## Tocotrienol Rich Fraction in Nicotine-Induced Embryos: Cytoskeletal Changes of Actin and Tubulin

Authors : Nurul Hamirah Kamsani, Mohd Hamim Rajikin, Nor Ashikin Mohamed Noor Khan, Sharaniza Abdul Rahim Abstract : Cytoskeletal structures, in particular actin and tubulin, provide a fundamental framework in all cells, including embryos. Under influence of nicotine, the cytoskeletal organization may be subjected to oxidative stress (OS) insult and cause alteration. Tocotrienol-rich fraction (TRF) is proven to enhance fertility better than the other sub-group of Vitamin E, tocopherols (TCPs). The objective of this study was to evaluate the effects of TRF on 1) actin and tubulin of 2- and 8-cell murine embryos and 2) the regulation of reactive oxygen species (ROS)-scavenging enzymes; induced by nicotine. Twenty four female Balb/C were subjected to either subcutaneous (sc) injection of 0.9% NaCl; sc injection of 3.0 mg/kg bw/day nicotine; sc injection of 3.0 mg/kg bw/day nicotine + oral gavage (OG) of 60 mg/kg bw/day TRF; or OG of 60 mg/kg bw/day TRF for 7 consecutive days. After superovulation and mating, animals were euthanized. 2-cell developing embryos were retrieved. 50% of the retrieved embryos were visualized under confocal laser staining microscopy (CLSM) for alterations of actin and tubulin. The remaining amount of embryos was cultured in vitro until 8-cell stage followed by CLSM visualization. Blood plasma was subjected to OS assays. Plasma malondialdehyde (MDA), superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx) were determined and analysed accordingly. At both 2- and 8-cell developing stages, actin intensities were significantly reduced in the nicotine group (p<0.001). After the intervention, actin intensity was significantly increased compared to that of the nicotine group (p<0.001). The same trend was seen in tubulin at both cell stages. TRF has minimized the deleterious effects of nicotine in actin and tubulin of both 2- and 8-cell developmental stages during pre-implantation embryonic development in mice in vitro. Levels of endogenous anti-oxidative enzymes were sustained close to control accompanied by decreased levels of OS biomarker.

Keywords : actin, nicotine, pre-implantation embryos, tocotrienol rich fraction, tubulin

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