



ENHANCEMENT OF SEED GERMINATION AND SEEDLING GROWTH IN *MAGNOLIA GRANDIFLORA* L. USING PRE-SOWING TREATMENT WITH GROWTH REGULATORS (IAA AND GA3)

OMER IBRAHIM¹, VALERIA IVANOVA², PETYA GERCHEVA³, LILYANA NACHEVA³

¹ Horticulture Dept., Fac. Agriculture, Assiut Univ., Assiut, Egypt, E-mail: omer_hooo@yahoo.com

² Faculty of Viticulture and Horticulture, Agricultural Univ., Plovdiv, Bulgaria, E-mail: valeriasi1@abv.bg

³ Fruit-growing Institute, 4004 Plovdiv, Bulgaria. E-mail: gercheva_p@abv.bg

Abstract

The efficiency of pre-sowing treatment of *Magnolia grandiflora* seeds with GA3 and/or IAA at different concentrations (1000, 1500, 2000 and 2500 ppm) was investigated. The highest germination percentage was obtained with 2500 ppm GA3 or IAA+GA3 (66.25%, 62.50%, respectively). Meanwhile, IAA with or without GA3 induced the best seedling growth. Using of 2500 ppm GA3+IAA is, therefore, recommended to attain high germination percentage and vigorous seedlings.

Key words: *Magnolia grandiflora*, GA3, IAA, Auxins, Germination, Propagation

INTRODUCTION

Magnolia grandiflora, Southern Magnolia, is an evergreen tree belonging to family Magnoliaceae and native to Southeastern the United States from which the name southern magnolia was derived. It was introduced to Europe in 1731 (Gary, 1994; Shi *et al.*, 2002).

M. grandiflora is a tree of remarkable aesthetic, medicinal and timber values. Its showy, large, fragrant white flowers appearing in late spring make it a valuable ornamental species (Dirr, 2002). It is a noble urban landscape tree for it is resistant to acid deposition. Besides, it provides timber and yields a variety of extracts with potential applications as pharmaceuticals (Halls, 1977; Shi *et al.*, 2002).

M. grandiflora is successfully propagated by several vegetative methods, i.e. cutting, grafting and layering, yet seeds provide the easiest method of mass propagation (Callaway, 1994). Germination in *M. grandiflora* is epigeal and usually occurs the first or second spring following seedfall. Even though viable, seeds rarely germinate under the parent tree because of reported inhibitory effects (Blaisdell *et al.*, 1973). The underdeveloped embryo of *Magnolia*, which is about 1 mm long and 0.4 mm in diameter and is located at the micropyle end of the seed, needs to undergo a stratification treatment (Barbour, 2008 and references therein).

Considerable research has been performed on enhancing the germination of *Magnolia* seeds. *M. grandiflora* seeds germinated up to 61-63% when pre-sowing stored at 5°C for 3 months (Misiha and El-Ashry, 1991). Hanchey and Kimbrough

(1954) have reported higher rooting percentage 88% using 2-month stratification at 15°C. Increasing the temperature (15-35°C) was reported to get a great total germination percentage (Evans, 1933). Naked stratification, i.e. stratification without substrates, of *M. grandiflora* seeds proved not to be suitable (Mihailo and Dragana, 2004).

Another common pre-germination treatment is growth regulators, where both GA3 and BA have been reported for treating seeds of *M. grandiflora*. Rates as high as 1000 p.p.m. of GA3 were found to have 69-70% germination percentage (Misiha and El-Ashry, 1991). While 50 mg/l (50 ppm) of GA3 or BA had relatively lower percentages, 51.06% and 45.80%, respectively (Bhat *et al.*, 1991). Not only seed germination but also seedling growth is known to be regulated by exogenous hormones (Kumaran *et al.*, 1994). Therefore, this research was initiated right from investigating the effect of pre-sowing treatment with GA3 and/or IAA not only on seed germination but also on subsequent seedling growth of *M. grandiflora*.

MATERIALS AND METHODS

Dispersal ripe seeds were gathered from a single *Magnolia grandiflora* tree at the City Park, Plovdiv, Bulgaria. Seeds were extracted, cleaned, air dried at room temperature and stored at 1-5°C in paper bags until they were used. The trial was conducted at the Stat Forestry Nursery, Plovdiv.

At the end of February/ beginning of March, seeds were treated by soaking in 1000, 1500, 2000 and 2500 ppm solutions of gibberellic acid (GA3), indole-3-acetic acid (IAA) and their combination IAA+GA3 for 24 h at room temperature. Seeds soaked in water served as a control. Treated seeds were sown immediately in a 1:1 peat-perlite mixture on raised nursery beds in a randomized complete block design with four replications of 25 seeds per experimental unit. Observations were taken on germination percentage 45 days after sowing and stem height (cm), number of leaves, mean leaf area (cm²) and volume of root system (cm³) were recorded on seven-month-old seedlings. The same procedures were repeated under the same aforementioned conditions for three successive seasons of 2008, 2009 and 2010. The results were subjected to analysis of variance (ANOVA) and differences among means were tested by Duncan's Multiple Range Test (DMRT).

RESULTS AND DISCUSSION

Significant differences were found among pre-sowing treatments of *M. grandiflora* seeds with IAA and/or GA3 at 1000, 1500, 2000 and 2500 ppm regarding all recorded parameters except mean leaf area (Fig 1, Table 1). Using of any of the growth regulators at any level induced higher germination percentage (G%) and better seedling growth comparing to the control (untreated seeds).

With respect to G%, it could be deduced from data illustrated in Fig 1 that G% increased as the concentration of either growth regulators was increased. GA3 alone or in combination with IAA was superior to the sole application of IAA. The highest values of G% was induced by the highest level of either GA3 (66.25%) or IAA+GA3 (62.50%) with no significant difference in between. This percentage is more than twice as much as the control. Although applying IAA alone was inferior

to the former treatments, its highest concentration–2500 ppm improved seed germination up to 50% over the control.

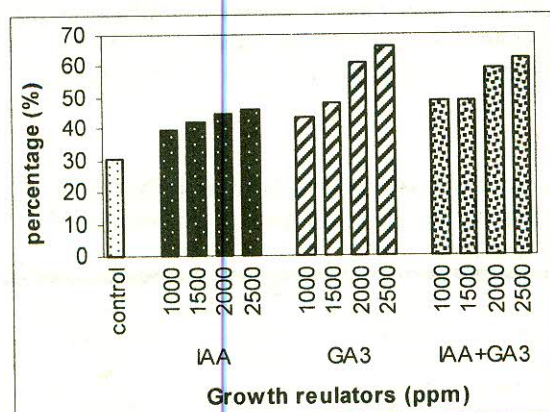


Fig 1. Effect of pre-sowing treatment with IAA and/or GA3 at 1000, 1500, 2000 and 2500 ppm on germination percentage (G%) of *M. grandiflora* seeds.

The preferable effect of GA3 on seeds of *M. grandiflora* was previously reported by several investigators. Bhat *et al.* (1991) found that 50 ppm GA3 had G% of 51.06%. Misiha and El-Ashry (1991) who obtained G% of 69-70% using 1000 ppm GA3 whilst lower concentration (250 or 500 ppm) resulted in 46-56%. It is worth to mention that they used seeds with their testae removed before soaking in GA3. In our study, however, intact seeds were used. This is, probably, the reason of the enhancement effect of GA3 at 1000 ppm in Misiha and El-Ashry (1991) comparing to the same level in our study as the impermeability of the seed coat

could inhibit the promotive effect of GA3 on seed germination.

The promotive effect of growth regulators and pre-sowing chemicals on germination may be attributed to their indirect effect through change in membrane permeability (Shivanna *et al.*, 2007). The positive effect of GA3 on seed germination has been reported on many woody species; *Ginkgo biloba* (West *et al.*, 1970; Jhonson and Wickliff, 1974) *Cassia obtusifolia* (Singh and Murthy, 1987), *Morus nigra* (Koyuncu, 2005), *Abies pindrow*, *Cupressus torulosa* and *Picea smithiana* (Rawat *et al.*, 2006), *Pyracantha crenulata* (Joshi *et al.*, 2010), as has been IAA and IBA on *Picea smithiana* (Singh, 1990), IBA on *Azadirachta indica* (Sivgnanam, 1995).

In spite of the preferable effect of GA3 on G%, seedling growth was significantly improved by the treatment of IAA alone or in combination with GA3 (Table 1). Increasing the growth regulator concentration caused a linear increase in almost all seedling growth characteristics. The application of 2500ppm IAA alone or in combination with GA3 resulted in the best values of stem height (8.53 cm & 8.05 cm, respectively) as well as root volume (3.30 cm³, 3.30 cm³). Mean leaf area insignificantly varied among treatments, except for GA3 at 2000 ppm (18.08 cm²). Though Total leaf area was affected by number of leaves and therefore treatments characterized by the highest number of leaves (2500 ppm IAA/ or IAA+GA3– 3.90/ 3.75, respectively) had the highest total leaf area (68.84/ 66.75 cm², respectively).

Our results are consistent with Kumaran *et al.* (1994), where, application of IAA was effective on seed germination of *Azadirachta indica* accompanied by enhancement in seedling growth compared to GA3 application. Pre-treatment with

IAA at 350 ppm concentration enhanced seed germination and vigour of the *Prosopis Cineraria* seedlings (Shivanna *et al.*, 2007). GA3 treatment was found to increase seedling stem length and fresh weight in *M. grandiflora* (Misiha and El-Ashry, 1991). Other studies showed that application of several plant growth regulators such as auxins may increase the germination ability of seeds and seedling vigour in a wide range of plants (Swaminathan and Srinivasan, 1996; Balestri and Bertini, 2003).

Table 1. Effect of pre-sowing treatment with IAA and/or GA3 at 1000, 1500, 2000 and 2500 ppm on seed germination % (G%) and seedling growth of *M. grandiflora*

Treatment	Stem height	No. leaves	Mean leaf area	Total leaf area	Vol. root system
ppm	cm		cm ²	cm ²	cm ³
Control	4.85 ^B	1.65 ^B	17.83 ^{ab}	29.42	2.05 ^f
IAA					
1000	4.80 ^B	1.98 ^f	17.50 ^{ab}	34.65	2.05 ^f
1500	5.25 ^{ef}	2.55 ^{de}	17.75 ^{ab}	45.26	2.13 ^{def}
2000	7.23 ^c	3.73 ^a	17.35 ^b	64.72	2.53 ^c
2500	8.53 ^a	3.90 ^a	17.65 ^{ab}	68.84	3.30 ^a
GA3					
1000	4.88 ^B	2.15 ^f	17.83 ^{ab}	38.33	2.08 ^{ef}
1500	4.85 ^B	2.38 ^c	17.80 ^{ab}	42.36	2.25 ^{def}
2000	5.05 ^{fg}	2.98 ^c	18.08 ^a	53.88	2.30 ^{de}
2500	5.18 ^{ef}	3.18 ^b	17.73 ^{ab}	56.38	2.33 ^{cd}
IAA+GA3					
1000	4.83 ^B	2.65 ^d	17.28 ^b	45.79	2.28 ^{def}
1500	5.35 ^c	2.98 ^c	17.33 ^b	51.64	2.15 ^{def}
2000	6.70 ^d	3.35 ^b	17.88 ^{ab}	59.90	2.75 ^b
2500	8.05 ^b	3.75 ^a	17.80 ^{ab}	66.75	3.30 ^a

Values in the same column followed by the same letter are not significantly different at 1% level by DMRT.

Information on the effects of auxins on seed germination is still limited, but there is evidence that exposure of seedlings of different plants to auxins may influence root development (Reed *et al.*, 1998; Balestri and Bertini, 2003). The effect of different auxins on seed germination process includes direct and indirect evidences indicating the involvement of auxins in the seed germination (Chiwocha *et al.*, 2005).

CONCLUSIONS AND FUTURE PROSPECTIVE

It could be inferred from the above results that germination of *M. grandiflora* was enhanced by either GA3 or IAA+GA3 and seedling growth was enhanced by either IAA or IAA+GA3 at the highest concentration. We recommend that the

advantage of both high germination % and vigorous seedlings could be guaranteed using a combination of GA3 and IAA at 2500 ppm.

ACKNOWLEDGEMENT

This research has been supported by Erasmus Mundus External Cooperation Window (EMECW) programme, funded by the European commission, in cooperation with Agricultural University, Plovdiv, Bulgaria. The authors express sincerest appreciation to Mr. krasimiz Kissyov, the manager of the State Forestry Nursery, Plovdiv for his assistance with this research.

REFERENCES

1. Balestri, E. and S. Bertini 2003. Growth and development of *Posidonia oceanica* seedlings treated with plant growth regulators: Possible implications for meadow restoration. *Aquat. Bot.* 76: 291-297.
2. Barbour, j. 2008. *Magnolia* L. In: *Woody Plant Seed Manual*. USDA Forest Service, Agriculture Handbook 727, pp. 700-705.
3. Bhat, S.M.; A.Q. Jhon and A.H. Lone 1991. Propagation of *Magnolia grandiflora* through seed. *Progressive Hort.* 23 (1-4): 30-33.
4. Blaisdell, R.S.; J.Wooten and R.K. Godfrey 1973. The role of magnolia and beech in forest processes in the Tallahassee, Florida, Thomasville, Georgia area. *Proc. 13th Ann. Tall Timbers Fire Ecol. Conf.*, pp 363-397.
5. Callaway, D.J. 1994. *The World of Magnolias*. Portland, OR: Timber Press.
6. Chiwocha, S.D.S.; A.J. Cutler; S.R. Abrams; S.J. Ambrose; J. Yang; A.R.S. Ross and A.R. Kermode 2005. The *etr1-2* mutation in *Arabidopsis thaliana* affects the abscisic acid, auxin, cytokinin and gibberellin metabolic pathways during maintenance of seed dormancy, moist-chilling and germination. *Plant J.* 42: 35-48.
7. Dirr, M.A. 2002. *Dirr's Trees and Shrubs for Warm Climates*, an illustrated encyclopedia. Timber Press Inc., Portland, Oregon, pp 216-218.
8. Evans, CR. 1933. Germination behaviour of *Magnolia grandiflora* L. *Bot.Gaz.* 94: 729-754.
9. Gary, W. 1994. *Magnolias*. The Environmental Horticulture Department, Florida Cooperative Extension Service, Institute of Food and Agricultural Sciences, University of Florida. Circular 1089. (<http://edis.ifas.ufl.edu>).
10. Halls, L.K. 1977. Southern Magnolia - *Magnolia grandiflora* L. In *Southern fruit-producing woody plants used by wildlife*. USDA Forest Service, pp 196-197.
11. Johnson, M.I. and J.L. Wickliff 1974. Seed germination in *Ginkgo biloba* L. I. Influences of cold treatment, gibberellic acid and red light. *Arkansas Academy Sci. Proc.*, vol. XXVIII: 34-36.
12. Joshi, M.R.S.C.; Debarati; Preeti; S.S. Parihar and HCS. Negi 2010. Effect of GA3 on seed germination of *Pyracantha crenulata*. *New York Sci. J.* 3(9): 55-57.
13. Koyuncu, F. 2005. Breaking seed dormancy in black mulberry (*Morus nigra* L.) by cold stratification and exogenous application of gibberellic acid. *Acta Biol. Cracov. Bot.* 47 (2): 23-26.

14. Kumaran, K.; M. Palani; R. Jerlin and C. Surendran 1994. Effects of growth regulators on seed germination and seedling growth of neem (*Azadirachta indica*) J. Trop. For. Sci. 6 (4): 529-532.
15. Mihailo, G. and S. Dragana 2004. Breaking embryo dormancy in evergreen *Magnolia*, aromatic samach and service-tree by naked stratification. Proc. Intl. sci. conf., Sofia, Bulgaria, 1-5 October 2003, 2: 139-145.
16. Misiha, A. and A. El-Ashry 1991. Seed germination and seedling growth of *Magnolia grandiflora* L. Bull. Fac. Agric., Cairo Univ. 42 (3): 869-879.
17. Rawat, B.S.; C.M. Sharma and S. K. Ghildiyal 2006. Improvement of seed germination in three important conifer species by Gibberellic acid (GA3). *lyonia*, 11 (2): 23-30.
18. Shi, S.; Y. Zhong and W.A. Hoch 2002. Distribution and Commercial Cultivation of *Magnolia*. In: Sarker, S.D. and Y. Maruyama (eds.). *Magnolia: The genus Magnolia*. Taylor and Francis, London, pp 156-180.
19. Shivanna, H.; H.C. Balachandra and N.L. Suresha 2007. Influence of Growth Regulators and Pre-Sowing Chemicals on Germination and Growth Parameters of *Prosopis Cineraria* (L.) Druce. Karnataka J. Agric. Sci., 20 (2):328-329.
20. Singh, C. and Y.S. Murthy 1987. Effect of some growth regulators on the seed germination and seedling growth of *Cassia obtusifolia*. *Acta Bot Indica* 15: 77 - 79.
21. Singh, V. 1990. Influence of IAA and IBA on seed germination of spruce. *Indian Forester* 116: 450-453.
22. Sivagnanam, K. 1995. Growth biology, reproductive strategy and seed production spectrum in Neem (*Azadirachata indica* A Jus.), Ph.D. Thesis, Tamilnadu Agric. Univ., Coimbatore.
23. Swaminathan, C. and V.M. Srinivasan 1996. Seedling invigoration through plant growth substances in teak (*Tectona grandis*). J. Trop. For. Sci. 8: 310-316.
24. West, W.C.; F.J. Frattarelli and K.J. Russin 1970. Effect of stratification and gibberellin on seed germination in *Ginkgo biloba*. Bull. Torrey Bot. Club, 97 (6): 380-384.