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**RESEARCH ARTICLE……!!!**

# *IN VIVO* EVALUATION OF ANALGESIC ACTIVITY OF WHOLE PLANT EXTRACTS OF *PHYSALIS ALKEKENGI* USING VARIOUS EXPERIMENTAL MODELS

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**KEYWORDS:**

*Physalis alkekengi*, writhings, Eddy's hot plate, Aspirin.

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## ABSTRACT

The present study is performed on the whole plant ethanolic extract of *Physalis alkekengi* (EEPA) and successive ethanolic extract of *Physalis alkekengi* (SEEPA) to screen the phytochemical compounds and analgesic activity. Analgesic activity is being assessed on mice using acetic acid induced writhings and on rats using Eddy's hot plate method.. The extracts are used in the doses of 150mg/kg and 300mg/kg *p.o.* showed the significant peripheral and central analgesic activity comparing with the control group which is given with normal saline 0.9%. Aspirin (150 mg/kg *p.o.*) is being used as a standard for acetic acid induced writhing method and Tramadol (5mg/kg) for Hot plate method.

**INTRODUCTION:**

The role of ancient healing systems which include Ayurveda, Siddha and Unani has been increased in the market due to serious obsession with the modern medicinal system since many decades (Biswa Nath Das *et al*., 2012). During the past two decades the traditional system has gained importance in the field of medicine. The usage of these herbal medicines has increased due to their quality, safety and efficacy. The emerging output of these systems lead to screen scientifically the various claims of herbal medicines (Anil *et al*., 2011)(Kumbhar *et al*., 2012).

According to international association for the study of pain (IASP) the term "Pain" can be defined as an unpleasant sensory and emotional experience associated with actual or potential tissue damage or in terms of such damage (Abdullahi *et al*., 2013). Analgesics act by increasing the pain threshold to external stimuli and relieve pain as a symptom, without affecting its cause. Previous studies reveal that the imbalance of pro-oxidants and antioxidant factors in the living systems might be a possible chance of causing pain. Thus, endogenous antioxidant compounds might be beneficial on the systems that are deprived from required amounts of endogenous antioxidants as in the case of some cardiovascular diseases, tumour etc (Raquibul *et al*., 2009). Currently used drugs have reported adverse effects which leads to search for alternatives preferably the plant products (Hassan *et al*., 2013).

Noxious pain can be produced by using various chemicals, physical pressure or by thermal methods. The nociceptive afferent fibres located in the dorsal horn of spinal cord releases two neurotransmitters glutamate and substance P which stimulate the second order sensory neurons (Akriti *et al*., 2011).

*Physalis alkekengi* belongs to the family solanaceae is widely distributed in Asia and Europe. It is extensively used in herbal medicine as antibacterial, antileishmanial, antitumour, antispermatogenesis, anti-conception, antitussive , expectorant (Jasem *et al*., 2011), anti-diabetic (Sanchooli N, 2011) and as an antioxidant (Mahdieh *et al*., 2012). According to Jasem Estakhr *et al.*, 2011 the extracts of leaves and fruits of *Physalis alkekengi* possess analgesic activity. The extracts are widely used to treat kidney & bladder stone, inflammation, general edema, febrile diseases and arthritis.(Veronica *et al*., 2014). The main phytochemical constituents of *Physalis alkekengi* include alkaloids, flavanoids, sterols, fattyacids, amino acids and sterols. Chemical studies have demonstrated the presence of physalin, citric acid and Vitamin C as major components in the extract (Sanchooli N, 2011). Physalins and theophysains have tremendous biological activities (Wan-Xuan *et al*., 2013). There are many known toxicities of the synthetic antioxidants therefore the interest in antioxidants from plant origin preferably increased in these years (Raquibul *et al*., 2009). The main

aim of the study is to evaluate the analgesic activity of various doses such as 150mg/kg & 300mg/kg of EEPA and SEEPA using acetic acid induced writhing and hot plate method.

## MATERIALS AND METHODS

### Plant Materials:

*Physalis alkekengi* was obtained from Tirumala Hills, tirupathi and was authentificated by botanist Dr. T. Madavachetty, SVU, Tirupathi. The whole plant of *Physalis alkekengi* was shade dried, powdered and extracted with ethanol for EEPA and successively extracted with petroleum ether, chloroform, ethyl acetate followed by ethanol to obtain SEEPA. The obtained extracts of EEPA and SEEPA are dried using rotary evaporator and used for phytochemical evaluation.

### Animals:

Healthy male swiss albino mice weighing 18-25g were selected for acetic acid induced writhing method and healthy male wistar rats weighing 180-220g were selected for hot plate method. The animals are obtained from teena labs, bachupally, Hyderabad. The experimental animals are acclimatized for a week in the standard cages at 23 ± 2°C under 12 hrs dark/light cycle and standard conditions in the animal house at CMR College of pharmacy, Hyderabad approved by CPCSEA. The study was conducted in accordance with accepted principles outlined in the "Guide for the Care and use of Laboratory Animals" prepared by the National Academy of Sciences and published by the National Institutes of Health.

### Analgesic activity

The analgesic activity is being evaluated by using following methods

1. **Acetic acid induced writhing in mice**

The experimental animal's mice are divided into six groups containing six in each. Group 1 serves as control treated with normal saline and group 2 as standard treated with aspirin 150 mg/kg (Sattayasai *et al*., 2012). Group 3 and group 4 were treated with EEPA 150 mg/kg and 300 mg/kg. Group 5 and group 6 are given SEEPA 150 mg/kg and 300 mg/kg respectively. After 30 mins of the drug treatment 1 % acetic acid (1ml/100g) is given by *i.p* route. The number of writhings are recorded after 5 mins of 1% acetic acid administration for the duration of 10 mins (Bhattacharya *et al*., 2014).

2. **Hot plate method**

Male albino rats of Wistar strain were divided into six groups of six each. The animals received the treatment as per the treatment schedule: Group 1 serves as control treated with normal saline and group 2 as standard treated with Tramadol 5 mg/kg. Group 3 and group 4 were treated with EEPA 150 mg/kg and 300 mg/kg. Group 5 and group 6 are given SEEPA 150 mg/kg and 300 mg/kg respectively. After 30 mins of the treatment schedule the animals are positioned on eddy's hot plate which is maintained at a temperature of 55 ± 0.5°C with a cut off period of 20 seconds (Zulfiker *et*

*al*., 2010) (Ezeja *et al*., 2011). The reaction time was recorded when animals licked their fore or hind paws or jumped at 0, 30, 60, 90 and 120 mins after oral administration of the drugs as per the treatment schedule (Sagar Raut *et al*., 2011).

**Statistical Analysis:**

Values were expressed as mean ± SEM (n=6 animals per group). The statistical analysis was carried out using one way ANOVA and unpaired student's 't' test. Values are considered statistically significant at P<0.05, P<0.01, and P<0.001

## RESULTS

**Phytochemical screening**

| EXTRACTS | Alkaloids | Glycosides | Tannins | Flavanoids | Fatty acids | Saponins |
|---|---|---|---|---|---|---|
| EEPA | + | + | + | ++ | + | + |
| SEEPA | + | + | + | ++ | + | + |

Table 1: Phytochemical screening of the *Physalis alkekengi* extracts.

**Analgesic activity**

**a) Acetic acid induced writhing in mice**

Table 2: Effect of *Physalis alkekengi* on acetic acid induced writhing in mice.

| Group | Treatment (mg/kg) *p.o* | Mean No. of writhing in 10 mins ± SEM | % Inhibition |
|---|---|---|---|
| 1 | Normal saline 10ml/kg | 35.5 ± 2.50 | - |
| 2 | Aspirin 150mg/kg | 16.2 ± 1.60* | 54.46 |
| 3 | EEPA 150mg/kg | 25.5 ± 1.87* | 28.17 |
| 4 | EEPA 300mg/kg | 20.7 ± 1.97* | 41.69 |
| 5 | SEEPA 150mg/kg | 29.2 ± 1.83* | 17.75 |
| 6 | SEEPA 300mg/kg | 24.2 ± 1.33* | 31.83 |

Values were expressed as mean ± SEM (n=6 animals per group). *P<0.001 vs control group(Group 1).

**b) Hot plate method**

Table 3: Effect of *Physalis alkekengi* on latency to hot plate test

| Group | Treatment (mg/kg) *p.o* | Latency of paw withdrawl (Seconds) | | | | |
|---|---|---|---|---|---|---|
| | | **0 Mins** | **30 Mins** | **60 Mins** | **90 Mins** | **120 Mins** |
| 1 | Normal saline 10ml/kg | 9.5 ± 1.05 | 8.8 ± 0.75 | 7.7 ± 0.52 | 6.8 ± 0.41 | 5.8 ± 0.41 |
| 2 | Tramadol 5mg/kg | 8.8 ± 1.20** | 15.7 ± 1.51* | 14.7 ± 1.21* | 14.2 ± 0.75* | 13.3 ± 1.21* |
| 3 | EEPA 150mg/kg | 8.5 ± 1.04* | 13.0 ± 0.63* | 13.5 ± 0.84* | 13.2 ± 0.75* | 11.7 ± 0.82* |
| 4 | EEPA 300mg/kg | 07.8 ± 1.7*** | 14.3 ± 0.52* | 14.0 ± 0.90* | 13.5 ± 1.05* | 12.7 ± 0.82* |
| 5 | SEEPA 150mg/kg | 8.2 ± 0.75** | 12.7 ± 1.03* | 13.0 ± 0.63* | 12.2 ± 0.99* | 10.8 ± 0.73* |
| 6 | SEEPA 300mg/kg | 7.2 ± 0.75* | 13.2 ± 0.75 | 13.5 ± 0.84* | 13.3 ± 1.03* | 11.3 ± 0.52* |

Values were expressed as mean ± SEM (n=6 animals per group). *P<0.001, **P<0.01 and ***P<0.05 vs control group(Group 1)

**DISCUSSION:**

These studies help in identifying the valuable lead compounds for analgesic drugs, keeping in mind the side effects of NSAIDS and corticosteroids. In the present study, analgesic activity of EEPA and SEEPA was observed centrally and peripherally using hot pate method and acetic acid induce writhing method.

Acetic acid increases the visceral pain by triggering localized inflammation due to release of free arachidonic acid from the tissue phospholipid by cycloxygenase which leads to the elevation of peritoneal prostaglandin levels and finally involves in the release of pain mediators such as bradykinin, histamine and 5-hydroxytryptamine (Zulfiker *et al*., 2010). The analgesic principles in the plant extract would possibly act on the prostaglandin pathways. Acetic acid induced writhings method reveals the significant activity at the dose of 300mg for both extracts with inhibition of 41.69% for EEPA and 31.83% for SEEPA when compared with control group.

Hot plate method is sensitive to centrally acting analgesics. The latency of reaction time was gradually decreased with the dose of 300mg/kg of both EEPA and SEEPA extracts. It is a well established fact that any agent that causes prolongation of the hot plate latency using this test must be acting centrally (Zulfiker *et al*., 2010). The results of this method reveal that the extract shows significant central analgesic activity at the dose of 300mg/kg of EEPA at 30 mins interval and 300mg/kg of SEEPA at 60 mins. Therefore, the ethanolic extracts *Physalis alkekengi* might have central analgesic sssactivity.

The preliminary qualitative phytochemical screening shows the presence of alkaloids, flavanoids, saponins, tannins, and glycosides in the whole plant extracts. The analgesic activity may be attributed by the phytochemical constituents. *Physalis alkekengi* is well known for its antioxidant activity (Mahdieh *et al*., 2012). Studies had reported that flavanoids have a role in analgesic action by targeting the prostaglandins and alkaloids are known for their ability to inhibit pain perception. (Ibironke and Ajiboye, 2007)

The analgesic activity of the ethanolic and successive ethanolic whole plant extracts of *Physalis alkekengi* used in acetic acid induced writing in mice and Eddy's hot plate method in rats suggests that they have both central and peripheral analgesic activity.

**CONCLUSION**

In conclusion, the present study has revealed that the ethanolic extracts possess a significant analgesic activity both centrally and peripherally. Further studies has to be done to understand the precise mechanism as well identify and isolate the fractions responsible for the analgesic activity in the development of a potent analgesic compound.

## REFERENCES


1. Abdullahi MN , Ilyas N and Ibrahim H, (2013), Evaluation of phytochemical screening and analgesic activity of aqueous extract of the leaves of *Microtrichia perotitii* dc (Asteraceae) in mice using hotplate method. Med Plant Research. 3, 37-43
2. Akriti Pal and Pawar RS, (2011), A study on *Ajuga bracteosa* wall ex. Benth for analgesic activity. Int J. of Current Biological and Med Sci, 1, 12 – 14.
3. Anil Kumar, Kavita Gahlot, Jyotsna Dora and Pankaj Singh, (2011), Analgesic activity of methanolic extract of *Flemingia Strobilifera* (R.Br). Int J. of Research in Pharmacy and Chemistry, 1, 825-827.
4. Bhattacharya A, Agarwal D, Sahu PK, Kumar S, Mishra SS and Patnaik S, (2014), Analgesic effect of ethanolic leaf extract of Moringa oleifera on albino mice. Indian J Pain, 28, 89-94.
5. Biswa Nath Das and Muniruddin Ahmed, (2012), Analgesic activity of the fruit extract of *Averrhoa Carambola.* Int. J. Lifesc. Bt & Pharm. Res. 1(3): 1-7.
6. Hassan MM, Khan SA, Shaikat AH, Hossain ME, Hoque MA, Ullah MH and Islam S, (2013), Analgesic and anti-inflammatory effects of ethanol extracted leaves of selected medicinal plants in animal model, Vet World, 6, 68-71.
7. Ibironke GF and Ajiboye KI, (2007), Studies on the anti-inflammatory and analgesic properties of chenopodium ambrosiodies leaf extract in rats. Int J. of Pharmacology, 3, 111-115.
8. Jasem Estakhr, Asim Javdan, (2011), Study of the analgesic activity of methanolic extract of *Physalis alkekengi* in adult rats, Pharmacologyonline, 3, 768-772.
9. Jasem Estakhr and asim Javdan, (2011), Preliminary study of phytochemical screening and antibacterial activity of *Physalis alkekengi* against staphylococcus aureus. Pharmacologyonline, 3, 97-103
10. Kumbhar ST, ghaisas MM, Chavan DL, Pawar YD and Shaik AA, (2012), Analgesic activity of ethanolic extract of *Ficus glomerata* leaves: In vivo evaluation. Int J. of Research in Ayurveda & Pharmacy, 3, 706-709.
11. Mahdieh Hoshani, Manijeh Mianabadi, Mahnaze Aghdasi and Majid Azim-Mohseni, (2012), Inhibitory effect of *Physalis alkekengi* extracts in different phenological stages on xanthine oxidase activity. Physiology and Pharmacology, 16, 300-309.
12. Raquibul Hasan SM, Mariam Jamila, Muntasir Mamun Majumder, Raushanara Akter, Mokarram Hossain, Ehsanul Hoque Mazumder *et al*., (2009), Analgesic and antioxidant activity of the hydromethanolic extract of *Mikania scandens (*L.) willd. leaves. American J. of Pharmacology and Toxicology, 4, 1-7.

**13.** Sanchooli N, (2011), Antidiabetic Properties of *Physalis alkekengi* Extract in Alloxan-Induced Diabetic Rats. Research Journal of Pharmaceutical, Biological and Chemical Sciences, 2, 168-173.

**14.** Sattayasai J, Bantadkit J, Aromdee C, Lattmann E and Airarat W, (2012) Antipyretic, analgesic and antioxidative activities of *Aquilaria crassna* leaves extract in rhodents. J Ayurveda Integr Med, 3, 175-179.

**15.** Veronica Sanda Chedea, Adela Pintea, Andrea Bunea, Cornelia Braicu, Andreea Stanila and Carmen Socaciu, (2014), *Physalis alkekengi* carotenoidic extract inhibitor of soybean lipoxygenase-1 activity. BioMed Research Int. http://dx.doi.org/10.1155/2014/589168

**16.** Wan-Xuan Xu, Jian-Chao Chen, Jie-Qing Liu, Lin Zhou, Yi-Fen Wang, and Ming-Hua Qiu, (2013), Three new physalins from *Physalis alkekengi* var. *Franchetii. Nat. Prod. Bioprospect*, 3, 103–106.

**17.** Zulfiker AHM, Rahman MM, Hossain MK, Hamid K, Mazumder MEH & Rana MS, (2010), In vivo analgesic activity of ethanolic extracts of two medicinal plants – *Scoparia dulcis* L. and *Ficus racemosa* L. Biology and medicine, 2, 42-48.