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Production of callus from different parts of *Balanites Aegyptiaca* and study the effect of callus extract on different cell lines

Presented by

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ABSTRACT

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Title of the thesis: Production of callus from different parts of *Balanites Aegyptiacae* and study the effect of callus extract on different cell lines.

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Study objective: the aim of this study to evaluate the role of plant growth regulator (PGR) and different elicitors *in vitro* to induce the enhancement of bioactive compound (Diosgenin) production in callus culture from different parts of *Balanites Aegyptiacae* plant and study the effect of diosgenin extract free and functionalized with IONPs synthesized by coprecipitation method on cell viability of HepG2 carcinoma and A549 lung carcinoma and determination of P53 levels in HepG2. **Results and conclusion:** the highest diosgenin content obtained from root part culture elicited with tryptophan 300mg/l was 3.12 mg/100g DW, the cytotoxic activity of the root callus extract supplemented with tryptophan was determined against HepG2 (hepatic carcinoma), A549 (Lung carcinoma cell lines) and the result was 51.26 $\mu\text{m/ml}$, 28.9 $\mu\text{m/ml}$ respectively. Also the results of this study showed that the FTIR and HRTEM confirm the formation of IONPs with size ranging from (6-14)nm, FTIR confirm the conjugation chemistry between diosgenin and IONPs via citric acid as a linker by the presence of iron oxide signature peak at 501 cm^{-1} and signature peak of diosgenin at 2927.06, 1050.56 and 1640.32 cm^{-1} . also our results demonstrated that the cytotoxic effect of functionalized diosgenin IONPs-D increased against HepG2 and A549 and became IC50 at 20.38 μg and 0.795 μg respectively, Our study reported that Diosgenin free and functionalized induced cell cycle arrest and apoptosis via activation of P53 expression to be 2.3 and 3.4 respectively when compared with control.

Keywords: *B.Aegyptiacae*, callus induction, diosgenin bio-production, Elicitation, IONPs synthesis, functionalized diosgenin, hepatocellular carcinoma, A549 lung carcinoma.

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List of Abbreviation

B. Aegyptiacae	<i>Balanites Aegyptiacae</i>
MS Media	Murashige and Skoog medium
NAA	Naphthaline acetic acid
2,4D	Dichlorophenoxy acetic acid
BAP	Benzyl amino purine
HPLC	High performance liquid chromatography
IONPs	Iron oxide nanoparticles
IONPs-CA	Citrate Capped iron oxide nanoparticles
IONs-D	Diosgenin functionalized iron oxide nanoparticles
HRTEM	High resolution transmission electron microscopy
FTIR	Fourier transform infrared
ATCC	American Tyoe culture collection
A549 NSCLC	<i>Human lung carcinoma</i>
HepG2	Hepatocellular carcinoma
KBr	Potassium bromide
CCL4	Carbon tetra chloride
GRD	<i>Glutathione reductase</i>
MDA	Malondialdehyde
DPPH	Diphenyl-2,4,6 trinitrophenyliminoazanium
2-CEPA	Ethylene generating agent 2- chloro ethylphosphonic acid
HMGR	<i>3-hydroxy 3- methylglutaryl-coenzyme A reductase</i>
CAS	CRISPER
HCC	Hepatocellular carcinoma
JAK 1	Janus Kinase Inhibitor
c-SR	Calcium sensing receptor

MDA-MB-231	Epithelial like cell line
<i>TOR signaling</i>	<i>Target of rapamycin</i>
Vav2	Guanine nucleotide exchange factor
Cdc42	Cell division control protein 42 homolog
TNF-κB	Tumor necrosis factor beta
STAT3	Signal transducer and activator
U87MG	Human primary glioplastoma cell line
MRI	Magnetic resonance imaging
FCS	Fetal Calf serum
RPMI 1640	L-glutamine , phenol red, reduced serum
MTT	3-(4,5 dimethylthiazole-2-yl)-2,5 diphenyl tetrazolium bromide
dCTP	Deoxycytidine triphosphate
dGTP	Deoxyguanosine triphosphate
dTTP	Thymidine triphosphate
PGR	Plant growth regulator
TNF-α	Tumor necrosis factor alfa