Experiment No: 2

STERILIZATION OF GLASSWARE, PREPARATION, AND STERILIZATION OF MEDIA

Aim: To perform the sterilization of glassware, preparation, and sterilization of media

Sterilizing methods for glassware in a laboratory hot air oven: Sterilizing glassware such as bottles, Petri dishes, and test tubes requires dry heat in a hot air oven. The ideal temperature of the oven needs to reach at least 160'c and the content needs to be regulated at this 45 to 50 min. The content must not be removed from the oven immediately as a slow cooling period is necessary when the temperature has reduced down 50'c.

AUTOCLAVING LAB GLASSWARE

Autoclaves are used to sterilize instruments, glassware, plasticware, solutions, and media, and decontaminate biological wastes. Because of the physical hazards associated with autoclaving, extra care must be taken to ensure their safe use.

The following safety practices are followed when autoclaving laboratory glassware:

- Never autoclave items containing corrosives or radioactive materials
- Use only borosilicates, glassware's, which can be better withstand the stresses of high autoclave temperatures and pressures.
- Load the autoclave properly as per the manufacturer's recommendations.
- Individual glassware vessels should be placed within a heat-resistant plastic or metal tray on a shelf or rack and never placed directly on the autoclave bottom or floor.
- Add ¼ to ½ inch of water to the tray so the glassware will heat more evenly
- Check any plastic caps, tubing, or other items to ensure they can be safely autoclaved with the glassware
- Fill glassware only half full with liquids to be sterilized. Take into account the volume of liquid to be autoclaved.

PREPARATION AND STERILIZATION OF CULTURE MEDIA

1. The constituents or ingredients of that particular medium are weighed accurately

2. The required amount of water is measured and poured into stainless steel pan or heat resistant beaker and the weighed ingredients are putted one by one into it.

3. The pan is kept on heat source and each ingredients is gradually dissolve by contsnt stirring with glass rod and heat is reduced to prevent boiling over or burning of the medium.

4. Small amount of preparation is taken out, cooled, pH is checked, and if necessary, pH is adjusted.

5. The medium is now poured into test tubes, usually in 10 ml aliquots in 500 ml Erlemeyer flasks or other containers. While in a liquefied state, solid media can be added in test tubes, which are allowed to cool and harden in a diagonal position, producing agar slants.

6. The container is now closed with cotton plugs, screw caps, or caps all covers.

7. The medium is now sterilized in an autoclave, usually at 121'C for 15 min, and allowed to cool for use.