Among females frequency of Breast Cancer (BC) is very high. The onset of BC is well understood by CA15-3 which is a monomeric glycoprotein of with varying molecular mass of 250 - 350 kDa.

The goal of this study is the production and downstream processing of cell-derived CA15-3 a BC antigen for its potential applications in development for in vitro diagnostics. Raising monoclonal antibodies against the purified cell derived antigen and utilizing these antibodies for sandwich immunoassay development. To the best of our knowledge, the recent study shows that homogenous CA15-3 was first generated and purified through in-vitro cell culture methods at a large scale ensuring homogeneity profile in CA 15-3 antigen.

Whereas, traditionally CA15-3 is isolated and purified from biomedical fluids containing heterogeneous population of different cancer antigen.

The designed study describes a simple and economic purification approach for CA 15-3, which enables to explore various bio clinical as well as bio diagnostic applications and requirements. The purified CA15-3 was tested by SDS-PAGE, immunoblotting and chemiluminescence immunoassays (CLIA) and Fourier transform infrared (FTIR) spectrometry. This project also aimed to describe the development of an Immunoassay based on sandwich ELISA for estimation of CA15-3 in India, reducing the large amount of expenses incurred by the patients for performing diagnostic tests and with higher accuracy to save upon the precious time wasted during the diagnosis phase due to which treatment is delayed.

Thus we hope that this project will dramatically change the diagnostic scenario in reference of CA15-3 estimation in BC patients in Indian population and other developing countries with reference of cost effectiveness and enhanced availability.
During the course of project homogenous Cell derived CA 15-3 antigen was efficiently purified from harvest CA15-3 immunoassay using Mabs developed using cell derived CA15-3 antigen generated and purified from BC cell line T47-D. The murine immune system was challenged with purified CA 15-3 antigen and monoclonal antibodies synthesising clones were produced. The so produced monoclonal antibodies were tested for their activity in sandwich assay system and finally a Sandwich ELISA was developed.

To the best of our knowledge we generated and purified homogenous CA15-3 at large scale through *in vitro* cell culture method. Our immunoassay was validated against commercially available CA15-3 assay (Commercial assay utilizes antibodies raised against HMFG). It also had the benefits of simplified interpretation cost efficiency and reduced improvement time.

The cell derived CA15-3 could replace isolation and purification of CA15-3 from native source, as the purification of CA15-3 is a tedious process. Cell derived CA15-3 holds all conformational epitopes needed to develop MAb with equivalent and better affinity towards native antigen.

Therefore cell derived CA15-3 to an extent minimizes dependency on biomedical fluids and tissues. The immunoassay developed demonstrated good co-relation with commercially available CA15-3 immunoassay (CALBIOTECH CA240T).