COMPARISION OF IN VITRO AND EX VIVO EXPERIMENTAL ANALYSIS OF ALOE VERA EXTRACTS ON THE ANTIOXIDANT AND ANTIMICROBIAL ACTIVITY

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ABSTRACT

Medicines generally found in market are prepared from the gel and from the juice. In this study, *Aloe* explants were selected and propagated through *in vitro* propagation. The efficiency of various solvents such as ethanol, water and acetone extract were analysed in this study. *In vitro* and *ex vivo* samples of succulent plants were compared for disease resistance and anti-oxidant activity. It was found that the *in vitro* explants possess elevated disease resistance and higher concentrations of anti-oxidants. Thus it can be used as an alternative to those cosmetics and medicines made out of *aloe barbadensis*.

Key words: Aloe Vera, Ex vivo, In vitro, Antioxidant property, anti-microbial property.

1. INTRODUCTION

Aloe Vera is a unique plant of Genus aloe, which is rich source of many chemical compounds. Aloe Vera gel contains powerful antioxidants belong to a large family of *polyphenols* helps to inhibit the bacterial growth causing human infections [1]. The processed Aloe Vera gel is used in preparation of Drugs, Cosmetics and in Healthcare product [2]. *Aloe Vera* contains powerful antioxidant nutrients, presence of A, C, and E vitamins are free radical components helps in

getting rid of the toxins [3]. Now a day's gaining resistance against antibiotics by bacteria is becoming a serious issue. The gel extracted from *Aloe Vera* leaf proofs to inhibit the Gram-positive bacteria growth such as *Shigella flexneri* and *Streptococcus progenies* [4]. Also, specific plant compounds such as anthraquinones and dihhydroxyanthraquinones as well as saponins as a phytochemical have been found to perform direct attack on the microbes in the body and many research papers suggest its role in providing immunity [5,6]. In this study we multiply the identified Aloe species through in vitro propagation and we aimed to compare the antioxidant and anti-bacterial activity of the in vitro and ex vivo aloe plants.

Fig: 1 *Aloe Vera* (*Aloe barbadensis*)

2. MATERIALS AND METHODS

Explants were collected from Genewin Biotech and Maintained in green house. Explants were collected from healthy mature aloe Vera plant and for 10 minutes washed thoroughly in running tap water to eliminate muddy particles from explants. New bud Explants were soaked in antifungal, antibacterial, carbendazim and streptocycline (0.1%) for 15 minutes for sterilization. Then the explants were wiped with sterile cotton containing 70% ethanol. Finally explants were treated using detergent (Tween 20 for 20 min), Then for complete removal of foam washed with sterile water. The explants collected were soaked in 10% Sodium hypochlorite for 40 min and the first layer was removed. Again the explants were soaked in 10% NaOCl for 40 min and the second layer was removed. The further sterilization was carried out inside laminar air flow

chamber and 2 sets of Experiments were performed. In Set 1 Mercuric chloride (0.1%) were used as surface sterilant. After removing the explants from the mercuric chloride solution and washing with sterile water 3 times (for eliminating the toxic effects of Mercuric chloride) Sodium hypochlorite (NaOCl) solution was used as surface sterilant in second set. In each trials in 3-4 weeks bud break was recorded.

3. RESULTS AND DISCUSSION

QUANTITATIVE ANALYSIS

The quantitative analysis of invitro and exvivo was done for comparing the antioxidants such as flavanoids and DPPH.

I. TEST FOR TOTAL PHENOLIC CONTENT

Table 1: Table showing ex vivo activity of total phenolic content test

ex vivo

S.No	Samples	Acetone extract (mg/ml)	Ethanol extract (mg/ml)	Water extract (mg/ml)
1.	Aloe leaf	40	51	46
2.	Aloe gel	40.1	46.8	49.2

Table 2: Table showing In vitro activity of total phenolic content test

In vitro

S.No	Samples	Acetone extract (mg/ml)	Ethanol extract (mg/ml)	Water extract (mg/ml)
1.	Aloe leaf	49.5	53	49
2.	Aloe gel	59.8	55	51.3

II. TEST FOR FLAVANOID CONTENT

Table 3: Table showing ex vivo activity of flavanoid test

ex vivo

S.No	Samples	Acetone extract (mg/ml)	Ethanol extract (mg/ml)	Water extract (mg/ml)
1.	Aloe leaf	24	22	23.6
2.	Aloe gel	20	22	21

Table 4: Table showing In vitro activity of flavanoid test

in vitro

S.No	Samples	Acetone extract (mg/ml)	Ethanol extract (mg/ml)	Water extract (mg/ml)
1.	Aloe leaf	32	39.2	26
2.	Aloe gel	20.6	22.4	25

III. TEST FOR DPPH FREE RADICAL SCAVENGING ASSAY

Table 5: Table showing Ex vivo activity of DPPH assay

ex vivo

S.No	Samples	Acetone extract (%)	Ethanol extract (%)	Water extract (%)
1.	Aloe(leaf)50µl	22	14.20%	17.80%
2.	Aloe(Leaf)100µl	55.70	16.50%	15.50%
3.	Aloe(leaf)150µl)	55.20	17.90%	19.99%
4.	Aloe(leaf)200µl	44.60	15.10%	22.30%
5.	Aloe(gel)50µl	24.90	28.10%	17.80%
6.	Aloe(gel)100µl	16.60	20%	24.20%
7.	Aloe(gel)150µl	14.10	33.40%	17.70%
8.	Aloe(gel)200µl	17.20	15%	16.30%

Table 6: Table showing In vitro activity of flavanoid test

in vitro

S.No	Samples	Acetone extract (%)	Ethanol extract (%)	Water extract (%)
1.	Aloe(leaf)50µl	66%	43%	42%
2.	Aloe(Leaf)100µl	78%	48%	58%
3.	Aloe(leaf)150µl)	92%	59%	71%
4.	Aloe(leaf)200µl	93%	71%	77%
5.	Aloe(gel)50µl	63%	53%	37%
6.	Aloe(gel)100µl	73%	62%	59%
7.	Aloe(gel)150µl	92%	66%	82%
8.	Aloe(gel)200µl	92%	75%	94%

IV. TEST FOR TOTAL REDUCING POWER

Table 7: Table showing In vitro and Ex vivo of total reducing power

ex vivo

S.No	Samples	Acetone extract (mg/ml)	Ethanol extract (mg/ml)	Water extract (mg/ml)
1.	Aloe leaf	620	955	820
2.	Aloe gel	840	890	892

in vitro

S.No	Samples	Acetone extract (mg/ml)	Ethanol extract (mg/ml)	Water extract (mg/ml)
1.	Aloe leaf	625	1020	842
2.	Aloe gel	6352	675	590

ANTIMICROBIAL ACTIVITY

Ex vivo

The zone of inhibition in bacterial culture plates were found to be higher in *Pseudomonas fluorescens*(40µl) and *Staphylococcus aureus*(20µl) with 0.15 mm diameter and in fungal culture plates were found to be higher in *Trichodermaviridae*(20µl) with 0.15 mm diameter.

Pseudomonas species ***Streptococcus species*** ***Trichoderma species***

Fig 4: Bacteria showing antimicrobial activity in Ex vivo type

Table 8: Antimicrobial activity in In vitro type

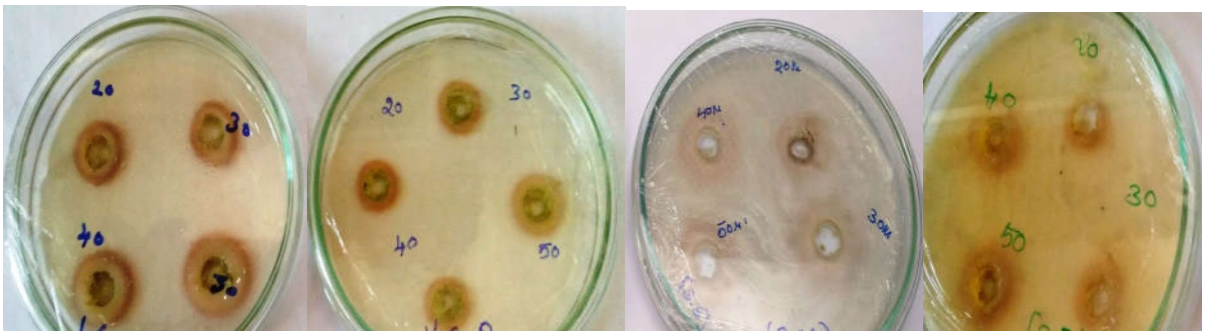
Sl. No	Microbes	20µl	30µl	40µl	50µl
1.	*Pseudomonas fluorescence*	0.1 mm	-	0.15mm	-
2.	*Staphylococcus aureus*	0.15 mm	0.22 mm	0.2 mm	0.13 mm
3.	*Aspergillusniger*	-	-	0.2 mm	0.1 mm
4.	*Trichodermaviridae*	0.15 mm	0.2 mm	0.13 mm	0.12 mm

Table 9: Antimicrobial activity in In vitro type

In vitro

Sl. No	Microbes	20µl	30µl	40µl	50µl
1.	*Pseudomonas fluorescence*	0.3 mm	0.35mm	0.3mm	0.43 mm
2.	*Staphylococcus aureus*	0.15 mm	0.25 mm	0.3 mm	0.33 mm
3.	*Aspergillusniger*	-	0.41mm	0.46 mm	0.25 mm
4.	*Trichoderma viridae*	0.2mm	0.3 mm	0.5 mm	0.2 mm

The zone of inhibition in bacterial culture plates were found to be higher in *Pseudomonas fluorescens*(50µl) with 0.43 mm diameter and in fungal culture plates were found to be higher in *Aspergillusniger*(40µl) with 0.46 mm diameter.



Pseudomonas species ***Streptococcus species*** ***Aspergillus niger*** ***Trichoderma species***

Fig 5: Bacteria showing antimicrobial activity in In vivo type

DISCUSSION

There is considerable use of *A. barbadensis*in folk medicine in the southern most United States, and some cosmetics and patent medicines generally found on the market are prepared from the gel in the leaves and from the juice. This demand for *Aloe vera* interferes with the fastest mode of propagation along with the large quantity. The phenolics content that was found to be higher in acetone extract of *aloe* gel in *in vitro* with 59.8 mg/ml. *Ex vivo* plant extracts were equally performing but the Flavonoids presence shown higher in the *Aloe in vitro.* The reducing power that was found to be higher in acetone extract of aloe *leaf* in both *ex vivo* and *in vitro* with 955 mg/ml and 1020 mg/ml respectively. The DPPH free radical content that was found to be higher in acetone extract of aloe *leaf* (100µl) in ex *vivo* with 55.7% and higher in acetone extract of *aloe gel* (200µl) in *in vitro* with 94%.

When antimicrobial tests were performed, the zone of inhibition in bacterial culture plates were found to be higher in *Pseudomonas fluorescens*(40µl) and *Staphylococcus aureus*(20µl) with 0.15 mm diameter and in fungal culture plates were found to be higher in *Trichoderma viridae*(20µl) with 0.15 mm diameter followed bythe zone of inhibition in bacterial culture plates were found to be higher in *Pseudomonas fluorescens* (50µl) with 0.43 mm diameter and in fungal culture plates were found to be higher in *Aspergillus niger*(40µl) with 0.46 mm diameter. *In vitro* and *ex vivo* samples of succulent plants were compared for disease resistance and anti-oxidant activity. It was found that the *in vitro* explants possess

elevative disease resistance and higher concentrations of anti-oxidants. So it can be used as an alternative to those cosmetics and medicines made out of *Aloe barbedensis.*

CONCLUSION

The in-vitro and ex vivo anti-oxidant and anti-microbial activities of Aloe barbadensis were studied successfully. Efficient estimation of total flavonoid content was studied. It was inferred that Alkaloids were present in the ethanol and acetone extracts also followed by flavonoids present in the ethanol and acetone extracts except water extracts. The flavanoids can be responsible for the various pharmacological activities. In vitro and ex vivo samples of succulent plant were compared and it was found that the contents present in in vitro, was found to be Higher. While compared to ex vivo, such as phenolic, Flavonoids and DPPH free radical and in vitro explants possess elevative disease resistance and higher concentrations of antioxidants. So it can be used as an alternative to those cosmetics and medicines made out of aloe barbadensis.

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