



## Original Research Article

## The spontaneous motor action of alcoholic excerpt of *Withania coagulans* fruits in Swiss albino mice by Actophotometer

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## ABSTRACT

**Background:** People spend about one third of their life in sleeping. The various sedative and hypnotic drugs currently used have various adverse drug reactions. In 1977 Budhiraja et al. have done insufficient work on the *Withania coagulans* – a susceptible species that is not found plentiful in the world. Therefore, it was critical to discover the Central Nervous System (CNS) depressant activities of alcoholic extract of *Withania coagulans* fruits in Swiss albino mice by means of actophotometer.

**Materials and Methods:** The spontaneous locomotor action was assessed by means of the actophotometer. The CNS depressant medications reduce the exploratory action in rodent as they weaken the motor coordination so that rodent is stationary for a considerable time. Thus, there is fewer interruptions to the rays of light. This spontaneous locomotor movement duration is statistically compared with the control, standard and the test drugs.

**Results:** There was statistically highly significant ( $p$ -value < 0.001) relationship witnessed between alcoholic excerpt of *Withania coagulans* fruits with spontaneous locomotor action in Swiss albino mice on the photoactometer.

**Conclusion:** The alcoholic excerpt of *Withania coagulans* fruits established the CNS depressant action in Swiss albino mice by photoactometer.

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### 1. Introduction

The importance of sleep can be understood from the fact that people spend about one third of their duration in sleep every day. The prevalence of sleep disorders is about 15% in USA,<sup>1</sup> and that in the adolescent is 6%.<sup>2</sup> Yet the prevalence in India is not clear.<sup>3</sup> Insomnia causes depression and vice versa.<sup>4</sup> Though benzodiazepines are most widely used for the treatment of insomnia among various medications prescribed, they are not devoid of the side effects. Thus, there is always a need to invent the new drugs.

The word Solanaceae means soothing. The Solanaceae family covers 84 genera comprising approximately 3,000

species. The two genera namely *Withania* and *Physalis* play a vital role in the Unani and Ayurvedic systems in the South East Asia. The 23 acknowledged *Withania* species are extensively scattered in the drier portions of tropical and subtropical zones.<sup>5–7</sup> Out of all these species, the *W. coagulans* and *W. Somnifera* are financially and curatively important.<sup>8–10</sup> As the name suggest the herb has milk - coagulating property due to the presence of an enzyme, which is a plant hormone. The active principle named “Withanin” found in the seeds of the fruits is ferment like animal rennet. Thus, it is also called as vegetable rennet or Indian rennet.<sup>11,12</sup> It is also used to control the Diabetes Mellitus.<sup>13</sup>

Spontaneous motor action (SMA) is also referred as spontaneous locomotor action (SLA) in mice was used to

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assess the Central Nervous System (CNS) depressant effect of *Withania coagulans*. This was assessed by means of actophotometer (photocell action cage). It was found that if the depressant had the sedative property then there was less disruption of the beams of light in the actophotometer as the rodents did not try to explore much because of the sedation.<sup>14</sup> On the other hand, stimulants had the opposite effect.<sup>15</sup> The *Withania coagulans* was not explored much for its CNS depressant effect. Thus, it was thought worthwhile to assess the CNS depressant action of the alcoholic excerpt of medicinal herb in Swiss albino mice by acto-photometer for spontaneous locomotor action.

## 2. Materials and Methods

### 2.1. Actophotometer apparatus

It was 30×30×30 cm in dimension. The two photocells were fixed 3 cm above the floor. Each wall of the cage had 3 equidistant holes in a horizontal array, 7 cm above the floor. Through the holes 3 light beams were passed from all sides of the apparatus. They crossed in the centre of the cage. There were the infra-red filters to minimize the effect of light on behaviour. The number of interruptions of the light beams were recorded on digital counters in the instrument automatically.

### 2.2. Procedure

Each mouse was kept for 5 minutes in the action cage. If mouse started the exploratory action, then the beam of light was interrupted. Each interruption was picked up automatically by the digital recorder in the instrument and was counted. If the animal stopped the locomotor action, the digital counter stopped recording. Again when animal started walking, the action was recorded. The "Walks" were defined as the number of durations the mouse moved with all four feet in the space between two opposite walls of the cage. The total duration consumed in walking was recorded as above.

### 2.3. Control, standard and experimental medications

Distilled water was given as vehicle for control. Diazepam was used as the standard medication. The animals were treated 30 min prior the experiment with the test medications (WCFAIcE of 200 mg/kg, 500mg/kg and 1000 mg/kg dosages p. o.). Yet, the experimental medication was given every day for 30 days throughout the period of experiment. The mice were witnessed for 5 min. Recordings were done on Day 1, Day 8, Day 15, Day 23 and Day 30 for all the groups. The recordings were taken half an hour after medication administration to the respective groups. After each trial the equipment was cleaned with super hypochlorous water to prevent the bias based on olfactory cues.

Medications were given in the following manner:

Control: Vehicle (Distilled Water) 2 ml/kg p. o. O.D. for 30 days.

Standard: Standard medication (Diazepam) 5mg/kg i. p. once half an hour prior experiment.

ALC-200: WCFAIcE 200 mg/kg p. o. O.D. for 30 days.

ALC-500: WCFAIcE 500 mg/kg p. o. O.D. for 30 days.

ALC-1000: WCFAIcE 1000 mg/kg p. o. O.D. for 30 days.

Where WCFAIcE = *Withania coagulans* fruits alcoholic excerpt.

## 3. Results

As shown in the Table 1, on Day 1 to Day 8 there was no statistically significant difference in the spontaneous locomotor action by mice. Yet, on days 15, 23 and 30 spontaneous locomotor action by mice in actophotometer fell highly significantly ( $p < 0.001$ ) for all the 3 dosages of 200 mg/kg, 500 mg/kg and 1000 mg/kg of WCFAIcE compared to control. Furthermore, dose response relationship was witnessed for these dosages. To conclude, this fall in spontaneous locomotor action by experimental medication was similar to that of the standard diazepam.

## 4. Discussion

As shown in the Table 1, on Day 1 to Day 8 there was no statistically significant difference in the spontaneous locomotor action by mice. Yet, on days 15, 23 and 30 spontaneous locomotor action by mice in actophotometer fell highly significantly ( $p < 0.001$ ) for all the 3 dosages of 200 mg/kg, 500 mg/kg and 1000 mg/kg of WCFAIcE compared to control. The dose response relationship was also witnessed.

Our study is unique one as there are no other experiments reported which have investigated the effect of any part of *Withania coagulans* on locomotor action of mice by means of actophotometer. Yet, in 1977 Budhiraja et al. reported the significant ( $p < 0.001$ ) fall in spontaneous locomotor action at the dosages of 1 g/kg, 200-400 mg/kg and 5 ml/100 g of the alcoholic excerpts of *Woliithania coagulans*.<sup>16</sup> He further concluded the CNS depressant action based on the pentobarbitone sleeping duration.<sup>16</sup> In another study, at the dosages of 100 mg/kg and 200 mg/kg intraperitoneal, leaf alcoholic excerpt of the *Withania Somnifera* (a similar species as that of *Withania coagulans*) significantly ( $p < 0.001$ ) fell the locomotor action and dose response relationship was also witnessed.

It was used to record the spontaneous locomotor action of mice. In this action "walks" and "rears" can be measured.<sup>17</sup> Yet, this instrument does not measure the "grooming" and "washing" by rodents.<sup>17</sup> In our experiment we focused on the "walk" part to measure the locomotor action. Since, it took 15 days for the effect to appear, the results cannot be

**Table 1:** Effect of oral administration of WCFAlcE on spontaneous locomotor action of mice in seconds (mean±SD) in actophotometer.

	Control	Standard	ALC-200	ALC-500	ALC-1000
Day 1	254.50± 15.63	258± 30.35	242.16± 36.36	234.83± 26.97	227.66± 27.06
Day 8	270.33± 82.41	231.33± 20.14	235± 48.55	229.66± 48.66	219± 66.66
Day 15	263.50± 7.55	176.66± 40.91***	170.33± 17.69***	149.50± 22.13***	131.83± 20.52***
Day 23	259.66± 7.28	158.50± 27.34***	142.16± 23.73***	123± 15.77***	105.66± 24.90***
Day 30	251.66± 6.43	128.50± 26.28***	111± 17.82***	93.50± 15.04***	70.33± 24.32***

WCFAlcE : Withania coagulans fruits alcoholic excerpt

Control: Vehicle (Distilled Water) 2 ml/kg p. o. O.D.for 30 days.

Standard: Standard medication (Diazepam) 5 mg/kg i. p. half an hour prior experiment.

ALC-200: WCFAlcE 200 mg/kg body mass p. o. O.D.for 30 days.

ALC-500: WCFAlcE 500 mg/kg body mass p. o. O.D.for 30 days.

ALC-1000: WCFAlcE 1000 mg/kg body mass p. o. O.D.for 30 days.

extrapolated in acute condition, rather the test medication can be used in chronic conditions.

The stimulants rise the spontaneous locomotor action in contrast to the depressants which fall the same.<sup>18</sup> Thus, the test substance might have CNS depressant action. Hence it can be concluded that similar with anolides in both the species might be responsible for this action. To conclude, our study herb W. coagulans demonstrated the CNS depressant action in the Swiss albino mice for the 3 dosages of WCFAlcE.

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## 6. Conflict of Interest

There is no conflict of interest among authors

## 7. Ethical Approval

Research was approved by Institutional as well as the Animal Ethics Committee of MGIMS, Sevagram.

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