ARTICLE

Cite This:J. of Pharmaceu. and Formul. yyyy, vol, page

Sciforce

Journal of Pharmaceuticals and Formulations

Journal homepage: www.sciforce.org

Manufacturing and formulation of topical products based on the Artemisia annua herb as active ingredient in the forms of Solid and Liquid Soaps and Cream

Mohammadreza Gholibeikian[1*],Amirhosein Firouzi[2], Masoumeh Lari[3], Mahdi alaeddini[4], Saeid Taherkhani[5], Afsaneh oshli[6], Somayeh faraji[7]

[1]*Department of Organic Chemistry, Faculty of Chemistry, University of Kashan, Kashan, Iran.*

[2]*Department of chemistry, Faculty of science, Golestan university, Gorgan, Iran.*

[3]*Alzahra University, Faculty of Economics, Iran.*

[4]*Homapharmed pharmaceutical company, Department of research and development, Iran.*

[5]*Faculty of Chemistry, Azad University, Tehran North Branch, Iran.*

[6]*Department of Chemical Engineering, University of science & Technology, Iran.*

[7]*Faculty of new sciences and technology, University of Tehran, Iran.*

ARTICLE INFO

Article history:
Received 20240122
Received in revised form
Accepted 20240128
Available online 20240223

Keywords:
Artemisia annua,
Hydroalcoholic method,
Ingredient,
Anti-inflammations,
Skin

ABSTRACT

The purpose of this work is to make herbal topical products of solid and liquid soaps and cream based on the active ingredient of Artemisia annua herb. The herb used in this work is the dry extract of Artemisia annua herb, which was extracted by hydroalcoholic method.Physical and chemical analysis, identification test of Artemisia extract as active ingredient and auxiliary materials and also non-sensitizing test of these products have been done on the rabbit's skin that did not show no sensitivity and inflammation. It seems that herbal products based on of Artemisia extract as effective products have a useful and promising role as skin anti-inflammations on the skin of the body.

.

∗Corresponding author; e-mail: mgholibeikian@gmail.com

Introduction

The Artemisia genus belongs to the Asteraceae family and the Anthemideae tribe, which has about 400 species. Most of its species have a specific smell and taste, which is caused by the monoterpene and sesquiterpene compounds in them called artemisin. This medicinal herb contains a substance called santonin, which has been considered the most famous anti-helminthic drug for a long time (1-3). Artemisia herb is considered a valuable herb in Chinese medicine. The Chinese consider this herb as the prince of medicinal herbs that can affect respiratory infections or improve the respiratory system and relieve fever. Also, this herb can have an effect on the skin in addition to improving the respiratory and pulmonary system and lead to the improvement and treatment of eczema, psoriasis, itching and inflammation, acne, freckles and skin infections.

In addition to anthelmintic activity, many biological activities such as microbicide, antifungal, virucidal, antiparasitic, and also have analgesic and antioxidant properties have been proven (4-9). One of the species of this herb is the mountain wormwood, which according to the studies has flavonoid, santonin, coumarin compounds, bitter substances and volatile essential oil (2).

Since this herb has many biological properties, including antimicrobial, anti-inflammatory, it seems that it can be effective in the wound healing process and increase the speed of wound healing. In this study, the manufacture and formulation of topical products containing dry extract of Artemisia herbal have been discussed. Artemisia annua herb has great antiseptic properties and is used to treat fungal skin infections and disinfect wounds. This herb is used in the pharmaceutical industry to make

antiseptic lotions for skin fungi. It is also used as a lotion to strengthen and prevent skin wrinkles. The use of Artemisia drops cures ringworm of the nails and has anti-inflammatory effects and is effective in the healing process of the wound and its rapid recovery.

There are several possible reasons for how Artemisia annua herb accelerates wound healing. Preventing wound infection and treating the wound with antibiotics accelerates the wound healing process (4). It has also been reported that the herb has antimicrobial effects (4, 5).

The analysis of the essential oil of Artemisia annua herb has shown that this herb contains various compounds such as simene, sabinene, cineol, linalool, eugenol, borneol, farnesol, ester and other compounds. Artemisia annua contains various chemical compounds such as santonin compounds, coumarin compounds, as well as flavonoids such as quercetin and rutenoid. The aerial part and especially the leaves of this herb contain cineole and thujone essential compounds. This herb contains substances such as triterpenes, adenine, amyrin, myrcene, squaline, nerol, vulgarin, myrcene, anthelmintic substances containing artemisin, and these substances have given special therapeutic properties to the medicinal herb. The presence of borneol, cimen and cineol in the essential oil of the herb has a germicidal effect on Staphylococcus aureus, Streptococcus and Escherichia coli, which are the most important causes of wound infection (1).

It is possible that this herb accelerates the wound healing process by preventing infection and the growth of microorganisms. Anti-inflammatory and antioxidant activity has been reported in many types of herbs, including Artemisia annua (2), and the modulation of inflammation and the use of antioxidants accelerate wound healing. Antioxidants in the herb can protect the skin from sunlight and reduce the risk of skin cancer (10, 11). Therefore, it can be concluded that the hydroalcoholic extract of Artemisia annua herb probably improved the wound healing process by reducing inflammation, scavenging free radicals and oxidant substances. In this work, Artemisia topical products were made in the form of soap, hand wash and wound healing cream. The herbal cream is 60 grams in the form of a tube and contains active ingredient solid artemisia extract, wheat germ oil, jojoba oil, avocado oil and pomegranate seed oil. Physico-chemical studies, stability studies of Artemisia topical products have been reviewed and analyzed under the supervision of the collaborating laboratory of food and drug (Detergent and Cosmetic Industry Association of Iran) and these products contains analysis sheets from the collaborating laboratory. The oils in the cream have been analyzed and checked by the research institute of medicinal plants of Jihad University, which is a laboratory cooperating with food and drug, and all the oils have analysis sheets from the research institute of medicinal plants of Jihad University. Also, animal studies of Artemisia topical products in this work on rabbits have been conducted for 14 days at the Department of Toxicology, Shahid Beheshti Faculty of Pharmacy, Tehran, and it was observed that during the study for 14 days on rabbit skin, there were no side effects such as inflammation and redness with topical products containing herbs extract of Artemisia annua. All of the topical products made from the active ingredientof Artemisia annua herb have an analysis sheets and animal certificates.

2. Materials and Methods

castor oil, coconut oil, olive oil, palm oil, citric acid, (sodium lauryl sulfate (SLS), sodium lauryl ether sulfate (SLES), Glycerin, Sodium EDTA, Methyl isothiazolinone, Gzantan Gam, Polyquaternium 7, Vitamin B5, Sodium chloride, distilled water, wheat germ oil, jojoba oil, avocado oil, pomegranate seed oil, Orand® base cream, vitamin c, Himalayan salt, Tween 80, Sodium benzoate and Potassium sorbate, were purchased from Sadra Pajohesh company.

Experimental

The Preparation of Artemisinin annua herb

The Artemisia annua herb is prepared by a botanist of the Research Institute of Medicinal Plants of Jihad University. After identifying the herbarium code, the herb is dried in the shade and at room temperature and extracted using 70% ethanol and percolation method within 72 hours, and then the remaining solvent in the extract is completely removed using a rotary evaporator. Then, with a spray dryer, extract powder is obtained for use in formulation.

Standardization:

In order to standardize the extract, the indicator substance of Artemisia annua herb named Artemisinin and total phenolic index of the extract are measured.

Formulation of topical products of Artemisia annua herb:

The Making of Artemisia solid soap by cold method:

Transfer the amount of 30% castor oil, 25% coconut oil, 25% olive oil, 20% palm oil to a glass container so that all the oils are well mixed together and pour all the oils. Melt all the oils at a temperature of 60℃. Dissolve soda in water and continue dissolving until all undissolved particles of soda in water are reached. Filter the dissolved solution in water. Add citric acid to the soap to neutralize the excess soda in the water. Add the dry extract obtained from the Artemisia plant by the hydro alcoholic method to the soap mixture. Stir the soap mixture well so that the dry Artemisia extract is well distributed in the texture of the soap (Fig1).

The Making of Artemisia herbal liquid soap per 100 grams:

Transfer 75 grams of water to the mixer tank. Add 8 grams of sodium lauryl sulfate to the tank of the device at a low speed so that it does not foam, and after the complete dissolution of sodium lauryl sulfate in water, add 5 grams of sodium lauryl ether sulfate to the tank of the device and proceed until complete dissolution. Then add 5 grams of glycerin to the mixture, followed by 0.05 grams of sodium edta. Then add 5 ml of water in a glass beaker and then add 0.1 g of preservative, 0.5 g of humectant or emollient vitamin B5, 0.5 g of Artemisia annua extract. Add the solution to the mixing tank. Then add 0.5 grams of thickener (xanthan gum) to the tank of the machine and continue stirring until all the xanthan gum dissolves in the mixture and the mixture becomes viscous. Then dissolve the polyquaternium 7 in a small amount of water and then add to the mixture and continue the stirring operation and then let the mixture rest for 24 hours until the foam of the mixture disappears. Then slowly add sodium chloride to the tank mixture to obtain a viscous mixture (Table 1, Fig 2).

Formulation of the cream based on Artemisia annua herb as active ingredient:

The Orand® base cream was stirred for 30 minutes by a mechanical stirrer. wheat germ oil, jojoba oil, avocado oil and pomegranate seed oil were added to the base cream. Tween 80 as an emulsifier was added to base cream. The base cream was stirred for 20 minutes at room temperature. vitamin cwas dissolved in Distilled water, then Himalayan salt added to the solution. The solution was slowly added to the cream base. The base cream was stirred for 15 minutes at room temperature.The first, Sodium benzoate as preservative was added to the mixture. The base cream was stirred for 10 minutes at room temperature. The second, Potassium sorbate as preservative was added to the mixture. The base cream was stirred for 10 minutes at room temperature. Artemisia annua solid extract was added to the cream mixture as an active ingredient.Table 1. shows the percentage of each component for making Artemisia annua cream (Table 2, Fig 3).

Methodology Acute Dermal Irritation/Corrosion for products based on Artemisia annua herb:

Good Laboratory Practice compliance statement: This study was conducted in compliance with current OECD Good Laboratory Practices Standards. Study Title: Acute Dermal Irritation Study in Rabbits. Irritation Test: In Vivo. Test Animal: Male Albino Rabbit. Test Guideline: Based on OECD Guideline for the Testing of Chemicals. Performing Laboratory: Quality Control Laboratory, Department of Pharmaceutics, school of pharmacy, Shahid Beheshti University of Medical Sciences, 2660 Vali-e-Asr Ave., Tehran, Iran. Results: The sample showed no irritation.

The aim of this study was to evaluate the skin irritation/ corrosive potential and the reversibility of dermal effects of the topical products based on the Artemisia annua herb as active ingredient in the forms of Solid and Liquid Soaps and Cream following a 4-hour dermal exposure in albino rabbit. The results of the study were used to determine the approximate toxicity classification. Dermal erythema and edema are evaluated and scored at approximately 60 minutes, and 24, 48, and 72 hours following the removal of the test substance at the end of the 4-hour exposure in rabbit. The reversibility of any dermal effects is assessed for up to 14 days. If necessary. Dermal effects are quantified according to the Draize scale (Table 3) (12). In this study, one male adult New Zeland albino rabbit was obtained from the Animal House of School of Pharmacy, Shahid Beheshti University of Medical Sciences, Tehran, Iran. The rabbit was housed singly in stainless steel, wire-mesh cages and kept under standard animal laboratory conditions, 12 h of light and dark cycles, at controlled temperature 22±2□, with a relative humidity of 50±5%, and free access to food and water. Around 24 hours prior to treatment, the four of male New Zealand White rabbit was closely shaved to exposure the skin (approximately 2.5×2.5) from scapular to the lumbar region of the back.

Solid Soap, Liquid Soap and Cream was applied as a single dermal dose to the shaved intact skin of the rabbit. The application area was covered with a 2-ply gauze square which was held in place with non-irritating tape. The rabbit was returned to cage after treatment. No other substances were tested on the rabbit. The rabbit was exposed to the test substance for 4 hours after which the test substance was removed. Test sites were evaluated and scored by the method of Draize (Table 3) for erythema, edema, and other evidence of dermal effects approximately 60 minutes, and 24, 48, and 72 hours after test substance removal. Additionally, the rabbit was examined for clinical signs of toxicity at each observation period. The test substance appeared to be stable under the conditions of the study. No evidence of instability, such as a change in color or physical state, was observed. For data analysis and interpretation of the results, values for each lesion (erythema and edeme) were calculated from numerical scores obtained at the 60 minutes, 24, 48, and 72 hour observations (Table 4). The final results were calculated as a primary irritation index (PII). The mean scores were summed and averaged to obtain the primary irritation index (Table 5). No dermal irritation, erythema and edema was observed at 60 minutes, and 72 hours after removal of the substance in the rabbit. No clinical signs were observed. The dermal scores from rabbit with respect to observation time are presented in Table 6.

3-Result:

To make the herbal Artemisia annua cream with the oils used in the cream, tween as an emulsifier with these oils and with the ratio of water achieves a soft creamy texture. As shows figure 4&5, fatty acids palmitic acid, oleic acid, linoleic acid, stearic acid have the highest percentage of abundance in herbal cream.

Fig 1: Artemisia annua herb solid soap with Brand PACOLOREN

Fig 2: Artemisia annua herb liquid soap with Name Artemsan and Brand PACOLOREN

Fig 3: Artemisia annua herb Cream with Name Hermisafe and Brand PACOLOREN

PROFESSIONAL CENTER OF ANALYSIS

Attachment

Lab Number	99-OTH-200-432

Gas chromatography–mass spectrometry (GC/MS) Analysis

No	RT	%	Components	KI	Type
1	17.63	10.28	Cyclohexadecane	1891	Other
2	18.04	10.66	Palmitic acid, methyl ester	1932	C16:0
3	19.71	25.53	Linoleic acid, methyl ester	2053	C18:2(n-6)
4	19.75	30.49	Oleic acid, methyl ester	2056	C18:1(n-9)
5	19.97	20.61	Stearic acid, methyl ester	2068	C18:0
6	21.73	0.64	Arachidic acid methyl ester	2268	C20:0
7	23.84	0.99	Hexacosane	2600	Other
8	24.62	0.81	Heptacosane	2646	Other
		100.00	Total Identified		

Monoterpene Hydrocarbons MH *Oxygenated Monoterpenes MO*
Sesquiterpene Hydrocarbons SH *Oxygenated Sesquiterpenes* SO

Operator : Z.Tarassoli
Sample : OTH-200/432
Misc :
ALS Vial : 1 Sample Multiplier: 1

Search Libraries: C:\Database\NIST05a.L Minimum Quality: 0
C:\Database\wiley7n.l Minimum Quality: 0

Unknown Spectrum: Apex
Integration Events: Chemstation Integrator - autoint1.e

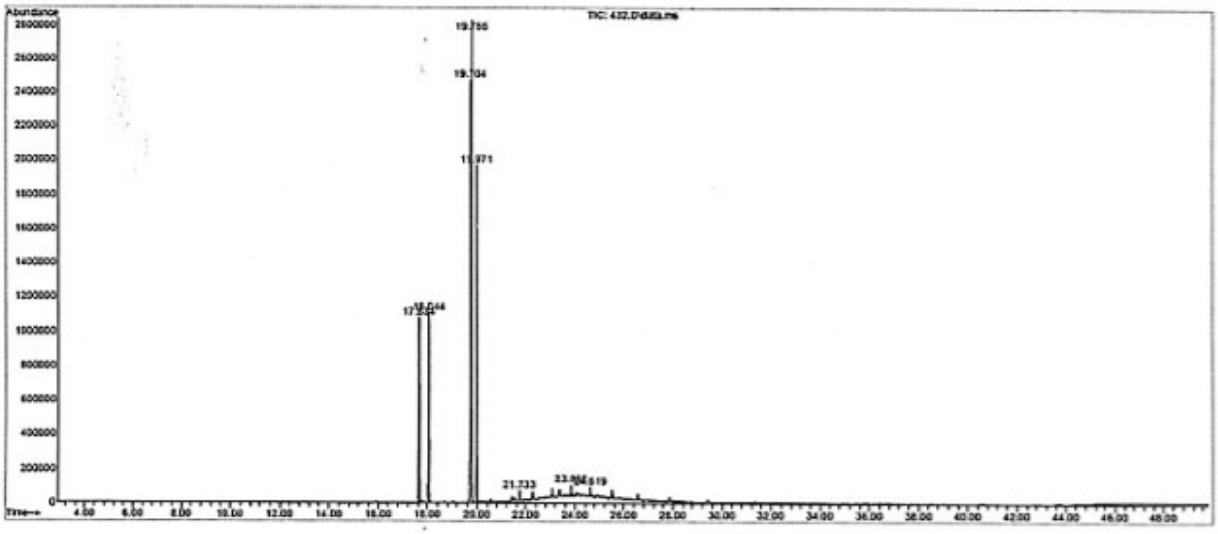


Fig 4. Fatty acids of the first category

PROFESSIONAL CENTER OF ANALYSIS

Attachment

Lab Number	99-OTH-200-433

Gas chromatography–mass spectrometry (GC/MS) Analysis

No	RT	%	Components	KI	Type
1	18.05	9.33	**Palmitic acid, methyl ester**	1932	**C16:0**
2	19.74	38.94	**Linoleic acid, methyl ester**	2055	**C18:2(n-6)**
3	19.79	35.59	**Oleic acid, methyl ester**	2058	**C18:1(n-9)**
4	19.98	12.94	**Stearic acid, methyl ester**	2068	**C18:0**
5	21.53	0.48	11-Eicosenoic acid, methyl ester	2256	**C20:1(n-9)**
6	21.74	0.76	Arachidic acid methyl ester	2268	C20:0
7	23.36	0.86	Behenic acid, methyl ester	2469	C22:0
9	24.95	0.28	Lignoceric acid methyl ester	2667	**C24:0**
		99.19	**Total Identified**		

Monoterpene Hydrocarbons MH *Oxygenated Monoterpenes MO*
Sesquiterpene Hydrocarbons SH *Oxygenated Sesquiterpenes* SO

Operator : Z.Tarassoli
Sample : OTH-200/433
Misc :
ALS Vial : 1 Sample Multiplier: 1

Search Libraries: C:\Database\NIST05a.L Minimum Quality: 0
C:\Database\wiley7n.l Minimum Quality: 0

Unknown Spectrum: Apex
Integration Events: Chemstation Integrator - autoint1.e

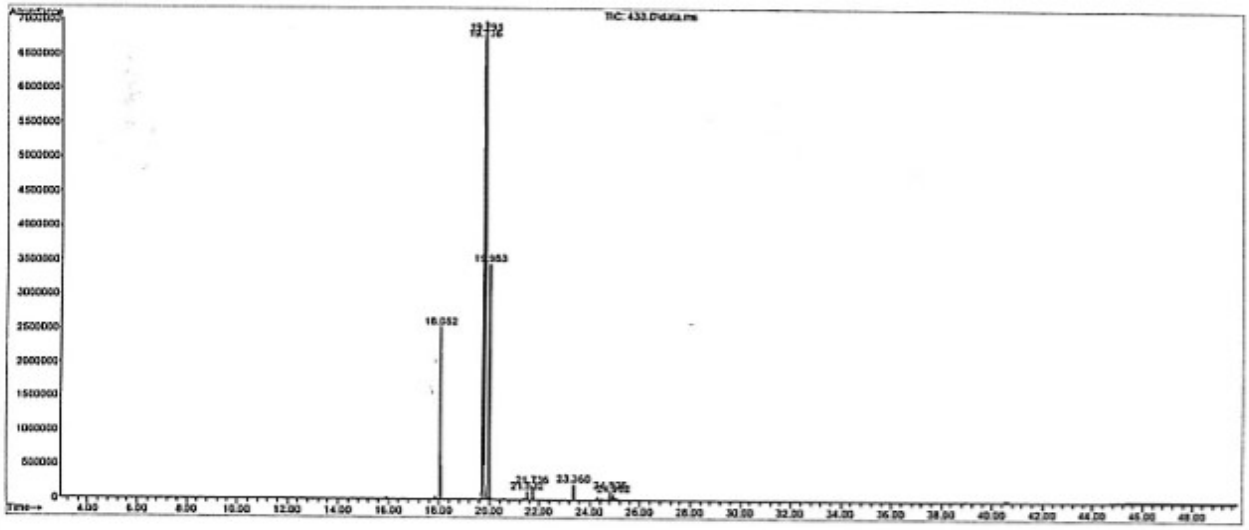


Fig 5. Fatty acids of the second category

Table 1. percentage of each component for making Artemisia annua liquid Soap.

Artemsan Liquid Soap	
Chemical Name	Amount (gram)
(sodium lauryl sulfate (SLS)	8
sodium lauryl ether sulfate (SLES) 100% or 70%	5
Glycerin (humectant or emollient)	5
Sodium EDTA (Na_2EDTA)	0.05
Methyl isothiazolinone (preservative)	0.1
Gzantan Gam (thickener)	0.5
Polyquaternium 7 or 10 (hemooctant)	0.3
Vitamin B5 (humectant or emollient)	0.5
Sodium chloride (viscosifier or thickener)	3
Artemisia annua Extract	0.5
Distilled water	80

Table 2. percentage of each component for making Artemisia annua cream.

Product Name: Artemisia annua cream		
Entry	Material Name	Amount of material
1	Artemisia annua solid extract	1% base cream by weight
2	wheat germ oil	2 grams in the base cream
3	jojoba oil	2 grams in the base cream
4	avocado oil	2 grams in the base cream
5	pomegranate seed oil	2 grams in the base cream
6	Orand® base cream	80 grams of cream base
7	vitamin c	1.5% base cream
8	Distilled water	2 grams in the base cream
9	Himalayan salt / Himalayan salt should be dissolved in 2 cc of warm distilled water and vitamin C	1.5% Himalayan salt in the base cream
10	Tween 80 as an emulsifier	3.9 grams in the base cream
11	Sodium benzoate	0.05 grams in the base cream
12	Potassium sorbate	0.05 grams in the base cream

Table 3. Dermal effects quantified according to the Draize scale

scores	Arrhythmia rate
0	no erythema
1	Very mild erythema
2	Specific arrhythmia
3	Moderate to severe erythema
4	Severe erythema to scar formation

Table 4: Comparison of the test group and the control group on the rabbit body with topical products

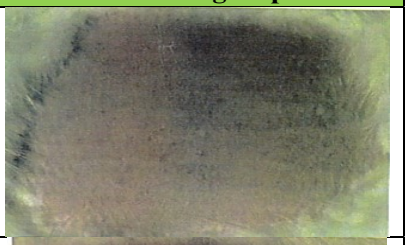
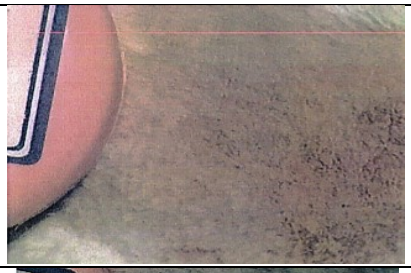
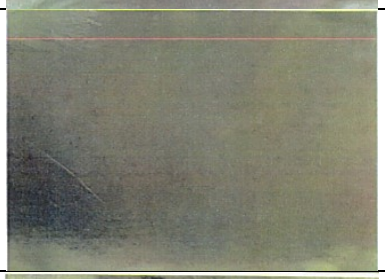
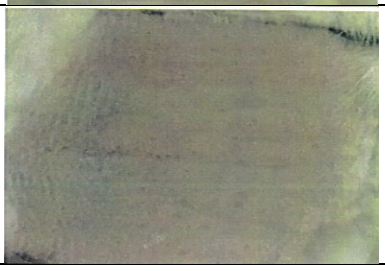
Time	Test group	Control group
After 24 hr		
After 60min		
After 48 hr		
After 72 hr		
After 14 day		

Table 5: Descriptive rating for mean primary irritation index (PII).

Primary irritation index (PII)	Classification
0	Negligible
0<PII≤2	Slight

0<PII≤5	Moderate
5<PII	Severe

Table 6: Dermal response observed in Rabbit

	Evaluation after removal of test substance				
	0 minutes	60 minutes	24 hours	48 hours	72 hours
Erythema	0	0	0	0	0
Edema	0	0	0	0	0

4- Conclusion:

Solid and Liquid Soaps and Cream containing the active ingredient Artemisia annua herb can be good products as skin anti-inflammations on the skin of the body. there was not no dermal irritation in Irritation Test in Male Albino Rabbit. No clinical signs were observed. Under the conditions of this study, Solid and Liquid Soaps and Cream, produced no erythema and no edema when applied to the skin of rabbit. Solid Soap, Liquid Soap and Cream showed a negligible irritation response.

Acknowledgements:

Arezo Lari, Fatemeh Hosseinnia and Zahra Mirhashemi and Sepideh Moafi played an important role in production and construction. I thank and appreciate all these dear ones.

References:

[1]. Allahtavakoli M, Khaksari Haddad M, AssarSh. Comparison of topical application of Mummify and Phenytoin cream on skinwound healing in rat. J Babol Univ Med Sci**2003**; 5(18): 7-13.

[2]. Dinani NJ, Asgary A, Madani H, NaderiG, Mahzoni P. Hypocholesterolemic and antiatherosclerotic effect of artemisia aucheriin hypercholesterolemic rabbits. Pak J Pharm Sci **2010**; 23(3): 321-325.

[3]. Zargari A. Herbal plants. 6th ed, Tehran: Tehran University Publisher; **1999**. P 63.

[4]. Hakimi Maybodi MH, Afkhami Aghdaee M, Mijalili BF. An investigation into biological activities of Artemisia Persia's essential oil. Pajoohesh and Sazandegi**2003**; 16(61): 2-5.

[5]. Shakarami J, kamali K, Moharrmi Pour S, Meshkatossadat MH. Fumigant toxicity and repellency of essential oil of Artemisia aucheri on four species of stored pest. Pests and Diseases of Plants **2003**; 71(2): 61-76.

[6]. Han J, Zhao YL, Shan LM, Huang FJ, Xiao XH. An experiment on standardized cell culture assay in assessing the activities of Composite Artemisia Capillaris Tablets against hepatitis B virus replication in vitro.

[7]. Chin J Integr Med **2005**; 11(1): 54-56.

[8]. Willcox M, Rasoanaivo P, Sharma VP, Bodeker G. Comment on: Randomized controlled trial of a traditional preparation of Artemisia annua L. (Annual Wormwood) in the treatment of malaria. Trans R Soc Trop Med Hyg**2004**; 98(12): 755-756.

[9]. Said Fernández S, Ramos Guerra MC, Mata Cárdenas BD, Vargas Villarreal J, Villarreal Treviño L. In vitro antiprotozoal activity of the leaves of Artemisia ludoviciana. Fitoterapia**2005**; 76(5): 466-468.

[10]. Sadeghi Fard H, Zareian P. Survey on analgesic effect of Hydro-alcoholic extract of Artemisia aucheri in two models of acute and chronic pain. J Kordestan Univ Med Sci**2008**; 13(4): 30-36.

[11]. Khaksari M, Mardani M, Rezaee zadeh A. Effect of oral ascorbic acid on histological indices of wound healing in chronic diabetic rat. J Babol Univ Med Sci **2005**; 8(29): 12-21.

[12]. Jarrahi M, Zahedi M, Ajorlu M. Effect of Aloe barbadensis Miller on skin wound healing in rat. J Gorgan Uni Med Sci **2009**; 11(29): 13-17.

[13]. Draize, J.H., "Dermal Toxicity". Appraisal of the safety of chemicals in foods, Drugs and Cosmetics. The Editorial Committee of the Association of Food and Drug officials of the United States, Austin, Texas. **1959**.pp.45-59.