SUMMARY AND CONCLUSIONS

Chemoprevention is regarded as one of the most promising and realistic approaches in the prevention of cancer. Several marine bioactive compounds present in phlorotannins have revealed their cancer curative potential on hepatocellular carcinoma. Dieckol is a naturally occurring phlorotannins found in seaweeds. To validate and authenticate the protective role of dieckol during NDEA-induced hepatocarcinogenesis in wistar rats by analyzing the phase I cytochrome P450 and phase II glutathione S-transferase (GST) enzymes, extent of redox status and liver marker enzymes and also evaluated the lipid peroxidation and antioxidant were also investigated. In addition, examined the molecular mechanisms by which dieckol inhibited growth, inflammation, metastasis and angiogenesis of NDEA induced hepatocellular carcinoma in rats.

**Effect of Dieckol during hepatocarcinogenesis**

Administration of NDEA induced hepatocellular carcinoma (HCC) as evidenced by changes in morphological, histological and ultrastructural architecture, increased activity of cytochrome P450 and decreased activity of GST, enhanced lipid peroxidation and liver marker enzymes and decreased antioxidant status.

Metabolic biotransformation of NDEA by cytochrome P450 enzymes produces O\_6-ethyldeoxyguanosine and O\_4 and O\_6-ethyldeoxythymididine, active ethyl radical metabolite (CH\_3 CH\_2\^+) which is responsible for initiation of carcinogenesis. A subsequently reactive product of NDEA can be detoxified by phase II enzymes such as GST. Increased activity of cytochrome P450 and decreased activity of GST observed in the present study provide evidences for the development of HCC in NDEA treated animals. Our results provide evidences that dieckol could act as a dual acting agent by decreasing phase I cytochrome P450 and increasing phase II enzyme GST and could block initiation of NDEA-induced hepatocarcinogenesis. Modulation of
XMEs by dieckol can be correlated with equilibrium between oxidant and antioxidant balance, which is tilted towards the antioxidant side.

The reactive metabolite of NDEA and the free radicals generated by P-450-dependent enzymes disturb antioxidant status and facilitate lipid peroxidation (LPO) and decreased antioxidant status thereby producing several toxic products, such as malondialdehyde (MDE) and 4-hydroxynonenal. These products can attack cellular targets including DNA, thereby inducing mutagenecity and carcinogenicity. Dieckol inhibits lipid peroxidation and decreases the generation of free radicals by enhancing antioxidant status as evidenced by increasing the levels of SOD, catalase and GPx and non-enzymatic antioxidants, vitamins E and D, and GSH. Generally liver damage induced by NDEA reflects instability of liver cell metabolism and membrane instability subsequently caused distinctive changes in the serum enzyme activities. Upon liver injury, liver marker enzymes (AST, ALT, and ALP) enter into the circulatory system due to altered permeability of membrane. Administration of Dieckol attenuated NDEA induced hepatocarcinogenesis, as shown by the reverted activities of AST, ALT, ALP, LDH, and decreased the concentration of bilirubin and α-fetoprotein.

Our results suggest that pre and post-treatment of dieckol effectively suppressed the NDEA-initiated hepatocarcinoma and its preneoplastic lesions by modulating xenobiotic-metabolizing enzymes (XMEs) and alleviating lipid peroxidation through scavenging free radicals and enhancing antioxidant status and reverting liver marker enzymes. This could be the initial step in the prevention of HCC in Wistar rats.
Modulatory effect of dieckol on markers of cell proliferation, apoptosis, invasion, metastasis and angiogenesis

NDEA usually causes genomic damage in exposed cells. As a consequence, the damaged cells may be triggered to proliferate with genomic damage, leading to the formation of cancerous cells that showed apoptosis evasion, increased cell proliferation, angiogenesis, invasion and metastasis. Enhanced cell proliferation and apoptosis evasion in NDEA-induced hepatocarcinogenesis were associated with imbalance in pro-apoptotic and anti-apoptotic proteins together with upregulation of PCNA and downregulation of caspase-3.

Treatment of dieckol decreased the expression of PCNA and Bcl-2 and increased the expression of Bax and caspase-3, which showed the antiproliferative and apoptotic effects, respectively. Administration of NDEA increased the tumor expression of NF-κB, COX-2, VEGF, MMP-2, and MMP-9 that was correlated with more aggressive lesions that sustains inflammation, tumour growth, progression and metastasis. Treatment of dieckol suppressed various NF-κB regulated gene products including VEGF, proinflammatory enzyme COX-2, MMP-2 and MMP-9 which are associated with the inhibition of tumour angiogenesis, invasion and metastasis and thus resulting in suppression of liver tumour induced by NDEA. Our results suggest that dieckol could acts as a legitimate agent to inhibit cell proliferation, inflammation, angiogenesis, invasion and metastasis and induce apoptosis in cancer chemoprevention.

CONCLUSION

Our findings suggest that dieckol is inhibits chemically induced carcinogenesis through modulation of XMEs, cellular redox status, liver marker enzymes, inhibition of cell proliferation, induction of apoptosis, inhibition of inflammation, invasion, metastasis, angiogenesis which are against liver cancer development. Dieckol could play a major role by interacting between different types of genes
and enzymes. Molecular mechanisms involved in prevention of cancer include carcinogen inactivation, antiproliferation, cell cycle arrest, induction of apoptosis, inhibition of angiogenesis, antioxidation and reversal of multidrug resistance and or a combination of these mechanisms. Therefore further studies are required to address the role of dieckol role by interacting between different types of genes and enzymes involved in various mechanisms of chemoprevention and/or carcinogenesis.
References
REFERENCES


Arii S, Mise M, Harada T, Furutani M, Ishigami S, Niwano M, Mizumoto M, Fukumoto M and Imamura M. Overexpression of


Gaosawara S, Yano H, Momosaki S, Nishida N, Takemoto Y, Kojiro S and Kojiro M. Expression of matrix metalloproteinases (MMPs)


Kang KA, Lee KH, Chae S, Zhang R, Jung MS, Ham YM, Baik JS, Lee NH and Hyun JW. Cytoprotective effect of phloroglucinol on


against \( H_2O_2 \)-induced oxidative stress in murine hippocampal HT22 cells, *Environ. Toxicol. Pharmacol.*, 2012; 34: 96-105.


Kelly PN and Strasser A. The role of Bcl-2 and its pro-survival relatives in tumourigenesis and cancer therapy. *Cell Death Differ.*, 2011; 18: 1414-1424.


Lee SH, Han JS, Heo SJ, Hwang JY and Jeon YJ. Protective effects of dieckol isolated from *Ecklonia cava* against high glucose induced oxidative stress in human umbilical in endothelial cells. *Toxicol. Invitro.*, 2010; 24: 375-381.


Li Y, Qian ZJ, Ryu B, Lee SH, Kim MM and Kim SK. Chemical components and its antioxidant properties *in-vitro*: an edible


Mesallamy HO, Metwally NS, Soliman MS, Ahmed KA and Abdel Moaty MM. The chemopreventive effect of *Ginkgo biloba* and *Silybum marianum* extracts on hepatocarcinogenesis in rats. *Cancer Cell Int.*, 2011; 11: 38.


Small AR, Neagu A, Amyot F and Sackett D, Chernomordik V, Gandjbakhche A. Spatial distribution of VEGF isoforms and


Thompson HJ, Zhu Z and Jiang W. Weight control and breast cancer prevention: are the effects of reduced energy intake equivalent to those of increased energy expenditure?. *J. Nutr.*, 2004; 134: 3407-3411.


Verna L, Whysner J and Williams GM. N-nitrosodiethylamine mechanistic data and risk assessment: bioactivation, DNA-


