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The Sanitation of Herbal Substances Using Infrared Radiation as Exemplified by Calendula officinalis L.

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ABSTRACT
The key parameters of raw material sanitation are the temperature and the time. These two factors can be seen as microbiological in sanitation processes because they are responsible for the elimination of microorganisms. The study object is represented by herbal substances of Calendula officinalis L. The material was dried in natural conditions and contaminated with yeast and mold fungi, colibacilli and aerobic bacteria. The reduction of bacterial content in dry herbal material was achieved by applying infrared radiation. The results show that the raw material made of Calendula officinalis flowers shall be heat-treated at temperatures of 70–80° with 30–60-second exposure and exposure rate of 7,5 kW/m^2. Heating the calendula material up to 80° makes the extractive substance reduce by 10 % of the original value. If the exposure rate is at 2,5 and 5 kW/m^2, the sanitation is not efficient. At the exposure rate of 7,5 kW/m^2, the sanitation effect was achieved in three trials out of four. The results of the research show that the suggested energy-saving sanitation method allows reducing the microbial content of the contaminated herbal substance of Calendula officinalis til the normal level set out in the State Pharmacopoeia and the disease control authorities. It also allows for the production of medicinal substances of higher quality and optimum composition of extractive substances from 38,8 % (control value) to 35 % after the treatment.

Keywords: *sanitation, crude medicine, infrared radiation, Calendula officinalis*

1. INTRODUCTION

The microbial cleanliness of medical plants is very important for chemical and pharmaceutical production. One way of achieving it is by creating new methods and means of reducing the bacterial content of non-sterile medical plants to the levels set out in scientific and technical documentation. Pharmacopoeial requirements on microbial cleanliness are applied to the plant materials.

Key parameters of the sanitation process include the temperature that shall be maintained in the sterilizer and the time during which the material is heated. These two factors can be seen as microbiological in sanitation processes because they are responsible for the elimination of microorganisms. However, it is impossible to talk about the lethal time without taking into consideration the sanitation temperature, just as it is impossible to talk about the temperature not taking into consideration the duration of the treatment. Lethal conditions for certain bacteria species cannot be determined by temperature alone. They can only be described by a specific combination of lethal temperature and time.

The objective of this research is calculating the perfect duration of infrared radiation exposure (IR exposure) for the material of *Calendula officinalis* L. to achieve the necessary levels of microbiological cleanliness.

Calendula officinalis is an annual herbaceous plant of the family *Asteraceae*. It has a stalk of 60–90 cm and a taproot. Leaves alternate, lower leaves are oblong, egg-shaped and petiolate; upper leaves are oblong or lanceolate, and sessile. Athodium blooms reach 3 to 8 cm in diameter, external florets are semifloscule, orange or yellow, and pistillate; internal florets are tubular, reddish or reddish-orange, staminate. Achene fruits. The weight of 1000 seeds is between 6.8 and 14.9 grams.

Calendula officinalis is widely cultivated in Russia, and it is a valuable medical and ornamental plant. Its athodiums and semifloscules are harvested as raw material [1]. The pharmacological activity profile of the *Calendula officinalis* flowers is quite large. They have antiinflammatory, regenerating, antimicrobial, bile-expelling, and expectorant properties due to the presence of various bioactive substances, such as carotenoids, flavonoids (kaempferol glycoside, quercetin and isorhamnetin), saponins, etc. Calendula products have a soothing effect, reduce arterial tension and reflex excitability, increase the amplitude of cardiac contraction and make them slower. When applied topically, calendula products hasten tissue regeneration and reduce inflammation. This species also has some antibacterial properties, especially against staphylococci and streptococci [2, 3].

2. METHODS AND MATERIALS

The study object was represented by various kinds of raw material made of medical plants. The results are presented as exemplified by *Calendula officinalis* [4, 5]. The raw material was dried in natural conditions and contaminated with yeast and mold fungi (over 105/g), colibacilli (104/g) and aerobic bacteria (over 107/g).

The research was carried out taking into consideration the conventional and contemporary methods of food dehydration [6–9].

Due to the application of the mathematical planning method for the experiment, we received the following optimum heat treatment parameters for the given object: infrared exposure duration, treatment temperature, infrared exposure intensity, and layer thickness.

In order to measure electrical parameters, such as the voltage, the current, the power and power consumption, we used both separate electric meters and K-505 elecromeasuring unit. The temperature of the heat-treated material was measured using a more delicate chromel-copel thermocouple (with the response time of 2 seconds) 0.5 mm in diameter that was connected to a recording potentiometer PP-01. We also took into consideration the intrinsic error of the thermocouple (no more than ±0.1 % of the reading at 23 °C), and the temperature effect (no more than ± 0.01 % per each degree of the deviation from 23 °C). We set the temperature of the radiating unit using a voltage controller, measured the radiating unit heating with thermocouples connected to the PP-63 potentiometer, and we monitored it using OPPIR-09 optical pyrometer. The irradiancy of the medical substance was measured with an albedometer connected to a GSA galvanometer.

The extractive substance content assessment and the statistical analysis of the experimental data were carried out using common methods.

In order to reduce the microbial content of the dry raw material made of *Calendula officinalis*, we used an industrial-grade infrared unit (IR unit) that we developed and produced ourselves (Fig. 1).

The unit consists of a radiator, a drying chamber, a radial fan and a control panel with start-up and metering instrumentation. The laboratory unit is designed so that the radiator can be replaced easily. As radiating units, we used factory-made equipment, such as tubular heating elements, silicon carbide heaters, ceramic radiators, lamp radiators like ZS, IKZ, IKZK, KI and KG, and laboratory-made radiators based on nichrome and micacious laminate electric heaters. We placed four flat infrared radiators in the heat treatment chamber, sized 1x1 meters. The overall power of all of the radiators made up 30 kW. The maximum dehydration output reached 30 kg/h. Between the two flat radiators, from one to five material cassettes can be placed, and in the chamber, from 3 to 15 or even more.

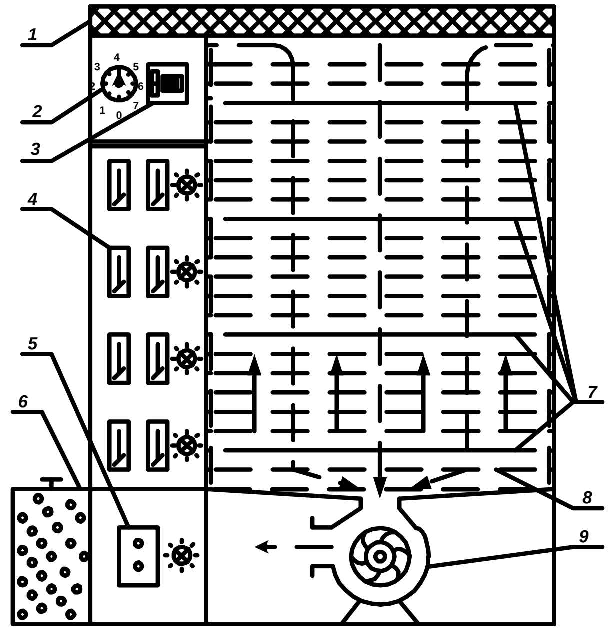

Figure 1 IR unit structural layout: 1 – deflector; 2 – power level switch; 3 – cycle timer; 4 – IR radiator control panel; 5 – radial fan control panel; 6 – power level knob; 7 – IR radiators; 8 – retiform cassette with the material; 9 – radial fan; ←--- – airflow; ← – material cassette movement direction. The section containing the description of methods and materials used in the article

The methods for calculating the material layer thickness in a cassette and the layerwise movement of cassettes depending on the original moisture level are described in the works of V. N. Karpov from the standpoint of the Bouguer law [10].

A set of controlling equipment allows adjusting the power of the three-phase infrared radiation using three methods: incremental power adjustment using automatic switches and a seven-position controller; smooth control using controlled thyristors; a combination of the incremental and the smooth methods.

Infrared radiation control for the automation of the experiment and providing intermittent radiation was achieved using electronic and electromechanical devices. Some cycle timer-based devices were developed in order to maintain intermittent radiation mode, and a program temperature controller Termodat 14 was used for the smooth adjustment of the power for three-phase infrared radiation.

3. RESULTS

Concerning crude medicines made of plants, we studied the impact infrared radiation has on the microflora and the preservation of extractive substances in the treated material. All of the samples underwent microbiological and phytochemical analyses according to pharmacopoeial articles in the Certification Center for the Medicine Quality Assurance at the Pharmaceutical and Medicine Production Committee of the Administration of the Irkutsk region and the Laboratory of the Irkutsk State Center of the State Committee for Sanitary and Epidemiological Oversight.

The infrared sanitation of plant material has a thermal influence on microorganisms and suppresses them. Applying vibrations to the material promotes its smoother treatment due to the constant rotation of the radiated surface [11–12].

During the research, we studied the effects of infrared radiation intensity and duration on the microbial content value, and the quality and the quantity of the extractive substances in the crude drugs made of *Calendula officinalis* (Table 1). During the experiments, we applied various exposure times (15 to 60 seconds), power density values (2.5 to 7.5 kW/m^2) and temperatures (20 to 80°).

The results show that when the flowers of *Calendula officinalis* were heat-treated at 20–66°, the content of extractive substances is between 38.8 and 35.6 %, and material sanitation was only achieved in one sequence out of three (at a temperature of 60–66°). This leads us to the conclusion that if the temperature of the *Calendula officinalis* material is between 20° and 66°, the sanitation procedures are inefficient.

When the material was heated up to 70–80° and its exposure time was 30–60 seconds, the sanitation proved to be effective in all of the samples. Heating the *Calendula officinalis* material up to 80° makes the extractive substance reduce by 10 % of the original value.

If the exposure rate is at 2,5 and 5 kW/m^2, the sanitation is not efficient. At the exposure rate of 7,5 kW/m^2, the sanitation effect was achieved in three trials out of four.

Table 1 IR radiation parameter impact on the microbial content and extractive substance content of crude medicines made of *Calendula officinalis* (for the ground material layer thickness of 7 mm)

Option #	**Treatment parameters**			**Microbial contamination|replication**			**Extractive substance content after treatment in %**
	Exposure in seconds	**Power density in kW/m^2**	**The heating temperature in °C**	**1**	**2**	**3**	
1	60	7,5	80±2.10	-	-	-	35.0±0.33
2	45	7,5	76±1.00	-	-	-	35.1±0.56
3	30	7,5	70±0.54	-	-	-	35.5±0.05
4	15	7.5	66±0.38	+	+	-	35.6±0.14
5	60	5.0	60±2.60	+	+	-	36.2±0.40
6	45	5.0	55±1.15	+	+	+	36.9±0.06
7	30	5.0	50±0.40	+	+	+	37.1±0.11
8	15	5.0	45±0.66	+	+	+	37.3±0.32
9	60	2.5	40±0.48	+	+	+	38.0±0.06
10	45	2.5	30±1.01	+	+	+	38.6±0.09
11	30	2.5	24±1.50	+	+	+	38.8±0.65
12	15	2.5	20±0.55	+	+	+	38.8± 0.33
Control	0	0	0	+	+	+	38.8± 0.74

Table 2 The impact of IR radiation on the quality of crude medicine material of *Calendula officinalis*

Quality indicator	**Quality requirements**	**Results**
External features	Athodiums are intact or partially slough without flower spikes	Compliant
Microscopy	Compliant with regulatory documents	Compliant
Quantitative indicators. Ground (shredded) material:	Compliant with regulatory documents	Compliant
Moisture	Up to 14 %	5.75 %
Ash total	Up to 11 %	3.08 %
Ash insoluble in 10% hydrochloric acid solution	Up to 5 %	0.98 %
Flower spike and fruit bits	Up to 10 %	4.78 %
Stem and leaf bits	Up to 3 %	None
Flower spike remains	Up to 6 %	0.32 %
Slough athodiums	Up to 20 %	15 %
Discolored (brown) athodiums	Up to 3 %	1,29 %
Particles that cannot pass through 0.18 mm sieve	Up to 10 %	1.81 %
Organic impurities	Up to 0.5 %	None
Mineral impurities	Up to 0.5 %	None
Extractive substances	Starting from 35 %	35 %
Microbial cleanliness		
- Aerobic bacteria	Up to 10^7	$9*10^6$
- Yeast and mold fungi	Up to 10^4	10^4
- *Escherichia coli*	None	None
- *Salmonella*	None	None
- *Staphiylococcus aureus*	None	None
Other colibacilli	Up to 10^2	10^2
Radioactivity	Compliant with regulatory documents	Compliant
Packaging	Compliant with regulatory documents	30 grams per paper bag, bags placed in cardboard packs
Marking	Compliant with regulatory documents	Compliant

All of the samples underwent microbiological and phytochemical analyses according to pharmacopoeial articles in the Certification Center for the Medicine Quality Assurance at the Pharmaceutical and Medicine Production Committee of the Administration of the Irkutsk region and the Laboratory of the Irkutsk State Center of the State Committee for Sanitary and Epidemiological Oversight (Table 2).

4. CONCLUSION

The results show that the raw material made of *Calendula officinalis* flowers shall be heat-treated at temperatures of 70–80° with 30–60-second exposure and exposure rate of 7,5 kW/m^2.

The suggested energy-saving sanitation method using an industrial-grade infrared unit allows reducing the microbial content of the herbal substance of *Calendula officinalis* to the normal level set out in the State Pharmacopoeia and the disease control authorities of Russia. It also allows for the production of substances of higher quality and optimum composition of extractive substances from 38,8 % (control value) to 35 % after the treatment.

The studies infrared radiation method is efficient for sanitizing other medical plants, including Hedysarum alpinum L., Origanum vulgare L., Symphytum officinale L., Symphytum caucasicum Bieb., Thymus serpyllum L., Tussilago farfara L., Urtica dioica L.etc. Sanitation modes are specific for different types of material and they depend on the level of contamination. The results of the research conducted show that when sanitizing crude medicine material contaminated with aerobic bacteria, yeast and mold fungi, colibacilli, the temperature on the surface of the material in most cases should be between 55 and 80° with the exposure time of 15–60 seconds and the IR intensity of 5–7.5 kW/m. These parameters promote sanitation effects and allow preserving the optimum amount of bioactive substances in the material.

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