SUMMARY AND CONCLUSION
SUMMARY

The study can be summarized as follows:

1. MCT model of PH was standardized using subcutaneous administration of single dose of 60 mg/kg of MCT which led to a significant increase in RVP and RVH, two hallmarks of PH, after 35 days of its administration.

2. The model of PH was validated using clinically used endothelin receptor antagonist, Bosentan (100 mg/kg) which reduced RVP, RVH and pulmonary vascular remodeling.

3. The standardized dose of PARP-1 inhibitor 1,5- Isoquinolinediol (3 mg/kg) was effective in lowering RVP and checking the RVH in MCT treated rats. The standardized dose of 3 mg/kg dose of ISO was also effective in reversing established PH when administered from 21 to 35 days.

4. PARP-1 inhibition by ISO, reduced oxidative stress markers i.e. MDA, ROS and nitrite and increased in endogenous antioxidant GSH in both lungs and right ventricle in MCT challenged rats.

5. PARP-1 inhibition decreased the expression of gH2AX (DNA damage marker), and expression and activity of PARP-1 in lungs and RV of MCT treated rats.

6. PARP-1 inhibition ameliorated the pulmonary vascular remodeling and increased expression of PCNA (a marker of cell proliferation) in MCT treated rats.
7. PARP-1 inhibition attenuated apoptosis resistance in lungs of MCT treated rats as evident by increase in the number of TUNEL positive cells and caspase 3 activity.

8. Inhibition of PARP-1 favorably modulated the expression of a number of proteins like HIF-1α, VEGF, MMP-2, MMP-9, TIMP, GSK3β, which participate in the pathophysiology of PH.

9. PH is characterized by pulmonary endothelial dysfunction, decreased vasoreactivity and eNOS expression which was restored by PARP-1 inhibitor, ISO.

10. In hypertrophied RV, PARP-1 inhibition, improved mitochondrial function as shown by increased NAD and ATP levels along with preserving the mitochondrial membrane integrity. PARP-1 inhibition with ISO also prevented the release of cell death factors like AIF and cytochrome c, and further protected RV from apoptotic cell death.

11. The effect of *Withania somnifera* (WS) in MCT induced PH was also studied. WS treatment attenuated right ventricular pressure and hypertrophy in preventive as well as therapeutic treatment. WS also decreased oxidative stress, inflammation, improved endothelial dysfunction, reversed the pulmonary vascular remodeling and weakened the apoptosis resistance in the lungs of MCT exposed rats. The WS treatment also reduced the DNA damage, by decreasing expression of gH2Ax, and expression of PARP-1. Thus, this study reconfirmed the status of WS as a vaso- and cardioprotective herbal in Indian medicinal system and showed that protective effect of WS is also mediated by inhibition of PARP-1.
Schematic representation of the involvement of PARP-1 in the pathophysiology of pulmonary hypertension
CONCLUSION

This is the first report where PARP-1 involvement in the endothelial dysfunction and right ventricle hypertrophy in pulmonary hypertension has been found. The pharmacological modulation using PARP-1 inhibitor (ISO) attenuated RVP and RVH in preventive as well as curative treatment in PH. Also, ISO reversed the complications associated with PH like pulmonary vascular remodelling and pulmonary vascular dysfunction. Therefore, the results of this study indicate that PARP-1 can be a potential therapeutic target in PH. Further, Withania somnifera, a well known herbal drug in Ayurveda was protective in MCT model of pulmonary hypertension and its protective effect was also mediated by inhibiting PARP-1.

FUTURE PROSPECTS

This study has provided adequate data to underline the PARP-1 as a potential target in PH, however, further studies in transgenic models of PH are needed to confirm the involvement of PARP-1 in PH pathophysiology. In the present work, all the studies remained confined only in in vivo model of PH, however, in vitro studies should be carried out to dissect the detailed molecular mechanism of PARP-1 involvement in PH. Detailed studies are also needed for exploring the possible role of other analogues of PARP (eg. PARP-2, etc.) in PH. Lastly, several PARP inhibitors, used clinically for cancer treatment, may be explored (experimentally as well as clinically) for their effect in PH because PH shares its pathophysiology with cancer also.