The Effect of Multiple Environmental Conditions on Acacia Senegal Seedling's Carbon, Nitrogen, and Hydrogen Contents: An Experimental Investigation

Abdoelmoniem A. Attaelmanan, Ahmed A. H. Siddig

Abstract**—**This study was conducted in light of continual global climate changes that projected increasing aridity, changes in soil fertility, and pollution. Plant growth and development largely depend on the combination of availing water and nutrients in the soil. Changes in the climate and atmospheric chemistry can cause serious effects on these growth factors. Plant carbon (C), nitrogen (N), and hydrogen (H) play a fundamental role in the maintenance of ecosystem structure and function. Hashab (*Acacia senegal*), which produces gum Arabic, supports dryland ecosystems in tropical zones by its potentiality to restore degraded soils; hence, it is ecologically and economically important for the dry areas of sub-Saharan Africa. The study aims at investigating the effects of water stress (simulated drought) and poor soil type on *Acacia senegal* C, N, and H contents. Seven-day-old seedlings were assigned to the treatments in split-plot design for four weeks. The main plot is irrigation interval (well-watered and water-stressed), and the subplot is soil types (silt and sandy soils). Seedling's C%, N%, and H% were measured using CHNS-O Analyzer and applying Standard Test Method. Irrigation intervals and soil types had no effects on seedlings and leaves C%, N%, and H%, irrigation interval had affected stem C% and H%, both irrigation intervals and soil types had affected root N% and interaction effect of water and soil was found on leaves and root's N%. Application of well-watered irrigation with soil that is rich in N and other nutrients would result in the greatest seedling C, N, and H content which will enhance growth and biomass accumulation and can play a crucial role in ecosystem productivity and services in the dryland regions.

Keywords**—***Acacia senegal*, Africa, climate change, drylands, nutrients biomass, Sub-Sahara, Sudan.

I. Introduction

THE amalgamation of soil moisture and mineral nutrients is the major limiting growth factor on which plants mostly depend on for their growth, fitness and nutrient biomass accumulation. Nevertheless, climatic changes and atmospheric carbon concentration can have serious effects on these growth factors [1]. Global climate changes such as changed precipitation patterns and temperature will result in water scarcity and worsening land degradation [2]. Africa has been identified as one of the most vulnerable parts of the world to the impact of climate change [3], [4].

In particular, sub-Saharan region is the most ecological diverse and developing part that experienced extensive climatic changes [5]. According to the UN Department of Economic and Social Affairs [6], the population size of sub-Saharan Africa will reach 2 billion people in 2050 with greatest population growth ratio constituting the largest proportion of people living below the poverty line worldwide [7]. About one in five people (21% of the population) was facing hunger in Africa in 2020 – more than double the proportion of any other region in the world. Moreover, the situation in sub-Saharan Africa is worse with one in four people (24% of the population) affected by hunger [8].

Acacia senegal, as N_2 fixing species, has the ability to restore poor soil by adding nitrogen; hence, it is to be used for traditional intercropping in drylands of sub-Saharan Africa playing a crucial role in socio-ecological resilience in the region [9]. *A. senegal* is a leguminous multi-use species that produces gum Arabic which is economically and ecologically very important, as it has many different uses in addition to supporting the people and the state [10], [11].

Plant nutrient biomass, such as carbon, nitrogen and hydrogen, has a crucial role in ecosystem processes. Carbon (C) is major part of a chemical components of photosynthesis reactions and biochemistry; nitrogen (N) is the main part of nucleic acids and proteins of plant's cell [12]. Plant development depends mostly on C and N metabolism though, stoichiometry of C and N can be used for prediction of plant growth in era of changes [13], [14]. Therefore, investigation of plant C%, N% and C/N is a center of attention to evaluate and monitor forest ecosystem adaptation to changes. On the other hand, and according to recent studies, e.g. [15], [16], plant hydrogen (H) of trees is a part of the hormone processes that can lead to enhancing plant adaptation to climatic extreme events such as aridity, frost, salinity and heavy metals.

For many regions, the climatic scenarios projected that soil moisture content and fertility will be decreased and changed [17]. For instance, and at universal level, soil chemical and physical properties are to be degraded due to changes in land utilization, excessive and unorganized use of fertilization and N deficiencies [18]. Within these continual global climatic changes, it is important to understand how plants will deal and evolve with the most important limiting growth factors that are projected to be changed. Also, better knowledge about the effects of multiple environmental drivers in the growth of

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important dryland species such as *A. Senegal* should be of great significance to forest rehabilitation and restoration planning. Therefore, the study aims to investigate the effects of water stress and poor soil on *A. senegal* nutrients biomass of the percentages of C, N and H.

II. Methodology

A. Study Site and Settings

The experiment was conducted in the nursery as split-plot design with the water interval as main plots, and the soil types as subplots. The nursery of the Faculty of Forestry, University of Khartoum, is located at the Shambat Campus, Khartoum North, Sudan (15° 40' 5" North, 32 32' 1" East). Shambat has a subtropical desert and sub-humid climate with low-latitude. The irrigation intervals were well-watered and water stressed (200 ml/every day and 200 ml/every two days, respectively), while the soil treatments were rich and poor soils (silt with C% 0.26, N% 0.046 and C/N 5.7; sand with C% 0.24, N% 0.0226, C/N 10.6, respectively).

Bulk seeds were collected from El-Damazeen forests. They were obtained from the National Tree Seed Center. Then seeds were germinated in polymer bags of 10 cm × 20 cm filled with silt or sandy soils and irrigated daily by 200 ml of water. After 7 days (one week), 60 seedlings (30 of silt and 30 of sand) were selected with minimum morphological variations, then assigned randomly into six experimental plots. Three blocks of 1 m × 2 m plots were prepared and each one was divided into two subplots for irrigation interval treatments either well-watered (200 ml/every day) or water stressed (200 ml/every two days). Then five seedlings from those raised in silt soil and five from sandy soil were assigned randomly to each of the subplot for four weeks.

B. Measured Variables

Seedlings were harvested after four weeks from the start of the experiment and separated into leaves, stems and roots then converted into powdered-dry matter. The seedlings and its compartments C, N and H percentage content were determined using CHNS-O Analyzer and applying Standard Test Method (ASTM International, model D 5291-02. 2002, USA) for instrumental determination of carbon, nitrogen, and hydrogen contents for plants' samples.

C. Data Analysis

The Analysis of Variance (ANOVA) procedures and Duncan's Multiple Range Test to separate means of the same factor at significance were carried out using Statistical Analysis Software (SAS). The model is a split-plot with three blocks: irrigation interval is the main plot and soil type is the subplot within each block: Y (dependent variable) = B (block effect) + W (irrigation interval) + B*W + S (soil type) + W*S.

III. Results

A. Effects of Irrigation Intervals and Soil Types on Seedling's C%, N% and H%

Irrigation intervals and soil types had no significant effects on seedling's C, N and H% (Table I).

TABLE I
Effects of Irrigation Intervals and Soil Types on Seedling's C%, N% and H%

Treatments	C%	N%	H%
Irrigation interval			
Well-watered	41.3(5.7)	2.9(0.6)	5.8(0.4)
Water stressed	42.5(5.6)	3(0.6)	6.2(0.4)
P value	**0.73**	**0.73**	**0.36**
Soil type			
Silt	42.7(7.3)	3(0.8)	6.1(0.7)
Sand	41.1(6)	2.9(0.5)	5.9(0.4)
P value	**0.65**	**0.93**	**0.58**

B. Effects of Irrigation Intervals and Soil Types on Leaves' C, N and H%

Irrigation intervals and soil types had not significantly affected leaves' C, N and H% (Table II). However, a significant interaction effect of irrigation intervals and soil types was found on leaves' N% (P = 0.039). But the interaction effect on leaves' C% was slightly insignificant (P = 0.059).

On one hand, the leaves' N% dropped significantly by 10% from well-watered to water stressed in silt soil (P = 0.05) and it increased by about 8% from well-watered to water stressed in sand soil but not significantly. On the other hand, the leaves' N% dropped significantly by 10% from silt to sandy soils in well-watered (P = 0.05) and it increased by about 7% from silt to sandy soils in water stressed but not significantly (Table III; Fig. 1).

TABLE II
Effects of Irrigation Intervals and Soil Types on Leaves' C%, N% and H%

Treatments	C%	N%	H%
Irrigation intervals			
Well-watered	41.55(1.5)	4.5(0.3)	6.16(0.2)
Water stressed	41.8(1.9)	4.44(0.4)	6.33(0.3)
P value	**0.66**	**0.84**	**0.08**
Soil types			
Silt	41.48(1.6)	4.51(0.2)	6.28(0.3)
Sand	41.96(2.2)	4.44(0.4)	6.22(0.3)
P value	**0.53**	**0.79**	**0.48**

TABLE III
Interaction Effects of Water and Soil Factors on Leaves' N%

Levels	Silt	Sand	P value
Well-watered	4.7 (0.2)	4.26(0.4)	**0.05**
Water stressed	4.28(0.2)	4.61(0.4)	**0.38**
P value	**0.05**	**0.36**	

C. Effects of Irrigation Intervals and Soil Types on Stem's C%, N% and H%

Irrigation intervals had significantly affected stem's C% and H% but not N% (P = 0.022 and 0.003, respectively). Hence, well-watered had increased C content by about 32% over that irrigated with water stressed, while, well-watered had resulted in higher stem's N% (40%) than that of water stressed.

Soil types had no significant effects on the stem's C%, N% and H%. However, soil types had slight insignificant effects (P

= 0.06) on the stem's N%, as it increased by about 33% in silt than in sandy soil (Table IV).

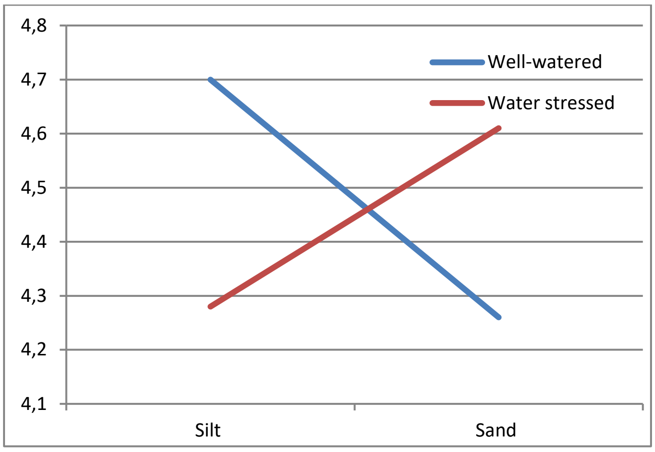

Fig. 1 Interaction effect of water and soil factors on leaves' N%

TABLE IV
EFFECTS OF IRRIGATION INTERVALS AND SOIL TYPES ON STEM'S C%, N% AND H%

Treatments	C%	N%	H%
Irrigation interval			
Well-watered	45.66(5.1)	2.67(0.5)	6.50(0.3)
Water stressed	30.88(8.5)	1.75(0.9)	3.94(0.7)
P value	**0.022**	**0.09**	**0.003**
Soil type			
Silt	40.70(9.2)	2.76(0.8)	5.37(0.7)
Sand	35.82(2.1)	1.84(0.7)	5.06(0.4)
P value	**0.29**	**0.06**	**0.486**

D. Effects of Irrigation Intervals and Soil Types on Root's C%, N% and H%

Irrigation interval had significantly affected root's N% while, it had no effects on root's C% and H% (P = 0.05; Table V). Accordingly, the root's N% of well-watered was higher (about 27%) than that of water stressed.

Soil type had significant effects on the root's N% (P = 0.02), however, it had no effects on root C% and H%. Hence, the root N% in slit soil was higher (about 35%) than that in sand soil (Table V). Nevertheless, the interaction effect of water and soil factors was found on the root's N% (P = 0.05).

TABLE V
EFFECTS OF IRRIGATION INTERVALS AND SOIL TYPES ON ROOT'S C%, N% AND H%

Treatments	C%	N%	H%
Irrigation interval			
Well-watered	40.1(15)	2.33(0.6)	1.85(0.4)
Water stressed	39.7(8)	1.71(0.6)	2.33(0.4)
P value	**0.96**	**0.05**	**0.12**
Soil type			
Silt	39.8(8)	2.47(0.8)	2.47(0.7)
Sand	40(15)	1.60(0.5)	1.72(0.4)
P value	**0.98**	**0.02**	**0.07**

On one hand, root's N% insignificantly decreased (about 5%) from well-watered to water stressed in silt soil but it dropped significantly (54%; P = 0.022) in sand soil. On the other hand, the root's N% insignificantly decreased (14%) from silt to sandy soil in well-watered, however, it dropped significantly (58%) from silt to sandy soil in water stressed (P = 0.004; Table VI; Fig. 2).

TABLE VI
INTERACTION EFFECT OF WATER AND SOIL FACTORS ON ROOT'S N%

Levels	Silt	Sand	P value
Well-watered	2.55(0.4)	2.19(0.2)	**0.542**
Water stressed	2.41(0.6)	1.01(0.5)	**0.004**
P value	**0.772**	**0.022**	

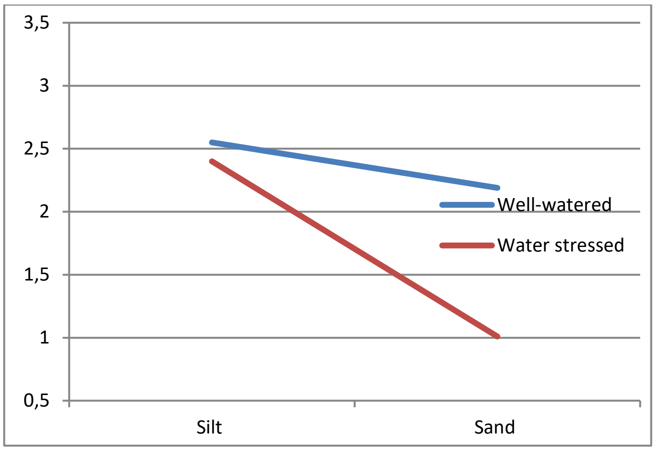

Fig. 2 Interaction effect of water and soil factors on root's N%

IV. DISCUSSION

The results of our study (e.g., leaves' and total seedling's C, N and H, stem's N% and root's C contents) are partially in line with [19] and [20] which concluded that most plant species have flexibility to take up nutrients under conditions of unbalanced nutrients availability. In contrast, recent field experiments showed that dominant species (*Paspalum dilatatum*) are the most sensitive species to water stress but can still increase their C and N biomass contents [21], [22]. The responsive results of our study show that, stem C, H% and root N% to water and soil treatments are in agreement with [14] who reported that plant C and C:N ratios in both shoots and roots increased with increasing soil fertility and decreased with increasing aridity. However, the results are in contrast with finding that the seedling's nutrients biomass was not affected by other environmental factors because the basic molecular mass and structure of the light harvesting chlorophyll–protein are genetically determined [23]. The positive interaction effect of water and soil on leaves' and root's N content are in line with [24] and [25] which concluded that irrigation water dissolved the nitrogen and made it available to plants, subsequently influencing on all growth parameters significantly.

The *Acacias* are important C_3 and Savanna trees species dominating sub-Saharan Africa and are suffering from ongoing anthropogenic and climate-mediated degradation [26], [27]. For instance, *A. senegal* has an important role in restoring these eroded fields due to the tree's ability to associate with N_2-fixing bacteria that improve the nutrient-depleted soil. Carbon stocks of African drylands also have declined because of land degradation and loss of soil fertility/water retention while planting acacia trees in these areas could considerably increase soil organic carbon stocks [28]. Changing or altering of *A. senegal* C, N and H content can cause a dangerous effect on ecosystem functions and services and overall resilience in this fragile region in Africa [29]. Also, *A. senegal* is a multi-purpose tree producing gum Arabic, a high-value export commodity dominantly from Sudan and some African countries. After all, affecting the stoichiometry of the tree can reduce the commodity's production and/or quality.

Finally, *Acacias* play an especially important role in rehabilitating sandy or clay soils. By fixing N_2, *A. senegal* improves the soil, which permits crop growth in restored gum Arabic agroforestry systems of sub-Saharan Africa [30]. From the above, it could be concluded that planting *acacias* in soil that is rich in N_2 and probably other nutrients with applying well irrigation would result in greatest tree growth and biomass accumulation (i.e., C from atmosphere; N from soil and atmosphere) which can play an important role in ecosystem function and services in this era of global changes. These findings give insights on the responses of key growth traits of dryland tree species to global change from a single tissue now to the whole community and ecosystem in the future.

V. Conclusion

- Water stress/simulated drought has partially affected stem's C and H% and root N% but no other nutrients' biomass.
- Soil types have partially affected root N% but not others.
- However, the combined effects, i.e., the interaction of water and soil, seem to have an effect on leaves' and root's N%.

Nevertheless, the study findings have been confounded by some issues including sample size, duration and design of the experiment. Furthermore, studies on the effect of these factors (i.e., soil type and water stress) under elevated carbon dioxide and/or elevated temperature will give a better picture about responses of acacia seedling's biomass and nutrients budget. Finally, long-term experiments on *A. senegal* are recommended to evaluate the effects these factors on different growth parameters under different global change scenarios.

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