**Antitussive effect of *Portulaca oleracea* L. in guinea pigs**

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**Abstract**

Several therapeutic effects including diuretic, anti-pyretic and anti-scurvy have been reported for *Portulaca oleracea*. In previous studies the analgesic, anti-inflammatory, anti-hypertensive, anti-oxidant, muscle relaxant effects and neuropharmacological effects of the aqueous extract of *Portulaca oleracea* have been demonstrated on experimental animals. In the present study the antitussive effect of this plant was evaluated. The antitussive effects of aerosols of two different concentrations of boiled extract, codeine, and saline were tested by counting the number of coughs induced by citric acid aerosol 10 min following exposure of animals. The effective concentration of extract causing 50% reduction of cough number (EC50) was also determined. The results showed significant reduction (p<0.001) in cough numbers following the use of both concentrations of the boiled extract (10.7 0.16 and 6.7 0.33 for %2.5 and %5 extracts respectively) compared to saline (14.8 0.30). In addition there was a significant difference (p<0.01) between the cough numbers of the 5% extract with that of codeine (10.2 0.38). In this study, EC50 of the plant extract was 4.5%. These results showed that the antitussive effect of *Portulaca oleracea* L. was comparable to codeine.

**Keywords:** *Portulaca oleracea*; antitussive effect; medicinal plants; guinea pig; codeine.

**Introduction**

*Portulaca oleracea* is a grassy plant with small-yellow flowers and height of 10-30 cm, which grows in different areas of the world including north and northwest of Iran. It contains water (92-95%), mucilage, pectin and lipids (0.3-0.4%). Several therapeutic effects including anti-pyretic, anti-scurvy, anthelmentic, pain relief in migraine headaches and gastrointestinal tract and anti-inflammatory have been described for *Portulaca oleracea* in Iranian folk medicine (1, 2). There is evidence for relaxant effect of the aqueous extract of this plant on skeletal and smooth muscles (3-7), analgesic and anti-inflammatory effects (8), antioxidant effect (9), and neuropharmacological activity (10). Our laboratory findings also showed a relaxant effect of the boiled aqueous extract of this plant in airways of asthmatic patients and also on isolated guinea pig tracheal chains. In the present study the antitussive effect of the boiled extract (2.5 and 5%, w/v) of *Portulaca oleracea* was evaluated.

**Experimental**

**Plant and extracts**

The whole plant was collected from the farm of school of Pharmacy, Mashhad University of Medical Sciences, identified by Mr. Ahi, dried and powdered. The extracts were prepared as follows: ten g of the dried plant powder was added to 100 ml boiled water for 15 min and then filtered. The solvent was then removed under reduced pressure until the extracts volume reached to 10 ml. The concentration in the final extracts was 10% w/v.
Protocol
The Dunkin-Hartley guinea pigs (Razi Institute, Mashhad, Iran) of both sexes (body weight 500-600g) were used in this study. The method used has been described previously (11). Unanaesthetized unrestrained animals were placed individually in a transparent Perspex chamber, dimensions 30×20×20 cm and exposed to a nebulized aqueous solution of 0.1 g/ml citric acid for 7 min. The aerosol was produced by an air flow of 8 L/min through a Wright nebulizer (producing an aerosol with particles with an aerodynamic mass median diameter of 0.9 mm) nebulizing 0.65±0.04 ml solution per minute. During the exposure, a trained observer continuously watched the animals, and the number of coughs produced during the last 5 min of exposure was counted. Coughs could easily be distinguished from sneeze, since there is a clear difference in sound as well as in behaviour of the animal (11).

The above method was performed 10 min after exposing animals to 4 different aerosols of following solutions (n=6 for each solution):

i) Normal saline (baseline measurements)
ii) Boiled extract (2.5% w/v)
iii) Boiled extract (5% w/v)
iv) Codeine solution (0.03 g/ml, positive control)

All of the experiments were performed randomly with 2 h resting period between each experiment. The effective concentration of extract causing 50% reduction of cough number (EC50) was also determined.

Statistical analysis
All data were expressed as mean±SEM. Comparison of baseline data with number of cough obtained in the presence of plant extracts and codeine were made using ANOVA. Comparison of data obtained in the presence of two different concentrations of boiled extracts was made using paired "t" test. Significance was accepted at p<0.05.

Results and Discussion

Both concentrations of boiled extract and codeine caused significant reduction in number of citric acid-induced coughs compared to saline (p<0.001). However, the antitussive effect of 5% concentration of boiled extract was significantly different with that of codeine (p<0.01), but 2.5% extract showed no significant difference from codeine. There was also significant difference between antitussive effects of both boiled extracts (p<0.001) (Fig. 1). EC50 of the plant extract was 4.5%.

In the present study the antitussive effect of boiled extract of Portulaca oleracea was evaluated using a standard method applied previously by several investigators (11, 12). Data showed relatively a potent antitussive effect for boiled extract of the plant that was even greater than the effect of codeine. Codeine at concentration of 3% reduced number of coughs from 14.8 to 10.2 and caused 31.1% protection, while the 2.5% and 5% solutions of plant extract reduced number of coughs to 10.7 and 6.7 and caused 27.7% and 54.7% protection, respectively. The EC50 for plant extract was 4.5%. It means that effect of 3% codeine solution is almost equivalent to 2.5% extract. Opioids such as morphine and codeine are generally considered to be the most potent and effective antitussive drugs available and are believed to inhibit cough through suppression of cough centre in the CNS (13, 14). Although the antitussive effect of plant extracts was similar to that of codeine, the mechanism(s) of antitussive effect of this plant cannot be concluded from the results of the present study.

Cough may be caused by several mechanisms including; mechanical stimulation of normal afferent system, chemicals,
inflammatory mediators, and neurotransmitter stimulation of normal afferent system (15). Although the modulating effects of inflammatory mediators and neurotransmitters on the cough reflex are likely to be very important but it needs further investigations (16-18). Results of studies to date attest the complexity of this issue. For example, in human, prostaglandin F2 potentiates cough responses to both inhalation of low chloride solution and capsaicin, but prostaglandin E2 potentiates cough resulting from capsaicin only (18). In another study, it has been shown that iv injection of serotonin, decreases cough in response to inhaled low chloride solution but not capsaicin (17). In another study, Richard et al., (2001) have tabulated chemicals that could induce cough. Some of these agents are acetic acid, capsaicin, acetylcholine, citric acid, histamine, HCl, Tear gas, phosphoric acid, sulfuric acid, bradykinin, ether, lobeline, ammonia vapor and tobacco smoke (19).

Morphine was recently shown to reduce a vagally mediated bronchoconstriction produced by inhaled distilled water in asthmatics (20) and in healthy human subjects. The mechanism behind this inhibitory effect is unknown, but suppression of neurotransmitter release has been suggested (21). Some experimental data indicated that opioids might interact with the peripheral nervous system of the tracheobronchial tree. A partial antagonism of a noncholinergetic neurogenic bronchoconstriction in the guinea pig by opioid agonists has been reported (22-24). Kerlsson et al., (1990) also showed that nebulized codeine and morphine could inhibit bronchoconstriction and cough induced by citric acid using a method similar to that of the present study (12). Therefore, the similar antitussive effect of extracts from *Portulaca oleracea* and codeine may indicate that this plant may have bronchodilator activity that has been shown in our previous studies both in human (25) and experimental animals (26).

In conclusion the results of this study indicated that the antitussive effect of *Portulaca oleracea* was comparable to that of codeine. The probable antitussive mechanism(s) of this plant, may be due to its relaxant effect, and/or suppression of neurotransmitter release or may be due to its relaxant effect, but the exact mechanism(s), should be clarified in future studies. Finally, since the 2.5% extract showed equivalent effect with 3% codeine solution and bascd of our previous data on the airways of asthmatic patients (25) and its fantastic relaxant effect on tracheal chain in guinea pig (26), it is suggested that the aqueous extract of this plant is a good candidate to be used in clinic.

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