Association of Age-Related Cataract Risk with High Polygenetic Risk Scores Involved in Galactose-Related Metabolism and Dietary Interactions

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Age-related cataract · Polygenetic risk score · Galactose · Lactose · Milk · Dairy product

Abstract

Introduction: Cataracts are associated with the accumulation of galactose and galactitol in the lens. We determined the polygenetic risk scores for the best model (PRSBM) associated with age-related cataract (ARC) risk and their interaction with diets and lifestyles in 40,262 Korean adults aged over 50 years belonging to a hospital-based city cohort.

Methods: The genetic variants for ARC risk were selected in lactose and galactose metabolism-related genes with multivariate logistic regression using PLINK 1.9 version. PRSBM from the selected genetic variants were estimated by generalized multifactor dimensionality reduction (GMDR) after adjusting for covariates. The interactions between the PRSBM and each lifestyle factor were determined to modulate ARC risk.

Results: The genetic variants for ARC risk related to lactose and galactose metabolism were SLC2A1_rs3729548, ST3GAL3_rs3791047, LCT_rs2304371, GALNT5_rs6728956, ST6GAL1_rs2268536, GALNT17_rs17058752, CSGALNACT1_rs1994788, GALNT4_rs10831608, B4GALT6_rs1667288, and A4GALT_rs9623659. In GMDR, the best model included all ten genetic variants. The highest odds ratio for a single SNP in the PRSBM was 1.26. However, subjects with a high-PRSBM had a higher ARC risk by 2.1-fold than a low-PRSBM after adjusting for covariates. Carbohydrate, dairy products, kimchi, and alcohol intake interacted with PRSBM for ARC risk, where participants with high-PRSBM had a much higher ARC risk than those with low-PRSBM when consuming diets with high carbohydrate and low dairy product and kimchi intake. However, only with low alcohol intake, the participants with high-PRSBM had a higher ARC risk than those with low-PRSBM.

Conclusion: Adults aged >50 years having high-PRSBM may modulate dietary habits to reduce ARC risk.

Introduction

A cataract is defined as eye lens opacity causing light not to focus on the retina, which is the most common cause for preventable low vision. Cataracts may be congenital or developed at an advanced age, considered an age-related cataract (ARC). ARC commonly occurs in people over 50 years old [1]. The potential risk factors of cataracts are classified as genetic and environmental factors. Environmental factors include age, smoking, diabetes, eye injury, chronic use of steroids, heavy alcohol drinking, and ultraviolet exposure. ARC is induced by in-
creased osmotic pressure, oxidative stress, and inflammation [2].

When monosaccharides, including glucose and galactose, are not properly metabolized in the lens, they can cause a cataract. Galactose can be produced in the body, and it is metabolized through the DeLey-Doudoroff pathway [3]. If the genes in the pathway are mutated, the expressions and functions of the mutated genes are changed, and galactose metabolism is impaired. Glucose and galactose are converted into sorbitol and galactitol by aldose reductase, and the concentrations of sorbitol and galactitol increase in the lens [3]. As a consequence of the process, oxidative stress is elevated, and sugar alcohol induces osmotic damage in the lens [4], causing cellular damage, leading to the development of cataracts. Additionally, galactokinase deficiency is an autosomal recessive genetic disease that causes hypergalactosemia, which induces cataracts [5]. Galactokinase (GALK) missense mutations (Ala198Val and Pro28Thr) and UDP-galactose 4-epimerase-A (Val94Met) can lead to hypergalactosemia [6, 7]. Persons with the GALK_Aal198Val variant have a higher risk of congenital cataract formation [7]. Mutation of some genes, including FOXE3, HSF4, MAF, PITX3, and CRYAB, are reported to be associated with cataract risk [8–10].

Dairy products, mainly milk, have a reputation for causing cataracts. However, there is no evidence that milk and dairy product consumption itself increases cataract risk. This belief may come from the association of GALK mutations and cataract risk [10]. Galactose mostly comes from the lactose in milk and dairy products by lactase, an inducible gene, in the intestines [11, 12]. However, galactose is also endogenously produced in the body, and it needs to be metabolized [3]. GALK is necessary to metabolize galactose, and persons with attenuated galactose utilization may be at increased risk of developing ARC. Continuous dairy product consumption by infants produces lactose- and galactose-metabolizing genes to metabolize lactose and galactose [13]. However, discontinuation of dairy products prevents the symptoms of lactose intolerance and impairs galactose metabolism in the body. Dairy product intake may protect against the impairment of galactose metabolism to protect against ARC. Since lactic acid bacteria also participate in lactose digestion [14, 15], bacteria from fermented foods such as kimchi may utilize lactose and galactose in the intestines. Therefore, lactose and galactose metabolism in the intestines may interact with modulating ARC risk [12].

Galactose is converted into glucose by several enzymes, including GALK, galactose-1-phosphate uridylyltransferase (GALT), aldose transferase, galactose dehydrogenase (GALDH), and UDP-glucose/galactose pyrophosphorylase (UGP), and other genes in the body [16, 17]. Lactose intolerance due to an inborn error of metabolism with nonfunctional GALT is known to result in cataracts at a young age. However, genetic variants of GALT and other enzymes related to lactose and galactose metabolism may be associated with an increased risk of ARC due to disruption of normal galactose metabolism. The genetic variants may interact with each other, and they may have environmental interactions, including food intake (vegetables, kimchi, fruits, and dairy products), physical activity, smoking, alcohol drinking, and steroid usage, which may increase the risk of ARC. Therefore, we hypothesized that (1) the genetic variants related to lactose and galactose metabolism are associated with ARC risk and (2) the polygenetic risk score generated from the ARC-related genetic variants with gene-gene interactions interacts with dairy products and other foods and nutrients. These hypotheses were examined in 40,262 Korean adults >50 years old in a hospital-based city cohort.

Methods

Participants

Middle-aged and elderly Korean adults volunteered to participate in this hospital-based city cohort study. During 2004–2013, 40,262 adults aged >50 years participated in the city hospital cohorts under the Korean Genome and Epidemiology Study (KoGES) established by the Korean Center for Disease and Control. The present study, including KoGES procedures, followed the Helsinki Declaration. The Institutional Review Board on KoGES approved this study (KBP-2015-055). All subjects that participated in KoGES provided written informed consent. The Institutional Review Board of Hoseo University approved the present study (1041231-150811-HR-034-01).

Definition of ARC and Metabolic Syndrome

The presence of ARC was defined by using a questionnaire whether participants had a diagnosis of cataracts by a physician after 50 years old, and those who answered yes were considered to have ARC. Since ARC primarily develops after age 50, only participants over 50 were included in this study. Among the participants, 1,972 participants were previously diagnosed with cataracts after age 50 and 38,290 without a cataract diagnosis.

Metabolic syndrome comprised 3 or more of the following categories: (1) waist circumference ≥90 cm for men and ≥85 cm for women (abdominal obesity); (2) average systolic blood pressure ≥130 mm Hg or diastolic blood pressure ≥85 mm Hg (hypertension); (3) <40 mg/dL for men and <50 mg/dL for women (hypothyroidism); (4) ≤150 mg/dL (hypertriglyceridemia); and (5) ≥110 mg/dL (hyperglycemia) [18]. Participants who were taking medication for dyslipidemia, hyperglycemia, and hypertension were included in the metabolic syndrome group.
Clinical Parameters Measurements
Trained researchers interviewed the participants to collect health interview including age, education and income levels, the status of smoking and alcohol intake, and physical activity [19, 20]. We divided education levels into three groups: less than graduation of high school, graduation of high school, and graduation of college, or more. We categorized household income (USD/month) into four groups: very low (<USD 1,000), low (USD 1,000–2,000), intermediate (USD 2,000–4,000), and high (>USD 4,000) [21]. Current and no smoking status was defined as quitting smoking for at least the last 6 months [21]. Alcohol consumption status was categorized into four groups according to average daily alcohol consumption: non-drinker (0 g), moderate drinker (0–20 g), and heavy drinker (>20 g). The consumption of dairy products including cheese, yogurt, and milk was classified into <0.75 serving/day (low dairy products) and ≥0.75 serving/day (high dairy products).

A standardized procedure was adopted to measure waist circumference, height, and body weight [22]. We have calculated BMI as the weight in kilograms per height in meter squared (kg/m²). Blood sampling was performed after an overnight fast. Biochemical measurements were conducted using plasma and serum samples [22]. An Automatic Analyzer (Hitachi 7600; Hitachi, Tokyo, Japan) was used for the determination of concentrations of serum glucose levels and hemoglobin A1c in serum. We measured blood pressure on the right arm at the same height as the subject’s heart in a sitting position.

Genotyping
The Center for Genome Science in the Korea National Institute of Health provided the genotype data. Genomic DNA was extracted from whole blood, and genotypes determined on Korean Chip (Affymetrix, Santa Clara, CA, USA) made available to the scientists. Korean Chip was implemented for studying Korean genetic variants and included the critical SNPs related to metabolic diseases (diabetes, hypertension, dyslipidemia, osteoporosis, kidney disease, and autoimmune diseases) and cancers [23]. The genotyping accuracy was examined by Bayesian Robust Linear Modeling using the Mahalanobis Distance (BRLMM) Genotyping Algorithm. DNA samples included genotyping accuracies (≥98%), missing genotype call rates (<4%), low heterozygosity (<30%), or no gender biases. We included genetic variants that satisfy the inclusion criteria of HWE (p > 0.05) [24].

Association of ARC with the Genetic Variants Related to Lactose and Galactose Metabolism
Subjects were categorized into having ARC (case; n = 1,971) and noncataract (control; n = 38,290). The genes related to lactose and galactose metabolism were selected from the Kyoto Encyclopedia of Genes and Genomes database (https://www.kegg.jp/kegg/kegg2.html) using keywords of lactose and galactose metabolism including the DeLey-Doudoroff pathway. The locations of the related genes were used to explore the genetic variants associated with ARC. The association of ARC with the genetic variants related to lactose- and galactose-related metabolism was conducted with covariates including age, sex, residence area, education, income, BMI, and serum glucose concentrations using PLINK 1.9 version, a free, open-source whole genome association analysis toolset (https://zzz.bwh.harvard.edu/plink/). Covariates were used to eliminate the confounding effects to find the genetic variants associated with ARC. The genetic variants associated with ARC were selected at p < 0.05. Linkage disequilibrium (LD) analyses were performed in the selected genetic variants in the same chromosome using Haploview 4.2 in PLINK, and the 10 potential genetic variants for the best model in the same chromosome did not show a strong correlation (r² < 0.4) in LD.

Identification of the Best Model for Genetic Variant-Genetic Variant Interaction by Generalized Multifactor Dimensionality Reduction
The genetic variants selected were used to find the best genetic variant-genetic variant interaction model using the GMMDR method [19]. The best genetic variant-genetic variant interaction model was searched in a sign rank test of trained balanced accuracy (TRBA) and testing balanced accuracy (TEBA) with or without adjusting for covariates using a GMMDR program and a p value threshold of 0.05 [20]. The covariates included age, gender, residence area, education, income level, and BMI. Ten-fold cross-validation was also checked by cross-validation consistency (CVC) since the sample size was greater than 1,000 [20]. Ten out of 10 in CVC met the perfect cross-validation. Therefore, the best model was selected from the criteria of the sign test result between TRBA and TEBA and CVC value.

The risk allele of each SNP in the selected best model was counted as 1. If persons had AA, AG, GG of one SNP, and the A allele was the risk allele, the genetic score for the SNP was 2, 1, and 0. Polygenetic risk scores of the best model (PRSBM) were computed by the summation of risk allele scores in each SNP in the PRSBM. According to the PRSBM, the participants were divided into low-PRSBM, medium-PRSBM, and high-PRSBM, corresponding to the low-risk, medium-risk, and high-risk groups of genetic impacts. A high score of the PRSBM indicated a higher number of risk alleles in the SNPs in the best gene-gene interaction model.

Assessment of Food and Nutrient Intake by Using the Semiquantitative Food Frequency Questionnaire
Dietary intake was estimated using an SQFFQ developed and validated in the KoGES [25]. This questionnaire requested information regarding the participants’ consumption of food items. The SQFFQ included 106 food items, and they were used to calculate the intake of 23 nutrients from the Korean food composition table [25]. Average daily nutrient intake was calculated from the food intake measured by SQFFQ using the Computer-Aided Nutritional Analysis Program (CAN Pro) 3.0, a nutrient database developed by the Korean Nutrition Society. Intakes of carbohydrate, protein, and fat were calculated as energy percent, and they were categorized into 2 groups by 65, 13, and 15 energy percent, respectively, and they corresponded to about 25, 75, and 70 percentiles of Korean adults represented in KoGES and KNHANES [26].

Statistical Analyses
Statistical analysis was performed using PLINK version 2.0 (http://pngu.mgh.harvard.edu/~purcell/plink/) for GWAS and SAS (version 9.3; SAS Institute, Cary, NC, USA) for other statistical analyses. Proc univariate were used to check the normality of the variables, and they all had a normal distribution. Statistical differences of continuous variables between ARC and control groups were compared by analysis of covariance after adjusting covariates. Those of the categorical variables were determined by the χ² test. The association of general characteristics with ARC incidence was
assessed through logistic regression analysis after adjusting for covariates including residence area, sex, age, levels of income and education, BMI, habits of smoking and alcohol, the amount of daily energy intake, dairy product consumption, physical activity, and medication for dermatitis and arthritis [27].

For the descriptive statistics, the numbers and percentages of categorical variables such as gender, education, income, exercise, and smoking status and alcohol and coffee intake were calculated according to the three groups of PRSBM (low-, middle-, and high-PSBM groups). Frequency distributions of the classification variables were analyzed using the chi² test. Means and standard deviations were calculated for the continuous variables according to the PRSBM categories. The significant differences were determined by one-way analysis of variance with or without the adjustments for covariates. Multiple comparisons among the groups were determined with the Tukey test.

The association of PRSBM in the best model with ARC risk was evaluated using logistic regression analysis after adjusting for covariates to exclude the effect of confounding factors. ARC risk was assessed as the odds ratios (ORs) and 95% confidence intervals (CI) using low-PSBM as a reference. The variables were divided into two groups with the cutoff points to analyze their associations with PRSBM for ARC risk in logistic regression analysis. The variables’ cutoffs were mainly 70th percentiles or known values such as dietary recommended intake of energy intake. The logistic regression analysis was conducted considering two adjusted models. The first model included adjustment for residence area, sex, age, BMI, education, and income. The second model included adjustments for residence area, sex, age, BMI, education and income levels, smoking and drinking status, daily energy intake, dairy product consumption, physical activity, and medication for arthritis and dermatitis. The covariates were selected because they are known to influence ARC risk. Arthritis and dermatitis medicine intakes were used to adjust taking corticosteroids for treating dermatitis and arthritis since corticosteroid intake is involved in cataract etiology. According to the dietary intake, unadjusted and adjusted ORs and 95% CI for ARC risk compared to the low-PSBM were calculated.

We divided the participants into two groups with high or low intake using the classification criteria mentioned above to determine the interaction between the PRSBM and each lifestyle factor, including intake of fat, carbohydrate, protein, kimchi, milk, dairy products, and alcohol. The interaction between PRSBM and lifestyles was examined using two-way analysis of variance with PRSBM and lifestyles as the main effects with their interaction terms, and the model also included covariates. p value <0.05 indicated statistical significance.

Results

Socioeconomic and Lifestyle Characteristics of the Participants

Of 40,262 participants in the hospital-based cohort, 1,972 (5.2%) had ARC. The adjusted mean of age was higher in the cataract group than the noncataract group, and men had a lower incidence of ARC than women. The covariates to calculate adjusted means were as follows: residence area, surveyed year, BMI, gender, age, the status of smoking and alcohol, education levels, income, job, energy intake, and medication for arthritis and dermatitis. BMI was not significantly different between the two groups (Table 1). However, adjusted ORs indicated that age and BMI were positively associated with ARC incidence by 4.2- and 1.4-fold, respectively. Adjusted means of serum glucose levels and hemoglobin A1c levels were higher in the cataract group than the noncataract group. Adjusted ORs for serum glucose and HbA1c were also elevated by 1.4- and 1.6-fold in the cataract group, respectively (Table 1). ARC incidence was lower in the participants with higher education (p < 0.001), higher income (p < 0.001), moderate alcohol drinking (p < 0.001), and high coffee intake (p < 0.01) than those with lower education, lower income, heavy exercise, no alcohol, and coffee intake without any adjustment. However, adjusted ORs for cataract incidence did not show any association with education, income, exercise, and alcohol and coffee drinking after adjusting for residence area, survey year, BMI, gender, age, levels of education and income, the status of smoking and alcohol, job, and energy intake. The cataract incidence was lower with higher daily energy and kimchi intake by 0.90- and 0.82-fold, respectively, although the adjusted means of daily energy and kimchi intake were not significantly different between the cataract and noncataract groups (Table 1). High carbohydrate (≥65 energy %), fat (≥15 energy %), and protein (≥13 energy %) intakes were associated with cataract incidence by 1.37-0.79-, and 0.84-fold, respectively (Table 1). The cutoffs of carbohydrates, fat, and protein intake were based on the 75th percentiles of Korean intake [28].

Characteristics of 10 Selected Genetic Variants Related to Lactose and Galactose Metabolism

According to the candidate gene-based approach, ten SNPs were selected to be significantly associated with ARC risk among the genes related to lactose and galactose metabolism (online suppl. Fig. 1S; for all online suppl. material, see www.karger.com/doi/10.1159/000521548). The selected SNPs and their characteristics are shown in Table 2. The minor alleles of SLC2A1, ST3GAL3, LCT, and GALNT5 had a negative association with ARC (ORs = 0.8646–0.9033) after adjusting for cataract risk factors, whereas those of ST6GAL1, GALNT17, CSGALNACT1, GALNTL4, B4GALT6, and A4GALT had a positive association (ORs = 1.111–1.256; Table 2) (online suppl. Table 1S). All SNPs met the criteria of MAF (≥0.05%) and Hardy-Weinberg equilibrium (HWE; p > 0.05) criteria (Ta-
ble 2). Each selected SNP exhibited a low OR value (0.8646–1.256), indicating the weak association between each SNP and ARC. Therefore, a stronger association of ARC with genetic variants related to lactose and galactose metabolism needed to be found. We hypothesized that polygenetic risk scores of the genetic variant-genetic variant interactions might be better to show the impact of lactose- and galactose-related genes on ARC risk.

Table 1. Socioeconomic and lifestyle characteristics of the participants according to ARC incidence

|                | Non-ARC (n = 38,290) | ARC (n = 1,972) | Adjusted OR (95% CI) |
|----------------|---------------------|----------------|---------------------|
| Age, years     | 57.8±5.5            | 61.8±5.4***    | 4.161 (3.722–4.652)*** |
| Gender (male)  | 14,001 (36.6)       | 805 (40.8)***  | 1.081 (0.971–1.204)  |
| BMI, kg/m²     | 19.8±2.8            | 20.1±2.9       | 1.112 (1.006–1.230)* |
| Fasting serum glucose, mg/dL | 96.5±20.2 | 100.4±25.6***  | 1.385 (1.250–1.535)*** |
| HbA1c          | 5.79±0.7            | 5.91±0.9***    | 1.636 (1.388–1.928)*** |
| Blood pressure | 1.169 (4.17)        | 803 (6.56)***  | 1.454 (1.314–1.609)*** |
| Income (per month) |            |                |                     |
| <USD 1,000/yr  | 4,862 (13.5)        | 433 (22.9)***  |                     |
| USD 1,000–2,000| 8,791 (24.4)        | 530 (28.7)     |                     |
| USD 2,000–4,000| 14,849 (41.3)       | 641 (34.7)     |                     |
| >USD 4,000     | 7,480 (20.8)        | 254 (13.8)     |                     |
| Exercise       | No                  | Yes            |                     |
| No             | 16,593 (43.5)       | 808 (41.2)*    | 1.082 (0.979–1.196) |
| Yes            | 21,589 (41.2)       | 1,153 (58.8)   |                     |
| Smoking        | No                  | Yes            |                     |
| No             | 35,113 (91.7)       | 1,831 (92.9)   |                     |
| Former smoking | 1,994 (5.21)        | 90 (4.57)      | 1.069 (0.787–1.452) |
| Current smoking| 1,168 (3.1)         | 50 (2.54)      |                     |
| Alcohol intake | No                  | Yes            |                     |
| No             | 23,123 (60.4)       | 1,290 (65.4)***| 0.901 (0.783–1.036) |
| Medium (3–16 g/day) | 6,539 (17.1) | 332 (16.8) | 0.970 (0.878–1.071) |
| High (≥16 g/day) | 13,846 (36.2) | 643 (32.6) |                     |
| Energy intake, kcal | 1,713±531 | 1,691±516 | 0.902 (0.814–0.999)* |
| CHO percent intake | 70.5±20.8 | 71.0±20.0*** | 1.368 (1.142–1.639)*** |
| Fat percent intake | 13.5±8.7 | 13.1±8.0** | 0.786 (0.681–0.907)** |
| Protein percent intake | 13.2±5.8 | 13.1±5.6* | 0.840 (0.722–0.979)* |
| Dairy product intake (0.75 serving/day) | 113±133 | 115±142 | 0.997 (0.895–1.111) |
| Ca intake, mg/day | 442±256 | 443±265 | 0.991 (0.875–1.121) |
| Kimchi, g/day | 138±113            | 130±142        | 0.817 (0.732–0.913)*** |

The values represent adjusted means±standard deviations or the number of the subjects (percentage of each group). BMI, body mass index. Statistical significance was set at *p < 0.05, **p < 0.01, and ***p < 0.001. 1 By ANCOVA after adjusted covariates. 2 Adjusted ORs by logistic regression analysis with adjustment of covariates such as the location of residency, examination year, BMI, gender, age, the status of smoking and alcohol, levels of education and income, job, energy intake, and medication for arthritis and dermatitis in logistic regression models. 3 By χ² test. In logistic regression analysis, each parameter needed to be categorized into the low and high groups with cutoff point that was as following: 4 <60 years old, 5 <25 kg/m² BMI, 6 <126 mL/dL fasting serum glucose or medication for diabetes, 7 <6.5% HbA1c or medication for diabetes, 8 <130/90 mm Hg for blood pressure, 9 <college, formal and current smoking, 10 <USD 2,000/month, 11 nonsmoking, 12 <20 g/day alcohol drinking, 13 <3 g/day coffee drinking, 14 <estimated energy intake, 15 <65 energy % carbohydrate intake, 16 <15 energy % fat, 17 <13 energy % protein, 18 <0.75 serving/day of dairy products adding milk, yogurt, and cheese, 19 <500 mg/day Ca intake, and 20 <100 g/day kimchi intake.
Table 2: The characteristics of the ten genetic variants of genes related to lactose and galactose metabolism in age-related cataract risk used for the generalized multifactor dimensionality reduction analysis

| Chr | SNP  | Gene       | Feature   | Location | MAF | OR | Location 95% CI | p value for OR | Genes     |
|-----|------|------------|-----------|----------|-----|----|-----------------|--------------|-----------|
| 1   | rs10831608 | ST6GAL1    | Intron    | 13.276548 | 0.0383 | 1.006 | 1.00-1.04       | 0.8949       | LCT       |
| 2   | rs1667288  | ST3GAL3    | Intron    | 13.276548 | 0.0384 | 1.006 | 1.00-1.04       | 0.8949       | LCT       |
| 3   | rs2268536  | GALNT5     | Intron    | 13.276548 | 0.0385 | 1.006 | 1.00-1.04       | 0.8949       | LCT       |
| 4   | rs9623659  | GALNTL4    | Intron    | 13.276548 | 0.0386 | 1.006 | 1.00-1.04       | 0.8949       | LCT       |
| 5   | rs17058752 | CSGALNACT1 | Intron    | 13.276548 | 0.0387 | 1.006 | 1.00-1.04       | 0.8949       | LCT       |
| 6   | rs3729548  | GALNTL4    | Intron    | 13.276548 | 0.0388 | 1.006 | 1.00-1.04       | 0.8949       | LCT       |
| 7   | rs6728956  | GALNT17    | Intron    | 13.276548 | 0.0389 | 1.006 | 1.00-1.04       | 0.8949       | LCT       |
| 8   | rs10831608 | ST6GAL1    | Intron    | 13.276548 | 0.0390 | 1.006 | 1.00-1.04       | 0.8949       | LCT       |
| 9   | rs1994788  | GALNTL4    | Intron    | 13.276548 | 0.0391 | 1.006 | 1.00-1.04       | 0.8949       | LCT       |
| 10  | rs10831608 | ST6GAL1    | Intron    | 13.276548 | 0.0392 | 1.006 | 1.00-1.04       | 0.8949       | LCT       |

The Best Model for the Association with ARC Assessed by Gene-Gene Interaction Using GMDR

Since each SNP selected for ARC was weakly associated with ARC, a gene-gene interaction study was conducted to better explain the ARC genetic effect by combining multiple genetic variants. The GMDR approach is used to identify the gene-gene interactions through multifactor dimensionality reduction [29]. GMDR provided multiple sets of the best SNP combinations with gene-gene interaction effects. Although SNPs involved in lactose metabolism were identified, 10 SNPs were considered core SNPs to explain ARC risk. In the process of generating the best model using GMDR, TRBA, TEBA, and CVC were used for validating the gene-gene interaction model [30, 31]. The best set of gene-gene interactions was selected by minimum prediction error to represent high balance accuracy and maximum CVC values [30, 31]. TRBA calculated the accuracy of the prediction model of gene-gene interaction with 80% of the participants after training the program [19]. TEBA represented the accuracy of the prediction model in the rest of the participants. A sign test was used to analyze the statistical significance of TEBA. CVC indicated that the number of genetic variants had cross-validation, and the maximum was 10 out of 10 genetic variants. GMDR was conducted with the risk alleles of the 10 SNPs adjusting for age, gender, and BMI or age, gender, survey year, residence area, BMI, education, and income. The best model for ARC with gene-gene interaction was explored with the selected 10 SNPs by GMDR in consideration of CVC (p value for the GMDR model was 0.001 in both models, and CVC was 10/10) (Table 3). Model 9 plus GALNTL4_rs10831608 showed the highest training accuracy (0.7962) and CVC (10/10). The best model included SLC2A1_rs3729548, ST3GAL3_1097407, beta-Galactoside alpha-2,3-sialyltransferase 3; LCT, lactase; GALNT5, polypeptide N-acetylgalactosaminyltransferase 5; ST6GAL1; ST6 beta-galactoside alpha-2,6-sialyltransferase 1; GALNT17, polypeptide N-acetylgalactosaminyltransferase 17; CSGALNACT1, chondroitin sulfate N-acetyl-galactosaminyltransferase 1; GALNTL4, alpha 1,4-galactosyltransferase. Chromosome.

Adjusted ORs for ARC according to the PRSBM

The PRSBM was calculated by summing the number of risk alleles, and it was divided into three categories: low-PRSBM, 0–7; medium-PRSBM, 8–9; and high-PRSBM, ≥10. High-PRSBM indicated a genetically higher incidence of ARC. The ARC incidence was higher in the high-PRSBM group by 1.874- and 2.140-folds in models 1 and 2 compared to the low-PRSBM (Table 4). Models 1 and 2 included different adjusting covariates. Covariates of model 1 were age, gender, residency location, survey year, BMI, lev-
els of income and education, and job. Those of model 2 were age, gender, location of residency, examination year, the status of smoking and drinking, levels of income and education, job, energy intake, amount of physical activity, hypertension, dairy products, percent intake for carbohydrate and fat, and medication of arthritis and dermatitis. The PRSBM groups did not significantly associate metabolic syndrome and its individual components including BMI, waist circumferences, fasting serum glucose concentrations plus diabetic drug intake, \( <6.5\% \text{ HbA1c} \) plus diabetic drug intake, \( <130/90 \text{ mmHg} \) in systolic blood pressure/diastolic blood pressure, \( <160 \text{ mg/dL} \) for plasma LDL, \( >40 \text{ and } 50 \text{ mg/dL} \) in men and women, respectively, for plasma HDL, and \( <150 \text{ mg/dL} \) for plasma TG. Statistical significance from low-PRSBM in logistic regression analysis was set at \( * \ p < 0.05, ** \ p < 0.01, \) and *** \( p < 0.001. \)

**Table 3.** GMDR results of multilocus interaction with genes related to ARC

| GMDR models | Adjusted for age, gender, BMI | Adjusted for age, gender, survey year, residence area, BMI, education, income |
|-------------|--------------------------------|--------------------------------------------------------------------------------|
| Model 1 GALNT5_rs6728956 | TRBA\(^1\) 0.5215, TEBA\(^2\) 0.4988, p value\(^3\) 4 (0.828), 5/10 | TRBA\(^1\) 0.5216, TEBA\(^2\) 0.4957, p value\(^3\) 3 (0.945), 4/10 |
| Model 1 plus ST6GAL1_rs2268536 | 0.5314, 0.5041, 8 (0.055), 6/10 | 0.5313, 0.5041, 8 (0.0547), 6/10 |
| Model 2 plus B4GALT6_rs1667288 | 0.5431, 0.4957, 5 (0.623), 2/10 | 0.5431, 0.4966, 5 (0.623), 2/10 |
| Model 3 plus GALNT17_rs17058752 | 0.5614, 0.5277, 9 (0.011), 4/10 | 0.5613, 0.5276, 9 (0.011), 4/10 |
| Model 4 plus SLCL2A1_rs3729548 | 0.5863, 0.5280, 9 (0.011), 7/10 | 0.5862, 0.5271, 8 (0.005), 7/10 |
| Model 5 plus ST3GAL3_rs3791047 | 0.6179, 0.5310, 8 (0.055), 7/10 | 0.6178, 0.5295, 8 (0.055), 7/10 |
| Model 6 plus A4GALT_rs9623659 | 0.6606, 0.5223, 8 (0.055), 7/10 | 0.6605, 0.5223, 8 (0.055), 6/10 |
| Model 7 plus LCT_rs2304371 | 0.7096, 0.5212, 7 (0.172), 10/10 | 0.7096, 0.521, 7 (0.172), 10/10 |
| Model 8 plus CSGALNACT1_rs1994788 | 0.7580, 0.5232, 7 (0.172), 10/10 | 0.7581, 0.5237, 7 (0.172), 10/10 |
| Model 9 plus GALNTL4_rs10831608 | 0.7962, 0.5250, 10 (0.001), 10/10 | 0.7962, 0.5261, 10 (0.001), 10/10 |

BMI, body mass index. \(^1\) Trained balanced accuracy; \(^2\) test balance accuracy; \(^3\) sign rank (p value for the significance of the GMDR model by the sign test with adjusting for covariates designated in the table); \(^4\) cross-validation consistency.

**Table 4.** Adjusted odds ratios for ARC according to the PRSBM for gene-gene interaction after covariate adjustments

| Model | Low-PRSBM \((n = 14,420)\) | Medium-PRSBM \((n = 21,641)\) | High-PRSBM \((n = 4,201)\) | Medium-PRSBM \((n = 21,641)\) | High-PRSBM \((n = 4,201)\) |
|-------|-----------------|-----------------|-----------------|-----------------|-----------------|
| ARC   | 1               | 1.411 (1.219–1.634) | 1.874 (1.507–2.332)** | 1.369 (1.135–1.650) | 2.140 (1.622–2.822)** |
| BMI\(^1\) | 1               | 0.973 (0.912–1.063) | 0.984 (0.912–1.063) | 0.956 (0.897–1.051) | 0.946 (0.852–1.051) |
| Metabolic syndrome | 1               | 1.027 (0.964–1.095) | 0.931 (0.838–1.034) | 1.028 (0.964–1.096) | 0.926 (0.832–1.029) |
| Waist circumference\(^2\) | 1               | 1.065 (0.994–1.141) | 1.115 (0.997–1.247) | 1.085 (0.986–1.195) | 1.135 (0.971–1.327) |
| Fasting serum glucose\(^3\) | 1               | 1.004 (0.935–1.077) | 1.002 (0.893–1.124) | 0.953 (0.811–1.119) | 1.033 (0.797–1.339) |
| HbA1c\(^4\) | 1               | 1.001 (0.907–1.104) | 1.066 (0.911–1.247) | 1.008 (0.921–1.114) | 1.080 (0.921–1.268) |
| Hypertension\(^5\) | 1               | 1.021 (0.918–1.136) | 1.055 (0.890–1.250) | 0.964 (0.835–1.114) | 1.034 (0.829–1.300) |
| Plasma LDL cholesterol\(^6\) | 1               | 1.132 (0.993–1.291) | 1.040 (0.843–1.283) | 1.050 (0.888–1.243) | 0.997 (0.763–1.304) |
| Plasma HDL cholesterol\(^7\) | 1               | 0.990 (0.942–1.040) | 0.946 (0.873–1.026) | 0.968 (0.904–1.037) | 0.949 (0.848–1.062) |
| Plasma TG\(^8\) | 1               | 1.015 (0.910–1.132) | 0.891 (0.747–1.064) | 1.024 (0.884–1.187) | 0.806 (0.634–1.026) |

Data were expressed as odds ratios and 95% confidence intervals. PRSBM from 10 SNPs were divided into three categories (0–7, 8–9, and >10) corresponding to the low-risk, medium-risk, and high-risk groups, respectively. Low-PRSBM was the reference for both model 1 and model 2. Model 1: adjusted for sex, age, location of residence, examination year, BMI, levels of income and education, and job. Model 2: adjusted for covariate of model 1 and job, hypertension, activity, energy, milk, percent intake of fat and carbohydrate, and medication for arthritis and dermatitis. The cut-off points of the parameters were: \(^1\) <25 kg/m\(^2\) BMI, \(^2\) <90 and 85 cm in men and women, respectively, for waist circumferences, \(^3\) <126 mg/dL fasting serum glucose concentrations plus diabetic drug intake, \(^4\) <6.5% HbA1c plus diabetic drug intake, \(^5\) <130/90 mmHg in systolic blood pressure/diastolic blood pressure, \(^6\) <160 mg/dL for plasma LDL, \(^7\) >40 and 50 mg/dL in men and women, respectively, for plasma HDL, and \(^8\) <150 mg/dL for plasma TG. Statistical significance from low-PRSBM in logistic regression analysis was set at \* \( p < 0.05, ** \( p < 0.01, \) and *** \( p < 0.001. \)
Interaction of PRSBM for the Best Model and Nutrient and Food Intakes in ARC Incidence

ARC incidence exhibited a significant interaction with the intake of carbohydrates, alcohol, dairy product (milk, yogurt, and cheese), and kimchi at the significance level of 0.05 (Table 5). Other dietary factors, including fruits, vegetables, and nuts, did not interact with PRSBM for ARC risk (data not shown). High carbohydrate intake (≥65 energy %), but not low carbohydrate intake (<65 energy %), was associated with ARC by 1.803-energyfold. PRSBM was positively associated with ARC risk in both low and high intake of dairy products (milk, yogurt, and cheese) and kimchi (Table 5). Only a low alcohol intake showed a positive association of 1.803-energyfold with ARC (Table 5). In Figure 1a, ARC risk was much higher in the high-PRSBM group than the low-PRSBM in the low-carbohydrate category, but there was no significant association with PRSBM with ARC risk in the high-carbohydrate category. Interestingly, ARC risk was higher in subjects with the high-PRSBM than those with the low-PRSBM with low dairy product consumption (Fig. 1b). In the high dairy product category, subjects with high-PRSBM had a higher ARC risk, but they had a lower risk in the low dairy product category. Kimchi intake showed a similar trend to dairy product intake. In low kimchi intake, ARC risk was higher in the high-PRSBM than the low-PRSBM (Fig. 1c). The trend for higher ARC risk in the low kimchi intake was similar in the high-kimchi intake, but the ARC risk in the high-PRSBM was lower in the high-kimchi category than the low-kimchi category. The participants with high-PRSBM had a high ARC risk only in low alcohol intake but not moderate alcohol intake (Fig. 1d). With moderate alcohol intake (<20 g/day), PRSBM did not associate with ARC incidence.

Discussion

In this study, we established a PRSBM for cataracts using GMDR and evaluated whether they are related to the incidence of cataracts. We also evaluated the gene-nutri-
ent interaction by analyzing the nutritional effect on the risk of ARC by PRSBM. We observed that the cataract risk was 2.1 times higher in the high-PRSBM than in the low-PRSBM after adjusting for covariates. The genetic variants in the PRSBM were selected from the genes related to galactose metabolism since the disturbance of galactose metabolism, especially GALK, is associated with ARC risk. The A198V mutation of GALK is prevalent in Asians (4.1% in Japanese and 2.8% in Koreans) but not Caucasians and Blacks [5]. Genetic variants in GALK had no association with ARC in the present study. ARC’s genetic risk was modified by nutrients, including consuming carbohydrates and foods such as alcohol, dairy products, kimchi, and alcohol intake. Specifically, ARC’s genetic risk significantly increased in groups with high carbohydrates, low alcohol drinking, low dairy product consumption, and low kimchi consumption.

The origin of galactose is mostly from lactose derived from dairy products, mainly milk. Galactose is converted to glucose, primarily in the liver and brain [32]. Three enzymes that convert galactose to glucose-6-phosphate are encoded by the GALK, GALT, and phosphoglucomutase (PGM) genes. UDP-galactose and UDP-glucose interconversion require the enzyme encoded by the GALE gene UDP-glucose pyrophosphorylase 2 encoded by UGP2. Other genes related to galactose and lactose metabolism were also considered possible links to ARC, and 10 SNPs were selected for gene-gene interaction since no individual SNP had a significant impact on ARC risk. In GMDR, the best model included all 10 SNPs, and PRSBM had much higher adjusted ORs (2.14) for ARC risk. Interestingly, the PRSBM had interactions with dietary intakes that lower ARC incidence in Koreans.
Low dairy product consumption (<150 g/day) was found to increase the genetic risk of ARC, which increased in the participants with high-PRSBM in the present study. These results suggest that low dairy product intake might reduce the expression of the selected genes and that the galactose metabolism might be increased with high-PRSBM when galactose metabolism is needed. It remains controversial whether dairy product consumption is a risk factor for cataract formation [33]. An experimental animal study provided quantitative evidence that excessive milk intake aggravates ARC in rats due to oxidative damage caused by increased reactive oxygen species [34]. However, a recent longitudinal epidemiologic study reported no significant association between dairy product consumption and the risk of incident cataracts in 5,860 subjects [35]. The finding in the present study suggests that the cataractogenic action of lactose in dairy products, which is linked to the ARC genetic risk, could be exaggerated in low dairy product intake groups, possibly due to less expression of the related genes. Therefore, a low intake of dairy products may reduce the expression of genes encoding lactose and galactose enzymes, reducing the activity of those enzymes in people with high-PRSBM, resulting in reduced galactose metabolism and a concomitant increased ARC risk. Therefore, the continuous and proper amount of dairy product intake may normalize the gene expression related to lactose and galactose metabolism, and ARC might be lessened, especially in adults with high-PRSBM. However, this hypothesis has not been tested in human trials and needs confirmation.

ARC risk was highly positively associated with a high carbohydrate intake (≥65 energy %) by 1.37-fold in the present study. It was a consistent finding given that high carbohydrate consumption was regarded as a risk factor for cataracts. A cross-sectional epidemiologic study with 1,609 subjects reported that nuclear cataract was associated with total carbohydrate intake, although this study failed to show a dose-dependent relationship [36]. A recent meta-analysis confirmed that the risk of ARC was 1.18 and 1.15 times higher for the highest versus the lowest category of carbohydrate intake and glycemic index [37]. The effect of high carbohydrate intake on ARC risk in Koreans can explain this finding; an environmental risk factor could attenuate the impact of the genetic risk over the ARC risk. Furthermore, only the group with high carbohydrate intake showed a significant genetic risk of developing cataracts. Persons with high-PRSBM could reduce their risk of cataracts by decreasing carbohydrate intake (<65 energy %).

Alcohol drinking is a well-known risk factor for cataracts. Alcohol drinking also modifies the genetic risk factor of cataract development. However, alcohol intake did not play as a risk factor for ARC incidence in the present study. A meta-analysis involving a total of 119,706 participants demonstrated that relative risks in heavy drinkers, which were defined by >20 g/day of alcohol intake versus non-drinkers, were 1.25 for cataract surgery, 1.06 for cortical cataracts, and 1.26 for nuclear cataracts, respectively. These association between alcohol consumption and cataracts is not significant, consistent with the present study [38]. On the other hand, alcohol intake had an interaction with PRSBM in the present study. We found that the ARC risk by PRSBM was significant only in groups with low alcohol consumption, but more than 20 g/day alcohol intake was not associated with ARC’s genetic risk. It suggested that moderate alcohol intake had a tendency to lower risk in the participants with high-PRSBM who elevated ARC incidence in low alcohol intake. The possible mechanism is that alcohol metabolism generates free radicals, aggregates lens proteins, and subsequently forms cataracts, and the genetic variants in the PRSBM may be involved in removing free radicals and preventing the aggregation of lens proteins by alcohol [39]. This strong environmental risk factor of alcohol consumption may attenuate the genetic risk factor’s effect on the overall risk of cataract development.

On the other hand, physical exercise did not exhibit a significant association with ARC in the present study. It may be related to spending more time outside with greater exposure to UV light from the sun during exercise. A recent meta-analysis has demonstrated that increased physical activity is associated with reduced ARC risk by 10%, related to improving oxidative stress [40]. Although a positive relationship between ARC and physical activity has been reported [40], ambient UV light exposure increases ARC risk [41]. Therefore, UV light exposure needs to be avoided during outdoor physical activity to prevent ARC in older persons.

Interestingly, the consumption of kimchi, the Korean traditional probiotic vegetable food, significantly modified the genetic risk effect on ARC in this study. The group with low kimchi intake had a higher odds ratio for genetic risk of cataract of 1.254 and 1.622 for medium- and high-PRSBM, respectively, whereas the group with the intake of kimchi over 100 g/day showed that OR for genetic risk of cataract was 1.049 and 1.639 for medium- and high-PRSBM, respectively. Although ARC’s genetic risk showed significantly different patterns, with the medium intake being adequate for decreasing ARC risk but not high intake of Kimchi consumption, we could not
draw any meaningful clinical conclusion due to the small effect size and inconsistent pattern.

This study has limitations as well as strengths. This study’s strength includes a relatively large number of participants aged >50 years old, including 1,971 subjects having ARC and 38,290 participants having no cataracts, which could increase the robustness of the present study. Our study also has several limitations. First, the cataract incidence was assessed by using a questionnaire of cataract diagnosis by a physician, which may not be as accurate as a slit-lamp examination of the lens due to the recall bias. The second limitation is that the current study has a cross-sectional design, which makes inferring causality difficult. However, it is unlikely that the presence of ARC changes the eating patterns. The third limitation was no duplication study in another cohort since no cohort analyzing genetic variants included sufficient ARC patients to conduct a duplication study. The fourth limitation is that the SQFFQ method used in this study for nutrition evaluation may have some shortfalls. It might overestimate the nutrition intakes at individual levels since it relies on the participant’s memory to recall past intake. However, SQFFQ is widely used in many other studies after developing and validating the KoGES [25, 42, 43].

In conclusion, ARC incidence might be positively associated about 2-fold with PRSBM involved in lactose and galactose metabolism and the nutrition-gene interaction for the ARC risk. The lactose- and galactose-related genetic impact on ARC risk interacted with carbohydrate intake, alcohol drinking, dairy products, and kimchi intake. Adults aged >50 years with high-PRSBM might benefit from consuming dairy products and kimchi daily and avoiding a high carbohydrate diet (≥65 energy %) associated with a lower incidence of ARC. Given that cataract is the most common cause for low vision, further study is warranted to assess the utility of nutrition-gene interactions to modify and decrease cataract risk.

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Statement of Ethics

The study was conducted according to the Declaration of Helsinki’s guidelines and approved by the Institutional Review Board of the Korean National Institute of Health (1041231-190902-BR-099-01). We additionally obtained approval from the Institutional Review Board of Hoseo University (1041231-150811-HR-034-01). We obtained written informed consent from all participants involved in the study.

Conflict of Interest Statement

The authors declare no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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Author Contributions

S.P. and D.J. participated in the experimental design, analyzed the data, and wrote the manuscript. S.K. designed the research and analyzed the data. S.P. and S.K. had primary responsibility for the final content. All authors listed as authors have substantially contributed to the work and seen and approved the submitted version.

Data Availability Statement

All data generated or analyzed during this study are included in this article. Further enquiries can be directed to the corresponding author.

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