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## Callus induction in Marigold-Nemo Mix (Tagetespatula L) required for diesel toxicity evaluation

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Three-week-old leaf explants of in vitro germinated seedlings of Tagetespatula were used to develop a callus induction medium suitable for diesel toxicity evaluation. The adaxial side of the excised leaves was placed on the basal Murashige and Skoog (MS) medium, supplemented with various combinations of different auxins including 1 naphthalene acetic acid (NAA), 2, 4-dichlorophenoxyacetic acid (2, 4-D) and picloram (PIC) with different concentrations of 6 benzyladenine (BA). High frequency of the induction of two morphologically distinct calli resulted dependent on the combination of auxin to cytokinin in the medium. Creamy yellowish friable calli were induced on the basal medium supplemented with the combination of 5.71  $\mu$ M PIC and 4.44  $\mu$ M BA but the callus changed to dark brown during subculture on to the medium of the same composition. Green compact callus was formed on the basal MS medium supplemented with 1.82  $\mu$ M NAA and 66.6  $\mu$ M BA. This callus texture and morphology were maintained during subculture on the medium of the same composition. The combination of 13.5  $\mu$ M 2, 4-D and 4.4  $\mu$ M BA had the lowest effect on the callus induction. The results show interaction effects of plant growth regulators with young leaf explants for callus initiation. The compact green callus formed from the MS medium supplemented with 1.82  $\mu$ M and 66.6  $\mu$ M was selected for use in the diesel toxicity evaluation due to their relative ease of manipulation compared to the friable.

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