ADPKD protects against diabetogenic effects associated with genetically-predicted lactase persistence.

Keywords
obesity, insulin resistance, genetic polymorphism, ADPKD, lactase persistence

Abstract
Introduction
The LCT: -13910C>T (rs4988235) polymorphism is associated with lactase persistence. Our previous study revealed impairment of pancreatic beta-cell function after an oral glucose tolerance test (OGTT) without a significant decrease in insulin sensitivity in ADPKD patients. The aim of the present study was to analyse clinical and biochemical parameters including indices of insulin sensitivity in regard to LCT polymorphism both in ADPKD patients and controls.

Material and methods
The study group consisted of 49 ADPKD patients and the control group comprised 50 healthy subjects. Plasma glucose, insulin and C-peptide concentrations were measured during WHO OGTT. The LCT polymorphism was identified by PCR-RFLP assay of samples of genomic DNA extracted from peripheral blood leukocytes.

Results
In the ADPKD group no significant differences in clinical and biochemical parameters were found between patients with lactase non-persistence (LNP) and individuals with lactase persistence (LP). In healthy subjects with LP (LCT: CT or TT genotype) body fat, the levels of insulin, C-peptide and insulin/glucose ratio (at OGTT times 0 and 120 minutes); area under curve ratios of secretory 1st and 2nd phase, homeostasis model assessment (HOMA)-insulin resistance and % beta, were significantly higher as compared with CC homozygotes (LNP). In addition, HOMA% sensitivity and indices of insulin sensitivity in LP controls were significantly lower as compared with healthy subjects with LNP.

Conclusions
Our results suggest that ADPKD patients are protected against a lactase-persistence-associated diabetogenic increase in insulin resistance.

Explanation letter
Dear Editors,
thank you very much for the evaluation of our article entitled „ADPKD protects against diabetogenic effects associated with genetically-predicted lactase persistence”. Please find below our answers to Reviewers’ comments.
Reviewer #1:
COMMENT: Dear authors. In the manuscript, there is not any scientific error, only the grammatical errors are clear in all parts of the manuscript. Therefore, I select the “Minor revision” option. After solving the grammatical errors, the recent manuscript can be accepted at the Archives of Medical Sciences.
Reviewer #3:
COMMENT: Dear authors. There isn't any scientific error in the recent manuscript. Only the authors should solve the grammatical errors. Dear authors. There isn't any scientific error in the recent manuscript. Only the authors should solve the grammatical errors.
RESPONSE: we fully agree with the Reviewers comments. English in the revised manuscript has been checked by a native speaker experienced in scientific and medical English. All grammar corrections have been marked using red font in the revised manuscript.

Summary
- line 9, 15 and 21: healthy subjects
- line 11: of
- line 22: a
- line 23: in
Manuscript body
- line 10: a mutated T allele (with
- line 14 and 15: a
- line 21: with fasting insulin levels
- line 31: higher compared with healthy controls
- line 35: parameters, including glycemic traits,
- line 36: polymorphism, both
- line 50: (kg)/(height, m)2.
- line 52: at
- line 90: analyses.
- line 93 and 94: tests were
- line 95: analyses of interactions between the LCT genotypes
- line 96: phenotypes
- line 111: distributions, in
- line 113: equilibria
- line 145: provides an
- line 147: homogeneous
- line 151: A
- line 152: have been found
- line 156: its
- line 158: its
- line 199: focused
- line 222-225: In addition, HOMA-IR ≥ 2.5 (the cut-off point for insulin resistance) in the control group was eight times more frequent in LP subjects (7 out of 23 subjects) than in CC homozygotes with lactase non-persistence (1 of 27 individuals).
- line 234: by
- line 248: A
- line 252: have
- line 253: phenotyping and analyses
- line 259: in
- line 262-264: We thank the anonymous native speaker (experienced in scientific and medical English) at the Department of Clinical and Molecular Biochemistry, Pomeranian Medical University, Szczecin, Poland, for proofreading the manuscript.
- line 371 and 376: the
Table II
-notes: cell
Reviewer #2:
COMMENT: It is well designed and described study concerning the relationship between diabetogenic effects correlated with genetically-predicted lactase persistence (LP) and autosomal dominant polycystic kidney disease (ADPKD). The final conclusion that ADPKD patients appear to be protected against LP- associated diabetogenic increase of insulin resistance is firmly supported by the presented analysis. The author also discuss their results in the context of relevant available data. I would suggest to consider in Discussion the paper by Vareesangthip et al, 1997 (doi: 10.1038/ki.1997.360) as well as the emerging contribution of LP to protection against insulin resistance in ADPDK. n.a.
RESPONSE: we fully agree with the Reviewer's comment. As recommended, we have added the suggested paragraphs in the Discussion:
- lines 165-176: In 1997, Vareesangthip et al. were the first to report that the prevalence of insulin resistance in ADPKD patients was significantly higher as compared with age- and sex-matched healthy subjects [23]. The authors assessed insulin sensitivity by a short insulin tolerance test (ITT) to derive the first-order rate constant for the disappearance of glucose (KITT) and found the KITT value indicating an insulin resistance in 10 of 15 (66.7%) ADPKD patients and in only 2 of 20 (10.0%) controls. In contrast, using homeostasis model assessment-insulin resistance (HOMA-IR) we revealed an insulin resistance state in 9 of 49 ADPKD patients and in 8 of 50 healthy controls (18.4% and

16.0%, respectively). However, it is worth noting that Inchiostro found a significant association between these two measures which was quite small and therefore they could not consider interchangeable estimates of insulin sensitivity [24].
-□lines 181-184: This suggests that lactase persistence contributes to protection against insulin resistance in ADPKD patients. These findings are supported by Abaturov and Nikulina who reported that CT or TT LCT genotypes (LP-related genotypes) were associated with a decreased risk of insulin resistance in Ukrainian children [25].
Therefore, we had to add 3 new references: [23], [24] and [25], and as the consequence to change the numbering of subsequent references.
Reviewer #4:
COMMENT: line 147: homogeneous instead of homogenous table II: beta cell function instead of beta cel function.
RESPONSE: we fully agree with the Reviewer's comment. We corrected both misspelings as requested (line 147: homogeneous instead of homogenous; legend for table II: cell instead of cel).
Once again, we do appreciate Your helpful comments. We have found them very helpful and useful in guiding our revision.

Yours sincerely
Andrzej Ciechanowicz

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Introduction

Lactose, a disaccharide and the main carbohydrate in milk, requires hydrolysis in the intestinal tract to release the monosaccharides galactose and glucose. This hydrolysis is catalyzed by the enzyme lactase (LPH, Lactase-Phlorizin Hydrolase) which is encoded by the *LCT* gene on chromosome 2q21 [1]. Lactase non-persistence (LNP) is a common autosomal recessive condition which results in physiological decline in LPH activity in intestinal cells after weaning. Sequence analysis of the *LCT* region carried out by Enattah et al. has revealed that the homozygous wild-type variant of the *LCT*: rs4988235 (-13910C>T) polymorphism ([C;C], referred to here as CC) completely co-segregates with biochemically verified LNP and a mutated T allele (with CT or TT genotypes) is associated with lactase persistence (LP) [2].

Lactase persistence, confirmed either by rs4988235 genotyping or by breath-hydrogen production, has been positively associated with body mass index (BMI) [3-5], diabetes mellitus [6], metabolic syndrome [7], body fat [5], a lower concentration of LDL-cholesterol [5] or a lower concentration of HDL-cholesterol [5,8]. However, other authors have not confirmed these associations [9-14].

In 2016 de Campos Mazo et al. reported that lactase non-persistence (*LCT*: CC genotype) in Brazilian patients with non-alcoholic steatohepatitis (n=93) was associated with insulin resistance [12]. In contrast, Yang et al. in a Mendelian randomization study revealed no association of *LCT*: rs4988235 polymorphism with insulin resistance and other glycemic traits except with fasting insulin levels [13]. Recently, the association of LNP with insulin resistance has been reported by de Luis et al. in 96 menopausal obese females in Spain [14].

Autosomal dominant polycystic kidney disease (ADPKD) is the most common hereditary kidney disease with a prevalence of 1:400 to 1:1000 live births among

subjects of European descent [15]. In most cases (85%) this is related to mutations of the *PKD1* gene encoding polycystin 1, with the remainder caused by mutations in the *PKD2* gene encoding polycystin 2 [16]. Previously, we have reported that in ADPKD patients with normal kidney function and no prior diagnosis of diabetes, the waist-to-hip ratio, systolic blood pressure, diastolic blood pressure and fasting glycemia were significantly higher compared with healthy controls [17]. In addition, we also found that ADPKD was associated with impaired pancreatic β-cell function after an oral glucose load without a significant decrease in insulin sensitivity [18].

Therefore, the aim of our study was to analyse clinical and biochemical parameters, including glycemic traits, with indices of β-cell function and indices of insulin sensitivity in regard to *LCT*: -13910C>T (rs4988235) polymorphism, both in ADPKD patients and healthy controls.

Material and methods

The study group consisted of 49 ADPKD patients (19 males and 30 females) and the control group comprised 50 gender- and age-matched healthy individuals (22 males and 28 females). All subjects gave informed, written consent to participate in the study, which was approved by the bioethics committee at the Pomeranian Medical University, Szczecin, Poland. Both ADPKD patients and controls were recruited according to a protocol described previously [18]. Briefly, at baseline, full medical history was reviewed with clinical examination of each subject. Clinical data included: age; sex; body mass; height; waist and hip circumference; body fat and water content measured using an infrared body composition analyser (Futrex 5000A/ZL, Futrex Inc., Hagerstown, USA); waist-to-hip ratio (WHR) and body mass index (BMI) calculated as body mass (kg)/(height, m)^2. In all subjects an oral glucose tolerance test (OGTT) with

75 g of glucose was performed according to WHO guidelines. Before glucose administration (at t = 0 minutes (min)), venous blood was collected to measure concentrations of glucose (GLU), insulin (INS), C-peptide, creatinine, triglycerides (TG), total cholesterol (TC), LDL-cholesterol (LDL-C) and HDL-cholesterol (HDL-C) as well as levels of glycated haemoglobin (HbA_{1C}). Venous blood was also taken at the 30^{th}, 60^{th} and 120^{th} minute of the OGTT to measure the concentrations of glucose and insulin. All laboratory measurements were performed as described previously [18]. The following beta-cell function or insulin sensitivity indices, based on the concentrations of glucose and insulin concentrations measured at several time points of the OGTT, were calculated using formulae described previously [18]: ratio of insulin-to-glucose concentrations (INS/GLU 0 and INS/GLU120); homeostasis model assessment-% beta (HOMA%B); ratio of the area under curve of insulin concentration to the area under curve of glucose concentration (SECR AUC), secretory 1^{st} phase (SECR1P) and 2^{nd} phase (SECR2P) calculated from the first 30 or 60 minutes of OGTT (SECR1P 30 and SECR1P 60 or SECR2P 30 and SECR2P60, respectively); index of beta cell function (INDXBETA); insulinogenic index (INSGENIN) or homeostasis model assessment-% sensitivity (HOMA%S) and its reciprocal homeostasis model assessment-insulin resistance (HOMA-IR); insulin sensitivity indices (ISI0 and ISI120); insulin sensitivity composite index (ISI COMP); and Cederholm sensitivity index (ISI CEDE).

Genotyping

Genomic DNA was isolated from human whole blood (using QIAapm® DNA Mini Kit; Qiagen, Hilden, Germany). PCR was performed using: 5'-CATGGAGGATTACAGTGCG ACAGC-3' as the forward primer and 5'-CCTTGGTTGAAGCGAAGATGGGA-3' as the reverse primer to generate a 576-bp

amplicon. The reaction was performed in 20 µl volumes containing 2 µl DNA (80 ng); 10 µl buffer (2x PCR Master Mix; Thermo Fisher Scientific, Waltham, MA, USA)); 0.2 µl forward primer [20 pmol/µl]; 0.2 µl reverse primer [20 pmol/µl] and 7.6 µl H2O. The PCR was carried out (in a Mastercycler Gradient; Eppendorf, Hamburg, Germany) and included an initial denaturation at 94°C for 5 min followed by 34 cycles of denaturation at 94°C for 30 s, primer annealing at 58°C for 40 s, extension at 72°C for 30 s, and a final extension at 72°C for 10 min. The amplified product was digested with FaqI (BsmFI) (Thermo Fisher Scientific), generating for the wild-type allele (*LCT*: -13910C) three fragments of 401 bp, 151 bp and 24 bp in length or for the mutated allele (-13910T) four fragments of 253 bp, 151 bp, 148 bp and 24 bp in length. Electrophoresis of RFLP products was performed in 2% agarose gel containing ethidium bromide and photographed using a gel documentation system (G:BOX BioImaging System; Syngene, Bangalore, India).

Statistical analyses.

Data are presented as numbers and percentages for qualitative variables or mean ± standard deviation for quantitative variables. As most quantitative variables were not distributed normally, Mann-Whitney tests were used for comparison between groups, while Fisher's exact tests were applied for qualitative variables. General linear models (GLMs) were created for bivariate analyses of interactions between the *LCT* genotypes and ADPKD phenotypes as independent variables associated with quantitative dependent variables, which were transformed logarithmically when not distributed normally. Results with $p<0.05$ were considered statistically significant. Commercial software (Statistica 13, Statsoft Polska, Krakow, Poland) was used for all statistical analyses.

Results

There were 22 CC homozygotes (45%), 19 CT heterozygotes (39%) and 8 TT homozygotes (16%) in the ADPKD group. The frequency of the minor *LCT*: -13910T allele in ADPKD patients was 36%. There were 27 CC homozygotes (54%), 18 CT heterozygotes (36) and 5 TT homozygotes (10%) in the control group consisting of healthy subjects. The frequency of the minor *LCT*: -13910T allele in controls was 28%. No significant differences in frequency distribution of genotypes or alleles were found between ADPKD patients and healthy subjects ($p = 0.544$ or $p = 0.244$, respectively). In addition, no significant differences ($p = 0.366$) in the frequency of genetically-predicted lactase persistence (CT or TT genotype) were found between the ADPKD group (55%) and controls (46%). The *LCT*: rs4988235 genotype distributions, in the ADPKD group, in the control group and in the combined group (ADPKD patients and control group) conformed to expected Hardy-Weinberg equilibria ($p = 0.276$, $p = 0.449$ and $p = 0.168$, respectively).

In the ADPKD group no significant differences in clinical and biochemical parameters (Table I), as well as in values of glycemic traits (Table II), were found between patients homozygous for the wild-type *LCT* allele (-13910C) and individuals having at least one mutated allele (CT or TT genotype). In healthy subjects with lactase persistence (CT or TT *LCT* genotype), body fat and total body fat were significantly higher and total body water was significantly lower as compared with lactase non-persistent controls (subjects homozygous for wild-type *LCT*: -13910C allele). In addition, the frequency of impaired fasting glucose (IFG) in LP controls was significantly higher as compared with LNP (Table I). The values of the

Table 2

following glycemic traits: INS 0, INS 120, C-peptide 0, C-peptide 120, HOMA-IR, HOMA-%B, INS/GLU 0, INS/GLU 120, SECR1P 60 and SECR2P 60 were significantly higher in healthy subjects with genetically-predicted LP as compared with LNP. In addition, HOMA%S, ISI 0, ISI 120 and ISI COMP in controls with lactase persistence were significantly lower as compared with healthy subjects homozygous for the *LCT*: -13910C allele (Table II).

Table II

Bivariate analysis was performed to search for interaction between *LCT* genotype and ADPKD phenotype in their association with percentage of fat in body weight and fasting plasma insulin concentration. The general linear model (GLM) in both cases confirmed a significant interaction (p=0.035 for percentage of fat and p=0.033 for fasting plasma insulin transformed logarithmically, (Figures 1 and 2) which can be interpreted in the following way: the *LCT*:g.-13910T allele was associated with a higher percentage of fat, in body weight and fasting plasma insulin only in controls without ADPKD, while the presence of ADPKD phenotype broke these associations.

Figure 1

Figure 2

Discussion

Our study provides an association analysis of the *LCT*: -13910C>T (rs4988235) polymorphism responsible for lactase persistence with anthropometry as well as lipid and glucose metabolism parameters in a homogeneous group of ADPKD patients of European descent.

149 The mechanism responsible for lactase persistence is the existence of cis-
150 element mutations mapping to a gene other than *LCT* (the regulatory enhancer region
151 *MCM6*). A total of twenty-three single nucleotide polymorphisms (SNPs) within the
152 *MCM6* have been found to be associated with LP so far in human populations, among
153 them the only -13910C>T mutation responsible for lactase persistence in subjects of
154 European ancestry. These variants seem to have arisen during the same period but
155 independently in different human populations [19]. The frequency of LP genotypes (CT
156 heterozygotes or TT homozygotes) in our ADPKD patients was similar to its prevalence
157 in control subjects (45% and 54%, respectively) indicating good matching and no
158 population bias in our recruiting of ADPKD patients. The frequency of LP in the
159 combined group was 51%. This value is equal to the LP frequency reported previously
160 in Polish subjects (51.0%) by Ploszaj et al. [20]. In addition, Fojcik et al. and Mądry et
161 al. reported even higher LP prevalence in Poles (69.0% or 68.5%, respectively)[21,22].
162 This reported high LP frequency is also found in other northern European populations,
163 but frequencies decrease across Southern Europe and the Middle East, and the
164 frequency is low in non-pastoralist Asian and African communities [19].
165 In 1997, Vareesangthip et al. were the first to report that the prevalence of
166 insulin resistance in ADPKD patients was significantly higher as compared with age-
167 and sex-matched healthy subjects [23]. The authors assessed insulin sensitivity by a
168 short insulin tolerance test (ITT) to derive the first-order rate constant for the
169 disappearance of glucose (K_{ITT}) and found the K_{ITT} value indicating an insulin
170 resistance in 10 of 15 (66.7%) ADPKD patients and in only 2 of 20 (10.0%) controls. In
171 contrast, using homeostasis model assessment-insulin resistance (HOMA-IR) we
172 revealed an insulin resistance state in 9 of 49 ADPKD patients and in 8 of 50 healthy
173 controls (18.4% and 16.0%, respectively).

However, it is worth noting that Inchiostro found a significant association between these two measures which was quite small and therefore they could not consider interchangeable estimates of insulin sensitivity [24].

Our results have shown that genetically-predicted lactase persistence (LP) in adult healthy subjects of European descent, but not in age- and gender-matched ADPKD patients, was associated with indices of increased beta-cell function and decreased insulin sensitivity (the latter equivalent to indices of increased insulin resistance). This suggests that lactase persistence contributes to protection against insulin resistance in ADPKD patients. These findings are supported by Abaturov and Nikulina who reported that CT or TT LCT genotypes (LP-related genotypes) were associated with a decreased risk of insulin resistance in Ukrainian children [25].

In addition, we also found that body fat percentage and body fat mass in LP controls were significantly higher as compared to healthy subjects with lactase non-persistence. Previously, association of genetically-predicted lactase persistence with higher body fat content has been reported by Manco et al. in young adults (mean age 20.8 years) from Portugal [26], by Malek et al. in a group of multiethnic children (n = 296, 7–12 years old)[27] and by Vimaleswaran et al. in a meta-analysis of data from three large-scale population-based studies (1958 British Birth Cohort, Health and Retirement study, and the UK Biobank) with up to 417 236 participants [5]. In addition, Popadowska et al. in a group of Polish healthy blood donors revealed that higher intake of milk and dairy products was associated with higher fat body mass but only among men with lactase persistence [28]. However, Almon et al. reported that lactase persistence was not linked to higher body fat mass in children (n=298, mean age 9.6 years) or adolescents (n=386, mean age 15.6 years) belonging to the Swedish component of the European Youth Heart Study [29].

In contrast to the above studies which have focused on analysis of the association of lactase persistence with body fat, reports concerning indices of insulin sensitivity or pancreatic beta-cell function in regard to *LCT*: rs4988235 polymorphism are scarce and, in contrast to our results, previous results have suggested that lactase non-persistence rather than lactase persistence is associated with insulin resistance [12,14]. Mazo et al. analysed the *LCT*: -13910C>T polymorphism in 93 Brazilian patients with non-alcoholic steatohepatitis (NASH). Among these subjects, 60.7% suffered from type 2 diabetes, 83.5% had insulin resistance (defined as a HOMA-IR value ≥ 2.5), and mean age was 56.5 ± 10.1 years old and mean BMI was 31.2 ± 5.9 kg/m^2. The authors used univariate analysis to reveal that lactase non-persistence (CC homozygous genotype) was associated with significantly higher insulin levels (23.47 ± 15.94 μU/mL vs 15.8 ± 8.33 μU/mL, p = 0.027) with a significantly higher frequency of insulin resistance (91.84% vs 72.22%, p = 0.02) as compared with the lactase persistence. Moreover, LNP in NASH patients was an independent risk factor for insulin resistance even after adjusting for gender and age [OR = 5.0 (95%CI: 1.35-20; P = 0.017)[12]. Recently, de Luis et al. investigated a group of 86 Spanish menopausal females with obesity defined as BMI ≥ 30.0 kg/m^2 (15% of them suffered from type 2 diabetes). The authors found that LNP females had significantly higher fasting glucose levels, insulin levels, and HOMA-IR values as compared with LP ones [14].

However, we are confident that the results of studies by Mazo et al. [12] and de Luis et al. [14] were carried out in subjects not representative for the general population and do not undermine the reliability of our findings. Our study was carried out in adult healthy Poles aged from 22 to 55 years. No studied subjects suffered from diabetes and only 10% had BMI ≥ 30.0 kg/m^2. In addition, HOMA-IR ≥ 2.5 (the cut-off point for insulin resistance) in the control group was eight times more frequent in LP subjects (7

out of 23 subjects) than in CC homozygotes with lactase non-persistence (1 of 27 individuals). We also hypothesize that the association of lactase non-persistence with insulin resistance reported above in Brazilian NASH patients [12] or in Spanish menopausal women with obesity [14] may be linked in those subjects with the avoidance of milk and dairy products and their replacement by more diabetogenic food products (the ongoing nutrition transition). It is noteworthy that Mosley et al. showed that the rise in metabolic syndrome is accompanied by a decrease in milk and dairy consumption and an increase in sugar-sweetened beverage (SSB) consumption, with SSB possibly displacing dairy products in the diet [30].

Despite the identification of *PKD1* and *PKD2* genes more than 20 years ago, the mechanisms by which their mutations cause ADPKD and, as revealed by our study, attenuate insulin resistance remains unclear. Over the years, numerous interacting proteins have been identified, using directed and unbiased approaches, which have been shown to modulate the function, cellular localization, and protein stability and turnover of polycystins. *PKD1* encodes a large plasma membrane protein (PKD1, PC1, or Polycystin-1) with a long extracellular domain which has been speculated to function as an atypical G-protein-coupled receptor. *PKD2* encodes an ion channel of the Transient Receptor Potential superfamily (TRPP2, PKD2, PC2, or Polycystin2) [31]. Kim et al. revealed that WNT proteins bind to the extracellular domain of PKD1 and induce whole cell currents and Ca^{2+} influx dependent on TRPP2. Pathogenic *PKD1* or *PKD2* mutations that abrogate complex formation, compromise cell surface expression of PKD1, or reduce TRPP2 channel activity suppress activation by WNTs [32]. It is worth noting that we have previously reported an increase in Ca^{2+} concentration in the erythrocytes of our ADPKD patients with normal renal function [33].

A major limitation of our study is the fairly low sample size. However, in contrast to the reports of de Campos Mazo et al. or de Luis et al. who analysed the association of LCT polymorphism with insulin resistance in 93 Brazilian patients with non-alcoholic steatohepatitis [12] or in 96 menopausal obese females in Spain, respectively [14], we have rather focused on healthy subjects (n=50) with extensive phenotyping and analyses of biochemical characteristics. In addition, the results in healthy subjects were compared with results in age- and gender-matched ADPKD patients (n=50).

Conclusion

Our results suggest that ADPKD patients are protected against a lactase persistence-associated diabetogenic increase in insulin resistance.

Acknowledgements

We thank the anonymous native speaker (experienced in scientific and medical English) at the Department of Clinical and Molecular Biochemistry, Pomeranian Medical University, Szczecin, Poland, for proofreading the manuscript.

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Legends of figures

Figure 1. Bivariate analysis of the interaction between *LCT*: -13910C>T genotype and ADPKD phenotype with association with percentage of body fat using a general linear model (GLM). Marginal means with 95% confidence intervals are presented. The interaction is statistically significant (p=0.035).

Figure 2. Bivariate analysis of the interaction between *LCT*: -13910C>T genotype and ADPKD phenotype with association with fasting plasma insulin, transformed logarithmically, using a general linear model (GLM). Marginal means with 95% confidence intervals are presented. The interaction is statistically significant (p=0.033).

Table I. Clinical and biochemical characteristics of the ADPKD patients and control subjects in regard to *LCT*: -13910C>T polymorphism.

Variable	ADPKD patients			Control subjects		
	CC (n=22)	CT+TT (n=27)	*p* [a]	CC (n=27)	CT+TT (n=23)	*p* [a]
Age, years	35.6 ± 11.5	36.0 ± 11.0	0.882	35.4 ± 7.7	38.2± 10.7	0.487
Gender (M/F), n	7/15	13/14	0.381	13/14	9/14	0.577
Current smokers, n (%)	8 (36%)	9 (33%)	1.000	6 (22%)	9 (39%)	0.228
Family history of DM, n (%)	1 (4%)	8 (30%)	0.059	4 (15%)	6 (26%)	0.480
SBP, mmHg	136.4±21.2	131.9±18.7	0.454	122.7±16.2	121.5±13.6	0.862
DBP, mmHg	93.6±12.8	92.3±12.3	0.597	82.6±8.9	83.2±9.93	0.908
Waist, cm	81.3 ± 14.6	86.7 ± 14.4	0.150	79.1 ± 11.4	83.2 ± 14.6	0.428
Hip, cm	97.9 ± 11.1	102.3 ± 8.5	0.121	99.0 ± 6.3	101.4 ± 6.8	0.157
WHR	0.83 ± 0.09	0.84 ± 0.10	0.629	0.79 ± 0.08	0.82 ± 0.11	0.685
Weight, kg	70.2 ± 16.0	78.4 ± 16.9	0.089	72.1 ± 14.5	74.6 ± 15.7	0.743
BMI, kg/m^2	24.2 ± 5.2	25.8 ± 4.7	0.252	23.7 ± 3.7	25.2 ± 3.4	0.140
Obesity, %	4 (18%)	5 (26%)	0.272	2 (7%)	3 (17%)	0.439
Body fat, kg	20.1 ± 7.0	22.9 ± 7.9	0.117	17.2 ± 6.5	22.6 ± 7.3	0.005
Total body fat, %	28.3 ± 6.2	28.8 ± 7.2	0.494	24.1 ± 7.2	30.7 ± 7.7	0.003
Total body water, %	54.7 ± 4.1	54.5 ± 4.8	0.546	57.6 ± 4.6	53.1 ± 4.9	0.003
TC, mg/dL	192.7 ± 38.7	196.2 ± 37.1	0.665	189.8 ± 38.7	197.0 ± 36.7	0.373
LDL, mg/dL	124.4 ± 37.5	127.6 ± 34.4	0.833	118.4 ± 32.6	120.1 ± 36.7	0.847
HDL, mg/dL	57.8 ± 13.2	55.2 ± 12.5	0.475	60.7 ± 15.1	62.2 ± 22.1	1.000
TG, mg/dL	98.2 ± 61.3	99.2 ± 58.1	0.825	96.8 ± 46.9	128.6 ± 81.5	0.168
HbA_{1C}, %	5.4 ± 0.4	5.4 ±0.3	0.755	5.2 ± 0.2	5.4 ±0.4	0.201
IFG, n (%)	5 (23%)	5 (18%)	0.736	0 (0%)	5 (22%)	0.016
IGT, n (%)	1 (4%)	0 (0%)	0.449	0 (0%)	1 (4%)	0.460
Creatinine, mg/dl	0.85 ± 0.21	0.82 ± 0.17	0.597	0.81 ± 0.17	0.77 ± 0.11	0.451
e-GFR, ml/min/1.73m^2	94.0 ± 24.1	101.2 ± 18.1	0.323	103.2 ± 22.5	103.0 ± 16.7	0.923

[a] Mann-Whitney tests for quantitative variables and Fisher's exact tests for qualitative variables. Notes: M, male; F, female; DM, type2 diabetes; SBP, systolic blood pressure; DBP, diastolic blood pressure; WHR, waist to hip ratio; BMI, body mass index; TC, total cholesterol; LDL, low density lipoprotein-cholesterol; HDL, high density lipoprotein-cholesterol; HDL, high density lipoprotein-cholesterol; TG, triglycerides; HbA_{1c}, glycated haemoglobin; IFG, impaired fasting glucose; IGT, impaired glucose tolerance; e-GFR, estimated glomerular filtration rate.

Table II. Glycemic traits in the ADPKD patients and control subjects in regard to *LCT*: -13910C>T polymorphism.

Variable	ADPKD patients			Control subjects		
	CC (n=22)	CT+TT (n=27)	*p* [a]	CC (n=27)	CT+TT (n=23)	*p* [a]
GLU 0 (mg/dl)	90.7±9.32	92.4±11.6	0.54	85.2±7.78	90.1±10.2	0.091
GLU 120 (mg/dl)	93.3±22.5	86.59±20.3	0.23	82.5±17.9	94.2±29.4	0.253
INS 0 (µU/mL)	8.81±5.2	9.16±4.49	0.68	6.52±2.62	10.55±5.23	<0.001
INS 120 (µU/mL)	45.3±45.3	36.6±27.2	0.73	32.83±22.3	67.2±62.1	<0.001
C-peptide 0 (µU/mL)	2.26±0.59	2.27±0.77	0.98	1.78±0.46	2.37±0.75	0.003
C-peptide 120 (µU/mL)	7.77±2.7	7.31±3.23	0.48	6.42±2.47	8.96±2.95	<0.001
INS/GLU 0	1.7 ± 0.9	1.8 ± 0.8	0.71	1.4 ± 0.6	2.1 ± 0.9	0.002
INS/GLU 120	7.7 ± 6.9	7.3 ± 4.7	0.94	6.8 ± 3.6	12.2 ± 9.01	<0.001
HOMA%B	122.6 ± 64.8	127.9 ± 78.3	0.89	119.2 ± 67.9	149.5 ± 66.7	0.025
SECR AUC	7.7 ± 3.9	8.9 ± 3.8	0.15	8.6 ± 3.6	10.5 ± 4.0	0.071
SECR1P 30	-33.3 ± 617.7	-1.5 ± 436.3	0.32	31.4 ± 389.3	222.9 ± 439.2	0.125
SECR2P 30	325.1 ± 144.4	331.4 ± 99.1	0.33	335.5 ± 85.6	382.9 ± 104.1	0.107
SECR1P 60	141.8 ± 562.6	314.6 ± 518.6	0.079	184.8 ± 305.8	465.8 ± 538.6	0.044
SECR2P 60	348.5 ± 138.6	389.6 ± 126.9	0.063	355.4 ± 74.9	426.1 ± 132.9	0.038
INDEXBETA	76.2 ± 436.9	22.4 ± 939.1	0.57	187.6 ± 211.8	215.1 ± 450.7	0.537
INSGENIN	46.6 ± 25.8	49.2 ± 26.9	0.69	59.7 ± 30.9	60.6 ± 24.6	0.599
HOMA%S	64.9 ± 32.4	61.7 ± 31.6	0.76	91.9 ± 66.3	54.6 ± 28.3	0.002
HOMA-IR	2.02 ± 1.38	2.13 ± 1.21	0.76	1.38 ± 0.54	2.42 ± 1.41	0.002
HOMA-IR≥2.5, n (%)	3 (14)	6 (22)	0.488	1 (4)	7 (30)	0.017
ISI 0	0.08 ± 0.02	0.08 ± 0.02	0.47	0.1 ± 0.01	0.08 ± 0.02	0.009
ISI 120	0.09 ± 0.04	0.1 ± 0.02	0.99	0.1 ± 0.02	0.08 ± 0.04	0.002
ISI COMP	5.8 ± 2.6	5.4 ± 2.6	0.45	7.3 ± 3.4	4.7 ± 2.6	0.004
ISI CEDE	27.3 ± 9.4	27.5 ± 7.6	0.79	31.4 ± 8.4	26.1 ± 9.2	0.059

Notes: GLU, glucose concentration; INS, insulin concentration; HOMA%B, homeostasis model assessment-%beta; SECR AUC, ratio of the area under curve of insulin concentration to the area under curve of glucose concentration; SECR1P 30, secretory phase 1 calculated from the first 30 minutes of OGTT; SECR1P 60, secretory phase 1 calculated from the first 60 minutes of OGTT; SECR2P 30, secretory phase 2 calculated from the first 30 minutes of OGTT; SECR2P 60, secretory phase 2 calculated from the first 60 minutes of OGTT; INDEX BETA, index of beta cell function; INSGENIN, insulinogenic index; HOMA%S, homeostasis model assessment-% sensitivity; HOMA-IR, homeostasis model assessment-insulin resistance; ISI, insulin sensitivity index; ISI COMP, insulin sensitivity composite index; ISI CEDE, Cederholm sensitivity index.

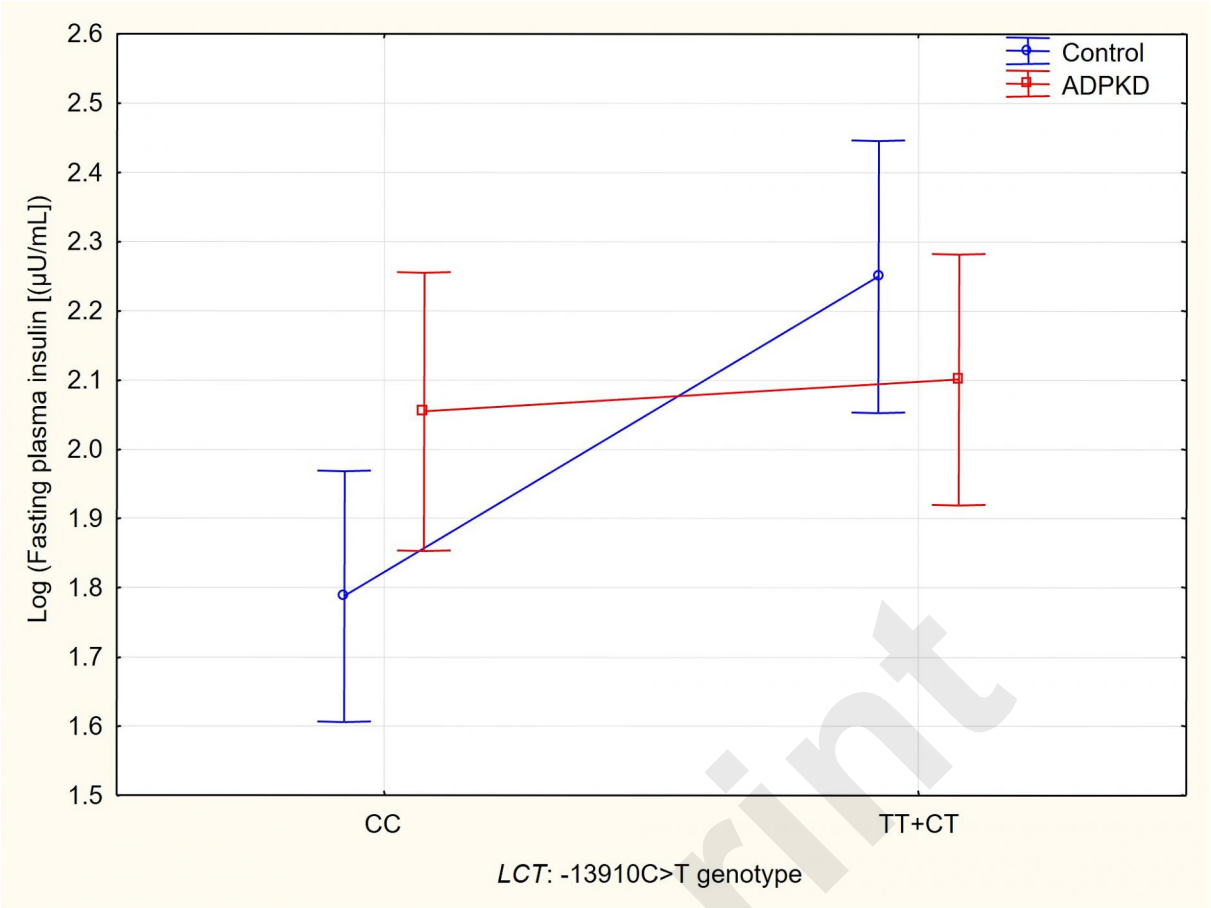

Figure 1. Bivariate analysis of interaction between LCT: -13910C>T genotype and ADPKD phenotype with association with percentage of body fat using a general linear model (GLM). Marginal means with 95% confidence intervals are presented. The interaction is statistically significant (p=0.035).

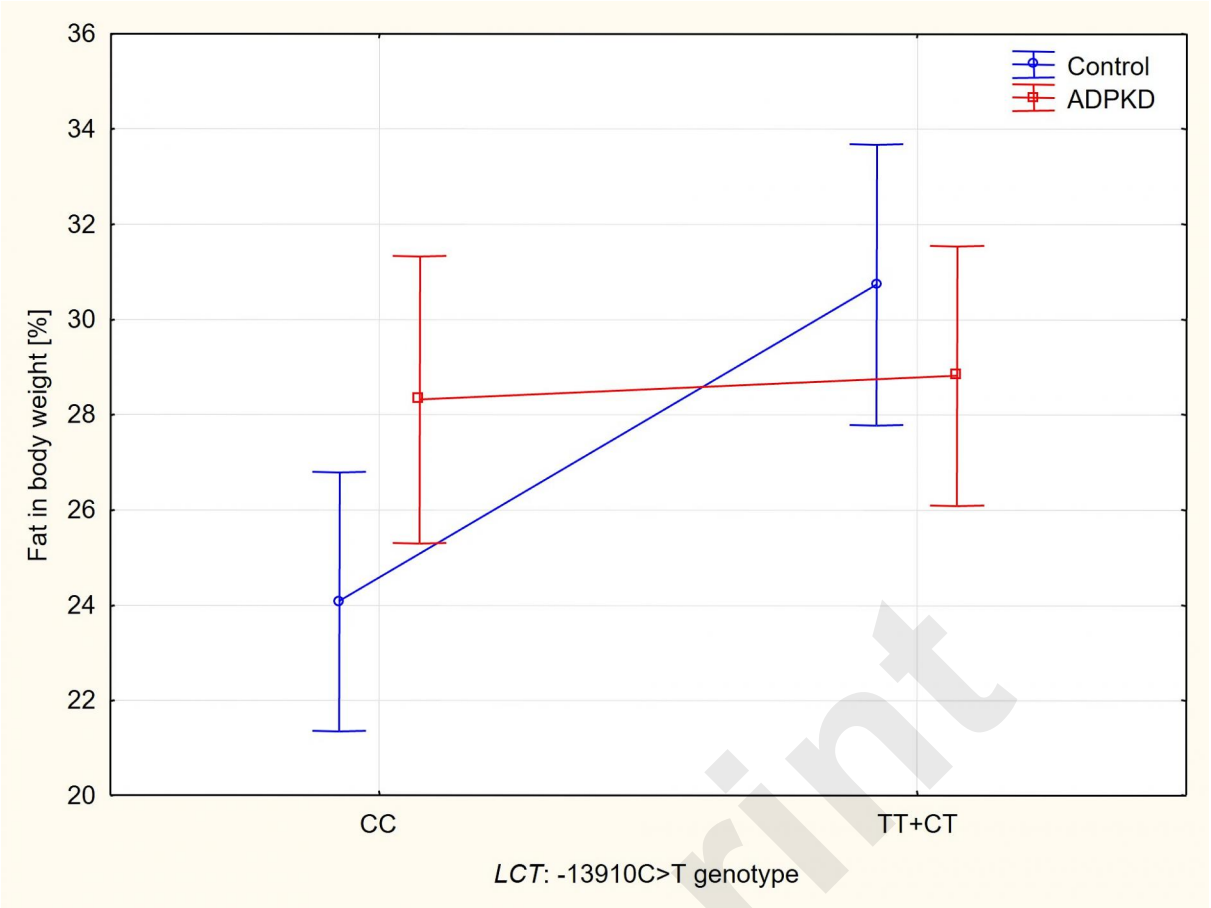

Figure 2. Bivariate analysis of interaction between LCT: -13910C>T genotype and ADPKD phenotype with association with fasting plasma insulin transformed logarithmically using a general linear model (GLM). Marginal means with 95% confidence intervals are presented. The interaction is statistically significant (p=0.033).