

Manufacturing And Formulation of Topical Products Based on The Artemisia Annu Herb as Active Ingredient in The Forms of Solid and Liquid Soaps and Cream

Mohammadreza Gholibeikian^{1*}, Amirhosein Firouzi², Masoumeh Lari³, Mahdi alaeddini⁴, Saeid Taherkhani⁵, Afsaneh oshli⁶ and Somayeh faraji⁷

¹Department of Organic Chemistry, Faculty of Chemistry, University of Kashan, Kashan, Iran.

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

³Alzahra University, Faculty of Economics, Iran.

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

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

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

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

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*Corresponding Author Email: kanishk.kala@gmail.com

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 Artemisia extract as effective products have a useful and promising role as skin anti-inflammations on the skin of the body.

KEY WORDS:

Artemisia annua, hydroalcoholic method, ingredient, anti-inflammations, skin

1. 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 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 and 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).

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, amyryl, myrcene, squalene, 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 ingredient of Artemisia annua herb have 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°C. 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).

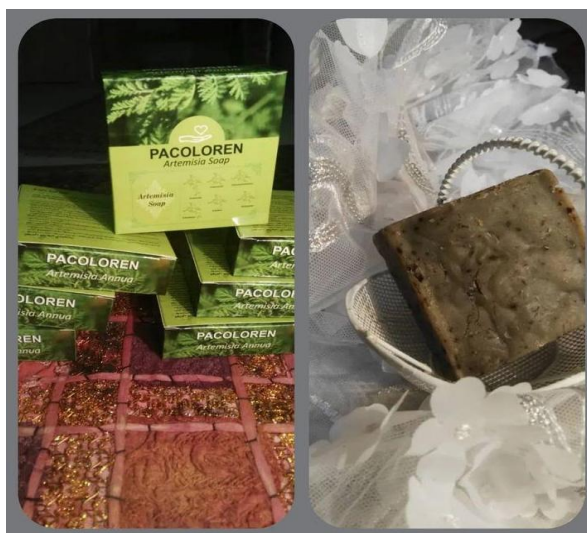


Fig 1: *Artemisia annua* herb solid soap with Brand PACOLOREN

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).

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

Artemisan 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 ₂ EDTA)	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
<i>Artemisia annua</i> Extract	0.5
Distilled water	80



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

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 c was 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. 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).

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	Tweat 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



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

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 $22\pm 2^{\circ}\text{C}$, with a relative humidity of $50\pm 5\%$, 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.

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

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 the 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 edema) were calculated from numerical scores obtained at the 60 minutes, 24-, 48-, and 72-hour observations (Table 4). The 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.

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 60 min		
After 48 hr.		

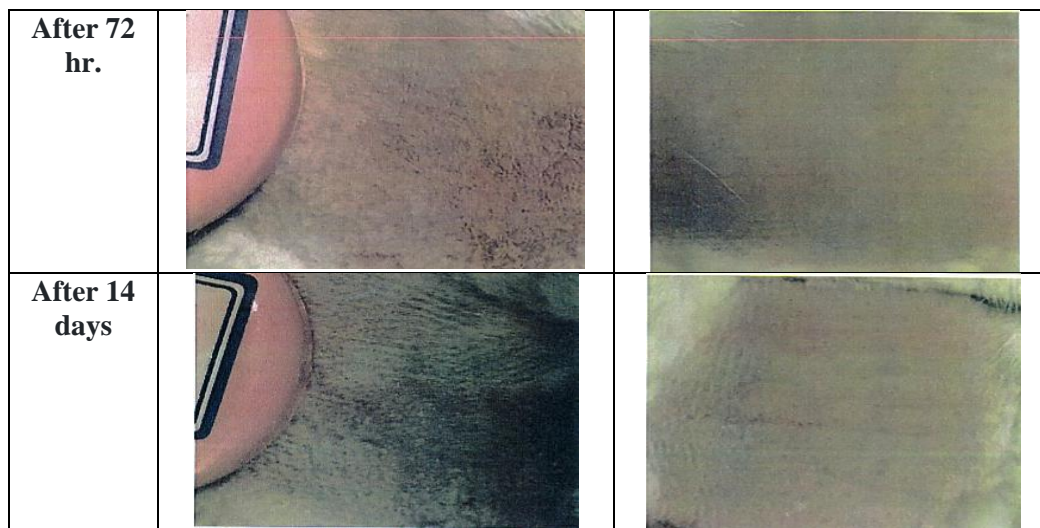


Table 5: Descriptive rating for mean primary irritation index (PII).

Primary irritation index (PII)	Classification
0	Negligible
$0 < PII \leq 2$	Slight
$0 < PII \leq 5$	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

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 shown in Figure 4 & 5, fatty acids palmitic acid, oleic acid, linoleic acid, stearic acid has the highest percentage of abundance in herbal cream.

Fig 4. Fatty acids of the first category

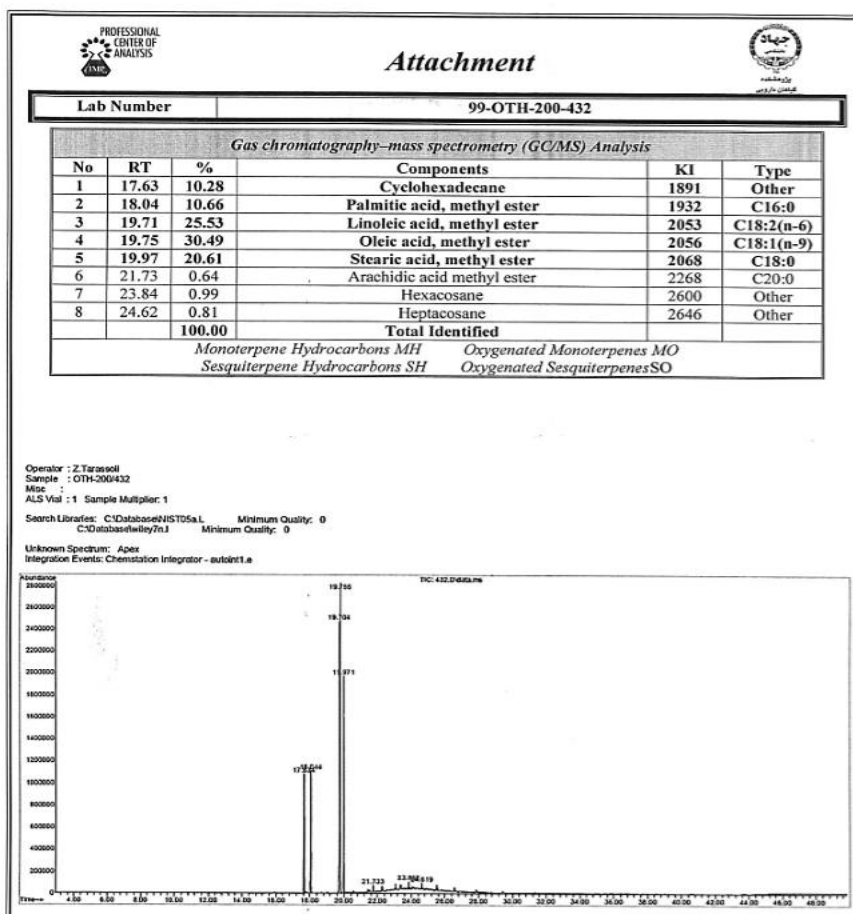
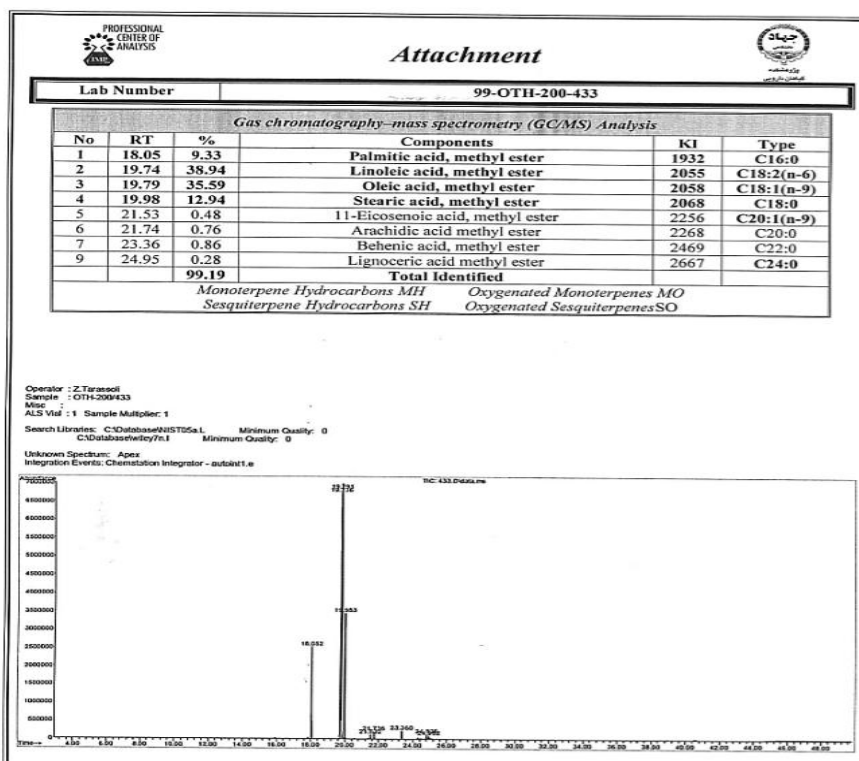


Fig 5. Fatty acids of the second category



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.

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References:

1. Allahtavakoli M, Khaksari Haddad M, Assar Sh. Comparison of topical application of Mummify and Phenytoin cream on skin wound healing in rat. *J Babol Univ Med Sci* 2003; 5(18): 7-13.
2. Dinani NJ, Asgary A, Madani H, Naderi G, Mahzoni P. Hypocholesterolemic and antiatherosclerotic effect of *artemisia aucheri* in 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. *Pajooresh 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 assessing the activities of Composite *Artemisia Capillaris* Tablets against hepatitis B virus replication in vitro. *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.