Functional Role of TET3 Enzyme in Breast Tumorigenesis

By:
Sondos M. Odeh
Supervisor:
Zaidoun Salah. PhD
Cancer

• Cancer is a complex disease.
• Characterized by rapid and uncontrolled cell proliferation within a tissue.
• Cancers are thought to be genetic diseases of somatic cells:
  - activate oncogenes
  - inactivate tumor suppressor genes
Breast Cancer

• Breast cancer has the highest morbidity and mortality rates of all cancers in women.

• Manifestation of different malignancies in the mammary glands (heterogeneous).
Epigenetics and Epigenetic Modifications

Balogh & Engelmann 2011
Methylation

- DNA methylation is one of the pivotal epigenetic mechanisms that controls cell proliferation, apoptosis, differentiation, cell cycle, and transformation in eukaryotes.

Alokail & Alenad, 2017
Methylation/Demethylation

- DNA methylation has been considered a stable, persistent and heritable mark. However, emerging research indicates that transcription factors and related proteins not only protect sequences from methylation but also initiate active DNA demethylation.

- DNA demethylation is defined as the process of removal of a methyl group from cytosines. This process can be passive or active.
Active/Passive DNA Demethylation

Ambrosi et al. 2017
TET Enzymes

• Ten-Eleven Translocation Enzymes

• A family of three enzymes that convert 5-methylcytosine (5mC) to 5-hydroxymethylcytosine (5hmC).

• These enzymes are differentially expressed in several tissues during development and can regulate several conserved signaling pathways, such as Wingless (WNT), Notch, Sonic Hedgehog (SHH) and Transforming Growth Factor Beta (TGF-β).
TET3

- Studies have shown that the role of TET3 is different from one cancer type to another.

- Predominantly expressed in oocytes and zygotes and neurons.
Study Aims

• Investigating the expression pattern and functional role of TET3 in breast cancer.

• Elucidating the effect of TET3 overexpression in breast cancer cell lines under different cellular conditions in order to understand the behavior of this enzyme in different cancer hallmarks.
Results
Production of TET3 Overexpressing Cells
Production of TET3 Overexpressing Cells
Production of TET3 Overexpressing Cells
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Target Gene (TET3) → Cut → Mix → Ligate → TET3
Expression Pattern of TET3 Enzyme in Breast Cancer Cell Lines

![Bar graph showing expression levels of TET3 in different breast cancer cell lines.](image-url)
Transfection and Infection

TET3

MDA MB 231 cells
Generation and Validation of TET3 Clones

* Indicates P value is <0.05
Effect of TET3 Overexpression on MDA MB231 Cell Phenotypes

- Different hallmarks of cancer were studied:
  - Cell proliferation
  - Motility and cell migration
  - Cell survival
TET3 Overexpressing Cells Result in an Increase in Cell Proliferation

* Indicates P value is <0.05
TET3 Overexpressing Cells Result in a Higher Rate of Cell Migration
TET3 Overexpressing Cells Result in an Increase in Cell Survival
Effect of TET3 Overexpression on other Genes like Oncogenes and TSGs

* Indicates P value is <0.05
TET3 Enzyme Expression in Breast Cancer Tissue Samples.
High TET3 Expression Predicts Poor Prognosis in Breast Cancer

- +ve lymph nodes & grade 3
  N=369

- Grade 3
  N=903

- TNBC & grade 3
  N=138

- TNBC
  N=255

- Protein level

N=1113
Conclusion

• Different expression patterns in different cell lines.

• Our results show that TET3 has an oncogenic behavior at least in specific breast cancer subtypes.

• There are other factors that need to be further studied that play a role in regulating the expression of other genes.
Future Studies

- Cloning the different isoforms of TET3 in order to have a wider and generalized vision of the function and behavior of TET3 in breast cancer.

- Study the function of this enzyme by knocking out the different isoforms of TET3 and its effect on different cellular phenotypes.
Thank You!