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## RESEARCH ARTICLE

### NEW POLYMER IMPROVED HEMOSTATIC WITH ANTIBACTERIAL PROPERTIES

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#### ABSTRACT

The aim of research was the evaluation of blood clotting activity and hemostatic effect of Salvia polymers using developed set of laboratory tests and preparation of Salvia polymers with high blood clotting activity. Salvia polymers have antibacterial effect with rapid coagulation time and local drug applied to site of bleeding to stop bleeding. **Material and Method:** Salvia officinalis is one of the essential herb. In this study take leaves of plant. extraction alcoholic & water extract and study chemical composition of this extract by UV spectrophotometer and prepare polymer that load the extract of Salvia Officinalis then the use Differential scanning calorimetry for Salvia officinalis. In addition, study the new polymer with antibacterial study with study prothrombin time toxicity level of salvia polymer. **Result:** results The study of the leaf extract shows the effect of S. aureus through the chemical structure containing Tammi, the main oil that is a basic component (p-cymene and-terpinene) which corresponds to the previous results and its effect on the bacteria. Indicates that it is anti-bacteria with hemostatic effect. New polymer can used in stop bleeding with antibacterial effect. **Conclusion:** The advantage of this drug is that the few side effect (local drug). In addition to the ease of its position and its control over the place of bleeding, it can easily applied in cases of venous and arterial hemorrhage, as well as the wounds as well as an antibacterial substance. In addition, quick effect rapid period of coagulation and anti-bacterial and thus reduce the mortality caused by blood poisoning and blood loss.

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## INTRODUCTION

Salvia officinalis is one of the essential herb. This plant family includes approximately 230 genera and 7100 species all over the world. (Abdelkader et al. 2014) postulated that this family includes several species, which have great value due to their effect in medicament, cooking, makeup, beauty products and production of essential oils. Salvia officinalis is one of the essential herbs. Essential oil of sage contains cineole, borneol, and thujone. Leaf contains tannic acid, oleic acid, ursolic acid, niacin, nicotinamide, flavones, flavonoid glycosides, camphor, ursolic acid, fumaric acid, chlorogenic acid, caffeic acid, and estrogenic substances (Harley et al., 2004). Salvia is one of the major genera of Lamiaceae family (Ilkiu-Vidal et al., 2010). This genus includes about 900 species, which cultivated in many countries due to its traditional usefulness in medicine

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and for domestic applications (Ayatollahi. A et al., 2009). This plant historically known from the early 1960s until now by its therapeutic and domestic applications due to its high economic value (Akhondzadeh et al., 2004). The recent studies suggested that the plant extracts is naturally rich sources of antioxidants, which used in the prevention of diseases occurred because of oxidative stress and free radical attack. (Pierozan, M.K. et al. 2009). (Behradmanesh et al., 2013) showed that this plant used in the food and beverage industries due to its tasty and spicy flavor. In addition, it added to several food products as preservative (Baranauskiene et al., 2011). This is due to its powerful antimicrobial activity against several gram positive and negative bacteria such as L. monocytogenes, B. cereus, B. subtilis, E. coli S. aureus and all various food borne pathogens (Nickavar B et al., 2007) & (Aleksovski et al., 2007). (Miura et al., 2002) postulated that this plant species used widely for treatment of some diseases. They added that the genus name Salvia comes from the Latin names S. officinalis meaning to heal and means medicinal. The recent studies reported by (Behradmanesh et al. 2013).

emphasized that *Salvia officinalis* a common herbal plant known as sage, common sage, garden sage, golden sage, true sage, culinary sage, kitchen sage and broadleaf sage. Antioxidant and antimicrobial activities of the extracts depend on the type of plant material and the extraction conditions ([http://www.who.int/whosis/whostat/EN\\_WHS09](http://www.who.int/whosis/whostat/EN_WHS09)). *S. officinalis* L leaves characterized by antioxidant properties and the antioxidants are effective against blood sugar. It was reported that *S. officinalis* L has wide range of biological activities, such as antioxidant, antibacterial, fungistatic, virustatic, astringent, hypoglycemic and anti-inflammatory properties (Longaray Delamare AP *et al.*, 2007). The literature data showed that carnosic acid, carnosol and methyl carnosate are responsible for the antioxidant properties of sage extracts. Due to presence of the essential oil, this species showed hypotensive properties, central nervous system-depressant actions and anti-spasmodic activity. *S. officinalis* tincture showed anti-inflammatory effects by reducing marrow acute phase response and NO synthesis. Although *S. officinalis* tincture had a smaller inhibitory effect than diclofenac, it significantly reduced the total leukocyte and monocytes percentages and the activation of circulating phagocytes (Moein MR *et al.* 2015).

Presence of the organic compound, diterpene, is the major constituent, which appears to relate to the anti-inflammatory properties of the plant. This is in addition to presence of compounds caryophyllene oxide and caryophyllene, both of which have displayed anti-inflammatory activity. Baracevic *et al.* (2001) showed that ursolic acid was the main component of different fractions of *S. officinalis* L. The antioxidant effects of this extract have often been attributed to phenolic and monoterpene compounds. Flavonoids which are considered as groups of the polyphenols and rosmarinic acid possess several modulatory effects, either inducing or decreasing the expression of SOD and CAT enzymes. Rosmarinic acid belongs to the predominant phenolic compound in sage. Its effect attributed to the antioxidant properties through scavenging of reactive oxygen species. Sage extract has the ability to prevent GSH depletion by their main phenolic compounds, rosmarinic acid and luteolin-7-glucoside. Brandstetter *et al.* (2009) postulated that sage extract (especially the methanolic extract) was able to induce the glutathione synthesis and hence increase basal GSH levels.

Bleeding is the one of leading cause of death after civil and combat trauma and affective hemostasis is a key challenge for emergency medicine. Current review focused on modern topical hemostatic agents based on chitosan. Review classified all topical hemostatic agents and show advantage of chitosan-based dressing. In addition, it gives perspectives in hemostatic dressing research. Adequate hemostasis after trauma and during surgical operation is a big challenge in modern medicine. About the 40% traumatic and more than 90% of combat deaths took place in pre-hospital settings. In addition, about the 50% from these reported due to massive blood loss (Ersoy G, 2007). Sauaia A. reported 80% of civilian trauma fatalities within the United States caused by uncontrollable hemorrhage (Sauaia A., 1995). In addition, hemorrhage in trauma patients is a leading cause resulting reoperation (Hirshberg A, 1993). Topical hemostatic treatment applied since ancient time. They used herbs, mixture of wax, grease, barley, and animal hides mixed with hot sand to stop bleeding (Hardean E. Achneck, 2010).

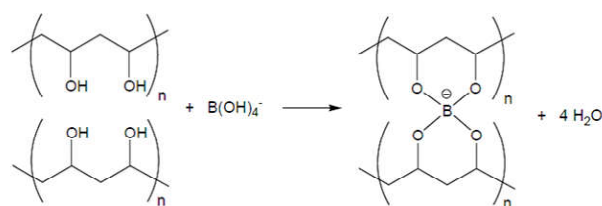
Advances in biotechnology have resulted in an explosive growth of topical hemostatic agents in the last two decades. The plant is a known herb plant since ancient times. Many species are native to the region, where they have used since time or several medicinal uses due to its effectiveness as a preservative, disinfectant and anti-inflammatory agent, which extracted by alcohol and water extraction and then made as polymer clay. The main cause of hemostatic effect of saliva polymers is aggregation and activation. Hemostatic action are most promising due to effective blood stop and possible additional properties like antibacterial and stimulatory that widely used both in battlefield and civil emergence.

## MATERIALS AND METHODS

**Extraction of Plant:** The plant collected and the drying of the plant extracted by the extraction of alcohol and water. Grind the dried leaves thoroughly and weigh 100 g of the substance. Add 400 ml ethanol concentration 70% and cool well after it coated with a light insulating material to prevent the oxidation of light materials with continuous shaking and continue for 3 days. The filter is then filter into the centrifuge at 5000 cycles per minute for 10 minutes then the leachate is taken and converted to other tubes and then dried in the incubator 45 ° C to obtain the powder material. The leaves of the saliva official's plant obtained from the local markets and confirmed to classify in the Department of Life Sciences / University of Holler. The boiling water extract of the leaves of the plant *S. officinalis* obtained using the method.

### Preparation of gelatin polymer

- Prepare 4% of the poly-phenyl alcohol solution (4 g of poly-ethyl alcohol was prepared in 100 of hot water 70 ml).
- Prepare 4% of the borax solution was prepared Na<sub>2</sub>B<sub>4</sub>O<sub>7</sub> • 10H<sub>2</sub>O (4 g of borax in 100 ml of water) as the borax decomposes to 4 (B (OH))
- Mixed two solution 10: 1 (Borax: Poly Vinyl alcohol). With continuous stirring, we will get thick and mixed polymers as shown below.



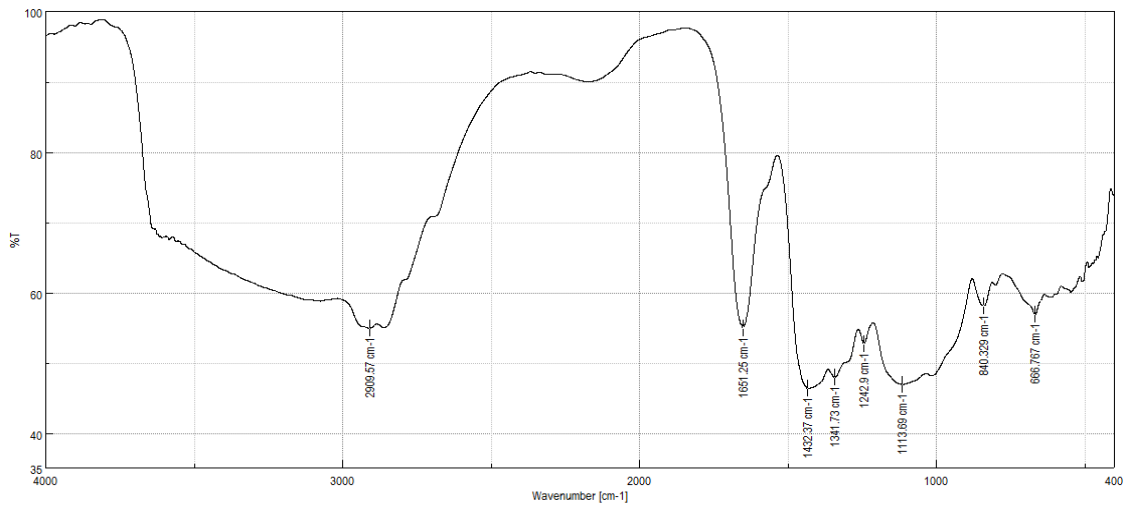
- Allow to dry for 24 hours in a 50 m oven to get a solid polymer material to be grinded for powder.
- Take 10 ml of the extract of alcohol and water to the plant saliva .official's in Baker and add the polymer powder with steel stirring for one hour at room temperature will note the swelling of the polymer with the extract of saliva .official's plant and obtain a gelatin polymer.

Studying the gelatinous polymer network with the extract of saliva. official's alcohol and water. The swelling behavior of the gels polymers can studied by measuring the water content of the gel (Water Content). The swelling ratio, symbolized by the symbol Q, calculated as the ratio between the weight of water absorbed by the gel and the dry gel weight, Puffiness (gm solution absorbed / gm polymer) with the following equation:



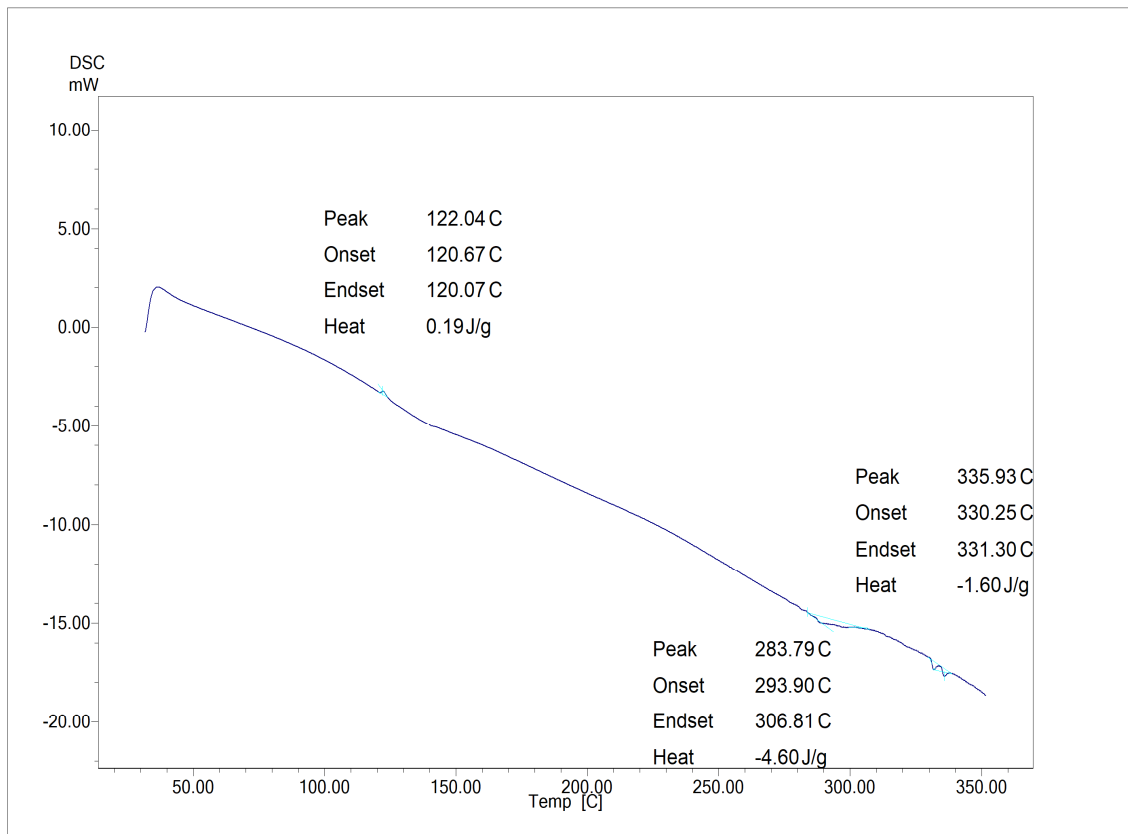
Area was cleaned with povidone Iodide solution disinfectant

**Figure (3.6) Rabbit cleaned with povidone iodide**



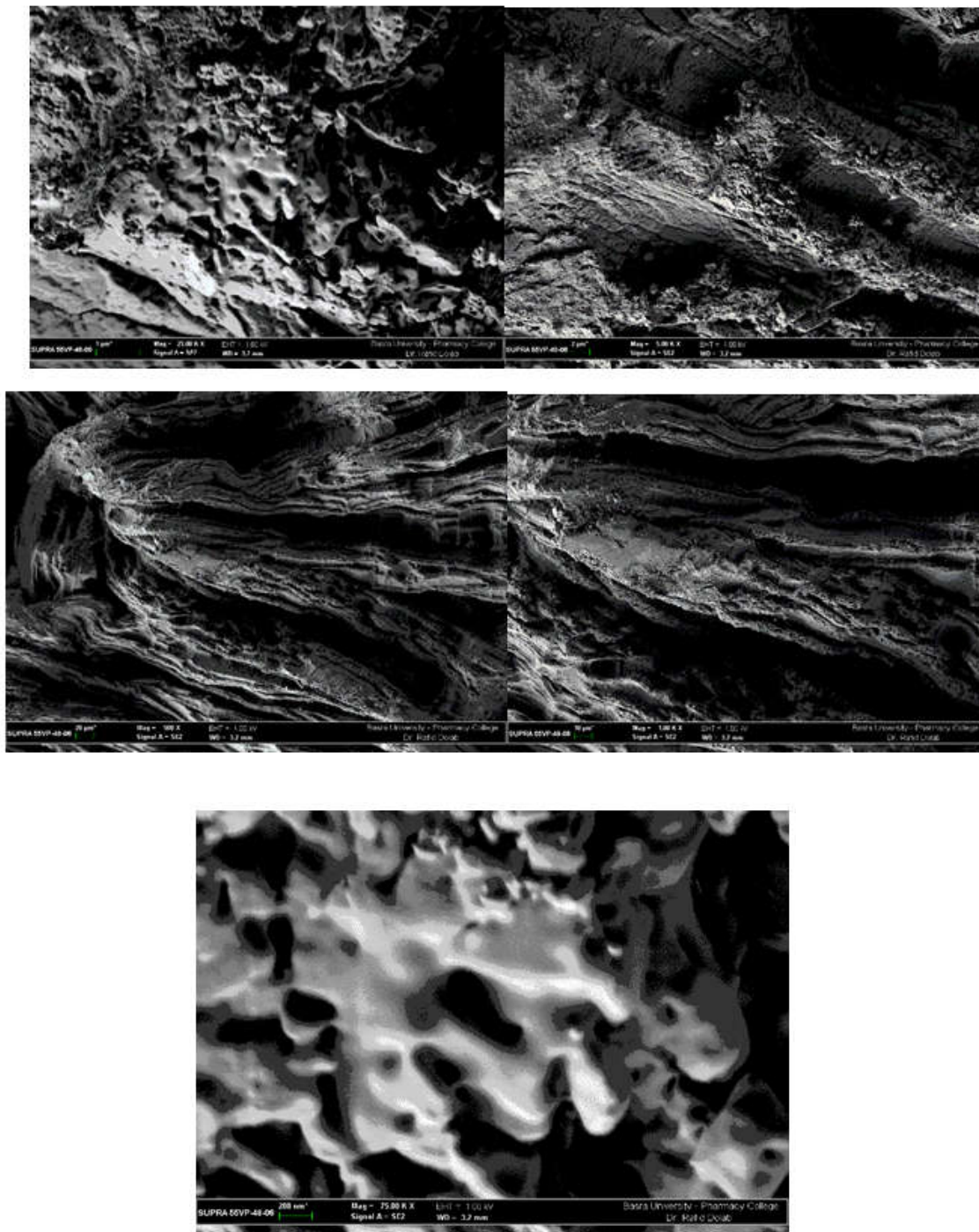
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**Figure 7. The infrared spectrum of the gelatin polymer loaded on the saliva**



**Figure 8. DSC of saliva polymers DSC (Differential scanning calorimetry)**

Test compound	Bacterium		MIC	Result
	MBC	MIC		
<i>S. officinalis</i>		2.5	1.25	Effect against <i>S.aureus</i>



Q= Taken as a percentage according to the following equation:

$$100 \times \% Q = \quad (2)$$

Swelling ratio: (Swelling ratio (g solution / g polymer). = Q

Ws= weight polymer swollen

Wd = weight dry polymer

Thrombin generation and Thrombin formation:

Thrombin–antithrombin complex (TAT), a marker of thrombin neutralization, is an indicator of how much thrombin formed over a period.

Figure (4) shows the thrombin withdrawal and its deposition on a coagulation device. Blood clotting was performed on a total of 4 rabbits aged between 6 months and one year. Developed under similar administrative conditions and examined for disease safety, the surgical site of the gluteal region was surgically prepared for injuries of 3 x 2 cm after anesthesia.

The polymer network installed and the coagulation process followed every half an hour clinically. Less than two hours after the blood clot was stopped and the bleeding stopped completely in less than an hour of rabbits and two rabbits stopped bleeding completely after an hour and a half.

:(LD50 ) Median lethal dose

This study used agroups of white mice four group (Albino mice) provided with natural food. Each group consists of (20) mice (10 females and 10 males). Different doses (10,7,5,2.5 , 2 , 1.5 , 1) were given from the polymeric network orally using a gastric tube with a control group. The number of deaths was recorded within (48) hours, noting differences between the treatment groups and the control group. This experiment conducted at the University of Basra (Schoenecker *et al.*, 2001). In addition, the study antibacterial effect (*S. aureus*).

## RESULTS

The figure below shows the process of polymerization of the gelatinous polymer network with alcohol extract solution ( $\Delta$ ) and water ( $\blacklozenge$ ). Where we note that the absorption ratio of the alcohol extract represents 500% of the weight of the gelatinous polymer network while the absorption rate of the water extract is 410% of the weight of the polymer network. infrared spectrum samples analyzed from the main sage components, although they found in different concentrations. That carnosic acid, carnosol and methyl carnosate are responsible for the antioxidant properties of sage extracts. Due to presence of the essential oil, this species showed hypotensive properties, central nervous system-depressant actions and anti-spasmodic activity. *S. officinalis* tincture showed antiinflammatory effects by reducing acute phase response and NO synthesis. Although *S. officinalis* tincture had a smaller inhibitory effect than diclofenac, it significantly reduced the total leukocyte and monocytes percentages and the activation of circulating phagocytes .Presence of the organic compound, diterpene, is the major constituent, which appears to related to the anti-inflammatory properties of the plant. This is in addition to presence of compounds caryophyllene oxide and caryophyllene, both of which have displayed anti-inflammatory activity.

Baracevic *et al.* (2001) showed that ursolic acid was the main component of different fractions of *S. officinalis* L. chloroform extract. It has a strong anti-oxidant, free radicals and anti-bacterial antibiotics. It contains phenolic acid in its essential components And it contains uricolic acid as a major component involved in its anti-inflammatory activity. The anti-inflammatory effect of ursolic acid (ID50 = 0.14  $\mu\text{mol} / \text{CM}^2$ ) was two times more potent than indomethacin (ID50 = 0.26  $\mu\text{mol} / \text{CM}^2$ ), which was used as a non-steroidal anti-inflammatory reference and suggested oresolic acid content Carnosic acid and rosemarynic acid These are the components of the sage present in high concentrations in the sage extract as an antioxidant, a preservative 8, Ursolic acid Differential scanning calorimetry Used widely for examining HYPERLINK "https://en.wikipedia.org /wiki/ Polymer" polymeric materials to determine their thermal transitions. The observed thermal transitions can be utilized to compare materials, although the transitions do not uniquely identify composition. The composition of unknown materials may be completed using complementary techniques such as IR spectroscopy results The study of the leaf extract shows the effect of *S. aureus* through the chemical structure containing T.ammi, the main oil that is a basic component (p-cymene and-terpinene) which corresponds to the previous results and its effect on the bacteria Indicates that it is anti-bacteria. Minimum inhibitory concentration (MIC) is the lowest concentration of a chemical, which prevents visible growth of

a bacterium. This is in difference to the minimum bactericidal concentration (MBC), which is the concentration resulting in microbial death as defined by the inability to re-culture bacteria. The closer the MIC is to the MBC, the more bactericidal the compound Amyes *S et al.* 1996. Uricolic acid as a major component involved in its anti-inflammatory activity. The anti-inflammatory effect of uricolic acid (ID50 = 0.14  $\mu\text{mol} / \text{CM}^2$ ) was two times more potent than endometacin (ID50 = 0.26  $\mu\text{mol} / \text{CM}^2$ ), which was used as a non-steroidal anti-inflammatory reference and suggested oresolic acid content. TAT is a sign of the formation of the thrombosis over time for the measurement of the coagulation test in the tube containing the blood in which the saiva polyme is the highest percentage than the control blood group that is not treated(control blood & saliva officialis only). Thrombin generation over time, as measured by the levels of thrombin–antithromb in The results showed that there was no loss in all treated rats as well as control mice. The behavior was normal compared with control mice for five days of observation. The mice that injected with concentrations above 15 g / kg reported to have mortality after 48 hours with 40% of the animals, and these results are excellent compared to half of the lethal dose (Ersoy *et al.*, 2007). The microscopic imaging of the new polymeric polymer carried out at the College of Pharmacy / University of Basra in the electronic microscope. The images appeared as follows

## Conclusion

New herbal medicine and form and the preparation of a unique combination of polymer gelatin, alcohol and poranex. The gel components are homogenous and not absorbed by the skin. We have noticed gradual degeneration of the uterus as it helps prevent hemorrhage in stopping bleeding in war after it is manufactured and stopping deaths. Resulting from severe bleeding can be applied to a wide range of people who suffer from bleeding in the event of being transferred to the hospital needs time and time is not in the patient's benefit we are forced to lose the result of the lack of control of bleeding in wars need to stop bleeding fast time In order to avoid the loss of huge amounts of blood and fluids and thus the patient's loss of consciousness and enter into a coma. The advantage of this drug is that the few side effect (local drug). In addition to the ease of its position and its control over the place of bleeding, it can easy applied in cases of venous and arterial hemorrhage, as well as the wounds as well as an antibacterial substance. In addition, quick effect rapid period of coagulation and anti-bacterial and thus reduce the mortality caused by blood poisoning and blood loss

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