

## ABSTRACT

The present study deals with tissue culture of *Punica granatum* L. Nodal stem explant was found to be the best explant which gave the highest shoot number ( $3.8\pm 0.2$ ) on MS medium supplemented with 1.5 g/L BAP, while shoot tip explant ( $1.9\pm 0.1$ cm) on MS medium with 2.0 mg/L BAP gave the best shoot height. MS supplemented with 1.5 mg/L BAP was observed to give the highest shoot production ( $4.1\pm 0.2$ ) with combination of 40 g/L of sucrose in shoot tip explant. Complete plant regeneration was achieved for both shoot tips and nodal stem explants cultured on MS medium supplemented with 1.5 mg/L BAP. In scanning electron microscopy studies, *in vitro* leaf exhibited higher stomata density compared to *in vivo* leaf sample. No irregular characteristics were found in *in vitro* leaf and stem samples when compared to the *in vivo* leaf and stem samples. In callus production from *P.granatum* L., the highest callus weight was obtained from root explants cultured on MS supplemented with 1.5 mg/L BAP and 1.5 mg/L NAA ( $0.48\pm 0.08$ g) and the highest callus weight using other auxins and cytokinins (IAA, IBA, 2,4-D and Kinetin) was obtained from stem explants supplemented with 2.0 mg/L IBA ( $0.26 \pm 0.01$ g). The highest percentage of radical scavenging activity was obtained from *in vivo* leaf extract with 67.4% at 100  $\mu$ g/ml of sample concentration. Peel extract exhibited the highest percentage of radical scavenging activity of all samples (77.6%) and also the strongest  $IC_{50}$  value (2.21 mg/ml), at 100  $\mu$ g/ml of sample concentration. Phytochemical contents that were presence in *in vivo* *P.granatum* L. sample were also presence in *in vitro* samples such as saponin, tannins, flavonoids and reducing sugar. Between peel and seed extracts of *P.granatum* L., terpenoid was detected but tannins and reducing sugars were detected only in peel extract.

## ABSTRAK

Kajian ini melibatkan kultur tisu ke atas *Punica granatum* L. Didapati eksplan batang bernod adalah eksplan yang terbaik dan menghasilkan jumlah pucuk yang tertinggi ( $3.8 \pm 0.2$ ) manakala eksplan pucuk ( $1.9 \pm 0.1$ cm) memberikan panjang pucuk yang tertinggi. Media MS diperkaya dengan 1.5 mg/L BAP memberikan keputusan terbaik untuk jumlah pucuk tertinggi ( $4.1 \pm 0.2$ ) dengan kombinasi 40 g/L sukrosa menggunakan eksplan pucuk. Regenerasi tumbuhan lengkap didapati apabila eksplan pucuk dan batang bernod dikultur di atas media MS yang ditambah dengan 1.5 mg/L BAP. Di dalam kajian mikroskopi elektron pengimbas, daun *in vitro* telah menunjukkan ketumpatan stomata yang lebih tinggi jika dibandingkan dengan sampel daun *in vivo*. Tiada sifat luar biasa dapat dikesan dari sampel daun dan batang *in vitro* apabila dibandingkan dengan sampel daun dan batang *in vivo*. Dalam produksi kalus daripada *P.granatum* L, berat kalus tertinggi didapati dari eksplan akar apabila dikultur di atas MS media diperkaya dengan 1.5 mg/L BAP dan 1.5 mg/L NAA ( $0.48 \pm 0.08$ g) dan berat tertinggi kalus dari hormon auksin dan sitokinin yang lain pula diperolehi dari eksplan batang diperkaya dengan 2.0 mg/L IBA ( $0.26 \pm 0.01$ g). Peratusan cerapan radikal tertinggi di antara kesemua sampel *in vitro* dan *in vivo* diperolehi dari ekstrak daun *in vivo* dengan 67.4% pada 100  $\mu$ g/ml kepekatan sampel. Ekstrak kulit mempunyai peratusan cerapan radikal tertinggi di antara semua sampel (77.6%) dan juga nilai Setengah Kepekatan Perencatan Maksimum, atau  $IC_{50}$  terendah (2.21), pada kepekatan sampel 100  $\mu$ g/ml. Kandungan fitokimia seperti saponin, tannin, flavonoid dan gula penurun yang wujud di dalam sampel *in vivo* daun dan batang *P.granatum* L. didapati juga di dalam sampel daun dan batang *in vitro*. Di antara ekstrak sampel kulit dan biji benih dari *P.granatum* L., terpenoid dikesan pada kedua-dua ekstrak, manakala tannin dan gula penurun hanya dikesan pada ekstrak kulit.

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## ABBREVIATIONS

BAP	- 6-Benzylaminopurine
NAA	- $\alpha$ -Naphthaleneacetic acid
IBA	- Indole-3-butyric acid
IAA	- Indole-3-acetic acid
2,4-D	- 2,4-dichlorophenoxyacetic acid
Kinetin	- 6-furfurylamino purine
cm	- centimeter
m	- meter
ml	- mililitre
g	- gram
g/L	- gram per liter
Mg/L	- milligram per liter
°C	- degree celcius
v/v	- volume per volume
w/v	- weight per volume
NaOH	- Natrium Hydroxide
HCl	- Hydrochloric acid
DMRT	- Duncan's Multiple Range Test
mM	- miliMolar
$\mu$ M	- microMolar
MS	- Murashige and Skoog
Nm	- nanometer
PVA	- PolivinyI-Alcohol
LDL	- Low-density lipoprotein
DPPH	- 2,2-diphenyl-1-picrylhydrazyl
IC <sub>50</sub>	- Half Maximal Inhibitory Concentration