and red radish genotypes

HA (kg ha ⁻¹)	Red radish genotypes			Mean
	(V ₁) Red Dragon	(V ₂) Red Prince	(V ₃) Red Meat	
HA ₁ = 0	7.60 g	8.75 efg	9.30 def	8.55 d
HA ₂ = 25	8.01 fg	8.43 efg	11.33 bc	9.26 c
HA ₃ = 50	7.78 g	9.60 de	12.31 b	9.90 bc
HA ₄ = 75	10.50 cd	11.15 bc	13.79 a	11.80 a
HA ₅ = 100	9.69 de	10.70 cd	11.27 bc	10.55 b
Mean	8.71 c	9.73 b	11.60 a	–

Different letters indicated considerable variation at $P \leq 0.05$, LSD ($P \leq 0.05$) HA = 0.67, LSD ($P \leq 0.05$) Genotype = 0.44, LSD ($P \leq 0.05$) HA × Genotype = 1.45.

3.9 Root diameter (RD)

3.9.1 Results

In the HA₄ treatment (75 HA kg ha⁻¹), the maximum recorded RD reached 2.68 mm. This was followed by the HA₅ (100 kg ha⁻¹) and HA₃ (50 kg

ha⁻¹) treatments, which displayed diameters of 2.21 mm and 2.11 mm, respectively. Both of these treatments showed no significant statistical difference. Conversely, the HA₁ (0 kg ha⁻¹) and HA₂ (25 kg ha⁻¹) treatments exhibited the minimum RD of 1.44 mm and 1.99 mm, respectively. Notably, these outcomes showed significant differences when compared to each other. The relationship between RD and increased humic acid levels demonstrated an initial increase up to a certain threshold (75 kg ha⁻¹), beyond which the diameter started to decline. This negative impact on RD indicates the adverse effects of surpassing the optimal humic acid level. The Red Prince genotype (V₂) recorded the highest RD data, measuring 3.19 mm, which was a significant finding. It was followed by Red Dragon (V₁) and Red Meat (V₃) with diameters of 1.72 mm and 1.34 mm, respectively.

3.9.2 Discussion

All of these genotypes exhibited significant mathematical differences from each other. Additionally, the Mino Early Long cultivar of radish also produced a significantly larger RD compared to other cultivars. In the HA₄V₂ treatment, the maximum RD of 4.55 mm was recorded, making it the highest among the treatments. It was followed by HA₃V₂ and HA₅V₂, which displayed RD of 3.20 mm and 3.04 mm, respectively. These two treatments exhibited a non-significant relationship. Similarly, the HA₄V₁ and HA₃V₁ treatments exhibited comparable results, with RD measuring 1.82 mm and 1.79 mm, respectively. These two interactions showed statistically similar outcomes. Intermediate results were observed in the HA₅V₃, HA₃V₃ and HA₂V₃ treatments, with RD measuring

1.38, 1.34, and 1.28 mm, respectively. **Table 10** illustrates that none of these interactions displayed a significant response when compared to each other. The HA₁V₃ and HA₁V₁ treatments demonstrated the lowest response in terms of RD, measuring 1.08 mm and 1.14 mm, respectively. Interestingly, these two interactions displayed non-significantly different behavior when compared to each other. On the other hand, all the other treatments produced intermediate results in terms of RD^[43–45].

3.10 Root fresh weight (RFW)

3.10.1 Results

The maximum recorded RFW was 21.78 g, observed in the HA₄ treatment, followed by HA₅, HA₃ and HA₂ with weights of 16.10 g, 15.23 g and 15.10 g, respectively. These treatments showed significant mathematical differences from each other. On the other hand, the lowest response of 14.52 g was recorded in the HA₁ treatment. These results demonstrated a significant improvement in RFW of red radish with the application of humic acid (HA), particularly up to a level of 75 HA kg ha⁻¹. Reproductive traits were adversely affected as the root weight declined beyond that level. The Red Prince genotype (V₂) recorded the highest numerical value for root fresh weight, measuring 27.60 g. Following closely were Red Dragon (V₁) with a weight of 15.03 g and Red Meat (V₃) with 6.10 g. All of these genotypes exhibited significant variations in terms of RFW. In the Red Prince cultivar, our findings suggest that the positive influence of increased leaf area, leaf chlorophyll content, plant weight, and root diameter may have contributed to the production of maximum root fresh weight.

Table 10. Root diameter (mm) as influenced by various HA levels and red radish genotypes

HA (kg ha ⁻¹)	Red radish genotypes			Mean
	(V ₁) Red Dragon	(V ₂) Red Prince	(V ₃) Red Meat	
HA ₁ = 0	1.14 h	2.10 cd	1.08 h	1.44 d
HA ₂ = 25	1.61 efg	3.04 b	1.28 gh	1.99 c
HA ₃ = 50	1.79 de	3.20 b	1.34 fgh	2.11 bc
HA ₄ = 75	1.82 de	4.55 a	1.64 ef	2.68 a
HA ₅ = 100	2.23 c	3.04 b	1.38 fgh	2.21 b
Mean	1.72 b	3.19 a	1.34 c	–

Different letters indicated considerable variation at $P \leq 0.05$, LSD ($P \leq 0.05$) HA = 0.16, LSD ($P \leq 0.05$) Genotype = 0.11.

The HA₄V₂ treatment statistically exhibited the highest recorded value for maximum root fresh weight (41.50 g), followed by HA₃V₂ with 28.79 g and HA₁V₂ with 25.79 g, in that order. The behavior of these two treatments showed no significant difference. Similar results for RFW were observed in the HA₅V₂, HA₅V₁ and HA₄V₁ treatments, with weights of 19.13 g, 18.12 g and 17.10 g, respectively.

3.10.2 Discussion

The treatments HA₁V₁, HA₅V₃ and HA₃V₁ demonstrated statistically similar outcomes, with

root fresh weights of 13.10 g, 11.43 g and 12.10 g, respectively. These levels showed significant numerical differences from each other, as depicted in **Table 11**. The treatments HA₁V₃, HA₃V₃, HA₄V₃ and HA₂V₃ exhibited the lowest response, with corresponding weights of 4.71 g, 5.10 g, 6.71 g and 7.14 g, respectively. Comparatively, these interactions did not show any significant differences amongst each other. Likewise, the remaining treatments resulted in moderate outcomes in relation to the recorded RFW (g)^[42,46–48].

Table 11. Root fresh weight (g) as influenced by various HA levels and red radish genotypes

HA (kg ha ⁻¹)	Red radish genotypes			Mean
	(V ₁) Red Dragon	(V ₂) Red Prince	(V ₃) Red Meat	
HA ₁ = 0	13.10 fg	25.79 bc	4.71 h	14.52 c
HA ₂ = 25	15.11 ef	23.00 c	7.14 h	15.10 bc
HA ₃ = 50	12.10 g	28.79 b	5.10 h	15.23 bc
HA ₄ = 75	17.10 de	41.50 a	6.71 h	21.78 a
HA ₅ = 100	18.12 de	19.13 d	11.43 g	16.10 b
Mean	15.03 b	27.60 a	6.10 c	–

Different letters indicated considerable variation at $P \leq 0.05$, LSD ($P \leq 0.05$) HA = 1.43, LSD ($P \leq 0.05$) Genotype = 0.95, LSD ($P \leq 0.05$) HA × Genotype = 3.14.

3.11 Root dry weight (RDW)

3.11.1 Results

The HA₄ treatment exhibited the highest recorded data for RDW, measuring 6.67 g, followed by HA₅, HA₃ and HA₂ with weights of 4.26 g, 3.52 g and 2.75 g, respectively. **Table 12** clearly demonstrates significant statistical variances among all of these treatments. Conversely, the HA₁ treatment displayed the lowest data for RDW, measuring 2.46 g. The RDW response exhibited a linear increase with the increment of HA levels, reaching a

maximum at a threshold of 75 kg ha⁻¹. However, beyond that level, there was a decline in root dry weight, adversely affecting its growth. The highest recorded RDW data, numerically, was observed in the Red Prince genotype (V₂) with a weight of 6.75 g. This was followed by Red Dragon (V₁) and Red Meat (V₃) with weights of 3.50 g and 1.56 g, respectively. These treatments exhibited statistically significant differences from each other. HA₄V₂ treatment exhibited the significantly maximum RDW, measuring 12.79 g, followed by HA₅V₂ with 7.31 g.

Table 12. Root dry weights (g) as influenced by various HA levels and red radish genotypes

HA (kg ha ⁻¹)	Red radish genotypes			Mean
	(V ₁) Red Dragon	(V ₂) Red Prince	(V ₃) Red Meat	
HA ₁ = 0	2.73 de	4.67 c	0.85 f	2.46 c
HA ₂ = 25	1.99 ef	3.80 cd	1.69 ef	2.75 c
HA ₃ = 50	2.07 ef	7.31 b	1.19 ef	3.52 b
HA ₄ = 75	5.37 c	12.79 a	1.87 ef	6.67 a
HA ₅ = 100	5.31 c	5.18 c	2.3 def	4.26 b
Mean	3.50 b	6.75 a	1.56 c	–

Different letters indicated considerable variation at $P \leq 0.05$, LSD ($P \leq 0.05$) HA = 0.76, LSD ($P \leq 0.05$) Genotype = 0.50, LSD ($P \leq 0.05$) HA × Genotype = 1.66.

3.11.2 Discussion

Likewise, the treatments HA₄V₁, HA₅V₁, HA₅V₂ and HA₁V₂ exhibited similar results in terms of RDW, yielding 5.37 g, 5.31 g, 5.18 g and

4.67 g, respectively. Importantly, all of these interactions demonstrated statistically comparable outcomes. The treatments HA₃V₁, HA₂V₁, HA₄V₃, HA₂V₃ and HA₃V₃ exhibited statistically intermediate results in terms of RDW, with recorded weights of 2.07 g, 1.99 g, 1.87 g, 1.69 g and 1.19 g, respectively. These interactions displayed non-significant differences when compared to each other. On the other hand, the minimum recorded data of 0.85 g for RDW was found in the HA₁V₃ treatment. On the other hand, the remaining treatments displayed intermediate outcomes for the total RDW, measured in g^[49-51].

3.12 Total yield (TY)

3.12.1 Results

The HA₄ treatment (75 HA kg ha⁻¹) exhibited the highest significant TY, recording a yield of 14.15 metric tons per hectare (t/ha). It was followed by HA₅ (100 kg ha⁻¹) with a yield of 10.46 t/ha, HA₃ (50 kg ha⁻¹) with a yield of 9.90 t/ha and HA₂ (25 kg ha⁻¹) with a yield of 9.80 t/ha. These three treatments exhibited similar numerical outcomes. In contrast, the lowest yield of 9.43 t/ha was observed in the HA₁ treatment with a HA level of 0 kg ha⁻¹. The TY (t/ha) exhibited an increasing trend in response to higher HA levels up to a threshold of 75 kg ha⁻¹. However, beyond this

point, there was a decline in yield, negatively impacting the TY (t/ha). In terms of statistical analysis, the Red Prince genotype (V₂) exhibited the highest yield, with a recorded value of 17.93 t/ha. It was followed by Red Dragon (V₁) and Red Meat (V₃) with yields of 9.76 t/ha and 4.54 t/ha, respectively. These genotypes displayed significant differences in yield.

3.12.2 Discussion

The HA₄V₂ treatment demonstrated the highest TY, recording a significant yield of 26.97 t/ha. This was followed by HA₃V₂ and HA₁V₂ treatments, which yielded 18.71 t/ha and 16.76 t/ha, respectively. The HA₅V₁ and HA₄V₁ treatments displayed statistically comparable results. Likewise, numerically similar outcomes were observed in the HA₂V₁ and HA₁V₁ treatments, with yields of 9.82 t/ha and 8.51 t/ha, respectively. The HA₁V₃, HA₃V₁, HA₁V₃ and HA₂V₃ treatments exhibited the lowest response in terms of TY (t/ha), with yields of 3.04 t/ha, 3.24 t/ha, 4.35 t/ha, and 4.64 t/ha, respectively. In **Table 13**, it can be observed that these interactions displayed statistically similar results when compared to each other. Additionally, all the remaining treatments yielded intermediate outcomes in terms of TY (t/ha)^[52-54].

Table 13. Total yield t/ha. as influenced by various HA levels and red radish genotypes

HA (kg ha ⁻¹)	Red radish genotypes			Mean
	(V ₁) Red Dragon	(V ₂) Red Prince	(V ₃) Red Meat	
HA ₁ = 0	8.51 fg	16.76 bc	3.04 h	9.43 c
HA ₂ = 25	9.82 ef	14.94 c	4.64 h	9.80 bc
HA ₃ = 50	7.74 g	18.71 b	3.24 h	9.90 bc
HA ₄ = 75	11.11 de	26.97 a	4.35 h	14.15 a
HA ₅ = 100	11.65 de	12.30 d	7.43 g	10.46 b
Mean	9.76 b	17.93 a	4.54 c	–

Different letters indicated considerable variation at $P \leq 0.05$, LSD ($P \leq 0.05$) HA = 0.93, LSD ($P \leq 0.05$) Genotype = 0.61, LSD ($P \leq 0.05$) HA × Genotype = 2.04.

4. Conclusion

The objective of this study was to assess the effects of different levels of HA on various growth and yield parameters of three red radish genotypes. The findings of the study indicated that the application of HA at a rate of 75 kg ha⁻¹ led to significant enhancements in the growth and yield characteristics of red radish, surpassing the outcomes of

other treatments. Notably, Red Prince genotype exhibited the most favorable outcomes across various parameters. Red Prince genotype, when treated with 75 kg ha⁻¹ of HA, exhibited the maximum plant height of 24.00 cm, 7.50 leaves per plant, leaf area of 23.11 cm², canopy cover of 26.76%, leaf chlorophyll content of 54.60%, leaf fresh weight of 41.16 g, leaf dry weight of 8.20 g, root length of

9.73 cm, root diameter of 3.19 mm, root fresh weight of 27.60 g, root dry weight of 6.75 g, and total yield of 17.93 t/ha. These findings suggest that cultivating the Red Prince genotype along with the application of 75 kg ha⁻¹ of humic acid is highly recommended for achieving optimal production in the specific agro-climatic conditions of Dera Ismail Khan.

Author contributions

Kashif Hussain performed the experimental work. Mehwish Kiran revised the topic several times and provided her suggestions and gave the final approval of the last version to be submitted. Fazal Haq revised the final version and provided his comments focusing on the synthetic routes. Kashif Waseem and Muhammad Ajmad Nadeem revised the topic from the beginning to the end providing his suggestions about the contents and the format. Ghazanfar Ullah and Arshad Farid participated in jotting down the application section. Tariq Aziz provided technical support regarding the discussion part of application.

Conflict of interest

The authors declare no conflict of interest.

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