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Propagation of Ishwarmul (*Aristolochia indica*) on Culture Medium

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Different culture mediums were used for *in vitro* plant regeneration through different explants viz., Cotyledons, Stem segment and Leaf base in Ishwarmul. For *in vitro* regeneration of Ishwarmul, seven media were tested out of which five were based on Murashige and Skoog's medium (MS), one on Gamborg's B₅ and one on White's media for their response on induction of callus from Stem segment, Leaf base and Cotyledons. Media based on MS responded well irrespective of explants used. Leaf base proved to be the best for callusing percent and fresh weight of callus in MS based media. The highest callusing efficiency was observed in MS medium with full strength of MS salts, 5 mg/l 2,4-D + 1.6mg/l BAP. Observation on shoot regeneration capacity suggested that MS medium contained MS salt +5mg/l 2,4-D+1.6 BAP has shown superior performance and was highly effective in inducing multiple shoots from callus. The MS medium with half strength of basic MS salts in combination with 1.2 mg/l BAP/ 0.6 mg/l IBA was also found to be the best for root regeneration. The Darkness supports the fast root regeneration in Ishwarmul. The rooted plantlets were successfully transplanted in pots in poly house, after 25 days, these plants with pots were transferred in to green house for further acclimatization under natural environments. The survival rate was 63%.

Introduction

Aristolochia indica Linn., a divine herb which can fight against any poisonous bite. Indian Birthwort, Snakeroot, Ishwarmul, Iswarballi are the common names of *Aristolochia indica*. It is a native of India and belongs to family Aristolochiaceae, distributed throughout the tropical, subtropical and Mediterranean countries. In India it is found in all most all types of forest throughout the country particularly in low hills and moist plains but due to indiscriminate harvesting from forest now it became a rare endangered species. It is a twining herb, semi woody, leaves are cordate or ovate, exstipulate; flowers are irregular, often offensively smelling, perianth is globose with a purple dilated and trumpet-shaped mouth with a strap-shaped brown purple appendage or lip behind; fruit is a sub globose capsule. It is used in India to induce vomiting and to treat poisons, intestinal parasite, swelling, menstrual irregularities, dropsy, low appetite, ulcers and fever (Dey, 2011). The roots of plant are used as antidote in scorpion sting, bites of poisonous insects and snake bite. This plant is used both internally and externally. For white leprosy, the roots are rubbed with honey. The plant possesses emmenagogue, abortifacient, anti-spermatogenic,

anti-fertility, anti-arthritic, anti-inflammatory, antiperiodic, diuretic and anti-bilious properties. The leaves of plant are applied externally in skin diseases. Though the plant is useful in many ways, it is also nephrotoxic and carcinogenic. Over dosage of the plant components may lead to serious complications. The propagation of ishwarmul is done mainly through the seeds. Its seeds have poor germination and need specific care to grow, hence an effort has been made in this investigation to draw a protocol for In-Vitro plantlets of ishwarmul.

Material and Methods

The research experiment was carried out at Tissue Culture Laboratory, College of Agriculture, (Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya), Indore, (M.P.) during the year 2015-2016. The experiment was conducted in Completely Randomized Design (CRD) with five replications for each explant under each medium. Explants viz., Stem segment, Leaf base and Cotyledons were collected from pretreated mother plants of ishwarmul available in Medicinal plant nursery of College of Agriculture Indore. Seven combinations and concentrations of hormones with full strength basic salts and vitamins of MS, B₅ and White's media were used to test the response of different explants for organogenesis, shoot regeneration and root regeneration capacity. Aseptic environment was maintained during the investigation period. Observations were recorded on various explants viz., Cotyledons, Stem segment and leaf base for induction of callus, regeneration on different culture media, callusing percentage, weight of callus and regeneration capacity of callus into shoots and roots.

Result and Discussion

All the media under investigation responded for callusing percentage and fresh weight of callus irrespective of explants used, except B₅ and White's media. All the three explants viz., Stem segment, Leaf base and Cotyledons showed their highest level of callusing percent and fresh weight of callus on M₃ medium. On the basis of callusing efficiency M₃ media was the best medium among all the media tested in the present investigation. The M₃ media was supported with full strength of MS salts, 5mg/l 2,4-D + 1.6mg/l BAP. It was also reported earlier that the presence of 2,4-D alone or with other hormones in MS basal media enhanced the callusing efficiency. Sahaya et al. (2011) observed the highest percentage of callus induction (82.3 ± 0.57) from internodal segment on Murashige and Skoog's medium (MS) supplemented with 1.5 mg/l of 2,4-D in *Aristolochia bracteata*. Wani et al. (2010) studied on *Tridax procumbens* revealed that the Leaf and apical bud explants showed maximum callus induction by using MS media with the combination of 2, 4-D 0.5 mg/lit and BAP 0.5 mg/lit. Soniya (2006) and Wani et al. (2010) suggested that the combination of 2,4-D and BAP in different concentrations may enhance the callusing efficiency of medium.

In present investigation three explants viz. Cotyledons, stem segment and leaf base were evaluated for their effectiveness for induction of callus on different media. The leaf base was proved as the best explant on the basis of callusing percentage and averaged fresh weight of callus. Least effective explant was cotyledon.

The evaluation of media for shoot regeneration capacity suggested that medium M₃ contained MS salt +5mg/l 2,4-D+1.6 BAP has shown superior performance and was highly effective in inducing multiple shoots from callus. The efficiency of M₃ medium may be due to the presence of high concentration of 2,4-D and BAP. The efficacy of 2,4-D and BAP in combination or alone in MS media for induction of shoot has already been reported by number of scientists. Siddiqui et al. (2006) in their two different studies on ishwarmul suggested that the highest percentage of shoot regeneration was obtained in MS medium fortified with 2.5 mg/l Kn+ 1.0

mg/l BAP and 1.0 mg L-1 BAP + 2.5 mg L-1 NAA respectively. Biswas et al. (2007) and Sahaya et al. (2011) also suggested that combination of BAP and NAA in MS media enhanced the shoot multiplication and proliferation in calli derived from nodal and inter-nodal segment. The MS medium with half strength of basic MS salts in combination with 1.2 mg/l BAP/ 0.6 mg/l IBA was also found to be the best for root regeneration. The Darkness supports the fast root regeneration in Ishwarmul. The rooted plantlets were successfully transplanted in pots in poly house, after 25 days, these plants with pots were transferred in to green house for further acclimatization under natural environments. The survival rate was 63%. Biswas et al. (2007) and Chandraprabha et al. (2010) reported in *Aristolochia atagala* Champ that excised shoot roots were cultured on half-strength MS medium containing 0.5 mg/l and 1.0 mg/l IBA respectively. While Remya et al. (2016) observed the well developed shoots when rooted on MS medium supplemented with indole acetic acid (1.5 μ M), Kinetin (1.5 μ M) and 6-benzylaminopurine (0.5 μ M).

Conclusion

Ishwarmul is an important medicinal plant and it can be multiplied by *in-vitro* culture. Among Murashige and Skoog (MS), Gamborg's B₅ and White's media only MS media were responded for callusing percentage and fresh weight of callus. Among the MS media, the callusing efficiency of media (M₃) which supported with full strength of MS salts, 5 mg/l 2, 4-D + 1.6 mg/l BAP was highest. The callusing percent and fresh weight of callus of all the three explants i.e., Stem segment, Leaf base and Cotyledons were high on M₃ medium. The shoot regeneration capacity of MS media M₃ was proved to be the best irrespective of explants used but the highest number of shoots were induced through the leaf base. The MS medium with half strength of basic MS salts in combination with 1.2mg/l BAP/ 0.6 mg/l IBA was also found to be the best for root regeneration. The Darkness supports the fast root regeneration in Ishwarmul.

References

- Biswas A, Bari MA, Mohashweta R and Bhadra SK (2007). In-vitro Regeneration of *Aristolochia tagala* Champ. A rare Medicinal Plant of Chittagong Hill Tracts. J. Bio. Sci. Res. **15**; 63-67.
- Chandraprabha A and Rama Sabbu R (2010) Micropropagation of *Aristolochia atagala* Champ. A Rare and Endemic Medicinal Plant from Western Ghats. J. Bio. Sci. Res. **1**(2):70-73.
- Dey Abhijit and De Nath Jitendra (2011). *Aristolochia indica* L.: A Review. Asian J. Plant Sci., **10**: 108-116.
- Remya M, Narmathabai V and Mutharaian VN (2016). Changes in bioactive components of *Aristolochia tagala*. Cham, a rare species of medicinal importance during its in vitro development through direct regeneration. Department of Biotechnology, Vinayaka Missions University, Aarupadai Veedu Institute of Technology, Kancheepuram Tamil Nadu, India.
- Siddique NA, Kabir MH and Bari MA (2006). Comparative in vitro study of plant regeneration from nodal segments derived callus in *Aristolochia indica* Linn. and *Hemidesmus indicus* (L.) R. Br. Endangered Medicinal Plants in Bangladesh. J. Plant Sci. (1): 106-118.)
- Soniya KV and Sujitha M (2006). An efficient in vitro propagation of *Aristolochia indica*. Biol. Plant., (**50**): 272-274.
- Wani Minal, Pande Snehal and More Nitin (2010). Callus Induction Studies in *Tridax procumbens* (L). Int. J. of Biotech Applications 2 (1): 11-14.