Journal of Materials and Environmental Sciences
ISSN : 2028-2508

JMES, 2017 Volume 8, Issue 3, Page 942-948

http://www.jmaterenvironsci.com

ICMES2016, 1-3 Dec. 2016, Oujda

Effect of gibberellic acid (AG3) on the germination of seeds of *Thymus satureioides L and Lavandula dentata*

M. Chetouani, I. Mzabri, A. Aamar, A. Boukroute, N. Kouddane, A. Berrichi

Laboratory of Biology of Plants and Microorganisms, Faculty of Sciences,
B.P. 717, Oujda 60.000, Morocco.

Received
Revised
Accepted

Keywords

- ✓ medicinal plants;
- ✓ optimization;
- ✓ gibberellic acid;
- ✓ *Thymus satureioides*;
- ✓ *Lavandula dentata*

:chetouanimohammed@gmail.com

Phone :00212677624147

ABSTRACT

Although Morocco has long been considered a reservoir rich in medicinal and aromatic plants, this heritage is still under exploitation. In fact the diversity of the Moroccan seed bank in medicinal and aromatic plants requires a particular interest through the optimization of the production of some species, such as *Thymus satureioides.L* and *Lavandula dentata*. To do this, an experiment was carried out on seeds of these species at the Laboratory of Biology of Plants and Microorganisms of the Faculty of Sciences in Oujda to study the effect of gibberellic acid on their germinations. The experimental protocol includes four treatments on *Thymus satureioides* and six treatments on *Lavandula dentata*. The comparison of the different results shows that the treatment of the seeds of *Thymus satureioides* L and *Lavandula dentata* significantly improved the germination's percentage and kinetics. The highest effect observed in the treatment which was treated with 50 ppm AG3 for *Thymus satureioides*.L showed an increase of 27% in the germination's percentage compared to the control and that the seeds of the *Lavandula dentata* treated with gibberellic acid at 1000ppm marked a maximum germination of 67% compared to the control which did not exceed 1%.

1. Introduction

Moroccan flora is one of the richest in the Mediterranean basin and has many aromatic and medicinal species such as *Lavandula dentata* and *Thymus satureioides*.L which are represented respectively by 10 spontaneous species of which 5 are endemic for *Lavandula dentata* and by 21 species of which 12 are endemic for *Thymus satureioides* [1]. This richness and diversity of the plant palette adds to the exponential demand of the national and international market for products from medicinal plants where 25% of medicines in developed countries are derived directly or indirectly from plants [2]. Morocco will become a major producer of aromatic and medicinal plants especially with the support of the Moroccan Green Plan program which gives particular interest to the rational exploitation of this national heritage in the ecologically and geographically marginalized regions.

The success of seed germination is hampered by many endogenous and exogenous factors leading to different types of dormancy, such as thyme's and lavender's, which is known to be slow [3]. There are several methods for initiating germination: stratification, scarification and the use of growth regulators such as gibberellic acid known for its positive effect on seed dormancy emergence [4, 5].

This study will focus on the optimization of the production of two medicinal and aromatic plants namely *Thymus satureioides* and *Lavandula dentata* in an attempt to improve their germination rates.

2. **Materials and methods**

2.1. Seeds' origin

Thymus satureioides'seesds were harvested in July 2014 in the region of Talsint which is located south-west of the province of Figuig in eastern Morocco whose coordinates are 32 ° 51'54 " north 3 ° 27'02 " West at an altitude of 1344m.

Lavandula dentata's seeds were harvested in March 2015 at the experimental station of the Faculty of Science of Oujda at an altitude of 661m, a latitude of 34 ° 39 '07' 'North and a longitude of 01 ° 53' 01 'West (GPS Back Track Bushnell), the climate is from semi-arid to mild winter.

2.2. Experimental conditions

The germination tests were carried out in the laboratory of Biology of Plants and Microorganisms of the Faculty of Sciences in Oujda.

All the germination tests were carried out in a chamber considered to be phytotronic where the temperature is set at 25 ° C. ± 2 with a relatively saturating humidity, the substrate used consists solely of black peat placed in transparent sterile plastic Petri dishes of 10cm of diameter.

2.3. Tests installation

In order to improve the germination of Thymus satureiodes and Lavandula dentata, 300 seeds for each treatment and for each species (with 3 replicates) were selected and disinfected with 12 ° bleach diluted to 5 % for 5 minutes and then thoroughly washed 3 times with distilled water to remove the traces of sodium hypochlorite as well as the traces of other products which have been adhered to the seed.

2.3.1. Treatments for *Thymus satureioides*

Treatment 0: without imbibition (Control).
Treatment 1: imbibition for 24 hours in distilled water.
Treatment 2: imbibition for 24 h in gibberellic acid (AG3) at 50ppm.
Treatment 3: imbibition for 24h in AG3 at 100ppm.

2.3.2. Treatments for *Lavandula dentata*

Treatment 0: imbibition for 24 hours in distilled water
Treatment 1: imbibition for 24h in gibberellic acid (AG3) at 200ppm.
Treatment 2: imbibition for 24h in AG3 at 400ppm.
Treatment 3: imbibition for 24h in AG3 at 600ppm.
Treatment 4: imbibition for 24h in AG3 at 800ppm.
Treatment 5: imbibition for 24h in AG3 at 1000ppm.

100 seeds thus treated were sown in Petri dishes filled to 2/3 of its volume with black peat and sprayed with a methyl thiophanate fungicide (2cc / hl) to prevent mold growth before being put into the phytotronic enclosure. A daily control of humidity and temperature was carried out in order to avoid any desiccation of the substrate in the boxes.

2.4. Experimental device

The experimental device adopted is a complete randomized block device for both species, with 4 treatments and 3 repetitions for Thymus satureioides, and 6 treatments in total with 3 repetitions for Lavandula dentata.

2.5. Measured parameters

2.5.1**.** Germination's percentage (%)

The germination's percentage corresponds to the number of germinated seeds relative to the total number of seeds germinated.

A seed is considered germinated when it emits a radicle and gemmule of 2mm [6].

Seed counting is carried out from 48 hours of germination and spread over a period of 15 days.

2.5.2 Germination's Kinetics:

It corresponds to the curve of the evolution of the daily rates of the seeds from the beginning to the end of the germination and calculated on the basis of the number of the seeds newly germinated at each observation [7].

2.5.3 Statistical Analysis

The results are subjected to a descriptive statistical analysis and an analysis of the variance (ANOVA), using the software "SPSS for Windows version 20" and the comparison of the averages is made by the test of Scheffe at the probability threshold of 5 %.

3. Results

3.1 *Thymus satureioides*' germination Percentage

The results obtained show that seeds' germination percentage (Fig.1) has been improved in a statistically significant manner following the pretreatments applied. A simple imbibition in distilled water resulted in an 11% increase in the germination's percentage compared to the control, while hormone therapy showed increases of 27% and 23%, respectively, compared to the control for concentrations of 50 and 100 ppm respectively. Thus a germination rate of 62% was obtained following the application of AG3 at a concentration of 50 ppm, while the germination rate of the control did not exceed 35%.

.

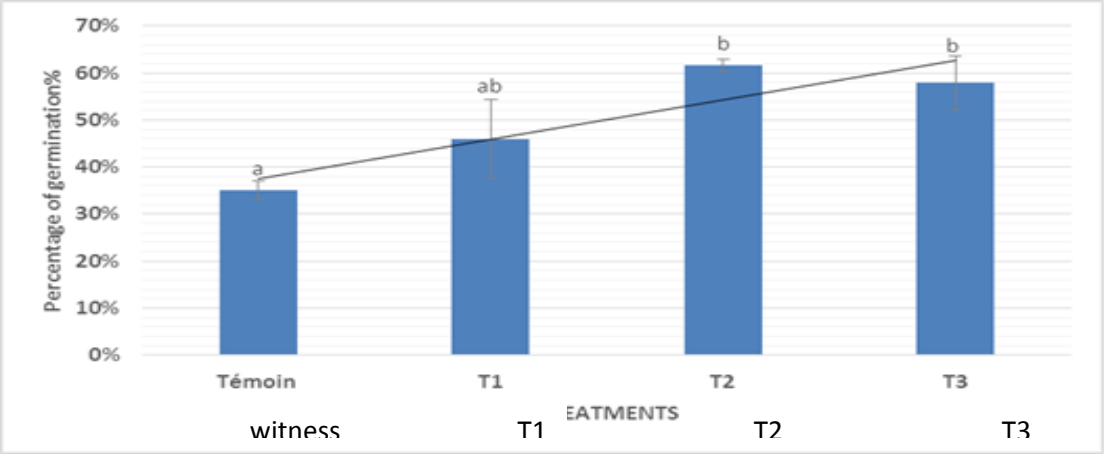

Figure 1: *Thymus satureioides*'s seed germination percentage

3.2 *Lavandula dentata*'s germination Percentage .

Seed germination was significantly improved following hormonal treatments compared to those imbibed in distilled water where the germination rate did not exceed 1% (Fig. 2).

The curve's trend also shows that the germination's percentage of the seeds treated with gibberellic acid at 1000ppm marked a maximum germination equal to 67% while the other hormonal treatments did not exceed 39%.

3.3 *Thymus satureioides'* germination Kinetics.

The Thymus Satureioides's germination kinetics shown in fig3 describe a sigmoidal shape comprising three phases: a latency phase, corresponds to the imbibition of the seeds, an exponential phase where we notice an acceleration of germination and finally a stationary phase indicating germination stop. The control showed a 4% germination percentage observed from the 3rd day after sowing to a final rate of 35% after 15 days,

the exponential germination phase lasted 12 days. While a simple imbibition in water at ambient temperature for 24 hours (T1) allowed the latency phase which lasted only one day to elapse. The first germinations (7%) were observed on the second day reaching a maximum rate of 46% after an exponential phase of 13 days.

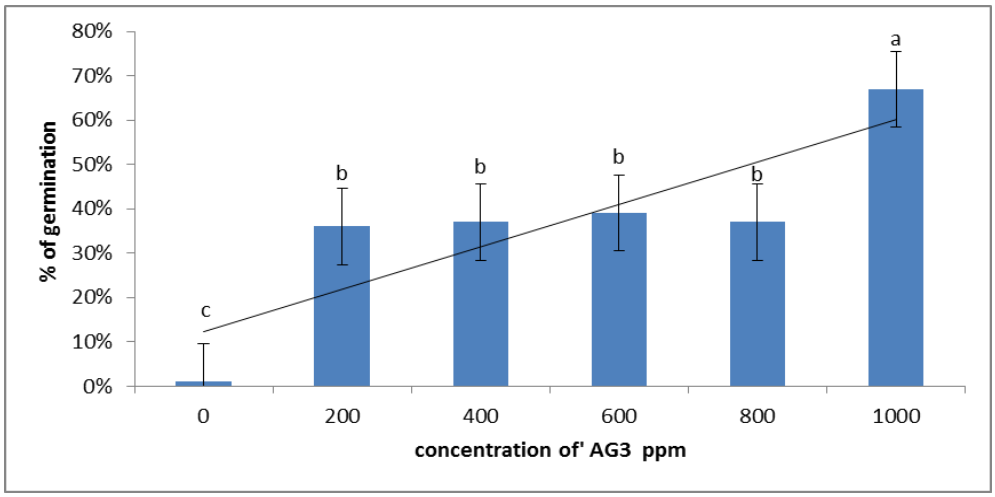

Figure 2: *Lavandula dentata* seeds' germination percentage

The imbibition for 24 hours in gibberellic acid (AG3) at 50 ppm (T2) showed the first germinations (28%) after a latency phase of 3days and an exponential phase lasting only 7 days, before reaching the stationary phase where germination stops with a maximum germination of 62%.

Treatment with AG3 at 100 ppm marked the first germinations from the 3rd day after sowing, rapidly reaching a rate of 37% on the 5th day. This level was stabilized at 58% after an exponential phase of 11 days.

From the results obtained, treatment of the seeds of Thymus satureioides with AG3 significantly improved the kinetics of the germination whose curve shape is modified in the direction of shrinkage, resulting in an acceleration of the germination rate.

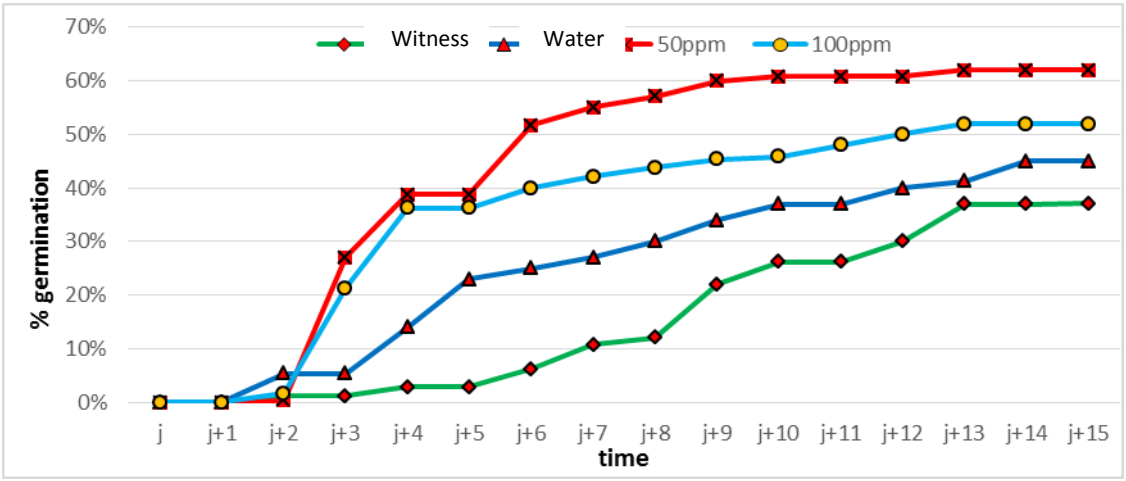

Figure 3: Thymus Satureioides germination's Kinetics

3.4 *Lavandula dentata*'s germination Kinetics

Lavandula dentata's seed germination kinetics illustrated in figure 4 illustrates 3 stages, a latency stage due to the massive absorbance of the water by the seeds, a second stage indicating an exponential mobilization of the embryonic stocks and then a Stationary stage indicating the cessation of germination.

The germination's start was after 3 days for T0 to T5, then an exponential phase reaching the maximum germination of 39% for the concentration of 600ppm except for the concentration of 1000ppm where the exponential phase recorded from the 6th day stabilized at 67% and then a stationary phase indicating the germination's end.

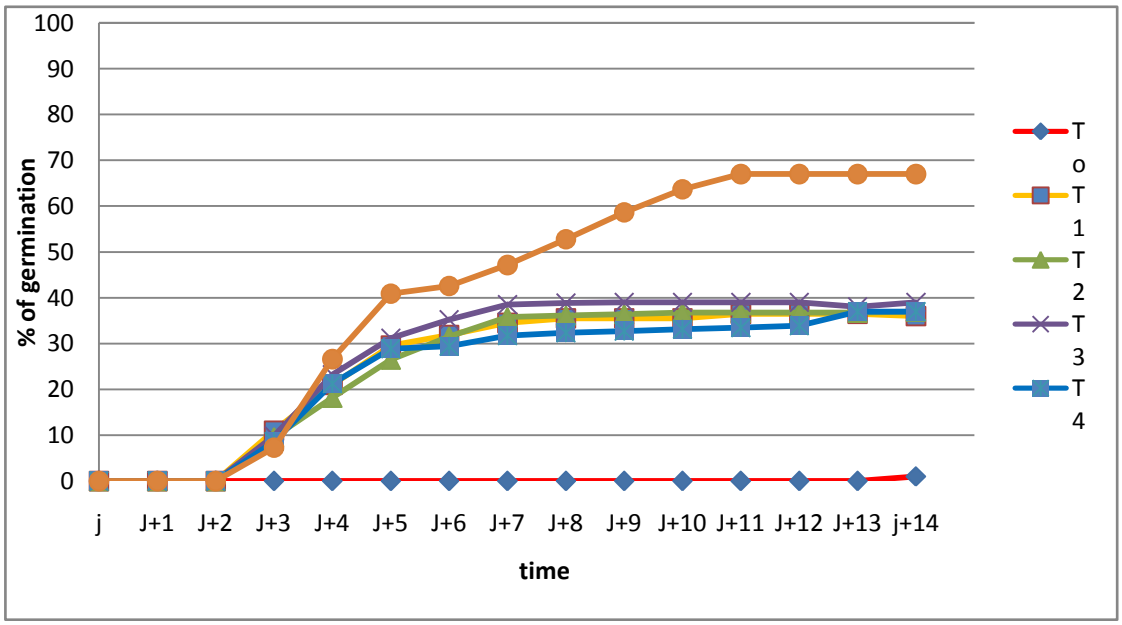

Figure 4: *Lavandula dentata*'s germination kinetics

4. Discussion

Overall, for all the seeds that underwent germinating hormone treatment with a level of more than 50% for thyme and 67% for lavender, the germination's percentage did not exceed 45% in the control of *thymus satureioides*'s seeds and 1% for *Lavandula dentata*'s. The difficult and random germination of *Lavandula dentata*'s seeds is reported by many authors to have a very clear improvement of the germination's rates by the positive effect of certain treatments with gibberellic acid. Several authors also confirm the existence of a dormancy that can be of the morpho-physiological type for *Lavandula dentata* [8], which may be a combination of physiological dormancy and morphological dormancy that originate from an inhibition mechanism which prevents emergence of the radicle. The causes of this dormancy are still poorly known and are probably at the level of interactions between the embryo and the tegumentary structures.

The most significant effects are obtained in seeds receiving a concentration of 50 ppm AG3 for thyme and 1000 ppm for *Lavandula dentata*. Similar results were obtained by Felipe [9] on the almond tree which showed that the use of AG3 with a concentration of 50 ppm made it possible to obtain a total germination of the almonds without being stratified. Also the soaking of the seeds of capering in the gibberellic acid at concentrations of 50 to 100 ppm for 1 hour allowed an increase in their percentage of germination to 80% [10]. In contrast to treatment (T2) for *thymus satureioides* with the highest gelation rate (62%), treatment (T3) did not exceed a rate of 52%. Although this decrease is not statistically significant, it may be related to a surplus of the hormone content used, which could be considered as an onset of AG3 toxicity. According to Abdel Hady [11], similar results were obtained from Roselle (Hibiscus sabdariffa L. var. Sabdariffa) [12], similarly Ozguven et al [13], Yaaqobi et al [14,15] showed the positive effect of gibberellic acid on seeds of Pistacia atlantica and Pistacia Vera. Ayfer and Serr [16] also showed the positive effect of AG3 on the rate and the percentage of germination of the seeds of the cultivar Trabonella germinated in vivo. This reflects the primordial action of the AG3 hormone on seed germination through its physiological effect, which translates into the replacement of exogenous cold requirements by an improvement in the level of endogenous AG3 required for germination. It is well known that during the

germination process gibberellic acid is released from the embryo to activate genes specific for the transcription of α-amylase [17, 18], in order to accelerate the synthesis Enzyme required for the hydrolysis of the starch.
It appears that the exogenous AG3 induces not only the longitudinal and diametrical growths of the stems and those of the foliar surfaces of the plants but also the increase of the seed germination rates known in the literature.
Gibberellin treatments have caused increases in germination's percentage and resulted in rapid and the mechanisms of this hormone's action are now fairly well known. [19] Gibberellin acts through the Stimulation of the inter-meristem division's between the nodes of the stems. [20] This stimulation comes from the coordinated effect of two independent actions: the induction of meristematic proliferation and cellular elongation [21]. Cell proliferation consists of an activation of the meristematic divisions, leading to the differentiation of new tissues, and therefore to the formation of the cortical and epidermal parenchyma [22]. Such actions have been noted in corn cells [23], wheat [24] or Cajanus caja [25] treated with AG3. Cellular elongation, on the other hand, results from the plastic extension of the cell walls. [26] Parietal distensions induce cellular elongations which lead to tissue growth and to the dislocation of the internodes [27] longitudinal growth.
The most significant effects were obtained at concentrations of 50 ppm and 1000 ppm respectively for thymus satureioides and Lavandula dentata. Gibberellin is therefore an important growth stimulating hormone for plants *thymus satureioides* and *Lavandula dentata*. Similar results have already been reported on tomato [28], pea [29] and cotton [30]. The use of gibberellic acid promotes a 10% increase in the germination of inoculated seeds in a culture medium immediately after harvest. However, the use of 2.48 mg L-1 (calculated value) leads to 100% germination [31].
In our work, the results of this study also showed that concentrations exceeding 50 ppm AG3 for *thymus satureioides* produced low levels of germination, these results may be related to deficiencies in hormone levels used, such as l 'Suggested [32] on Brassica campestris [33] on tomato. Indeed, up to a threshold limit (specific to each plant), the effects of a hormone are proportional to the concentrations used, the smallest inducing doses, the lowest effects, and the highest concentrations causing the greatest impacts [34].
The decrease in the growth parameters of treated plants at concentrations greater than 10-2 M seems related to the beginning of chemical toxicity of gibberellin, as proposed [35] on Atropa belladonna (Solanaceae).
According to these authors, gibberellin is a pesticide whose high concentrations cause acute toxicities on the animal and plant cells, if long exposed to its action. In Vigna radiata (Fabaceae), for example, the work [36] revealed cell poisoning after one week of treatment with GA-3 concentrated at 100 mg / l a threshold limit, the effects of a hormone are proportional to the concentrations used; The smallest inducing doses, the lowest effects, and the highest concentrations causing the greatest impacts[37] .

Conclusion

The study of the improvement of hormonal germination using gibberellic acid (AG3) has improved the germination's rate and kinetics *Thymus satureioides* and *Lavandula dentata*. Thus, following the results obtained by the different pretreatments used, we observed that the imbibition of seeds for 24h in gibberellic acid (AG3) at 50ppm for *Thymus satureioides* and 1000ppm for *Lavandula dentata* germination is 62% and 67%. This will enable nurserymen to optimize the production of these species for extension in marginalized regions with low rainfall and to develop products with high added value corresponding market's demand.
Gibberellic acid can therefore be recommended in programs to improve *Thymus satureioides* and *Lavandula dentata*, provided that the recommended concentration ranges are observed.

References

1. Benabid A. *Édition Ibis Press*. (2000) 159- 161.
2. Damintoti K., Mamoudou H.D., Jacques S., Saydou Y., Souleymane S., Alfred S.T. *Mémoire Ouagadougou*. (2005).
3. Elsheikh A.M., *J. Coll. Sci., King Saud Univ*., 15 (2) (1984) 389-400.
4. Finkelstein R., W Reeves.,T Ariizumi. Steber. *Plant* . 59 (2008) 387-415.
5. Bacchetta G., Belletti P., Brullo S., Cagelli L., Carasso V., Casas, J., Guemes J., *Manuel* (2006) 217.
6. Sayar R., Bchini H., Mosbahi M., Khemira H. *Plant Breed*. 2 (2010) 54-63.
7. Hajlaoui H.,Denden Met.Bouslama M *Tropicultura* .25 (2007) 168-173.
8. Baskin CC., Baskin. Jerry M. *Elsevier*. (1998) 87-100.
9. Felipe A.J Ciheam : *Opts Médit* (1981) 139-140.
10. Bulletin de Transfert de technologie en agriculture. 37 (1997) 12.
11. Abdel-Hady M.S., Okasha E.M., Soliman S.S.A., Talaat M., *J. Austr. Appl. Sci*. 2 (2008) 401-405.
12. Lepengue A., N Daouda., S Kouadio.,Zouzou M.,M'Batchi, B. *J Appl Biosc* .49 (2012) 3435-3443.
13. Ozguven AK., Nikpeymay A.I., *Acta Hortic*. 419 (1995) 115-117.
14. Yaaqobi A., El Hafid L., Haloui B. *Biomatec Echo*. 3 (2009) 39-49.
15. Simonnet X., Quennoz M.,Sigg S., Carlen C. *MAP-Mountain*. 955 (2011) 309-.313.
16. Ayfer M., Serr E. F., *J. Amer. Soc. Hart. Sci.* 77 (1961) 308-315.
17. Yaniaguchi S., *Annu. Rev. Plant Biol* .59 (2008) 225-251.
18. Bialecka B., Kepczynski J., *Acta Biologìca Cracoviensïa.* 51/2 (2009) 119-125.
19. Heller R., Esnault R., Lance C. *Editions Dunod*. (2006) 366.
20. Hooley R. *Plant Mol. Biol* .26 (1994) 1529-1555.
21. Inglese P., Chessa I., La Mantia T., Nieddu G. *Sci. Hort.* 73(1998) 45-51.
22. Pharis RP. And King RW. *Annu. Rev. Plant Physiol.*36 (1985) 517-568.
23. *Carpita NC. Kanabus J., Plant Physiol.*88 (1988) 671-678.
24. Keyes G., Sorrells ME., Setter TL. *Plant Physiol.* 92(1990) 242-245.
25. Chudasama, V. Thaker. *Plant Physiology* .19 (2007) 1.
26. Heller R., Esnault R., Lance C. *Editions Dunod (2006) 366.*
27. Nakayama A., Park S., Zheng-Jun X., Nakajima M., Yamaguchi I., *Plant Physiol.* 129 (2002) 1045-1053.
28. Richards DE., King KE., Ait-Ali T., Harberd NP., *Plant Physiol. Plant Mol. Biol.* 52 (2001) 67-68.
29. Groot SPC. Bruinsma J., Karssen CM., *Plant.*71 (1987) 184-190.
30. Swain SM., Reid JB., Kamiya Y., *Plant J.*, 12 (1997) 1329-1338.
31. Gokani SJ. Thaker VS., *J. Agric. Sci.* 138 (2002) 255-260.
32. Banyal S., Rai VK. *R. Physiol. Plant* .59 (1983) 111-114.
33. Damiani.C.R ,*Afr. J. Biotechnol.* 15(32) (2016) 1731-1737.
34. Groot SPC. Bruinsma J., Karssen CM. *Physiol. Plant*. 71 (1987) 184-190.
35. Hopkins WG., *Edition de Boeck* (2003) 532.
36. Abdel hady M.S., Okasha O.M., Soliman S.S.A., tallat M. *Aust. J. Basic Appl. Sci.* 2 (1991) *401-405.*
37. Akbari N., Barani M., Ahmadi H, *World Applied Sci Journal*.5 (2008) 199-203.

(2017) ; http://www.jmaterenvironsci.com/