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# EVALUATION OF ESSENTIAL OIL OF *Myrtus communis* LEAVES FOR ANALGESIC AND GISTROINTESTINAL MOTILITY PROFILE

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## Summary

In the present research work the essential oil of *Myrtus communis* Leaves were tested for their analgesic and gastrointestinal motility. The analgesic effect was evaluated in mice, using acetic acid induced writhing test and effect on GIT motility was also tested in mice using charcoal as marker in intestine. The essential oils showed dose dependent analgesic effect in compression with standard drug and significantly ($P<0.05$) inhibited the writhing at 100 and 150 mg/kg, while the analgesic effect was non-significant at the dose of 50 mg/kg. In case of GIT motility the oils were good laxative at low dose (50 mg/kg) while with increasing the dose the motility was not significant. It can be concluded that the oils should be use for constipation in low dose while for analgesic effect its high dose is required.

Key words: *Myrtus communis, Analgesic and Gastrointestinal motility*

## Introduction

*Myrtus communis* L. (myrtle) is a common and widespread annual shrub, and the sole representative of the family, Myrtaceae in the Mediterranean Basin, that has been used since ancient times for medicinal, food and spices purposes. The leaves contain tannins, flavonoids such as quercetin, catechin and myricetin derivatives and volatile oils (Baytop, 1999). In the essential oil of *M. communis* species, eucalyptol was the predominant component (50.13%). The other important components were linalool (12.65%), -terpineol (7.57%) and limonene (4.26%). The major components were -pinene, limonene and 1,8-cineole of the essential oil of leaves of *M. communis* (Flamini *et al.*, 2004) . The decoction of leaves and fruits was useful for sore washing. The decoction of the leaves is still used for enemas and against respiratory diseases (Maccioni *et al.*, 2007) . The essential oil obtained from the leaves by steam distillation is also important in perfumery (Baytop, 1999). It is known to possess antioxidant, antibacterial, and antiseptic agent (Zanetti *et al.*, 2010). This study on the essential oil of *Myrtus communis for* analgesic and anti-constipating actions is the part of our research work on Pakistani medicinal plants (Muhammad *et al.*, 2012, Barkatullah *et al.*, 2011, Muhammad and Saeed, 2011, Rahman *et al.*, 2011, Raziq *et al.*, 2011, Rahman *et al.*, 2011, Ismail M *et al.*, 2011, Muhammad N *et al.*, 2012).

## Material And Methods

### Plant material

The fresh leaves of *Myrtus communis* were collected from Peshawar, Pakistan in May 2011. After the botanical authentication, the plant material were washed under running tap water to remove adhering dust, The plant was air dried under shade and crushed into small pieces. The resulting material was subjected to distillation (Muhammad and Saeed, 2011).

### Extraction of Oil

The isolation of essential oils from *Myrtus communis* leaves is usually obtained by hydro distillation method with a Clevenger-type apparatus, according to the Italian Official Pharmacopoeia. These essential oils were screened for analgesic and anti-constipating effect.

### Chemicals

Diclofenac sodium, castor oil, activated charcoal, Acetic acid, sterile normal saline was used in all experiments as control while extracts was prepared in normal saline.

### Animals

Balb-C mice of either sex were used in all experiments. Animals were purchased from the Pharmacology Section of the Department of Pharmacy, University of Peshawar. The animals were maintained in standard laboratory conditions (25 °C and light/dark cycles i.e. 12/12 h and were fed with standard food and water.

### Analgesic activity

All animal were fasted for 2 h before starting experiment. Animals were divided in five groups. Group I was injected with normal saline (10 ml/kg i.p) as control while group II was treated with standard drug diclofenac sodium (10 mg/ kg i.p) and the remaining three groups were treated with essential oils at the doses of 50, 100 and 150 mg/kg i.p. After 30 min of saline, diclofenac sodium and plant oils injection, the animals were treated i.p. with 1% acetic acid. The writhing was counted after 5 min of acetic acid injection and the number of abdominal constrictions (writhing) was counted for 10 min (Butterweck *et al.*, 2004). The percent analgesia was calculated using the following formula

Percent analgesia = 100- no, of writhing in tested animals/ no, of writhing in control animals×100

### GIT Motility test

For this purpose the animals were fasted 18 – 24 hr before the start of experiment. Animals were divided in five groups each of six animals. First group were given normal saline i.p, groups II was treated with castor oil as stander drug, remaining three groups were treated with plant essential oil (50, 100 and 200 mg/kg i.p), after 30 min of injecting saline, castor oil and essential oils 0.5 ml a 10 % charcoal suspension in 5 % gum acacia was administer P.O. After 15 min of administering charcoal the animal was kill by cervical dislocation and dissected out. The dissected animal was place on clean surface and measures the distance cover by charcoal (Marona and Lucchesi, 2004). The percent motility was calculated using the following formula

Percent Motility = 100- Distance covered / total length of intestine ×100

### Statistical Analysis

The results were articulated as mean ± SEM of six animals. For statistical analysis, ANOVA was followed by post hoc Dunnetts test for multiple comparisons. Effects were considered to be significant at the $P < 0.05$ level.

## Results and Discussion

### Analgesic effect

The analgesic effects of essential oil were tested in mice of either sex at the dose of 50, 100 and 150 mg/kg body weight. The numbers of writhing were significantly decreases by the essential oils of our selected plant. The analgesic effect of our tested essential oils (150 mg/kg) and that of positive control drug was similar as shown in table 1. The dose dependent analgesia was noticed, the percent analgesia was 43.54, 70.20 and 72.67 for 50, 100, 150 and diclofenac sodium respectively as shown in figure 1. The essential oil increased the intestinal motility very interesting. The motility was increased event more than the standard drug (castor oil) as shown in table 2. The highest activity was observed at 50 mg/kg with increasing dose the motility was decreased. The percent increased in GIT motility is presented in figure 2 .The analgesic effects of the crude extract of the leaves of *Myrtus communis* were investigated in acetic acid induced writhing pain model. This model was used for the analgesic affect, because of its sensitivity that could give different grades of injurious stimuli in chemically induced tissue damage (Victor *et al.*, 2009). Similarly, the acetic acid induced writhing has been used to evaluate analgesic effects of drugs and the response is thought to be mediated by peritoneal mast cells, acid sensing ion channels and the prostaglandin pathways (Ranjit *et al.*, 2006). The essential oils of *Myrtus communis* has a significant inhibition in number of the writhing in each mouse (Nuhu *et al.*, 2010 ). The intraperitoneal injection of acetic acid produces an abdominal writhing response due to sensitization of chemo-sensitive nociceptors by prostaglandins. Increase level of protanoids as well as lipoxygenase products have been found in the peritoneal fluid after the injection of the acetic acid. The analgesic effect of any plant extract may therefore be due to either its action on visceral receptors sensitive to acetic acid, to the inhibition of the production of algogenic substances or the inhibition at the central level of the transmission of painful message (Ranjit *et al.*, 2006).

| Treatment | Dose | No, of writhing(mean) |
|---|---|---|
| Normal saline | 10ml/kg | 65±3.46 |
| Diclofenac sodium | 10mg/kg | 17.6**±1.14 |
| Essential oil | 50 mg/kg | 35.00*±2.00 |
| | 100 mg/kg | 19.37**±1.53 |
| | 150 mg/kg | 17.70**±2.7 |

Table 1. Analgesic activities of essential oil from the leaves Myrtus communis

Values (mean ± SEM) present the writing after treatment with diclofanec sodium (10mg/kg), saline (10 ml/kg) and oils (50, 100 and 150 mg/kg). The data was analyzed by ANOVA followed by Dunnett's test. Asterisks indicated statistically significant values from control. *P < 0.05, **P<0.01.

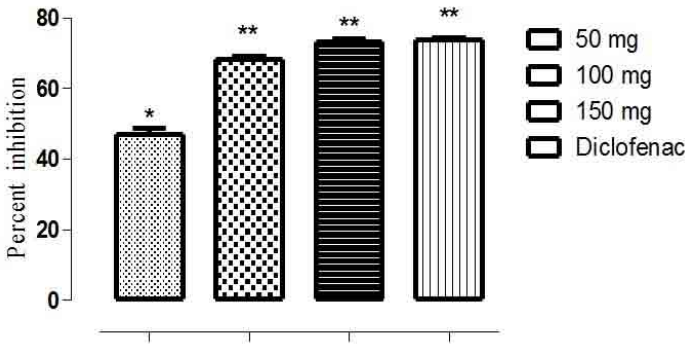


Figure 1. Analgesic effect essential oil of Myrtus communis in mice. Bars present the percent inhibition of writing after treatment with diclofenac sodium (10mg/kg), normal saline (10 ml/kg) and oils (50, 100 and 150 mg/kg). The data was analyzed by ANOVA followed by Dunnett's test. Asterisks indicated statistically significant values from control. *P < 0.05, **P<0.01.

| Treatment | Dose | Total length of intestine | Distance cover by charcoal |
|---|---|---|---|
| Control | 10ml/kg | 59.33±0.07 | 15.17±0.27 |
| Castor oils | 10ml/kg | 55.75±0.10 | 33.25**±0.17 |
| Essential oils | 50 mg/kg | 45.57±0.58 | 10.67**±0.58 |
| | 100mg/kg | 47±2.65 | 18.00**±1.00 |
| | 150 mg/kg | 47±2.65 | 28.00*±1.00 |

Table 2. GIT mortality activities of essential oil from the leaves Myrtus communis

Values (mean ± SEM) present the movement of charcoal in GIT after treatment with castor oils (10ml/kg), normal saline (10ml/kg) and oils (50, 100 and 150 mg/kg). The data was analyzed by ANOVA followed by Dunnett's test. Asterisks indicated statistically significant values from control. *P < 0.05, **P<0.01

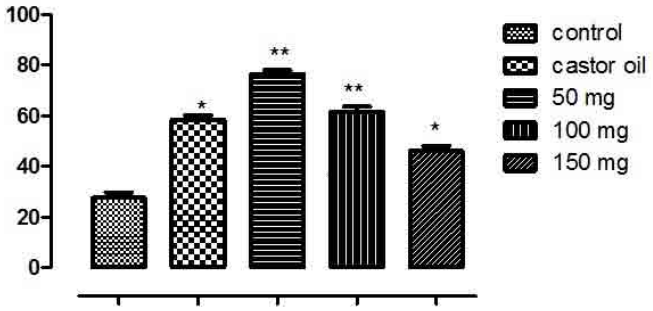


Figure 2. GIT motility effect of essential oil from Myrtus communis in mice. Bar presents the percent movement of charcoal in GIT after treatment with castor oils (10ml/kg) and oils (50, 100 and 150 mg/kg). The data was analyzed by ANOVA followed by Dunnett's test. Asterisks indicated statistically significant values from control. *P < 0.05, **P<0.01.

## GIT motility

Constipation is one of GIT disorder and its major complication is pills/hemorrhoid, which is chronic disorder and need surgery. Although there is number of constipation reliving compounds in market but having a lot of side effects, therefore the search for natural anti-constipating agents is carried out in various research institutes. In this research work *Myrtus communis* is tested for their anti-constipating effects on mice, using activated charcoal as marker. This method is simple and non expensive for testing the chemicals or plant extracts for GIT motility study. It is shown that the traveling of charcoal in intestine was mostly upto 20 cm while the movement of charcoal was significant with essential oil of *Myrtus communis.* It is still vey interesting to say that these essential oils are anti-constipating at low dose and good analgesic at high dose. So if these oils are used for the analgesic purpose there will be no chance of loss motion as these oils are not laxative at high dose.

## Conclusion

In conclusion these essential oils can be used as analgesic and laxative in traditional medicine.

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