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Roles of *ATP7B* gene to maintain the copper-transporting ATPase in a HepG2 cell line against excess copper toxicity

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Wilson disease (WD) is an autosomal recessive disorder of copper transport with a worldwide frequency of ~1 in 30000. Wilson's disease is characterized by chronic liver and neurological disease and also reported in kidney. Hepatic copper levels vary among normal individuals and WD patients depending upon on dietary copper intake and bioavailability, as well as genetic factors. In this study we examined that abnormal copper accumulation in human hepatocarcinoma (HepG2) cell line. Copper chloride ($CuCl_2$) caused dose dependent cell viability reduction of human hepatocarcinoma (HepG2) cell line which was measured through MTT assay. We used different concentration of $CuCl_2$ in their log doses but maximum cell viability reduction was recorded at 15 µg/ml. It also induces cell cycle arrest and DNA damage due to intracellular ROS generation. $CuCl_2$ induces Ca^{2+} release from endoplasmic reticulum (ER) and leads to apoptotic cell death. It causes the up-regulation of WD stress marker genes *ATP7B* and Cyp1A1, Cyp1A2 at transcription levels. The similar response of *ATP7B* and Cyp1A1, Cyp1A2 proteins was recorded at translation levels. Heavy dietary intake of $CuCl_2$ induces mitochondria and reduced the mitochondrial membrane potential analyzed through JC-1 staining. It further increases Bax/Bcl2 ratio and promotes the release of cytochrome C, finally leads to caspase-dependent apoptosis. Up-regulation of APAF1 in $CuCl_2$ treated cells supports the mitochondrial-mediated apoptotic cell death. The results support the involvement of ER and mitochondria in ROS mediated $CuCl_2$ toxicity. Therefore, the heavy dietary intake of $CuCl_2$ in food products may be deleterious to users.

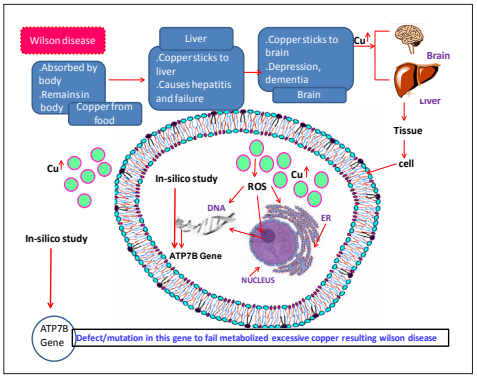

Figure-1: *Graphical abstract of copper toxicity*

References

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Biography

Shikha Agnihotry has been working as Research Assistant at Sanjay Gandhi Postgraduate Institute of Medical Sciences, Lucknow of ICMR since 2013. She has been involved in training biomedical research scholars in the field of bioinformatics and has also assisted as Research Assistant under an ICMR-funded project.

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