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Original Research Article

Effect of Aqueous Extract of Terminalia Bellirica Fruit Pulp: An Animal Experimental Study

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Abstract

Aim: Effect of aqueous Extract of Terminalia bellirica fruit pulp on Alcohol affected learning in swiss albino mice.

Methods: This study was conducted in the Department of Pharmacology, ANMMCH Gaya, Bihar, India for 1 year. 64 Swiss albino mice of either sex weighing 20-30 grams bred and maintained under standard conditions in the central animal house in AJIMS and RC were used. The standard drug Piracetam was obtained from micro labs. The dried fruit of Terminalia bellirica was obtained by Ayurvedic Dispensary, BIHAR. Aqueous Extract of Terminalia bellirica fruit pulp [AETB] -Investigational product AETB (dose1) – 9mg/kg orally. AETB (dose2) – 18mg/kg orally. AETB (dose 3) - 36mg/kg orally.

Results: Piracetam (200mg/kg) and AETB (36mg/kg) significantly reduced the time taken to reach the reward chamber ($p < 0.001$), while Ethanol (1.5g/kg) significantly increased the latency when compared to vehicle treated groups ($p< 0.001$). Animals pre-treated with Piracetam (200mg/kg) and AETB (9mg/kg) significantly reduced the enhanced latency induced by Ethanol when compared to Ethanol alone treated group ($p< 0.001$).

Conclusion: Our study indicates that the test drug enhances the learning process and is comparable to the standard drug Piracetam at higher doses (36mg/kg). Also, it can oppose the alcohol induced learning impairment at lower doses (9mg/kg).

Keywords: Piracetam, Mice, AETB.

Introduction

Ayurveda, the oldest healing science, focuses on treating different ailments through balancing the three pillars of life, *vat*, *pitta* and *kaf*. The role of different plant extracts in maintaining this balance and also treating various diseases is also well documented. However, in recent past in depth research is focusing more on the phytochemical analysis of these extracts and their effects on various disease conditions *in vitro*. These plant extracts have proven to be important for re-establishing body's equilibrium and providing resistance against infection. They also possess the restorative and rejuvenating powers as they act on the immune system and positively affect the response of the body towards infection [1]. Selective COX 2 inhibitors han advantages over conventional NSAIDs, it is known for

its adverse effects on cardiovascular system and moreover many drugs in this group have been already withdrawn from the market. Nimuselide even though initially has shown promising results but has been banned in many countries due to its liver toxicity effects. On the contrary, paracetamol had very efficacious analgesic and antipyretic effect but has very poor anti- inflammatory activity thus exhibiting its drawbacks and also not safe in overdose [2]. Ani-inflammatory activity of corticosteroids is well known but are devoid of analgesic and also antipyretic activity and moreover they carry their own adverse effects in overdose and especially on long term use. They also delay the wound healing process in the body [3]. Opioids are known to act by central mechanism but is devoid of anti-inflammatory and antipyretic effect and moreover carries high risk of abuse and dependence liability [4]. Several evidence suggest the role of herbal medicine in treatment of diseases and Ayurveda stands as a backbone for the rationalized treatment in Indian system medicine [5]. From many centuries, plants and their products or byproducts are known to possess analgesic, anti-inflammatory and antipyretic affects [6]. *Terminalia bellerica* is well mentioned in Indian system of medicine. It is a tree, and its fruit pulp is mentioned to be having many therapeutic uses in the management of pain, diarrhea, hypertension, infections, spasms, asthma and other conditions.

Materials and methods

This study was conducted in the Department of Pharmacology, ANMMCH Gaya, Bihar, India for 1 year. after taking the approval of the protocol review committee and institutional ethics committee. 64 Swiss albino mice of either sex weighing 20-30 grams bred and maintained under standard conditions in the central animal house in AJIMS and RC were used. They were kept at the animal house of the institute, in clean, clear polypropylene cages in groups of six and maintained at standard laboratory temperature and humidity (42-60%) with light/dark cycle of 12:12 hours. Animals were provided with standard diet and water ad libitum. They were allowed to acclimatize to the laboratory conditions for one week.

The experiment was performed as per the Committee for Purpose of Control and Supervision on Experiments on Animals (CPCSEA) guidelines.

The standard drug Piracetam was obtained from micro labs. The dried fruit of Terminalia bellirica was obtained by Ayurvedic Dispensary, BIHAR. Ethanol was obtained from Changshu Yanguan Chemical, China. About 1000 g of air-dried crude powder of Terminalia bellirica fruit pulp was extracted with water in Soxhlet extractor for 36.0 hours. It was dried and reduced under controlled pressure and temperature (40-50°C) using a rotator evaporator. The aqueous extract had a yield of brownish mass weighing 150g. The yield obtained was 15% w/w with respect to dried powder [7]. Piracetam 200mg/kg Body weight, administered orally. Alcohol (15%) 1.5g/kg, administered per oral [8] Aqueous Extract of Terminalia bellirica fruit pulp [AETB] -Investigational product AETB (dose1) – 9mg/kg orally. AETB (dose2) – 18mg/kg orally. AETB (dose 3) - 36mg/kg orally [7].

Methodology

Swiss albino mice (n=64) of either sex weighing 20-30g were divided into 8 groups of 8 mice each. Drugs were given orally after 12 hours of fasting. Group I mice received 10ml/kg of Normal Saline, Group II mice received Piracetam 200mg/kg, Group III received AETB 36mg/kg, Group IV received ethanol 1.5g/kg orally, Group V received ethanol (1.5g/kg) + piracetam (200mg/kg), Group VI mice received ethanol (1.5g/kg)+AETB(9mg/kg), Group VII mice received ethanol (1.5g/kg) + AETB (18mg/kg), Group VIII mice received ethanol (1.5g/kg) + AETB(36mg/kg). Time taken by the animal to reach the reward

chamber from the start chamber (TRC) in Hebb-William maze was used as a parameter to evaluate the learning. [8]

Apparatus

Hebb- William Maze [9]

It is a tool for incentive based exteroceptive behavioural model. The total time taken by the mice to reach the reward chamber from the start box indicates learning. The animals are acclimated to the maze environment through timed exposure periods. During the testing phase, the mice are placed in the start box and the door is opened to facilitate the entry of the mice into next chamber. The door of the start box is closed immediately after the animal moves into the next chamber to prevent back entry. The time taken by the animal to reach the reward chamber from the start box is recorded. (TRC)

Statistics

Results were analysed by ANNOVA followed by post hoc Dunnet's multiple comparision test. The observations were mean SD $p < 0.05$ was considered statistically significant.

Results and Discussion

Acute alcohol administration showed increase in TRC. Whereas acute administration of Aqueous extracts of Terminalia bellirica fruit pulp showed a decrease in TRC when compared to the control group. The TRC values for the groups that were administered AETB along with acute alcohol administration showed decrease in TRC values compared to the negative control.

Table 1: Number of animals

Animals	No.
Swiss albino mice	64

Ethanol can cause direct alteration of various ion channels, receptors and enzymes. These actions contribute to changes in synaptic function and plasticity. Ethanol augments the actions of certain GABA receptors and inhibits the effects of glutamate. Ethanol also promotes neorosteroid synthesis locally in the brain. This increases steroid levels in the hippocampus within few minutes of exposure. These are likely to cause changes in cognition [10].

Table 2: Parameter

PARAMETER	P
Piracetam (200mg/kg) and AETB (36mg/kg)	< 0.001
Piracetam (200mg/kg) and AETB (9mg/kg)	< 0.001

In the present study, Piracetam (200mg/kg) and AETB (36mg/kg) significantly reduced the time taken to reach the reward chamber ($p < 0.001$), while Ethanol (1.5g/kg) significantly increased the latency when compared to vehicle treated groups ($p< 0.001$). Animals pre-treated with Piracetam (200mg/kg) and AETB (9mg/kg) significantly reduced the enhanced latency induced by Ethanol when compared to Ethanol alone treated group ($p< 0.001$). However, the test drug AETB at higher doses (18mg/kg and 36mg/kg) failed to reduce the enhanced latency induced by Ethanol. Decrease and increase in latency period in Hebb- William Maze is indicative of improvement and impairment of learning process respectively. In the present study, the test drug alone at the highest dose tested, produced the effect similar to that of Piracetam and opposite to that of Ethanol. This suggests that the test drug enhances learning process.

The terpenoids (belleric acid and chebulagic acid), saponins (bellericoside and bellericanin) and tannins in general are known to have neuroprotective activity [11]. This was a single day study, and the protective action of Terminalia Bellirica was evaluated for acute alcohol induced impairment in learning process. Further chronic studies would be use full in evaluating the long-term effects of Terminalia Bellirica and its effects on chronic alcohol induced memory impairment.

Conclusion

Our study indicates that the test drug enhances the learning process and is comparable to the standard drug Piracetam at higher doses (36mg/kg). Also, it can oppose the alcohol induced learning impairment at lower doses (9mg/kg).

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