In vitro effects of essential oils of *Tanacetum balsamita* and carvone on the contractility of bovine ileum smooth muscles

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**Abstract**

*Tanacetum balsamita* (*Asteraceae*) is a medicinal plant previously used in human medicine to solve gastrointestinal problems such as abdominal pain. Anti-inflammatory, analgesic, immune-modulatory, and antioxidant effects of *T. balsamita* have been proven in previous studies. The present study investigated the effects of *T. balsamita* essential oil (TBOE) on ruminant smooth muscle contractions. The experiment was performed on the circular smooth muscle of ileum samples taken from slaughtered bulls in the organ bath. Nine cumulative concentrations of TBOE from 0.10 to 1000 µg mL−1 were added to tissue samples. The solution used was Tyrode’s solution aerated with a mixture of 95.00% oxygen and 5.00% carbon dioxide, and the temperature was set at 37.00 °C. The effect of TBOE on baseline contractions and three induced contractions with potassium chloride, barium chloride, and carbachol was investigated. In GC-MS analysis of TBOE, carvone was identified as a major ingredient. The effects of eight concentrations of 0.001 to 10.00 µM of carvone on all contractions were investigated under similar conditions. The effects of TBOE, carvone, and verapamil (standard calcium channel blocker) on calcium channels were assessed. The results revealed that TBOE and carvone significantly inhibit spontaneous contractions as well as all spasmogen-induced contractions. The TBOE and carvone exert their myorelaxant properties by inhibiting Ca2+ channels in smooth muscle. The anti-spasmodic properties of *T. balsamita* can be employed for the treatment of intestinal spasms or hypermotility.

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**Introduction**

Gastrointestinal dysmotility is an important part of alimentary problems in ruminants. Some stimulants may increase the intestinal motor activity and cause cramps, abdominal pain, and colic through causing problems such as diarrhea syndrome. Stressful situations can cause excessive motility in the gastrointestinal tract, and the hypermotility affects intestinal permeability and nutrient absorption. Moreover, constipation, intestinal obstruction, terminal ileitis, and intussusception are associated with intestinal movement disorders in ruminants. Spasm of intestinal smooth muscle in cattle is one of the reasons for abdominal pain; thus, anti-spasmodics can help relieve abdominal pain symptoms.

Medicinal plant products are economically viable in livestock that can be used as adjunctive or alternative therapies. The plants used in ethnoveterinary medicine vary in different region, and different parts of plants such as roots, leaves, flowers, and seeds are used in powder, ointment, or decoction. Studies showed the beneficial effects of essential oils and extracts of medical plants on digestive problems of ruminants such as diarrhea, gastrointestinal inflammation, abdominal pain, colic, impaired digestion, and tympany. Some medical plants may stimulate salivation, restore rumination, promote growth, and improve liver function.

*Tanacetum balsamita* (costmary) is an aromatic plant used in medicine from ancient times. *Tanacetum* genus, belongs to the *Asteraceae* family, is called “Shahesparam” in the local language. *T. balsamita* is one of the 26 species of Tanacetum genus in Iran widely grown in East and West Azarbaijan province in Iran. This plant has been used in traditional Iranian medicine for years as a pain

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reliever, tranquilizer, and heart tonic.³° Tanacetum species are used as stomach strengthen, food preservatives, food additives, cosmetics, insecticides, balsams and dyes.⁹,¹⁰ The T. balsamita, as one of the medicinal plants in human and animal medicine, has been considered by researchers worldwide. Interesting reports have been recorded about the medicinal effects of T. balsamita extracts and essential oils. Their antibacterial, antioxidant, anti-inflammatory, immune-modulatory, and analgesic effects have been confirmed.⁷,⁷¹⁻¹⁵ The T. balsamita, in the formulation of massage lotion, has shown analgesic, anti-congestion, anti-irritant, muscle relaxant properties, and blood circulation booster.¹⁶ Previous in vitro studies in organ bath have revealed the spasmyloytic properties of T. parthenium and T. vulgare.¹⁷⁻¹⁹

Carvone belongs to Terpenoids and is a main component of T. balsamita, Mentha longifolia, and Mentha spicata.⁴,²⁰,²¹ Carvone has potent antioxidant, anti-inflammatory, and anti-spasmodic properties.²²,²³ A previous study showed that carvone had an anti-spasmodic effect on the guinea pig ileum.²³

Due to the history of successful use of T. balsamita in the treatment of human gastrointestinal problems⁶,⁹,¹⁰ and the various effects proven for this plant,⁷,¹¹⁻¹⁶ this plant seems to have good potential for the treatment and prevention of gastrointestinal problems. However, the effects of plants on gastrointestinal motility were unknown. Dysmotility is an important part of the ruminant GI disease’s pathophysiology, and natural remedies of these problems is important in the production of organic food for human. This experiment was aimed at evaluating the in vitro effects of T. balsamita and its major compounds on contraction of small intestine in ruminant.

Materials and Methods

**Plant material.** The fresh T. balsamita plant was collected under the supervision of a botanist from Urmia, Iran, in July 2020. The plant was authenticated at the Department of Medicinal and Aromatic Plants of Urmia University. Hydro-distillation was performed to extract the T. balsamita essential oil (TBEO) from its aerial parts. TBEO was stored in a refrigerator at 4.00 °C until being used.

**Gas chromatography-mass spectrometry (GC-MS).** It was performed to identify the components of the plant’s essential oil. For chemical analysis, an Agilent 7890 (Agilent, Santa Clara, USA) A gas chromatograph coupled to a 5975 A mass spectrometer (Agilent) using a HP-5 MS capillary column (5.00% Phenyl Methylpolysiloxane, 30.00 m, 0.25 mm inner diameter, 0.25 μm film thickness) was used. The oven temperature was programmed as follows: 3 min at 80.00 °C, subsequently increased 8.00 °C per min to 180 °C, held for 10 min at 180 °C. Helium was used as carrier gas at a flow rate of 1.00 mL per min, and electron impact (EI) was 70.00 eV. The split mode injector was set in a split ratio of 1:500, and mass range acquisition was from 40.00 to 500 m/z. The Wiley 2007 (https://www.wiley.com/en-us) and NIST 2005 (https://www.nist.gov) libraries were used to identify the TBEO compounds. The GC-MS data processing was performed using Chemstation Software (Agilent).

**Chemicals.** Standard carvone, acetylcholine chloride (Ach) verapamil hydrochloride, and carbachol chloride (Cch) were obtained from Sigma Chemicals Co. (St. Louis, USA). Calcium chloride (CaCl₂), potassium chloride (KCl), sodium chloride (NaCl), glucose, sodium dihydrogen phosphate (NaH₂PO₄), magnesium chloride (MgCl₂), sodium bicarbonate (NaHCO₃), barium chloride (BaCl₂), ethylene-diaminetetraacetic acid (EDTA) and dimethyl sulfoxide (DMSO) were purchased from Merck (Darmstadt, Germany).

**Tissue samples collection.** Ileum tissue samples were taken from slaughtered bulls between 2 and 4 years old from Urmia Industrial Slaughterhouse. Bulls were evaluated before sampling, and those with a history or symptoms of gastrointestinal problems were excluded from the sampling process. The ileum was excised from the gastrointestinal tract less than 20 min after slaughter. A complete 15.00 cm piece of ileum was cut, and a longitudinal incision was then made to open the inner (mucosal) surface of the ileum. Tyrode’s solution containing NaCl (136.90 mM), NaHCO₃ (11.90 mM), glucose (5.60 mM), KCl (2.70 mM), CaCl₂ (1.80 mM), MgCl₂ (1.10 mM) and NaH₂PO₄ (0.40 mM) was used as the rinse, transportation and incubation medium. The mucosal and serous surface of the tissue samples were immediately rinsed with cooled (4.00 °C) and aerated Tyrode’s solution to remove the digestive contents.²⁴ Specimens were immersed in 4.00 °C Tyrode’s solution and kept at this temperature until reaching the laboratory. The solution was exchanged 10 min after the initial collection to remove digestive contents and supply the vital material to the tissue. In the laboratory, whole pieces of ileum tissue were placed on the dissected board filled with Tyrode’s solution. The non-smooth muscle (mucosa and submucosa) was carefully separated from the ileum smooth muscle. The tissue strips (5.00 × 20.00 mm) were cut in the direction of the circular muscle layer.²⁴

**Recording of smooth muscle activity.** To mount the samples in chambers, one end of each strip was joined to the bottom hook of the chamber, and the other end was connected to an isometric transducer with suture thread (TRI 202P; PanLab, Barcelona, Spain). Six transducers connected to an amplifier (ML224; AD Instruments, Castle Hill, Australia) and Power Lab data acquisition system (ML870; AD Instruments) were used for data collecting. Lab chart software (version 8.0; AD Instruments) was employed to view and record data. The organ bath chambers filled with 25.00-mL Tyrode’s solution was set at 37.00 °C and gassed continuously with a mixture of 95.00% O₂ and 5.00% CO₂.²⁴


Design of experiments. First, the ileum muscle samples were rested in the Tyrode’s medium for 1 hr to adapt to the new environment. In this step, the solution was changed every 15 min, and two 1.20 g tensions were applied to the tissues at 15-min intervals. To evaluate the viability and contractile function, the samples were tested before the main experiment through adding 10.00 µM of acetylcholine. After the acetylcholine, tissues were washed to achieve basal contraction of muscles. This procedure was repeated three times and if the results were the same, the tissue was considered acceptable for testing.24 The effects of Tanacetum balsamita essential oils (TBEO) on the contractions of bovine ileal circular smooth muscle were investigated in four groups. The experiment groups were designed to observe the effects of TBEO on spontaneous, Cch, KCl, and BaCl2 induced contractions on bovine ilium muscles.23 In the first group, the cumulative concentrations of TBEOs were added to the chambers when the muscle tissue reached a stable spontaneous contraction. In the other groups, after the stable induced contraction of Cch (1.00 µM), KCl (20.00 and 60.00 mM), and BaCl2 (3.00 mM), TBEO concentrations added to the organ bath and its cumulative effects on contractions were recorded. TBEO dissolved in DMSO 5.00% and dilutions of 0.10, 0.30, 1.00, 3.00, 10.00, 30.00, 100, 300, and 1000 µg mL−1 were prepared by adding the Tyrode solution. All concentrations of 0.10 to 1000 µg mL−1 TBEO in separate groups were added to the medium at 2-minute intervals. Concentrations of 0.001, 0.003, 0.01, 0.03, 0.10, 0.30, 1.00, 3.00, and 10.00 µg of carvone were prepared and tested in the same TBEO method. At the end of the test, the tissues were washed with thyroid solution. The viability of muscles was confirmed by their response to the addition of 10.00 µM acetylcholine.23

Mechanism of action. Effect of TBEO and carvone on calcium channels was investigated in bovine ilium smooth muscle. Tissue strip samples were separately treated with concentrations of 30.00 and 100 µg mL−1 TBEO and 0.10 and 0.30 µM carvone, and the concentration-response curves (CRCs) were then obtained by adding calcium to the Ca++ free medium. Furthermore, CRCs of 0.10 and 0.30 µM verapamil concentrations as standard calcium channel blockers were evaluated in distinct groups.25 The mechanism test was performed by placing ileum samples in a Ca++ free medium containing: NaCl (136.90), NaHCO3 (11.90), glucose (5.60), KCl (2.70), MgCl2 (1.10), Na2HPO4 (0.40), and EDTA (0.10) mM for 30 min and then replacing it with depolarizing solution (containing: NaCl (91.04), KCl (60.00), NaHCO3 (11.90), glucose (5.55), MgCl2 (1.05), Na2HPO4 (0.42), and EDTA (0.10) mM for 45 min. Before doing this, the samples passed 1 hr adaptation period in thyroid solution and their viability and contractive activity were evaluated with acetylcholine. Exchanging the solutions in each step was done every 15 min. After the K+ rich and Ca++ free (depolarizing) solutions, two control CRCs were recorded with the addition of calcium concentrations. After each period, washing the tissues with the depolarizing solution restores the contractions to baseline. In the third round, the rates of Ca++ CRCs were recorded 10 min after adding the essential oil (30.00 and 100 µg mL−1), carvone (0.10 and 0.30 µM), or verapamil (0.10 and 0.30 µM).25 Cumulative concentrations of 0.003, 0.01, 0.03, 0.10, and 0.30 µM verapamil were added to the induced contractions of 60.00 mM KCl (K60) to investigate the effect of standard calcium channel blocker. Cumulative concentrations of TBEO and carvone were added to the KCl 20.00 mM (K20) induced contractions in separate groups to assess their effects on low dose KCl (K20) and compare it with high dose KCl (K60).23

Statistical analysis. The assumption of data normality was tested by the Shapiro-Wilkes test. Moreover, data were graphically assessed using histogram and box plots for assumptions of normal distribution and homogeneity of variance. Since the assumptions were not met, the nonparametric Friedman repeated measures analysis of variance on ranks was employed to compare the results. Pairwise comparison between each concentration and the control were identified by the use of the Dunnett’s test. The significance level was set at p < 0.05. Results were expressed as medians and interquartile ranges (25th-75th percentiles). IBM SPSS Statistics for Windows, (version 25.0; IBM Corp., Armonk, USA) was used for statistical analysis of data.

Results

The overall result of this study was that TBEO has spasmolytic effects on bovine ileum smooth muscle. TBEO reduces both spontaneous and spasmogen-induced contractions. TBEO significantly reduced ileal circular smooth muscle baseline contractions at concentrations of 100 to 1000 µg mL−1. The cumulative addition of TBEO at concentrations of 100 to 1000 µg mL−1 significantly relaxed each of the contractions induced by BaCl2, KCl, and Cch (p < 0.05), (Fig. 1).

The GC-MS analysis of TBEO revealed the main ingredient in this essential oil (Table 1). The results showed that in our essential oil sample, as in previous studies, carvone was the major (63.92%) ingredient. Cumulative concentrations of 1.00 to 10.00 µM carvone led to a significant reduction in spontaneous basal contraction of the ileum smooth muscle. The Cch-induced contractions were significantly relaxed at 1.00 to 10.00 µM concentrations of carvone. The spasmolytic effect of carvone on KCl induced contractions was significant at 3.00 and 10.00 µM concentrations of carvone. The BaCl2-induced contractions were significantly inhibited only at 10.00 µM concentration of carvone (p < 0.05), (Fig. 1).
Fig. 1. The spasmolytic effect of TBEO and carvone on spontaneous and induced contractions (by Cch, KCl, and BaCl₂) on bovine ileum. Results are reported as the percentage of stable initial maximum contraction (SIMC) of each spasmogen and the percentage of stable initial basal contractions (SIBC) about spontaneous contractions. ★ Indicates significant reduction from SIBC or SIMC of each group at $p < 0.05$. 
The greatest amount of contraction reduction was in Cch-induced contractions, in which contractions average with TBEO reached 1.83% and with carvone 6.83% of the control contractions. The significant effect of TBEO in all spontaneous and induced contractions by Cch, KCl, and BaCl$_2$ started at 100 µg mL$^{-1}$ concentration, reaching the most amounts at 1000 µg mL$^{-1}$ concentration. The maximum concentration of TBEO (1000 mg L$^{-1}$) reduced spontaneous contractions less than induced contractions. The highest resistance to carvone’s myorelaxant effect was in BaCl$_2$-induced contractions, significantly reducing the contraction with only one concentration of carvone (10.00 µM). The next resistance was in KCl contractions, significantly decreasing at 3.00 and 10.00 µM concentrations of carvone. Baseline and Cch-induced contractions were most impressible against carvone and showed a significant reduction at 1.00 to 10.00 µM of concentrations. Evaluation of the results revealed that the myorelaxant effect of TBEO and carvone is a concentration-dependent that increases with increasing concentrations. The most potent effect of TBEO was at a concentration of 1000 µg mL$^{-1}$, and the most potent effect of carvone was in 10.00 µM concentration.

Two concentrations of TBEO reduced the contractions created by adding calcium to the depolarizing solution (K$^+$-rich and Ca$^{2+}$-free). Besides, two different concentrations of carvone inhibited contractions due to the influx of calcium into the depolarizing solution. The effect of standard calcium channels blocker (verapamil) on bovine ileum smooth muscle contractions was shown, too. Comparison of the results obtained from the blockade of calcium channels by TBEO and carvone with verapamil proved that TBEO and carvone also have the property of inhibiting the calcium channels of smooth intestinal muscles (Fig. 2). The 0.10 and 0.30 µM concentrations of verapamil caused a significant reduction in K60 contractions. None of TBEO or carvone concentrations significantly reduced the K20 contractions.

The control treatments showed that the contractions created with the three spasmogens Cch, KCl, and BaCl$_2$ survived for 25 min. The primary solvent of essential oils and carvone (i.e. DMSO) had no significant effect on ileal contractions.

Table 1. The main components of Tanacetum balsamita essential oil (TBEO) based on GCMS findings.

| Components            | RI  | RT  | Percentage |
|-----------------------|-----|-----|------------|
| 1,8-Cineole           | 1032| 7.21| 3.39       |
| Beta thujone          | 1099| 8.95| 11.53      |
| Alpha thujone         | 1115| 9.17| 1.20       |
| Menthatriene          | 1119| 9.25| 2.15       |
| Para menthatriene     | 1133| 9.58| 1.42       |
| Pinocarvone           | 1162| 10.26| 0.54      |
| Borneol               | 1165| 10.33| 0.63      |
| Terpinen-4-ol         | 1176| 10.58| 0.56      |
| p-Diethylbenzene      | 1186| 10.82| 2.77      |
| trans-Dihydrocarvone  | 1196| 11.04| 1.33      |
| trans-Piperitol       | 1200| 11.13| 1.52      |
| Mentha-1,4,8-triene   | 1219| 11.58| 2.19      |
| Nerol                 | 1229| 11.81| 2.04      |
| Carvone               | 1249| 12.25| 63.92     |

RI: Retention index, RT: Retention time.

Fig. 2. Concentration-response curves (CRCs) of bovine ileal smooth muscle to increasing Ca$^{2+}$ dose in the absence and presence of two different concentrations of each of TBEO (A), carvone (B), and verapamil (C).
Discussion

This study investigated the effects of TBEO and its main ingredient (carvone) on contractions induced by several spasogens in bovine ileum smooth muscle in an organ bath.

Due to the necessity of research ethics and animal rights rules, in vitro studies were preferred. Many in vitro (organ bath) studies have been performed on the gastrointestinal motility of animals, and it is believed that these experiments may reflect in vivo conditions. Although the ileum is used to evaluate bowel motilities in most species, there are few studies on ileal motilities in cattle. However, one of the sites involved in bovine gastrointestinal syndromes such as diarrhea syndrome is the ileum, which may play a significant role in the pathophysiology of GI diseases such as bovine viral diarrhea virus (BVDV) and Johne’s disease. The ileums of ruminants and non-ruminants have many anatomical and physiological similarities, making it possible to extend the results of this study to other animals and even humans.

The exact effect of *T. balsamita* and its mechanism of action on gastrointestinal motility are undefined. *T. balsamita* in the formulation of massage lotion has muscle relaxant properties, agreeing with its anti-spasmodic properties in the gastrointestinal muscles in the present study. In other members of *Tanacetum* genus, the antispasmodic effect on smooth muscles has been explained for *T. parthenium* and *T. vulgare*. The extract of *T. parthenium* reduced the spontaneous and induced contraction of the ear artery and aortic smooth muscle as well as the anococcygeal body in rabbits. The effects of *T. parthenium* were concentration-dependent and aortic irreversible. However, the myorelaxant effect of *T. balsamita* was concentration-dependent and reversible (the present study). The *T. parthenium* may inhibit migraine pain by the anti-spasmodic effect on cerebrovascular smooth muscles. The aqueous extract of *T. vulgare* (*Tansy*) relaxed the KCl-induced contraction in the smooth muscle of the rat aorta. The significant effect of *T. parthenium* came from concentrations of 50.00 to 500 µg mL⁻¹, and 1000 µg mL⁻¹ completely blocked the contractions. Significant effect of *T. vulgare* was observed from 100 to 800 µg mL⁻¹. In the present study, the first significant effect of TBEO concentrations was 100 µg mL⁻¹, peaking at 1000 µg mL⁻¹, but complete inhibition occurred only in some of the Cch-induced contractions. These results showed that the spasmylocytic potency of *T. balsamita* was similar to *T. vulgare* and slightly lower than *T. parthenium*.

The GC-MS analysis results revealed that, as in previous studies, carvone was the major ingredient in TBEO. In various TBEO samples, components other than carvone have been identified as major one, including quercetin, bornyl acetate, and Trans-chrysantheno. In various TBEO samples, carvone, beta-thujone, quercetin, bornyl acetate, and Trans-chrysantheno have been identified as major components. In previous studies, the authors identified carvone, beta-thujone, quercetin, bornyl acetate, and Trans-chrysantheno as major components of tested TBEO. Differences in the chemical composition of the TBEO in previous studies may be due to the different geographical origins of the plant. Moreover, carvone had an anti-spasmodic effect on the smooth muscle of the bovine ileum (Fig. 1), justifying the hypothesis of carvone being responsible for TBEO anti-spasmodic activity.

Carvone could reduce Cch-induced contractions in the aorta of rats and guinea pig trachea smooth muscles. Different carvone reversals have the same effect on the relaxation of vascular and respiratory smooth muscles. However, in another study, the anti-spasmodic effect of carvone on guinea pig ileum smooth muscle was stronger than carvone. In an experiment, carvone inhibited the contractions of mercury and lead on rat aorta. In this experiment carvone had no significant effect on the passive tension of the aorta. The spasmolytic effects of carvone on contractions induced by Cch, KCl, and BaCl₂ in guinea pig ileum smooth muscle have been proved. In this study, carvone had no significant effect on spontaneous contractions of the ileum. The Cch-induced contraction significantly reduced by carvone but did not decrease to baseline. In line with the study of de Sousa et al., in the present study the largest decrease of contractions due to carvone was observed in the Cch-induced contractions. Although the average of contractions did not reach the baseline, in half of the samples the contractions were completely inhibited (Fig. 1). Carvone had no significant effect on spontaneous smooth muscle contractions in guinea pig ileum and rat aorta, while in the present study antispasmodic effects of 0.01 to 0.001 µM carvone on bovine ileum spontaneous contractions were significant. Cumulative concentrations of 0.01 to 0.001 µM carvone showed a significant reduction in all contractions. This difference may be due to differences in the animal species being tested. The significant concentrations of carvone in previous studies were from 1.00 to 300 µM, which were significant from 0.01 to 0.001 µM in our study. These results could indicate that the bovine ileum is more sensitive to carvone than the guinea-pig ileum.

Antispasmodics can either block specific receptors or act in a non-specific way. Different pathways and receptors are involved in causing each of the spontaneous and induced (Cch, KCl, and BaCl₂) contractions. In the present study, TBEO and carvone inhibited all types of these contractions; therefore, their mechanism of action is
nonspecific. The Cch-induced contraction in smooth muscle is highly dependent on intracellular calcium stores, and in present study, the more significant effect of TBEO and carvone on Cch-induced contractions reinforced the hypothesis of their inhibitory effect on calcium channels. Comparing the effects of TBEO and carvone with the standard calcium channel blocker (verapamil) showed that the muscle relaxant effect in TBEO and carvone was exerted by blocking calcium channels. Other researches have not provided sufficient evidence to the mechanism of the muscle relaxant effect of T. balsamita. In addition, the impaired calcium transfer has been observed in other Tanacetums, and in carvone consistent with the results of the present study. Studies on carvone mechanism of action have been investigated by comparing the effects of carvone and verapamil on induced contractions in the aorta of rats and guinea pig ileum. KCl works both at high doses (K60) and at low doses (K20) by opening calcium channels and opening potassium channels, respectively. The significant effect of TBEO and carvone on K60 contractions and their ineffectiveness on K20 confirm the CCB mechanism in smooth muscle.

Concentrations of carvone caused a significant decrease in all spontaneous and induced contractions. These concentrations were different for each spasmodogen. On the other hand, significant concentrations of TBEO were equal in all groups. This difference may be due to the presence of non-carvone spasmylytic compounds in TBEO. These compounds (non-carvone) may have functional mechanisms other than CCB. In an experiment, the spectrophotometry method showed the inhibitory activity of T. balsamita extract on acetylcholinesterase. Thus, the components with contradictory properties in the composition of this plant are possible.

Muscle relaxants may have beneficial clinical uses. In-vitro myorelaxant effect of herbal extracts can be expected to reduce gastrointestinal transit and aim to diarrhea treatment in live animals. Using modulators of intestinal motility may lead to better results than using antibiotics alone for diarrhea. Intestinal smooth muscle spasm is one of the causes of the acute abdominal syndrome in ruminants. TBEO can be a natural alternative to antispasmodics such as atropine and hyoscine butyl bromide in this syndrome. TBEO products can also help relieve the physical obstruction of the intestinal lumen by relaxing the intestines. Anti-spasmodic action of TBEO may be useful in facilitating the passage of calcium and reducing the pain caused by uroliths in ruminants. The T. balsamita can concomitantly exert several beneficial medicinal (anti-inflammatory, antimicrobial, antioxidant, analgesic, and myorelaxant) effects in gastrointestinal disorders associated with hypermotility and/or spasm. A promising finding of the clinical use of T. balsamita is that TBEO consumption in animal diets had no adverse effects on carcasses and nutritional value of the meat.

In conclusion, the ability of the TBEO to reduce spontaneous and induced contractions in-vitro on bovine ileum reveals its ability to relieve spasms and control intestinal hypermotility in living ruminants. Carvone, as the major component in TBEO, has similar muscle relaxant effects on ileal smooth muscle contractions and it is probably responsible for an important part of TBEO’s antispasmodic effects. Both TBEO and carvone relax smooth muscles by calcium channel blocking. T. balsamita can be an excellent candidate for producing a gastrointestinal motility modulator in bovine medicine and even in human medicine. The products of this plant or its major ingredient (carvone) may be used to treat ruminant gastrointestinal disorders such as acute abdomen or intestinal hypermotility (such as diarrhea syndrome). However, further research should be done to evaluate the effects of T. balsamita on gastrointestinal motility in vivo.

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Conflict of interest

The authors declare no competing financial interest.

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