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Article

Effects of Oxidized Brown Coal Humic Acid Fertilizer on the Relative Height Growth Rate of Several Tree Species

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Abstract: Field experiments were carried out during 2011-2014 at the Research and Experimental Center to Combating Desertification located in the Elsen Tasarkhai station of central Mongolia. The study was aimed to identify the effects of oxidized brown coal humic acid fertilizer on the relative growth rate of several trees for forest rehabilitation. The trees used were *Populus sibirica*, *Salix ledebouriana* and *Acer tataricum*. The experiment was carried out with the concentrations of 2,000, 10,000 and 20,000 mg L^{-1} of humic acid fertilization treatments. The measurement of relative height growth rate (RHGR) was conducted for a period of 4 years. The treatments showed significant differences within humic fertilizer concentrations which differed depending on the species. Compared to the monthly RHGR over the study, the treatment of fertilizers were significantly better for tree growth. *P. sibirica* in the 10,000 and 20,000 mg L^{-1} humic acid fertilizers had significant height growth rates. In addition, when the humic acid treatments were compared to control, *A. tataricum* decreased over the years which were statistically significant for high growth rate and a positive effect of humic acid fertilization treatments was observed. Furthermore, results showed that oxidized brown coal humic acid fertilizers as organic fertilizer can have a positive effect for the growth of *A. tataricum* during the study years. The results showed that the soil chemical properties EC, CO_2, NO_3, and K_2O were significant among all the treatments compared to the control. The effect on P_2O_5 statistically significantly increased among all the treatments. However, pH and Mg were not significant effect among all the treatments. Combining the results obtained from the oxidized brown coal humic acid fertilization with sustainable land management practices can help improve soil organics for environmental issues in degraded sandy soil regions.

Keywords: Forest rehabilitation; humic acid fertilizer; relative height growth rate; soil chemical properties

1. Introduction

Scientists have found that the organic matter plays an important role in controlling the physiochemical properties of soils [1,2]. Residues from the partial oxidation of dead biomass from plants are considered as the source of the organic substances that add to the soils as humus [3,4,5]. Currently, there is a rise in the quantity of humic based products available to plant growers. They come from various sources ranging from hard black coal that are composted plant matter and manure to complex and richer humic sources such as brown coal, peat and leonardite [4,6,7]. Leonardite is medium brown coal that contains the most complex and bio-active form of humic [8]. Humic substances (HS) are the major organic components of the Earth's soils and sediments that were created from decayed bio matter by a process called humification [1]. HS are found especially in high

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concentrations of peat and brown coals [9]. Different grades of coal are formed by geologic compression of soil layers over millions of years. The lower ranks of coal such as lignite and sub-bituminous coals are not efficient as fuel for energy but contain large amounts of organic matter [10]. During humification with the aid of millions of microorganisms, simple products of decomposition like amino acids, carbohydrates and phenols turn into very complex, high molecular weight, long chain organic compounds called humic acids (HA) that are derived from vegetations. This material is very rich and beneficial for plant growth with environment-friendly resources [11]. HA are complex molecules that exist naturally in soils, peats, oceans and fresh waters [12,13]. The best source of HA is the soft sedimentation layers of brown coal called leonardite, an organic material found beneath the earth's surface in the cold climes of the United States, Russia and Mongolia [14]. Often referred to as the oxidized lignite, they are the richest as well as the most economical source of HA, which is readily available from leonardite, a naturally occurring oxidized form of lignite coal [15,16]. HS, the major components of soil organic matter, are the subject of study in different fields of agriculture, which include soil chemistry, fertility, plant physiology and environmental sciences. This is due to the multiple roles played by these substances that has great beneficial effects on plant growth directly or indirectly [4,17,18]. HS are the end products of the organic decomposition of biotic compounds which constitute of a range of 50 to 90% of the organic matter of peat, lignite's, sapropels, non-living organic matter of soil and water ecosystems [19-21]. These substances affect the content and characteristics of soil organic matter and hence play a vital role in the soil's function and structure [22]. These can be useful for living organisms to develop food, enzyme metabolism and substrate material; a carrier of nutrition; as catalysts of biochemical reactions; and in antioxidant activity [23,24,25] for instance argued that HS can both directly and indirectly affect the physiological processes of plant growth.

Commercial products derived from lignite (brown coal), sold mainly as humate preparations, are widely developed as plant growth stimulants which are responsible for the increase in crop yield [3,26]. These products are also claimed to improve key indicators of soil health including soil pH and microbial biomass [27]. Sorption of sulfathiazole besides three structural analogs to leonardie HA was investigated in single and binary solute systems to elucidate the sorption mechanism of sulfonamides to soil organic matter. High affinity cation binding explains absorption and adsorption of polar sulfonamides within crop soils and the strong relationship of adsorption and absorption on soil organic matter content and pH [28]. It is difficult to identify the difference between the direct and indirect effects of these substances. In fact, some of their positive consequences can be assigned to the general enrichment of the soil's fertility, hence higher availability of nutrients [29]. Mongolian hereditary manure has been used for farming experience, but other types of fertilizers used in the neighboring countries are imported. Rapid population growth, desertification and land degradation due to reforestation and agriculture to improve the productivity of locally produced low-cost, environmentally friendly bio-fertilizer use is driven by necessity. In Mongolia, there are several mines and seams of carbon-rich in mineral deposits which are good agricultural grade humic substances [30,31]. A study by the Mongolian Academy of Sciences, Institute of Chemistry and Chemical Technology on the non-energy derived from oxidized brown coal humic fertilizers were used for the experiment. The locally produced brown coal humic soil fertility is useful for forest rehabilitation, impact identification and desirable volume suitable for fertilizing. HA can reduce the use of chemical fertilizers which are the cause of environmental pollution, and also due to lower expenditure of these fertilizers has consequent on lower costs [7,32]. The aim of the recent study was to determine the effects of non-energy derived from oxidized brown coal HS, degraded sandy soil region, on reforestation activity. In reforestation applications, HS has the advantage that they are 100% organic, compatible with sustainable land management practices and help to address environmental issues such as desertification and land degradation. The present study highlights the plant growth promoting oxidized brown coal as an alternative to fertilizers and are also considered environment friendly.

2. Materials and Methods

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2.1. Study area

The study was carried out at the Research and Experimental Center to Combating Desertification Elsen Tasarkhai station (47°27′N, 103°68′E; 1967 m a.s.l), located at the Khugnu-Tarna National Park in Rashaant district of Bulgan province, central Mongolia. The study area has a semi-arid continental climate that is characterized by average annual minimum and maximum temperatures of 22°C and -20, respectively, whereas maximum absolute temperature is 36°C, mean annual precipitation has been reported to be 200-250 mm. The soil of the field was sandy loam with a pH range between 7.80 to 8.92.

2.2. Experimental design and fertilization experiment

This study was conducted in 2011-2014, by the Mongolian Academy of Sciences, (Institute of Chemistry and Chemical Technology) on the non-energy derived from oxidized brown coal humic acid fertilizers which were used for the experiment. The applied humus acid fertilizers with the following characteristics (wt %) were used: carbon (C) 60.5; hydrogen (H) 3.9; nitrogen (N) 0.9; oxygen (O) 34.7, respectively [33]. These concentrations were approximately equivalent to field application rates of control (no humic), 2,000, 10,000 and 20,000 mg L^{-1} oxidized brown coal humic fertilizers m^2 yr^{-1}. The humic acid fertilizers mixed with water were irrigated twice to the experimental site during the growing seasons for four months with a watering can, 100 liters of water with 20, 100 and 200 g of humic acid per solution preparation. Ten liters spray were used for each 1 m^2 area each time with irrigation done in a flooding manner. *P. sibirica*, *S. ledebouriana* and *A. tataricum* species were used for monitoring studies as deciduous trees and shrubs. *P. sibirica* and *S. ledebouriana* are a widely distributed natural plants in Mongolia and are the most widely used plants in the forest rehabilitation [34]. However, *A. tataricum* is a successful naturally occurring plant in Mongolia [35], also being widespread across central and southeastern Europe, temperate Asia [36], from Russia and China. The experimental study size within the research area was (25x25 m), the nurse-tree treatment (2x1.5 m) and the order of planting rows of target tree species were randomized. Plant growth was measured at intervals of 2 times a year and soil analysis was carried out in September of 2014. The soil profile was demarcated according to field morphology and other properties in 0-30 cm identifiable layers. The soil samples were air dried and sieved throught 2 mm mesh size steel sieves. The soil's chemical properties were analyzed at the soil laboratory in the Institute of Geography & Geoecology for electrical conductivity (EC) and potential of hydrogen (pH) in a dS/m^{-1} (1:2.5) [37]. Carbon dioxide (CO_2) was determined through the calcimeter method and organic matter (OM) content using the Walkley-Black method [38]. Assimilable phosphoric acid (P_2O_5) and potassium oxide (K_2O) by the spectrophotometry method [39]. Exchangeable magnesium (Mg) and calcium (Ca) were determined with the atomic absorption spectrometer and the nitrate (NO_3) content was determined by titration method [39].

2.3. Statistical analyses

Since growth may be related to initial tree size at the beginning of the growth period, relative height growth rate (RHGR) was calculated using the following equation. In the first estimator, plant height is averaged before in-transforming, whereas in the second estimator, the height is in-transformed before averaging. The RHGR was considered as it's a type and was computed according to the following:

$$\mathrm{RHGR} = \frac{1n\ HT_2 - 1n\ HT_1}{t_2 - t_1})$$

Where in *HT* is the natural logarithm of seedling height, t is the time (in months), and the subscript refers to initial and final seedling height [40-42]. All data were statistically analyzed by one-way analysis of variance (ANOVA) followed by Duncan's Multiple Range Tests using Statistical Package for the Social Sciences (SPSS) Version 21 (IBM Corp., USA). Statistical significance was accepted at $p<0.05$.

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3. Results

3.1. Relative height growth rate during monitoring period

The number of relative height growth rate (RHGR) showed significantly different for the treatments with increased exposure to brown coal humic fertilizer compared to control treatments (Table 1).

Table 1. Effect of humic fertilization on the relative height growth rate (RHGR) of *Populus sibirica.*

Treatments	Years			
	2011	2012	2013	2014
	Treatment effect (cm month^{-1})			
HA_0	5.62 ± 0.89^{b}	5.32 ± 0.85^{b}	5.99 ± 0.79^{b}	6.57 ± 0.89^{b}
$HA_{0.2}$	8.30 ± 0.63^{a}	7.79 ± 0.60^{a}	9.12 ± 0.60^{a}	9.25 ± 0.63^{a}
HA_{10}	7.98 ± 0.77^{a}	7.70 ± 0.67^{a}	7.23 ± 0.77^{ab}	8.15 ± 0.67^{ab}
HA_{20}	6.24 ± 0.62^{ab}	5.92 ± 0.43^{ab}	7.18 ± 0.67^{ab}	7.61 ± 0.47^{ab}

Data are shown as mean ± SEM; Mean followed by the same letter are not significantly different at 0.05 level. Mean of three different rates of humic addition: control HA_0 (0 mg L^{-1}), $HA_{0.2}$ (2,000 mg L^{-1}), HA_{10} (10,000 mg L^{-1}), and HA_{20} (20,000 mg L^{-1}) of oxidized brown coal humic fertilizers m^2 yr^{-1}, respectively.

The brown coal humic fertilizer treatments marginally affected tree RHGR between 2011 and 2014. Positive RHGR effect was recognized at 2,000 mg L^{-1} and weak RHGR effect at 20,000 mg L^{-1} treatments. Between treatments, HA_{10} (2011-2012) and HA_{20} (2011-2014) were more than most RHGR. In the course of the survey, P. sibirica RHGR range was 5.32±9.25 cm month^{-1} or uneven growth.

The number of RHGR was significantly larger for the treatments with increased exposure to brown coal humic fertilizer as compared to the control treatments (Table 2).

Table 2. Effect of humic fertilization on the relative height growth rate (RHGR) of Acer tataricum.

Treatments	Years			
	2011	2012	2013	2014
	Treatment effect (cm month^{-1})			
HA_0	6.74 ± 0.74^{a}	5.12 ± 0.89^{b}	3.54 ± 0.61^{b}	2.45 ± 0.70^{b}
$HA_{0.2}$	6.53 ± 0.68^{a}	5.49 ± 0.79^{ab}	4.15 ± 0.56^{ab}	3.32 ± 0.85^{b}
HA_{10}	7.10 ± 0.65^{a}	7.48 ± 0.77^{a}	5.59 ± 0.85^{a}	5.61 ± 0.47^{a}
HA_{20}	7.54 ± 0.60^{a}	7.80 ± 0.63^{a}	5.97 ± 0.59^{a}	5.70 ± 0.67^{a}

Data are shown as mean ± SEM; Mean followed by the same letter are not significantly different at 0.05 level. Mean of three different rates of humic addition: control HA_0 (0 mg L^{-1}), $HA_{0.2}$ (2,000 mg L^{-1}), HA_{10} (10,000 mg L^{-1}), and HA_{20} (20,000 mg L^{-1}) of oxidized brown coal humic fertilizers m^2 yr^{-1}, respectively.

The total RHGR of A. tataricum significantly decreased over the years (6.98, 6.47, 4.81 and 4.27 cm month^{-1}, respectively). However, in 2011 the RHGR was not significantly different, in 2012, 2013 and 2014, there was a rapid increase in RHGR for HA_{10} and HA_{20} humic fertilizer treatments. In the course of the survey, A. tataricum RHGR range was 2.45±7.80 cm month^{-1} or uneven growth.

The RHGR was significantly moderate for the treatments with increased exposure to brown coal humic fertilizer as compared to control treatments (Table 3).

Table 3. Effect of humic fertilization on the relative height growth rate (RHGR) of Salix ledebouriana.

Treatments	Years			
	2011	**2012**	**2013**	**2014**
	Treatment effect (cm month^{-1})			
HA$_0$	6.74±0.74^a	5.12±0.89^b	3.54±0.61^b	2.45±0.70^b
HA$_{0.2}$	6.53±0.68^a	5.49±0.79^{ab}	4.15±0.56^{ab}	3.32±0.85^b
HA$_{10}$	7.10±0.65^a	7.48±0.77^a	5.59±0.85^a	5.61±0.47^a
HA$_{20}$	7.54±0.60^a	7.80±0.63^a	5.97±0.59^a	5.70±0.67^a

Data are shown as mean ± SEM; Mean followed by the same letter are not significantly different at 0.05 level. Mean of three different rates of humic addition: control HA$_0$ (0 mg L^{-1}), HA$_{0.2}$ (2,000 mg L^{-1}), HA$_{10}$ (10,000 mg L^{-1}), and HA$_{20}$ (20,000 mg L^{-1}) of oxidized brown coal humic fertilizers m^2 yr^{-1}, respectively.

The highest RHGR increase was observed in HA$_{20}$ in brown coal humic acid fertilizer treatments, between 2011 and 2013. However, HA$_{10}$, HA$_{0.2}$ and HA$_0$ treatments had less effect on RHGR over the study years. There were no significant differences in the treatments within 2014 and in comparison with other years, all treatments had a significant increased RHGR. In the course of the survey, S. ledebouriana RHGR range was 5.87±9.97 cm month-1 or uneven growth.

3.2. Relative height growth rate during monitoring period

The response of plant growth characteristic to the humic acid application and different treatments is presented in Figure 1.

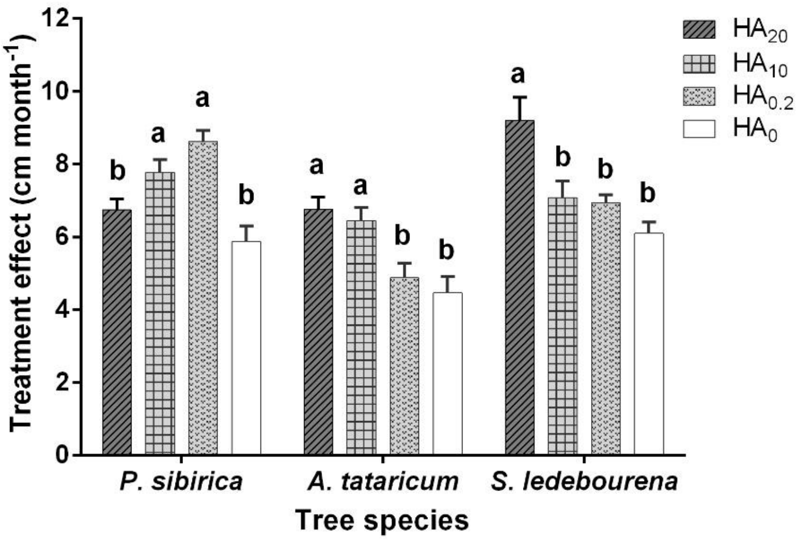

Figure 1. The comparative analysis of different treatment and years with relative height growth rate (RHGR). Humic acid was applied at HA$_{20}$ (20,000 mg L^{-1}), HA$_{10}$ (10,000 mg L^{-1}), HA$_{0.2}$ (2,000 mg L^{-1}) and HA$_0$ (0 mg L^{-1}) oxidized brown coal humic fertilizers m^2 yr^{-1}, respectively. Mean followed by the same letter stand are not significantly different at the 0.05 level. (Duncan's multiple range test).

Results indicate that humic acid treatments significantly increased relative growth rate traits of trees over time. Based on the observed increment, the RHGR mean ratios obtained for P. sibirica, A. tataricum, and S. ledebouriana were 7.25, 5.63 and 7.32 cm month^{-1}, respectively. The interaction effect between oxidized brown coal humic fertilizers with trees RHGR ranging from HA$_{20}$, HA$_{10}$, HA$_{0.2}$ and HA$_0$ treatments for each area evaluated by comparing the highest between S. ledebouriana and A. tataricum alternative, humic is shown to lower the intensity of the norm, P. sibirica HA$_{0.2}$ and HA$_{10}$ scenario has shown the highest intensity value (Fig 1). S. ledebouriana of different RHGR traits

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ranged from 12.6 to 26.8, with the value of HA_0 and $HA_{0.2}$ treatments being the lowest and that of HA_{10} and HA_{20} treatments being the highest.

3.3. Effect of soil chemical properties on fertilization

The effect of oxidized brown coal humic acid fertilization treatments on soil chemical properties was significantly ($p<0.05$) affected by the results of compared to control treatment (Table 4).

Table 4. Chemical properties of brown coal humic fertilizer used in the test estimates within treatments, for soil samples.

Treat	*pH*	*EC*	*CO_2*	*OM*	*NO_3*	*Assimilable*		*Exchangeable*	
						P_2O_5	*K_2O*	*Ca*	*Mg*
HA_0	8.32 ± 0.04^{a}	7.58 ± 2.01^{b}	0.75 ± 0.15^{a}	0.39 ± 0.09^{b}	0.96 ± 0.04^{a}	0.86 ± 0.08^{d}	10.2 ± 2.28^{b}	8.60 ± 0.54^{a}	3.20 ± 0.82^{a}
$HA_{0.2}$	8.25 ± 0.08^{a}	9.92 ± 0.54^{a}	0.45 ± 0.02^{b}	0.41 ± 0.17^{ab}	0.70 ± 0.21^{b}	1.66 ± 0.37^{c}	17.2 ± 1.30^{a}	7.00 ± 1.58^{b}	4.20 ± 0.44^{a}
HA_{10}	8.23 ± 0.08^{a}	11.4 ± 0.05^{a}	0.37 ± 0.01^{b}	0.57 ± 0.10^{a}	0.71 ± 0.16^{b}	2.24 ± 0.66^{b}	17.4 ± 2.19^{a}	8.52 ± 0.86^{a}	3.52 ± 0.86^{a}
HA_{20}	8.23 ± 0.11^{a}	9.92 ± 0.90^{a}	0.40 ± 0.01^{b}	0.42 ± 0.05^{ab}	0.69 ± 0.08^{b}	2.88 ± 0.31^{a}	16.2 ± 1.47^{a}	6.74 ± 1.08^{b}	4.24 ± 1.47^{a}

Data are shown as means ± SEM; pH and EC (1:2.5) (dS/m^{-1}); CO_2 and OM (g kg^{-1} %s.s); P_2O_5, K_2O, Ca and Mg (meq 100 g^{-1}); Mean followed by the same letter stand for not significantly different at 0.05. Mean of three different rates of humic addition: control HA_0 (0 mg L^{-1}), $HA_{0.2}$ (2,000 mg L^{-1}), HA_{10} (10,000 mg L^{-1}), and HA_{20} (20,000 mg L^{-1}) of oxidized brown coal humic fertilizers m^2 yr^{-1}, respectively.

The highest RHGR increase was observed in HA_{20} in brown coal humic acid fertilizer treatments, between 2011 and 2013. However, HA_{10}, $HA_{0.2}$ and HA_0 treatments had less effect on RHGR over the study years. There were no significant differences in the treatments within 2014 and in comparison with other years, all treatments had a significant increased RHGR. In the course of the survey, S. ledebouriana RHGR range was 5.87±9.97 cm month-1 or uneven growth.

4. Discussion

The findings from this study suggested that humic acid fertilizer concentrations had a significant effect on all growth traits with the treatments of HA_{20}, HA_{10}, $HA_{0.2}$, and HA_0. The monthly RHGR during the study years showed that plants treated with humic fertilizer treatments were significantly better. In other related experimental studies, HA application increased the growth rate of planted tree seedlings [43-46]. The cumulative effect was the enhanced growth of plants and an increased yield of dry matter [47,48]. The arouse of root growth in most cases is more visible than the stimulation of shoot growth [46,49]. Studies on the effects of HS on plant growth, under conditions of adequate mineral nutrition, consistently show positive effects on plant biomass [13,47]. Overall, random effects meta-analysis estimated shoot dry weight increase of 22% and root dry weight increase of 21% in response to HS application [3]. This study also showed that HA fertilization treatments when compared to control, *A. tataricum* growth rate significantly decreased over the years. HS affected most plant metabolic processes, nevertheless their source, they controlled enzymatic systems related to primary, secondary, and defense metabolisms as a recation to environmental stress [49,50]. The results of this study is consitent with whereby humic matter affected plant growth and development via different metabolic pathways, hormonal, carbon and nitrogen metabolisms and stress response [29,46,51]. These results are quite useful, especailly to the field of agronomic use of HS because soil weathering, climate change, limited water and nutrient resources are becoming increasingly important challenges to agricultural production, and guidnce for using HS are often directed at alleviating these stress [19,49,52]. The height growth rate values of *P. sibirica* in HA_{10} and $HA_{0.2}$ treatments were similar to those in the HA_{20} treatments, and the $HA_{0.2}$ treatments had significant height growth rates. In the related study, typical response curve showed increased growth with increasing HS concentration in nutrient solutions, followed by a decrease in growth at very high concentrations [13,53]. The typical response curve showed increasing growth with increasing HS

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concentration in nutrient solutions, followed by a decrease in growth at very high concentrations [25,54].

In the present study, the treatments of $HA_{0.2}$,HA_{10}, HA_{20}, significantly increased the soil's EC, and K_2O, whereas the CO_2 and NO_3 contents were significantly reduced as compared to the control. This results are consistent with previous study reports on soil physicochemical properties; in particular, soil organic matter content and nutrient concentrations increased [32,46,49,55]. P_2O_5 was statistically different and increased among all the treatments. In the related study, phosphorus concentrations increased with increasing levels of humic acid regardless of the yield response [32,45,56]. The effect of phosphorus absorption was the opposite of that relating to nitrogen; absorption increasing with higher doses [57]. HA and phosphorus applications increased the growth parameter of plants [53]. HA technically is not a fertilizer,eventhough, in some studies, people do account for it [7]. In some instances, the usage of fertilizers could be stopped at once if there is enough organic material and the soil can be fully dependent on microbial processes and humus production to sustain it's self [9,49,58].

5. Conclusions

Our study revealed a positive effect of oxidized brown coal humic fertilizers on plant growth even though the humic fertilizer concentrations may differ depending on the species. Planted tree growth treated with HS was compared with RHGR test. Specifically, *A. tataricum* decreased over the years which were statistically significant for high growth rates. However, there was a variety of impacts on growth due to adaptability to tree species and ecological environment. The experiment site soil with a very high sodium content (pH of 8.23 to 8.32) and Mg, was significantly not-changed for HA fertilization treatments. Nonetheless, the increase in the assimilable K_2O, P_2O_5 and OM may have positively affected plant growth. Findings from this experiment show that the application oxidized brown coal humic fertilizer can positively affect the RHGR of planted trees and some adaptability traits related to it. Therefore, further studies would be needed for long term monitoring, which includes species of trees and soils are necessary for specific views in different ecological conditions.

Supplementary Materials: The code used and data files produced in this study are available online at www.mdpi.com/....-..../././s1.

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