



Original Article

Nutrient Solution on Aloin Content and other Quality Characteristics of *Aloe vera*

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Abstract

One of the main constraints in *Aloe vera* production is poor information about optimum nutrients that are helpful for growth and production. The objective of this study was to optimize nutrient solution for *Aloe vera* cultivation in soilless culture. Therefore, to determine the best nutrients the experiment was conducted in a greenhouse during 2011 in the Agricultural Faculty, University of Guilan, Rasht, Iran (37 °16 'N). Experimental design was a completely randomized with three replications and each replication contained ten pots. Aloe (*A. vera* L.) sprouts were irrigated with nutrient solution containing different level of NO₃, NH₄ and potassium starting from March and harvesting took place during September. The research indicated that nutrient solution with the highest level of nitrogen increased *Aloe* vegetative growth without any negative effect on qualitative indices including Aloin, total phenol, total antioxidative activity and element contents; it was possible to produce the highest level of vegetative growth in *Aloe vera*.

Key words: Soilless culture, Phenol, Antioxidant, Protected cultivation

Introduction

Aloe vera belonging to the family Asphodelaceae [1], is one of a few Aloe species that has been explored by pharmaceutical and cosmetics industries [2,3]. Biological activities are ascribed to the pulp gel of *A. vera* such as antiviral, antibacterial, antifungal, anticancer, wound healing, and many other characteristics [4]. Those characteristics have prompted industrial and commercial increase in the production of *A. vera* throughout the world.

Hydroponic systems include all systems that deliver the nutrients in a liquid form, with or without an aggregate medium to anchor the plant roots [5]. Various medicinal crops were observed in different hydroponic systems. Production of medicinal plants in controlled environments provides opportunities for improving the quality, purity, consistency, bioactivity, and biomass production of the raw

material [5]. Hydroponic systems in controlled environments can produce high quality medicinal plant free from accidental adulteration by weeds, soil or environmental toxins such as heavy metals in soils. In some species it may be possible to optimize for higher yields of specific secondary metabolites or for higher yields of target organs [5]. Application of nitrogen (N) and phosphorus (P) fertilizers increased the yield of *A. vera* [6,7]. Nitrogen is essential for plant growth. As a macro-element, it is part of protein structure and participates in metabolic processes and energy transfer. It is absorbed by the plant as ammonium (NH₄⁺) or nitrate (NO₃⁻) ions. The form in which N is absorbed is, in part, dependent on pH [8]. In hydroponics, nitrate and ammonium forms are used in nutrient solutions. A balance between ammonium and nitrate favors plant growth and that the degree of benefit varies among crops [9].

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For most plant species, NO_3^- supply combined with low quantities of NH_4^+ favors growth, but the response depends on the species and the age of the plant. Mengel and Kirkby [9] reported plant species grow better when nitrogen is administered as NO_3^- instead as NH_4^+ . Other reports have indicated that the incorporation of nitrogen in N-NH_4^+ form is toxic for many species, even in low concentrations [10]. In other hands the yield depends to a large degree on the content of photosynthetically active pigments. Authors of numerous papers showed a close correlation between the level of these pigments and nitrogen content in leaves determined by the dose and time of fertilization [11-14].

Little work has been done on the cultivation of *A. vera* in the soilless systems. One of the main constraints in this production chain is to obtain suitable nutrient solution with the highest yield and quality. If *Aloe vera* has cultivated in soilless with poor nutrition, it is difficult for *A. vera* to grow in this system without nutrient amendments. Our previous research [5] indicated that nutrient solution with the highest level of nitrogen increased *Aloe* vegetative growth. The objective of this study was to determine nutrient solution with the highest vegetative growth on *Aloe* quality to optimized nutrient solution to reach the highest yield and quality in soilless culture of *Aloe*.

Material and Methods

The experiment was conducted in a greenhouse during 2011 in the Agricultural Faculty, University of Guilan, Rasht, Iran ($37^\circ 16' \text{N}$). Experimental design was a completely randomized design with three replications and each replication contained ten pots. *Aloe* (*A. vera* L.) sprouts were cultured in pot with 29 cm diameter containing cocopeat and perlite (50:50 v/v) and irrigated with nutrient solution (Table 1) containing different level of NO_3^- ,

NH_4 and potassium on March and harvesting took place on September. All nutrient solution contained 0.05, 1.5, 2, 0.25, 1 and 10 $\text{mg}\cdot\text{L}^{-1}$ $(\text{NH}_4)_6\text{Mo}_7\text{O}_2/4\text{H}_2\text{O}$, H_3BO_3 , $\text{MnSO}_4/4\text{H}_2\text{O}$, $\text{CuSO}_4/5\text{H}_2\text{O}$, $\text{ZnSO}_4/7\text{H}_2\text{O}$, and sequestrene Fe 136 respectively. Stock solutions were used to produce treatment solutions with different nitrogen and potassium levels and different ratios of ammonium to nitrate (Table 1). Temperatures during the experiment were $25 \pm 3^\circ \text{C}$ during the day and $20 \pm 2^\circ \text{C}$ during the night.

Ascorbic acid was quantitatively determined according to the 2, 6-dichloro-phenolindophenol dye method [15]. Ascorbic acid was extracted by grinding 10 g of fresh sample in a mortar with pistil and 3% metaphosphoric acid (v/v) as a protective agent. The extract was made up to a volume of 100 mL and centrifuged at 3000 g for 15 min at room temperature. Ten mL were titrated against 2, 6-dichlorophenolindophenol dye, which had been standardized against ascorbic acid. Phosphorus, calcium and magnesium were measured by spectrometry (JENWAY 6105 U.V/V) [16]. K was determined by flame photometer [17,18]. One g of dry matter was burned to produce ash at 550°C for 6 h [19]. The methanol extracts of *Aloe* were used for the determination of total phenolics. Total phenolic content was evaluated by colorimetric analyses using Folin-Ciocaltaue's phenol reagent [20]. The content of total phenolics was expressed as mg galic acid equivalent per 100 g of fruits. The free radical-scavenging activity against DPPH radical was evaluated according to the method of Leonge and Shui [21] and Miliouskas *et al.* [22] with minor modification. According to principle of this method, in the presence of an antioxidant, the purple color intensity of DPPH solution decays and the change of absorbance are followed Spectrophotometrically at 517 nm. The scavenging activity was expressed as IC50 (mg/ml).

Table 1 Macronutrients used in nutrient solutions.

Solution	$\text{meq}\cdot\text{L}^{-1}$										
	KNO_3	KH_2PO_4	NaCl	CaNO_3	MgSO_4	NH_4NO_3	K	NH_4^{a}	NO_3^{b}	N^{c}	Total salt ^d
1	3.2	3.3	0.2	5.2	1.5	0.1	4.6	0.1	8.5	8.6	13.5
2	3.8	3.3	0.2	5.2	1.5	0.1	5.2	0.1	9.1	9.2	14.1
3	4.4	3.3	0.2	5.2	1.5	0.1	5.8	0.1	9.7	9.8	14.7
4	2.6	3.3	0.2	5.2	1.5	0.1	3.8	0.1	7.9	8.0	12.9

^a total concentration of NH_4 in salts in solutions.

^b total concentration of NO_3 in salts in solutions.

^c Nitrogen equivalent as sum of NO_3 and NH_4 .

^d refers to salt concentrations in nutrient solution presented in the first 6 columns containing compounds.

Aloin content was measured with high performance liquid chromatography method [23]. The analytical column was a II 5-C18 RS WK (4.6 mm × 250 mm) filled with a 5 microns stationary phase. The mobile phase consisted of methanol-water (95:5 and 5:95); the flow-rate was 1 mL.min⁻¹. The injection volume was 5 micromole. The DAD detector was set at 297 nm. The calibration curve was linear over the range of 0.17-5.9 micrograms ($r = 0.9999$). The average recovery of the method was 98.6%, RSD 1.32% ($n = 6$).

Data for each parameter were subjected to one-way ANOVA and significant differences between treatment means were determined by Tukey test using the SAS software package (9.2).

Results and Discussion

Nutrient solution did not affect all measured qualitative characteristics (Table 2) while our previous research indicated that nutrient solution with the highest level of nitrogen increased *Aloe* vegetative growth [5]. Therefore, without any negative effect on qualitative indices we are able to produce the highest level of vegetative growth in *Aloe vera*. In other hand we may be possible to optimize for higher yields of target organs as mentioned previously by Saliqedar *et al.*, [5].

In fact application of nitrogen fertilizers increased the yield of *A. vera* [6-7] without any negative effect on qualitative indices including Aloin, total phenol, total antioxidative activity, element content and other measured characteristics (Table 2) indicating that Aloin quantity per hectare will increase significantly. The antioxidant activity of *Aloe vera* was 33.64-37.33%, which is similar to the one reported by Narsih *et al.*, [24], who found the antioxidant activity of their sample was 37.2-86.6%. In hydroponics, nitrate and ammonium forms are used in nutrient solutions. A balance between ammonium and nitrate favors plant growth and that the degree of benefit varies among crops [9] but it seems that these ions ratio did not have any positive or negative effect on *Aloe vera* quality.

Aloin also known as barbaloin is a bitter, yellow-brown colored compound noted in the exudates of at least 68 *Aloe* species at levels from 0.1 to 6.6% of leaf dry weight (making between 3% and 35% of total exudates) and in another 17 species at indeterminate levels [25]. By increasing NO₃ in nutrient solution Aloin content increased to 40.6 and 45.96% by nutrient solution 2 and 3 respectively while the differences between treatments was not significant.

Table 2 Effect of nutrient solution on measured characteristics

Nutrient solution	Antioxidant	Total Phenol	Vit. C	pH	TSS	Aloin	Ash
1	33.64 ^a	9.92 ^a	4.99 ^a	4.46 ^a	0.92 ^a	33.64 ^a	0.36 ^a
2	34.57 ^a	8.29 ^a	5.88 ^a	4.46 ^a	0.90 ^a	40.60 ^a	0.33 ^a
3	36.97 ^a	6.00 ^a	4.67 ^a	4.52 ^a	1.02 ^a	45.96 ^a	0.40 ^a
4	37.33 ^a	7.78 ^a	4.27 ^a	4.45 ^a	0.95 ^a	26.67 ^a	0.34 ^a

Numbers followed by same letters in each column are not significantly different according to the Tukey test ($P \leq 0.01$)

Table 2 Continue

T	N	K	P	Ca	Mg
1	0.95 ^a	3.48 ^a	0.12 ^a	3.56 ^a	1.89 ^a
2	1.05 ^a	4.26 ^a	0.08 ^a	3.12 ^a	1.44 ^a
3	1 ^a	4.18 ^a	0.13 ^a	3.8 ^a	1.80 ^a
4	1.25 ^a	4.5 ^a	0.15 ^a	3.72 ^a	1.58 ^a

Numbers followed by same letters in each column are not significantly different according to the Tukey test ($P \leq 0.01$)

Conclusion

Little work has been done on the cultivation of *A. vera* in the soilless systems. One of the main constraints in this production chain is poor knowledge about optimum nutrient solution best

suitable for its growth and production. Our research optimized nutrient solution for *A. vera* cultivation in soilless culture using nutrient solution containing 9.7 meq·L⁻¹ NO₃ and 5.8 meq·L⁻¹ K which produce the highest vegetative growth without negative effect on *A. vera* quality.

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