ABSTRACT

Objective: The objective of this study is to assess the hepatoprotective nature of Siddha formulation manjal noi kudineer (MNK) against Paracetamol induced hepatotoxicity in Zebrafish Danio rerio model.

Methods: Pharmacological studies were carried out in the adult zebra fish model in the category of four groups; each individual group contained ten animals. Groups 1, 2, 3, and 4 were considered as control groups, disease control groups, and drug treated groups at different concentrations, respectively. Histopathological analysis was recorded.

Results: The observed research findings were confirmed that this novel Siddha formulation MNK possess potent hepatoprotective effect at the both dose level of 250 mg/l–500 mg/l against paracetamol induced toxicity in Zebrafish.

Conclusion: Siddha medicinal formulation has been in practice for more than 1000 years. Liver diseases are the highly prevalent one among all the diseases. Although huge drugs are available for liver diseases, there is a need for cost effectiveness drugs without any adverse effect. From the study, it was concluded that the drug MNK has promising hepatoprotective activity in dose-dependent manners and restores the basic liver architecture by means of its rejuvenating potential against paracetamol induced toxicity in Zebrafish model. We can strongly suggest that the Sastric Siddha drug MNK to treat various kinds of liver diseases such as liver cirrhosis and hepatic carcinoma. This research work may be highly beneficial to the people who suffered from various liver diseases if the drug will be given clinically.

Keywords: Siddha medicine, Poly herbal, Traditional, Sastric, Hepatoprotective, Kudineer.
The weight/ml calculation of the drug MNK=0.050 gm (5000 mg or 50 mg)/ml.

Grouping
- Group I: Control
- Group II: Paracetamol 5 mM (755.8 mg) per liter concentration
- Group III: Paracetamol 5 mM+MNK Low Dose 250 mg/l
- Group IV: Paracetamol 5 mM+MNK High Dose 500 mg/l.

Treatment
Animal belongs to Group I left untreated and Group II treated with Paracetamol at the concentration of 5 mM (755.8 mg) per liter concentration for the period of 7 days. Animal belongs to Group III received test drug MNK at the concentration of 250 mg /l and Group IV received test drug MNK at the concentration of 500 mg/l along with paracetamol 5 mM for the period of 7 days.

Histopathology
After a 1-week exposure period, fishe were sacrificed and the livers of Zebrafish were dissected and fixed in 10% formalin at 4°C for 24 h. Subsequently, the fixed liver tissues were dehydrated in gradient ethanol, hyalinized in xylene, and embedded in paraffin wax at 56°C. At that point, the paraffin blocks were separated at 4-μm thickness. The segments were gathered on glass slides and recolored with hematoxylin and eosin (H and E) utilizing a H and E Staining Kit. Histologic lesions were watched utilizing an optical microscope instrument outfitted with a computerized camera.

RESULTS AND DISCUSSION
The histological microscopy findings belong to the normal control Group I show the presence of regularly arranged hepatocytes exist in polygonal shape cells and with prominent nucleus further sinusoids appears with regular intervals. In Group II, severe discrete cytoplasmic vacuoles and rare foamy cytoplasm were observed. This prominence indicates the presence of inflammatory changes in it. The histopathological findings of Group III retain the basic structure of polygonal shaped hepatic parenchyma with occasional Pyknotic nuclei and of Group IV shows that regenerated liver cells with Occasional bi nucleated hepatocytes were observed. This is due to the hepatoprotective nature of the drug MNK treated. The reasons behind the hepatoprotective effect of this formulation are possibly due to the presence of herbals in it.

The pharmacological research findings confirms that the aqueous extract of *Phyllanthus niruri* exhibits hepatoprotective effects against carbon tetrachloride induced liver toxicity in mice. The herbals *T. terrestris* and *Bauhinia acuminata* present in this formulation were already documented for its immunomodulatory effects. In addition to that the other herbal...
The results of the present investigation indicate that paracetamol treated groups show severe liver degeneration and inflammation associated fibrosis whereas treatment with test drug MNK at both the dose levels fundamentally constricted the paracetamol prompted harm in Groups III and IV. Hence, from the study, it was concluded that the drug MNK has promising hepatoprotective activity in dose-dependent manners and restores the basic liver architecture by means of its rejuvenating potential against paracetamol induced toxicity in Zebrafish model. We can strongly suggest that the Sastric Siddha drug MNK to treat various kinds of liver diseases such as liver cirrhosis and hepatic carcinoma; however, further study is required to identify the hepatoprotective constituents present in the formulation as well as elucidating the mechanism of action.

CONFLICTS OF INTEREST STATEMENT
No conflicts of interest.

FINANCIAL SUPPORT AND SPONSORSHIP
Nil.

ACKNOWLEDGMENT
Our sincere thanks to Palpandian, Dr. Thamodaran, Dr. Balasubramanian, Dr. Natarajan, Rathinambal for their constant support.
AUTHOR CONTRIBUTION

This study was designed, directed, and coordinated by principal investigator provided conceptual and technical guidance for all aspects of the project by Dr. R. Gomathi, Dr. E. Preetheekha, Dr. P. Shanmuga Priya, and Dr. A. Mamallan Suggested and commented on the design of the experiments.

REFERENCES