

## World News of Natural Sciences

An International Scientific Journal

WNOFNS 48 (2023) 40-45

EISSN 2543-5426

# Retention of leaf senescence of aquatic angiosperms using IAA

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#### **ABSTRACT**

One of the plant growth promoters, IAA (Indole-3-acetic acid) on deferral of leaf senescence was analysed for plant species namely, *Salvinia* sp. and *Jussiaea* sp. Changes of some biochemical parameters like chlorophyll, protein, soluble and insoluble carbohydrates as well as activity of catalase enzyme were analysed as reliable senescence indices during detached leaf senescence. With the progress of ageing duration from zero to 48 hours the levels of chlorophyll and proteins in leaf discs gradually declined in both control and IAA treated samples. However, in the hormone treated samples the rate of decline was found to be much slower. Concomitantly the level of insoluble carbohydrate started declining right from 12, 24 and 48 hours of observation period both in control and IAA treated samples.

Keywords: Senescence, IAA, biochemical changes, leaf ageing, Salvinia, Jussiaea, aquatic angiosperms

#### 1. INTRODUCTION

Senescence is a programmed deteriorative phenomenon occurring within cells, tissues, organs and organisms which is culminated in the death of the concerned plant part or the organisms as a whole [1]. As the process of senescence takes place at an exceedingly faster rate under detached condition of plant parts, the effect of any chemical having influence on the regulation of senescence can be quickly determined [2]. Deferral of senescence by plant hormones like cytokinins is well established [3-5]. However, some gibberellins and auxins have also been reported to defer senescence in a number of plant species, but their efficiency is mostly not at par with cytokinins [6-7].

In the present experiment, an attempt was made to ascertain whether IAA, a member of auxin class of growth promoter, can regulate senescence of two aquatic plant species namely *Salvinia* and *Jussiaea* under detached leaf condition and the principal objective of this investigation was to evaluate the efficiency of IAA towards possible response on senescence retardation at least in case of the three aquatic plant species.

#### 2. MATERIALS AND METHODS

The experimental plants used were two aquatic angiosperms namely *Salvinia* sp. (Family: Salviniaceae) and *Jussiaea* sp. (Family: Onagraceae). The plant species were first carefully surface blotted using blotting paper. Uniformly sliced leaf discs, taken from mature leaves of the plants were treated with aqueous solution of IAA (200  $\mu$ g/ml) or distilled water (control) in Petri dishes containing filter paper.

The experimental set-up was kept in dark condition and thus allowed the leaf discs to experience treatment with IAA for 48 hours. At an interval of 12 hours the filter papers were remoistened with the test chemical or distilled water and the biochemical data recorded include: chlorophyll, protein, soluble and insoluble carbohydrate contents as well as activity of the enzyme catalase.

To determine the level of chlorophyll the absorbance of the chlorophyll extracts was measured at 650 nm and total chlorophyll content was estimated following Arnon's principle [8]. Extraction and estimation of protein and carbohydrates (both soluble and insoluble) was done as per the method of Lowry *et al.* and McCready *et al.* respectively [9-10]. Extraction and estimation of the enzyme catalase was made following the method of Snell and Snell [11]. For assaying the enzyme, a blank was taken as zero time control and the activity was expressed as  $(\Delta OD \times T_V)/(t \times v)$ , where  $\Delta OD$  is the difference of OD of the blank and sample,  $T_V$  is the total volume of the filtrate, t is the time (min) of incubation with the substrate and v is the volume of filtrate taken for incubation [12].

All the data were statistically analysed at the treatment and replication levels and least significant difference (LSD) values were calculated at 95% confidence limits [13].

#### 3. RESULTS AND DISCUSSION

Results showed that both chlorophyll and protein contents started declining rapidly with the advancement of leaf ageing irrespective of the treated and control samples. However, IAA arrested the rapid loss of both chlorophyll and protein levels (Table 1). Soluble carbohydrate was found to increase while insoluble carbohydrate steadily declined with the progress of ageing duration (Table 2).

IAA ameliorated the ageing-induced rapid rise of the soluble sugar levels as well as the progressive loss of the insoluble carbohydrate levels. IAA was found to significantly arrested reduction in nucleic acid levels and the effect was particularly significant when data were recorded after 12 hours of leaf ageing and subsequent observations. The activity of enzyme catalase decreased with the progress of stress-induced ageing duration (Table 3). The chemical treatment of leaves with IAA alleviated the ageing-induced rapid loss of catalase.

Senescence of detached leaves starts immediately after the separation from the plants and occurs at a rapid rate with the progressive increase of catabolic activities and these ultimately result in death and decay of leaves. Although the senescence pattern of terrestrial plants has been profusely studied, the literature on the senescence pattern and the effect of hormones on regulation of senescence in aquatic plants are rather scanty [14].

The precise mechanism of senescence of terrestrial and aquatic plants may be somewhat different but the overall deteriorative changes during senescence are almost similar in both the plant types as well as in attached and detached leaf senescence types. The basic difference is that in case of detached leaf senescence the physiological and biochemical changes occur at an exceedingly faster rate [15]. Similarly, the reversal effects of the above types of senescence by senescence retardants are more or less same.

Numerous reports exist in the literature that during all types of senescence loss of some vital macromolecules like chlorophyll and protein takes place which is due to their degradation and /or subdued rate of biosynthesis [16-17]. Any chemical or external agents possessing the property to maintain the chlorophyll and protein levels during senescence are regarded as senescence retardants [18-19].

In this investigation, IAA-induced partial arrestation of the rapid loss of chlorophyll, protein, insoluble carbohydrates is indicative of the senescence deferral action of IAA. Catalase is regarded as a scavenger enzyme [20-21], and higher activity of this enzyme is the index of plant vigour.

In this investigation the IAA-induced retention of catalase activity during dark-induced detached leaf senescence is indicative of the retardation of senescence.

**Table 1.** Effect of IAA (200 μg/ml) on changes in chlorophyll; CHL and protein; PR (mg/g fr. wt. each) levels in leaf discs of plants analysed.

Plant materials	Treatments (μg/ml)	Hours after leaf ageing									
		0		12		24		48			
		CHL	PR	CHL	PR	CHL	PR	CHL	PR		
Salvinia	Control	3.28	2.35	2.22	1.78	1.58	1.00	0.95	0.47		
	IAA	3.28	2.45	3.05	1.77	2.85	1.14	1.75	0.88		
	LSD (P=0.05)	NC	NC	0.47	0.46	0.56	0.17	0.85	0.17		
Jussiaea	Control	3.85	2.53	2.52	1.60	1.70	1.10	0.85	0.74		
	IAA	3.85	2.53	2.70	2.05	1.61	1.62	1.01	1.15		
	LSD (P=0.05)	NC	NC	0.22	0.28	0.17	0.21	0.15	0.09		

NC: Not calculated.

**Table 2.** Effect of IAA (200  $\mu$ g/ml) on the changes in soluble carbohydrate; SC. and insoluble carbohydrate; ISC (mg/g fr. wt. each) levels in leaf discs of plants analysed.

Plant materials	Treatments (μg/ml)	Hours after leaf ageing								
		0		12		24		48		
		SC	ISC	SC	ISC	SC	ISC	SC	ISC	
Salvinia	Control	1.85	4.52	2.10	4.15	2.95	3.12	3.30	1.95	
	IAA	1.85	4.51	1.92	4.38	2.25	3.05	2.52	2.25	
	LSD (P=0.05)	NC	NC	0.07	0.28	0.21	0.25	0.25	0.11	
Jussiaea	Control	1.96	5.77	2.17	5.09	2.69	4.82	3.18	3.08	
	IAA	1.96	5.78	2.05	5.70	2.15	5.15	2.57	4.24	
	LSD (P=0.05)	NC	NC	0.17	0.79	0.32	0.15	0.34	0.72	

NC: Not calculated.

**Table 3.** Effect of IAA (200  $\mu$ g/ml) on the changes in the activity of catalase ( $\Delta$ OD  $\times$  Tv / t  $\times$  v) enzyme in the leaf discs of plants analysed.

Plant materials	Treatments	Hours after leaf ageing						
	(μg/ml)	0	12	24	48			
Salvinia	Control	57.5	48.2	38.0	31.7			
	IAA	57.8	52.7	48.2	40.5			
	LSD (P=0.05)	NC	4.01	1.07	0.17			
Jussiaea	Control	60.8	58.7	52.3	37.9			
	IAA	60.9	60.1	58.7	46.79			
	LSD (P=0.05)	NC	0.92	2.28	3.15			

NC: Not calculated.

#### 4. CONCLUSION

Results show that IAA is a potent growth promoter for maintenance of membrane integrity as well as arrestration of the detached leaves senescence of the plants analysed. Thus, can be concluded that IAA can be considered as a potent senescence deferral agent for the aquatic plant species analysed in this investigation.

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