In Vitro Diagnosis

A Industrial Training Report submitted

For the partial fulfilment of the Degree of Master of Science By

Riya Bera

210621039

[M.Sc. Biotechnology]

Under the supervision of

Anjali Ranghani

Head of QA & QC

DEPARTMENT OF BIOTECHNOLOGY ATMIYA UNIVERSITY

'YOGIDHAM GURUKUL' KALAWAD ROAD

RAJKOT (GUJARAT) – 360005

2022-23

ATMIYA UNIVERSITY
(Established under the Gujarat Private University Act 11, 2018)
Yogidham Gurukul, Kalawad Road, Rajkot - 360005, Gujarat (INDIA)

CERTIFICATE

This is to certify that this training report entitled "In Vitro Diagnosis" was successfully carried out by Miss Riya Bera 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 her under the guidance of Anjali Ranghani for a period of 3 months during the academic year of 2022-23. The content of this report, in full or in parts, has not been submitted for the award of any other degree or certificate in this or any other University.

Nutan Prakash Vishwakarma

Anjali Ranghani

DECLARATION

I hereby declare that the work incorporated in the present an industrial training 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: March 31, 2023 Riya Bera

INDEX

Acknowledgement

I profoundly thank Mr. Kashyap Bhatiya Manager of SR Bioera Pvt. Ltd and Ms.Anjali Ranghani Senior Biotechnologist of SR Bioera who has been an excellent guide and also a great source of inspiration to my work.

1) I also would like to thank my lab mates and my colleagues for their kind help and support.

2) The satisfaction that a company the successful completion of the task would be great but incomplete without the mention of the people who made it possible with their constant guidance and encouragement crowns all the efforts with success.

3) My special gratitude is to my brother, my sister and my family for their loving support. Lastly, and most importantly, I wish to thank my parents. They have always supported and encouraged me to do my best in all matters of life. To them I dedicate this report.

Abstract:

- Lateral flow assays (LFAs) are the technology behind low-cost, simple, rapid and portable detection devices popular in biomedicine, agriculture, food and environmental sciences.
- This review presents an overview of the principle of the method and the critical components of the assay, focusing on lateral flow immunoassays.
- This type of assay has recently attracted considerable interest because of its potential to provide instantaneous diagnosis directly to patients.
- The range and interpretation of results and parameters used for evaluation of the assay will also be discussed.

- The main advantages and disadvantages of LFAs will be summarized and relevant future improvements to testing devices and strategies will be proposed.
- Finally, the major recent advances and future diagnostic applications in the LFA field will be explored.
- A variety of biological samples can be tested using LFAs, including urine, saliva, sweat, serum, plasma, whole blood and other fluids. Further industries in which LFA-based tests are employed include veterinary medicine, quality control, product safety in food production, and environmental health and safety.
- In these areas of utilization, rapid tests are used to screen for animal diseases, pathogens, chemicals, toxins and water pollutants, among others.

About Company

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 offers only the best quality products at competitive prices and has one of the widest ranges of medical products in south-eastern Europe.

Our experience and flexibility towards the client make us a sought -after business partner, which has been recognized on the global market.

Product Detail

SR Bioera do believe in deliver each of product discovery at economic platform to industry.SR Bioera have inspired by innovation and have the vision to support advancement in human healthcare.

SR Bioera have dedicated to provide affordable innovated products with precise quality and services.

Product

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

Consumables: Sample pad, Conjugate pad, Absorbent pad, PVC card, nitrocellulose membrane Rapid Test Kit: Infectious diseases, Drug of Abuse, animal tests etc.

Objective

- Handle practical activity with minimum error by following Good Laboratory Practice.
- To gain knowledge about production process.
- To gain knowledge about HCG kit work of diagnosis company.
- Learn handling of cassette assembly, sample pad dipping. Sample pad, nitrocellulose membrane, conjugate pad and absorbent pad overlapping

Production Process

PVC card

Nitrocellulose membrane

Primary treatment of conjugate pad

Gold dipping

Sample pad

Absorbent pad

Card lamination

Antibody

- Although the physical components of the test strip, construction techniques and buffers play the major role in optimizing the test
- The heart of these processes are the antibodies, which need to be carefully designed and highly purified.
- It is very important to ensure a consistent antibody supply with proven affinity and specificity.
- Use of monoclonal antibodies is preferable, as it allows the production of specific antibodies in large quantities.

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

- Sample pad made of binder free glass fibre that exhibits high absorption capacity
  - The sample is applied at one end of the strip, on the adsorbent sample pad,
  - Sample pad is the platform where sample is placed at the time of testing.
- A sample pad can be used for urine, whole blood, plasma and serum specimen,

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

Conjugate Pad

The important function of the conjugate pad is to deliver the test agent particles as a constant volume of sample on each test strip to the membrane.

- 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 complex of gold conjugate and analyte then moves into and up the membrane.

[1] Buffer Dipping

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

[2] GOLD DIPPING

- Primary treated strips are dipped into a pure gold solution.
- Placed in the hot oven for 4 hrs.

Absorbent Pad

- These are specially treated absorbent pads made with compressed cellulose to absorb the reaction mixture of the rapid test assay and hold the reaction mixture for a longer duration of time.
- Absorbent pads, when used, are placed at the distal end of the test strip. The primary function of the absorbent pad is to increase the total volume of sample that enters the test strip.
- This increased volume can be used to wash unbound detector particles away from the test and control lines, thereby lowering the background and enhancing assay sensitivity.
- Since the volume of sample that ultimately contributes to signal is controlled by the volume required to solubilize the detector particles, and not by the total volume of sample that enters the device.

- If the strip design does not include an absorbent pad, the volume of sample analysed in the strip is determined solely by the bed volume of the membrane.
- There are two major considerations associated with the use of absorbent pads.
- First, a suitable material must be identified, specified, purchased, and integrated into the manufacturing process.
- Ultimately, this leads to a higher cost for the finished product. Second, an absorbent pad makes it difficult to incorporate an end-of-assay indicator in the test device.
- Most absorbent pads are made from cellulose filters. The material should be selected on the basis of thickness, compressibility, manufacturability, and, most of all, uniformity of bed volume.
- Once an absorbent material has been chosen, optimizing the overall volume absorbed by the test strip is best managed by changing the dimensions (usually the length) of the absorbent pad.
- The role of the absorbent pad is to wick the fluid through the membrane and to collect the processed liquid.
- The absorbent pad allows the use of larger sample volumes, which results in increased test sensitivity. The most popular absorbent pads are made of cellulose filters.

Nitrocellulose Membrane

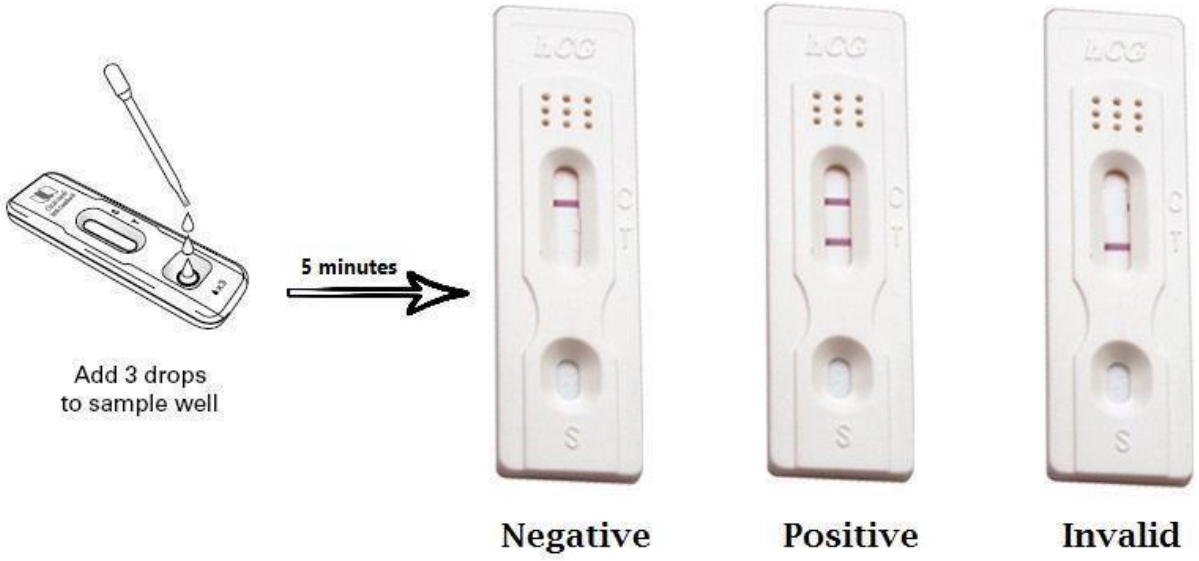
- The membrane is considered the most critical element in LFA strips and nitrocellulose is by far the most commonly used material
- Moreover, there are also pillar based capillary LFA devices used for deoxyribonucleic acid (DNA) hybridization detection (where micropillar arrays

replace the membrane), which have the advantage of more precise control of the capillary flow**.**

- Important parameters characterizing a good membrane material are the capillary forces, as well as the ease of binding and immobilizing proteins necessary for subsequent selection, reaction and detection.
- A range of nitrocellulose pore sizes are available, from 0.05 to 12 μm. However, as the pores are not equally distributed (because of the manufacturing process), capillary flow time is a more accurate parameter and it should be used when selecting the most effective strip material.
- The capillary flow time is the time required for the liquid to travel to and completely fill the strip of the membrane.
- Microporous nitrocellulose membranes are used in lateral-flow assays as the substrate upon which immune complexes are formed and visualized to indicate the presence or absence of an analyte in a liquid sample.
- The capillary flow time is the time required for the liquid to travel to and completely fill the strip of the membrane
- Microporous nitrocellulose membranes are used in lateral-flow assays as the substrate upon which immune complexes are formed and visualized to indicate the presence or absence of an analyte in a liquid sample.
- The pore sizes of membranes used in this application are comparatively large, ranging from 3 to 20 μm.
- Several attributes have resulted in nitrocellulose being the preferred substrate for lateral-flow assays.
- Nitrocellulose adsorbs protein at a high level.
- To facilitate the utilization of nitrocellulose in lateral-flow assays, the membrane can be cast directly into a polyester backing.
- The backing does not interfere with the function of the nitrocellulose.

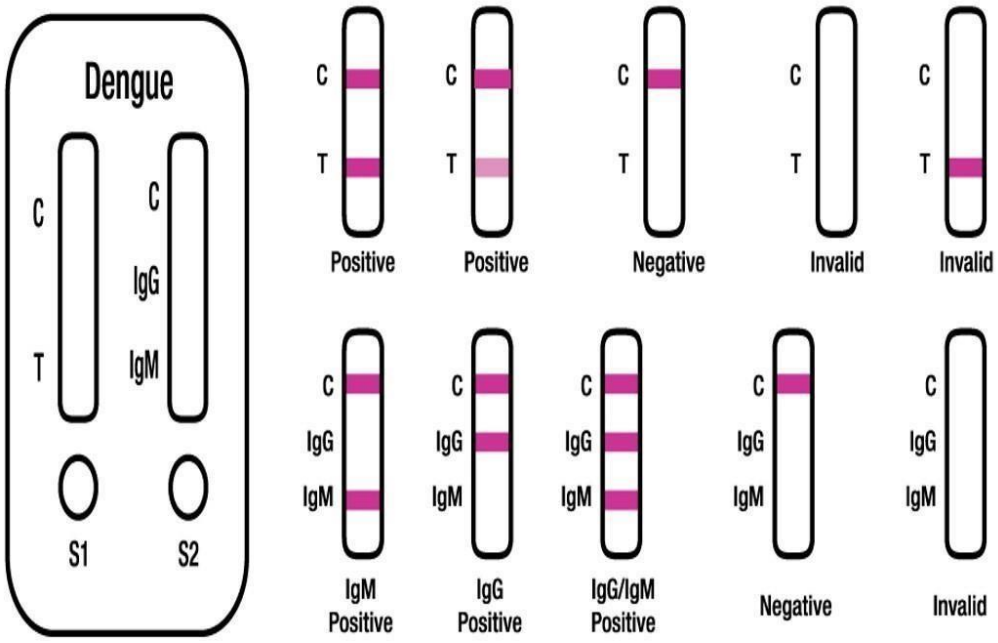
Results

HCG Result



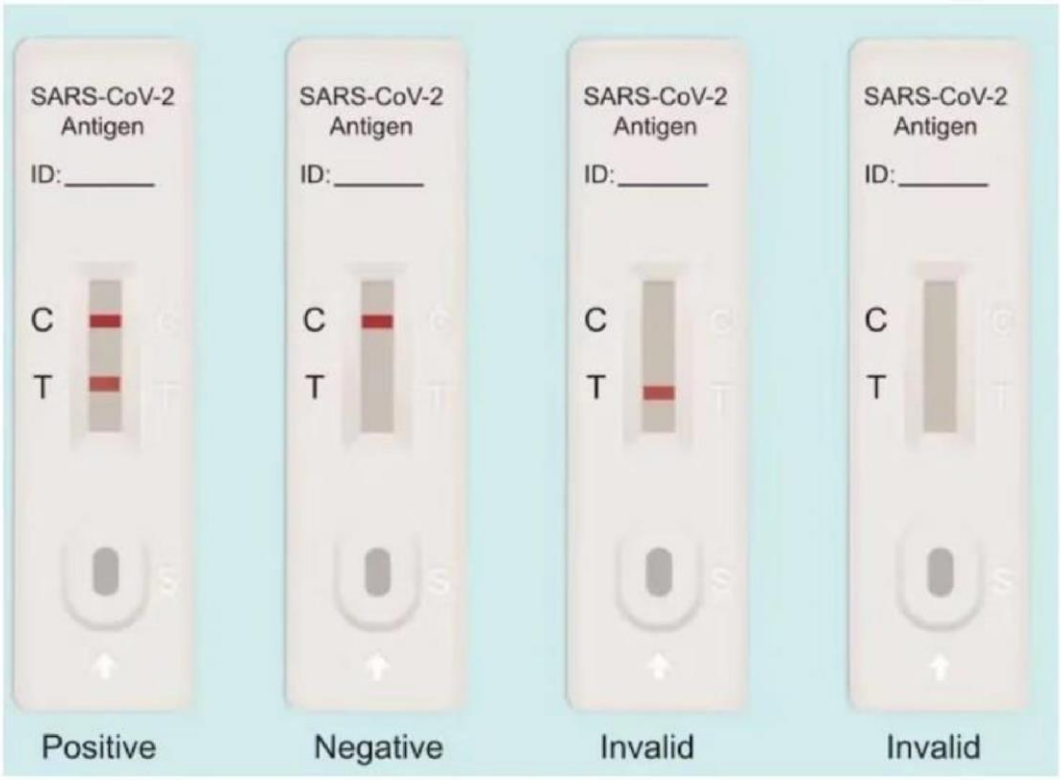
- People must read the urine test instructions and follow them carefully. Most tests use lines to show when a test is positive. The test line does not have to be as dark as the control line to be positive. Any line at all indicates the test is positive.
- An individual must check the test within the time frame the instruction indicates. This is typically around 2 minutes.
- Test stripes can change colour as they dry. Some people notice an evaporation line after several minutes. This is a very faint line that may look like a shadow.

Dengue Result



- A positive NS1 test indicates dengue virus infection but does not reveal the serotype.
- A negative NS1 test result does not rule out the absence of illness.
- People who have a negative NS1 test should be tested for dengue IgM antibodies to see if they have recently been exposed to the virus.

Covid Test Kit



- If SARS-CoV-2 RNA is detected then the test is positive.
- If SARS-CoV-2 RNA is not detected then the test is negative.