INTERNSHIP REPORT

K.K WAGH EDUCTION SOCIETY'S

K.K WAGH ARTS ,COMMERCE, SCIENCE AND

COMPUTER SCIENCE COLLEGE,

SARASWATI NAGAR,NASHIK.

- **NAME : ARBAZ ISMAIL KHAN**
- **ROLL.NO: 09**
- **FACULTY: BSC.BIOTECHNOLOGY**

COMPANY DETAILS

COMPANY NAME: JAI BIOTECH

ADRESSS: D29 ,NICE Area, MIDC,satpur colony,NASHIK,Maharashtra

Guide name : Nilima MAM,

Introduction of the company:

Jai Biotech Industries are one of the leading processors, marketers and suppliers of a wide range of products from Bio-instrumentation, Agricultural & Environmental Sectors. In addition to these, we also offer our clients services for R & D related to Biotechnology, Establishment of Laboratory, Bio-control & Biofertilizer plants, Hi-tech Agriculture services and Training, Seminars. Our business endeavors are well supported by our Partners Ambika plast & Chem Pvt. Ltd. & Sequence Biotech (P) Ltd., which are leading companies in the biotech field. We understand the fact that for development of Biotechnology, Instruments plays major role. Therefore, our biotech instruments like Plant Growth Promoter, Organic Plant Growth Promoter, Natural Plant Growth Promoter, Laboratory Fermenters, Bottle Fermenters, Information & Magnifying systems & Electrophoresis Systems are manufactured under the leadership of Mr. C. Santaram who is a senior Director in organization and has more than 35 years of experience in this field. Owing to our wide distribution network, we have been able to reach out to our clients in an effective manner. Moreover, the customization facility and flexible payment modes offered by us ensure that our clients remain completely satisfied.

Basic Information

Nature of Business	Manufacturer
Additional Business	Trader
Company CEO	Rajendra Patil
Total Number of Employees	11 to 25 People
Legal Status of Firm	Individual - Proprietor
Annual Turnover	Rs. 2 - 5 Crore

INDEX

PRODUCTS

#Bio-fertilizers:-

o AzoBact - Azatobacter

o Azospire – Azospirallum

o Rhizobact – Rhizobium

o Acetobact – Acetobacter

o Zincbact – Zinc Solubilizing Bacteria (ZSB)

o Bactophose – Phospate Solubilizing Bacteria(PSB)

o JK-Bact – Potassium Mobilizing Biofertilizers (KMB)

o Jaimix – Carried Based Consortia

o Jaibiomix(L) Liquid Consortia

Biopesticides:-

o JaiMold – Trichoderma viride

o Beavera – Beauveria bassiana

o Vertimust – Verticillium leccani

o Metaz - Metarhizium anisopliae

Biocontrol Agents:-

o Pheromone trapes

o Lures

Justification that why choose this company:

We choose Jai Biotech industry because it is one of the best biotech industry in Nashik. Gives us plenty time to learn about biotech lab.

Introduction to staffs:

When we enter Jai Biotech Industry their was Nilima Patil Ma’am who is QA & QC Manager of the company. She gives us information and introduction of company. After that she introduced us to Kalyani ma’am who is QC executive of Jai Biotech, she show their labs and how they works.

1.LAB INTRODUCTION /INSTRUMENTS IN LAB

A. PH METER:

Electric device used to measure hydrogen ion activity

(acidity/ alkanity)in solution.

Standerization /ph meter calibration on daily basis is compulsory.

How **to calibrate a pH meter**

1. Examine the pH electrode

First, check the pH electrode for contamination or damage. If it's damaged, then fix it or toss it. If it's just dirty, then use the cleaning solution according to its directions. Whether we use acids, washing liquid, or alkali, choose a solution appropriate for your process and the contamination.

2. Flush the pH sensor

Next, flush your sensor with distilled water. We can do this, even if we didn't have to clean it, to rinse away anything that may contaminate the buffer solution you'll use in Step 3. After flushing, dab or pat away excess water with the paper towels. We should not generally rub it more: you might charge or damage the sensor.

3. Immerse the pH electrode

Fill a beaker with your first buffer solution, then immerse your electrode in it. It's easy to just drop the sensor directly into the buffer bottle, but we can avoid contamination and extend the life of a buffer if we use the beaker.

4. Calibrate the pH meter

Now we can start your calibration/adjustment. Keep an eye on the stability of the value; an old pH meter might react sluggishly. When the value stabilizes, set the device to accept this calibration/adjustment point.

5. Rinse the pH sensor and repeat

Flush the pH sensor with distilled water again, then immerse it in another clean beaker with the second buffer solution.

MAGNETIC STIRRER:

A **magnetic stirrer** or **magnetic mixer** is a laboratory device that employs a rotating magnetic field to cause a stir bar (or *flea*) immersed in a liquid to spin very quickly, thus stirring it. The rotating field may be created either by a rotating magnet or a set of stationary electromagnets, placed beneath the vessel with the liquid. It is used in chemistry and biology where other forms of stirring, such as motorized stirrers and stirring rods, may not be viable for use.

Calibration of temperature:

6.3.1 Start the calibration procedure by using calibrated thermometer ranging up to 300°C.
6.3.2 Place the beaker or flask containing sample in the mid most of hot plate stirrer.
6.3.3 Operate only hot plate without using magnetic stirrer.
6.3.4 Set temperature of the hot plate by temperature regulator.
6.3.5 Dip calibrated thermometer at one side of the beaker.
6.3.6 Record both control panel temperature display and calibrated thermometer temperature.
6.3.7 Record the result in annexure-1.
6.3.8 Similarly, repeat the procedure at different temperature. And record the temperature.
6.4 Record the result on the **"Calibration Record",**

C. AUTOCLAVE :

An **autoclave** is a machine used to carry out industrial and scientific processes requiring elevated temperature and pressure in relation to ambient pressure/temperature. Autoclaves are used in medical applications to perform sterilization and in the chemical industry to cure coatings and vulcanize rubber and for hydrothermal synthesis. Industrial autoclaves are used in industrial applications, especially in the manufacturing of composites.

Many autoclaves are used to sterilize equipment and supplies by subjecting them to pressurized saturated steam at 121 °C (250 °F) for around 15–20 minutes depending on the size of the load and the contents.[1] The autoclave was invented by Charles Chamberland in 1879,[2] although a precursor known as the steam digester was created by Denis Papin in 1679.[3] The name comes from Greek *auto-*, ultimately meaning self, and Latin *clavis* meaning key, thus a self-locking device.[4]

Calibration of Autoclave by Biological Indicator:

- Put five indicators of Bacillus steriothermophilus spores at five different locations in the chamber.
- Run Autoclave at 121°C & 15 lbs pressure for 15 minutes.
- After completion of sterilisation cycle remove biological indicators from autoclave.
- Inoculate the biological indicator in sterile Soyabean casein digest medium tubes.
- Inoculate an unsterilized indicator in sterile Soyabean casein digest tube as positive control and incubate one sterile Soyabean casein digest medium tube without any inoculation as negative control.
- Incubate all the tubes at 55° to 60° for 72 hours.
- If purple colour changes to yellow after 15 to 18 hours: Sterilisation is faulty.
- If purple colour changes to reddish brown after 72 hours: Material is overheated.
- Incubate unopened Ampoules at 56° C for o72 hours if purple colour is retained after 72 hours of incubation. Sterilisation is perfect.

Calibration of Autoclave by using Heat labile Physical Indicator:

- lace five strips of steam clox at five different places in the chamber.
- Run autoclave at 121°C & 15 lbs pressure for 15 minutes.
- After completion of sterilization cycle remove the indicators from the autoclave.
- Observe the colour changes in indicator.
- The colour should change from pink to green.

D. HOT AIR OVEN :

Hot air ovens are electrical devices which use dry heat to sterilize. They were originally developed by Pasteur.[1] Generally, they use a thermostat to control the temperature. Their double walled insulation keeps the heat in and conserves energy, the inner layer being a poor conductor and outer layer being metallic. There is also an air filled space in between to aid insulation. An air circulating fan helps in uniform distribution of the heat. These are fitted with the adjustable wire mesh plated trays or aluminium trays and may have an on/off rocker switch, as well as indicators and controls for temperature and holding time. The capacities of these ovens vary. Power supply needs vary from country to country, depending on the voltage and frequency (hertz) used. Temperature sensitive tapes or biological indicators using bacterial spores can be used as controls, to test for the efficacy of the device during use.

Operate the Hot air oven according to the SOP.

1. For the calibration of hot air oven use a standard thermometer ranging up to 300ºC.
2. Start the calibration procedure after 1 hour of starting the oven.
3. Set the oven at desired temperature.
4. Put the standard thermometer for 30 min in upper shelf of hot air oven and close the door of it.
5. After 30 min open the door of hot air oven and read the temperature of standard thermometer randomly and match the observed temperature of thermometer.
6. Repeat the above procedure by putting the thermometer in lower shelf for 30 min.
7. Record the observed temperature in calibration record of hot air oven as per annexure-1.
8. The observed temperature of thermometer in both shelves should be ±2.0ºC tolerance limit to the set temperature value.
9. Record the result on the **“Calibration Record**

E. LAMINAR AIR FLOW :

In fluid dynamics, **laminar flow** is characterized by fluid particles following smooth paths in layers, with each layer moving smoothly past the adjacent layers with little or no mixing.[1] At low velocities, the fluid tends to flow without lateral mixing, and adjacent layers slide past one another like playing cards. There are no cross-currents perpendicular to the direction of flow, nor eddies or swirls of fluids.[2] In laminar flow, the motion of the particles of the fluid is very orderly with particles close to a solid surface moving in straight lines parallel to that surface.[3] Laminar flow is a flow regime characterized by high momentum diffusion and low momentum convection.

When a fluid is flowing through a closed channel such as a pipe or between two flat plates, either of two types of flow may occur depending on the velocity and viscosity of the fluid: laminar flow or turbulent flow. Laminar flow occurs at lower velocities, below a threshold at which the flow becomes turbulent. The velocity is determined by a dimensionless parameter characterizing the flow called the Reynolds number, which also depends on the viscosity and density of the fluid and dimensions of the channel. Turbulent flow is a less orderly flow regime that is characterized by eddies or small packets of fluid particles, which result in lateral mixing.[2] In non-scientific terms, laminar flow is *smooth*, while turbulent flow is *rough*.

CALIBRATION PROCEDURE:

The performance of the HEPA filters could be checked by the following two methods.
(i)DOP (Di Octyl Pthlate) test: (Done on contract basis)
(ii) Anemometer Test: This is one of the suitable tests for validation of the HEPA Filter. During the time of validation, place the anemometric sensor on the Laminar Air Flow at different location in front of the HEPA Filter.
Efficient -- 90 to 110 feet/minute
(iii) Plate method: Prepare nutrient agar plates (or) soyabean casein digests agar plates and exposes it in different corners of the instrument base. Then incubate these plates at 350C for 48 hours. No growth is observed, the HEPA Filter is working in good condition.

C. GENERAL CARE & PRECAUTIONS

Proper handling of the instrument.
Clean the instrument with 70% Iso propyl alcohol, before and after use.
A routine cleaning of filters should be done by blowing air.

F.DISTILATION UNIT :

Distillation unit

A method of separating mixtures based on differences in volatilities of components in a boiling liquid mixture is called distillation unit.It's a process that works a little like a still. Instead of removing contaminants from your water, a distiller reverses the equation and removes the water from your contaminants.

lfur / **Distillation** / Vapor Pressure / Flash Point / Octane Index / Chromatograph **analyzer calibration** main difficulty is due to the coexistence between gaseous and liquid phases in equilibrium in the container when the boiling point of different fractions is different .
The quantity of worldwide implementations in the oil refinery and petrochemical industries established the superiority of **piston cylinders** over the nitrogen sky tank , the siphon cylinders and the membrane accumulators for Sulfur / **Distillation** / Vapor Pressure / Flash Point / Octane Index / Chromatograph **analyzer calibration.**

G.INCUBATOR :

An **incubator** is a device used to grow and maintain microbiological cultures or cell cultures. The incubator maintains optimal temperature, humidity and other conditions such as the CO_2 and oxygen content of the atmosphere inside. Incubators are essential for much experimental work in cell biology, microbiology and molecular biology and are used to culture both bacterial and eukaryotic cells.

The simplest incubators are insulated boxes with an adjustable heater, typically going up to 60 to 65 °C (140 to 150 °F), though some can go slightly higher (generally to no more than 100 °C). The most commonly used temperature both for bacteria such as the frequently used E. coli as well as for mammalian cells is approximately 37 °C (99 °F), as these organisms grow well under such conditions. For other organisms used in biological experiments, such as the budding yeast Saccharomyces cerevisiae, a growth temperature of 30 °C (86 °F) is optimal.

The simplest incubators are insulated boxes with an adjustable heater, typically going up to 60 to 65 °C (140 to 150 °F), though some can go slightly higher (generally to no more than 100 °C). The most commonly used temperature both for bacteria such as the frequently used E. coli as well as for mammalian cells is approximately 37 °C (99 °F), as these organisms grow well under such conditions. For other organisms used in biological experiments, such as the budding yeast Saccharomyces cerevisiae, a growth temperature of 30 °C (86 °F) is optimal.

More elaborate incubators can also include the ability to lower the temperature (via refrigeration), or the ability to control humidity or CO_2 levels. This is important in the cultivation of mammalian cells, where the relative humidity is typically >80% to prevent evaporation and a slightly acidic pH is achieved by maintaining a CO_2 level of 5%.

H. B.O.D INCUBATOR :

The front panel has two circular doors which allow the insertion of the hands of the operator into the chamber to conduct inoculation, transfer of culture, etc. The cabinet on top is provided with an UV lamp as well as an ordinary tube light. The UV lamp is used for sterilizing the internal environment while the tube light provides illumination if and when required.

Procedure for use :

- (i) The interior of inoculation chamber is cleaned using a suitable disinfectant such as 70% alcohol.
- (ii) The materials required for experimentation inside the chamber are kept through the doors present on the opposite sides of the chamber.
- (iii) The UV light is switched on for about 15 minutes to sterilize the interior of the chamber.
- (iv) Transfer of materials to conduct inoculation, etc. can be carried out after this period.
- (v) It is preferable to keep a spirit lamp which can be used for flaming the inoculation needles or loops before and after inoculation.

I .Colony Counter

Colony counters are used to estimate a liquid culture's density of microorganisms by counting individual colonies on an agar plate, slide, mini gel, or Petri dish. The counting can be accomplished manually, often with touch pressure and a digital counter, or can be semi- or fully automatic.

Spectrophotometer

Spectrophotometry is a tool that hinges on the quantitative analysis of molecules depending on how much light is absorbed by colored compounds. ... A spectrophotometer is commonly used for the measurement of transmittance or reflectance of solutions, transparent or opaque solids, such as polished glass, or gases.

FUMIGATION SECTION :

Fumigation chamber is tomaintain environmental condition monitoring high concentation of toxic chemicals has been proposed to reduce microbial agents on hospital surfaces and to controlinfections fumigation with formal dehyde vapor is recognized and most commonly used method because it is a cost effective procedure potassium permagnet and formaldehyde Are burned in china dishes lethal gas to exterminate pests within an enclosed space

1.Media and broths

1. Nutrient Agar:

- Nutrient agar is a general purpose medium supporting growth of a
- wide range of non-fastidious organisms. It typically contains,
- . 0.5% peptone - this provides organic nitrogen
- 0.3% beef extract/yeast extract - the water-soluble content of
- these contribute vitamins, carbohydrates, nitrogen, and salts
- 1.5% agar - this gives the mixture solidity
- 0.5% sodium chloride - this gives the mixture proportions similar
- to those found in the cytoplasm of most organisms
- distilled water - water serves as a transport medium for the agar's
- various substances
- pH adjusted to neutral (6.8) at 25 °C (77 °F).
- These ingredients are combined and boiled for approximately one
- minute to ensure they are mixed and then sterilized by autoclaving,
- typically at 121 °C (250 °F) for 15 minutes. Then they are cooled to
- around 50 °C (122 °F) and poured into Petri dishes which are covered immediately. Once the dishes hold solidified agar, they stored upside down and are often refrigerated untilused. Inoculation takes place on warm dishes rather than coolones: if refrigerated for storage, the dishes must be rewarmed to room temperature prior to inoculation.

2. King's B media:

King Agar B enhances the elaboration of fluorescein and inhibits the pyocyanin formation. Mixed peptone provide the essential nitrogenous nutrients, carbon, sulphur and trace elements. Glycerol serves as a C-source and Dipotassium hydrogen phosphate buffers the medium.

Distilled water	1 L
proteose peptone #3 (Difco)	20 g
K 2 HPO 4	1.5 g
MgSO 4 •7H 2 O	1.5 g
Glycerol	10 mL
Agar	15 g

3.Potato dextrose agar:

Potato Dextrose Agar (PDA) is used for the cultivation of fungi. Potato Dextrose Agar (PDA) is a general purpose medium for yeasts and molds that can be supplemented with acid or antiobiotics to inhibit bacterial growth. It is recommended for plate count methods for foods, dairy products and testing cosmetics. PDA can be used for growing clinically significant yeast and molds. The nutritionally rich base (potato infusion) encourages mold sporulation and pigment production in some dermatophytes.

Potato infusion	200 gm
Dextrose	20 gm
Agar	20 gm
Distilled water	1 liter

4.Nutrient broth:

Nutrient Broth is a liquid medium used for the cultivation of a wide variety of organisms from clinical specimens and other materials. This medium can be enriched with other ingredients such as blood, serum, sugars, etc., for special purpose.

Distilled water	1L
beef extract	1g
Yeast extract	2g
Peptone	5g
Sodium chloride (NaCl)	5g

Slide plate technique:

Culture:

7-10 day old fungal culture.

Media:
Sabouraud agar

Preparation of agar (pH-5.6)
Sabouraud agar supplemented with aureomycin
Peptone-10g/liter
Dextrose-40g/liter
Agar-15g/liter
Aseptically add aureomycin ,10µg per ml, to the sterile, molten and cooled medium

Equipments:

- Sterile Petri dish
- Filter paper (9cm diameter)
- U-shaped glass rod
- Microscope slides and coverslips (Sterile)
- Sabouraud's plate with mixed culture of fungi
- Sterile Sabouraud's agar plate
- Lactophenol cotton blue stain
- Glass capillary tube
- Scalpel
- Inoculating needle
- Sterile distilled water
- 95% ethanol
- Forceps

Procedure:

A) Slide Culture Preparation

Aseptically, with a pair of forceps, place a sheet of sterile filter paper in a Petri dish.

- Place a sterile U-shaped glass rod on the filter paper. (Rod can be sterilized by flaming, if held by forceps.)
- Pour enough sterile water (about 4 ml) on filter paper to completely moisten it.
- With forceps, place a sterile slide on the U-shaped rod.
- Gently flame a scalpel to sterilize, and cut a 5 mm square block of medium from the plate of Sabouraud's agar or Emmons' medium.
- Pick up the block of agar by inserting the scalpel and carefully transfer this block aseptically to the centre of the slide.
- Inoculate four sides of the agar square with spores or mycelial fragments of the fungus to be examined. Be sure to flame and cool the loop prior to picking up spores.
- Aseptically, place a sterile cover glass on the upper surface of the agar cube.
- Place the cover on the Petri dish and incubate at room temperature for 48 hours.
- After 48 hours, examine the slide under low power. If growth has occurred there will be growth of hyphae and production of spores. If growth is inadequate and spores are not evident, allow the mold to grow for another 24–48 hours before making the stained slides.

Streak plate technique

Principle:
The sample/inoculum is diluted by streaking it across the surface of the agar plate. While streaking in successive areas of the plate, the inoculum is diluted to the point where there is only one bacterial cell deposited every few millimeters on the surface of the agar plate. When these lone bacterial cells divide and give rise to thousands and thousands of new bacterial cells, an isolated colony is formed. Pure cultures can be obtained by picking well-isolated colonies and re-streaking these on fresh agar plates.

Materials required:

- A source of bacteria (stock culture, previously streaked agar plate or any other inoculum)
- Inoculation loop
- Wire loop
- Bunsen burner
- Ethanol (70%)
- Agar plate (nutrient agar or any other agar medium)

Procedure

1. Sterilize the inoculating loop in the bunsen burner by putting the loop into the flame until it is red hot. Allow it to cool.
2. Pick an isolated colony from the agar plate culture and spread it over the first quadrant (approximately 1/4 of the plate) using close parallel streaks or Insert your loop into the tube/culture bottle and remove some inoculum. You don't need a huge chunk.
3. Immediately streak the inoculating loop very gently over a quarter of the plate using a back and forth motion (see area 1 in the figure above).
4. Flame the loop again and allow it to cool. Going back to the edge of area 1 that you just streaked, extend the streaks into the second quarter of the plate (area 2).
5. Flame the loop again and allow it to cool. Going back to the area that you just streaked (area 2), extend the streaks into the third quarter of the plate (area 3).

6. Flame the loop again and allow it to cool. Going back to the area that you just streaked (area 3), extend the streaks into the center fourth of the plate (area 4).
7. Flame your loop once more.