

ACUTE AND SUBACUTE ORAL TOXICITY STUDIES OF MANJAL NOI KUDINEER: A PRECLINICAL INVESTIGATION SUPPORTING SAFE SIDDHA PRACTICE

^{1*}Senthil Kumar P, ²Elansekaran S and ³Bharath Kumar G

¹Velumailu Siddha Medical College and Hospital, Sriperumbudur, Kancheepuram, Tamil Nadu, India.

²Department of Noi Naadal, National Institute of Siddha, Tambaram, Chennai, Tamil Nadu, India.

³The Tamil Nadu Dr MGR medical University, Guindy, Chennai, Tamil Nadu, India.

Article History: Received 19th September 2025; Accepted 11th November 2025; Published 1st December 2025

ABSTRACT

Manjal Noi Kudineer is a classical Siddha polyherbal formulation traditionally used for liver ailments, inflammatory conditions, and detoxification. Despite its long history of therapeutic use, scientific evidence on its safety profile remains limited. This study aimed to evaluate the acute and repeated 28-day oral toxicity of MNK in Wistar rats following OECD guidelines to establish preliminary safety data. Acute toxicity was assessed using OECD 423 at a single dose of 2000 mg/kg, while subacute toxicity followed OECD 407 with daily oral administration of three dose levels for 28 days. Animals were monitored for clinical signs, body weight, hematological and biochemical markers, organ weights, and histopathology. MNK produced no mortality, no clinical toxicity, and normal body weight gain across all groups. Hematological parameters, liver and kidney function markers (AST, ALT, ALP, bilirubin, urea, creatinine), lipid profile, and electrolytes remained within physiological limits. Organ weights showed no dose-dependent deviations, and histopathology of major organs confirmed absence of structural damage. MNK demonstrated a favorable safety profile, supporting its traditional use and warranting further pharmacological and clinical investigation.

Keywords: Manjal Noi Kudineer, Acute toxicity, Subacute toxicity, Siddha medicine, Herbal safety evaluation.

INTRODUCTION

The Siddha system of medicine is among the earliest traditional healthcare frameworks in India, characterized by its holistic therapeutic principles that integrate diet, lifestyle, and the use of herbal, mineral, and animal-derived preparations. Within this system, polyherbal decoctions known as Kudineer formulations hold a central role in managing fever, inflammation, metabolic imbalances, hepatic dysfunction, and toxin accumulation (Government of India, 2008). Among these formulations, Manjal Noi Kudineer (MNK) has been traditionally prescribed for conditions related to impair liver function, chronic inflammatory states, and systemic detoxification. The formulation comprises six botanicals such as *Phyllanthus niruri*, *Eclipta prostrata*, *Trichosanthes cucumerina*, *Piper longum*, *Foeniculum vulgare*, and *Aegle marmelos*, each of which is widely documented for its diverse pharmacological actions, including antioxidant,

hepatoprotective, antimicrobial, and immunomodulatory properties (Loha *et al.*, 2019). Collectively, these attributes highlight MNK's therapeutic relevance within classical Siddha practice.

Despite its longstanding traditional use, scientific validation of MNK remains crucial in the context of growing global interest in herbal therapeutics. The increasing dependence on phytomedicine has led international bodies, including the World Health Organization, to emphasize the importance of establishing safety, efficacy, and quality profiles for herbal formulations (WHO, 2011). Long-term or unregulated consumption of botanicals can pose potential risks, particularly in polyherbal preparations where multiple phytoconstituents may interact synergistically or antagonistically, thereby altering their pharmacodynamic or toxicological profiles. Although MNK continues to be widely utilized in Siddha clinical settings, there remains a scarcity of systematic

*Corresponding Author: Senthil Kumar P, Professor, Velumailu Siddha Medical College and Hospital, Sriperumbudur, Kancheepuram, Tamil Nadu, India Email: senthilsamrat.sk@gmail.com

toxicological studies evaluating its safety, especially in its standardized decoction form.

Preclinical toxicity assessments play a vital role in determining the biological tolerability of herbal medicines before clinical application. Rodent models, particularly Wistar rats, are considered reliable for initial safety evaluations due to their physiological stability and reproducible responses. OECD Guideline 423 for acute oral toxicity provides a structured approach for identifying potential adverse effects following a single high-dose exposure, while OECD Guideline 407 outlines a standardized protocol for assessing repeated-dose toxicity over 28 days (OECD, 2001; OECD, 2008). These guidelines offer internationally recognized methodologies to evaluate systemic toxicity, organ-specific damage, hematological alterations, biochemical disturbances, and histopathological changes.

Given the therapeutic importance of MNK, its widespread use, and the lack of comprehensive safety data, a structured toxicological study becomes essential to ensure its suitability for continued clinical use. The present investigation was therefore designed to evaluate both acute and subacute (28-day) oral toxicity of MNK in Wistar rats using OECD-recommended protocols. The study assessed a broad range of parameters, including clinical signs, behavioral responses, mortality, body weight progression, feed and water intake, hematological indices, serum biochemical markers of hepatic and renal function, lipid profiles, relative organ weights, and microscopic architecture of major organs. By generating robust toxicological evidence through standardized procedures, this study aims to support the safe therapeutic application of MNK and contribute to the scientific foundation necessary for integrating traditional Siddha formulations into evidence-based modern healthcare.

MATERIALS AND METHODS

Preparation and authentication of herbal ingredients

All raw herbal components used for the preparation of Manjal Noi Kudineer (MNK) were procured from a certified GMP-compliant herbal supplier. Each botanical ingredient such as *Phyllanthus niruri*, *Eclipta prostrata*, *Trichosanthes cucumerina*, *Piper longum*, *Foeniculum vulgare*, and *Aegle marmelos* were authenticated at the Department of Maruthuva Thavaraiyal, National Institute of Siddha, Chennai. Authentication followed macroscopic, microscopic, and organoleptic standards described in the Siddha Pharmacopoeia of India (Government of India, 2008) and classical pharmacognostic guidelines (Trease & Evans, 2009). The plant materials were cleaned, shade-dried at ambient temperature to prevent phytochemical degradation (Sasidharan *et al.*, 2011), and coarsely powdered. The MNK decoction was prepared following traditional Siddha procedures by boiling 100 g of the composite mixture in 800 mL of water and reducing it to one-fourth volume, as standardized in Indian traditional medicine protocols (Mukherjee, 2019; Loha *et al.*, 2019).

Experimental animals and husbandry

Healthy Wistar albino rats (*Rattus norvegicus*), weighing 100–120 g, were obtained from a CPCSEA-registered breeding facility. Animals were housed in polypropylene cages containing autoclaved paddy-husk bedding, maintained at controlled temperature (25 ± 3 °C), humidity (45–55%), and a 12-h light/dark cycle, consistent with internationally accepted animal care guidelines (National Research Council, 2011). Standard pellet diet and filtered water were provided *ad libitum*. A 7-day acclimatization period was ensured, as recommended for stabilizing physiological parameters prior to toxicological studies (OECD, 2008). Ethical approval was granted by the Institutional Animal Ethics Committee (IAEC), and all procedures followed CPCSEA regulatory standards for laboratory animal welfare.

Dose selection

Dose selection for MNK was based on the human therapeutic dose, converted to rat-equivalent values using the body surface area normalization method proposed by Paget and Barnes (1964) and further validated in interspecies dose extrapolation studies (Reagan-Shaw *et al.*, 2008). Low, intermediate, and high doses were selected to represent no-observed-effect, moderate physiological effect, and potential threshold effect levels, respectively.

Acute oral toxicity study (OECD 423)

Acute toxicity was assessed using the OECD Guideline 423 for acute oral toxicity (OECD, 2001), which employs a stepwise dose-level approach for toxic class determination. A limit dose of 2000 mg/kg body weight was administered orally to overnight-fasted female rats using a stainless-steel oral gavage. Animals were monitored continuously during the first 4 hours for signs of toxicity including behavioral changes, tremors, piloerection, diarrhea, and convulsions and subsequently observed daily for 14 days for morbidity and mortality, following recommendations by Parasuraman (2011).

Subacute 28-day repeated dose toxicity study (OECD 407)

The 28-day repeated oral toxicity study followed OECD Guideline 407 (OECD, 2008). Forty rats were randomly distributed into four groups ($n = 10$; five males and five females). Group I served as the vehicle control, while Groups II–IV received low, intermediate, and high doses of MNK, respectively. MNK was administered orally once daily for 28 consecutive days. Clinical observations, body weight, food intake, and water consumption were recorded weekly, following standard subacute toxicity assessment protocols (Hall *et al.*, 2012).

Hematological, biochemical, and histopathological evaluation

Terminal blood samples were collected via retro-orbital puncture under light anesthesia. Hematological parameters

such as Hb, RBC, WBC, platelet count, and differential leukocyte count were analyzed using an automated hematology analyzer, following standard methodologies (Yogananth *et al.*, 2015). Serum biochemical markers, including liver enzymes (AST, ALT, ALP), renal indices (creatinine, urea), electrolytes, glucose, lipid profile, and total protein, were measured using validated diagnostic kits (Revathi Chitra *et al.*, 2024). Major organs were excised, rinsed, weighed, and preserved in 10% buffered formalin. Histopathological analysis was performed on paraffin-embedded tissue sections stained with hematoxylin and eosin, following established protocols (Chitra *et al.*, 2020).

Statistical Analysis

Data were expressed as mean ± SD. Statistical comparisons among groups were performed using one-way ANOVA, with p < 0.05 considered statistically significant.

RESULTS AND DISCUSSION

Oral administration of MNK at a limit dose of 2000 mg/kg in female rats did not induce mortality or any treatment-related signs of toxicity throughout the 14-day observation period. Behavioral parameters including alertness, motor activity, autonomic reflexes, and gait remained comparable to the control group. The absence of clinical abnormalities indicates that MNK is well tolerated at high doses, with the LD₅₀ estimated to be greater than 2000 mg/kg. All animals

survived without any behavioral deviations, confirming the non-toxic nature of MNK at higher doses. 28-Day Repeated Oral Toxicity Study Daily clinical evaluation revealed no morbidity, mortality, or treatment-related behavioral changes in any of the MNK-treated groups during the 28-day period. All animals remained active with normal grooming, feeding, and reflex responses. Body Weight, Food and Water Intake Body weight profiles showed progressive, normal growth in all experimental groups.

Although minor variations were noted, no statistically significant reduction was observed in comparison with controls, indicating absence of MNK-induced metabolic or systemic stress. Food and water intake patterns remained stable across all groups, with no significant deviations from the control. This suggests MNK does not interfere with appetite, hydration, or gastrointestinal function (Table 1b). Hematological evaluation after 28 days of MNK administration demonstrated that all parameters remained within normal physiological limits, with no dose-related variations. The maximum RBC count was recorded in the 250 mg/kg group (8.32±0.18 mm³), while the minimum occurred in the 50 mg/kg group (7.98±0.28 mm³). Hemoglobin levels showed minimal variation, ranging from 14.92±0.25% (lowest, 50 mg/kg) to 15.21±0.49% (highest, 250 mg/kg). Leukocyte count remained stable across groups, with values ranging from 10274±117.64×10⁶/mL to 10389±118.48×10⁶/mL.

Table 1a. Behavioral Observations During Acute Toxicity Testing of MNK.

Dose (mg/kg)	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
2000	+	-	-	+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	+	-

1. Alertness 2. Aggressiveness 3. Piloerection 4. Grooming 5. Gripping 6. Touch response 7. Decreased motor activity 8. Tremors 9. Convulsions 10. Muscle spasm 11. Catatonia 12. Muscle relaxation 13. Hypnosis 14. Analgesia 15. Lacrimation 16. Exophthalmos 17. Diarrhoea 18. Writhing 19. Respiration 20. Mortality

Platelets exhibited slight differences, where the lowest count was seen in the 50 mg/kg group (1254±22.38/μL) and the highest in the 250 mg/kg group (1317±14.58/μL). Differential leukocyte counts and erythrocytic indices (MCV, MCH, MCHC, PCV, ESR) displayed no abnormalities. Overall, the data confirm that MNK does not exert hematotoxic effects at any tested dose (Table 2). The hepatic biochemical assessment showed no significant alterations across MNK-treated groups compared with the control. Total bilirubin values ranged from a minimum of 0.250 mg/dL (500 mg/kg) to a maximum of 0.274 mg/dL (control), indicating stable liver function. SGOT levels showed a slight decline from 156.19 U/L (control) to 150.65 U/L (500 mg/kg), while SGPT remained within normal physiological limits (40.68–43.9 U/L). Total protein, albumin, globulin, and GGT values also exhibited normal fluctuations. Overall, the findings confirm that MNK does not exert hepatotoxic effects at the tested doses

(Table 3). Renal function parameters in MNK-treated rats showed no significant deviations compared with control animals, indicating preserved kidney function. Urea levels ranged from 63.48 mg/dL (50 mg/kg) to 65.48 mg/dL (control), while creatinine remained stable between 0.88–0.99 mg/dL. Uric acid varied minimally, with a maximum of 1.8 mg/dL (control) and a minimum of 1.64 mg/dL (250 mg/kg). Electrolytes, including sodium, potassium, and chloride, also remained within normal physiological limits across all dose groups. These results confirm that MNK administration up to 500 mg/kg does not induce nephrotoxicity in Wistar rats (Table 4). The lipid profile of MNK-treated rats remained within physiological limits, suggesting no adverse effects on lipid metabolism. Total cholesterol ranged from 41.56 mg/dL (50 mg/kg) to 43.55 mg/dL (500 mg/kg), while HDL varied slightly between 12.58–13.68 mg/dL. LDL levels were consistent across groups, with a maximum of 36.58 mg/dL (250 mg/kg) and

a minimum of 35.81 mg/dL (50 mg/kg). VLDL and triglycerides also showed minimal fluctuations. The TC/HDL ratio and blood glucose remained stable. These results indicate that repeated administration of MNK up to 500 mg/kg does not alter lipid metabolism or glycemic status in Wistar rats (Table 5). The assessment of organ weights in MNK-treated rats indicated no significant alterations compared to the control group, suggesting the absence of organ-specific toxicity. The liver ranged from

4.84 g (250 mg/kg) to 5.26 g (500 mg/kg), and the heart varied between 0.73–0.78 g. Lung weights ranged from 2.01–2.12 g, while spleen showed a slight decrease in treated groups (0.67–0.72 g) compared to control (0.81 g). Other organs, including kidneys, brain, stomach, ovaries, and testes, demonstrated stable weights across all doses. These findings indicate MNK is non-toxic at doses up to 500 mg/kg (Table 6).

Table 1b. Body Weight, Food and Water intake in rats treated with MNK.

Dose (mg/kg)	Body Weight (g)	Food Intake (g/day)	Water Intake (ml/day)
Control	128.75 ± 0.99	35.48 ± 1.48	42.60 ± 2.84
50	131.62 ± 5.02*	35.12 ± 2.34	41.05 ± 2.49
250	133.27 ± 8.95*	36.46 ± 1.84	41.58 ± 3.08
500	130.48 ± 7.33*	35.28 ± 1.25	41.98 ± 2.49

Table 2. Effect of MNK on Hematological Parameters.

Parameter	Control	50 mg/kg	250 mg/kg	500 mg/kg
RBC (mm ³)	8.12±0.22	7.98±0.28	8.32±0.18	8.24±0.16
Hb (%)	15.01±0.42	14.92±0.25	15.21±0.49	15.12±0.58
Leukocytes (×10 ⁶ /mL)	10389±118.48	10274±117.64	10297±118.68	10342±114.58
Platelets/μL	1301±13.48	1254±22.38	1317±14.58	1306±14.59
MCV (fl)	52.48±2.37	53.67±1.49	54.59±1.43	53.46±1.81
Neutrophils (%)	4.36±1.28	4.27±1.48	4.87±1.49	4.52±1.34
Lymphocytes (%)	85.18±4.49	84.42±3.17	85.12±3.67	86.48±2.66
Monocytes (%)	2.02±0.98	2.18±1.24	2.05±0.87	2.27±0.72
Eosinophils (%)	1.42±0.48	1.39±0.68	1.46±0.43	1.43±0.18
Basophils (%)	0	0	0	0
ESR (mm)	1±0.00	1±0.00	1±0.00	1±0.00
PCV	46.85±2.48	47.26±2.87	47.35±2.67	47.10±2.36
MCH (pg)	17.15±1.37	17.38±0.94	17.35±0.68	17.68±0.58
MCHC (g/dL)	32.67±1.68	32.66±0.59	33.18±2.59	31.67±1.28

Table 3. Effect of MNK on Hepatic Parameters.

Parameter	Control	50 mg/kg	250 mg/kg	500 mg/kg
Total Bilirubin (mg/dL)	0.274 ± 0.87	0.265 ± 0.34	0.252 ± 0.48	0.250 ± 1.08
Direct Bilirubin (mg/dL)	0.13 ± 0.15	0.12 ± 0.13	0.10 ± 0.17	0.11 ± 0.18
Indirect Bilirubin (mg/dL)	0.10 ± 0.12	0.15 ± 0.16	0.13 ± 0.08	0.12 ± 0.09
SGOT (U/L)	156.19 ± 2.94	154.84 ± 2.64	152.68 ± 2.73	150.65 ± 2.48
SGPT (U/L)	40.68 ± 2.28	43.9 ± 2.48	43.49 ± 2.09	42.95 ± 2.97
Total Protein (g/dL)	11.98 ± 1.17	11.64 ± 0.49	10.81 ± 0.37	11.68 ± 0.67
Albumin (g/dL)	3.91 ± 0.37	3.57 ± 0.27	3.52 ± 0.28	3.64 ± 0.18

Globulin (g/dL)	4.98 ± 0.47	5.02 ± 0.34	4.68 ± 0.58	5.59 ± 0.58
A/G Ratio	0.49 ± 0.12	0.52 ± 0.68	0.47 ± 0.48	0.51 ± 0.38
GGT (U/L)	7.3 ± 0.4	7.6 ± 0.2	7.32 ± 0.1	7.41 ± 0.43

Table 4. Effect of MNK on Renal Parameters.

Parameter	Control	50 mg/kg	250 mg/kg	500 mg/kg
Urea (mg/dL)	65.48 ± 2.82	63.48 ± 3.58	63.59 ± 2.45	64.82 ± 1.55
Creatinine (mg/dL)	0.91 ± 0.58	0.88 ± 0.59	0.99 ± 0.58	0.98 ± 0.69
Uric Acid (mg/dL)	1.80 ± 0.27	1.70 ± 0.56	1.64 ± 0.37	1.72 ± 0.55
Na (m.mol)	138.62 ± 2.97	138.40 ± 3.27	136.65 ± 2.72	137.64 ± 2.18
K (m.mol)	19.80 ± 2.48	20.06 ± 2.47	21.23 ± 2.34	20.67 ± 2.24
Cl (m.mol)	99.65 ± 1.87	98.54 ± 3.48	98.24 ± 3.69	98.97 ± 3.65

Table 5. Effect of MNK on Lipid Profile.

Parameter	Control	50 mg/kg	250 mg/kg	500 mg/kg
Total Cholesterol (mg/dL)	42.67 ± 1.57	41.56 ± 1.38	42.65 ± 1.87	43.55 ± 2.57
HDL (mg/dL)	13.68 ± 1.18	12.58 ± 1.63	12.82 ± 1.24	13.28 ± 1.18
LDL (mg/dL)	36.56 ± 2.55	35.81 ± 2.18	36.58 ± 2.37	36.15 ± 2.97
VLDL (mg/dL)	15.98 ± 2.67	16.08 ± 2.46	15.46 ± 1.16	15.37 ± 1.26
Triglycerides (mg/dL)	83.16 ± 1.47	82.54 ± 1.46	83.54 ± 1.29	83.68 ± 1.21
TC/HDL Ratio	3.82 ± 2.17	3.64 ± 1.48	3.68 ± 2.34	3.78 ± 2.06
Blood Glucose (mg/dL)	114.45 ± 3.98	110.68 ± 3.59	113.49 ± 3.65	113.98 ± 3.35

Table 6. Effect of MNK on Organ Weight.

Organ	Control	50 mg/kg	250 mg/kg	500 mg/kg
Liver (g)	4.98 ± 0.18	5.08 ± 0.16	4.84 ± 0.27	5.26 ± 0.25
Heart (g)	0.73 ± 0.06	0.78 ± 0.04	0.74 ± 0.08	0.75 ± 0.02
Lung (g)	2.01 ± 0.34	2.12 ± 0.27	2.08 ± 0.26	2.10 ± 0.32
Spleen (g)	0.81 ± 0.05	0.72 ± 0.08	0.68 ± 0.06	0.67 ± 0.07
Ovary (g)	2.05 ± 0.17	1.98 ± 0.14	1.87 ± 0.19	1.98 ± 0.16
Testes (g)	1.43 ± 0.18	1.40 ± 0.13	1.38 ± 0.17	1.42 ± 0.12
Brain (g)	1.52 ± 0.15	1.57 ± 0.17	1.48 ± 0.19	1.46 ± 0.19
Kidney (g)	0.68 ± 0.03	0.72 ± 0.04	0.70 ± 0.07	0.72 ± 0.06
Stomach (g)	1.30 ± 0.15	1.22 ± 0.21	1.27 ± 0.18	1.23 ± 0.12

Histopathological evaluation of vital organs was performed to assess potential tissue-level toxicity following 28-day oral administration of MNK at a high dose (500 mg/kg). Heart: Examination of myocardial tissue revealed normal myocytes with intact architecture in both control and treated groups, indicating no cardiotoxic effects (Figure 1A & 1B). Lungs: Pulmonary tissue showed normal alveolar structure and bronchioles with no signs of congestion, edema, or inflammation (Figure 2A & 2B). Spleen: Trabecular and red pulp structures appeared normal, with no evidence of degeneration or lymphoid depletion (Figure 3A & 3B). Liver: Hepatocytes displayed normal morphology, with intact central veins and portal triads; no signs of necrosis or fatty changes were observed (Figure 4A & 4B). Kidney: Renal sections revealed normal glomeruli, Bowman's capsules, and tubular structures, with no indications of degeneration, inflammation, or necrosis (Figure 5A & 5B).

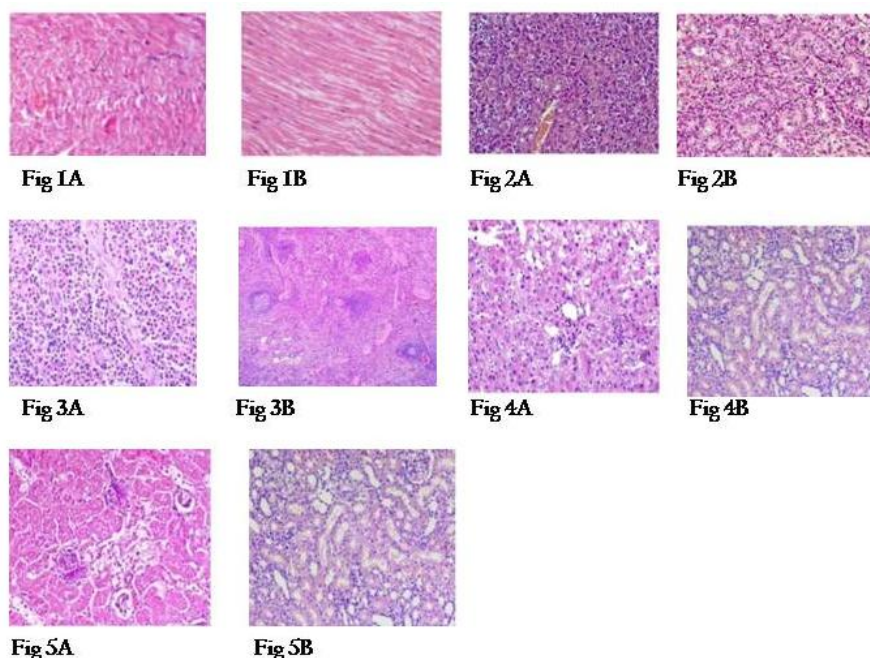


Fig 1: Histopathological observation of Heart (Fig 1A: Control, Fig 1B: 500 mg/kg MNK)
Fig 2: Histopathological observation of Lungs (Fig 2A: Control, Fig 2B: 500 mg/kg MNK)
Fig 3: Histopathological observation of Spleen (Fig 3A: Control, Fig 3B: 500 mg/kg MNK)
Fig 4: Histopathological observation of Liver (Fig 4A: Control, Fig 4B: 500 mg/kg MNK)
Fig 5: Histopathological observation of Kidney (Fig 5A: Control, Fig 5B: 500 mg/kg MNK)

The present study evaluated the acute and 28-day repeated oral toxicity of Manjal Noi Kudineer (MNK) in Wistar rats to establish its safety profile. In the acute toxicity study, administration of MNK at a single oral dose of 2000 mg/kg did not result in mortality, behavioral abnormalities, or clinical signs of toxicity, suggesting an LD₅₀ greater than 2000 mg/kg. This finding aligns with previous reports on other polyherbal decoctions used in Siddha medicine, which are generally considered safe at therapeutic doses (Yogananth *et al.*, 2021; OECD, 2001). The absence of acute toxicity indicates that MNK has a wide margin of safety, supporting its traditional use in humans. In the 28-day repeated oral toxicity study, all rats survived without any observable signs of morbidity or abnormal behavior. Body weight, food, and water intake remained comparable to the control group, indicating that MNK did not negatively impact growth or metabolic homeostasis. Stable body weight gain in treated animals corroborates the non-toxic nature of the formulation, as changes in body mass are often early indicators of systemic toxicity (Paget & Barnes, 1964; Yogananth *et al.*, 2020). Hematological analysis revealed no significant alterations in RBC, WBC, Hb, platelets, MCV, or differential leucocyte count across all dose groups. The minimum RBC value ($7.98 \times 10^6/\text{mm}^3$) and maximum value ($8.32 \times 10^6/\text{mm}^3$) fell within the physiological range, confirming that MNK does not exert hematotoxic effects. These findings are consistent with prior studies on polyherbal formulations showing negligible effects on hematopoiesis and immune cell homeostasis (Loha *et al.*, 2019). Biochemical assessments of hepatic function (AST, ALT, ALP, total protein, albumin, bilirubin) indicated no significant changes

compared to controls, suggesting that MNK is non-hepatotoxic. Minor fluctuations observed, such as a slight decrease in total bilirubin (0.25 mg/dL at 500 mg/kg), were not statistically significant and remained within normal ranges. Similarly, renal function markers (urea, creatinine, uric acid, electrolytes) showed no significant deviations, indicating preserved renal integrity and metabolic function. Stability in these parameters confirms that the formulation is unlikely to induce nephrotoxicity or disrupt electrolyte balance (WHO, 2011; OECD, 2008).

The lipid profile of treated animals also remained within normal limits, indicating that MNK does not adversely affect lipid metabolism or cardiovascular risk markers (Yogananth *et al.*, 2021; Loha *et al.*, 2019). Organ weight analysis further supported systemic safety, as all vital organs, including liver, kidneys, heart, lungs, spleen, brain, and gonads, displayed weights comparable to controls (OECD, 2008; WHO, 2011). Finally, histopathological examination of heart, lungs, spleen, liver, and kidney revealed no structural abnormalities, necrosis, or inflammatory changes. Normal architecture in these organs confirms the absence of tissue-level toxicity, corroborating the biochemical and hematological findings (Yogananth *et al.*, 2024; Government of India, 2008).

CONCLUSION

The present study demonstrates that Manjal Noi Kudineer (MNK), administered orally up to 500 mg/kg for 28 days, is safe and well-tolerated in Wistar rats. No mortality, behavioral abnormalities, or significant changes in body

weight, food and water intake were observed. Hematological, biochemical, and lipid profiles remained within normal ranges, while organ weights and histopathological examinations revealed no structural alterations or tissue damage. These findings collectively indicate that MNK exhibits a favorable safety profile, supporting its traditional use in Siddha medicine and providing a scientific basis for further pharmacological and clinical investigations.

ACKNOWLEDGMENT

The authors express sincere thanks to the head of the Velumailu Siddha Medical College and Hospital, Sriperumbudur, Kancheepuram, Tamil Nadu, India for the facilities provided to carry out this research work.

CONFLICT OF INTERESTS

The authors declare no conflict of interest

ETHICS APPROVAL

Not applicable

FUNDING

This study received no specific funding from public, commercial, or not-for-profit funding agencies.

AI TOOL DECLARATION

The authors declares that no AI and related tools are used to write the scientific content of this manuscript.

DATA AVAILABILITY

Data will be available on request

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