

## Supplementation of Annatto (*Bixa orellana*)-Derived $\delta$ -Tocotrienol Produced High Number of Morula through Increased Expression of 3-Phosphoinositide-Dependent Protein Kinase-1 (PDK1) in Mice

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**Abstract :** Several embryonic cellular mechanism including cell cycle, growth and apoptosis are regulated by phosphatidylinositol-3-kinase (PI3K)/Akt signaling pathway. The goal of present study is to determine the effects of annatto (*Bixa orellana*)-derived  $\delta$ -tocotrienol ( $\delta$ -TCT) on the regulations of PI3K/Akt genes in murine morula. Twenty four 6-8 week old (23-25g) female balb/c mice were randomly divided into four groups (G1-G4; n=6). Those groups were subjected to the following treatments for 7 consecutive days: G1 (control) received tocopherol stripped corn oil, G2 was given 60 mg/kg/day of  $\delta$ -TCT mixture (contains 90% delta & 10% gamma isomers), G3 was given 60 mg/kg/day of pure  $\delta$ -TCT (>98% purity) and G4 received 60 mg/kg/day  $\alpha$ -TOC. On Day 8, females were superovulated with 5 IU Pregnant Mare's Serum Gonadotropin (PMSG) for 48 hours followed with 5 IU human Chorionic Gonadotropin (hCG) before mated with males at the ratio of 1:1. Females were sacrificed by cervical dislocation for embryo collection 48 hours post-coitum. About fifty morula from each group were used in the gene expression analyses using Affymetrix QuantiGene Plex 2.0 Assay. Present data showed a significant increase ( $p < 0.05$ ) in the average number (mean + SEM) of morula produced in G2 (26.0 + 0.45), G3 (23.0 + 0.63) and G4 (25.0 + 0.73) compared to control group (G1 - 16.0 + 0.63). This is parallel with the high expression of PDK1 gene with increase of 2.75-fold (G2), 3.07-fold (G3) and 3.59-fold (G4) compared to G1 (1.78-fold). From the present data, it can be concluded that supplementation with  $\delta$ -TCT(s) and  $\alpha$ -TOC induced high expression of PDK1 in G2-G4 which enhanced the PI3K/Akt signaling activity, resulting in the increased number of morula.

**Keywords :** delta-tocotrienol, embryonic development, nicotine, vitamin E

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