

Summary

8.0. SUMMARY

Experiment-I focused on the effective dose standardization of arjunolic acid and B₁₂ against sodium arsenite mediated ill effects on female reproductive treatment. The results of experiment-I showed that arjunolic acid at the dose of 1.0 mg/100gm body weight is more effective than the other doses (0.5 mg/ 100gm body weight and 1.5mg/100gm body weight) and vitamin B₁₂ at the dose of 0.09µg/100gm body weight was more effective than other doses (0.07 µg/ 100gm body weight and 0.1 µg/100gm body weight) against arsenic at the dose of 1.0 mg/100gm body weight. The above-mentioned dose is the critical dose of arjunolic acid and B₁₂ that could effectively ameliorate sodium arsenite induced alterations of uterine oxidative stress and ovarian steroidogenesis by the restoration of lipid peroxidation, activities of enzymatic antioxidants, and steroidogenic enzymes.

From the above results of experiment-II, pretreatment of arjunolic acid and B₁₂ followed by arsenication showed the beneficial preventive effects in the restoration of body growth, organs weight, antioxidant status of ovary and uterus against arsenic-induced female reproductive hazards in Wistar rats. Arjunolic acid and B₁₂ could also improve the histoarchitecture of the ovary and uterus. The deterioration of tissue through possible apoptosis and necrosis was prevented probably by the elimination of arsenic from organs by reducing the levels of lipid peroxide end products and serum LDH and via the alteration of methylation process arsenic may be removed from the ovary and uterus where circulating levels of B₁₂, folate, and homocysteine execute an important role.

The results of experiment-III explored that co-treatment with arjunolic acid and or B₁₂ could able to give protection against the uterine and ovarian expression of enzymatic antioxidants and ovarian steroidogenesis as well as the protection of the genetic constituent of the uterus and

cellular synthetic mechanism. Necrotic tissue deterioration may be protected by arjunolic acid and or B₁₂ in this experiment as indicated from lowering of LDH level in arsenicated rats. It may be feasible due to the probable removal of arsenic from the ovarian and uterine tissue as indicated by the protection of circulating levels of B₁₂, folate, and homocysteine. This type of protective action is helpful in the rejuvenation of tissue structural materials and amino acid pool. In the way of the protection of arsenic-induced ovarian-uterine toxicity and carcinogenicity arjunolic acid and B₁₂ as active exogenous ingredients have imperative role alone or jointly. However, arjunolic acid perhaps trap the As^{III} due to its chelating property and it may be possible that arjunolic acid increases the bioavailability of vitamin B₁₂ and folate that enhance arsenic detoxification in the way of diminishing arsenic-induced oxidative stress in ovarian and uterine tissue.

Experiment-IV focused that arjunolic acid and B₁₂ have curative effects in the renovation of body growth, organ weight, antioxidant status, and histoarchitecture of ovary and uterus. Moreover, this experiment highlighted the status of pro-inflammatory cytokines such as uterine NF-κB, serum TNF-α, and IL-6. We observed that these inflammatory markers were well managed by the post-treatment B₁₂ and arjunolic acid in arsenicated rats. A possible elimination of arsenic from the organs as indicated by the improvement of the components of methionine pool such as B₁₂, folate, and homocysteine; that perhaps limit the inflammatory response and probable apoptotic and necrotic tissue deterioration.

In vitro assay in Experiment-V explored that arjunolic acid and or vitamin B₁₂ could reduce the toxicity with a higher concentration of arsenic and H₂O₂ from liver and uterus at short duration (3 and 6 hrs). The hepatic MDA and CD levels, uterine and hepatic SOD and catalase activity, ovarian steroidogenic enzyme level, and liver DNA damages were restored through the addition

of arjunolic acid and or vitamin B₁₂ in sodium arsenite containing assay media. Arjunolic acid and vitamin B₁₂ have better effective direct action in restoring ovarian and uterine tissue enzymatic antioxidants and steroidogenesis when incubated the organ slices for 6 hrs rather than 3 hrs in assay media containing arsenic and or H₂O₂.

However, this noninvasive treatment strategy with arjunolic acid and B₁₂ against arsenic highlighted an indirect action during the restoration of female reproductive function by the involvement of hypothalamico-pituitary-ovarian axis as indicated from the results of *in vivo* Experiment-I-IV; although *in vitro* study (Experiment-V) indicated the possible direct action of arjunolic acid and B₁₂ too against arsenic.