



ORIGINAL ARTICALE

**GINGIVITIS: ISOLATION AND DIAGNOSIS OF THE CAUSATIVE BACTERIA AND ITS
RELATIONSHIP TO OBESITY IN AL -DIWANIYAH GOVERNORATE**

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ABSTRACT

Background: Seventy clinical samples were collected from gingivitis patients between 3/1/2025 and 30/3/2025 under the supervision of a specialized physician. Samples were obtained from professional dental facilities in Al-Diwaniyah Governorate. The study included 29 male patients (41.43%) and 41 female patients (58.57%), with ages ranging from 7 to 80 years.

Objectives: The aim of this study was to investigate the prevalence and types of bacterial isolates in gingivitis patients and to examine the relationship between gingivitis and obesity.

Results: Out of 70 clinical samples, 55 bacterial isolates (78.57%) demonstrated growth. Microscopic examination with Gram staining showed that 44 isolates were Gram-positive (80% growth rate), including 28 streptococci, 11 staphylococci, and 5 Enterococcus faecalis. Eleven isolates were Gram-negative, with a growth rate of 20%. The findings indicated that bacteria associated with gingivitis were more prevalent in obese patients, with a bacterial growth rate of 70.91%, compared to 29.09% in patients with gingivitis only.

Conclusion: The study demonstrates a clear relationship between gingivitis and obesity, indicating that bacterial prevalence is higher in obese patients with gingivitis. These findings highlight the importance of considering obesity as a risk factor in the management and prevention of gingivitis.

Keywords: Enucleation, Healing Time, Marsupialization, Pediatric Dentistry, Recurrence Rates

INTRODUCTION

The mouth, like other areas of the body, or possibly more so, is distinguished by the presence of microorganisms in the majority of its components. These organisms have a balanced interaction with their hosts, but this relationship can worsen in the mouth, resulting in illness. This is frequently related with changes in the biology of the mouth. These alterations may be caused by external factors such as antibiotic treatment or an increase in the amount of fermentable carbohydrates in the diet. Or it could be internal sources, such as changes in the integrity of the host defenses following pharmacological treatment, which impact the usual presence of bacteria and possibly the existence of germs in difficult-to-reach areas. For example, when a person's teeth are pulled due to gum inflammation, the germs causing the inflammation are

transmitted into the bloodstream and spread to other organs of the body, resulting in secondary disorders. This sort of bacteria is known as opportunistic pathogens. Many oral bacteria can behave this manner. At some time in their lives, many people suffer from an imbalance of the natural bacteria in their mouth ¹. Oral bacteria are ordinarily in a state of dynamic equilibrium, and when this balance is upset, the natural flora in the mouth is disrupted, resulting in oral disorders ². Gingivitis and tooth decay are the two most frequent and widespread oral illnesses in the world ³.

Periodontal disease refers to a range of disorders that affect the tissues surrounding a tooth, Gum disease is the major cause of tooth loss and is very prevalent in developing countries, Gingivitis is a reversible inflammatory response due to the presence of dental

plaque. The gums usually return to their normal state before the infection if oral hygiene is taken care of. The infection is chronic and more severe in people who suffer from hormonal disorders, such as during pregnancy and puberty⁴. Gingivitis is a chronic inflammatory illness caused by the interaction of gum bacteria and the host's reaction to that contact⁵. This raises the risk of obesity, diabetes, and perhaps rheumatoid arthritis^{6,7}. In 2010,⁸ proposed a relationship between gingivitis and obesity. In 2011,⁹⁻¹² shown the tight relationship between obesity and gingivitis. Obesity is a chronic metabolic condition characterized by the abnormal buildup of fat¹³. Poor eating habits, as well as genetic, behavioral, cultural, social, and technological influences, all contribute to weight gain¹⁴. Obesity has a number of immediate life-altering repercussions, including stroke, osteoporosis, diabetes, gout, gallbladder disease, and coronary heart disease^{15,16} demonstrated that non-vital gut microorganisms have a significant role in obesity by enhancing insulin resistance produced by Gram-negative bacteria.

MATERIALS AND METHODS

2-1 "Clinical specimen collection"

70 clinical samples were gathered from patients with varying weights and disease lesions from specialized dental centers and private clinics in Al- Diwaniyah Governorate. The collection cases included inflammation and swelling of the gums of both sexes and of different ages.

Samples were gathered with sterile cotton swabs under the supervision of a specialized physician before being transferred to the scientific laboratories at Al-

Diwaniyah Teaching Hospital.

2-2 "Isolation and identification of bacteria" 2-2-1

'Culture diagnosis'

The initial stage in identifying bacteria was to culture

clinical samples on different medium using a planned approach appropriate to each bacterial species being studied. The plates were then incubated at 37°C for 18-24 hours. The diagnosis is made based on the morphological properties of bacterial development, such as colony size, shape, color, odor, and the ability to ferment lactose and hemolyze¹⁷.

2-2-2 ' Microscopic examination' A typical bacterial colony was transferred onto a glass slide on which a drop had previously been deposited. It was then spread across the slide and allowed to dry. Following that, the slide was exposed to a flame and stained with Gram stain. Following that, the bacterial cells were inspected using a light microscope at a magnification of 1000 to see the color, shape, and reactivity to the dye¹⁸.

RESULTS AND DISCUSSION

'Clinical specimen collection'

70 clinical samples were taken from gingivitis patients between 3/1/2025 and 30/3/2025, under the observation of a specialized physician. Samples were gathered from professional dental facilities in Al- Diwaniyah Governorate. The collecting process includes 29 clinical samples from men (41.428% of the total sample) and 41 clinical samples from women (58.571% of the total sample). Males and females' ages ranged between (7-80) years, as seen in Table 1. And, with varied weights, it was discovered during isolation that those with obesity and excessive weight gain are more vulnerable to gum inflammation than others, which is consistent with the findings of the researcher¹⁹.

Table 1. The collection of clinical samples from persons with gingivitis of both sexes, of various ages and weight.

| Number of clinical samples | Females | The age | The weight | Males | The age | The weight |
|----------------------------|---------|---------|------------|-------|---------|------------|
| 10 | 6 | 7-10 | 30-55 | 4 | 8-11 | 25-50 |
| 11 | 7 | 10-12 | 35-60 | 4 | 9-12 | 30-45 |
| 9 | 4 | 13-15 | 50-70 | 5 | 15-17 | 45-70 |
| 8 | 5 | 16-22 | 60-85 | 3 | 17-20 | 65-90 |
| 11 | 5 | 23-30 | 80-90 | 6 | 25-40 | 80-105 |
| 11 | 9 | 40-70 | 75-85 | 2 | 70-75 | 95-100 |
| 10 | 5 | 50-75 | 90-100 | 5 | 75-80 | 90-107 |
| 70 | | | | | | |

'Culture diagnosis and microscopic examination'

The findings of the culture diagnosis showed the growth of 55 bacterial isolates out of 70 isolates, with a growth rate of (78.571%), there were isolates, including anaerobic, spherical growth in the form of short chains, single and double spheres. This demonstrated its capacity to grow in the selective medium Mitis Salivarius agar, which is used to isolate streptococci. These colonies grew clearly on the medium and had a transparent blue tint. The size ranged from 0.5 to 2 mm, and they were both regular and irregular in shape, with some adhering to the medium and others not adhering (figure 1). The longer the anaerobic incubation period, the easier it was to identify the bacteria. This is consistent with the findings of the researcher ²⁰, who identified Staphylococci, Streptococci, Corynebacterium, Lactobacillus, and other anaerobes as the bacteria responsible for gingivitis and oral illnesses. Other bacterial colonies demonstrated the ability to grow on a medium Eosin methylene blue and under aerobic circumstances, forming small, flat, dark-green colonies with a metallic shine due to their ability to ferment lactose. These colonies were *Escherichia coli* (Figure 2), which is comparable to what was achieved by ²¹. Another bacterium was characterized by its capacity to grow in medium MacConkey agar, a differential medium. The bacteria that live on it can digest lactose and generate vivid pink mucous colonies. It was demonstrated that they are *K. pneumoniae* (Figure 3). This is what the researcher ²² demonstrated. Other bacterial colonies demonstrated their ability to thrive on the medium Cetrimide agar, producing bacterial colonies with blue-green and greenish-yellow colors and shapes. They were smooth, with a high core and flat corners, and smelled like grapes. After diagnosis, it was determined that they were *P. aeruginosa* (Figure 4). This outcome corresponds to what the researcher described ²³. Other bacteria grew in the medium Mannitol salt agar and formed smooth, elevated, circular colonies. Based on the physical characteristics of the colonies developing on the medium, the bacteria were identified as (staphylococcus). In addition to their ability to withstand salt concentration, yellow colonies emerged. After diagnosis, it was discovered that they were (*S. aureus*) and other pink-colored colonies that did not ferment lactose sugar. It was discovered that these were (*S. epidermidis*) (Figure 5) ²⁴. The bacterial isolates that were able to grow on the medium Esculin Agar, forming small white colonies with a circular shape, have the ability to turn the color of the medium to black due to their ability to secrete an enzyme that breaks down the esculin substance in this medium, forming the esculin compound, which combines with the iron ions present in the medium in the form of iron citrate, forming a black compound indicating the growth of *Enterococcus faecalis*. (Figure 6) ^{25,26}.

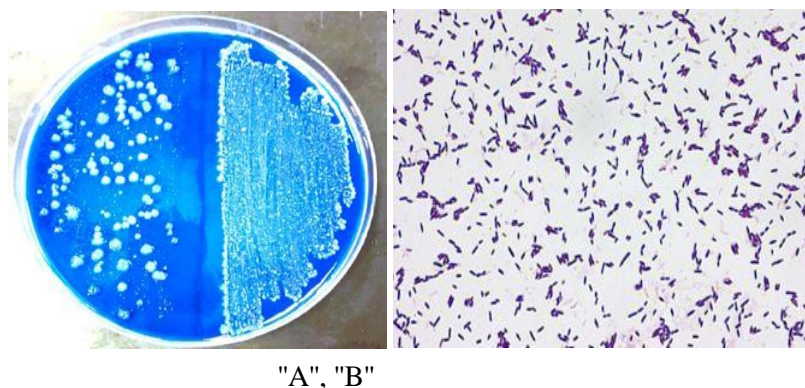


Figure1. 'A' *S. mutans* on mitis salivarius agar 'B' *S. mutans* under the microscope.

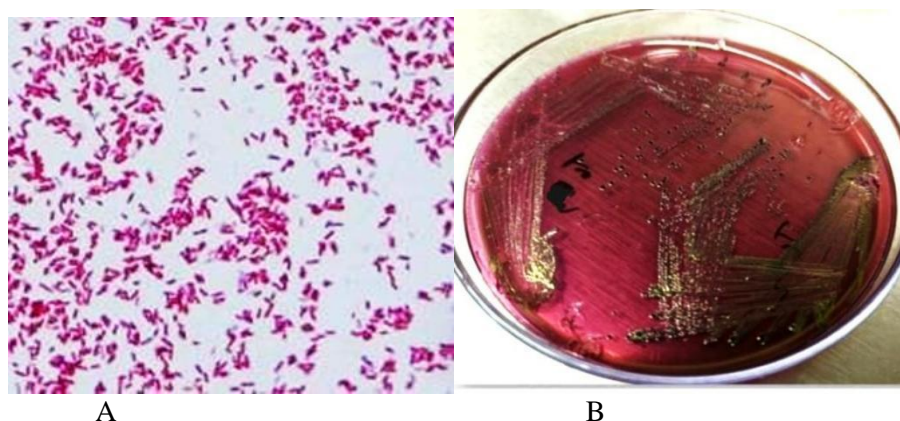


Figure2. 'A' *E. coli* on Eosin methylene blue 'B' *E. coli* under the microscope.

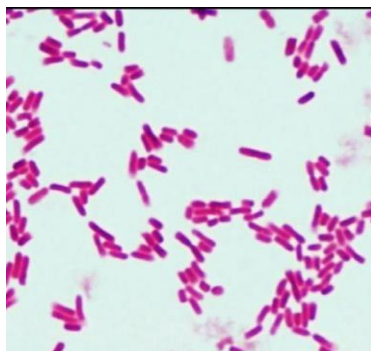
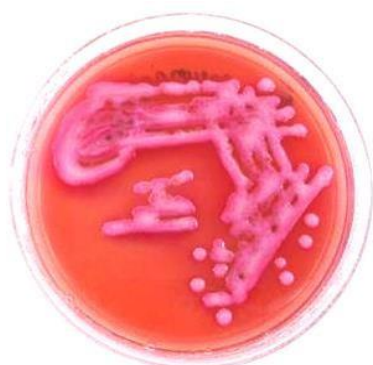


Figure 3. 'A' *K. pneumoniae* on MacConkey agar 'B' *K. pneumoniae* under the microscope.

"A"

"B"

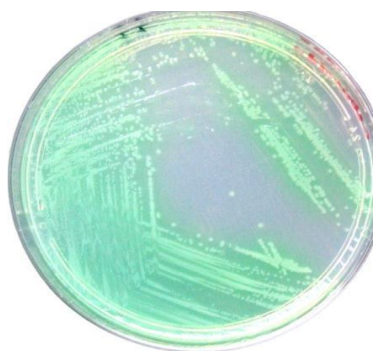
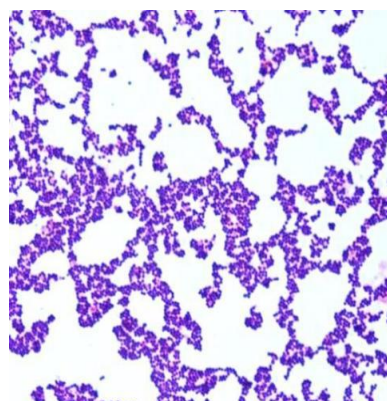


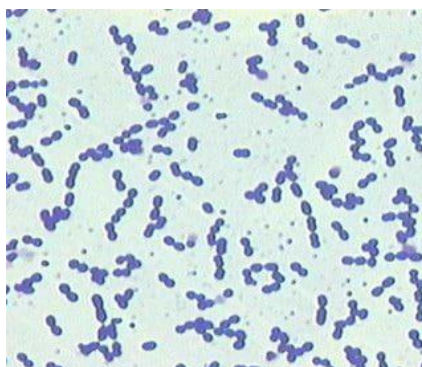
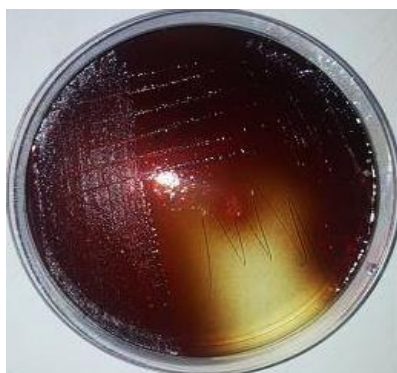
Figure 4. 'A' *P. aeruginosa* on Cetrimide agar 'B' *P. aeruginosa* under the microscope.



A

B

Figure 5. 'A' *S. aureus* on Mannitol salt agar 'B' *S. aureus* under the microscope.



"A"

"B"

Figure 6. 'A' *Enterococcus faecalis* on Esculin Agar 'B' *Enterococcus faecalis* under the micro

Following the culture diagnosis, a microscopic inspection with Gram stain was done on all of the developing isolates, which numbered 55 out of 70 and had a growth rate of 78.571%. Microscopic investigation, as shown in Table 2, revealed that 44 bacterial isolates tested positive for Gram stain, with a growth rate of 80%. 28 of the isolates were streptococci. This finding is consistent with ²⁷, which concluded that streptococci are the primary bacterial cause of gingivitis and tooth decay. 11 isolates were staphylococci, which contradicts the researcher's ^{28,29} claim that staphylococci are the predominant bacteria in gingivitis. Furthermore, Streptococcus bacteria induce opportunistic infections when combined with other Streptococcus species found on the skin, digestive tract, and nose ^{30,31}. They also cause serious hospital infections ³². This is owing to bacteria having quick resistance mechanisms that allow them to spread easily, such as plasmids during conjugation and transformation, it also has enzymes and surface antigens that allow it to infiltrate the body's tissues, similar to staphylococcus bacteria ³³. Gram-negative bacteria were found among the bacteria causing gingivitis, but at a lower rate than Gram-positive bacteria. Microscopic investigation revealed that 11 of the 55 bacterial isolates are Gram-negative, with an average growth rate of 20%. This is consistent with previous research, which has shown that the majority of Gram-negative bacteria originate from digestive and respiratory tract illnesses ^{27,34}.

Table 2. Number of bacterial isolates diagnosed by microscopic inspection and isolated from pathological illnesses characterized by gingivitis

| Bacterial isolates organized by genus and species | Microscopic examination | | number |
|---|-------------------------|---------------------|--------|
| S. mutans | Gram positive | | 28 |
| S. aureus | Gram positive | | 11 |
| E. faecalis | Gram positive | | 5 |
| E. coli | Gram negative | | 6 |
| P. aeruginosa | Gram negative | | 4 |
| K. pneumonia | Gram negative | | 1 |
| Total number of isolates | Gram positive 44 | Gram negative 11 | 55 |

Table 3 shows that bacteria that cause gingivitis are more prevalent in patients who are obese and have gingivitis. The growth rate of bacteria causing inflammation in people with gingivitis and obesity was 70.909%, which is higher than the rate of bacteria isolated from people with gingivitis only, which was 29.090%. The current investigation demonstrated the existence of a relationship between gingivitis and obesity. Obese people are more likely to develop gingivitis. This finding was reinforced by early research, including the claim ³⁵ that obesity increases the chance of developing gingivitis, indicating a faster and more visible development in overweight individuals. This is similar to what was stated by ³⁶, where it was proven that the risk of developing gingivitis is linked to an increase in the body mass index, ³⁷ also reported that excessive obesity increases the causes of diseases, ³⁸ suggested that obese individuals have adipose tissue that is a storehouse of inflammatory cytokines, which increases the likelihood of developing periodontitis, whereas ³⁹ reported difficulty in interpreting the association between obesity and gingivitis based on several observational studies.

Table 3. Depicts the connection between gingivitis and obesity

| Isolated bacterial species | Gingivitis + obesity | Gingivitis |
|----------------------------|----------------------|---------------|
| Gram-positive bacteria | Number of isolates | |
| S. mutans | 20 | 8 |
| S. aureus | 8 | 3 |
| E. faecalis | 3 | 2 |
| Gram-negative bacteria | Number of isolates | |
| E. coli | 4 | 2 |
| P. aeruginosa | 3 | 1 |
| K. pneumonia | 1 | |
| The total | 39 70.909% | 16 29.090% |

CONCLUSIONS

Among the many bacterial genera that live in the mouth, gram-positive streptococci are the most common cause of gingivitis. When conditions are favorable, they secrete acids, which cause a decrease in Hp levels and gingivitis. Obese and overweight individuals are also more likely to develop gingivitis and tooth decay.

Recommendations

We advocate performing a study on the wrong eating habits that contribute to gingivitis. We also recommend conducting a comprehensive study on foods that cause obesity and avoiding them due to their impact on oral health in particular and overall health, as well as conducting a comprehensive study similar to the current one to detect other types of bacteria that may cause oral infection. Raising knowledge about the importance of the mouth and how to protect it from gum inflammation and tooth decay by regular cleaning, avoiding foods that trigger inflammation, and frequent dental checkups.

DECLARATIONS

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Competing Interests

The authors have no competing interests to declare.

Ethical Approval

The study was approved by the appropriate ethics committee and conducted according to relevant guidelines and regulations.

Informed Consent

Not applicable.

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