Article

The impact of onions and figs extracts on *Streptococcus pyogenes* bacteria

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Abstract

Due to spread of infectious diseases and increase of bacterial resistance to antibiotics and side effects of chemical drugs which have expiration date, using herbal and organic drugs with faster effectiveness without side effects and limitation are proper replacement. Maceration to extraction was used to conduct this research in four methods using distilled water and alcohol as solvent and finally these extractions was used to measuring halos of blight. *Streptococcus pyogenes* bacteria were standard and the impact of extractions using impregnating discs on bacteria in Muller-Hinton agar in plate was conducted and their influence compared with required antibiotics and measuring halos of blight were accomplished, which were confirmed by MIC, MBC and FIC. The required extractions from onions and figs obtained by maceration which surveyed as synthetic and single on standard *S. pyogenes* bacteria and their impact on the tested bacteria in some resulted methods were logical compared to 8 used antibiotics and compared with some antibiotics was more, equal and less. Onions and figs extracts, both singly and specially synthetically by maceration on *S. pyogenes* bacteria are the causes of lung infections which have antibacterial feature. Onions and figs extracts synthetically with distilled water traditionally and the extant of drench for 10 days is more influential and has big halo of blight than 8 kinds of antibiotics such as gentamicin, neomycin, azithromycin and erythromycin. Also hypothesizes have been offered to explain the mechanism of their antibiotics feature. The FIC parameter result showed that, combination of figs and onions extractions had synergistic effect. Therefore the combinations of the two extractions are more effective.

Keywords *Streptococcus pyogenes*; onions and figs extract; azithromycin; rifampin; ampicillin; trimethoprim; sulphametoxazole; tetracycline; gentamicin; neomycin; erythromycin; synergistic.

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1 Introduction

Breathing organ is divided into two parts: the upper, and the lower

- Upper breathing organ includes: nose, throat, larynx and trachea.

- Lower breathing organ includes: lungs and their components. Lung includes bronchial, bronchioles and alveolus.

The causes of lung infection can be bacteria, virus and fungus. Parasites also cause infection. Bacterial infection is more than any other infections which are treated by antibiotics.

- Virus infections don't require drugs and automatically are recovered.
- The symptoms of lung infections due to fungus are the same as those of bacteria and virus. In some cases surgery is needed to remove fungus tumor.

One of the current lung infections is *Streptococcus pyogenes*. Streptococcus is positive and hot Cooksey pyogenes which are classified into group A in Lansfyld. When *S. pyogenes* grows on bloody agar, it is the Beta hemolytic. Streptococcus is negative in terms of producing catalase enzyme. Duration of bacteria incubation is 1 to 3 days. It is assumed that 700 million infections are produced by this Bactria annually that 650000 of them are acute. The amount of mortality caused by *S. pyogenes* is 25% (Ray, 2004).

Robecalansfyld invented the approach to serotyping *S. pyogenes* using protein M (surface protein and the main cause of disease in *S. pyogenes*) in 1928 (Lancefield). She designed serological classification on the base of surface protein T (Lancefield and Dole, 1946). So far 100 serotypes T, 20 serotypes M have been recognized in *S. pyogenes* (Facklam, 2002).

S. pyogenes produces different diseases factors. These bacteria can stick on host cells by disease factors, scape from safety system of host or distribute in body (Carroll et al, 2015).

This Bactria causes main diseases in human. Pharyngitis, Impetigo, Erysipelas, Cellulite, Gangrene are some diseases which are resulted from Streptococcus pyogenes. Some levels of bacteria produce toxins which play a role in producing disease. For example in scarlet, toxic shock syndrome, *S. pyogenes* can play a role. It is because of similarity between body tissue and the structure of protein M.

Antibodies which are produced against protein M react with body tissues that result autoimmune disease is produced. Rheumatic fever (heart), and Glomerulonephritis (lung) are autoimmune diseases related to Bactria (Ray, 2004).

Panahi and his colleagues (2013) in the study titled: studying the impact of liquid extraction of summer onion on *Albicans candid* in the laboratory conditions, concluded that densities of 5, 10, 20, 40 mg/ml have little effect on *A. candid*, but in density of 80 mg/ml, blight was more than lower densities (Panahi et al., 2013). Benkbelia and his colleagues surveyed the onion and garlic extractions on *Staphylococcus aureus* and Salmonella in 2004 that has effect on Salmonella (Benkeblia, 2004). Jiong and his colleagues surveyed figs extraction on oral Bactria in 2009 (Jeong et al., 2009). Shams and his colleagues studied Anti-Fungal feature of onion and garlic compared with Ketoconazole (Shams-Ghahfarokhi et al., 2006). Momeni and his colleagues (2010) conducted the study on antimicrobial effects of onion and ginger on albicans bacteria and fungus which isolated from Urine samples of People infected with genitourinary (Momeni and Zamanzad, 2010).

The purpose of this study was surveying the antimicrobial effects of the combination of onion and figs extractions on *S. pyogenes* on pulmonary infections (Machavarapu and Vangalapati, 2015). The problem of resistance against antibiotics which occur for different causes such as misusing, not only causes mortality, but results in production of more new antibiotics. In the other hand undesirable drug side effects is one of the biggest problems in treatment and drug side effects is the fourth cause of mortality in US. According to the above reasons, if we can use the herbal medicines which have proper antimicrobial effect, we take a big step towards solving the problem.

2 Study Area and Methodology

2.1 Preparing intended plant (yellow figs and local onions)

There are a variety of figs, such as *F.aspera*, *F.aurea*, *F. bengamina*, *F. bengalensis*, *F. cordata*, F. *dammaropsis*, *F. deltoidea*, *F. elastic*, *F.lyrata*, *F.pumila*, *F.religiosa*, *F.sur*, *F. stcomorus* (Lansky and Paavilainen, 2010). The figs used in this study with the scientific name Ficuscarica, which is a yellow figs type (Fig. 1).

A variety of onion is yellow onion, white onion, brown onion, red onion, pink onion with the scientific name *Allium cepa*, the type of onion is used in this research, which is pink type.

Fig. 1 Onions and figs.

2.2 Extracting

Extraction of figs and onion was conducted by maceration in four methods (Azmir et al., 2013)**:**

1) Dried figs and onion (powder) in 1/10 ratio
2) Dried figs and onion (powder) in 1/4 ratio
3) Sodden of powder figs and fresh onion in 1/1 ratio
4) Little components of figs and fresh onion in 1/4 ratio

Ratios of onions to figs were chosen by author observation, and the ratios of 1 to 4, 1 to 10, 1 to 1 were used in four ways (Table 1).

Method 1

The used solvent was distilled water and figs and onion were dried and powdered, then they were drenched for 10 days that during this time they were combined. In this method the ratio of substance and solvent was 1/10, that is 100 g onion, 1000 cc distilled water and 100 g figs, 1000 cc distilled water.

Then this solution was poured into 50 cc falcon tubes and was centrifuged in 2000 rpm for 10 minutes in temperature (20°C). The surface, pure and lucid solution was isolated and poured into the balloon of Rotary devices to isolate its solvent that is distilled water from extraction.

In this method Rotary devices took some hours for figs and onion and it was put in 10 to 120 rounds and 45 °c temperature and cooling valve of water was opened and vacuum device was turned on.

Method 2

In this method figs and onion were combined by digital scale in 1/4 ratio (20 mg of figs, 80 cc of distilled water, 20 mg of figs and 80 cc of alcohol), (20 mg of onion, 800 cc of distilled water, 20 mg of onion and 80 cc of alcohol) and were poured in beaker and their door were closed by paraffin and to keep out of reach of light, they were covered by paper and kept on shaker for 48 hours, then the extractions were centrifuged in 2000 rpm for 10 minutes and surface solution were rotated and solvent was isolated.

Method 3

The solution was distilled water that 150 g figs, 150 g onion with 300 cc distilled water which were combined and boiled and kept for some minutes, then were heated and after 8 hours they were extracted and poured into rotary device to isolate their solvent.

Method 4

In this method fresh and crushed components were drenched in 1/4 ratios with two kinds of solvent that is alcohol and distilled water and were poured into beaker and were put on shaker for 48 hours, and then they were centrifuged and rotated.

2.3 Preparation of their sample or vastok bacteria

Preparation of fresh cultivation (logarithm phase) was made from microorganism.

2.3.1 The way of testing disk diffusion

At first raw disks were put in the sterile empty plates and 20 µl of prepared extractions were poured on them and were heated for 1 hour in 60C° in oven. The examined extractions synthetically and 8 extractions were obtained and they were poured on 8 raw disks.

Table 1 Extractions for testing disk diffusion method.

Methods	Description
Method 1 (traditional way)	1) watery figs extraction 2) watery onion extraction 3) The combination of two extractions
Method 2 (the second series)	4) Figs and onion extraction in ¼ ratio 5) Figs and onion extraction in ¼ ratio with alcohol
Method 3 (the third series)	6) Traditional figs and onion extraction
Method 4 (fresh components)	7) Alcoholic figs and onion extraction in ¼ ratio 8) Alcoholic figs and onion extraction in ¼ ratio

Bacteria were poured into test tube that includes normal saline solution, so that its color gets like Half McFarland which was conducted by Spectrophotometer (Harrigan and McCance, 2014). Then bacteria were cultured fully into 8 plates containing Mueller-Hinton agar medium by swap from test tube and the disks containing extractions were put into these plates, and then they were placed in Incubator in 37C° for 24 h. After 24 h the plates were examined from which 5 plates had halos of blight and 3 plates had little halos and the former were cultured again (Jorgensen and Turnidge, 2015).

2.3.2 Examining MIC and MBC

Three plates which had high and average halos were isolated to examining MIC and MBC. 1cc of Mueller-Hinton agar medium were poured into 12 test tubes and then 1cc of the required extraction was added to tube one to dilution and then1cc of bacteria (the bacteria that was combined with saline solution and Half McFarland in test tube) was poured in all tubes and were placed in Incubator in 37C° for 24h.

2.3.3 FIC

$$\mathrm{FIC} = \frac{\text{MIC of antibacterial A in combination}}{\text{MIC of antibacterial A alone}}$$

FIC has the same formula for B and finally:

$$\sum \text{FIC} = \text{FIC of antibacterial A} + \text{FIC of antibacterial B}$$

FIC for 1, 2 and 3 extractions is:

$$\sum \text{FIC} = 0.624$$

According to the FIC parameter the result show that, combination of figs and onions extractions have synergistic effect. Therefore the combinations of the two extractions have more effect (Habiba et al., 2015).

2.3.4 Antibiogram test

At the end of research, Antibiogram test was conducted and antibiotics which in plates containing Mueller-Hinton medium in which bacteria was cultured, were used and after 24h placing in incubator, the growth halos were measured by caliper (Table 4 and Fig. 2).

3 Results

Effluence of disc watery figs extraction resulted from method 1 has greater blight halo than synthetic extraction of watery figs, and onion resulted from method 1 and this has greater halo than synthetic extraction of watery figs and onion resulted from method 4 (Table 2).

Table 2 The halo of blight.

big halo	**3 cm**
average halo	2.7cm
little halo	2.3 cm

3.1 MIC and MBC

The effective extractions on *S. pyogenes* have the least attractive and inhibitory density on the base of the Table 3.

Table 3 The least attractive and inhibitory density on effective extraction in 100 mg/ml^3.

	MIC	**MBC**
Synthetic extraction of watery figs and onion of method 1 (big halo)	3.12	25
watery onion extraction of method 1 (average halo)	6.25	25
extraction of watery onion of method 1 (little halo)	25	50

3.2 Antibiogram test

The results of antibiogram test are listed in Table 4.

Table 4 The blight halo of antibiotics.

Antibiotic	Size of blight halo in cm
Azithromycin(AZI)	2.2
Rifampin(RA)	4
Ampicillin(AM)	4.5
trimethoprim, sulphametoxazole(SXT)	4
Tetracycline(TE)	3.5
Gentamicin(GM)	2
Neomycin(Ne)	Without halo
Erythromycin (E)	2.2

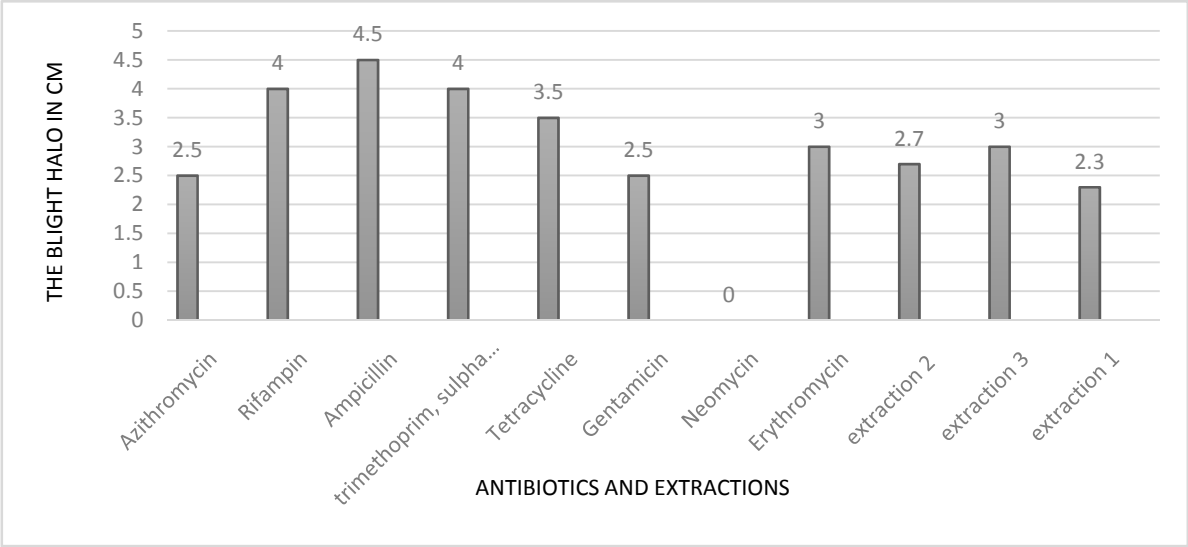

Fig. 2 The blight halo of antibiotics and extractions.

4 Conclusions

The amount of antibacterial effect of watery onion extraction of method 1 (Fig. 3-B), the extraction of watery fig of method 1 (Fig. 3-C) and synthetic extractions of watery figs and onions of method 1 (Fig. 3-A) on *S. pyogenes* is more than antibiotics such as Gentamicin, Neomycin, Erythromycin and Azithromycin, and the amount of antibacterial effect of all watery extractions like method 4 is more than Neomycin.

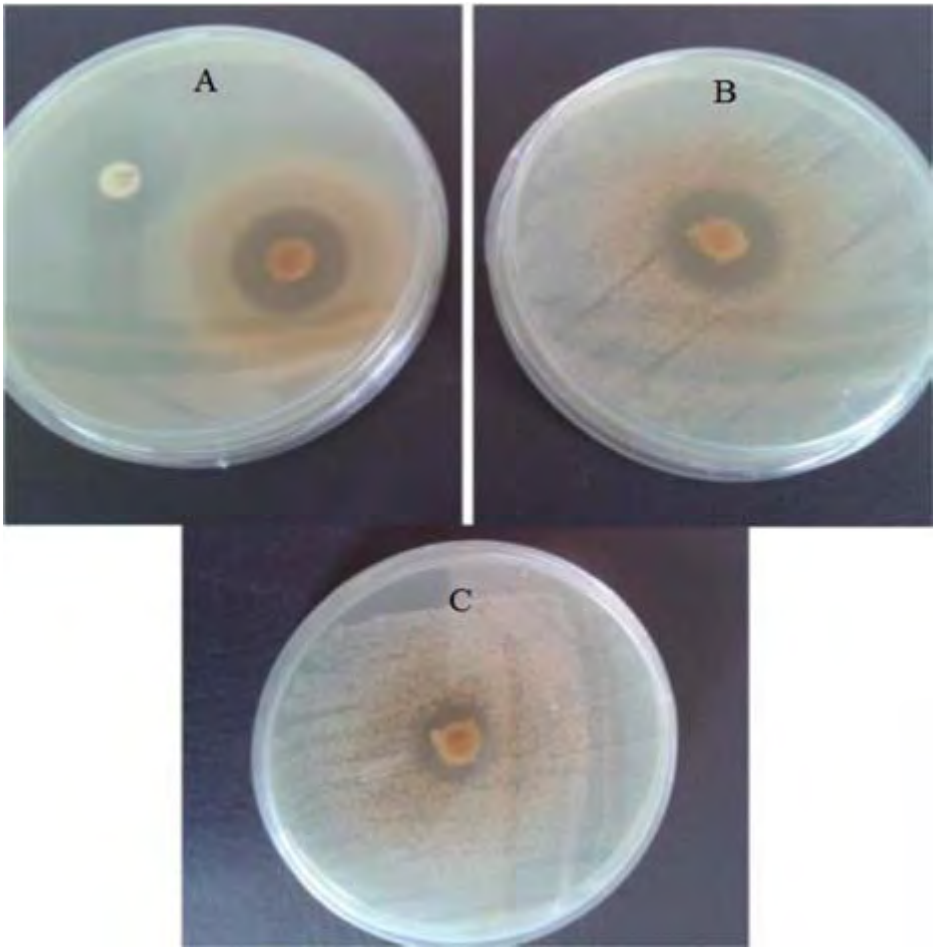

Fig. 3 The antibacterial effect of extractions.

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