Elza Freiberga (Kuzņecova)

ORCID 0000-0002-6855-6414

Molecular Subtype-Specific Gene Expression Profiles in Breast Cancer

Summary of the Doctoral Thesis for obtaining the scientific degree "Doctor of Science (*PhD*)"

Sector Group – Medical and Health Sciences

Sector – Basic Medicine

Sub-Sector – Medicinal Genetics

Riga, 2026

The Doctoral Thesis was developed at Institute of Oncology and Molecular Genetics
Rīga Stradiņš University, Latvia

Supervisors of the Doctoral Thesis:

Dr. med., Professor **Zanda Daneberga**,
Institute of Oncology and Molecular Genetics, Rīga Stradiņš University, Latvia

Dr. biol., Associate Professor **Miki Nakazawa-Miklaševiča**,
Institute of Oncology and Molecular Genetics, Rīga Stradiņš University, Latvia

Scientific Advisor:

Dr. biol., Professor **Edvīns Miklaševičs**,
Rīga Stradiņš University, Latvia

Official Reviewers:

Dr. biol., Tenured Professor **Dace Pjanova**,
Institute of Microbiology and Virology, Rīga Stradiņš University, Latvia

PhD, Professor **Sonata Jarmalaite**,
Vilnius University, Lithuania

MD, Associate Professor **Jānis Eglītis**,
University of Latvia

Defence of the Doctoral Thesis will take place at the public session of the Promotion Council of Basic Medicine on 11 May 2026 at 14.00 in the Hippocrates Lecture Theatre, 16 Dzirciema Street, Rīga Stradiņš University.

The Doctoral Thesis is available in RSU Library and on RSU website:
https://www.rsu.lv/en/dissertations

Secretary of the Promotion Council:

Dr. med., Associate Professor **Anna Miskova**

Table of Contents

Abbreviations used in the Thesis

AR	Androgen receptor
BRCA1	BRCA1 DNA repair associated coding gene
BRCA2	BRCA2 DNA repair associated coding gene
CARTPT	CART Prepropeptide coding gene
CGA	Glycoprotein Hormones, Alpha Polypeptide
CHGB	Chromogranin B
CI	Confidence interval
COL9A1	Collagen Type IX Alpha 1 Chain coding gene
CSN3	Casein Kappa
DEG	Differentially expressed genes
DNA	Deoxyribonucleic acid
ECM	Extracellular matrix
ER	Oestrogen receptor
FABP4	Fatty Acid Binding Protein 4 coding gene
FOXA1	Forkhead Box A1 coding gene
GATA3	GATA binding protein 3 coding gene
GPX2	Glutathione Peroxidase 2 coding gene
HER2	Human epidermal growth factor receptor 2
HR	Homologous recombination
IRS4	Insulin receptor substrate 4 coding gene
MMP9	Matrix metalloproteinase 9
mRNA	Messenger RNA
ORM1	Orosomucoid 1
PARP	poly (ADP-ribose) polymerase
PPI	Protein–protein interaction
PR	Progesterone receptor
RNA	Ribonucleic acid
SLC39A6	Solute Carrier Family 39 Member 6 coding gene
STRING	Search Tool for the Retrieval of Interacting Genes/Proteins.
TFF1	Trefoil factor 1
TFF3	Trefoil factor 3
TNBC	Triple negative breast cancer
TPSD1	Tryptase Delta 1
TRH	Thyroid releasing hormone

Introduction

Breast cancer is the most diagnosed malignancy among women globally, accounting for nearly 1 in 4 cancer cases in women, and representing significant public health challenges. In 2022 there were 2.3 million new cases of female breast cancer, making it the second leading cancer globally (11.6 % of all new cancer cases), and approximately 666 000 deaths, ranking it the fourth leading cause of cancer mortality (6.9 % of all cancer deaths) (Arnold et al., 2022; Bray et al., 2018). Although incidence rates are rising, mortality from breast cancer has gradually decreased due to improvement in early detection and therapeutic management (Kesson et al., 2012).

Triple negative breast cancer (TNBC) molecular subtype accounts for approximately 15–20 % of all breast cancer cases, characterised by absence of oestrogen (ER), progesterone (PR) and human epidermal growth factor receptor 2 (HER2) expression and is of particular concern due to its aggressive clinical behaviour and poor prognosis (Garrido-Castro et al., 2019; Shen et al., 2020). Predominantly TNBC affects younger women and is associated with higher relapse rates and higher likelihood of metastasis (Dent et al., 2007; Haffty et al., 2006). While most TNBC patients lack hormone receptor or HER2-targeted treatment options and therefore rely on chemotherapy, a subset of TNBC is associated with hereditary breast and ovarian cancer (HBOC) and may benefit from targeted therapies, highlighting the need for refined stratification and predictive biomarkers (Hwang et al., 2019).

Besides TNBC challenging management, breast cancer is heterogenous disease influenced by both genetic and environmental factors. Most common genetic factors include *BRCA1* and *BRCA2* genes, which play important role in genomic integrity through homologous recombination (HR) mediated DNA repair. Loss of function in these genes disrupts HR pathways which leads to genomic instability and tumour progression (Prakash et al., 2015). This can happen either through loss of function, or it can be inherited germline mutation or somatic alteration within tumour cells (Loboda et al., 2023). Somatic inactivation of *BRCA1* is known to be associated with distinctive molecular patterns, which includes genomic rearrangements and specific mutational signatures. These signatures are called “BRCAness” also known as predictive biomarkers for therapeutic response to platinum-based chemotherapies and PARP inhibitors, highlighting its clinical significance in treatment decision making (Bodily et al., 2020).

Development and advancement in high-throughput technologies such as RNA-sequencing has enabled scientists to look at transcriptomic profiles and molecular landscape of breast cancer (Chen et al., 2021; Hong et al., 2020; Rosati et al., 2024). This enables identification of differentially expressed genes and affected pathways within

specific cancer subtypes, including TNBC. Recent studies describe molecular heterogeneity of TNBC and highlights specific oncogenic drivers and evaluates tumour microenvironment (Kudelova et al., 2022; Shah et al., 2012). Transcriptomics also evaluates implications of somatic *BRCA1* inactivation therefore trying to clarify its role in tumour biology and therapy management (Arakelyan et al., 2021).

This Thesis integrates finding from two complementary studies. The first one explores transcriptome of TNBC, the hub genes and altered pathways associated with this particular breast cancer subtype, which can help in potential biomarkers and therapeutic targets identification. The second part of the study focuses on breast cancer with monoallelic somatic *BRCA1* inactivation, a condition of growing interest since even partial loss of *BRCA1* function may impair DNA repair and influence tumour behaviour. By examining its association with event-free survival, this study provides insights into the prognostic value of somatic *BRCA1* status. Together these studies help to enhance understanding of molecular mechanisms driving breast cancer progression and facilitate improvement in personalised treatment strategies which subsequently improve patient care outcome.

Aim of the Thesis

Characterise the gene expression profiles and altered pathways associated with distinct breast cancer molecular subtypes (triple-negative breast cancer and breast cancer with monoallelic somatic *BRCA1* inactivation) with the goal of identifying potential biomarkers and therapeutic targets, and to evaluate their potential clinical relevance.

Objectives of the Thesis

1. Perform RNA-seq, identify triple negative breast cancer (TNBC) subgroup and its specific transcriptome pattern.
2. Perform functional enrichment and protein-protein interaction analyses in TNBC.
3. Perform RNA-seq, identify transcriptomic pattern of tumours with monoallelic somatic *BRCA1* inactivation.
4. Perform functional enrichment and protein-protein interaction analyses in the *BRCA1* monoallelic inactivation group.
5. Analyse cancer free survival in the group with *BRCA1* monoallelic inactivation.

Hypothesis of the Thesis

Distinct transcriptomic signatures in breast cancer molecular subgroups may serve as predictive biomarkers, enabling personalised therapeutic strategies.

Novelty of the Thesis

Identification of gene expression alterations in TNBC and in breast cancers with BRCA1-associated homologous recombination impairment provides insight into tumour biology and may reveal biomarkers relevant for patient stratification and personalised therapeutic approaches.

1 Materials and methods

Study group

A total of 65 fresh-frozen breast cancer samples, obtained from patients diagnosed and surgically treated between 2012 and 2017, were collected from the tissue repository of the Rīga Stradiņš University Institute of Oncology (RSU IO) and the study design and workflow are shown in Figure 1.1. Ten patients were excluded from the analysis due to predefined exclusion criteria, including the presence of germline *BRCA1* mutations, unconfirmed histopathological diagnosis, and insufficient sequencing quality. Remaining 55 patients had a histologically confirmed breast cancer diagnosis and had not received neoadjuvant chemotherapy, hormone therapy, or radiotherapy prior to surgery. Clinical data including age at diagnosis, therapeutic interventions, and survival outcomes, were available through medical reports.

Following surgery, most patients received adjuvant chemotherapy, with or without hormone therapy (including trastuzumab/Herceptin) and/or radiotherapy. In some cases, patients received hormone therapy or radiotherapy alone or in combination.

The obtained cancer samples were used for two main research components of this Thesis:

1. Transcriptome profiling of TNBC – aiming to identify differentially expressed genes and alterations in signalling pathways associated with triple negative breast cancer vs non-triple-negative breast cancer (n = 19).
2. Transcriptome profiling of tumours with monoallelic somatic *BRCA1* inactivation vs tumours with both active *BRCA1* alleles, including Kaplan–Meier survival analysis to evaluate the impact of *BRCA1* status on event-free survival (n = 36).

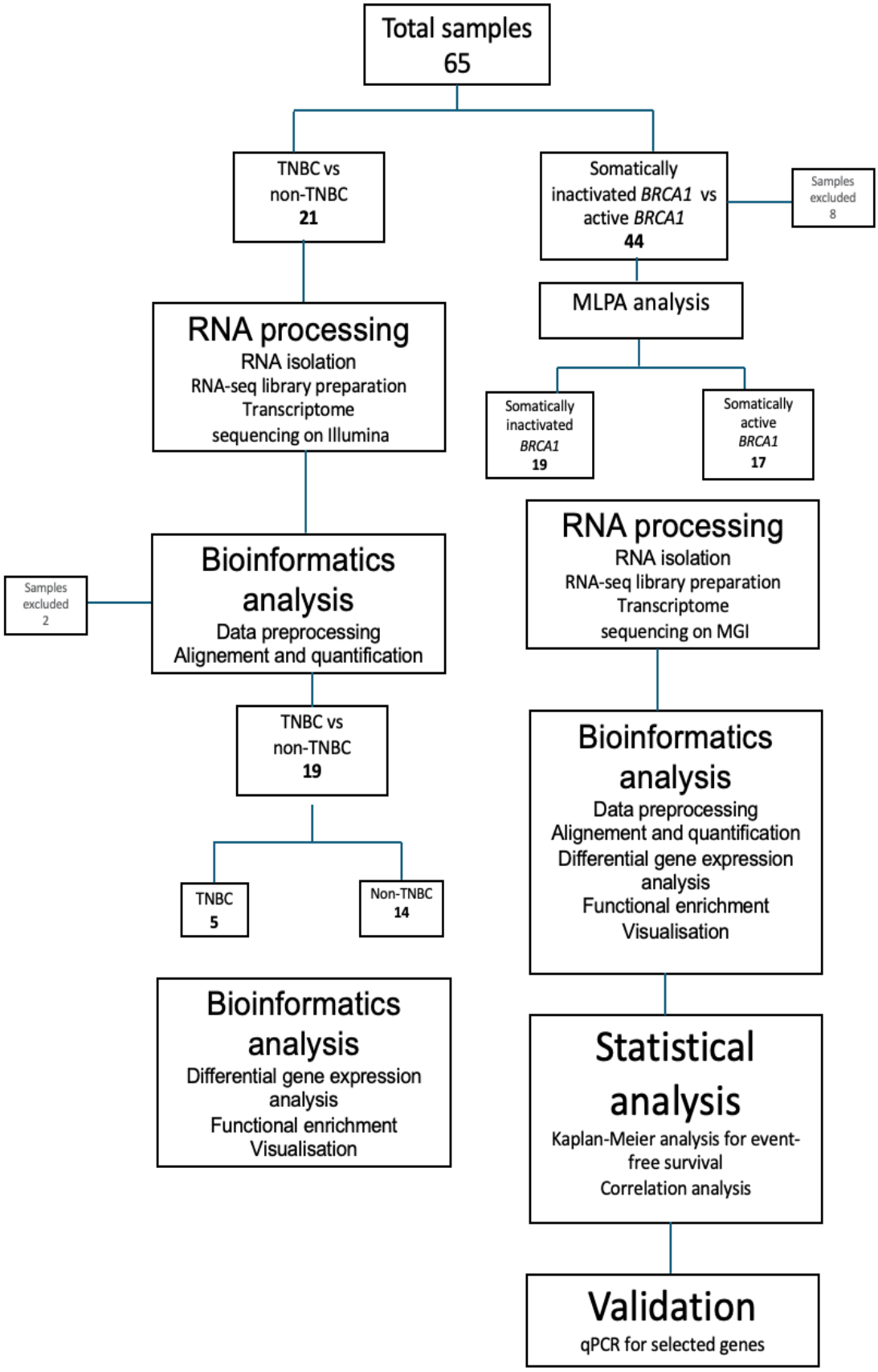

Figure 1.1 **Study design and analytical workflow**

2 Results

The study of this Thesis consists of two parts. The first part is focused on transcriptome analysis with the aim to reveal differentially expressed genes (DEGs) between triple negative breast cancer (TNBC) group and the non-TNBC group. The second part examined DEGs in patient samples with somatic monoallelic *BRCA1* inactivation compared to those with both active *BRCA1* alleles.

2.1 TNBC vs non-TNBC molecular profile

2.1.1 Clinical and pathological characteristics of the study groups

The clinical and pathological characteristics of the TNBC study cohort (n = 19) were analyzed after stratification into TNBC (n = 5) and non-TNBC (n = 14) groups based on acquired RNA-seq data. The median age was lower in the TNBC group (52 years) compared with the non-TNBC group (65.5 years), although this difference did not reach statistical significance. No significant differences were detected between the groups with respect to tumour (T) stage, nodal (N) status, or overall clinical stage. Most tumours in both groups were classified as T2 and clinically staged as I or II. Molecular subtype distribution reflected the expected biology: luminal and HER2-positive tumours were observed exclusively in the non-TNBC group, whereas the TNBC group consisted solely of triple-negative cases. Adjuvant treatment patterns, including chemotherapy, endocrine therapy, and radiotherapy, were comparable between groups. Trastuzumab therapy was administered only in HER2-positive non-TNBC cases, as expected. Although not statistically significant, overall survival tended to be lower in the TNBC subgroup at the last follow-up in 2025.

2.1.2 Transcriptome analysis

The study included sequencing of 19 samples, yielding a median Q30 quality score of 85.4 % and an average read count of 40 million per sample. The TNBC samples were sequenced at approximately 300× depth. Based on global transcriptome analysis, samples were classified into two distinct groups: the triple-negative breast cancer (TNBC) group, which consisted of 5 samples with low or not identified transcripts of ER, PR, and HER2 genes, and the non-TNBC group, comprising the remaining 14 samples. The sequencing results identified a total of 53,854 (DEGs). After applying Bonferroni correction ($p < 0.05$), 229 DEGs were statistically significant, including 124 genes that were downregulated and 105 genes that were upregulated in the TNBC group.

2.1.3 Gene ontology (GO) enrichment analysis

Gene Ontology enrichment analysis of the 42 upregulated genes revealed significant associations across molecular function, biological process, and cellular component categories. In the biological process group, 13 genes including *GDF*, *MYF5*, *COL9A1*, *COL11A2*, *COL19A1*, *DLK1*, *EXTL1*, *ZIC1*, *ALX1*, *NCAN*, *DLX6*, *MMP9* and *EN1* were enriched in skeletal system development ($p = 2.06 \times 10^{-7}$). A set of 10 genes (*COL9A1, MYF5, COL11A2, COL19A1, COL5A3, NCAN, LOXL4, LOXL3, MMP9, MIA*) showed strong enrichment for extracellular matrix and collagen organisation ($p = 4 \times 10^{-6}$), while *CD163, LOXL4, LOXL3,* and *MARCO* were implicated in scavenger receptor and cargo receptor activity ($p < 1 \times 10^{-4}$). At the molecular function level, enrichment was observed for extracellular matrix structural constituents conferring tensile strength and protein-lysine oxidase activity, driven largely by collagen and *LOXL* family members. Within the cellular component category, the most significant terms included collagen trimer (*COL11A2, COL19A1, COL5A3, C1QL2, C1QL4, FCN1, MARCO*; $p = 1.23 \times 10^{-9}$) and collagen-containing extracellular matrix ($p = 1.64 \times 10^{-5}$).

In contrast, the 11 downregulated genes (*ABCC8, KCNJ11, SHROOM3, RAB17, PDZK1, ABCC11, SHROOM1, P2RY2, RAB27B, GP2, TJP3*) showed enrichment only within cellular component categories, including inward-rectifying potassium channels and the apical plasma membrane.

2.1.4 PPI network analysis and hub gene detection

To examine the interaction among proteins encoded by the differentially expressed genes (DEGs), a protein–protein interaction (PPI) network was created using data from the STRING database. This network consisted of 188 nodes and 182 edges. Following this, clustering analysis identified three distinct functional modules within the network. Module 1 (Figure 2.1) is enriched in collagen and basement-membrane components, forming a tight extracellular matrix interaction cluster consistent with structural remodelling. Its connectivity suggests coordinated regulation of matrix organisation that can facilitate changes in tissue architecture linked to invasion. Module 2 (Figure 2.2) represents a canonical luminal/oestrogen receptor regulatory circuit, with *ESR1* linked to key cooperating transcription factors *GATA3* and *FOXA1* and downstream luminal markers (*TFF1*/*TFF3*, *SLC39A6*). Its presence within the network points to an ER/luminal-like program captured in the data, suggesting biological heterogeneity within the analysed samples. Module 3 (Figure 2.3) contains two connected subgroups: a cluster of tumour-associated surface proteins (*MSLN*, *LY6K*, *GP2*, *LYPD6B*) and a cluster cantered on *MMP9* that includes *MPO*, *HMOX1*, and *CD163*. The two subgroups are linked through *MSLN*, forming a single module with a denser interaction core around *MMP9* and *MSLN*.

Hub genes were then selected leading to the identification of eight key hub genes: *FOXA1, ESR1, TFF1, GATA3, TFF3, AR, SLC39A6,* and *COL9A1* (Figure 2.4). Notably, seven of these hub genes (*FOXA1, ESR1, TFF1, GATA3, TFF3, AR, SLC39A6*) showed reduced expression in the TNBC group when compared to the non-TNBC group, while *COL9A1* was the only gene with increased expression in the TNBC group. The genes were ranked according to their importance within the network: *ESR1* was the highest ranked, followed by *FOXA1* and *GATA3* in shared second position, *TFF1* and *TFF3* in third, *SLC39A6* in fourth, and both *COL9A1* and *AR* in fifth place.

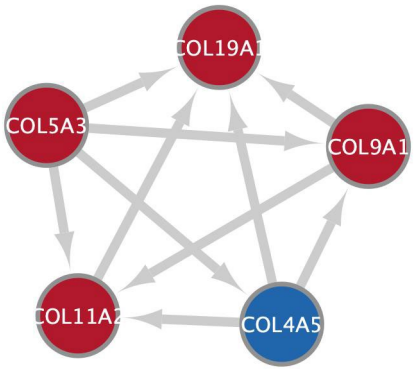

Figure 2.1 **Significant module-1 selected from PPI network including genes *COL5A3, COL19A1, COL9A1, COL5A5, COL11A2***

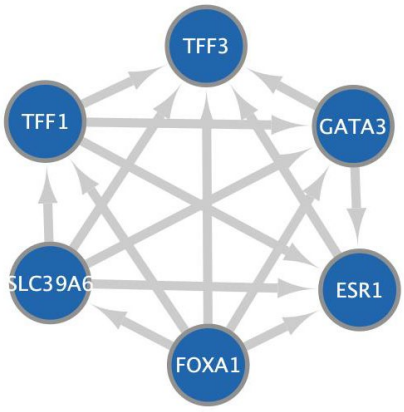

Figure 2.2 **Significant module-2 selected from PPI network including genes *TFF1, TFF3, GATA3, FOXA3, ESR1, SLC39A6***

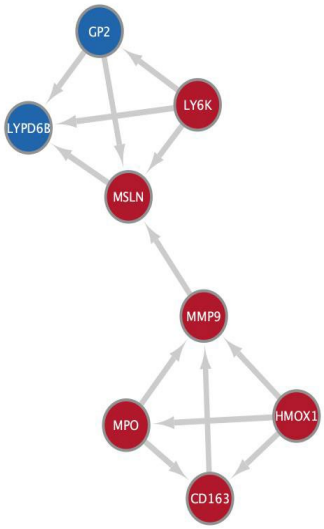

Figure 2.3 **Significant module-3 selected from PPI network including genes GP2, *LY6K, MSLN, LYPD6B, MMP9, HMOX1, CD163, MPO***

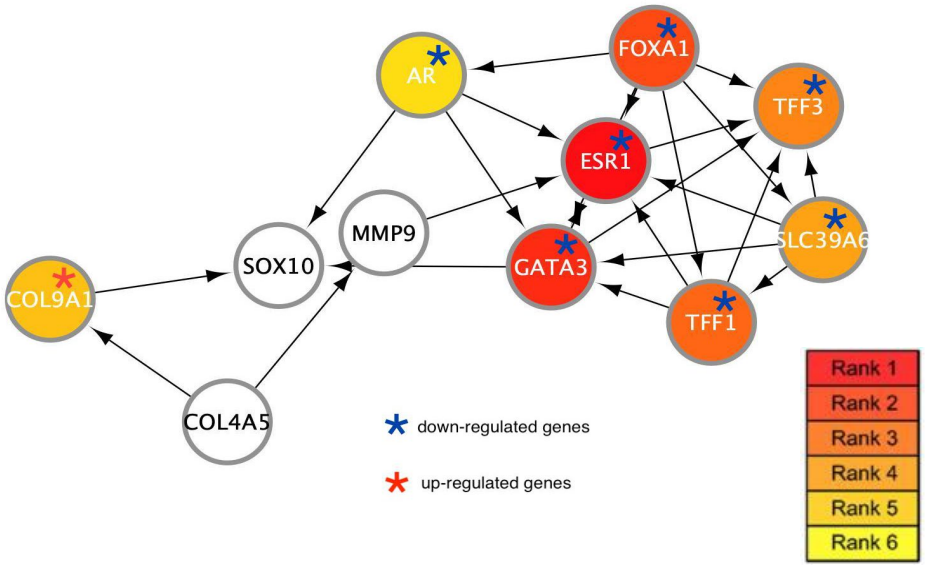

Figure 2.4 **Subnetwork of 8 hub genes (*ESR1, FOXA1, GATA3, TFF1, TFF3, SLC39A6, AR, COL9A1*)**

2.2 *BRCA1* monoallelic somatic inactivation molecular profile

2.2.1 Clinical and pathological characteristics of the study groups

A cohort of 36 tumour samples was classified into two study groups based on *BRCA1* status, which was determined using MLPA analysis: those with monoallelic somatic inactivation and those without. Among these, 16 samples exhibited a monoallelic deletion within the *BRCA1* promoter region, while one sample demonstrated promoter hypermethylation. The remaining 19 samples showed no evidence of deletion or methylation in the *BRCA1* promoter region. Accordingly, the cohort was divided into two groups: *BRCA1*−, representing cases with monoallelic somatic inactivation, and *BRCA1*+, representing tumours with two active *BRCA1* alleles.

The clinical and pathological characteristics of the study cohort (n = 36) were analysed after stratification into *BRCA1*− (n = 17) and *BRCA1*+ (n = 19) groups. The median age was slightly higher in the *BRCA1*+ group (68 years) compared with the *BRCA1*− group (58 years). No statistically significant differences were observed between groups regarding tumour (T) or nodal (N) stage, clinical stage, histological grade, Ki-67 proliferation index, or molecular subtype. Most tumours were classified as T2 stage, grade 2, and luminal A or luminal B molecular type. With respect to treatment, both groups received chemotherapy, endocrine therapy, and radiotherapy at comparable frequencies, while trastuzumab was administered only in the BRCA1− subgroup.

2.2.2 Differentially expressed genes

Analysis of RNA sequencing (RNA-seq) data identified 39 differentially expressed genes (DEGs) between the study groups, using Bonferroni-adjusted $p < 0.05$ and a maximum group mean > 10 as selection criteria. Of these, 23 genes were upregulated and 16 were downregulated in the *BRCA1-* group. It's noteworthy that, except for four transcripts, all identified DEGs were protein-coding genes. The remaining four corresponded to lncRNAs or rRNAs.

The volcano plot (Figure 2.5) displays the overall distribution of gene expression alterations, represented by log2 fold change. Among the most strongly upregulated transcripts were *TRH*, *MMP9*, *TPSD1*, and *CGA*, while *CARTPT*, *CHGB*, and *IRS4* showed notable downregulation.

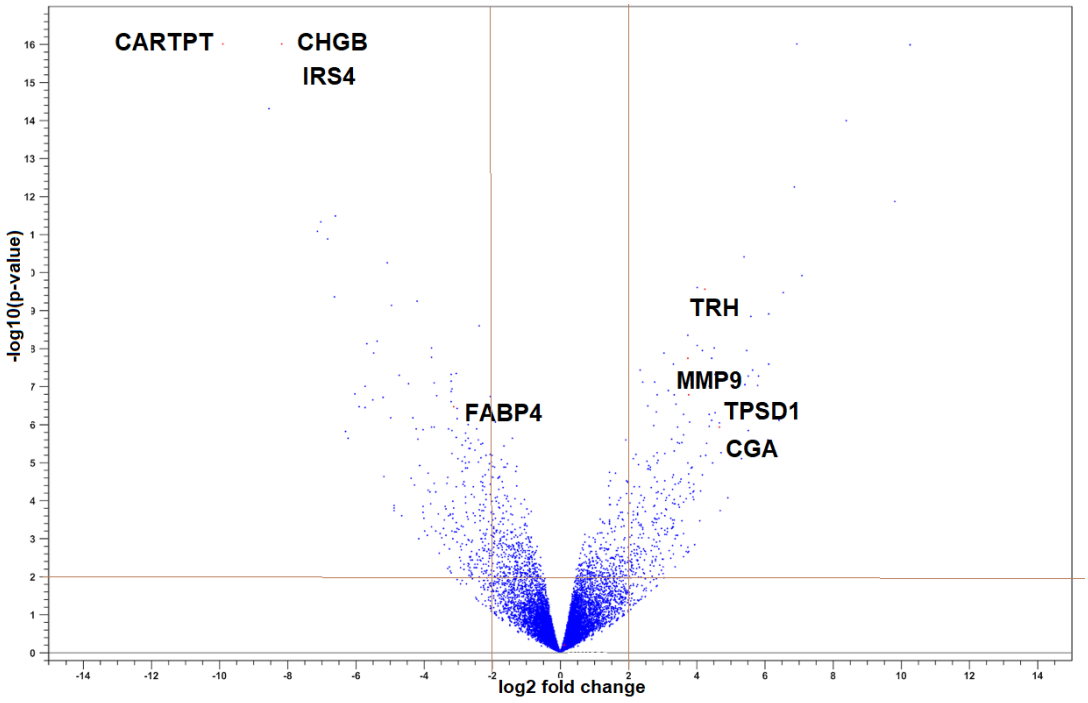

Figure 2.5 **Volcano plot of differentially expressed genes between *BRCA1*− and *BRCA*+ groups**

To investigate the functional connections among proteins encoded by the identified DEGs, we employed the STRING tool (Szklarczyk et al., 2019). Through this analysis, we gained valuable insights into the molecular pathways and processes possibly linked to monoallelic somatic inactivation of the *BRCA1* gene in breast cancer, as shown in Figure 2.6. Following hub gene analysis identified *MMP9* and *GPX2* as central nodes with the network.

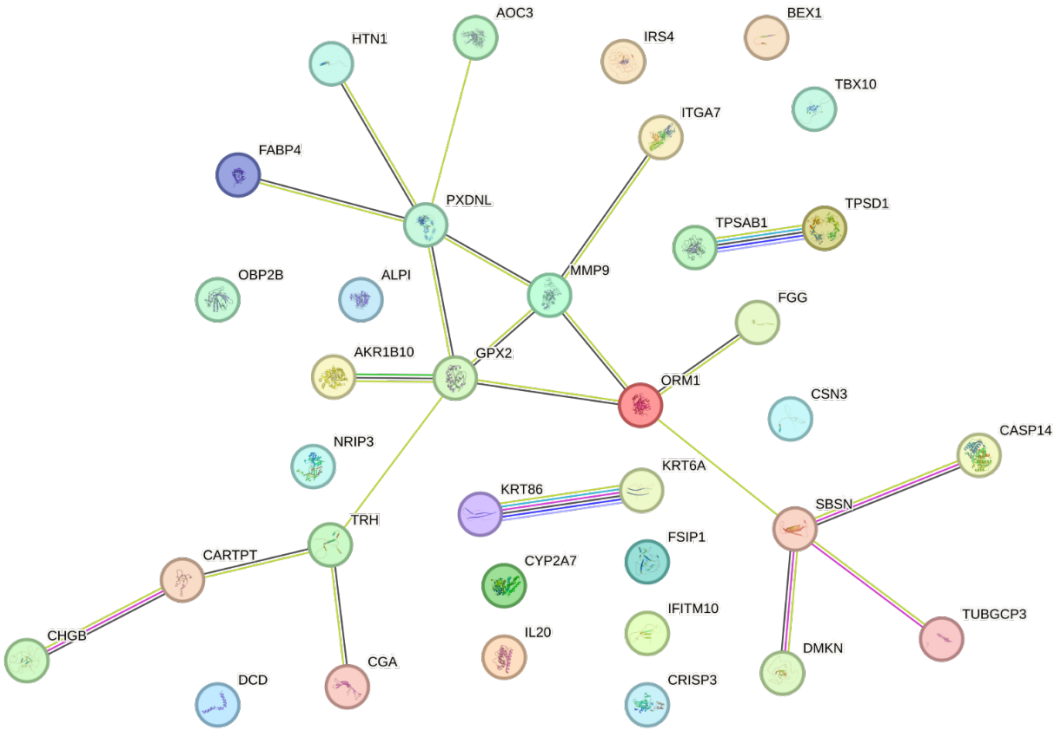

Figure 2.6 **Protein-protein interaction (PPI) network of DEGs in the *BRCA1* monoallelic inactivation group**

Gene Ontology (GO) cellular component analysis of the differentially expressed genes (DEGs) showed enrichment in the extracellular region and extracellular space. These terms exhibited the largest number of associated genes and the lowest false discovery rates (FDR), suggesting a strong overrepresentation of extracellular protein-encoding genes.

The enrichment analysis conducted using the STRING tool revealed significant enrichment of proteins within the extracellular region (GO:0005576) and extracellular space (GO:0005615). Notable genes associated with these categories include *TPSD1, FABP4, ORM1, ALPI, CARTPT, TRH, CSN3,* and *MMP9,* among others. Additionally, the identification of thyroid gland disease as an enriched category suggests potential connections between breast cancer and thyroid dysfunction, with enriched genes such as *TRH, IRS4, CHGB,* and *CGA* possibly contributing to the interaction between breast cancer and thyroid-related processes.

Conclusions

1. RNA-sequencing identified 53 854 expressed genes, with 229 differentially expressed between TNBC and non-TNBC tumours, defining a distinct TNBC-associated transcriptomic profile enriched for extracellular matrix-related pathways, including collagen organisation and extracellular structural functions.
2. Gene Ontology enrichment confirmed that TNBC-related DEGs are functionally linked to extracellular structure organisation and ECM integrity. Protein–protein interaction analysis identified hub genes *FOXA1, ESR1, TFF1, GATA3, TFF3, AR, SLC39A6,* and *COL9A1*, highlighting their central role in regulating the tumour microenvironment and endocrine signalling pathways.
3. Transcriptome analysis of tumours with monoallelic somatic *BRCA1* inactivation identified 39 differentially expressed genes, predominantly enriched in extracellular region–related processes, indicating altered tumour-stroma interactions.
4. Functional enrichment analysis of tumours with monoallelic *BRCA1* inactivation showed significant enrichment of genes in extracellular region categories, including *TPSD1, FABP4, ORM1, ALPI, CARTPT, TRH, CSN3,* and *MMP9*. Protein–protein interaction analysis identified *GPX2* and *MMP9* as hub genes, with *GPX2* consistently ranking highest across both algorithms.
5. A tendency toward prolonged event-free survival in patients with monoallelic somatic *BRCA1* inactivation was revealed by Kaplan-Meier analysis.

Proposals

The findings of this Doctoral Thesis demonstrate the value of transcriptomic profiling for improving molecular stratification of breast cancer, particularly in clinically challenging subgroups. It is proposed that future studies validate the identified subtype-specific gene expression signatures and hub genes in larger, independent cohorts, with special emphasis on triple-negative breast cancer. Such validation is necessary to determine their reliability as prognostic or predictive biomarkers and to assess their added value beyond standard clinicopathological classification.

The results further indicate that monoallelic somatic *BRCA1* inactivation represents a biologically distinct phenotype with specific transcriptomic alterations and potential prognostic relevance. Future research should therefore move beyond a binary interpretation of *BRCA1* status and systematically investigate partial *BRCA1* loss, its association with homologous recombination related pathways, and its impact on treatment response, particularly to DNA-damaging agents and PARP inhibitors.

In addition, functional validation of selected differentially expressed genes is proposed to clarify their role in tumour biology and therapy sensitivity. Finally, integrating transcriptomic data with clinical and pathological parameters in prospective studies could support more precise patient stratification and facilitate the development of personalised treatment strategies in breast cancer care.

List of publications, reports and patents on the topic of the Thesis

Publications:

1. **Kuzņecova, E.,** Daneberga, Z., Berga-Švītiņa, E., Nakazawa-Miklaševiča, M., Irmejs, A., Gardovskis, J., Miklaševičs, E. 2023. Identification of Altered Transcripts and Pathways in Triple Negative Breast Cancer. Proceedings of the Latvian Academy of Sciences. *Section B. Natural, Exact, and Applied Sciences.*, *77*(1), 33–40. https://doi.org/10.2478/prolas-2023-0004
2. **Kuznecova, E.,** Nakazawa-Miklasevica, M., Krike, N., Satcs, M., Sivina, E., Irmejs, A., Loza, P., Gardovskis, J., Miklasevics, E., Daneberga, Z. The Transcriptomic Profile Underlying Somatic Monoallelic *BRCA1* Inactivation: A Biomarker for Breast Cancer Prognosis. *Diagnostics*. 2025; 15(16):2037. https://doi.org/10.3390/diagnostics15162037

Reports and theses at international congresses and conferences:

1. **Kuzņecova, E**., Daneberga, Z., Nakazawa-Miklaševiča, M., Berga-Švītiņa, E., Pirsko, V., Miklaševičs, E., Irmejs, A., Maksimenko, J. HER2-positive Breast Cancer Gene Expression Influenced Pathway Analysis. Poster presentation at Rīga Stradiņš University International Research Conference on Medical and Health Care Sciences "Knowledge for Use in Practice": Abstracts, 1.–3.04.2019., 60.
2. **Kuzņecova, E**., Daneberga, Z., Nakazawa-Mikaševiča, M., Irmejs, A., Miklaševičs, E. Gene expression patterns as useful biomarkers for TNBC patients. Oral presentation at Rīga Stradiņš University International Research Conference on Medical and Health Care Sciences "Knowledge for Use in Practice": Abstracts 29.–31.03.2023.

References

1. Albergaria, A., Paredes, J., Sousa, B., Milanezi, F., Carneiro, V., Bastos, J., Costa, S., Vieira, D., Lopes, N., Lam, E. W., Lunet, N., & Schmitt, F. (2009). Expression of FOXA1 and GATA-3 in breast cancer: the prognostic significance in hormone receptor-negative tumours. *Breast Cancer Research*, *11*(3), R40. https://doi.org/10.1186/bcr2327

2. Al-Kabariti, A. Y., & Abbas, M. A. (2023). Progress in the Understanding of Estrogen Receptor Alpha Signaling in Triple-Negative Breast Cancer: Reactivation of Silenced ER-α and Signaling through ER-α36. *Molecular Cancer Research*, *21*(11), 1123–1138. https://doi.org/10.1158/1541-7786.MCR-23-0321

3. An, X., Yu, W., Liu, J., Tang, D., Yang, L., & Chen, X. (2024). Oxidative cell death in cancer: mechanisms and therapeutic opportunities. *Cell Death & Disease*, *15*(8), 556. https://doi.org/10.1038/s41419-024-06939-5

4. Arakelyan, A., Melkonyan, A., Hakobyan, S., Boyarskih, U., Simonyan, A., Nersisyan, L., Nikoghosyan, M., Filipenko, M., & Binder, H. (2021). Transcriptome Patterns of BRCA1- and BRCA2- Mutated Breast and Ovarian Cancers. *International Journal of Molecular Sciences*, *22*(3), 1266. https://doi.org/10.3390/ijms22031266

5. Arnold, M., Morgan, E., Rumgay, H., Mafra, A., Singh, D., Laversanne, M., Vignat, J., Gralow, J. R., Cardoso, F., Siesling, S., & Soerjomataram, I. (2022). Current and future burden of breast cancer: Global statistics for 2020 and 2040. *The Breast*, *66*, 15–23. https://doi.org/10.1016/j.breast.2022.08.010

6. Asano, Y., Kashiwagi, S., Goto, W., Takada, K., Takahashi, K., Hatano, T., Noda, S., Takashima, T., Onoda, N., Tomita, S., Motomura, H., Ohsawa, M., Hirakawa, K., & Ohira, M. (2017). Prediction of survival after neoadjuvant chemotherapy for breast cancer by evaluation of tumor-infiltrating lymphocytes and residual cancer burden. *BMC Cancer*, *17*(1), 888. https://doi.org/10.1186/s12885-017-3927-8

7. Babar, Q., Saeed, A., Tabish, T. A., Sarwar, M., & Thorat, N. D. (2023). Targeting the tumor microenvironment: Potential strategy for cancer therapeutics. *Biochimica et Biophysica Acta (BBA) - Molecular Basis of Disease*, *1869*(6), 166746. https://doi.org/10.1016/j.bbadis.2023.166746

8. Bacha, R., Alwisi, N., Ismail, R., Pedersen, S., & Al-Mansoori, L. (2024). Unveiling GATA3 Signaling Pathways in Health and Disease: Mechanisms, Implications, and Therapeutic Potential. *Cells*, *13*(24), 2127. https://doi.org/10.3390/cells13242127

9. Bai, F., Zheng, C., Liu, X., Chan, H. L., Liu, S., Ma, J., Ren, S., Zhu, W.-G., & Pei, X.-H. (2022). Loss of function of GATA3 induces basal-like mammary tumors. *Theranostics*, *12*(2), 720–733. https://doi.org/10.7150/thno.65796

10. Baldini, E., Lauro, A., Tripodi, D., Pironi, D., Amabile, M. I., Ferent, I. C., Lori, E., Gagliardi, F., Bellini, M. I., Forte, F., Pacini, P., Cantisani, V., D'Andrea, V., Sorrenti, S., & Ulisse, S. (2022). Thyroid Diseases and Breast Cancer. *Journal of Personalized Medicine*, *12*(2), 156. https://doi.org/10.3390/jpm12020156

11. Bodily, W. R., Shirts, B. H., Walsh, T., Gulsuner, S., King, M.-C., Parker, A., Roosan, M., & Piccolo, S. R. (2020). Effects of germline and somatic events in candidate BRCA-like genes on breast-tumor signatures. *PLOS ONE*, *15*(9), e0239197. https://doi.org/10.1371/journal.pone.0239197

12. Bray, F., Ferlay, J., Soerjomataram, I., Siegel, R. L., Torre, L. A., & Jemal, A. (2018). Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA: A Cancer Journal for Clinicians*, *68*(6), 394–424. https://doi.org/10.3322/caac.21492

13. Brennan, D. J., O'Connor, D. P., Laursen, H., McGee, S. F., McCarthy, S., Zagozdzon, R., Rexhepaj, E., Culhane, A. C., Martin, F. M., Duffy, M. J., Landberg, G., Ryden, L., Hewitt, S. M., Kuhar, M. J., Bernards, R., Millikan, R. C., Crown, J. P., Jirström, K., & Gallagher, W. M. (2012). The cocaine- and amphetamine-regulated transcript mediates ligand-independent activation of ERα, and is an independent prognostic factor in node-negative breast cancer. *Oncogene*, *31*(30), 3483–3494. https://doi.org/10.1038/onc.2011.519

14. Chen, G., Yu, M., Cao, J., Zhao, H., Dai, Y., Cong, Y., & Qiao, G. (2021). Identification of candidate biomarkers correlated with poor prognosis of breast cancer based on bioinformatics analysis. *Bioengineered*, *12*(1), 5149–5161. https://doi.org/10.1080/21655979.2021.1960775

15. Chin, C.-H., Chen, S.-H., Wu, H.-H., Ho, C.-W., Ko, M.-T., & Lin, C.-Y. (2014). cytoHubba: identifying hub objects and sub-networks from complex interactome. *BMC Systems Biology*, *8*(S4), S11. https://doi.org/10.1186/1752-0509-8-S4-S11

16. Collins, L. C., Cole, K. S., Marotti, J. D., Hu, R., Schnitt, S. J., & Tamimi, R. M. (2011). Androgen receptor expression in breast cancer in relation to molecular phenotype: results from the Nurses' Health Study. *Modern Pathology*, *24*(7), 924–931. https://doi.org/10.1038/modpathol.2011.54

17. Cortellino, S., & Longo, V. D. (2023). Metabolites and Immune Response in Tumor Microenvironments. *Cancers*, *15*(15), 3898. https://doi.org/10.3390/cancers15153898

18. Dai, X., Cheng, H., Chen, X., Li, T., Zhang, J., Jin, G., Cai, D., & Huang, Z. (2019). FOXA1 is Prognostic of Triple Negative Breast Cancers by Transcriptionally Suppressing *SOD2* and *IL6*. *International Journal of Biological Sciences*, *15*(5), 1030–1041. https://doi.org/10.7150/ijbs.31009

19. De Lara, S., Nyqvist, J., Werner Rönnerman, E., Helou, K., Kenne Sarenmalm, E., Einbeigi, Z., Karlsson, P., Parris, T. Z., & Kovács, A. (2019). The prognostic relevance of FOXA1 and Nestin expression in breast cancer metastases: a retrospective study of 164 cases during a 10-year period (2004–2014). *BMC Cancer*, *19*(1), 187. https://doi.org/10.1186/s12885-019-5373-2

20. Dent, R., Trudeau, M., Pritchard, K. I., Hanna, W. M., Kahn, H. K., Sawka, C. A., Lickley, L. A., Rawlinson, E., Sun, P., & Narod, S. A. (2007). Triple-Negative Breast Cancer: Clinical Features and Patterns of Recurrence. *Clinical Cancer Research*, *13*(15), 4429–4434. https://doi.org/10.1158/1078-0432.CCR-06-3045

21. Dustin, D., Gu, G., & Fuqua, S. A. W. (2019). *ESR1* mutations in breast cancer. *Cancer*, *125*(21), 3714–3728. https://doi.org/10.1002/cncr.32345

22. Elnagdy, M. H., Farouk, O., Seleem, A. K., & Nada, H. A. (2018). *TFF1* and *TFF3* mRNAs Are Higher in Blood from Breast Cancer Patients with Metastatic Disease than Those without. *Journal of Oncology*, *2018*, 1–8. https://doi.org/10.1155/2018/4793498

23. Fan, K. Y., Chehade, R., Qazi, M., Moravan, V., Nofech-Mozes, S., & Jerzak, K. J. (2023). Androgen Receptor Is Expressed in the Majority of Breast Cancer Brain Metastases and Is Subtype-Dependent. *Cancers*, *15*(10), 2748. https://doi.org/10.3390/cancers15102748

24. Furuhashi, M., Saitoh, S., Shimamoto, K., & Miura, T. (2014). Fatty Acid-Binding Protein 4 (FABP4): Pathophysiological Insights and Potent Clinical Biomarker of Metabolic and Cardiovascular Diseases. *Clinical Medicine Insights: Cardiology*, *8s3*, CMC.S17067. https://doi.org/10.4137/CMC.S17067

25. Garcia-Martinez, L., Zhang, Y., Nakata, Y., Chan, H. L., & Morey, L. (2021). Epigenetic mechanisms in breast cancer therapy and resistance. *Nature Communications*, *12*(1), 1786. https://doi.org/10.1038/s41467-021-22024-3

26. Garrido-Castro, A. C., Lin, N. U., & Polyak, K. (2019). Insights into Molecular Classifications of Triple-Negative Breast Cancer: Improving Patient Selection for Treatment. *Cancer Discovery*, *9*(2), 176–198. https://doi.org/10.1158/2159-8290.CD-18-1177

27. Gerratana, L., Basile, D., Buono, G., De Placido, S., Giuliano, M., Minichillo, S., Coinu, A., Martorana, F., De Santo, I., Del Mastro, L., De Laurentiis, M., Puglisi, F., & Arpino, G. (2018). Androgen receptor in triple negative breast cancer: A potential target for the targetless subtype. *Cancer Treatment Reviews*, *68*, 102–110. https://doi.org/10.1016/j.ctrv.2018.06.005

28. Giovanella, L., Marelli, M., Ceriani, L., Giardina, G., Garancini, S., & Colombo, L. (2001). Evaluation of Chromogranin a Expression in Serum and Tissues of Breast Cancer Patients. *The International Journal of Biological Markers*, *16*(4), 268–272. https://doi.org/10.1177/172460080101600408

29. Gonzalez-Avila, G., Sommer, B., Mendoza-Posada, D. A., Ramos, C., Garcia-Hernandez, A. A., & Falfan-Valencia, R. (2019). Matrix metalloproteinases participation in the metastatic process and their diagnostic and therapeutic applications in cancer. *Critical Reviews in Oncology/Hematology*, *137*, 57–83. https://doi.org/10.1016/j.critrevonc.2019.02.010

30. Govindan, S., Siraganahalli Eswaraiah, M., Basavaraj, C., Adinarayan, M., Sankaran, S., & Bakre, M. (2020). Androgen Receptor mRNA levels determine the prognosis in triple-negative breast cancer patients. *BMC Cancer*, *20*(1), 745. https://doi.org/10.1186/s12885-020-07218-0

31. Guijarro, L. G., Justo Bermejo, F. J., Boaru, D. L., De Castro-Martinez, P., De Leon-Oliva, D., Fraile-Martínez, O., Garcia-Montero, C., Alvarez-Mon, M., Toledo-Lobo, M. del V., & Ortega, M. A. (2023a). Is Insulin Receptor Substrate4 (IRS4) a Platform Involved in the Activation of Several Oncogenes? *Cancers*, *15*(18), 4651. https://doi.org/10.3390/cancers15184651

32. Haffty, B. G., Yang, Q., Reiss, M., Kearney, T., Higgins, S. A., Weidhaas, J., Harris, L., Hait, W., & Toppmeyer, D. (2006). Locoregional Relapse and Distant Metastasis in Conservatively Managed Triple Negative Early-Stage Breast Cancer. *Journal of Clinical Oncology*, *24*(36), 5652–5657. https://doi.org/10.1200/JCO.2006.06.5664

33. Hao, J., Yan, F., Zhang, Y., Triplett, A., Zhang, Y., Schultz, D. A., Sun, Y., Zeng, J., Silverstein, K. A. T., Zheng, Q., Bernlohr, D. A., Cleary, M. P., Egilmez, N. K., Sauter, E., Liu, S., Suttles, J., & Li, B. (2018). Expression of Adipocyte/Macrophage Fatty Acid–Binding Protein in Tumor-Associated Macrophages Promotes Breast Cancer Progression. *Cancer Research*, *78*(9), 2343–2355. https://doi.org/10.1158/0008-5472.CAN-17-2465

34. Hong, M., Tao, S., Zhang, L., Diao, L.-T., Huang, X., Huang, S., Xie, S.-J., Xiao, Z.-D., & Zhang, H. (2020). RNA sequencing: new technologies and applications in cancer research. *Journal of Hematology & Oncology*, *13*(1), 166. https://doi.org/10.1186/s13045-020-01005-x

35. Hu, H., Tong, K., Tsang, J. Y., Ko, C. W., Tam, F., Loong, T. C., & Tse, G. M. (2024). Subtyping of triple-negative breast cancers: its prognostication and implications in diagnosis of breast origin. *ESMO Open*, *9*(4), 102993. https://doi.org/10.1016/j.esmoop.2024.102993

36. Huang, H. (2018). Matrix Metalloproteinase-9 (MMP-9) as a Cancer Biomarker and MMP-9 Biosensors: Recent Advances. *Sensors*, *18*(10), 3249. https://doi.org/10.3390/s18103249

37. Hwang, K.-T., Kim, J., Jung, J., Chang, J. H., Chai, Y. J., Oh, S. W., Oh, S., Kim, Y. A., Park, S. B., & Hwang, K. R. (2019). Impact of Breast Cancer Subtypes on Prognosis of Women with Operable Invasive Breast Cancer: A Population-based Study Using SEER Database. *Clinical Cancer Research*, *25*(6), 1970–1979. https://doi.org/10.1158/1078-0432.CCR-18-2782

38. Iamaroon, A., Pongsiriwet, S., Jittidecharaks, S., Pattanaporn, K., Prapayasatok, S., & Wanachantararak, S. (2003). Increase of mast cells and tumor angiogenesis in oral squamous cell carcinoma. *Journal of Oral Pathology & Medicine*, *32*(4), 195–199. https://doi.org/10.1034/j.1600-0714.2003.00128.x

39. Ikink, G. J., Boer, M., Bakker, E. R. M., & Hilkens, J. (2016). IRS4 induces mammary tumorigenesis and confers resistance to HER2-targeted therapy through constitutive PI3K/AKT-pathway hyperactivation. *Nature Communications*, *7*(1), 13567. https://doi.org/10.1038/ncomms13567

40. Kankkunen, J.-P., Harvima, I. T., & Naukkarinen, A. (1997). Quantitative analysis of tryptase and chymase containing mast cells in benign and malignant breast lesions. *International Journal of Cancer*, *72*(3), 385–388. https://doi.org/10.1002/(SICI)1097-0215(19970729)72:3<385::AID-IJC1>3.0.CO;2-L

41. Kast, K., John, E. M., Hopper, J. L., Andrieu, N., Noguès, C., Mouret-Fourme, E., Lasset, C., Fricker, J.-P., Berthet, P., Mari, V., Salle, L., Schmidt, M. K., Ausems, M. G. E. M., Garcia, E. B. G., van de Beek, I., Wevers, M. R., Evans, D. G., Tischkowitz, M., Lalloo, F., … Engel, C. (2023). Associations of height, body mass index, and weight gain with breast cancer risk in carriers of a pathogenic variant in BRCA1 or BRCA2: the BRCA1 and BRCA2 Cohort Consortium. *Breast Cancer Research*, *25*(1), 72. https://doi.org/10.1186/s13058-023-01673-w

42. Kesson, E. M., Allardice, G. M., George, W. D., Burns, H. J. G., & Morrison, D. S. (2012). Effects of multidisciplinary team working on breast cancer survival: retrospective, comparative, interventional cohort study of 13 722 women. *BMJ*, *344*(apr26 1), e2718–e2718. https://doi.org/10.1136/bmj.e2718

43. Kim, S., Moon, B.-I., Lim, W., Park, S., Cho, M. S., & Sung, S. H. (2016). Expression patterns of GATA3 and the androgen receptor are strongly correlated in patients with triple-negative breast cancer. *Human Pathology*, *55*, 190–195. https://doi.org/10.1016/j.humpath.2016.04.013

44. Kudelova, E., Smolar, M., Holubekova, V., Hornakova, A., Dvorska, D., Lucansky, V., Koklesova, L., Kudela, E., & Kubatka, P. (2022). Genetic Heterogeneity, Tumor Microenvironment and Immunotherapy in Triple-Negative Breast Cancer. *International Journal of Molecular Sciences*, *23*(23), 14937. https://doi.org/10.3390/ijms232314937

45. Kumar, U., Ardasheva, A., Mahmud, Z., Coombes, R. C., & Yagüe, E. (2021). FOXA1 is a determinant of drug resistance in breast cancer cells. *Breast Cancer Research and Treatment*, *186*(2), 317–326. https://doi.org/10.1007/s10549-020-06068-5

46. Lehmann, B. D., Bauer, J. A., Chen, X., Sanders, M. E., Chakravarthy, A. B., Shyr, Y., & Pietenpol, J. A. (2011). Identification of human triple-negative breast cancer subtypes and preclinical models for selection of targeted therapies. *Journal of Clinical Investigation*, *121*(7), 2750–2767. https://doi.org/10.1172/JCI45014

47. Lehmann, B. D., Jovanović, B., Chen, X., Estrada, M. V., Johnson, K. N., Shyr, Y., Moses, H. L., Sanders, M. E., & Pietenpol, J. A. (2016). Refinement of Triple-Negative Breast Cancer Molecular Subtypes: Implications for Neoadjuvant Chemotherapy Selection. *PLOS ONE*, *11*(6), e0157368. https://doi.org/10.1371/journal.pone.0157368

48. Leifler, K. S., Svensson, S., Abrahamsson, A., Bendrik, C., Robertson, J., Gauldie, J., Olsson, A.-K., & Dabrosin, C. (2013). Inflammation Induced by MMP-9 Enhances Tumor Regression of Experimental Breast Cancer. *The Journal of Immunology*, *190*(8), 4420–4430. https://doi.org/10.4049/jimmunol.1202610

49. Li, Y., Zhan, Z., Yin, X., Fu, S., & Deng, X. (2021). Targeted Therapeutic Strategies for Triple-Negative Breast Cancer. *Frontiers in Oncology*, *11*. https://doi.org/10.3389/fonc.2021.731535

50. Liu, S., Wu, D., Fan, Z., Yang, J., Li, Y., Meng, Y., Gao, C., & Zhan, H. (2022). FABP4 in obesity-associated carcinogenesis: Novel insights into mechanisms and therapeutic implications. *Frontiers in Molecular Biosciences*, *9*. https://doi.org/10.3389/fmolb.2022.973955

51. Loboda, A. P., Adonin, L. S., Zvereva, S. D., Guschin, D. Y., Korneenko, T. V., Telegina, A. V., Kondratieva, O. K., Frolova, S. E., Pestov, N. B., & Barlev, N. A. (2023). BRCA Mutations – The Achilles Heel of Breast, Ovarian and Other Epithelial Cancers. *International Journal of Molecular Sciences*, *24*(5), 4982. https://doi.org/10.3390/ijms24054982

52. Lopez, M., Spehner, L., André, F., Viot, J., Seffar, E., Marguier, A., Curtit, E., Meynard, G., Dobi, E., Ladoire, S., Boidot, R., Loyon, R., Derangere, V., Bidard, F.-C., Borg, C., Mansi, L., & Kroemer, M. (2025). Exploring the role of ESR1 mutations in metastatic hormone receptor-positive breast cancer T cell immune surveillance disruption. *Breast Cancer Research*, *27*(1), 19. https://doi.org/10.1186/s13058-025-01962-6

53. Metovic, J., Borella, F., D'Alonzo, M., Biglia, N., Mangherini, L., Tampieri, C., Bertero, L., Cassoni, P., & Castellano, I. (2022a). FOXA1 in Breast Cancer: A Luminal Marker with Promising Prognostic and Predictive Impact. *Cancers*, *14*(19), 4699. https://doi.org/10.3390/cancers14194699

54. Miziak, P., Baran, M., Błaszczak, E., Przybyszewska-Podstawka, A., Kałafut, J., Smok-Kalwat, J., Dmoszyńska-Graniczka, M., Kiełbus, M., & Stepulak, A. (2023). Estrogen Receptor Signaling in Breast Cancer. *Cancers*, *15*(19), 4689. https://doi.org/10.3390/cancers15194689

55. Mohammed, A. A., Elsayed, F. M., Algazar, M., Rashed, H. E., & Anter, A. H. (2020). Neoadjuvant Chemotherapy in Triple Negative Breast Cancer: Correlation between Androgen Receptor Expression and Pathological Response. *Asian Pacific Journal of Cancer Prevention*, *21*(2), 563–568. https://doi.org/10.31557/APJCP.2020.21.2.563

56. Owe-Larsson, M., Pawłasek, J., Piecha, T., Sztokfisz-Ignasiak, A., Pater, M., & Janiuk, I. R. (2023). The Role of Cocaine- and Amphetamine-Regulated Transcript (CART) in Cancer: A Systematic Review. *International Journal of Molecular Sciences*, *24*(12), 9986. https://doi.org/10.3390/ijms24129986

57. Popova, N. V., & Jücker, M. (2022). The Functional Role of Extracellular Matrix Proteins in Cancer. *Cancers*, *14*(1), 238. https://doi.org/10.3390/cancers14010238

58. Prakash, R., Zhang, Y., Feng, W., & Jasin, M. (2015). Homologous Recombination and Human Health: The Roles of BRCA1, BRCA2, and Associated Proteins. *Cold Spring Harbor Perspectives in Biology*, *7*(4), a016600. https://doi.org/10.1101/cshperspect.a016600

59. Qiong, L., & Yin, J. (2021). Orosomucoid 1 promotes epirubicin resistance in breast cancer by upregulating the expression of matrix metalloproteinases 2 and 9. *Bioengineered*, *12*(1), 8822–8832. https://doi.org/10.1080/21655979.2021.1987067

60. Querzoli, P., Pedriali, M., Rinaldi, R., Secchiero, P., Rossi, P. G., & Kuhn, E. (2021a). GATA3 as an Adjunct Prognostic Factor in Breast Cancer Patients with Less Aggressive Disease: A Study with a Review of the Literature. *Diagnostics*, *11*(4), 604. https://doi.org/10.3390/diagnostics11040604

61. Rosati, D., Palmieri, M., Brunelli, G., Morrione, A., Iannelli, F., Frullanti, E., & Giordano, A. (2024). Differential gene expression analysis pipelines and bioinformatic tools for the identification of specific biomarkers: A review. *Computational and Structural Biotechnology Journal*, *23*, 1154–1168. https://doi.org/10.1016/j.csbj.2024.02.018

62. Shah, S. P., Roth, A., Goya, R., Oloumi, A., Ha, G., Zhao, Y., Turashvili, G., Ding, J., Tse, K., Haffari, G., Bashashati, A., Prentice, L. M., Khattra, J., Burleigh, A., Yap, D., Bernard, V., McPherson, A., Shumansky, K., Crisan, A., … Aparicio, S. (2012). The clonal and mutational evolution spectrum of primary triple-negative breast cancers. *Nature*, *486*(7403), 395–399. https://doi.org/10.1038/nature10933

63. Shen, M., Pan, H., Chen, Y., Xu, Y. H., Yang, W., & Wu, Z. (2020). A review of current progress in triple-negative breast cancer therapy. *Open Medicine*, *15*(1), 1143–1149. https://doi.org/10.1515/med-2020-0138

64. Sobiepanek, A., Kuryk, Ł., Garofalo, M., Kumar, S., Baran, J., Musolf, P., Siebenhaar, F., Fluhr, J. W., Kobiela, T., Plasenzotti, R., Kuchler, K., & Staniszewska, M. (2022). The Multifaceted Roles of Mast Cells in Immune Homeostasis, Infections and Cancers. *International Journal of Molecular Sciences*, *23*(4), 2249. https://doi.org/10.3390/ijms23042249

65. Sridhar, N., Iwase, T., Xie, X., Lee, J., Damodaran, S., & Ueno, N. T. (2025). Paving the path ForwARd: Advances and challenges in androgen receptor targeting in breast cancer. *Cancer Treatment Reviews*, *138*, 102958. https://doi.org/10.1016/j.ctrv.2025.102958

66. Sun, N., & Zhao, X. (2022a). Therapeutic Implications of FABP4 in Cancer: An Emerging Target to Tackle Cancer. *Frontiers in Pharmacology*, *13*. https://doi.org/10.3389/fphar.2022.948610

67. Szklarczyk, D., Gable, A. L., Lyon, D., Junge, A., Wyder, S., Huerta-Cepas, J., Simonovic, M., Doncheva, N. T., Morris, J. H., Bork, P., Jensen, L. J., & Mering, C. von. (2019a). STRING v11: protein–protein association networks with increased coverage, supporting functional discovery in genome-wide experimental datasets. *Nucleic Acids Research*, *47*(D1), D607–D613. https://doi.org/10.1093/nar/gky1131

68. Trubacova, R., Drastichova, Z., & Novotny, J. (2022). Biochemical and physiological insights into TRH receptor-mediated signaling. *Frontiers in Cell and Developmental Biology*, *10*. https://doi.org/10.3389/fcell.2022.981452

69. Vera-Badillo, F. E., Templeton, A. J., de Gouveia, P., Diaz-Padilla, I., Bedard, P. L., Al-Mubarak, M., Seruga, B., Tannock, I. F., Ocana, A., & Amir, E. (2014). Androgen Receptor Expression and Outcomes in Early Breast Cancer: A Systematic Review and Meta-Analysis. *JNCI Journal of the National Cancer Institute*, *106*(1), djt319–djt319. https://doi.org/10.1093/jnci/djt319

70. Vtorushin, S., Dulesova, A., & Krakhmal, N. (2022). Luminal androgen receptor (LAR) subtype of triple-negative breast cancer: molecular, morphological, and clinical features. *Journal of Zhejiang University-SCIENCE B*, *23*(8), 617–624. https://doi.org/10.1631/jzus.B2200113

71. Wang, Q., Wang, K., Tan, X., Li, Z., & Wang, H. (2022). Immunomodulatory role of metalloproteases in cancers: Current progress and future trends. *Frontiers in Immunology*, *13*. https://doi.org/10.3389/fimmu.2022.1064033

72. Weisbrod, A. B., Zhang, L., Jain, M., Barak, S., Quezado, M. M., & Kebebew, E. (2013). Altered PTEN, ATRX, CHGA, CHGB, and TP53 Expression Are Associated with Aggressive VHL-Associated Pancreatic Neuroendocrine Tumors. *Hormones and Cancer*, *4*(3), 165–175. https://doi.org/10.1007/s12672-013-0134-1

73. Wells, J. M., Gaggar, A., & Blalock, J. E. (2015). MMP generated matrikines. *Matrix Biology*, *44–46*, 122–129. https://doi.org/10.1016/j.matbio.2015.01.016

74. Winkler, J., Abisoye-Ogunniyan, A., Metcalf, K. J., & Werb, Z. (2020). Concepts of extracellular matrix remodelling in tumour progression and metastasis. *Nature Communications*, *11*(1), 5120. https://doi.org/10.1038/s41467-020-18794-x

75. Wroblewski, M., Bauer, R., Cubas Córdova, M., Udonta, F., Ben-Batalla, I., Legler, K., Hauser, C., Egberts, J., Janning, M., Velthaus, J., Schulze, C., Pantel, K., Bokemeyer, C., & Loges, S. (2017). Mast cells decrease efficacy of anti-angiogenic therapy by secreting matrix-degrading granzyme B. *Nature Communications*, *8*(1), 269. https://doi.org/10.1038/s41467-017-00327-8

76. Yang, Y., Lu, S., Zeng, W., Xie, S., & Xiao, S. (2017). GATA3 expression in clinically useful groups of breast carcinoma: a comparison with GCDFP15 and mammaglobin for identifying paired primary and metastatic tumors. *Annals of Diagnostic Pathology*, *26*, 1–5. https://doi.org/10.1016/j.anndiagpath.2016.09.011

77. Yi, J., Ren, L., Li, D., Wu, J., Li, W., Du, G., & Wang, J. (2020). Trefoil factor 1 (TFF1) is a potential prognostic biomarker with functional significance in breast cancers. *Biomedicine & Pharmacotherapy*, *124*, 109827. https://doi.org/10.1016/j.biopha.2020.109827

78. Yoon, E. C., Wang, G., Parkinson, B., Huo, L., Peng, Y., Wang, J., Salisbury, T., Wu, Y., Chen, H., Albarracin, C. T., Resetkova, E., Middleton, L. P., Krishnamurthy, S., Gan, Q., Sun, H., Huang, X., Shen, T., Chen, W., Parwani, A. V., … Ding, Q. (2022). TRPS1, GATA3, and SOX10 expression in triple-negative breast carcinoma. *Human Pathology*, *125*, 97–107. https://doi.org/10.1016/j.humpath.2022.04.006

79. Yu, T., & Di, G. (2017). Role of tumor microenvironment in triple-negative breast cancer and its prognostic significance. *Chinese Journal of Cancer Research*, *29*(3), 237–252. https://doi.org/10.21147/j.issn.1000-9604.2017.03.10

80. Zhao, Y., Simon, M., Seluanov, A., & Gorbunova, V. (2023). DNA damage and repair in age-related inflammation. *Nature Reviews Immunology*, *23*(2), 75–89. https://doi.org/10.1038/s41577-022-00751-y

Acknowledgments

I would like to begin by expressing my heartfelt appreciation to my scientific supervisors, **Zanda Daneberga** and **Miki Nakazawa-Miklaševiča**, whose guidance and support were essential throughout my doctoral studies. I am grateful to **E. Miklaševičs** for offering me the opportunity to initiate PhD studies and for proposing the initial research concept that laid the foundation for this work. I owe particular thanks to **Z. Daneberga** for her openness, trust, and continuous mentorship, as well as for patiently guiding me through each stage of this research. Without your expertise and encouragement, the completion of both the analyses and this Thesis would not have been possible.

I would also like to acknowledge the Breast Surgery Unit of Pauls Stradiņš Clinical University Hospital for their valuable assistance with patient material collection and sample selection, which was crucial for the success of this study.

My sincere thanks go to the reviewer of this Thesis, **Dace Pjanova**, for her careful evaluation of the work and for her insightful comments and constructive suggestions, which significantly enhanced its quality. I am likewise grateful to all members of the Doctoral Thesis Discussion Committee for their time, assessment, and thoughtful recommendations.

I extend my appreciation to Rīga Stradiņš University, Institute of Oncology and Molecular Genetics, for their support, collaboration, and professional advice during my doctoral studies.

Finally, I am deeply thankful to my friends and family for their enduring support and belief in me. I am especially grateful to my husband for his patience, understanding, and constant encouragement throughout this demanding journey. I thank everyone who has supported me along the way, these contributions have played an important role in both my academic development and personal growth.