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Micropropagation strategies for conservation of critically endangered medicinal plant *W. coagulans*

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Withania coagulans is an immensely important medicinal plant. Over the years the plant has become critically endangered due to extremely poor germination along with low propagation rate in nature and over exploitation for various purposes. The present study reports an efficient method of mass propagation of *W. coagulans* through culture of cotyledonary segment and also comparative biochemical analysis of mother and tissue culture raised plants. Seed of *W. coagulans* were pretreated with 10% HCl solution for 10 minutes to enhance germination rate. Germinated seeds were surface sterilized and excised cotyledons were cultured onto MS+NAA (8 μ M). Within 3-4 weeks of culture extensive callusing was obtained. Callus was further sub-cultured onto MS medium fortified with different PGR among which TDZ supplemented medium resulted in regeneration of shoot buds. After 8 weeks of culture a maximum of 35 shoot were regenerated onto MS+8 μ M TDZ. Regenerated shoots were elongated onto basal MS medium and *in vitro* rooting was achieved onto $\frac{1}{2}$ MS+20 μ M IBA. About 68.8% plants survived during the process of acclimatization. GC-MS analysis of methanolic extract of leaves obtained from wild and *in vitro* regenerated plant of *W. coagulans* revealed presence of 45 and 56 phytochemical compounds respectively. 1-Pentadecanamine, N, N-dimethyl; Palmitic acid, Methyl ester; Phthalic acid; butyltridecyl ester; 9-octadecenoic acid methyl ester (E); Betulin; Hexadecanoic acid, 2-hydroxy-1-(Hydroxy methyl) ethyl ester; Stigma sterol; Fuco sterol; Anthracane; Benzyl Benzoate and Tetradecanoic acid were major compounds present in mother plant whereas 9, 12 octadecadienoic acid (Z, Z); Hexadecanoic Acid, 2-hydroxy-1-(hydroxyl methyl) ethyl ester; n-hexadecanoic acid; Tetradecanoic acid; 1-Pentadecanamine, N, N-dimethyl; Benzoic acid, 4-ethoxy-ethyl ester; octanoic acid; Lycopene; gamma-tocopherol; Cholesterol and Stigmasta-5, 23-Dien-3-ol (3 Beta) were the major compounds present in tissue culture raised plants. Hence, the present study establishes tissue culture as an effective technique for mass propagation as well as enhanced production of bio-metabolites.

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SOCS5 in malaria vector: Annotation, domain organization and phylogenetic analysis in 18 *Anopheles* species

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Insects are amongst the most primitive creature and their encounter with varied groups of pathogen has led to the enormous diversity in their genome. Such diversity is seen to be more pronounced in *Anopheles*, a deadly vector of Malaria. With the advent of unannotated genome sequence of 18 *Anopheles* species, it will be feasible to understand the intricacy of this diverged genome. Here we identify and annotate SOCS gene, which known to be involved in immunity, growth and development of mosquitoes. SOCS gene from *An. culicifacies* was cloned and sequenced and was used to retrieve SOCS gene from unannotated genome of other *Anopheles* species. Sequence analysis of all *Anopheles* genome database confirmed the presence of three exons separated by two introns (~500 bp in the N terminal region and ~70-80 bp in the SH2 domain). SOCS in all *Anopheles* share a similar domain organization, with a central SH2 domain and a conserved C-terminal SOCS box, the N-terminal domains of SOCS proteins vary in length and amino acid sequence. The SH2 and SOCS box domains showed 99-100% similarity with each other and 80-85% similarity while comparing the whole SOCS protein of *Anopheles*. It indicates that SH2 and SOCS box domains are highly conserved during evolution due to their important role in receptor signaling. These observations indicate that SOCS N-terminal amino acids identity is solely similar, rather limited to, *Anopheles* SOCSs. This variability may indicate that all the domains of AcSOCS experienced differential selection pressures and it provides the evidence that N-terminal domain is under least selection pressure. Phylogenetic analysis of SOCS gene of all *Anopheles* mosquito species reveals that SOCS of same subgenus are similar to each other but diverged from other subgenus mosquitoes. This suggested that SOCS follows the same taxonomical pattern in which *Anopheles* are classified.

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