Original article

Lemon Balm (*Melissa officinalis L.*) Hydrolate Poorly Influences Isolated Rat Gastric Fundus Motility

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SUMMARY

Introduction. Lemon balm has been used for centuries due to its strong sedative, antimicrobial and spasmolytic effects. Besides different types of extracts and essential oils, hydrolates (by-products obtained during essential oil extraction) are preparations of aromatic medicinal plants with potential pharmacological activity.

Aim. To estimate *Melissa officinalis* leaves hydrolate effects on gastric motility.

Methods. The influence of the hydrolate was tested *in vitro* in a tissue bath in the rat fundus spontaneous contraction experimental model. Increasing volumes of sample (making the final concentration span 0.5 – 500 μl/ml) were successively added to the tissue bath.

Results. Lower doses of the hydrolate did not produce any significant changes in the contraction patterns, while the higher ones led to a statistically significant increase in basal tone of the stomach fundus. Yet, these changes in basal tonus were still modest. Phytochemical qualitative characterization, done by liquid chromatography with ultra-high performances, showed that rosmarinic acid was dominant compound of *M. officinalis* hydrolate.

Conclusion. Although the tested hydrolate poorly influences isolated rat gastric fundus motility, further studies could clarify its pharmacological activity and potential future application of this by-product of *M. officinalis* hydrodestilation.

Keywords: *Melissa officinalis*, hydrolate, gastric fundus, contractions

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INTRODUCTION

Melissa officinalis L. (*M. officinalis),* a plant species belonging to the family of Lamiaceae, was named lemon balm due to its lemon-like scent (1). Even though it can be found throughout the world, it commonly grows in Mediterranean region and west Asia, but it is also widely cultivated in Europe (2). Lemon balm extracts have complex chemical composition, which includes essential oil constit-uents such as citral (about 30%), geraniol, linalool and citronellal (about 40%), polyphenolic acids (rosmaric and caffeic acids), tanines, flavonoids, triterpens, etc. (3). As an herbal remedy *M. officinalis* has been used for centuries, due to its strong sedative, antibacterial, antiviral, and spasmolytic effects (4). It is generally considered safe for oral use and has already been placed on FDA list. The acute lethal dose (LD50) of *M. officinalis* essential oil in mice is 2.56 g/kg (5). The most common form for utilization of *M. officinalis* is herbal tea (water extract). This type of herbal remedy is already used in Germany for treating insomnia and headache, while in Austria it is used to mitigate gastrointestinal, liver, biliary, and neuronal symptoms (6). The Council of Europe has suggested that a leaf of this plant can be used for aromatizing food in category N2, which means that *M. officinalis* can be used as a form of meal seasoning. Also, it can be used for aromatizing wine, beer, or tea (7).

Disorders of gastric motility are common and patients often visit a doctor due to non-specific symptoms such as nausea, vomiting, abdominal pain, bloating, etc. Motoric gastric dysfunction includes two basic disorders: accelerated and delayed emptying of the stomach (8, 9). Accelerated emptying of the stomach almost always develops in patients subjected to the surgical procedure of modification or either partial or total removal of the stomach, which results in dumping syndrome. Typical symptoms include nausea, pain, bloating, diarrhea, syncope, redness, and palpitations lasting between 20 and 60 minutes (10). However, a much more common gastric motility disorder is gastroparesis, which represents a chronic, idiopathic disorder in the absence of any mechanical obstruction which leads to delayed emptying of the stomach (11). The main symptoms include early satiating and fullness after a meal, nausea, vomiting, abdominal pain, and bloating (12). Gastroparesis can be a characteristic of

other disorders or diseases, such as vascular disease of collagen, metabolic and endocrine disorders, as well as a side effect of anticholinergic and opioid drugs. However, in most cases in clinical practice, it is caused by vagotomy, diabetes mellitus, or is idiopathic in origin (13).

Symptoms in patients with gastric motility disorders can be alleviated by a change in dietary regime, through the regulation of glycemia in patients with diabetes mellitus, but mostly through therapy. The most important drugs for gastroparesis are prokinetic drugs, which enhance gastric motility by acting as an agonist of 5-HT4 receptors (cisapride, metoclopramide, domperidone, and tegaserod), and some as a D2 antagonist (metoclopramide) whose effect in alleviating nausea is known, but the effect on gastric motility is still being researched. Second-line prokinetics, which are rarely used, are erythromycin (an agonist of motilin receptors) and cholinergic drugs, most commonly anticholinesterase inhibitory drugs (14, 15). However, because of the significant side effects of these drugs (heart arrhythmias, depression, and extrapyramidal excitation), many national health organizations withdrew prokinetic drugs from the market, which led to a gap in the potential treatment of gastric motility disorders (16). This gave rise to alternative treatments such as herbal remedies (e.g. ginger extracts), psychotherapy, hypnosis, and others (17). Herbal remedies show a promising future due to their wider safe profile. Up to now, the only approved herbal remedy is STW-5 (*Iberogast*), which contains a mixture of plant extracts, used for treating dyspepsia and digestive tract motility disturbances, and can be considered as an alternative to prokinetic drugs (16). Moreover, some other plant extracts are being researched for their effect on gastrointestinal motility, and these include wide-garlic (18), bojungikki-tang (19), etc.

Since hydrolates (aromatic waters obtained during essential oil production) have moderate activity in comparison to corresponding essential oils, it makes make them potential daily soft drinks with therapeutic features (20). The aim of this study was to determine whether the hydrolate obtained during the hydrodestilation of *M. officinalis* leaves could influence the spontaneous contractions of isolated rat gastric fundus muscle.

METHODS

Drugs and chemicals

Commercially available *Melissa officinalis L.* hydrolate, produced by Promontis (Vilandrica, Serbia), was used in this study. All chemicals used were purchased from Sigma Aldrich (USA), Carl Roth (Germany), or Acros Organics (Belgium).

Chemical analysis

For qualitative characterization of the hydrolates, the chromatographic separation was performed by liquid chromatography with ultra-high performances (UHPLC) using a Dionex Ultimate 3000 UHPLC+ system equipped with a diode array (DAD) detector and LCQ Fleet Ion Trap Mass Spectrometer (Thermo Fisher Scientific, USA). The separation was obtained on Hypersil gold C18 column (50 × 2.1 mm, 1.9 μm) at 25°C. The mobile phase composed of two solvents, 0.1% formic acid in water (A) and methanol (B) at 0.250 mL/min flow rate with a gradient program is shown in Table 1. The injection volume was 50 μl. Absorption UV–Vis spectra were recorded on DAD-detector with a total spectral range between 200 and 800 nm. Mass spectrometric analysis was performed using a 3D–ion trap with electrospray ionization (ESI) in negative ion mode. The ESI–source parameters for both used methods were as follows: source voltage 4.5 kV, capillary voltage −41 V, tube lens voltage −95 V, capillary temperature 350°C, nitrogen sheath, and auxiliary gas flow 32 and 8 arbitrary units. Mass spectra were acquired by full range acquisition of m/z 100 – 900, with a tandem mass spectrometry analysis performed by a data-dependent scan – the collision-induced dissociation (CID) of detected molecular ions peaks ($[M−H]^-$) tuned at 30 eV. Additional MS analysis in positive ESI mode was provided for the detection of two characteristic compounds, thymol and carvacrol at 5.0 kV, 49 V, and 115 V for source voltage, capillary voltage, and tube lens voltage, respectively. Full range acquisition of m/z 130 – 900, with a tandem mass spectrometry analysis with CID of detected molecular ions peaks ($[M+H]^+$) at 30 eV. Xcalibur software (version 2.1) was used for instrument control, data acquisition, and data analysis.

The assignation of the detected compounds was based on their retention times, UV–Vis from DAD-detector and MS spectra with the corresponding molecular ion peaks as well as characteristic ion fragmentation of selected peaks (MS/MS) from corresponding UHPLC chromatograms. Identification of detected compounds was also provided by using reference standards for some compounds (citric acid, chlorogenic, and rosmarinic acid).

Animals and housing

Adult male disease-free Wistar rats (200 – 250 g) used in this study were obtained from the Animal Research Centre of the Medical Faculty, University of Niš. The animals were maintained under standard housing conditions (temperature 23 ± 2°C; humidity 55 ± 10%; 12/12 h light/dark cycle). All animals had constant access to standard (commercial) laboratory food pellets and water. The experiments were performed in accordance with the declaration of Helsinki and European Community guidelines for the ethical handling of laboratory animals (EU Directive of 2010; 2010/63/ EU) and the experimental protocols were commenced after being approved by the institutional animal Ethics committee (No. 323-07-06862/ 2016–05/1) and the one of the Republic of Serbia.

Gastric fundus isolation and preparation

Rats were fasted overnight and sacrificed on the morning of the experiment. Afterwards, their abdomen was opened and stomach was dissected out and placed in a Petri dish field with Tyrode's solution (136.89 mM NaCl, 2.68 mM KCl, 1.05 mM $MgCl_2$, 1.80 mM $CaCl_2$, 0.42 mM NaH_2PO_4, 11.09 mM $NaHCO_3$ and 5.55 mM glucose; pH 7.4). From each stomach, gastric fundus dissection was cut to make fundal strips (2 - 3 cm long), which were then suspended in a 10-ml tissue bath with Tyrode's solution which was kept at 37°C and aerated with 5% (v/v) carbon dioxide in oxygen mixture. An end of a fundal strip was attached to the bottom of the bath, while the other one was attached to an isotonic force transducer via a cotton thread (TSZ-04-E, Experimetria Ltd., Budapest, Hungary). Fundal responses were recorded and analyzed with a SPEL Advanced ISOSYS Data Acquisition System (Experimetria Ltd.). The segments of the isolated fundus were suspended under 1 g tensions and left to equilibrate for 30 min before the experiment.

Experimental procedure

At the start of the experiment, the first 5 min were recorded for spontaneous contractions without any changes in the environment. After that period, starting at the 5th minute, at each 5 min interval hydrolate was added to the organ bath cumulatively in the next order: 10, 50, 100, 250, 500 (0.5 – 910 µl/ml) of the tested substances, with no washing between the different concentrations (21). The change in the contractions were calculated as an area under the curve during the 5-min time interval when the tissue was exposed to the test concentration.

Statistical analysis

The results are presented as percent ± SD of increase/decrease from the baseline obtained from each fundal tissue strip before the application of the hydrolate. Statistically significant differences were determined by one-way analysis of variance (ANOVA), followed by Tukey's post hoc test for multiple comparisons (GraphPad Prism, ver. 5.03; San Diego, CA). Probability values (p) less than 0.05 were considered to be statistically significant.

RESULTS

Results of UHPC-DAD-MS/MS analysis are presented in Table 1. Since the most of hydrosols is water, the injection volume was 50 µl instead of the commonly applied 20 µl. Although there are 9 identified picks/5 identified compounds in the tested sample, the dominant one is undoubtedly rosmarinic acid.

The application of an increasing volume of *M. officinalis* hydrolate initially (10 and 50 µl) did not produce any significant changes in the basal tone of the isolated fundus (Figure 1 and 2). In the following dose of 100 µl, statistically significant increase in the basal tone was noted (Figure 2). The application of the fourth dose of hydrolate (250 µl) led to a statistically significant increase in the basal tone, however, this change was not as obvious as the one with 100 µl (Figure 2). The final, fifth dose, produced a slight decrease from a basal tone, however, this decrease was not significant compared to the untreated tissue (Figure 2). This kind of activity could be interpreted as an inverted U-shape curve (Figure 2).

Table 1. *Compounds detected in hydrolate using UHPLC-DAD-MS/MS*

Peak No.	t_R, min MS	λ_{max}, nm	Molecular Ion [M-H]⁻ m/z	Molecular Ion [M+H]⁺ m/z	MS/MS fragment ions	Assignment (reference)
1	0.74	-	191	-	111 (100%), 93.85	Quinic acid (22)
2	0.92	-	191	-	173.111 (100%)	Citric acid (standard)
3	2.05	278	-	-	-	*n.i.* phenolic acid
4	5.53	325 303sh	-	355/377	-	Chlorogenic acid (standard)
5	8.30	301	437	-	153 (100%)	*n.i.* phenolic acid
6	9.65	330	359	-	161 (100%)	Rosmarinic acid (standard)
7	10.51	263 299	-	193	175 (100%), 147.133.105	Senkyunolide A (23)
8	12.95	243 338	327	-	283, 229 (100%), 211	*n.i.* prenylated isoflavonoid
9	13.24	244	-	153	135,107,95 (100%),71	*n.i.*

sh – shoulder; *n.i.* - non identified
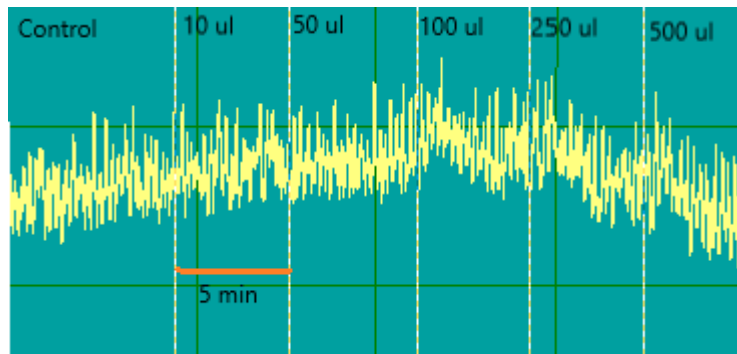
Figure 1. *Contraction patterns of the isolated gastric fundus observed during the course of the experiment*
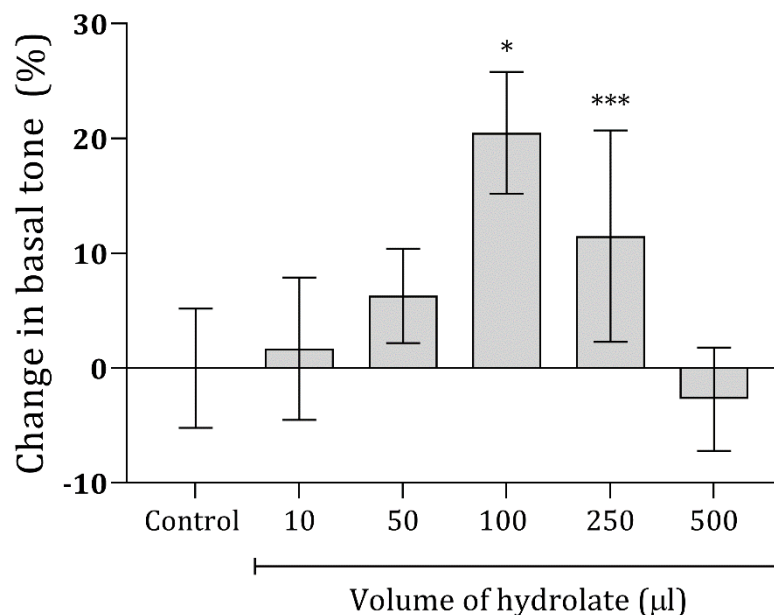
Figure 2. *Change in basal tone of rat gastric fundus after cumulative exposure to* M. officinalis *hydrolate. Data are given as mean ± SD (n = 6) and further compared with ANOVA followed by Tukey's post hoc test; *p < 0.001, ***p < 0.05 vs. control*

DISCUSSION

This study was designed to examine the impact of hydrolate, a by-product obtained during *M. officinalis* leaves essential oil isolation, on spontaneous gastric fundus contraction in *in vitro* conditions. Lower doses did not produce any significant changes in the contraction patterns, while the third and fourth dose (100 and 250 µl) led to a statistically significant increase in the basal tone of the stomach fundus (Figure 2). The final, fifth dose exerts a reduction potential on the spontaneous contraction of the stomach fundus (Figure 2), which could pos-

sibly be attributed to a change in the composition of the complex physiological solution (Thyroid solution). In the experiments performed under conditions identical (similar) to the ones in the present study, it is suggested that the volume of the examined drug should not exceed 5% of the total tissue bath volume (24). In this case, 500 µl is exactly 5% of the bath volume, which suggest that it might disturb the homeostatic environment achieved with Thyroid solution.

There are several major mechanisms through which a plant-based drug could exert its activity, which include voltage-gated Ca^{2+} channels, cholin-

ergic and serotoninergic system, as well as non-ad-renergic, non-cholinergic (NANC) pathways. Further, we will try to explain through which mechanism could the hydrolate act, and which are previously found to be involved in the spasmolytic action of *M. officinalis* extracts. Also, it should be noted that a dry residue of hydrolates is usually less than 1 g/L, and they could be considered a very diluted hydrosoluble fraction from essential oils (25).

During the process of the cell membrane depolarization, Ca^{2+} voltage-gated channels open, allowing Ca^{2+} entry and consequential contraction of the smooth muscle cells, and in *in vitro* conditions, this process can be induced by applying high K^+ concentrations (26). The inhibition of Ca^{2+} voltage-gated channel opening is known to be the underlying mechanism of action of a great many medicinal plants, as well as of their pure constituents (27). The same mechanism, demonstrated on isolated rat ileum tissue, was found to be associated with the activity of *M. officinalis* essential oil rich in citral (26). However, with an increase in the concentration of hydroalcoholic extract there is no increase in its inhibitory effect (28).

Acetylcholine (ACh), as a neurotransmitter released by the myenteric plexus, causes the contraction of smooth muscle cells of the digestive tract (29). The muscarinic Ach receptor family has five types (M1–M5) of membrane-GTP-associated proteins which trigger different intracellular signaling cascades following the interaction of an agonist with a receptor. The role of M1–M4 receptors in the motility of the gastric fundal strips isolated from rats has been previously shown (30). Thus, a decrease in spontaneous contractility of the fundus could be the consequence of decreased Ach release or the blockage of muscarinic receptors.

Serotonin (5-HT) represents one of the major signaling molecules, which conveys signals from the intestine lumen to sensory neurons via 5-HT receptors. The rat fundus is rich in 5-HT2B receptor, a subtype of the 5-HT2 receptor family, which is widespread throughout the body (31). This neurotransmitter participates in the initiation of the peristaltic reflex and segmentation of the gut, and it is believed that it could induce the peristaltic reflex, however, its presence is not of vital importance in this reflex (32). Previous studies showed that lemon balm essential oil and citral, as one of its major constituents, have potent inhibitory effects on the 5-HT induced ileum contraction.

Non-adrenergic, non-cholinergic (NANC) nerves also play an important role in intestine motility, due to their involvement in gastrointestinal reflexes, as well as in gastric receptive relaxation (33). *In vivo* animal studies showed that the storage of food in the fundus is a mechanism with involves various neurotransmitters such as nitric oxide, ATP, and vasoactive intestinal polypeptide. Apart from this, it is worth mentioning that nitric oxide is believed to be the dominant inhibitory neurotransmitter associated with the motility of the gastrointestinal tract (34, 35).

Rosmarinic acid was found to be one of the main compounds of lemon balm leaf hydroethanolic extract that Aubert and co-workers tested on the intestine contractions. These authors concluded that hydroethanolic extract possesses site- and dose-dependent effects since it affected the jejunum and ileum, but not the antrum and colon (36). Our study with *M. officinalis* hydrosol confirmed the weak activity of lemon balm hydrolate, but also rosmarinic acid, as the main component, on gastric tissue.

Bearing in mind that essential oils are rarely safely applied per os, hydrolates could be a potential way of ingestion diluted hydrosoluble fractions of essential oils. Yet, since hydrolates have their own composition (differing from the one of the essential oil obtained during the same hydrodistillation process), moderate pharmacological activities, and often some beneficial properties not possessed by the essential oil alone, the investigation of hydrolates helps in developing of some functional beverages and soft drinks (20).

CONCLUSIONS

The results of our study showed that *M. officinalis* hydrolate could significantly potentiate the spontaneous contraction of the isolated rat gastric fundus when applied in a dose of 100 and 250 µl. On the other hand, the highest tested dose of the hydrolate, 500 µl per bath, led to a reduction in basal tone of the isolated rat fundus. Hydrolates are promising new functional beverages and soft drinks and may be a safe way of administration of essential oils. Although the tested hydrolate poorly influences the isolated rat gastric fundus motility, further studies could clarify its pharmacological activity and potential future application of this by-product of *M. officinalis* hydrodestilation.

References

1. Kennedy DO, Scholey AB. The Psychopharmacology of European herbs with cognition-enhancing properties. Curr Pharm Des 2006; 12(35):4613–23. https://doi.org/10.2174/138161206779010387

2. Meftahizade H, Lotfi M, Moradkhani H. Optimization of micropropagation and establishment of cell suspension culture in *Melissa officinalis L*. Afr J Biotechnol 2010; 9(28):4314-21.

3. Mencherini T, Picerno P, Scesa C, Aquino R. Triterpene, antioxidant, and antimicrobial compounds from *Melissa officinalis.* J Nat Prod 2007; 70(12):1889–94. https://doi.org/10.1021/np070351s

4. Nasri H, Rafieian-Kopaei M. Oxidative stress and aging prevention. Int J Prev Med 2013; 4(9):1101–2.

5. Stojanović NM, Randjelović PJ, Mladenović MZ, et al. Toxic essential oils, part VI: Acute oral toxicity of lemon balm (*Melissa officinalis L.*) essential oil in BALB/c mice. Food Chem Toxicol 2019; 133:110794. https://doi.org/10.1016/j.fct.2019.110794

6. Ulbricht C, Brendler T, Gruenwald J, et al. Lemon balm (*Melissa officinalis L.*): an evidence-based systematic review by the natural standard Research collaboration. J Herb Pharmacother 2005; 5(4):71-114.

7. Barnes J, Anderson L, Phillipson JD. Herbal Medicines. A Guide for Healthcare Professionals. 2nd ed. Pharmaceutical Press, London, Chicago, USA; 2002.

8. Malagelada JR, Rees WD, Mazzotta LJ, Go VL. Gastric motor abnormalities in diabetic and postvagotomy gastroparesis: effect of metoclopramide and bethanechol. Gastroenterology 1980; 78(2):286-93.

9. Keller J, Bassotti G, Clarke J, et al. International Working Group for Disorders of Gastrointestinal Motility and Function. Expert consensus document: Advances in the diagnosis and classification of gastric and intestinal motility disorders. Nat Rev Gastroenterol Hepatol 2018; 15(5):291-308. https://doi.org/10.1038/nrgastro.2018.7.

10. Van Beek AP, Emous M, Laville M, Tack J. Dumping syndrome after esophageal, gastric or bariatric surgery: Pathophysiology, diagnosis, and management. Obes Rev 2017; 18(1):68–85. https://doi.org/10.1111/obr.12467

11. Camilleri M, Parkman HP, Shafi MA; American College of Gastroenterology. Clinical guideline: management of gastroparesis. Am J Gastroenterol 2013;108(1):18-38. https://doi.org/10.1038/ajg.2012.373

12. Parkman HP, Hasler WL, Fisher RS; American Gastroenterological Association. American gastroenterological association technical review on the diagnosis and treatment of gastroparesis. Gastroenterology 2004; 127(5):1592-622. https://doi.org/10.1053/j.gastro.2004.09.055

13. Minami H, McCallum RW. The physiology and pathophysiology of gastric emptying in humans. Gastroenterology 1984; 86(6):1592-1610.

14. Soffer EE. Gastric and intestinal dysmotility syndromes. Cleve Clin J Med 1997; 64(2):69-71.

15. Sanger GJ, Broad J, Andrews PLR. The relationship between gastric motility and nausea: Gastric prokinetic agents as treatments. Eur J Pharmacol 2013; 715(1-3):10–14. https://doi.org/10.1016/j.ejphar.2013.06.031

16. Madisch A, Vinson BR, Abdel-Aziz H, et al. Modulation of gastrointestinal motility beyond metoclopramide and domperidone: Pharmacological and clinical evidence for phytotherapy in functional gastrointestinal disorders. Wien Med Wochenschr 2017; 167(7-8):160-8. https://doi.org/10.1007/s10354-017-0557-3

17. Chen JDZ, Yin J, Hou X, Takahashi T. Complementary and Alternative Therapies for Functional Gastrointestinal Diseases 2016. Evid Based Complement Alternat Med 2017; 2017: 2089165. https://doi.org/10.1155/2017/2089165

18. Pavlović DR, Veljković M, Stojanović NM, et al. Influence of different wild-garlic (Allium ursinum) extracts on the gastrointestinal system: spasmolytic, antimicrobial and antioxidant properties. J Pharm Pharmacol 2017; 69(9):1208-18. https://doi.org/10.1111/jphp.12746

19. Kwon HE, Kim JN, Kwon MJ, et al. The traditional medicine bojungikki-tang increases intestinal motility. Phcog mag 2021;17(5):1-8. https://doi.org/10.4103/pm.pm_507_20

20. Hamedi A, Moheimani SM, Sakhteman A, et al. An overview on indications and chemical composition of aromatic waters (hydrosols) as functional beverages in Persian nutrition culture and folk medicine for hyperlipidemia and cardiovascular conditions. J Evid Based Complementary Altern Med 2017;22(4):544-561. https://doi.org/10.1177/2156587216686460

21. Stojanović NM, Mladenović MZ, Maslovarić A, et al. Lemon balm (*Melissa officinalis L.*) essential oil and citronellal modulate anxiety-related symptoms - *In vitro* and in vivo studies. J Ethnopharmacol 2022; 284:114788. https://doi.org/10.1016/j.jep.2021.114788

22. Masike K, Mhlongo MI, Mudau SP, et al. Highlighting mass spectrometric fragmentation differences and similarities between hydroxycinnamoyl-quinic acids and hydroxycinnamoyl-isocitric acids. Chem Cent J 2017; 11(1):29. https://doi.org/10.1186/s13065-017-0262-8

23. Wang S, Liu L, Wang L, et al. Structural characterization and identification of major constituents in Jitai tablets by high-performance liquid chromatography/diode-array detection coupled with electrospray ionization tandem mass spectrometry. Molecules 2012; 17(9):10470-93. https://doi.org/10.3390/molecules170910470

24. Dekić M, Radulović N, Stojanović N, et al. Spasmolytic, antimicrobial and cytotoxic activities of 5-phenylpentyl isothiocyanate, a new glucosinolate autolysis product from horseradish (Armoracia rusticana P. Gaertn., B. Mey. & Scherb., Brassicaceae). Food Chem 2017; 232:329-39. https://doi.org/10.1016/j.foodchem.2017.03.150

25. Aćimović M, Tešević V, Smiljanić K, et al. Hydrolates: By-products of essential oil distillation: Chemical composition, biological activity and potential uses. Advanced Technologies 2020; 9(2):54-70.

26. Sadraei H, Ghannadi A, Malekshahi K. Relaxant effect of essential oil of Melissa officinalis and citral on rat ileum contraction. Fitoterapia 2003; 74(5):445-52. https://doi.org/10.1016/s0367-326x(03)00109-6

27. Sanders KM. Ionic mechanisms of electrical rhythmicity in gastrointestinal smooth muscles. Annu Rev Physiol 1992; 54:439–53.

https://doi.org/10.1146/annurev.ph.54.030192.002255

28. Khalaja A, Khani S. Spasmolytic effects of hydroalcoholic extract of *Melissa officinalis* on isolated rat ileum. J Report Pharmaceut Sci 2018; 7(3):260–9.

29. Szerb JC. Storage and release of labelled acetylcholine in the myenteric plexus of the guinea-pig ileum. Can J Physiol Pharmacol 1976; 54(1):12–22.
    https://doi.org/10.1139/y76-003

30. Milovanović DR, Janković SM. Pharmacologic characterization of muscarinic receptor subtypes in rat gastric fundus mediating contractile responses. Indian J Med Res 1997; 105:239-45.

31. Vaccari A. Pharmacological receptors in the rat stomach fundus. Pharmacology 1971: 5(6):321–30.
    https://doi.org/10.1159/000136207

32. Cohen ML, Fludzinski LA. Contractile serotonergic receptor in rat stomach fundus. J Pharmacol Exp Ther 1987; 243(1):264-9.

33. Curro D, Preziosi P. Non-adrenergic non-cholinergic relaxation of the rat stomach. Gen Pharmacol 1998; 31(5):697-703.
    https://doi.org/10.1016/s0306-3623(98)00096-2

34. McConalogue K, Furness JB. Gastrointestinal neurotransmitters. Baillieres Clin Endocrinol Metab 1994; 8(1):51-76.
    https://doi.org/10.1016/s0950-351x(05)80226-5

35. Whittle BJ. Nitric oxide-modulating agents for gastrointestinal disorders. Expert Opin Investig Drugs 2005; 14(11):1347-58.
    https://doi.org/10.1517/13543784.14.11.1347

36. Aubert P, Guinobert I, Blondeau C, et al. Basal and spasmolytic effects of a hydroethanolic leaf extract of *Melissa officinalis L.* on intestinal motility: an ex vivo study. J Med Food 2019; 22(7):653-62.
    https://doi.org/10.1089/jmf.2018.0154

Hidrolat matičnjaka (*Melissa officinalis L.*) slabo utiče na motilitet fundusa želuca pacova

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S A Ž E T A K

Uvod. Matičnjak se vekovima koristio kao lekovita biljka zbog svojih sedativnih, antimikrobnih i spazmolitičnih efekata. Osim različitih vrsta ekstrakata i etarskih ulja, hidrolati, koji se dobijaju kao nusprodukt u procesu ekstrakcije eterskih ulja, preparati su aromatičnog medicinskog bilja sa potencijalnom farmakološkim aktivnošću.

Cilj. Procena uticaja hidrolata iz lista matičnjaka na poremećaje gastrične aktivnosti.

Metode. Aktivnost hidrolata je ispitivana *in vitro* na spontane kontrakcije preparata fundusa želuca pacova u kupatilu za izoloane organe. Hidrolati su dodavani u rastućim koncentracijama (sa konačnom koncntracijom 0,5 – 500 μl/ml), sukcesivno, u vodeno kupatilo.

Rezultati. Niže doze nisu uzrokovale značajnije promene u kontrakcijama fundusa želuca, dok su više dovele do statistički značajnog povećanja u bazalnom tonusu. Međutim, ove promene bile su umerene. Fitohemijska kvalitativna karakterizacija, uređena metodom tečne hromatografije sa ultra visokim performansama, pokazala je dominantnost rozmarinske kiseline u hidrolatu *Melissa officinalis*.

Zaključak. Iako testirani hidrolat ima slab uticaj na kontrakcije preparata fundusa želuca, dalja istraživanja bi mogla da razjasne farmakološki efekat i potencijalnu upotrebu ovog nusprodukta hidrodestilacije matičnjaka.

Ključne reči: Melissa officinalis, hidrolat, gastrični fundus, kontrakcije