Therapeutic effect of herbal infusion on abnormal uterine bleeding: interventional non-randomized pilot study [version 1; peer review: awaiting peer review]

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Abstract

Background: Abnormal uterine bleeding-menometrorrhagia due to uterine myomas is a health problem affecting women's quality of life and it is considered a frequent cause of emergency visits for women. If first line medication fails to treat symptoms, surgical procedures, such as hysterectomy could be indicated, which could interfere with women's physical, social, emotional, and material quality of life. The purpose of this pilot study was to investigate the effect of a mixture of two medicinal plants, *Mentha pulegium* L and *Artemisia abrotanum* L, on 13 women with menometrorrhagia, who were candidates for hysterectomy.

Methods: The herbs were taken as a tea infusion by the oral route. A total of 5 g of the dried aerial parts of the mixture were added to 100 ml of boiling water and the tea infusion was taken three times a day starting from the onset of menstruation for three consecutive days, and it was repeated for three consecutive months.

Results: The study results showed that 10 out of the 13 women involved avoided the surgical procedures. The mean number of bleeding days declined from 11.50 (±3.77) at baseline to 7.60 (±2.11) (*p* = 0.01). Participants confirmed a change in the bleeding intensity, regularization of their menstrual cycle, and improvement in their quality of life.

Conclusions: This preliminary study explores a new approach to treat abnormal uterine bleeding- menometrorrhagia, based on tea infusion consumption of a mixture of two medicinal herbs, and it paves the way for future studies.

Trial registration: This study is registered with ClinicalTrials.gov NCT05406960 (07/06/2022).
Keywords
Abnormal uterine bleeding, Menometrorrhagia, Tisane, Traditional Medicine, North Africa

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Introduction
Menometrorrhagia, caused by uterine myomas, is defined as excessive uterine bleeding resulting from a combination of two different conditions, namely menorrhagia or hypermenorrhea, which is blood loss of more than 80 ml per cycle or having a cycle longer than seven days or both, and metrorrhagia, which is uterine bleeding at irregular intervals, particularly between the expected menstrual periods. It is defined as abnormal uterine bleeding (AUB) and known as prolonged and excessive uterine bleeding in irregular intervals. AUB is a worldwide health problem affecting 20% of adolescent girls and more than 50% of women older than 45 years. Management of AUB includes hormonal and non-hormonal treatments. If first line medication fails to treat women with AUB, surgical treatment, such as hysterectomy could be indicated. This solution is often used without a clear treatment strategy. The hypoestrogenic state of these therapies can lead to rapid bone demineralization and menopausal symptoms, such as vaginal dryness and hot flushes, having negative effects on women’s quality of life. Thus, because of these serious side effects, it seems logical to investigate other available sources. The therapeutic uses of Mentha pulegium L, known as Pennyroyal, and Artemisia abrotanum L, known as Southernwood, as traditional medicinal treatments for painful menstruation, pave the way for the possibility of new health-related uses. A mixture of these two herbs is recommended as a strong emmenagogue. Only few studies have explored the effect of Pennyroyal on menstruation, however, to the best of the authors’ knowledge, no scientific data highlighting the therapeutic effect of a mixture of Southernwood and Pennyroyal tea infusion on menometrorrhagia in women are available.

Methods
Study design
This is an interventional, non-randomized, controlled, single center pilot study. It was conducted in the department of Gynecology and obstetrics at Farhat Hached University Hospital, Sousse, Tunisia.

Ethical approval
Retrospective registration was acquired (Clinicaltrials.gov NCT05406960, 07/06/2022) because the study was taken to be low risk; however, the study was approved by the Human Research Ethics Committee at the Faculty of Medicine of Sousse, Tunisia and it was registered under the number (Ref: CEFMS 27/2019, 29/08/2019).

After being informed about the research protocol, each participant signed a written informed consent form prior to the initiation of the study procedures. All clinical investigation procedures were conducted according to the principles expressed in the declaration of Helsinki guidelines. This study is reported in line with the Consolidated Standards of Reporting Trials (CONSORT) guidelines.

Recruitment procedures
Participants meeting the inclusion criteria were informed about the research protocol by their gynecologists. They underwent complete physical examination following their acceptance to be enrolled in the study (Figure 1). They were interviewed to complete a quality-of-life questionnaire to determine the impact of the illness on their daily life and to record all details of their gynecological and menstrual histories. All participants were asked to report all details with regard to the effect of the treatment on menstrual cycle, menstruation duration, and bleeding intensity measured using pictorial blood assessment chart (PBAC) during three consecutive menstrual cycles treatments and the three-month follow-up period. They were also asked to report any change in the bleeding pattern and any adverse effects. Participants were recruited between 2019 and 2021. The study was interrupted by the COVID-19 pandemic.

Dosage and administration
The herbal tea infusion was taken by the patients at home after providing them with a detailed oral and written explanation. Assistance was provided by phone or via visits if any participant had questions regarding the treatment.

Participants took 5 g of mixed herbs powder added to 100 ml of boiling water and infused for 10 to 15 min, in accordance with the traditional uses. Herbal tea infusion was taken through the oral route before food, three times a day at 4-hour intervals after the first administration for three consecutive days, starting from the first day of the onset of menstruation.

Selection criteria
Inclusion criteria
The study included women with menometrorrhagia who were candidates for hysterectomy, aged between 30 and 55 years, not pregnant and not lactating and having a menstrual period for more than seven days with uterine bleeding at irregular intervals.
Exclusion criteria

The exclusion criteria included women suffering from abnormal uterine bleeding with menometrorrhagia but who were not candidates for hysterectomy, those suffering from gastrointestinal problems or any chronic diseases requiring long-term treatment, participants with increasing menstrual bleeding during the treatment requiring emergency surgical procedures, and those refusing the instructions given or who were participating in other clinical trials.

Therapeutic intervention

Method of herbs powder preparation

Pennyroyal and Southernwood were collected in July 2019 and 2020 from two different areas in the region of Sousse, Tunisia. The aerial parts (leaves and stems) of each plant were dried at room temperature in the shade; then, they were powdered in a rotating knife grinder. The powder was packaged in sachets, each containing 5 g, 2.5 g Pennyroyal and 2.5 g Southernwood, mixed according to the traditional uses of the herbs.

Treatment duration and follow-up

The treatment duration was up to three consecutive menstrual cycles and the total duration of the participants’ involvement in the study ranged between six and nine months.

Participants were followed via phone interview during and after the treatment. They were followed up for six months. Monitoring for efficacy and safety was performed every month starting from the treatment initiation until the treatment completion.

Restriction parameters

During the treatment period, participants were restricted to use just oral iron therapy, opioid analgesics, or acetaminophen. They were strictly asked to stop using any hormonal therapy, other herbal treatments, tranexamic acid, or any other treatment in relation to their disease.
Safety assessments

All participants were asked to record any possible adverse effects occurring during the study period. The effect of mixed herbal tea infusion was evaluated in the first four days. If no improvement in bleeding intensity was noted, participants were asked to stop taking the infusion and to return to their gynecologist to undergo a surgical procedure.

Outcome measurements

Clinical criteria

Participants were evaluated based on the bleeding intensity using PBAC scores, the duration of menstrual bleeding in days, regularization of menstrual cycle, and the improvement in their quality of life.

Biological criteria

Complete blood count (CBC) was performed before the start of the trial and after the third cycle of taking the infusion to control anemia. The safety and hemostatic effects of the treatment were evaluated by the analysis of prothrombin time (PT), prothrombin ratio (INR), and fibrinogen (FIB) to detect bleeding disorders, and the analysis of serum creatinine to detect urinary toxicity. Laboratory tests were conducted in the department of hematology at Farhat Hached University Hospital, Sousse, Tunisia. A 4ml blood sample was collected from each participant during the first visit before the start of the trial and after the last infusion consumption (day 91) in the same laboratory. A heparinized catheter was inserted into the antecubital vein to collect venous blood samples in three different tubes: an EDTA-coated tube, used for the CBC analysis parameters that were assessed following the combining laser diffraction flow cytometry and spectrophotometric technique, a sodium citrate 3.2% tube used to collect blood samples for the hemostasis analysis FIB, PT, and INR that were assessed by the coagulometric method, and finally, a lithium heparin tube used for creatinine analysis that was determined by the Jaffe’s reaction.

Sample size and statistical analysis

Sample size

The sample size was assessed according to the following formula Kang et al. \(N = (Z_{\alpha}/2)^2 s^2/d^2\), where \(s\) is the standard deviation (SD = 2300.00), and \(d\) is the accuracy of estimate or how close it is to the true mean (d = 900.40). Given the pioneering nature of this study, the above two data were collected from a previous work including women suffering from abnormal uterine bleeding-menometrorrhagia, conducted by Qaraaty et al., where the mean number of bleeding days was 10.6 ± 2.7 and 8.2 ± 1.9, respectively, before and after the consumption of myrtle syrup. \(Z_{\alpha}/2\) is the normal deviate for a two-tailed alternative hypothesis at a level of significance \(Z_{\alpha}/2\) equal to 1.44 at an error rate of 0.20%). The assessed sample size as \(N = (1.44)^2 2,300.00 / (900.40)^2\) gives a sample of 14 women suffering from abnormal uterine bleeding-menometrorrhagia. Assumption of 40% for non-attendance during the second or the third session gives a revised sample of 24 women suffering from abnormal uterine bleeding-menometrorrhagia (24 = 14/ (1.0–0.40)).

Statistical analysis

Data entry and analysis were performed using SPSS software version 10 to test for normality, distribution of the studied variables was examined in reference to Shapiro-Wilk test. The significance level was fixed at p<0.05. Descriptive statistics were used to examine the characteristics of the studied population (means, standard deviation). Comparison of the biological parameters and scores of PBAC between baseline and the end of treatment was performed using paired t-test when variables were normally distributed and Wilcoxon signed ranks test, as a non-parametric test, when variables were not normally distributed.

Results

 Characteristics of the study population

A total of 24 women with abnormal uterine bleeding-menometrorrhagia were involved in this study. During the course of the study, the data of 11 participants were excluded from the final statistical analysis. Indeed, the study was interrupted by the Covid-19 pandemic during which five participants withdrew from the trial because they took other different species of plants in order to prevent or treat Covid-19. Among the 11 excluded participants, three underwent surgeries for other reasons and the remaining three withdrew for personal reasons. Therefore, 13 participants completed the study (Figure 1).
Table 1. Data of Baseline Characteristics and response of infusion herbal treatment.

| Participants | Age (years) | Marital status | Number of abortions | Using contraceptive | clots  | Endometrial finding | Episode of flooding | Response to treatment | Side effect |
|--------------|-------------|----------------|---------------------|--------------------|--------|---------------------|---------------------|----------------------|------------|
| 1            | 42          | Single         | 0                   | no                 | Yes    | Normal              | No                  | Positive             | No         |
| 2            | 43          | Married        | 2                   | yes                | No     | Fibroma             | Yes                 | Negative             | Headache   |
| 3            | 48          | Single         | 0                   | no                 | Yes    | Fibroma             | No                  | Positive             | No         |
| 4            | 50          | Married        | 2                   | yes                | Yes    | Fibroma             | No                  | Positive             | Strong contraction |
| 5            | 32          | Single         | 0                   | no                 | Yes    | Fibroma             | No                  | Positive             | Strong contraction |
| 6            | 53          | Married        | 1                   | no                 | Yes    | Fibroma             | No                  | Positive             | No         |
| 7            | 47          | Single         | 0                   | no                 | No     | Fibroma             | No                  | Positive             | Strong contraction |
| 8            | 47          | Married        | 1                   | yes                | Yes    | Fibroma             | No                  | Positive             | Headache   |
| 9            | 43          | Single         | 0                   | no                 | Yes    | Normal              | No                  | Positive             | No         |
| 10           | 51          | Married        | 0                   | yes                | Yes    | Fibroma             | No                  | Positive             | No         |
| 11           | 51          | Married        | 2                   | yes                | Yes    | Fibroma             | Yes                 | Negative             | No         |
| 12           | 47          | Married        | 0                   | no                 | Yes    | Fibroma             | No                  | Positive             | Strong contraction |
| 13           | 49          | Married        | 3                   | yes                | No     | Fibroma             | Yes                 | Negative             | No         |

Positive: decrease in bleeding intensity and duration, and recovery of the menstrual cycle regularity.
Negative: absence of improvement in bleeding intensity and/or duration, and/or persistence of menstrual cycle irregularity.

*Responses to the treatment of thirteen Tunisian women with menometrorrhagia candidate for hysterectomies treated with herbal infusion of 5 g mixed southernwood and pennyroyal plants 3 times a day, every 4h after the first administration by oral rout started from the onset of menstruation during 3*
Baseline characteristics, responses, and side effects of herbal tea infusion consumption on 13 Tunisian women with menometrorrhagia that are candidates for hysterectomy are shown in (Table 1). Participants’ ages ranged between 32 and 53 years. Among them, five were single. A total of nine participants were suffering from clot excretion. The infusion of mixed Pennyroyal and Southernwood extracts was effective in 10 participants; normal and less heavy bleeding flow and disappearance of blood clotting were noted. However, positive response means a decrease in bleeding intensity and duration, and recovery of the menstrual cycle regularity. Negative response corresponds to negative feedback reflecting patient unsatisfaction related to the absence of improvement in bleeding intensity and/or duration, and/or persistence of menstrual cycle irregularity. Negative feedback was noted in three participants after being treated for just two menstrual cycles. Persistence and an increase in bleeding was observed during the study, and they underwent hysterectomy. Strong uterine contraction, as a side effect, concomitant with bleeding cessation after three days of tea infusion consumption was reported by four participants. A total of two participants reported having a headache after using the herbal tea infusion in the first day. No other adverse effects or allergic reactions were observed.

Effect of herbal tea infusion consumption of mixed Pennyroyal and Southernwood extracts on menstrual cycle and the duration and intensity of bleeding

Results of the mean and standard deviation of the number of menstrual bleeding days before and after herbal treatment revealed a significant decline from 11.50 (∓3.17) at baseline to 7.60 (∓2.11) (\(p=0.01\)) to become close to the normal level (Table 2).

Results of the mean PBAC scores of the intensity of bleeding revealed a significant decrease from 219.30 (∓57.03) at baseline to 79.60 (∓31.09) (\(P=0.05\)) (Table 2).

All the participants reported regularization in the menstrual cycle, and improvement in their quality of life.

At the end of the study, herbal tea infusion consumption was found to help 10 participants out of the 13 women avoid the surgical procedure.

Effect of herbal tea infusion of mixed Pennyroyal and Southernwood extracts on blood chemical parameters

Results of the hematologic laboratory evaluation to control anemia before and after herbal treatment are detailed in (Table 2). For each parameter, the mean and the standard deviation were defined. An increase in the mean hemoglobin value (Hgb), from 9.740 (∓3.17) g/dl in baseline to 11.59 (∓1.6) g/dl after 90 days of treatment was noted. However, the parameters did not achieve a statistically significant level.

### Table 2. The effect of herbal tea treatment in clinical and laboratory features at baseline and post treatment.

| Parameter                  | N  | Baseline (±SD) | After herbal treatment (91 days) (±SD) | \(P\) value |
|----------------------------|----|---------------|---------------------------------------|------------|
| **Laboratory features**    |    |               |                                       |            |
| RBC                        | 10 | 4.18 (0.52)   | 4.25 (0.58)                           | 0.576      |
| Hgb g/dl                   | 10 | 9.74 (3.17)   | 11.59 (1.60)                          | 0.169      |
| Hct %                      | 10 | 32.49 (7.09)  | 35.53 (5.53)                          | 0.415      |
| MCV fl                     | 10 | 76.45 (10.73) | 83.63 (9.61)                          | 0.07       |
| MCH pg                     | 10 | 30.40 (4.00)  | 32.56 (3.87)                          | 0.515      |
| PLT *10^3/L                | 10 | 277.70 (92.35)| 242.50 (73.44)                        | 0.139      |
| Fibrinogen g/L             | 10 | 3.64 (1.67)   | 3.4 (0.41)                            | 0.344      |
| *Prothrombin (PT) %        | 10 | 88.27 (10.67) | 87 (7.77)                             | 0.593      |
| **Prothrombin ratio (INR)  | 10 | 1.08 (0.11)   | 1.1 (0.11)                            | 0.715      |
| **Clinical features**      |    |               |                                       |            |
| Menstrual period duration (days) | 10 | 11.50 (3.77) | 7.60 (2.11)                           | \(0.01\)  |
| PBAC Menstrual scores      | 10 | 219.30 (57.03)| 79.70 (31.4)                          | \(0.05\)  |

PBAC: Pictorial blood Assessment Charts, CBC: Complete blood count, RBC: Red blood cells, Hgb: Hemoglobin, Hct: Hematocrit, MCV: Mean corpuscular Volume, MCHC: Mean corpuscular haemoglobin concentration, PLT: Platelet count.

Data are expressed as mean ± standard deviation.
*Data are expressed as median value.
**Wilcoxon Signed Ranks Test, \(P \leq 0.05\).
No significant differences in PT and INR were observed before and after herbal treatment. They were at the normal level.

**Tolerability of mixed Pennyroyal and Southernwood**

The serum creatinine levels at baseline and after three months were in the normal range but the difference was not statistically significant ($p>0.05$) (Table 3). The tea infusion consumed was well-tolerated and participants did not report any serious adverse events.

**Discussion**

In the present preliminary study, the effect of mixed herbal *Pennyroyal* and *Southernwood* tea infusion consumption in a small sample size of women with abnormal uterine bleeding-menometrorrhagia that are candidates for hysterectomy was investigated. Used according to the Tunisian traditional remedies, these two herbs are considered as menstrual regulators and are known to stimulate uterus muscles during painful or strong menstruation. Our results revealed that mixed herbal *Pennyroyal* and *Southernwood* tea infusion consumption was effective in regulating menstrual problems in 10 women. This result is consistent with that of Firdose *et al.* showing that *Pennyroyal* and other species are effective in reducing the number of days of menstrual flow and in normalizing the cycle in patients with infrequent periods of more than 35 days or with scanty flow, both in amount and duration. This positive correlation of the tea infusion of both herbs may be due to their richness in polyphenols and flavonoids, as found in our results. Several studies have demonstrated that polyphenols and flavonoids have an effect on hormone regulation in premenopausal women. A study conducted by Taamalli *et al.* revealed that galloclatechin is a major compound in *Pennyroyal* from the north-west of Tunisia. Galloclatechin, as a phenolic compound, is a flavan-3-ol. According to a study performed by Ko *et al.* this compound has an osteoblastic activity. It is responsible for synthesizing dense cross-linked collagen and proteins. This activity may be related to the side effect mentioned in the results. This ascertainment is supported by the findings of Alekel *et al.* showing that bone mineral content increased by 10.1% in 40 postmenopausal women receiving Isoflavone extract. In their study, Baiceanu *et al.* showed that sinapic acid is a major active polyphenols compound in *Southernwood*. The most interesting effects of sinapic acid are its capacity to influence steroid hormone metabolism and its anti-inflammatory properties. The results of this study were similar to the results of Qaraaty *et al.* in the management of abnormal uterine bleeding-menometrorrhagia by myrtle fruit syrup.

**Study limitations**

This pilot study presents several limitations. First, it seems that there is a lack of information about the chemical composition of the aqueous extract of these two plants. Therefore, this study needs to be completed to identify the major active components of the two plants. Secondly, our study involved a small sample size of participants due to the inclusion criteria and the limited period of time of the study. Thirdly, investigation of the effect of herbal tea infusion would be strengthened if a placebo group was involved.

**Conclusion**

Through this pilot study, we tried to explore a new approach to treat menometrorrhagia in women candidates for hysterectomy, based on tea infusion consumption of a mixture of *Pennyroyal* and *Southernwood* extracts. The study demonstrated that consumption of this tea infusion during three consecutive menstrual cycles helped 10 out of the 13 participants avoid the surgical procedure and improve their quality of life with no major side effects. However, larger clinical trials involving placebo groups, variable doses, and extended treatment periods are required to investigate the effect of *Pennyroyal* and *Southernwood* herbal tea infusion on abnormal uterine bleeding.

**Data availability**

**Underling data**

Zenodo: ‘Therapeutic effect of herbal infusion on Abnormal Uterine Bleeding Extended data’, https://doi.org/10.5281/zenodo.6821462.
This project contains the following underlying data:

- data update version.xlsx (study subjects results (baseline and after three months))
- method of herbs powder preparation.pdf (the method used by subjects to prepare the herbal tea infusion)
- Pictorial Blood Assessment Chart.pdf (method used to assess menstrual blood loss in clinical trials)
- polyphenols and flavonoids content data.xlsx (data of total polyphenols and flavonoids content in the aerial parts (leaves and stems) of Tunisian Pennyroyal and Southernwood extracted by methanol solvent))
- Study protocol.pdf (outlining the study design and describing the objectives and methodology)
- Total polyphenol and flavonoids compounds contents in the aerial parts of Tunisian Mentha Pulegium.pdf (the total polyphenol and flavonoids compounds contents in the aerial parts of Mentha pulegium (pennyroyal) and artemisia abrotanum (southernwood))
- trendstatement_trend_checklist (1).pdf (Guide standardized reporting non randomized controlled trials)

**Reporting guidelines**

Zenodo: CONSORT checklist and flow chart for “Therapeutic effect of herbal infusion on Abnormal Uterine Bleeding: consort flowchart and checklist”: https://doi.org/10.5281/zenodo.6881765.

Data are available under the terms of the Creative Commons Zero “No rights reserved” data waiver (CC0 1.0 Public domain dedication).

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