Pharmacologyonline 3: 768-772 (2011) Newsletter *Estakhr* and *Javdan*

Study of the Analgesic Activity of Methanolic Extract of Physalis Alkekengi in Adult Rats

Jasem Estakhr*, Nasim Javdan

Science and Research Branch, Islamic Azad University, Fars, Iran.

***Corresponding Address:** Jasem Estakhr, j.estakhr@yahoo.com. Tel: +989179283966

Summary

In the present study, we evaluate the analgesic activity of methanolic extract of Physalis alkekengi, using chemical and thermal models which will induce acute pain in mice and rats. The methanolic extract was prepared by using maceration at room temperature (25°) over the period of 48 hours. The effect of extract of Physalis alkekengi was investigated for analgesic activity using acetic acid-induced abdominal writhings and Tail immersion method in rats. The analgesic activity of Physalis alkekengi extract at the dose of (100 and 200 mg/kg, body weight) showed significant ($p \leq 0.001$) in reducing abdominal writhings when compared with control and standard drug (Aspirin, 150 mg/kg, body weight). However this extract at the dose 150 mg/kg showed significant ($p \leq 0.001$) central analgesic action when compared with control and morphine (5 mg/kg, S.C.) as reference drug. In conclusion, the methanolic extract of Physalis alkekengi created significant analgesic activity in rats.

Keywords: Physalis alkekengi, Analgesic activity, Rat.

Introduction

Over the past two decades, there has been a tremendous increase in the use of herbal medicine; however, there is still a significant lack of research data in this field. Medicinal plants are now becoming more widely used by people all over the world. People understand the gentle strength of these natural remedies. Physalis alkekengi, belongs to the family Solanaceae. It is distributed in Asia (Iran, India, Japan and China) and Europe (Spain, Italy and Turkey) has a large history of herbal use, and an interesting chemistry but it is seldom used in modern practices (1).Chemical studies have demonstrated the presence of physalin, citric acid and vit C as the major components of P. alkekengi extract. The whole plant is claimed to possess medicinal properties. Physalin is the most chemical compound with various pharmacological characteristics including, anti bacterial, anti leishmanial and anti tumor and anti-spermatogenesis and anti-conception (2-7).The whole plant is anti phlogistic, anti pyretic, anti tussiveand expectorant (8-10). It is used in treatment of urinary and skin diseases (11). Its extract has been used for treatment of wide range of diseases, including kidney and bladder stone, febrile diseases, inflammation, general edema, and arthritis (12). The aim of the present study is to investigate the analgesic activity of *Physalis alkekengi* ethanolic extract using chemical and thermal acute pain models in rats.

Materials and Methods

Plant materials

Physalis alkekengi was collected from Guilan province, and then was identified by a botanist. Its leaves and fruits were dried under shade and powdered and were successively extracted with ethanol by maceration at room temperature (25°) over the period of 48 hours. 500 g of plant material and one liter of ethanol were used in the extraction. Methanol, containing the extract, was then filtered through Whatman paper and the solvent was vacuum distilled at 65c° in rotary evaporator. Final extract was a dark green semi-solid in percentage dray weight of 15%. This methanol extract was kept in deep freezer at -20 c° until use.

Animals:

wistar male rats (200-250g) were used in the pharmacological tests and were obtained from from Razi Institute, (Karaj, Iran). Animals were maintained under a 12:12-hour light/dark photoperiod, under controlled room temperature (23–25 ºC), humidity (60%). They were fed on standard rat diet and water ad libitum, allowed to acclimatize for one week before the experiments were started. The animals submitted to oral administration of the extracts or drugs were fasted for 18 h before the experiment (water was available).

Analgesic activity

The evaluation of the analgesic activity of the methanolic extract of Physalis alkekengi was carried out by using two different methods which used chemical stimuli (14) and thermal stimuli (13).

I. Acetic acid-induced writhing response in mice:

Koster et al. have described the method of this test (14). The total number of writhings following intraperitoneal administration of acetic acid solution (3% with 300 mg /kg, body weight) was recorded over a period of 20 min, starting 5 min after acetic acid injection, the mice were treated with extract of Physalis alkekengi (100 and 200 mg/kg, body weight) or standard drug (aspirin, 200 mg/kg, body weight), 30 min before administration of acetic acid. The number of writhings and stretching was recorded and permitted to express the percentage of protection using the following ratios (control mean-treated mean) x 100/control mean (15, 16).

II. Tail flick test:

The procedure is based on the observation that morphine like drugs are selectively prolonging the reaction time (in second) of the typical tail withdrawal reflex in rats induced by immersing the end of the tail about (4-5 cm) in warm water of 55 C°. Morphine (5 mg/kg s.c), was used as positive control and extract of Physalis alkekengi was administrated (100, 200 mg/kg, body weight). The tail withdrawal reflex was recorded before and after 15, 30, 60 and 120 min following oral administration of the extract to different groups of six animals each.

Statistical analysis:

The results were reported as mean ± S.E.M and analyzed by one-way ANOVA followed by student's t-test used for statistical evaluation. A value of $p< 0.05$ was considered significant.

Results

-Analgesic activity

In the acetic acid induced writhing test in mice, the extract of Physalis alkekengi (100, 200 mg/kg, body weight) demonstrated significant ($p \leq 0.001$) peripheral analgesic effect with an inhibition percentage of 38.15 % and 63.80 %, respectively, compared to Aspirin (49.11 %) as positive control (Table 1).

-Tail immersion assay

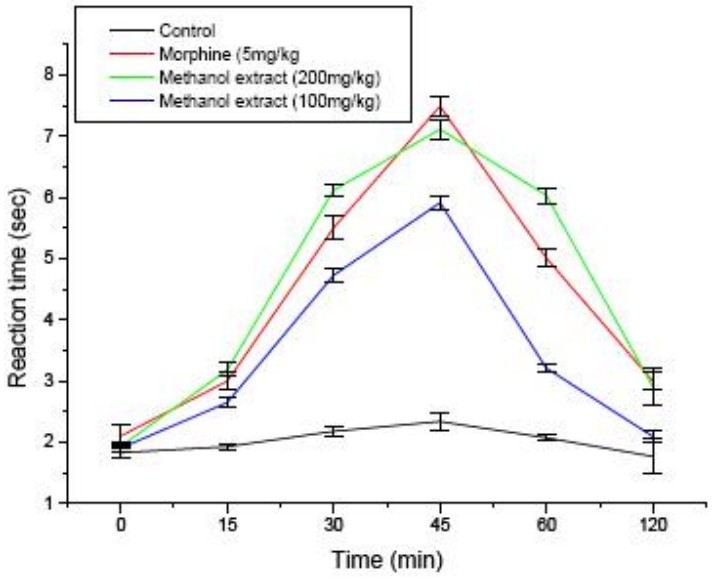
The anti-nociceptive activity of Physalis alkekengi extract on the tail immersion test is shown in (Figure 1). The dose of 200 mg/kg of the methanolic extract increased the reaction time (7.11 ± O.15 second) at 45min in the thermal stimulus significantly ($p \leq O.001$). Which comparable with morphine (5 mg/kg s.c), the reaction time was (7.49 ± 0.25 second) ($p \leq 0.001$).

Table 1: The effect of Physalis alkekengi methanolic extract on acetic acid induced writhes in mice.

Groups	Dose (mg/kg b. w.)	Number of writhes (per 20 min)	Inhibition of writhing %
Control	-	48.5 ± 2.71*	-
Aspirin	200	23.43 ± 3.52*	49.11
P. alkekengi extarct	100	29.36 ± 2.07*	38.15
P. alkekengi extarct	200	17.26 ± 2.41*	63.80

Data presented as mean ± standard error mean (M ± S.E.M.) $p \leq 0.001$ evaluated by one-way ANOVA against control group. N=6: number of mice for each test.

Figure 1: Evaluation of Analgesic activity of Physalis alkekengi methanolic extract by tail flick method. Data presented as mean ± standard error mean (M ± S.E.M.) $p \leq 0.001$ evaluated by one-way ANOVA against control group. N=6: number of mice for each test.



Discussion

The use of herbal medicine has become increasingly popular worldwide and has proved to be a rich source of new active compounds, especially to treat pain and inflammation processes. Pain is one of the classical signs of the inflammatory process in which sensitization of the nociceptors is the common denominator. This sensitization causes hyperalgesia or allodynia in humans, phenomena that involve pain perception, and is better described as nociception in animal models (17, 18). In general, acetic acid writhing tests are used to evaluate the compounds for peripheral anti-nociception activities (19). Acetic acid injection produces peritoneal inflammation which triggers response characterized by writhing. We analyzed the effect of different doses of methanolic extract of Physalis alkekengi for its analgesic activity. We observed that, animals treated with Physalis alkekengi methanolic extract (100 and 200 mg/kg), reduced significantly the number of abdominal writhing during 20 min in the peripheral analgesic ($p< 0.001$), while in the central analgesic at the dose 100 mg/kg, this extract don't exhibit significant analgesic activity when compared with control and morphine treated animal. By increasing the dose to 200 mg/kg, the methanolic extract of Physalis alkekengi produces significant ($p< 0.001$) central analgesic action, because this extract increase the reflex time of removal the tail of rats by inhibition the pain(7.11sec ± 0.15 at 45 min), these results were compared with morphine (7.49sec ± 0.25), at the same time. Related studies have demonstrated that acetic acid indirectly induces the release of endogenous mediators of pain (such as prostaglandin, kinin, histamine, etc) that stimulate the nociceptive neurons, which are sensitive to non-steroidal anti-inflammatory drugs and opioids (20-22). These observations suggest that, the methanolic extract has a significant inhibitory activity in inflammation pain, and this activity may be related with the suppression of synthesis and/or release of endogenous proinflammatory substances.

Conclusion

From the results it can be concluded that the methanolic extract of Physalsis alkekengi is a notable, central, and peripheral analgesic activity.

References

1- Unny R, Chauhan AK, Jashi YC. A review on potentiality of medicinal plants as the source of new contraceptive principles. Phytomedicine 2003; 10: 233-260.
2- Bown D. Encyclopedia of herbs and their uses. London: Dorling Kindersley; 1995.
3- Chaudhary M, Iqbal DH. Antileishmanial physalins from Physalia minima. Chemistry and Biodiversity 2005; 2(9): 1164-1173.
4- Janurio AH, Rodrigues Filho E, Pietro RC, Kashima S, Sato D, Franca S. Antimycobacterial physalins from Physalis angulata L. (Solonaceae). Phytotherapy Research 2002; 16(5): 445-448.
5- Masao K, Toichi O, Masaki N. Structure of physalin M isolated from Physalis alkekengi Var. fracheti. BCSJ 1988; 61(7): 2696-2698.
6- Launert E. Edible and medicinal plants. Hamlyn: 1981.
7- Amini A. Dictionary of therapeutic plant. Tehran: Tehran University; 2004: 157-159.
8- Zargari A. Medical plants. Tehran: Tehran University; 2003: 3595-3596.
9- Chopra RN, Nayar SL, Chopra IC. Glossary of Indian medicinal plants(including the supplement). New Delhi: CSIR; 1986.
10- Mirheydar H. Herbal information usage of plants in preventaion and treatment of diseases. Tehran: Tehran University; 2003. 39-42.

11- Vessal M,, Mehrani HA, Omrani GH. Effect of an aqueous extract of Physalis alkekengi fruit on estrus cycle, reproduction and uterine creatine kinase BB isozyme in rats. J of Ethnopharmacology 1991; 34(3): 69-78.
12- Montaserti A, Pourheydar M, Khazaei M, Ghorbani R. Antifertility effect of Physalis alkekengi alcoholic extract in female rat. IJRM 2007; 5(1): 13-16.
13- Hasard R, Cheymol J, Analgesique centraux, Manuel de Pharmacologie, 2eme edition, Masson et Cie 1970; 124-146.
14- Koster, R., Anderson, M., De-Beer, E.J, Acetic acid analgesic screen. Federation proceedings 1959; 18:418-420.
15- Dongmo, A.B., Nguelefack, T.B., Lacaille- Dubois, M.A, anti-nociceptive antiinflammatory activities of *Acacia pennata wild (Mimosaceae).* Journal of Ethnopharmacology 2005; 98: 201-206.
16- Edwige Laure Nguemfo, Theophile Dimo, Anatole Guy Blaise Azebaze, Emmanuel Acha Asongalem ,Katim Alaoui , Alain Bertrand Dongmo , Yahia Cherrah ,Pierre Kamtchouing, Anti-inflammatory and Antinociceptive activities of the stem bark extracts from *Allanblackia Staner L.C. (Guttiferae).* journal of ethino pharmacology 2007; 114:417-424.
17- Aline Cavalcanti de Queiroz,Daysianne Pereira de Lira,Thays de Lima Matos freire Dias et al. The antinociceptiveand anti-inflammatory activities of *Piptadenia stipulaceaBenth.(Fabaceae)*. Journal of Ethnopharmacology 2010; 128 :377-383.
18- Verri-JYnior,W.A., Cunha,T.M., Parada,C.A., Pool,S.,Cunha,F.Q., Ferreira,S.H, Hypernociceptive role of cytokines and chemokines:targets for analgesic drugs development. Pharmacology and therapeutics 2006; 112:116-138.
19- Le Bars, D., Gozariu, M., Cadden, S., Animal models of nociception, pharmacological Reviews 2001; 53:628-651.
20- Sanchez Mateo,C.C., Bonkanka,C.X., Hernandez Perez, M.,Rabanal,R.M., Evaluation of the analgesic and topical anti-inflammatory effects of *Hypericum reflexum L.fil.* Journal of Ethnopharmacology 2006; 107:1- 6.
21- Sulaiman,M.R**.,** Zakaria,Z.A., Bujarimin,A.S., Somchit,M.N., Israf,D.A., Moin,S., Evaluation of *Moringa oleifera* aqueous extracts for antinociceptive and anti-inflammatory activities in animal models. Pharmaceutical biology 2008; 46:838-845.
22- Belsem Marzouk, Zohra Marzouk, Ehsen Haloui, Nadia Fenina, Abderrahman Bouraoui, Mahjoub, Aouni, Screening of analgesic and anti-inflammatory activities of *Citrullus colocynthis* from Southern Tunisia. Journal of Ethnopharmacology 2010; 128: 15-19.