In-Vitro Diagnosis

A Internship Report submitted

for the partial fulfilment of the Degree of Master of Science

By

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C E R T I F I C A T E

This is to certify that this training report entitled **"In-Vitro Diagnosis"** was successfully carried out by Miss **Vrunda Chauhan** towards the partial fulfilment of requirements for the degree of Master of Science in Biotechnology of Atmiya University, Rajkot. It is an authentic record of his/her own work, carried out by him/her under the guidance of Name of Supervisor for a period of three months during the academic year 2022–23. The content of this manuscript, in full or in parts, has not been submitted for the award of any other degree or certificate in this or any other university.

Signature

Name of the Head of the Department

Signature

Name of the Supervisor

D E C L A R A T I O N

I hereby declare that the work incorporated in the present dissertation report entitled **"In-Vitro Diagnosis"** is my own work and is original. This work (in part or in full) has not been submitted to any University for the award of any Degree or a Diploma.

Date signature

Vrunda Chauhan

ACKNOWLEDGEMENT

- I would like to express my gratitude to all the people behind the screen who helped me to transform an idea into a real application.

- I would like to express my heart-felt gratitude to my parents without whom I would not have been privileged to achieve and fulfill my dreams. I am grateful to our H.O.D. Dr. Nutan Vishwakarma and Dr. Ragini raghav who must ably run the institution and has the major hand in enabling me to do my project.

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ABSTRACT

1. SR Bioera is synonymous to competence and innovation in raw material and consumable manufacturing catering customer worldwide with finest quality product and simultaneously elevating the norms of paramount business values. In the area of Lateral flow immunoassay test strips manufacturing.

2. SR Bioera have Offering The Finest And Purified Biomarker For Unique Sensitivity And Specificity For Immuno Chromatographic Assay.

3. covers the basic lateral flow test knowledge and formulations as well as the details experiment steps to make the test kit as you specified.

4. The LFIA is a paper-based (bio) analytical technique for the on-site detection of target.

5. When a lateral flow immunoassay is run, the test sample is added to a sample application pad at the end of the strip.

6. The sample then migrates to the conjugate release pad, where a detection particle (typically gold or latex) that has been conjugated to a biological component of the assay is held.

7. Next the sample and the detection reagent migrate to the reaction membrane; a second biological component of the assay will have been immobilized here to function as a capture reagent. The capture reagent usually exists as a test line which spans the width of the membrane. a control reagent will be immobilized in a second line further along the membrane.

8. The analyte is either captured at the test line, or continues to migrate until reaching the absorbent wicking pad at the other end of the strip

9. The detection reagent binds at the control line to indicate that the assay has run successfully.

COMPANY INFORMATION

Company name: SR Bioera Pvt. Ltd.

Location: Private Limited. Corporate H.O.India E-30, Hindwa Dreams, Dhoran Pardi, Surat-394155, Gujarat, India.

Nature of Company: Diagnosis

SR BioeraPvt. Ltd. has been a worldwide leader partnering with rapid test manufactures since 2016, SR Bioera is the unparalleled leader and innovator helping multitude of companies achieve substantial growth and prosperity, SR Bioera's entire business is in accordance with international ISO 9001, ISO 13485 & CE standards.

The company implemented ISO standards, having fulfilled the requirements of these standards, the company is completely in accordance with national legal regulation and the European union's medical device directive.

SR Bioera is synonymous to competence and innovation in raw material and consumable manufacturing catering customer worldwide with finest quality product and simultaneously elevating the norms of paramount business values.

Departments

In the area of Lateral flow immunoassay test strips manufacturing.

1. HR
2. Quality control
3. Quality assurance
4. Production
5. R & D
6. Engineering

Machine/Equipment

1. Strip cutter machine
2. Sheet cutter machine
3. Cassette pressing machine
4. Reagent dispenser
5. Sealer machine

Reagent:

1. Gold nanoparticles
2. Gold conjugates
3. Conjugation kit: Gold chloride salt, PH solution, Biomarker dilution buffer, Blocking Buffer, Gold dilution buffer.
4. Essential solution for lateral flow immune assay: Band enhancer
   Blocking solution
   Flow enhancer

OEM services:

An OEM (Original Equipment Manufacturer) builds a customer's product that is fully designed by that customer and then contracted out to produce.

Sr bioera products is machine/equipments, raw materials, reagent, consumables, rapid test kit, uncut sheet, plastic ware.

OBJECTIVES

Handle practical activity with minimum error by following Good Laboratory Practice.
To gain knowledge about production process.

I learn the hcg kit work of diagnosis company. Learn handling of cassette assembly, sample pad dipping. Sample pad, nitrocellulose membrane, conjugate pad and absorbent pad overlapping.

HCG Invitro Diagnosis kit as a product, studied the all procedure from beginning of making of lateral flow diagnosis.

Gain knowledge about Qualty Control analyzes the raw material, the final disposition of the raw material is assigned by the Quality Control Manager.

PRODUCT DETAILS

SR Bioera have dedicated to provide affordable innovated products with precise quality and service. SR Bioera have focused to design and develop each product as a Unique Blend of Art, Science and Technology.SR Bioera do believe in deliver each of productdiscovery at economic platform to industry.SR Bioera have inspired by innovation and have the vision to support advancement in human healthcare.

PRODUCTS

1. IVD Raw Materials :- gold nanoparticles, gold chloride salt, antigen and antibodies

2. Consumables :- sample pad, conjugate pad, absorbent pad, PVC backing cad, Nitrocellulose membrane

3. Plastic Ware :- dropper, test cassettes, etc.

4. Rapid Test Kit :- infectious diseases, Drug of Abuse, animal tests etc.

INTRODUCTION

- A pregnancy strip test works by measuring the amount of human chorionic gonadotropin (HCG) hormone in human urine.
- The hormone is produced by cells in placenta, 6-12 days after conception.
- HCG is eliminated from the mother's body through urine, and therefore urine is used as a sample during pregnancy test
- Based on the principle of lateral flow immunoassay (LFA)

Purpose of the test

The main use of a pregnancy test is to evaluate if you are pregnant. Both urine and blood tests for hCG can be used for this purpose, and repeat testing may be done to confirm a pregnancy.

Check for pregnancy prior to surgery or other medical treatments.

Test results

Test results are reported in milli-international units per milliliter (mIU/mL).

he results of quantitative test results may also be considered if hCG testing is being done related to an abnormal pregnancy or other health conditions.

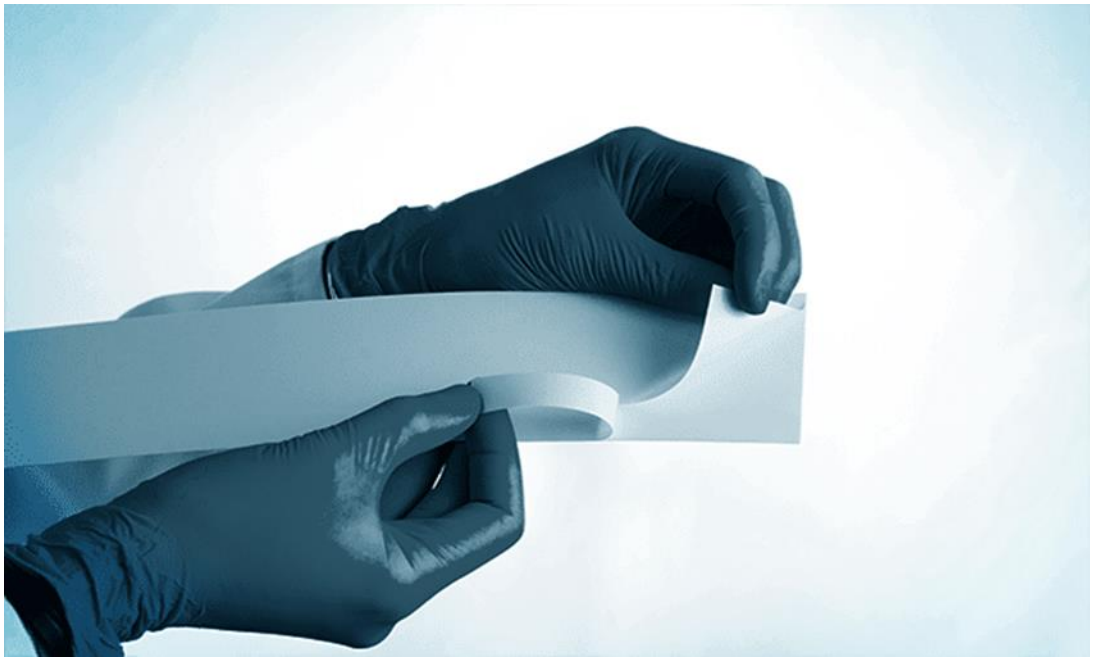
PRODUCTION PROCESS

- PVC card
- Nitrocellulose membrane
- Primary treatment of conjugate pad
- Gold dipping
- Sample pad
- Absorbent pad
- Card lamination

PVC BACKING CARD

- PVC Backing cards for lateral flow rapid diagnostic test kit is usually made of PVC materials, with main features of good flatness, strong adhesive.

- adhesive a non-reactive, non-volatile with a stable shelf-life of 2.5 years.



SAMPLE PAD

1. Sample pad made of binder free glass fiber that exhibits high absorption capacity.

2. the sample is applied at one end of the strip, on the adsorbent sample pad.

3. which is implied with buffer salts and surfactants that make the sample suitable for interaction with the detection system.

4. The sample pad ensures that the analyte present in the sample will be capable of binding to the capture reagents of conjugates and on the membrane.

5. The in vitro diagnostic sample pad is available in a wide range of thickness, absorption levels.

BUFFER DIPPING

- Sample pad are given a treatment because of the hydrophobic nature.

- Sample pad are dipped into a buffer

- Sample pad are dried into a hot air oven until it dry completely.

CONJUGATE PAD

- The conjugate pad is always of a synthetic material (at least when using a gold conjugate) to ensure the efficient release of its contents.

- The treated sample overlap through the conjugate pad.

- which contains antibodies that are specific to the target analyte and are conjugated to coloured or fluorescent particles-most commonly colloidal gold.

- The sample, together with the conjugated antibody bound to the target analyte, move along the strip into the detection zone.

BUFFER DIPPING (PRIMARY TREATMENT)

- Strips are given a primary treatment because of the hydrophobic nature.
- Strips are dipped into a buffer until stripes get wet.
- Now stripes are dried into a hot air oven until it dry completely.

GOLD DIPPING (SECONDARY TREATMENT)

- Primary treated strips are dipped into a pure gold solution.

  placed in the hot oven for 4 hrs.

NITROCELLULOSE MEMBRANE

- This is a porous membrane (usually composed of nitrocellulose) with specific biological components (mostly antibodies or antigens) immobilized in lines.

- Large pore size nitrocellulose membranes with pore size from about 2 to 20 microns.

- Their role is to react with the analyte bound to the conjugated antibody.

- The sample analyte results in an proper response on the test line,
  while a response on the control line indicates the proper liquid flow through the strip.

- Capture of colorimetric conjugate-antigen complexes by the immobilized antibody on each successive line, where the number of lines appearing on the strip is directly proportional to the concentration of the analyte.

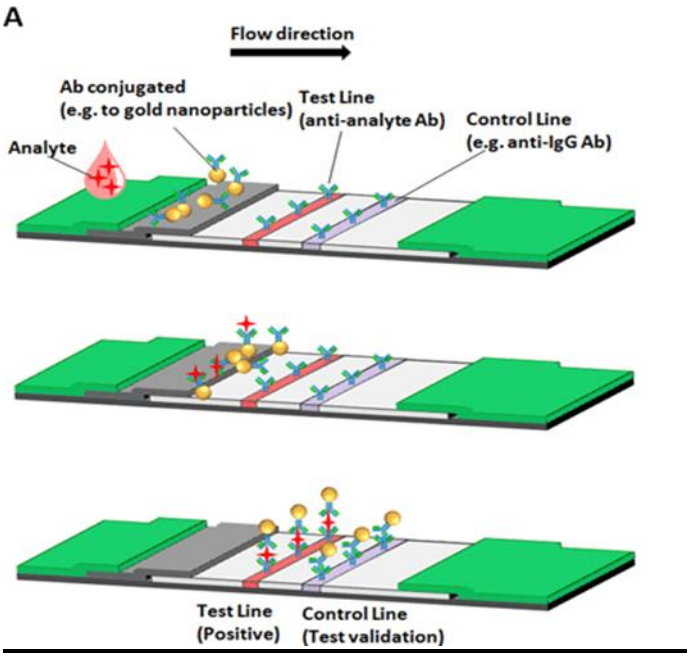
- In the direct test, the presence of the test line indicates a positive result and the control line usually contains species-specific anti-immunoglobulin antibodies, specific for the antibody in the particular conjugate.

ABSORBENT PAD

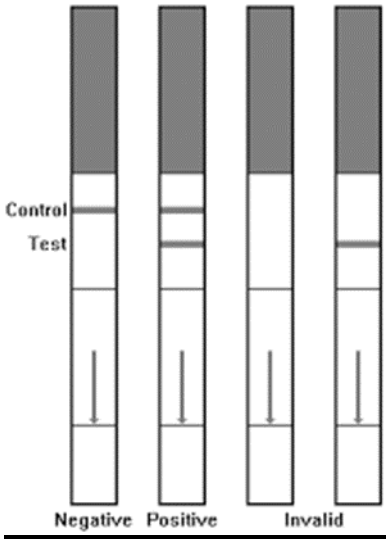
1) Absorbent pads are made from non-woven, cellulose fiber sheets.

2) The liquid flows across the device because of the capillary force of the strip material and, to maintain this movement, an absorbent pad is attached at the end of the strip.

3) The role of the absorbent pad is to wick the excess reagents and prevent backflow of the liquid.



TESTING



1. HCG KIT TESTING

- Results are given in milli-international units per milliliter (mUI/mL)anything above 25 mIU/mL is considered positive for pregnancy, This is generally around 12 to 14 days after conception.

- Negative Test: A coloured line appears in control line. No coloured line appears in test line.

- Positive Test: Two coloured lines appear, one in control area C and one in test area T. The intensity of the test line T may vary depending on the concentration of the antigen in the sample.

- Invalid Test: In the control area C, a red line must always appear when the test is performed. If this line does not appear, the test is invalid in any case. Please repeat the test with a new test cassette.

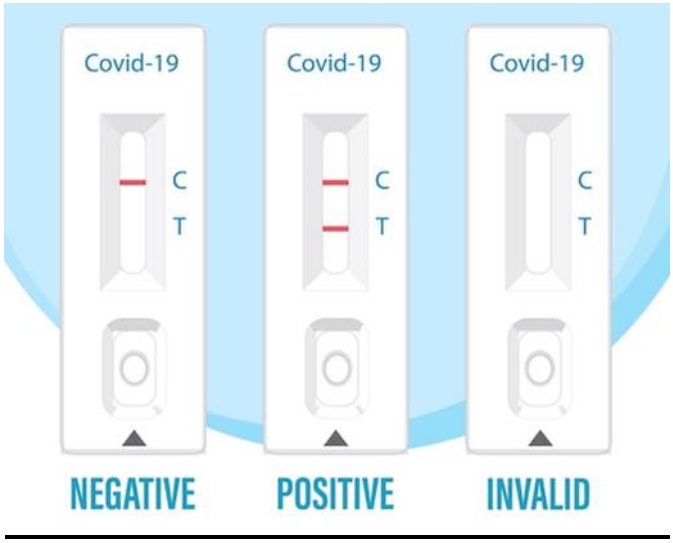
2. HIV KIT TESTING

- HIV self-tests provide results within 20 minutes.

- With a rapid antibody test, usually done with blood from a finger stick or with oral fluid, results are ready in 30 minutes or less.

- The rapid antigen/antibody test, done with blood from a finger stick, takes 30 minutes or less.

3. COVID KIT TESTING



Positive Test: Pink bands appear in the Control (top line) and IgG (bottom line). The test is positive for IgG antibodies to COVID19 virus.

Negative Test: Pink bands appear in the Control (top line) only. The negative test does not exclude COVID-19 infection.

Invalid Test: No pink band appear in the Control (top line). The test is invalid and should be repeated.

REFFERENCE

- https://pubmed.ncbi.nlm.nih.gov/26232440/
- https://www.futurelearn.com/info/courses/creating-the-amazing-engineering-the-future/0/steps/65276
- https://pubmed.ncbi.nlm.nih.gov/26355296/
- https://pubmed.ncbi.nlm.nih.gov/25660530/