

# **THE EFFECTS OF VITAMIN D ON INSULIN RESISTANCE AND NERVE CONDUCTION VELOCITY IN MALE RATS WITH TYPE 2 DIABETES**

*Thesis*

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*&*

*My children Salma, Talia & Marwan*

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## *Abstract*

**Background:** There is an emerging evidence of the neuroprotective roles for vitamin D. However its role in the pathogenesis of type 2 diabetes (T2 DM) and its exact mechanism of action in neuroprotection are still uncertain. The present work was designed to examine the effect of vitamin D supplementation on insulin sensitivity and nerve conduction velocity with and without insulin treatment in diabetic model.

**Methods:** The study was conducted on 50 male adult rats. They were divided into five groups :**Group 1:Control group**, **Group 2:Diabetic group**, in which T2DM was induced; **Group 3:Diabetic-insulin treated group** in which diabetic rats were treated with insulin alone; **Group 4:Diabetic - vitamin D treated group** in which diabetic rats were treated with vitamin D alone and finally,**Group 5:Diabetic with combined insulin and vitamin D treatment group**.At the end of the experimental period, blood samples were taken from all animals of all groups for measurement of serum glucose & insulin, insulin resistance index (HOMA-IR),together with the oxidative stress marker : Malondialdehyde (MDA) and inflammatory markers: Interleukins 1 $\beta$  &10 (IL1 $\beta$  and IL10) . VDR gene expression in pancreatic (VDR-P) and sciatic nerve (VDR-N) tissues were estimated. In addition Nerve conduction velocity was performed on dissected sciatic nerve .

**Results:** Data showed a significant reduction of nerve conduction velocity of sciatic nerve together with increased insulin resistance in diabetic rats that paralleled increased MDA and IL1 $\beta$  and the decreased IL10. Administration of insulin, vitamin D alone or combined after induction of diabetes improved the previous changes. This improvement was accompanied with significant enhancement of VDR –P and VDR-N gene expression.

**Conclusion:** The improvement of insulin sensitivity and the neuroprotection induced by vitamin D supplementation in T2DM is related to restoration of VDR-P and VDR-N expression. Thus vitamin D could be a novel approach to lower neuropathic risk in diabetes.

### **Keywords:**

**Vitamin D receptor; 1,25-dihydroxyvitamin D3; Type2 diabetes mellitus, Insulin resistance, Nerve conduction velocity.**

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## *List Of Abbreviations*

|                                                |                                                                  |
|------------------------------------------------|------------------------------------------------------------------|
| 1 $\alpha$ ,25(OH) <sub>2</sub> D <sub>3</sub> | 1 $\alpha$ ,25(OH) <sub>2</sub> Di hydroxyvitamin D <sub>3</sub> |
| 2HPP                                           | Two hours post prandial                                          |
| AGEs                                           | Advanced glycemc end products                                    |
| AIR                                            | Acute insulin response                                           |
| ANOVA                                          | Analysis of variance                                             |
| ARE                                            | Antioxidant response element                                     |
| BGP                                            | Bone gamma carboxyglutamic acid protein                          |
| BMD                                            | Bone mineral density                                             |
| BMI                                            | Body mass index                                                  |
| BNC                                            | Bayonet Neill-concelman                                          |
| CAPs                                           | Compound action potentials                                       |
| CGRP                                           | Calcitonin gene related peptide                                  |
| CIDP                                           | Chronic inflammatory demyelinating polyneuropathy                |
| COX                                            | Cyclo-oxygenase                                                  |
| CVD                                            | Cardiovascular disease                                           |
| CY                                             | Cytochrome                                                       |
| dATP                                           | Deoxyadenosine triphosphate                                      |
| DBP                                            | Vitamin D binding protein                                        |
| DCCT                                           | Diabetic control and complication trial                          |
| dCTP                                           | Deoxycytidine triphosphate                                       |
| dGTP                                           | Deoxyguanosine triphosphate                                      |
| DI                                             | Disposition index                                                |
| DIN                                            | Deutsches institute fur normung                                  |
| DKA                                            | Diabetic ketoacidosis                                            |
| DM                                             | Diabetes Mellitus                                                |
| DN                                             | Diabetic neuropathy                                              |
| DNA                                            | Deoxyribonucleic acid                                            |
| dNTPS                                          | Deoxynucleotide triphosphate                                     |

|         |                                                     |
|---------|-----------------------------------------------------|
| DPP     | Diabetes prevention program                         |
| DRIs    | Dietary reference intake                            |
| Drp1    | Dynamin related protein                             |
| Drp1    | Dyanamin related protein                            |
| DSPN    | Distal symmetrical peripheral neuropathy            |
| dTTP    | Deoxythymidine triphosphate                         |
| EAE     | Experimental autoimmune encephalomyelitis           |
| EAR     | Estimated average requirement                       |
| ELISA   | Enzyme linked immunosorbent assay                   |
| EMG     | Electromyography                                    |
| FBG     | Fasting blood glucose                               |
| FGF-23  | Fibroblast growth factor twenty three               |
| FOXO3   | Forkhead protein                                    |
| FPG     | Fasting plasma glucose                              |
| GDNF    | Glial cell derived neurotrophic factor              |
| GENNID  | Genetics of non-insulin dependent diabetes mellitus |
| GIP     | Gastric inhibitory polypeptide                      |
| GLP-1   | Glucagon like peptide one                           |
| GTC     | Guanidine thiocyanate                               |
| HAART   | High active antiretroviral treatment                |
| HbA1C   | Glycated haemoglobin                                |
| HFD     | High-fat diet                                       |
| HO-1    | Hemoxygenase one                                    |
| HOMA-IR | Homeostasis model assessment of insulin resistance  |
| HXXO    | Hypxanthine/xanthine oxidase system                 |
| IBD     | Inflammatory bowel disease                          |
| IENF    | Intra-epidermal nerve fiber                         |
| IFG     | Impaired fasting glucose                            |
| IGI     | Insulinogenic index                                 |
| IGT     | Impaired glucose tolerance                          |

|       |                                             |
|-------|---------------------------------------------|
| IL    | Interleukin                                 |
| iNOS  | Inducible nitric oxide synthase             |
| INS   | Insulin                                     |
| IOM   | Institute of medicine                       |
| IRR   | Insulin real risk                           |
| ISSI  | Insulin secretion sensitivity index         |
| LPO   | Lipid peroxidation                          |
| MAPK  | Mitogen activated protein kinase            |
| MDA   | Melondialdehyde                             |
| MMLV  | Molony murine leukemia virus                |
| NAD   | Nicotinamide adenine dinucleotide           |
| NADPH | Nicotinamide adenine dinucleotide phosphate |
| NCS   | Nerve conduction studies                    |
| NCS   | Nerve conduction studies                    |
| NCV   | Nerve conduction velocity                   |
| NGF   | Nerve growth factor                         |
| NPH   | Neutral protamine Hagedorn                  |
| NQO1  | Quinoneoxide reductase 1                    |
| Nr    | Nuclear factor                              |
| Nrf   | Nuclear factor                              |
| NT3   | Neutrotrophin three                         |
| OGTT  | Oral glucose tolerance test                 |
| P     | Probability                                 |
| PAD   | Peripheral arterial disease                 |
| PAD   | Peripheral arterial disease                 |
| PARP  | Poly(ADP-ribose)polymerase pathway          |
| PARP  | Poly(ADP-ribose) polymerase activity        |
| PCR   | Polymerase chain reaction                   |
| PKC   | Protein kinase c                            |
| PL    | Parometer logistic                          |

|      |                                             |
|------|---------------------------------------------|
| PPAR | Peroxisome proliferation activated receptor |
| PTH  | Parathyroid hormone                         |
| QST  | Quantitative sensory testing                |
| RCT  | Randomized controlled trials                |
| RDA  | Recommended dietary allowance               |
| RNS  | Reactive nitrogen species                   |
| ROS  | Reactive oxygen species                     |
| rpm  | Round per minute                            |
| RR   | Rapid response                              |
| RXR  | Retinoid X receptors                        |
| SC   | Subcutaneous                                |
| SHR  | Spontaneous hypertensive rats               |
| SPF  | Sun protection factor                       |
| SPPL | Osteoponin                                  |
| STZ  | Streptozotocin                              |
| SWMT | Semmes-Weinstein monofilament test          |
| T1DM | Type one diabetes mellitus                  |
| T2DM | Type two diabetes mellitus                  |
| Taq  | Thermos aquaticus                           |
| TBA  | Thiobarbituric acid                         |
| TCA  | Trichloracetic acid                         |
| TGF  | Transforming growth factor                  |
| TNF  | Tumour necrosis factor                      |
| TrKA | Tropomyosin related kinase receptor         |
| TZD  | Thiazolidinediones                          |
| UPS  | Ubiquitin- proteasome system                |
| UVB  | Ultraviolet B                               |
| VDDS | Vitamin D deficiency syndrome               |
| VDR  | vitamin D receptor                          |
| VDRE | Vitamin D response element                  |

|              |                                                     |
|--------------|-----------------------------------------------------|
| <b>VDR-N</b> | <b>Vitamin D receptor gene in nerve tissue</b>      |
| <b>VDR-P</b> | <b>Vitamin D receptor gene in pancreatic tissue</b> |
| <b>VPT</b>   | <b>Vibration perception threshold</b>               |
| <b>WC</b>    | <b>Waist circumference</b>                          |
| <b>WHO</b>   | <b>World Health Organisation</b>                    |

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*In Review of literature and Materials & methods*

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