Association of Low-Density Lipoprotein Receptor-Related Protein-5 4037C>T Variant with Nephropathy in Adolescents with Type 1 Diabetes Mellitus

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Abstract

Background: In people who have uncontrolled T1DM, microvascular problems such diabetic nephropathy (DN) occur. The co-receptor ligands of the Wnt signalling pathway is lower density lipoprotein receptors-related protein 5 (LRP5). The human LRP5 genes have several single nucleotide variations that have been linked to metabolic diseases. It has been suggested that the Wnt/-catenin pathway is crucial in a number of metabolic diseases, particularly diabetic nephropathy. Unfortunately, there are few research investigating the function of the LRP5 genes variation in T1DM.

Purpose: The progression of diabetes sequelae, including diabetic nephropathy and dyslipidemia, in addition to the relationship between the LRP5 variation 4037C>T and indicators of glycemic and metabolic control were examined in this cross-sectional case-control research of adolescents with T1DM.

Methods: Twenty age- and sex-matched control subjects and 40 juvenile T1DM participants participated in this study. All trial subjects underwent thorough history-taking, clinical evaluations, and lab tests to determine their glucose control and risk of diabetic microvascular complications. Employing real-time PCR, the LRP5 gene (rs3736228) variation assay was carried out.

Results: The current investigation found no statistically significant change in the LRP5 rs3736228 gene variant between identical control subjects and adolescents with T1DM. A lack of a statistically relevant correlation between the LRP5 gene variation and the onset of diabetic nephropathy was found. Furthermore, there was a statistically significant correlation between obesity and hypercholesterolemia in T1DM individuals with the CT genotypes. Teenagers with DN had HbA1c levels that were considerably greater than those without DN (p=0.018).

Conclusion: According to the research, there is no connection between the LRP5 4037C>T variation and diabetic nephropathy in hospitalized children. Furthermore, there is a statistically significant correlation between the genotype CT and elevated

BMI and hypercholesterolemia. Additional research is required to examine numerous gene variations associated with glycemic management and the emergence of diabetic complications.

Keywords: Low density lipoprotein receptor-related protein 5, type 1 diabetes, diabetes vascular complications, diabetic nephropathy

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Introduction

In youngsters and adolescents, type 1 diabetes mellitus (T1DM) is an autoimmune disease endocrine condition defined by the loss of pancreatic beta cells, an inability to produce enough insulin, and the emergence of hyperglycemia. The prevalence of it is rising around the world. **(El-Ziny, M. A et al.,2014)** As a result of its consequences' higher rates of morbidity and mortality, it is a serious health issue. The most major reason of end-stage kidney problems and a microvascular consequence of T1DM is diabetic nephropathy (DN). Earlier disease and severity detection is crucial as a result. **(Śnit, M. et al., 2017; Pan W. et al., 2020)**

A vital co-receptor of the Wnt/β-catenin signalling pathway, lower-density lipoprotein receptors-related protein 5 (LRP5) is a component of the family of lower-density lipoprotein transmitters. **(Kavanagh, D. H. et al., 2011)** The functioning of islet cells, pancreatic growth, and insulin release are all regulated by the LRP5-mediated Wnt signalling pathway. Different levels of the Wnt and insulin signalling pathways show cross-talk, and changing LRP5/Wnt activities can change how insulin acts and cause insulin resistance. **(Wang, J. et al., 2015)** It works in concert with the Frizzled family of receptors (FZD), which are in charge of activating the β-catenin pathways. **(Xiao, H. e l. 2019)**

When two or more different phenotypes exist in the same community, it is said to have a variance, which is a specific genetic difference. Any physical, behavioural, or physiological feature may exhibit this. **(Steinhart, Z. and Angers, S., 2018)** SNPs are changes to the DNA's construction blocks of guanines (G), cytosine (C), adenines (A), or thymine (T), which result in a single genetic coding change. **(Kochetova, O. V. et al., 2019)** Chromosome 11q13 is the location

of the LRP5 gene. The LRP5 genome has several SNPs that have been found to be linked to metabolic diseases such obese, hypercholesterolemia, and type 2 diabetes. **(You, H. F. et al., 2015)** In addition, the LRP5 has been implicated in a number of kidneys conditions, particularly diabetic nephropathy. **(Zhang, L. et al., 2017)** LRP5 genes expression control is still mainly unexplored. Furthermore, some research found and cloned the LRP5 gene's promoter region. They showed that the promoter region, which surrounds the transcriptional start point and is situated 40 nucleotides upstream of the 5' end of the DNA sequence, is a region rich in GC. Additionally, investigations revealed that the activation of the individual LRP5 promoters required both specificity protein 1 (SP-1) and Krüppel-like factor 15 motifs (KLF-15). **(Li, J. et al., 2010)**

Several different tissues and cells, including bones tissues, endothelium cells, stem cells, breast tissues, pancreatic beta cells, liver cells, **(You, H. F. et al., 2015)**, and renal, release the LRP5 peptide. **(13)** Several physiological functions, such as skeletal growth, ocular growth, lipids metabolism, and glucoses-induced insulin release, depend on the Wnt/β-catenin pathways being activated by LRP5. **(He, X. et al., 2020)** The LRP5 peptide is highly expressed in the liver, where it functions as receptors in the removal of chylomicrons and lipoproteins that comprise apolipoprotein E (Apo-E). **(Gao, H. et al., 2015, Mine, K. et al., 2017)** Furthermore, via inhibiting adipogenic transcription factors, the LRP5 and Wnt signalling pathway had been linked to the control of adipogenesis **(Montazeri-Najafabady, N. et al., 2017)**

Numerous LRP5 encoding SNPs, including rs3736228 in exons 18, rs41494349 in exons 2, and rs2306862 in exons 10, have been discovered, **(Kitjaroentham, A. et al., 2018)** rs566442 in exon 15, **(Ashouri, E. et al., 2014)** and rs4988321 in exon 9. **(Bernardes, M. et al., 2018)** Among the LRP5 operational variations that has been researched the most commonly is our LRP5 SNP of concern (rs3736228). **(Kitjaroentham, A. et al., 2018)** The LDL-repeated domains of the LRP5 proteins, which are involved for regulating the association between the receptors and the ligands, contains the rs3736228 in exon 18. This alternative causes the loss of functionality of the LRP5 proteins by changing the allele C to T, resulting in a missense mutations of LRP5 with an amino acid change from alanine to valines. **(Liu, K. et al., 2017)** Numerous researches have found links between the LRP5 gene's SNPs and a variety of clinical illnesses, including osteoporosis, pseudoglioma syndrome, familial exudative vitreoretinopathy, dyslipidemia, obesity, and renal ailments. **(Norwitz, N.G. et al., 2017) (Gilmour, D.F., 2015) (Gao, H. et al., 2015) (You, H. F. et al., 2015) (Cnossen, W. R. et al, 2016) (Ren, Q. et al., 2021)**

Aim of the study

Investigating the LRP5 variation 4037C>T in children and adolescents with T1DM was the purpose of this investigation, and its association with the development microvascular complications such as DN and its relation with clinical data and markers of glycemic and metabolic control.

Ethical considerations

The Faculty of Medicine at Ain Shams University's Research Ethical Committee gave its approval to this work under reference number FWA00017585 FMASU MS 499/2019. The Declaration of Helsinki was examined, and this research was given the go light (as revised in Brazil, 2013).

- - Before each individual agreed to participate in this research, their informed consent was obtained in writing.
- - Prior to patient recruitment, the report's objectives and procedures were described to the participants' carers.
- - The participants were free to leave the research at any moment.
- Participants have the option of keeping research findings, which is kept in confidence.

Conflict of interests

There are no disclosed conflicts of interest for the writers. The manuscript's elements have been reviewed and approved by each co-author, and there are no competing financial interests to disclose. We attest that the contribution is unique and is not currently being considered by another publisher.

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Study design

Forty juvenile T1DM patients, aged 8 to 18, participated in this case-control research. The International Society for Pediatric and Adolescent Diabetes (ISPAD) guidelines for diabetes diagnosis in youngsters, 2018, were used to make the T1DM diagnoses. Adolescents and children with T1DM for further than five years were recruited for the research (Group I). The patients' group was subsequently separated into subgroups Ia and Ib, each of which contained 20 patients. DN was identified in subgroup Ib using criteria set forth by the American Diabetes Association.

(American Diabetes Association, 2021). Twenty controls people who were matched for age and sex were chosen as the control group (Group II). The present research was carried out between January 2020 and January 2021. The Pediatrics and Adolescent Diabetic Clinic at Ain Shams University Hospital was used to enroll T1DM participants. Ain Shams University Hospital's paediatric outpatient clinics served as the source of the healthy participants.

Sample size

Using the EPI INFO sample size formula, the predicted sample size was calculated with an alpha error of 0.05, a confidence interval of 0.95, and a power of the research of 0.80. A minimum of 40 participants are required to explore the LRP5 mutation 4037C>T in children and adolescents with T1DM, which may help to partially understand the cause of the condition and its problems.

The relevant tests were administered to all respondents:

I. I. Obtaining a complete medical history, paying particular attention to demographic information, family history of diabetes, the number of years of diabetes (in years), the forms of insulin treatment used, the dosages of that treatment, how often blood glucose levels are checked, the historical background of DKA, and the existence of diabetes - related complications.

II. II. A thorough clinical evaluation with an emphasis on Vital signs such as blood pressure and anthropometric such as weight in kilogrammes (Kg) and height (cm) in millimetres were recorded. Based on WHO growth charts for weights, heights, and BMI Z ratings, body mass index was computed as Kg/m2 and represented on the age and sex standards percentiles. A neurological examination was conducted to look for any indications of neuropathy.

III. III. A skilled single ophthalmologist used direct ophthalmoscopy to do a fundus evaluation for the purpose of assessing retinopathy.

IV. IV. The diabetic peripheral neuropathy testing instrument known as the simple fast bedside neuropathic disability scoring (NDS) was accepted. The NDS was created by looking at vibrating perceptions (using a 128-Hz tuning fork), greater toe temperature and pin-prick perceptions, and the existence or lack of ankle reflexes. A rating of two or above on the sensory modalities was considered clinical diabetic peripheral neuropathy.

Sample collection:

Ten millilitres of venous blood were drawn from each patient by venipunctures while using strict aseptic techniques, and the blood was split into three tubes as indicated:

a simple, sterile tubes with gel for serum separation that can be used to measure lipid profiles, creatinine values, and fasting blood glucose. Two "K2-EDTA" vacutainers di-potassium ethylene diamines tetra acetic for the LRP5 mutant PCR assays and HbA1c concentration measurement.

1. 1. Glycemic and metabolic controls were assessed :

a- a- Applying reagents provided by the manufacturer, the HbA1c% was measured using an immunoturbidimetric technique on a Cobas 6000 autoanalyzers (Roche Diagnostics, 9115 Hague Road Indianapolis, IN46250). Two values from the past six months were used to establish the average HbA1c% value.

b- b- Utilizing Cobas Integra 800 (Roche Diagnostics GmbH, 68298 Mannheim, Germany) and reagents provided by the company, fasting serum triglycerides, total serum cholesterol, high density lipoprotein cholesterol (HDL-C), and low density lipoprotein cholesterol (LDL-C) were measured by absorbance photometry.

2. 2. Evaluation of the use of earlier diabetic nephropathy:

A. A. Roche Diagnostics GmbH, Indianapolis, IN 46250, USA, used chemicals provided by the firm to perform a quantitative immuno-turbidimetric analysis to measure the amount of microalbuminuria in a urine sample. Normally (30 mg/g creatinine), microalbuminuria (30-300 mg/g creatinine) are the standard ranges for the microalbumin/creatinine ratios. Patients with microalbuminuria were instructed to complete two more urine samples every three to six months. When two out of three specimens displayed an outflow rate of 30 to 300 mg/mg creatinine, persistent microalbuminuria was considered to exist.

Also, estimated glomerular filtration rate (eGFR) was calculated by Schwartz equation.

3. Determination of LRP5 gene variant using Real Time polymerase chain reaction (RT-PCR), detection of LRP5 variant (rs3736228) was done using TaqMan real time PCR kit supplied by Thermo scientific (Thermo scientific: 168 Third Avenue Waltham, MA USA 02451).

Statistical Analysis

Statistical analysis was done through Statistical Package for Social Science (IBM SPSS) for data analysis. Quantitative data were expressed as mean and standard deviation (SD) and qualitative data were expressed as number (n) and percentage (%). Student's t-test was applied in case of statistical comparison between quantitative parametric data of two independent groups. Chi-square test was done to compare categorical and qualitative data. Mann–Whitney U test was used to compare quantitative nonparametric data from two independent groups.

Results

The current study included 40 children and adolescents with T1DM; 21 females and 19 males with mean age 14.25 ± 2.90 years and mean diabetes duration 8.07 ± 1.97 years. They were compared with 20 age and sex matched healthy control group with mean age 13.70 ± 3.23 years. None of our patients had pubertal delay using Tanner staging for pubertal assessment. The results are represented in the following tables;

Table (1): Descriptive and Comparative Statistics of Demographic Data between Patients and Controls Using Student's t-Test and Chi-Square Test

Parameter		Group I (patients) n= 40	Group II (controls) n= 20	t / χ 2*	p-value
Age (years)	$\bar{x}$ ± SD	14.25 ± 2.90	13.70 ± 3.23	0.291	0.772
Gender	Female	21 (52.5%)	8 (40.0%)	0.834*	0.361
	Male	19 (47.5%)	12 (60.0%)		
Weight (kgs)	$\bar{x}$ ± SD	51.96 ± 14.08	47.95 ± 14.47	1.031	0.307
Height (cm)	$\bar{x}$ ± SD	152.93 ± 14.06	154.20 ± 14.75	0.326	0.746
BMI (percentiles)	$\bar{x}$ ± SD	71.13 ± 18.83	47.5 ± 16.05	4.804	0.014
BMI (percentiles)	Normal ($5^{th} – 85^{th}$)	28 (70.0%)	20 (100.0%)	7.500*	0.024
	Overweight	10 (25.0%)	0 (0.0%)		

	(≥85th)				
	Obese (≥95th)	2 (5.0%)	0 (0.0%)		

BMI: body mass index. p-value > 0.05: non-significant; p-value < 0.05: significant.

No statistically significant difference was found between patients and controls groups as regards age, gender, weight and height. A statistically significant difference was found between patients' group and controls in the different percentiles of BMI (p-value <0.05), where 25% of patients were overweight and 5% were obese compared to normal BMI in controls as shown in table (1).

Table (2): Descriptive and comparative statistical Analysis of lipid profile data between Patients and Controls Using Student's t-Test, Chi-Square Test and Mann-Whitney U Test

	Group I (patients)	**Group II (controls)**	**t / χ 2*/ Z≠**	**p-value**
	n= 40	**n=20**		
Hypercholesterolemia (≥200 mg/dL)	12 (30.0%)	0 (0.0%)	7.500*	0.006
Hypertriglyceridemia	15 (37.5%)	4 (20.0%)	1.887*	0.170
High LDL-C (≥130 mg/dL)	10 (25.0%)	0 (0.0%)	6.000*	0.014
Low HDL-C (<40 mg/dL)	10 (25.0%)	9 (45.0%)	2.465*	0.116

HbA1c: glycated hemoglobin. LDL-C: low-density lipoprotein cholesterol. HDL-C: high-density lipoprotein cholesterol. p-value > 0.05: non significant and p-value < 0.01: highly significant.

A statistically significant difference was found between both groups as regards presence of hypercholesterolemia and levels of LDL-C (p = 0.006, P = 0.014, respectively) as shown in table (2).

Table (3): Descriptive and Comparative Statistics of the Genotypes and Alleles Frequencies of LRP5 Gene Variant between Patients and Controls

		Group I (patients)		Group II (controls)		χ^2	p-value
		n= 40	%	n= 20	%		
Genotypes	**CC**	32	80.0%	14	70.0%	0.745	0.388
	CT	8	20.0%	6	30.0%		
Allele	**C**	72	90.0%	34	85.0%	0.647	0.421
	T	8	10.0%	6	15.0%		

P-value >0.05: non-significant. C: cytosine, T: thymine.

There was no statistically significant difference regarding the LRP5 gene variant rs3736228 between patients with T1DM and controls (p-value >0.05) as shown in table (3).

Table (4): Descriptive and comparative statistical analysis of the genotypes and alleles frequencies of LRP5 gene variant between patients' Subgroup Ia (without diabetic nephropathy) and patients' subgroup Ib (with diabetic nephropathy)

		Subgroup Ia		Subgroup Ib		χ^2	p-value
		n= 20	%	n= 20	%		
Genotypes	**CC**	17	70.0%	15	75.0%	0.625	0.429
	CT	3	30.0%	5	25.0%		
Allele	**C**	37	92.5%	35	87.5.%	0.556	0.456
	T	3	7.5%	5	12.5%		

P-value >0.05: non-significant

Children and adolescents with T1DM were further divided into 2 subgroups according to the presence of DN; subgroup Ia (without DN) and subgroup Ib (with DN) . Four patients in group Ib had elevated blood pressure. None of our patients had diabetic retinopathy by fundus examination nor manifestations of clinical neuropathy by NDS score. HbA1c was significantly higher in group Ib than group Ia (p= 0.018). However, there was no statistically significant difference regarding the LRP5 gene variant rs3736228 between the 2 subgroups (p-value >0.05) as shown in table (4).

Table (5): Comparative statistics between genotypes of LRP5 gene variant (rs3736228) in relation to clinical and laboratory data of T1DM patients

		CC	CT	$t/X^{2*}/Z^{\neq}$	p-value
		n= 32	n= 8		
Presence of hypertension		3 (9.4%)	1 (12.5%)	0.069*	0.792
BMI (percentiles)	Normal ($5^{th} – 85^{th}$)	24 (75.0%)	4 (50.0%)	8.571*	0.014
	Overweight ($\geq 85^{th}$)	8 (25.0%)	2 (25.0%)		
	Obese ($\geq 95^{th}$)	0 (0.0%)	2 (25.0%)		
FBG (mg/dL)	$\bar{x}$ **± SD**	113.60 ± 26.44	132.38 ± 23.22	1.835	0.074
	<100	5(15.6%)	1(12.5%)	0.049*	0.825
	>100	27(84.4%)	7(87.5%)		
HbA1c (%)	$\bar{x}$ **± SD**	9.97 ± 1.62	10.79 ± 1.77	1.261	0.215
Hypercholesterolemia (≥200 mg/dL)		6(18.8%)	6(75.0%)	9.643*	0.002
Hypertriglyceridemia		7(35%)	8(40.0%)	0.107*	0.744
High LDL-C (≥130 mg/dL)		3(15.0%)	7(35.0%)	2.133*	0.144
Low HDL-C (<40 mg/dL)		7(21.9%)	3(37.5%)	0.833*	0.361
Creatinine (mg/dL)	$\bar{x}$ **± SD**	0.069 ± 0.17	0.71 ± 0.11	0.0338	0.737
eGFR (mL/min/1.73m^2)	$\bar{x}$ **± SD**	97.72 ± 24.46	87.13 ± 14.10	1.170	0.396
Microalbumin/creatinine ratio (mg/g)	**Median (IQR)**	25.6 (9.65 – 43)	52.5 (10 – 87)	1.286*	0.063
	<30	17(53.1%)	3(37.5%)	$0.625^{\neq}$	0.429
	3-300	15(46.9%)	5(62.5%)		

BMI: body mass index FBG: fasting blood glucose, HbA1c: glycated hemoglobin, LDL-C: low-density lipoprotein, HDL-C: high-density lipoprotein, eGFR: estimated glomerular filtration rate. P-value >0.05: non-significant

Studying the relation between LRP5 genotypes (CC and CT) and clinical and laboratory data of T1DM patients revealed a statistically significant association between CT genotype and obesity (p= 0.014) as well as hypercholesterolemia (p= 0.002) as shown in table (5).

Discussion

T1DM is a major health problem as patients are at high risk of developing complications especially microvascular complications. DN is a common microvascular complication, occurring in approximately 40% of patients with diabetes. **(Mohammedi, K. et al., 2016) (Pan W. et al., 2020)** T1DM is a heterogeneous and multifactorial disease. However, genetic predisposition has been considered as an important risk factor modulating susceptibility to T1DM. **(Pang, H et al., 2021)** Moreover, its clinical course is characterized by a long silent period before appearance of any symptoms or signs. **(Battaglia, M. et al., 2017)**

LRP5 functions as a key co-receptor in the Wnt/β-catenin signaling pathway. It is highly expressed on pancreatic β-cells and is involved in the regulation of β-cell functions as well as regulation of cellular and whole-body metabolism. It is one of the systems that interacts with and modulates the insulin signaling network and is involved in glucose homeostasis. (**Kim S. P. et al., 2017) (Montazeri-Nafafabady, N. et al., 2019)** Moreover, experiments on animal models indicated that LRP5 is essential for normal glucose metabolism and LRP5-deficient mice showed markedly impaired glucose tolerance when fed with normal diet. **(Fujino, T. et al., 2003) (Souza, K. et al., 2018)**

The current work showed absence of a statistically significant difference as regards the LRP5 gene variant rs3736228 between T1DM patients and healthy controls. Souza et al. reported a statistically significant association between LRP5 rs3736228 variant and increased risk of T1DM in a group of 134 Brazilian patients with T1DM. **(Souza, K. et al., 2018)** This difference may be due to that genetic risk factors to diabetes may differ between different ethnic groups.

Our results found a non-significant statistical difference as regards LRP5 rs3736228 gene variant between patients with DN and patients without DN. This finding is in accordance with one study by Kavanagh et al. who investigated LRP5 (rs3736228) variant in a group of 1351 British and Irish patients with childhood onset T1DM (651 patient with DN and 700 patients without DN), they found no strong association of this SNP with diabetic nephropathy in the studied groups. **(Kavanagh, D. H. et al., 2011)**

In the present work, the relation between the observed genotypes (CC and CT) of LRP5 (rs3736228) gene variant, the demographic, clinical and laboratory characteristics were studied in patients with T1DM, results revealed a statistically significant association of the genotype CT with obesity and hypercholesterolemia. These findings are similar to the study performed by Jiang et al., who reported significant association between rs3736228 variant and increased total cholesterol

levels as well as reduced bone mineral density in an adult Chinese population. **(Jiang, X.Y. et al., 2010)** Moreover, Kochetova et al. who performed a study on 486 patients with type 2 diabetes and 444 persons with no evidence of diabetes, results detected a statistically significant association between CT and TT genotypes of LRP5 (rs3736228) variant and the risk of obesity in T2DM patients. **(Kochetova, O. V. et al., 2019)**

Conclusion

The present study revealed the absence of a significant association between LRP5 rs3736228 gene variant and T1DM as well as the development of diabetic nephropathy. However, the study showed that the CT genotype was significantly associated with obesity and hypercholesterolemia in young patients with T1DM. The presented association confirms the role this gene in the development of obesity and dyslipidemia but not with T1DM. Further gene variant analysis is essential for understanding the molecular mechanism of T1DM and its complications.

Recommendations

SNP in the LRP5 gene may act as a potential candidate of biomarker for diabetic nephropathy screening, diagnosis, and future treatment. To certain the current results, updated well-designed researches with larger sample size, in diverse ethnic populations particularly, are required in the future.

Limitations of the study

Limitations of our study must be addressed, firstly; sample size is relatively small and we recommend for researchers to do future studies with larger sample size. Secondary; many SNPs have been detected in the LRP5 gene and were reported to be associated with several metabolic disorders, more SNPs should be investigated on a wide scale in order to detect the SNP that mostly affect the disease course and the development complications in T1DM.

References

American Diabetes Association, 2021. Classification and diagnosis of diabetes: standards of medical care in diabetes-2021. Diabetes care. 44 (Suppl. 1): S15-S33.

Ashouri, E.; Meimandi, E. M.; Saki, F. et al., 2014. The impact of LRP5 variant (rs556442) on calcium homeostasis, bone mineral density, and body composition in Iranian children. Journal of Bone and Mineral Metabolism 33(6): 651–657.

Battaglia, M.; Anderson, M. S.; Buckner, J. H. et al., 2017. Understanding and preventing type 1 diabetes through the unique working model of Trial Net. Diabetologia. 60(11): 2139–2147.

Bernardes, M.; Durães, C.; Oliveira, A. et al., 2018. LRP5 gene variants and radiographic joint damage in rheumatoid arthritis patients. Osteoporosis International. 29: 2355-2368.

Cnossen, W. R.; te Morsche, R. H.; Hoischen, A. et al., 2016. LRP5 variants may contribute to ADPKD. European Journal of Human Genetics. 24(2): 237–242.

El-Ziny, M. A.; Salem, N. A.; El-Hawary, A. K et al., 2014. Epidemiology of childhood type 1 diabetes mellitus in Nile Delta, northern Egypt - a retrospective study. Journal of clinical research in pediatric endocrinology. 6(1): 9–15.

Fujino, T.; Asaba, H.; Kang, M. J. et al., 2003. Low-density lipoprotein receptor-related protein 5 (LRP5) is essential for normal cholesterol metabolism and glucose-induced insulin secretion. Proceedings of the National Academy of Sciences of the United States of America. 100(1): 229–234.

Gao, H.; Tian, Y.; Meng, H. et al., 2015. Associations of apolipoprotein E and low-density lipoprotein receptor-related protein 5 variants with dyslipidemia and generalized aggressive periodontitis in a Chinese population. Journal of Periodontal Research. 50(4): 509–518.

Gilmour, D.F., 2015. Familial exudative vitreoretinopathy and related retinopathies. Eye. 29: 1–14.

He, X.; Cheng, R.; Huang, C. et al., 2020. A novel role of LRP5 in tubulointerstitial fibrosis through activating TGF-β/Smad signaling. Signal Transduction and Targeted Therapy. 5(1): 45.

Jiang, X.Y.; Chen, Y.; Xu, L. et al., 2010. Association of LPR5 variant with bone mass density and cholesterol level in population of Chinese Han. Experimental and Clinical Endocrinology and Diabetes. 118(6): 388–391.

Kavanagh, D. H.; Savage, D. A.; Patterson, C. C. et al., 2011. Association Analysis of Canonical Wnt Signaling Genes in Diabetic Nephropathy. PLoS ONE.; 6(8): e23904.

Kim, S. P.; Frey, J. L.; Li, Z. et al., 2017. Lack of Lrp5 Signaling in Osteoblasts Sensitizes Male Mice to Diet-Induced Disturbances in Glucose Metabolism. Endocrinology. 158(11): 3805–3816.

Kitjaroentham, A., Hananantachai, H., Phonrat, B., 2016. Low density lipoprotein receptor-related protein 5 gene variants and osteoporosis in Thai menopausal women. Journal of Negative Results in BioMedicine. 15(1): 16.

Kochetova, O. V.; Avzaletdinova, D. S.; Sharipova, L. F. et al., 2019. An analysis of the associations of polymorphic variants of the LEPR (rs1137100), LRP5 (rs3736228), and LPL (rs320) genes with the risk of developing type 2 diabetes mellitus. Russian Journal of Genetics. 55(4): 495–503.

Li, J.; Yang, Y.; Jiang, B. et al., 2010. Sp1 and KLF15 regulate basal transcription of the human LRP5 gene. BMC Genetics. 11(1): 12.

Liu, K.; Tan, L.J.; Wang, P. et al., 2017. Functional relevance for associations between osteoporosis and genetic variants. PLoS ONE. 12(4): e0174808.

Ma, Y., 2016. LRP5: a novel anti-inflammatory macrophage marker that positively regulates migration and phagocytosis. Journal of Molecular and Cellular Cardiology. 91: 61–62.

Mine, K.; Hirakawa, K.; Kondo, S. et al., 2017. Subtyping of type 1 diabetes as classified by anti-GAD antibody, IgE Levels, and tyrosine kinase 2 (TYK2) promoter variant in the japanese. EBioMedicine. 23: 46–51.

Mohammedi, K.; Patente, T. A.; Bellili-Muñoz, N. et al., 2016. Glutathione peroxidase-1 gene (GPX1) variants, oxidative stress and risk of kidney complications in people with type 1 diabetes. Metabolism. 65(2): 12–19.

Montazeri-Nafafabady, N.; Dabbaghmanesh, M.H.; Mohamadian Amiri, R. et al., 2019. Influence of LRP5 (rs556442) variant on insulin resistance in healthy Iranian children and adolescents. Turkish Journal of Medical Sciences. 49(2): 490-496.

Montazeri-Najafabady, N.; Dabbaghmanesh, MH.; Omrani, GR. et al., 2017. Variant in LRP5 (rs556442) is associated with higher TG levels in Iranian children. Annals of Human Biology. 44 (4): 373-378.

Norwitz, N.G.; Mota, AS.; Misra, M. et al., 2019. LRP5, bone density, and mechanical stress: a case report and literature review. Frontiers in Endocrinology. 10:184.

Pang, H.; Xia, Y.; Luo, S. et al., 2021. Emerging roles of rare and low-frequency genetic variants in type 1 diabetes mellitus. Journal of Medical Genetics. 58: 289-296.

Pan, W.; Zheng, X.; Chen, G. et al., 2020. Nanotechnology's application in type 1 diabetes. WIREs Nanomedicine and Nanobiotechnology. e1645.

Ren, Q.; Chen, J. and Liu, Y., 2021. LRP5 and LRP6 in Wnt signaling: similarity and divergence. Frontiers in cell and developmental biology. 9: 670960.

Śnit, M.; Nabrdalik, K.; Długaszek, M. et al., 2017. Association of rs 3807337 variant of CALD1 gene with diabetic nephropathy occurrence in type 1 diabetes - preliminary results of a family-based study. Endokrynologia Polska. 68(1): 13–17.

Souza, K.; Ururahy, M.; Oliveira, Y. et al., 2018. The low-density lipoprotein receptor-related protein 5 (LRP5) 4037C>T variant: candidate for susceptibility to type 1 diabetes mellitus. Archives of endocrinology and metabolism. 62(4): 480–484.

Steinhart, Z. and Angers, 2018. S. Wnt signaling in development and tissue homeostasis. Development. 145: dev146589

Wang, J.; Zhao, J.; Zhang, J. et al., 2015. Association of Canonical Wnt/β-Catenin Pathway and Type 2 Diabetes: Genetic Epidemiological Study in Han Chinese. Nutrients. 7(6): 4763–4777.

Xiao, H.; Tong, Y; Zhu, Y. et al., 2019. Familial exudative vitreoretinopathy-related disease-causing genes and norrin/β-catenin signal pathway: structure, function, and mutation spectrums. Journal of Ophthalmology. 5782536.

Xu, GY.; Qiu, Y. and Mao, HJ., 2014. Common variant in the LRP5 gene may increase the risk of bone fracture and osteoporosis. Biomed Research International. 290531.

You, H. F.; Zhao, J. Z.; Zhai, Y. J. et al., 2015. Association between Low-density Lipoprotein Receptor-related Protein 5 Variants and Type 2 Diabetes Mellitus in Han Chinese: a Case-control Study. Biomedical and Environmental Sciences. 28(7): 510–517.

Zhang, L.; Wang, J.; Zhang, M. et al., 2017. Type 2 diabetes mellitus associated with the interaction of LRP5 variant and overweight and obesity in rural Chinese adults T2DM relate to interaction of LRP5 and obesity. Journal of Diabetes. 9 (11).