The Role of Staphylococcus Haemolyticus in Men Infertility

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Abstract. A total of 80 samples of seminal fluid from infertile men who attending to admitted to clinics and laboratories in Omara city during the period from 2016/6/1 to 2016/12/1 , samples were subjected to semen analysis as recommended by WHO. The age of patients from 20 - 59 year. Another 25 semen sample were collected from fertilized men considered as control group. For this purpose, seminal fluid was cultured on MacConkey agar, Blood agar, Chocolate agar. within the 80 samples recorded S. haemolyticus in 14 cases and the rate of infection 18%, all strains was diagnosed by Vitek system 2 Double. The highest percent (64%) record in age group (30 – 39). Primary infertility recorded 94% while 6% recorded for secondary infertility. Also, different species of bacterial isolates were identified in 32 cases by Vitek system 2 double. The bacterial infection of men genital system affected on fertility.

KEYWORDS: Infertility, semen analysis, genital pathogens

1-introduction
Infertility it means the inability to have a child after at least 12 months of unprotected sexual intercourse without using any contraceptive or condom use and affect the cause psychological and social of both spouses (Deka and Sarma , 2010). The incidence of male infertility means of a problem in the genital tract lead to an imbalance and lack of fertility (Askinazy-Elbhar,2005). Staphylococcus haemolyticus is a member of the coagulase – negative (Bertin  et al., 2004). It is a well- known opportunistic bacterial pathogen that is well-known for its highly antibiotic-resistant phenotype (Takeuchi et al., 2005). As a gram- positive Staphylococcus haemolyticus has a thick peptidoglycan wall outside (Billot- Klein et al., 1996). And it is highly cross- linked and reported cases of infections caused by S. haemolyticus dysfunction of organ systems resulting from immune response to a severe infection (Takeuchi, 2005). Primary infertility is infertility which affects the woman or man since the beginning of their lives of marital sexual and caused either hormonal or not integration organisms sexual for caused congenital (La vignera , et al.,2012).As for infertility secondary it is the infertility that affects woman after having a child or having a pregnancy end abortion child or ectopic pregnancy resulting from bleeding after birth or when the female is in a period of breastfeeding or because of the presence of infections resulting from bacteria . Men infertility occurs due to the inactivity of sperms and their inability to penetrate the membranes of the egg (Kay and Robertson, 1998).

2-Materials and Methods:
This study included the collection of seminal fluid (100) a married men infected infertility aged (20-59 year) 80 infertile men and 20 fertile men is considered control by collecting samples in sterile plastic (cup at the hospital) according to the instructions of where to abstain from ejaculation for at least 48 hours before
a routine examination of a sample of semen and then placed in the incubator for 15 - 30 minutes at least by converting it from a glass to the transparent liquid may be more than one hour if the high viscosity. Included measurements of volume sperm count and movement of sperms and type of sperms been collecting samples from reviewers to clinics and laboratories in Maysan - Iraq. As shown as in fig (1), well culture all samples on media (Blood, Chocolate, MacConky agar) and it was diagnosed by Vitek 2 system. Rapid identification (three hrs), a high level of automation, a simple methodology and taxonomically updated databases (Cheng et al., 2010). All men were instructed to abstain from sexual intercourse or masturbation for 5 days before attending the clinic up arrival at the clinic the men were asked to collect samples by masturbation on site into standard sterile containers know to be free of cystic effects on human spermatozoa (WHO,2010). Before semen collection the men were asked to wash their hands and genitalia with running water and soap. Soon after semen production the samples were promptly liquefied at 37 °C for about 30 minutes in an incubator before analysis. Finally, statistical analysis was done spss.

![Flow chart showing isolation and identification of Staphylococcus haemolyticus](image)

**Figure (1)** flow chart showing isolation and identification of *Staphylococcus haemolyticus*

### 3-RESULTS:

The results of the current study showed that can have isolated and diagnosis Staphylococcus haemolyticus of semen by Vitek 2 system, from (80) samples of seminal fluid of infertile men who the reviewers to the clinics and the private laboratories in the province of Maysan. Staphylococcus haemolyticus was diagnosed in 14 patients with rate 17.5%. When analysis semen for patients which infected by this bacterium their effect appeared on the size and nature semen and as well as on the number and efficiency of the tents the appearance of these bacteria also led to the appearance of abnormal sperm. Staphylococcu
haemolytic infection was affected bacteria in quantity and quality of sperm. Colonies were appeared after (24-48) hours and have coccis shape as shown in the 'figure2', 'figure3', 'figure4'.

**Figure 2.** colonies *Staphylococcus haemolyticus* on the blood agar medium.

**Figure 3.** colonies *Staphylococcus haemolyticus* on the chocolate agar medium.

**Figure 4.** colonies *Staphylococcus haemolyticus* on the MacConkey agar medium.

The colonies were stained with gram stain and it appeared Gram positive with purple color as show in 'figure 5'.

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Figure 5. Staphylococcus haemolyticus colonies stained with gram stain (100X).

Many bacteria were isolated from seminal fluid of infertile men (table 1). High percent of isolation from S. haemolyticus.

Table 1. bacterial which isolated from patients.

| Type of bacteria               | Number | Percent |
|-------------------------------|--------|---------|
| Staphylococcus haemolyticus   | 14     | 17.5%   |
| Enterobacter cloacae &        | 7      | 8.7%    |
| Escherichia coli              |        |         |
| Klebsiella pneumonia          | 6      | 7.5%    |
| Pseudomonas aeruginosa        | 6      | 7.5%    |
| Enterococcus faecium          | 4      | 5%      |
| Morganella morganii           | 2      | 4%      |

The volume of patient’s semen was ranged between 1.5- 3 ml. The infection was affected on appearance of abnormal sperm with high numbers. On the type of sperms, the infection was affected because type A (Active) sperms recorded low in numbers in contrat with of types B & C (Butti & Colorless movement) also, type D (Dead) sperms recorded low percent. (table 2).

Table 2. the influence of infection with Staphylococcus haemolyticus on seminal fluid analysis of infertile men.

| Count | Volume | Normal | Abnormal | A | B | C | D | RBC. | Pus |
|-------|--------|--------|----------|---|---|---|---|------|-----|
| 1     | 0      | 1.5    | 5        | 95 | 5 | 0 | 95 | 0    | 1   |
| 2     | 10^10  | 1.5    | 15       | 85 | 15| 20| 35 | 30   | 1   |
| 3     | 10^10  | 1.5    | 20       | 80 | 15| 30| 20 | 20   | 1   |
| 4     | 10^10×15 | 1.5     | 20       | 80 | 20| 40| 20 | 20   | 1   |
| 5     | 10^10×65 | 1.5      | 20       | 80 | 20| 25| 35 | 20   | 2   |
| 6     | 10^10×20 | 1.5     | 30       | 70 | 25| 65| 5  | 5    | 2   |
| 7     | 10^10×20 | 1.5     | 35       | 65 | 30| 30| 10 | 30   | 2   |
| 8     | 10^10×30 | 1.5     | 35       | 65 | 30| 40| 15 | 15   | 3   |
| 9     | 10^10×35 | 1.5     | 25       | 75 | 40| 20| 15 | 25   | 4   |
| 10    | 10^10×40 | 2.0     | 40       | 60 | 45| 40| 5  | 10   | 4   |
Type of infertility was determining in current study; Primary infertility percent was 94% while secondary infertility recorded only 6%. Among primary infertility cases a 79% found infected with \textit{Staphylococcus haemolyticus} and 21% in secondary infertility.

Patients were divided in four age groups. The high percent (64%) recorded in patients with age group (30-39). (table -3-).

| Age   | Number | Rate |
|-------|--------|------|
| 20-29 | 2      | 14%  |
| 30-39 | 9      | 64%  |
| 40-49 | 2      | 14%  |
| 50-59 | 1      | 7%   |

Table -5- explain the patient’s disease which recorded in this study beside infertility. A (4) patients have diabetes and (2) suffered with thyroid gland disease.

| Sickness | Number | Rate |
|----------|--------|------|
| Diabetes | 4      | 28%  |
| Thyroid  | 2      | 14%  |

Patients wives of study participants have infection with bacteria a 93.7% percent. Distributed patients among residents the province center Mayasan elimination and aspects, as recorded found \textit{Staphylococcus haemolyticus} in elimination and aspects more than the center of the civil, in elimination 9 infections (64.2%) and the center 5 infections (35.7%).

Finally, all medical examination showed statistically significant differences in mean (P<0.05).

4-Discussion:
The role of the genital tract microorganisms important factor in male infertility (Keck et al, 1998; Ramesh et al., 2004). Bacteria is transmitted to the genital tract of the urinary tract or from the digestive system due to the proximity of the opening device of the output of the reproductive system as it becomes opportunistic to pathogenic and caused infections in the epididymis prostate and semen and testicles and thus infertility (Alausa and Osoba, 1978; Hobson et al., 2013). More that presences bacteria in semen which cause infertility represented in total bacteria \textit{Staphylococcus} spp. (Cunningham and Beagley, 2008; Fraczek et al., 2012). Identified the current study bacteria \textit{Staphylococcus haemolyticus} in the rate 17.5% the presence of these bacteria it works paralysis in sperm. As it penetrates of sperm and break it excretion enzymes which on the occurrence of deformities (Veznik et al., 2004). In addition to the negative impact on the secretion of the gonads and its analysis of blood and appearance of RBC., WBC. and agglutinating of sperm (Qusada et al., 1968; Alekwe et al., 2013). More bacteria are present in samples she \textit{Staphylococcus haemolyticus} and Escherichia coli in terms of influence on movement and morphology of sperm (Prabha et al., 2010). Also the presence of \textit{Enterobacter cloacae} and \textit{Pseudomonas aeruginosa} influenced in appearance shapes of abnormal and lack of movement (Merino et al, 1995; Buonmimo et al, 1999). It was appearance of \textit{Klebsiella pneumoniae} and \textit{Enterococcus faecium} and \textit{Morganella morgani} role in infertility by
influencing the movement, quantity and quality of sperm (Claire et al., 2013). That presence *Staphylococcus haemolyticus* come through the proper environment for semen because it contains nutrients and this is basic basis in the presence of bacteria (Shalika et al., 1996).

Primary and secondary infertility occurs by having bacteria causing adverse and adverse changes in sperms chromosomes thereby causing abnormal and abnormalities of sexual chromosomes represented chromosomes(Y) as the bacteria work to lose semen sperm (Samara et al., 2011). Infertility increased in age groups (30-39). It is companion to the presence of bacteria *Staphylococcus haemolyticus* because as a defect in the function sex gland and the descent of abnormal secretions changed the characteristics of semen it is a period of sexual activity (Weng et al., 2014). Also recorded the current study accompanying bacteria with diabetes most people with diabetes suffer from cystitis which causes difficulty in dividing the bladder it is difficult to sweep the bacteria outside the body he added that high blood sugar is an appropriate environment for the presence of bacteria diabetes inhibits immunity which is caused by dysfunction in phagocytic cells (Jeanette et al., 2005). Thyroid diseases if caused by thyroid diseases lack of secretion of male hormone Testosterone (Paavonen, 1998). The current study was associated with infertility with the presence of bacteria in the districts more than the center of the city to raise health awareness (Lewis et al., 1981). 93.7% is the proportion of women infected with infections and because of sexual contract to infection or diseases in one of the spouses will move to the other partner and case to the diseases and thus infertility (Samara et al., 2011).

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