ANXIOLYTIC EFFECT OF *LAWSONIA INERMIS* LINN (HENA) ON LIGHT DARK BOX ACTIVITY

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ABSTRACT

*Lawsonia inermis* traditionally use as emollient on scalp as well as on palm to reduce warmness, extract made from leaves, pharmacological activities includes sedative, nootropic. The aim of current study was to evaluate Methanolic extract of *Lawsonia inermis* (MeLi) for its anxiolytic potential, for this purpose the light/dark box activity test used which is commonly used to assess anxiety-like behaviors and validate the pharmacological effects of neuroactive plant extracts and compounds. MeLi at 100 mg/kg i.p exhibited significant increase in time spent in light area with respect to control animals. Moreover the reduction in anxiety behavior, also demonstrated by significant increase in number of entries in the light compartment relative to the dark compartment of the testing apparatus. In conclusion MeLi possesses diazepam, buspirone like anti-anxiety activity in mice.

Key Words: *Lawsonia inermis*, anxiety, pharmacological, emollient and behavior

INTRODUCTION

*Lawsonia inermis* commonly known as Henna (Arabic)4,5 is a flowering plant, 2-6m in height, belongs to family Lythraceae and have been used in traditional herbal medicine for many years1. Ethnanopharmacological uses of *L. inermis* includes relieve unilateral headache, lumbago, bronchitis, boils, ophthalmia, syphilis, sores, amenorrhea, scabies, diseases of the spleen, dysuria, bleeding disorder, skin diseases, diuretic, antibacterial,
Antifungal, anti-amoebiasis, astringent, anti-hemorrhagic, hypotensive, sedative. *L. inermis* has been reported to possess antioxidant, anti-corrision, anti-inflammatory, analgesic, antipyretic, anti-parasitic, antimicrobial, antibacterial, anti-tumoral activity, cytotoxic, hypoglycaemic, hepatoprotective, protein glycation inhibitory, trypsin inhibitory, wound Healing, tuberculostatic. Acetone soluble fraction of petroleum ether extract of *Lawsonia inermis* leaves also possesses psychopharmacological activities sedative ([Meddah et al., 2012](#)), and the effect of acetone soluble fraction of petroleum ether extract of *Lawsonia inermis* leaves on memory, anxiety and behaviour was assessed using elevated plus maze and passive shock avoidance paradigms. In literature it is proposed that these CNS activities are due to presence of core chemical components are 2-hydroxynaphthoquinone (lawsone), mannite, tannic acid, mucilage and gallic acid ([Kathem et al., 2008](#)).

**MATERIAL AND METHODS**

**Plant Collection and Authentification**

*Lawsonia inermis* leaves were collected at the Botanical Garden of the Hamdard University. The leaves were identified and authenticated where a voucher specimen with the number (FHI 109601) was deposited.

**Extraction**

The leaves were air-dried, pulverized and 1000g was macerated for 72 h in 1 L of 50% methanol, was decanted, filtered several times using cotton wool and Whatman’s No.1 filter paper and concentrated using rotary evaporator at the Pharmacology Laboratory of Hamdard University the dark green colored extract obtained.

**Animals**

Albino mice (30 – 35 g) of either sex were obtained from the Animal House of Dr.HMI Institute of Pharmacology and Herbal Sciences, College of Medicine, and were housed in plastic cages at room temperature. They were fed with balanced rodent pellet diet and water ad libitum. The animals were acclimatized for at least 1 week before being used for experiments.

**Drugs and Chemicals**

Diazepam (Hoffman-La Roche, Switzerland), buspiron and Methanolic Extract of *Lawsonia inermis*. 
Experimental Design
Total thirty six (N=36) mice were randomly divided into six groups. For each of the model studied (n=6). The groups include control (vehicle) and standard drugs (Diazepam, Bupirone, 1mg/kg) and three groups of MeLi (50, 100 and 200 mg/kg.).

RESULTS
Student T-tests (two-tailed) were performed for analysis of zone preference in the light/dark activity test. MeLi at the dose of 100 mg/kg significantly (p>0.005) delayed latency with respect to control animals as shown in Figure 1. At same dose MeLi treated animals exhibited more number of entries in light area with respect to control animals Figure 2. Whereas the time spent and number of entries by MeLi treated animals were non-significantly different from diazepam and buspirone (1mg/kg).

![Fig.1. Student T-Test, p>0.005, p>0.0001 (n=6)](image1.png)

![Fig.2. Student T-Test, p>0.0001, p>0.005 (n=6)](image2.png)
Legends
Student T-tests (two-tailed) were performed for analysis of zone preference in the light/dark box activity test. The results are presented in Fig. 1. Number of entries of tested NMRI mice significantly more (p>0.0001) as compare to buspirone and approximately equal effect to diazepam that indicates herbal drug have capability to lead to relieve anxiety significantly.

DISCUSSION
In this study, we evaluated anxiolytic potential of MeLi using light/dark preference test in NMRI mice (n=6). The significant increase in time spent light area by MeLi treated animals indicates that MeLi possesses significant anxiolytic activity at the dose of 100 mg/kg. The furthermore MeLi treated animals displayed an immediate and strong avoidance of the dark zone and exhibited increased number of entries. These findings provide a clear demonstration that NMRI mice displayed diazepam like anti-anxiety property which at dose of 1mg/kg caused. Our results are in agreement with previous studies which suggested that …… Possesses anxiolytic activity as same manner (REF).11,12,13,14,15

The view that dark environments (encountered outside the night fall) appear to be perceived as aversive conditions, which stimulate the expression of anxiety-like behaviors in NMRI mice.

CONCLUSION
This behavioral assay is conclude that these plants have activity against anxiety disorder and can accommodate drug screening of both anxiolytic plants amenable to automation and high-throughput tendency in a future. These findings presented significant herbal pharmacological validation of the light/dark box activity test for NMRI mice and the nature of dark-avoidance behaviors as anxiety like behaviors. These findings show that NMRI mice are capable of showing anxiety-like behaviors. In sum, we show that the light/dark box activity test is a relatively simple and suitable behavioral assay for NMRI mice, and presents both face and pharmacological predictive validity. This behavioral-based assay is also versatile and can accommodate drug screening of both anxiolytic and anxiogenic compounds while eventually amenable to medium/high-throughput capacity in a near future. In the future, such behavioral models will improve pre-clinical drug screening methodologies towards the goal to uncover novel neuroactive drugs.
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REFERENCES