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Research Article

INVESTIGATION AND COMPARISON OF ANTIBACTERIAL EFFECTS OF HUMAN PLATELETS ON DIFFERENT STRAINS OF *PSEUDOMONAS*

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ABSTRACT
Nosocomial infections are a major medical problem in developing countries that cause the spread of infectious diseases in societies. Paying attention to nosocomial infections is of utmost importance in recent years. *Pseudomonas* is the most important pathogenic bacterium, especially in immunocompromised hosts. This bacterium is naturally resistant to antibiotics and/or earns resistant to them. In today's world treatment process occurs in other ways including the use of stem cells and blood cells, especially human platelets. Determination and comparison of antibacterial effects of human platelets on different strains of *Pseudomonas* will be studied in this research. This research is a pilot study. The population in this study is different samples of *Pseudomonas* strains. To select *Pseudomonas* type from its different types, its prevalence and frequency is placed at center of attention. According to obtained researches, *Pseudomonas aeruginosa* is the most frequent type of these infections. Cluster sampling method was used to select the strain among students of Kerman University of Medical Sciences. Sampling was completed by selection of 20 female and 20 male. Platelet-derived products (e.g., activated platelet-rich plasma (PRP), deactivated PRP and light PRP (LPRP)) were used as tools in this study. These products are obtained by laboratory methods such as centrifuging and freezing and de-freezing. Known and original disk diffusion method was used to find the effect of these products on bacterial strains. The results obtained from this study showed that sex influences on the size of halo diameter created around platelet-derived products including activated PRP, deactivated PRP and activated LPRP in bacterial culture medium (P-Value<0.05). There was also a significant relationship between the size of halo diameter created by activated PRP and LPRP platelet-derived products as well as *Enterococcus faecalis* and *Staphylococcus aureus* and *Bacillus cereus* bacteria (P-Value<0.05). There was no significant relationship with *E. coli* except *Pseudomonas Aeruginosa* strain (P-Value<0.05), but there was a significant relationship between activated PRP in other strains of *Pseudomonas* bacteria and *E. coli* (P-Value<0.05). So, platelet-derived products influence in *Pseudomonas* bacteria, especially in *Pseudomonas Aeruginosa* strain and these types of products can be used as antibacterial agents in infections. Based on this research, an important and applied conclusion is obtained that stem cells including blood platelets and so on play an important role as antibacterial to avoid the adverse effects of antibiotics. This finding is stronger in men than in women. Platelets cannot achieve this lonely, unless they become lysis or be used together with white blood cells and/or plasma, which contains other substances.

Keywords: *Pseudomonas Bacteria, Antibacterial, Platelet-Derived Products*

INTRODUCTION
Pseudomonas is one of the most pathogenic bacterium, especially in immunocompromised hosts. It is one of gram-negative bacteria associated with nosocomial infections. Infections caused by this bacterium are difficult to treat, since it is inherently resistant to antibiotics and/or acquires resistance to them. So, selection of appropriate antibiotics for treatment of infections caused by this bacterium is limited. *Pseudomonas* bacteria have high resistance to toxic effects of disinfection materials, such as disinfectants, antiseptics and protections. They are even able to grow inside some of these disinfection agents, which causes infection transfer and creation of cross-infection in hospitals. Etemadi and Shahraki Zahhedani

(1996) in their investigation on the role of *Serratia* and *Pseudomonas* in nosocomial infections have concluded that almost half of nosocomial infections, infections that hospitalized patients acquire it from the hospital, are Gram-negative bacteria that are ranked at the first degree of importance. Another important bacterium that can cause disease is *E. coli* or *Escherichia coli*, a gram-negative bacillus of the family *Enterobacteriaceae*. This bacterium is transferred from one person to another via the oral-fecal route (Retrieved, 2012). *Enterococcus faecalis* are gram-positive cocci in the form of catalase-negative chain and are the agent of urinary tract system and biliary tract infections, which are the most common infections. Another applied bacterium is *Bacillus anthracis* that is a gram-positive bacteria and causative agent of anthrax. *Bacillus cereus* lives as saprophyte in soil, water and air. Unlike *Bacillus anthracis*, this bacterium animates and produces hemolysin. Special platelet-derived products called platelet gel (PRP) are used to treat and/or prevent from spread of these bacteria in treatment of infected wounds. Platelet-derived products are taken from the blood. Platelet-rich plasma (PRP) is a blood product. This kind of plasma is taken from donor individual and is injected into damaged tissue and/or injury location of own individual for therapeutic purposes. Platelet gels are also derivatives of PRP blood products that generally obtain from activation of thrombin in platelet rich plasma (PRP). And today, they are used as adjuvant treatment for improvement of wounds and burns. Platelet lysis in repair of bones, tendons, ligaments and soft tissues after injury is a common phenomenon in most of patients. Since prolonged repairing time makes person to be prone to granulation tissue and/or fibrosis at the site of lesion, it can lead to a decrease in function of the organ and/or to not fused breakage. Nosocomial infections are considered as one of major medical problems in developing countries that cause spread of infectious diseases in societies (Adabi *et al.,* 2015). Paying attention to nosocomial infections in the recent years is of great importance, since a number of eighty eight deaths in the world have been reported only in 2016 due to these infections. The majority of nosocomial infections are created in hospitals' burn unit by opportunistic bacteria. Extensive wounds caused by burns facilitate entrance of organisms into the tissues and they are ideal for bacterial growth (Adabi *et al.,* 2015). Given that different strains are effective in creation of wound infections and *Pseudomonas aeruginosa* is a most important strain isolated from clinical samples and wounds, all individuals in hospitals try to control and prevent from it. This makes people to inevitably apply broad-spectrum antibiotics lead to resistance and incidence of severe side effects in patients, especially in hospitalized patients in burn unit. These patients usually suffer from substantially reduction in immune level.

So, treatment process in today's world occurs in other ways and that is the use of biological methods, which provide the possibility to reduce the destructive and harmful effects of chemical compounds, especially antibiotics. Among these methods can mention to the use of stem cells and blood cells, especially human platelets.

Given the important role of these cells in blood coagulation and body's defense against infection, and probably by having several compounds such as VEGF factor, vascular endothelial growth factor, transferring growth factor β, platelet-derived growth factor, PLT-derived growth factor and so on, it makes more critical the role of platelets in the wound healing process. One of more efficient products is platelet-rich plasma or PRP, which also termed as platelet gel and can be obtained from selective separation of solid and liquid components of whole blood from each other by means of plasmapheresis technique (Casper, 2005). The platelet gel derived from blood plasma of donor people and used for therapeutic purposes and tissue repair, is generally obtained from activation of thrombin platelet-rich plasma and is used today a as treatment adjuvant in wound healing caused by burn (Everstat, 2006). Since only the influence of platelet derivatives on various bacterial strains available in wounds have measured in numerous papers, and none of them have covered the impact of derivatives on different strains and that which of the products have had higher impact on inhibition of bacterial growth (Bagherifar, 2003; Tajbakhsh, 2013), obtained products from blood were used in this study. The products were obtained through blood separation method with centrifuge device. The upper layer of blood placed immediately on the layers is white or gray and is composed of white blood cells. A thin layer of platelets, which are not detectable with naked eye, covers white blood cells (Rahimi, 2014). The aim of this study was

determination and comparison of antibacterial effects of human platelets on different strains of *Pseudomonas*.

MATERIALS AND METHODS

Methods

This experimental study was performed 7 months in 2015 in standard condition of cell culture, in Laboratory of Bacteriology and Hematology of Kerman University of Medical Sciences and investigated the impact of antibacterial effects of human platelets on different strains of *Pseudomonas*. Cluster sampling method was used to select the sample among students of Kerman University of Medical Sciences.

Sampling was completed by selection of 20 women and 20 men. Platelet-derived products (e.g., activated platelet-rich plasma (PRP), platelet extract, light PRP (LPRP) and washed platelet) were used as tools in this study. These products were obtained by laboratory methods such as centrifuging and freezing and de-freezing and this paper will describe two of them including PRP and LPRP. To obtain PRP, blood samples were taken from the students. The samples were poured into Falcon tubes under sterile conditions.

By pouring 1cc of citrate and 9cc of blood, a solution with a ratio of 1/9 was obtained. The solution was then centrifuged at 800 RPM for 10 min. After centrifugation, a solution containing blood cells and platelets was obtained at the top of the tube. A solution rich of platelets, called PRP, is separated from 1cm above red blood cells or above buffy coat. The famous and original disk diffusion method was used to find the influence of these products on different strains of bacteria. The same method is used for extraction of LPRP.

But, after centrifugation, LPRP obtains by a completely sterile sampler from erythrocyte sedimentation area and surface plasma contains platelets and white blood cells that called buffy coat which is rich of white blood cells and also has platelets to some extent. The following compounds were respectively placed in centrifuge tubes: red blood cells deposit at the bottom of the tube, the buffy coat layer containing white blood cells and some platelets, then the surface layer containing plasma and platelets. The buffy coat layer was needed in this step and we separated it. Similar to before, the obtained solution was poured in micro-tube and applied and antibacterial evaluation was occurred by disk diffusion method. The method of disk diffusion was similar to well diffusion method, by the difference that not only the well was not created in the culture but also bacteria was cultivated inside plate after that the culture became cool.

The sterile disk was soiled to antibacterial material and placed on the culture at regular intervals. Then, the plates were incubated at 37 ˚C for 24 hours. Colony morphology, gram staining and biochemical characteristics including growth in pyrrolidonyl arylamidase (PYR) activity were used to verify the identity of *Pseudomonas* strain. Fermentation glucose test was used to verify the identity of *Pseudomonas* species. Then, the identified strain was cultured in blood agar plates and used in later stages. The main method was disk diffusion method. Cell counting was conducted by KX 21 Cell Counter device and the obtained data was recorded. To continue the tests and to determine antibacterial property of the samples, gram-negative bacteria such as *Pseudomonas* and *E. coli* were used in this study.

Data Analysis Method

SPSS statistics 20 was used to analyze the data. The extracted data were divided into two categories: descriptive and inferential statistics. Some indicators including median, mode, mean, frequency, frequency percentage and Pearson correlation coefficient were also used.

RESULTS AND DISCUSSION

Results

40 students including 20 men (50%) and 20 women (50%) were selected. Sexual frequency distribution and frequency distribution of white blood cell count of the students are given in Table 1 and Chart (bar graph) 1, respectively.

Table 1: Sexual Frequency Distribution of the Students

Sex	Frequency	Frequency Percentage
Male	20	50%
Female	20	50%
Total	40	100%

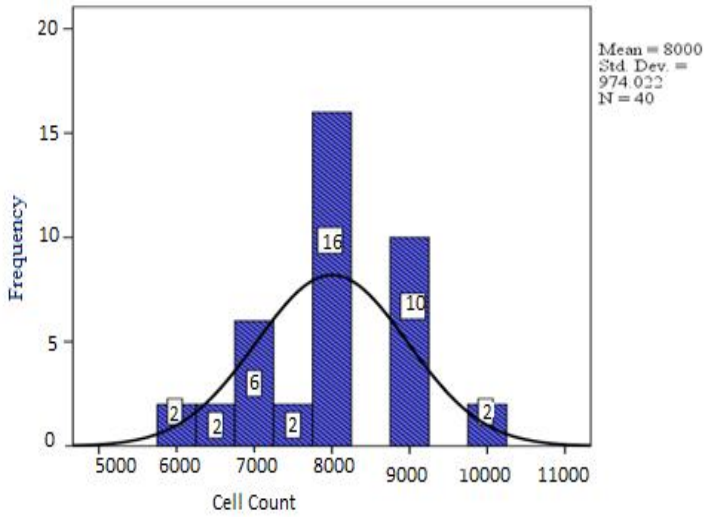

Chart 1: Frequency Distribution of the Students' White Blood Cell Count

Table 2: Comparison of the Size of Halo Diameter Created around Activated PRP Platelet-Derived Product Injected to the Bacterial Culture Medium in Terms of Gender

Bacterial Types		Sex	SD±Mean	Test Statistics	Degree of Freedom	P-Value
Pseudomonas	*Aeruginosa*	Male	14.175±1.6163	4.633	38	0.000
		Female	11.825±1.5917			
	Wound sample 1	Male	12.85±1.7704	3.217	38	0.003
		Female	11.15±1.5652			
	Wound sample 2	Male	12.85±1.7704	4.172	38	0.000
		Female	11.15±1.5652			
	Wound sample 3	Male	12.4±1.3917	4.303	38	0.000
		Female	11.6±1.2576			
	Strain 1	Male	4.8±1.78	3.314	38	0.002
		Female	3.1±1.4473			
	Strain 2	Male	5.35±1.1367	4.241	38	0.000
		Female	3.8±1.1743			
Enterococcus faecalis		Male	2.525±1.4279	2.665	38	0.011
		Female	1.475±1.0321			
Staphylococcus aureus		Male	2.575±1.4075	3.057	38	0.004
		Female	1.425±0.92			
E. coli		Male	8.6±0.8208	4.921	38	0.000
		Female	7.4±0.7182			
Bacillus cereus		Male	7.6±0.8208	4.982	38	0.000

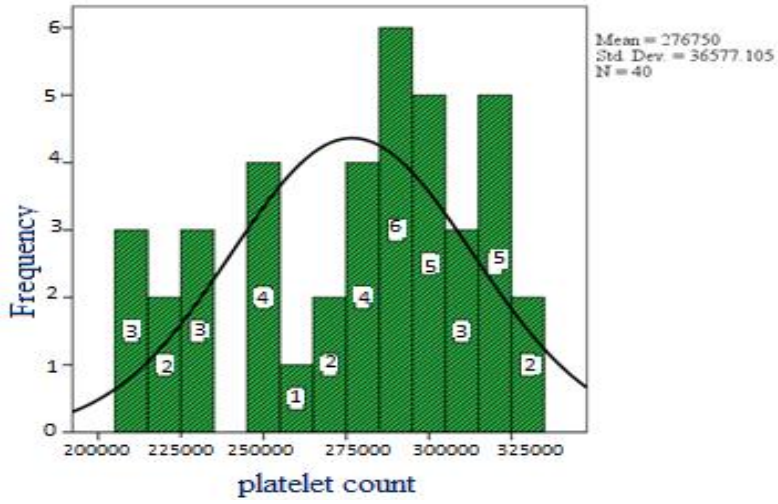

Chart 2: Frequency Distribution of the Students' Blood Platelet Count

Table 3: Comparison of the Size of Halo Diameter Created around Deactivated PRP Platelet-Derived Product Injected to the Bacterial Culture Medium in Terms of Gender

Bacterial Types		Sex	SD±Mean	Test Statistics	Degree of Freedom	P-Value
Pseudomonas	*Aeruginosa*	Male Female	8.175±0.9635 5.825±1.5917	5.648	38	0.000
	Wound sample 1	Male Female	6.975±1.5684 5±1.539	4.020	38	0.000
	Wound sample 2	Male Female	6±1.6543 4.075±1.3886	3.986	38	0.000
	Wound sample 3	Male Female	7.275±1.543 5.725±1.3715	3.358	38	0.002
	Strain 1	Male Female	3.75±1.2722 2.25±0.7164	4.595	38	0.000
	Strain 2	Male Female	4.275±0.8955 2.725±0.8656	5.565	38	0.000
Enterococcus faecalis		Male Female	2.525±0.8955 1.475±0.525	4.523	38	0.000
Staphylococcus aureus		Male Female	2. 5±1. 761 1.4±0.7539	3.744	38	0.001
E. coli		Male Female	5.6±0.9403 4.4±0.6806	4.623	38	0.000
Bacillus cereus		Male Female	4.55±0.8721 3.45±0.7931	4.173	38	0.000

The results of t-test showed that the mean size of halo diameter created around activated PRP injected to the bacterial culture medium in terms of gender is not the same (significance level less than 0.05). In other words, there is a significant difference between the mean sizes of halo diameter created around activated PRP injected to the bacterial culture medium in terms of gender. This means that gender has impact on the size of halo diameter created around activated PRP injected to the bacterial culture medium (P-Value<0.05). Hence, the above Table shows that the mean sizes of halo diameter created around activated PRP injected to the bacterial culture medium in the men group is more than in the women group (Table 2).

Table 4: Comparison of the Size of Halo Diameter Created around Activated LPRP Platelet-Derived Product Injected to the Bacterial Culture Medium in Terms of Gender

Bacterial Type		Sex	SD±Mean	Test Statistics	Degree of Freedom	P-Value
Pseudomonas	*Aeruginosa*	Male Female	10.675±1.2904 8.325±1.6565	5.005	38	0.000
	Wound sample 1	Male Female	6.9±1.3917 5.1±1.1877	4.4	38	0.000
	Wound sample 2	Male Female	5.975±1.3715 4.075±1.2904	4.512	38	0.000
	Wound sample 3	Male Female	7.375±1.4498 5.625±1.3066	4.01	38	0.000
	Strain 1	Male Female	1.1±0.4472 0.9±0.4168	1.463	38	0.152
	Strain 2	Male Female	2.825±1.0548 1.275±0.7691	5.31	38	0.000
Enterococcus faecalis		Male Female	1.375±0.5098 0.625±0.3582	5.384	38	0.000
Staphylococcus aureus		Male Female	2.025±1.1295 0.975±0.8347	3.344	38	0.002
E. coli		Male Female	3.575±0.9216 2.4±0.6996	4.541	38	0.000
Bacillus cereus		Male	4.55±0.8095	4.582	38	0.000

The results of t-test show that the mean sizes of halo diameter created around deactivated PRP injected to the bacterial culture medium in the sex group are not the same (significance level less than 0.05). In other words, there is a significant difference between the mean sizes of halo diameter created around deactivated PRP injected to the bacterial culture medium in terms of gender. This means that gender has impact on the size of halo diameter created around deactivated PRP injected to the bacterial culture medium (P-Value<0.05). Hence, the above Table shows that the mean sizes of halo diameter created around deactivated PRP injected to the bacterial culture medium in the men group is more than in the women group (Table 3).

The results of t-test show that the mean sizes of halo diameter created around activated LPRP injected to all bacterial culture medium are not the same except combination of strain (1) of *Pseudomonas* in the sex group (significance level less than 0.05). In other words, there is a significant difference between the mean sizes of halo diameter created around activated LPRP injected to all bacterial culture medium except combination of strain (1) of *Pseudomonas* in terms of gender. This means that gender has impact on the size of halo diameter created around activated LPRP injected to all bacterial culture medium except combination of strain (1) of *Pseudomonas* (P-Value<0.05). But the mean sizes of halo diameter created around activated LPRP injected to the bacterial culture medium of combination of strain (1) of *Pseudomonas* in the sex group is the same (significance level less than 0.05). In other words, there is no significant difference between the mean sizes of halo diameter created around activated LPRP injected to the bacterial culture medium of combination of strain (1) of *Pseudomonas* in terms of gender. This means that gender has no impact on the mean sizes of halo diameter created around activated LPRP injected to the bacterial culture medium of combination of strain (1) of *Pseudomonas* (P-Value>0.05). Hence, the above Table shows that the mean sizes of halo diameter created around activated LPRP injected to the

bacterial culture medium except combination of strain (1) of *Pseudomonas* in the men group is more than in the women group (Table 4).

Table 5: Duncan Test Results to Investigate and Determine the Best Human Platelet-Derived Products

Products	Count	Sub-Categories with α=0.05					
		1	2	3	4	5	6
Activated PRP	40						7.6550
Deactivated PRP	40				4.3975		
Activated LPRP	40			3.9562			
P-Value				1.000	1.000		1.000

As can be seen in the above Table, the difference between each of different levels of platelet-derived products is also significant and since mean of activated PRP is more than the others, so this test demonstrates the superiority of activated PRP than the other products (Table 5).

Table 6: Variance Analysis for Investigation of the Best Bacterium Compared to other Bacteria

Variables		Sum of Squares	Degrees of Freedom	Mean Squares	F Statistics	P-Value
Bacterium type	Intergroup	1827.570	9	203.063		
	Intra-group	331.445	390	0.850	238.938	0.000
	Total	2159.015	399	---		

The following result is obtained according to Table 6 that *Pseudomonas aeruginosa* is the best bacterium in comparison with the others.

Table 7: Duncan Test Results to Investigate and Determine the Best Bacterium among the Others

Bacterium Type	Count	Sub-Categories with α=0.05					
		1	2	3	4	5	6
Enterococcus faecalis	40	1.2562					
Staphylococcus aureus	40	1.4146					
Stain 1	40		1.9021				
Stain 2	40		2.2000				
Bacillus cereus	40			3.5917			
E. coli	40			3.7521			
Wound sample 2	40				5.6000		
Wound sample 1	40					6.0854	
Wound sample 3	40					6.2479	
Aeruginosa	40						7.4167
P-Value		0.443	0.149	0.437	1.000	0.431	1.000

In this test, the source of variations (variance) is divided into two groups of intergroup (difference between types of bacteria) and intra-group (error) (Table 6). Since the sum of squares related to intergroup (1827.570) is much more than the sum of squares related to intra-group (331.445), so the tests has good accuracy and the source of dispersion is caused by intergroup factors. High amount of test statistics and low amount of P-Value represents the difference between the sizes of halo diameter created in the bacterial culture medium. In other words, there is a significant difference between the sizes of halo diameter created in the bacterial culture medium (P-Value=0.000). However, to know whether or not

there is a significant difference between various levels of bacteria and to determine the best bacterium among them, complementary test should be performed. Here, since this work is a laboratory work, Duncan test is used (Table 7). As can be seen in this Table, the difference between each various levels of bacteria is also significant. And since the mean sizes of halo diameter of growth inhibition in *Pseudomonas aeruginosa* is more than the other bacteria, so this test and dot plot show the superiority of this type of bacterium than the others (Chart 3).

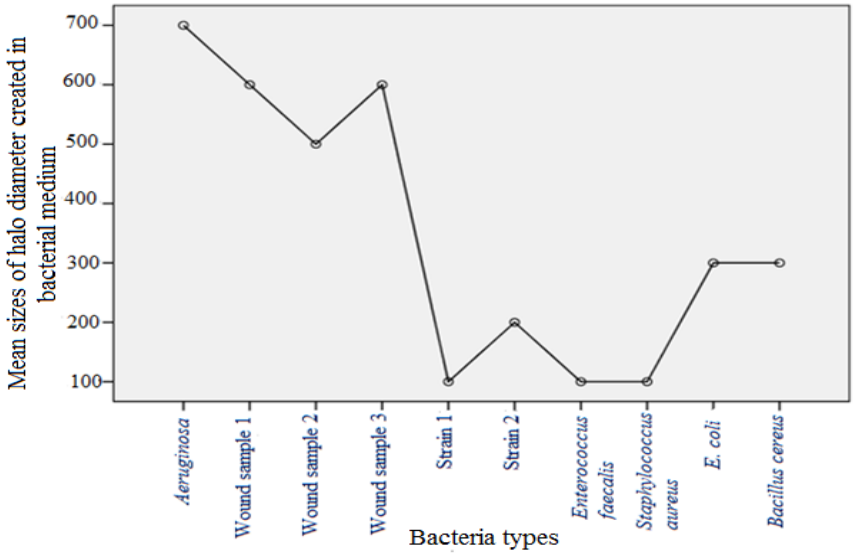

Chart 3: Determination of the Best Bacterium among the Others

Since the mean count of *Pseudomonas aeruginosa* is higher than the others, so this test shows the superiority of this type of bacterium than the others. Since in Figure 4, male sex lines in all bacteria are above female sex lines, this represents the male gender superior. And since the mean count of *Pseudomonas aeruginosa* in this Chart is higher than the other bacteria, so it is an emphasis on Duncan test.

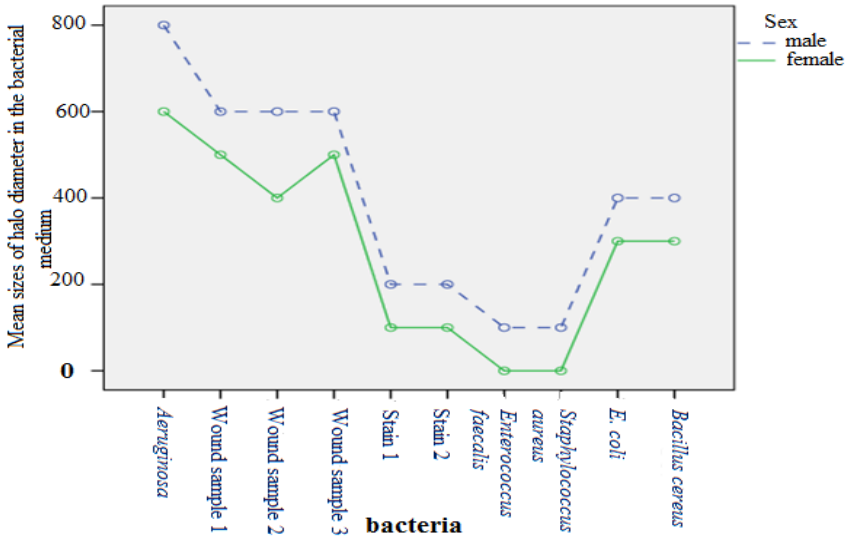

Chart 4: Determination of the most Effective Genus and the Best Bacterium among the Other Bacteria

Discussion

Many studies have shown the therapeutic effects of platelet-derived products. One of them is the study conducted by Amani *et al.,* (2009) that investigated the influence of platelet gel growth factors on proliferation and differentiation of mesenchymal stem cells. They found that platelet growth factors can be replaced by serum in three-dimensional culture medium. These factors provide a suitable medium for the growth of mesenchymal cells. Osteogenic differentiation potential of mesenchymal cells is preserved in this medium. In a research conducted by Kargar *et al.,* (2009), the influence of platelet gel in treatment of diabetic foot ulcers was investigated. They concluded that platelet gel can be used as an alternative method in the diabetic foot ulcers and by using platelet gel, it can be expected that most of diabetic ulcers heal and prevent from amputation. The results of the two studies are perfectly aligned and matched with the results of this research that platelet-derived products play antibacterial and regeneration role. Bielecki (2007) from UK performed a research in which the antibacterial effects of platelet gel (PLT Gels) were evaluated in vitro. PRP 20 sample of a volunteer was transformed to platelet gel using thrombin and calcium chloride. Its antibacterial effects were examined using Kirby-Bauer disk diffusion method. His results represented the antibacterial effects of platelet gel against *Staphylococcus aureus* and *E. coli*, but did not show this property against *Klebsiella pneumoniae, Pseudomonas aeruginosa* and *Enterococcus faecalis*. The results of the recent study and even the conducted method by the researcher are quite similar and consistent with the present study. Tohid Nezhad (2012) in his study conducted in Germany, analyzed the compounds inside platelet gel and proved the presence of defensin (2) peptide in this gel using Western blot and ELISA techniques. In continuous he also announced a part of antibacterial effects of platelet gel against gram-positive bacteria caused by activity of this compound in the platelet gel. He next examined the antibacterial property of this gel on a batch of bacteria including *E. coli, Enterobacter faecalis, Pseudomonas aeruginosa* and *Pseudomonas mirabilis*. Platelet gel was announced by him to have inhibitory effects on all of the above bacteria. The results of this study are perfectly aligned and consistent with the results of the conducted research that platelet gel or activated PRP plays antibacterial and inhibitory role on the above bacteria. Drago (2013) from Italy also performed a study in which the antibacterial property of platelet gel and other PRP-derived products was evaluated on a batch of microorganisms isolated from the mouth of 17 patients undergoing various surgeries of teeth and mouth. These microorganisms included *Enterococcus faecalis*, *Candida albicans*, *Streptococcus agalactiae*, *Streptococcus oralis* and *Pseudomonas aeruginosa*. The results of the research on the influence of platelet gel revealed its antibacterial property against all the evaluated bacteria except *Pseudomonas aeruginosa*. This research group announced that platelet gel and other PRP-derived products improve and accelerate tissue repair in surgeries of the oral cavity and moreover they have specific role in protection against pathogens and infectious factors. The results of last study is consistent and aligned with the results of the present study that platelet-derived products play restorative and antibacterial role against bacteria causing infection particularly *Pseudomonas aeruginosa*. Burnouf (2013) from Thailand conducted a study in which the antibacterial activity of various PRP-derived blood products including platelet gel was examined on bacteria-inoculated wounds. The results showed that all *E. coli* bacteria were killed by platelet gel after 48 hours. Other bacteria including *Klebsiella pneumonia, Pseudomonas aeruginosa* and *Staphylococcus aureus* were killed after 3 hours after treatment with other PRP-derived products. In the meantime the activity of platelet gel was weaker than other PRP-derived products and it showed similar results to other products in 48 hours later. Burnouf group finally announced that the activation of prothrombin to thrombin and formation of clots during preparation of platelet gel reduces its antibacterial activity. The results of the last study is opposite and incompatible with the results of this study that PRP is the best platelet-derived products and the highest antibacterial effect is against *Pseudomonas aeruginosa*. Li *et al.,* (2013) from China evaluated tissue repair ability and antibacterial properties of PRP and other PRP-derived products, which are rich in leukocytes on osteomyelitis in a rabbit model. 40 laboratory rabbits were experimentally affected by osteomyelitis caused by *Staphylococcus aureus* species resistant to methicillin by this research group. After 3 weeks and observation of the disease process, PRP was injected by them to the rabbits and diet therapy with vancomycin was also performed. The results showed

that treatment with vancomycin and LPRP had killed Staphylococcus aureus and they achieved complete remission, while the results for PRP and vancomycin were weaker. Although, the results of antibacterial of PRP-derived products were weaker than vancomycin, but it suggested that combination of antibiotic and PRP-derived products can be considered as an alternative treatment with the benefit of tissue repair. Some of results of the last study including the antibacterial property and regenerative role of platelet gel are consistent with the results of the present research. However, this result that LPRP is stronger than activated PRP is opposite to the result of the current study, and perhaps this is due to the use of vancomycin with these products as well as it may be due to the higher amounts of white blood cells in LPRP products and stimulation of them.

Safari *et al.,* (2008) conducted a study and investigated the impact of factors secreted by platelets on proliferation and differentiation of epithelial cells and came to the conclusion that these compounds play an important role in repairing of damaged tissues. The result that platelet-derived products, especially platelet gel have antibacterial, therapeutic and restorative impacts makes this study to be consistent with the mentioned studies. This study also examined the reaction of various bacteria including gram-positive and gram-negative bacteria and different strains of *Pseudomonas* against antibacterial activity of platelet-derived products isolated from human. It became clear that gram-negative bacteria, especially *Pseudomonas aeruginosa* among those bacteria evaluated in this research, showed the greatest influence against these products. The impact of platelet gel on treatment of diabetic foot ulcers was investigated by Kargar *et al.,* (2010). They came to the important conclusion that platelet gel can be used as an alternative treatment in the diabetic foot ulcers to prevent from amputation. The mentioned studies are consistent with this research and the treatment and regenerative role of platelet-derived products is completely clear and the side effects of antibiotics are reduced.

Conclusion

Overall, these results confirm the point that antibacterial effects of platelet-derived products, especially calcium chloride-activated PRP can be used against gram-positive and gram-negative bacteria, especially *Pseudomonas aeruginosa* as an alternative to antibiotics. Of course, its impact on male is more than on female. This method is a biological method that can be used in treatment plans as well as for prevention from nosocomial infections.

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