ASSESSMENT OF VITAMIN D RECEPTOR (VDR) GENE EXPRESSION IN GDM PATIENTS FROM NORTH KARNATAKA

Project Reference No.: *49S_MSC_0259*

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Keywords

RNA isolation, cDNA library, qRT PCR, VDR gene

Introduction

1. Gestational Diabetes Mellitus (GDM) is a metabolic disorder resulting in glucose intolerance detected during pregnancy. This condition most commonly occurs during the second or third trimester as a result of increased insulin resistance and inadequate pancreatic β-cell response.[1]
2. GDM has emerged as a significant health challenge worldwide, especially in developing countries such as India. In India, the prevalence varies between 3% and 35%[2].
3. Vitamin D plays a significant role in the regulation of insulin secretion and glucose metabolism. Vitamin D deficiency is commonly seen in pregnant women and is a risk factor for GDM.[3]
4. The human gene for VDR is located on the long arm of chromosome 12 and is designated as 12q13.11.The vitamin D effects are mediated by the Vitamin D Receptor (VDR), which is responsible for controlling the expression of genes. The VDR gene is implicated in metabolic pathways that affect insulin sensitivity and immune function.[4]
5. The altered expression of the VDR gene has also been found to influence insulin function, leading to glucose intolerance during pregnancy. It has been found that the expression of the VDR gene is associated with insulin resistance and dysfunction in β-cells.[5]

Significance:

Evaluation of VDR gene expression helps in understanding the molecular basis of GDM. It helps in understanding the relationship between vitamin D signalling and insulin regulation. Identification of abnormal expression of VDR can help in the early diagnosis of GDM. It can also help in developing therapeutic and preventive strategies for the disease.[6]

Relevance:

There is little information on the expression of VDR genes in patients with GDM in North Karnataka. [7]

Regional studies are necessary owing to genetic and environmental variations among different populations. This research may contribute to the development of 'personalized medicine' for mothers. It may help in the early management and prevention of complications related to GDM. In conclusion, the research is significant in reducing health risks for mothers and their babies during pregnancy.[8]

Aim: To study expression of the vitamin D receptor (VDR) gene in women with gestational diabetes mellitus.

Objectives:

1. Collecting blood samples from patients with GDM and normal individuals
2. Samples will be stored at 80°C
3. Extraction of RNA and conversion into cDNA
4. Quantification of VDR gene expression by qRT-PCR
5. Comparing the expression levels in patients and controls
6. Statistical analysis of the results
7. Investigating the relationship between VDR gene expression and GDM

Methodology:

Sample Collection

- Blood samples will be collected in K_2 EDTA tubes from GDM and non-GDM patients and will be stored at −80 °C.

RNA Isolation.

- Total RNA will be Isolated.
- Assess RNA quantity and quality using multimode plate reader and agarose gel electrophoresis.

cDNA Synthesis

- RNA will be converted to cDNA using reverse transcriptase with the Takara PrimeScript kit.

Gene Expression Analysis

- Perform quantitative real-time PCR (qRT-PCR) targeting VDR mRNA.

GENE	SEQUENCE
VDR Forward	5′-GAC TTTGACCGGAACGTGCC-3′
VDR Reverse	5′-CATCAT GCCGATGTCCACAC-3′
β actin Forward	5'-AAGATCATTGCTCCTCCTGAGC-3'

β actin Reverse 5’ -TCCTGCTTGCTGATCCACATC-3’

- Use housekeeping genes (Beta Actin) for normalization.
- Analyze relative expression using the ΔΔCt method.

Data Analysis

- Compare VDR expression levels between cases and controls.
- Correlate expression data with Vitamin D levels and clinical parameters.
- Use appropriate statistical tests (t-test, ANOVA, Spearman/Pearson correlation).

Result and Conclusion:

1. Ethical clearance for the study has been obtained.
2. Collection of samples is in progress.
3. Primers for the VDR gene and the housekeeping gene have been ordered.
4. Standardization of the experimental protocols (RNA isolation, cDNA synthesis, qPCR) is in progress.

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Project Outcomes

- Determination of VDR gene expression in GDM patients and controls
- Correlation between Vitamin D levels and gene expression
- Generation of experimental and statistical data

Learning's

- Hands-on experience in RNA isolation, cDNA synthesis, qRT-PCR
- Understanding of gene expression analysis (ΔΔCt method)
- Exposure to clinical data handling and analysis
- Improved research and teamwork skills

Industry Relevance

- Useful for diagnostic and biotech industries
- Supports development of molecular diagnostic kits
- Relevant to pharmaceutical sector

Future Scope:

The future scope of this project includes:

- Extension to larger population studies
- Development of biomarker-based diagnostics
- Scope for personalized treatment approaches