International Journal for Multidisciplinary Research (IJFMR)
E-ISSN: 2582-2160 ● Website: www.ijfmr.com ● Email: editor@ijfmr.com

Isolation And Identification of Bacteria Present on Spectacles

Dr. Siddharth N. Mendhe[1], Jivan Ingle[2]

Department of Microbiology, Shri Shivaji Science and Arts College, Chikhli, Dist. Buldhana.

Abstract:

The human eyes have a special anatomy that makes them vulnerable to infectious agents, including bacteria, as well as direct environmental exposure. The eyelids, conjunctiva, and cornea are the main parts of the eyes that are most frequently infected. The numerous microorganisms have been linked to eye infections caused by microbial contamination and colonization of eyeglasses. A study reported the presence of bacterial contaminants on spectacles used by students at Shri Shivaji Science and Arts College Chikhli, in July 2023 to August 2023, Such as *Pseudomonas sp.*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, and *Bacillus sp*. Total 40 samples are collect from different students such as 14 samples are collected from 11th Students, 12 samples are collected from 12th Students, 08 samples are collected from B.Sc. Students and 06 Samples collected from M.Sc. Students were taken by using sterile cotton swabs that had been moistened with sterile distilled water. After that it was inoculated on the Blood agar and MacConkey agar by streak plate method and incubated for 24 hours at 37°C. After incubation for 24 hours at 37°C, for confirmed the growing microorganism by using different tests such as Gram Staining, Hanging-Drop method, IMViC test, Carbohydrate fermentation, Enzyme production (Catalase Test and Oxidase Test), and Triple Sugar Iron Agar test are performed as per required for confirmation. All samples shows positive results, when we examined results, it shows different bacteria are present such as *Bacillus sp.* (55%), *Pseudomonas sp*. (42.50%), *Staphylococcus aureus* (22.50%), and *Staphylococcus epidermidis* (12.50%). The results shows that the there was contamination by pathogenic bacteria on spectacles used by the students of Shri Shivaji Science and Arts College Chikhli. It means spectacles provide suitable condition for survive of pathogenic bacteria then its leads to the cause of eyes infection.

Keywords*:* spectacles, Bacillus sp., Pseudomonas sp., Staphylococcus aureus, Staphylococcus epidermidis.

Introduction:

The eye has a variety of natural barrier defences against external stimuli. Lids, eyelashes and eyebrows protect against light, wind and particulate matter. The innate immune system is the first line of defence against invading organisms or microbial colonization and consists of anatomical barriers of the ocular surface (e.g. epithelium) (Lars Bräuer *et al.*, 2012).

The human eyes have a special anatomy that makes them vulnerable to infectious agents, including bacteria, as well as direct environmental exposure. The eyelids, conjunctiva, and cornea are the main parts of the eyes that are most frequently infected. Several ocular diseases, including conjunctivitis, keratitis, scleritis, choroiditis, iridocyclitis, retinal vasculitis, retinitis, and posterior

uveitis, have been linked to contamination of handles and lenses of eyeglasses. This is quite worrisome, because these bacteria are transferred to the eyewear through contact with infected hands and other methods (Willcox, 2007). Many frequently used devices, such as eyeglasses and contact lenses, are rarely sterilized or kept clean, which increases the likelihood of bacterial contamination and colonization (Francis *et al.*, 2016; Wu *et al.*,2015). Significant number of people wear eyeglasses for functional or purely cosmetic reasons (Kaschke *et al.*, 2013; Nwaugo *et al.*, 2008).

Numerous microorganisms have been linked to eye infections caused by microbial contamination and colonization of eyeglasses. A study reported the presence of bacterial contaminants such as *Staphylococcus sp., Streptococcus sp, Pseudomonas s., Aeromonas sp, Citrobacter sp, Haemophilus influenza, Chlamydia trachomatis* and *Neisseria gonorrhoeae* on surgeon's eyeglasses (Butt *et al.*, 2012). Microorganisms are transmitted from animated sources to inanimate environmental sources, which may become secondary reservoirs if they meet the needs of transmitted pathogens to survive and to multiply. In healthcare settings, however, contaminated surfaces, which may not always be optimal for microbial survival and multiplication, still may play a role in the chain of infection, since surfaces close to the patients' environment may be touched at high frequencies, allowing transmission from animated sources to others via contaminated inanimate surfaces (Kramer, *et al.*, 2014).

The lack of awareness of workers and students about personal hygiene, for example not washing their hands after rubbing their nose contributes to bacterial contamination of the glasses they use. In addition to hand washing habits, bacterial transmission can occur through handshakes with other people. The habit of cleaning glasses only with water or even just with a cloth is also a cause of bacterial contamination. The results also showed that the cleaning fluid specifically formulated to clean the lens cannot stop bacterial growth (Osaro-Matthew *et al.*, 2015).

Material and Methods:

Total 40 sample were collected from 11th, 12th, B.Sc. and M.Sc. students at Shri Shivaji Science and Arts College, Chikhli in July 2023 to August 2023. Samples were collected in the sterilized petriplate with differential media and they are labelled with place, date and time. These plates were transferred to research laboratory of the Shri Shivaji Science and Arts College, Chikhli. Out of total 40 samples, 14 samples are collected from 11th Students, 12 samples are collected from 12th Students, 08 samples are collected from B.Sc. Students and 06 Samples collected from M.Sc. Students were taken by using sterile cotton swabs that had been moistened with sterile distilled water. Out of that 24 are female and 16 are male. After that it was inoculated on the Blood agar and MacConkey agar by streaking and incubated for 24 hours at 37°C. After incubation for 24 hours at 37°C, for confirmed the growing microorganism by using different tests such as Gram Staining, Hanging-Drop method, IMViC test, Carbohydrate fermentation, Enzyme production (Catalase Test and Oxidase Test), and Triple Sugar Iron Agar test, etc.

Result and Discussion:

The main objective of the present study was to isolate and identify the bacteria present on spectacles to create public awareness about the health hazards resulting from these microorganisms. All the samples were first cultured on nutrient broth and nutrient agar, which ensured the presence of certain bacteria. For further confirmation and identification, the culture from the nutrient agar was streaked on

different selective media and the presence of pathogenic bacteria like *Staphylococcus epidermidis, Staphylococcus aureus, Bacillus sp.* and *Pseudomonas* sp. was confirmed.

Table 1:- Shows the Biochemical test results of swab sample.

Sr. No.	**1**	**2**	**3**	**4**
Bacteria	*Pseudomonas sp.*	*Staphylococcus epidermidis*	*Bacillus sp.*	*Staphylococcus aureus*
Shape	Coccobacillus,	Cocci	Rod shaped	Cocci
Gram Staining	– ve	+ve	+ve	+ve
Indole	-	N/A	-	-
Methyl Red	-	-	-	+
Voges Proskauer	-	+	+	+
Citrate	+	-	+	+
Motility	+	-	+	-
Catalase	+	+	+	+
Glucose	-	+	+	+
Lactose	-	+	-	+
Mannitol	+	-	+	+
Novobiosin	N/A	Sensitive	N/A	N/A
Urea	-	+	-	+
Oxidase	+	-	-	-
H2S	-	+	N/A	-
TSI	+	N/A	N/A	N/A

Key :- N/A : Not applicable

On the basis above observation table we conclude that the different bacteria identified by using the Gram staining and different biochemical tests. *Pseudomonas sp.* was identified with the help of Gram staining it shows negative results and having Coccobacillus shaped bacteria. For confirmation of the bacteria some biochemical tests are also done and showing results such as TSI test showed alkali/alkali without H2S and gas. Also Indole, MR, VP, glucose, Lactose, urea, H2S showing negative results while the bacterial motility, Oxidase, Catalase, Citrate tests were positive.

When we observed the above results, it shows the bacteria were found in highest number is Pseudomonas sp which were found in 17 samples (42.50 % out of total sample collected). Pseudomonas sp. bacteria can grow in different a variable environmental condition that's why it grows on spectacles. Pseudomonas sp. can be transmitted through direct contact with water, and these bacteria can even be found in disinfectant fluids in hospitals (Murray *et al.*, 2016). If Pseudomonas sp. Grow on the spectacles it can cause disease. As a contaminant on glasses, Pseudomonas sp. can cause various

diseases of the eye, such as corneal ulcers (Kim *et al.,* 2013). One of the species of Pseudomonas, Pseudomonas aeruginosa, is the most common bacteria found to be the cause of keratitis in contact lens wearers (Watson *et al.,* 2018).

Staphylococcus epidermidis was identified with the help of Gram staining; it shows positive results and having Cocci shaped bacteria. Its showing positive reaction in VP, Catalase, Glucose, Lactose, Urea and H2S while negative reaction in MR, Citrate, Mannitol, Oxidase tests. For further identification to check the novobiosin antibiotic test and its showing sensitive results (If diameter ≥ 16 mm= Sensitive).

Staphylococcus epidermidis found in 5 samples which is near about 12.50% out of total samples collected. Staphylococcus epidermidis are observed in least number on spectacles. Research conducted in 2015 found that Staphylococcus epidermidis is the most common bacterial contaminant in glasses (Fritz *et al.,* 2018). Staphylococcus epidermidis is a common flora of the skin and is one of the pathogenic bacteria that is often found in nosocomial infections (Willey *et al*., 2014). staphylococcus epidermidis can also cause conjunctivitis, blepharitis, corneal ulcer, and endophthalmitis (Flores-Páez *et al*., 2015).

Bacillus sp. was identified with gram staining, it shows positive results with the spores and rod shaped bacteria. It's showing positive reaction in VP, Citrate, Motility, Catalase, Glucose, and Mannitol while negative reaction in Indole, MR, Lactose, Urease and Oxidase tests.

When we observed the above results, it shows the bacteria were found in highest number is *Bacillus* species which were found in 22 samples (55% out of total sample collected). *Bacillus* species are bacteria that are spread in nature with high concentrations in soil, water, and plants-derived food (Schultz *et al.,* 2017). Bacillus sp. appear on the surface of the glasses because these bacteria are that can form spores (Osaro-Matthew *et al.,* 2015).

This causes them to survive in all types of environments and can colonize various types of surface objects. *Bacillus sp.,* except *Bacillus anthracis,* is considered to have a less significant role in the context of human infection. Endophthalmitis caused by *Bacillus cereus* can cause vision loss in just a few days (Callegan *et al*., 2017).

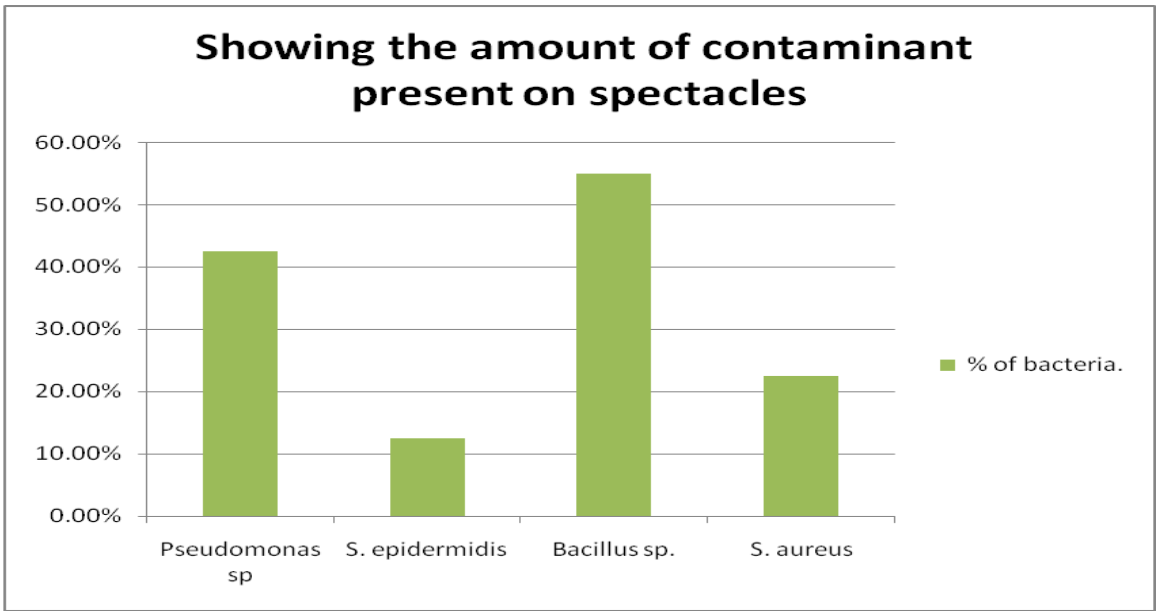
Staphylococcus aureus was identified with gram staining; it shows positive results with the Cocci shaped bacteria. The reaction on MR, VP, Citrate, Catalase, Glucose, Lactose, Mannitol and Urease tests are positive while Indole, Oxidase and H2S are showing negative results.

Total 9 sample shows the positive results of *Staphylococcus aureus,* which is near about 22.50 %. *Staphylococcus aureus* bacteria are grow on different environmental conditions. Staphylococcus aureus is the bacteria that can grow well on human skin (Willey *et al*., 2014). In addition, transmission of *Staphylococcus aureus* can also occur if someone does not wash his hands and shakes hands with others (Osaro-Matthew *et al*., 2015). *Staphylococcus aureus* infection leads to cause various types of diseases to the human, that's why we must clean our spectacles.

Table No. 2:- Showing the amount of contaminant spectacles in percentage.

Name of bacteria	% of bacteria occurred out of total sample.
Pseudomonas sp.	42.50 %
Staphylococcus epidermidis	12.50%
Bacillus sp.	55.00%
Staphylococcus aureus	22.50 %

Graph No. 1:- Showing the amount of contaminant present on spectacles.



Conclusion:

We observed the results and conclude that the microorganism mainly bacteria are present on spectacles which is used by the students of Shri Shivaji Science and Arts college Chikhli in 2023. Total four bacteria are identified namely *Pseudomonas sp.*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, and *Bacillus sp*. These bacteria are pathogenic for the eyes and spectacles having favourable environment for the bacterial growth leads to the continuous infection of that particular bacteria. We strongly recommend to the students wash your spectacles regularly with particular disinfectants.

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