Breast cancer is the most common cancer in women and second most common cancer overall, worldwide, with nearly 1.7 million new cases diagnosed in 2012. This makes about 12% of all new cancer cases and 25% of all cancers in women. Over the past two decades, our understanding of the mutations that drive breast cancer pathogenesis has grown enormously, which has rapidly led to the development of novel drugs. Among numerous approaches to treat breast cancer, large focus has been put on a particular type of pharmacological action that blocks activity of proteins called kinases. These drugs inhibit mistakenly activated signals that tell cells to divide. Such therapies however have been turned out to be less efficient than initially thought.

A more recent approach to stop growth and ultimately kill cancer cells is not to block wrongly activated signals but actions that the wrongly activated signals induce. Drugs that perform such actions are called transcription factors inhibitors. Transcription factors are proteins that activate gene expression in cells and therefore are necessary for a cell to perform a particular action e.g. growth, division or secretion of biochemical signals. Transcription factors can be activated by a number of signals, of which many may be pathogenic. Therefore by blocking a single transcription factor many sources of false signals can be inactivated. One class of transcription factors that is thought to be involved in onset and progression of breast cancers is the family of the so called STAT proteins. Numerous studies have described STATs as desirable targets for therapeutic interventions. In order to utilise STATs as drug targets, it is necessary to better understand their activation mechanisms in the specific types of breast cancer.

Typically, aberrations is cellular signalling and activity of transcription factors are examined by measurements of their activity in cells without external stimulation. It has been recently recognised that it is not the basal activity level that is most informative of cancer pathology but the manner in which such proteins perform upon activation. Thus, measuring dynamics of their activity in single cancer cells would be extremely insightful for recognising, distinguishing and understanding anomalies in different cancer cell types. Such method of measuring dynamical cell responses can be seen as interrogation of cells with specifically designed stimuli to reveal their pathogenic potential.

The main objective of our study is to provide experimental and theoretical analysis of activity of the most relevant of the STAT proteins STAT1, STAT3 and STAT5. For the experiments we will use 24 breast cancer cell lines that represent whole spectrum of breast cancer tumours. We will measure the dynamics of activity of these proteins after activation with a carefully designed set of biochemical stimulants that are thought to play a role in the genesis of breast cancer.

Cells will be stimulated with a low, medium and high dose of each stimulant. Activity of STAT1, STAT3 and STAT5 proteins will be quantified before stimulation, 30, 120 and 180 minutes after stimulation. For measurements, automated high content microscopy imaging will be used. Automated image analysis and quantification as well as pipetting robot will ensure high-throughput character of the project.

Single cell measurements will allow us to group cell lines according to signalling profiles and verify how such grouping relates to existing classifications of breast cancer cell lines. A detailed understanding of pathological mechanisms of transcription factor activation underlying cancerous phenotype would be extremely useful in developing new treatments. However, their complexity is such that their integrated functions are hardly to be determined without mathematical and computational approaches. In our project mathematical modelling will be used to explain differences in activity dynamics between cell lines. Literature knowledge as well as statistical methodology will be used to mathematicaly describe most likely origins of aberrations in activity of STAT proteins.

Building a mathematical model capable to explain activity dynamics of STAT proteins in a collection of cancer cell lines will provide a substantial progress in systematic understanding of the STAT proteins system. It will help in utilisation of their potential as effective drug targets Moreover, the classification of activity dynamics will be a valuable tool for selection of cell line models for breast cancer *in vitro* and *in vivo* research.