INTRODUCTION

Diabetes mellitus (DM), one of the most common chronic disorders in children and adolescents, is characterized by hyperglycemia and results from impaired insulin secretion, insulin action, or both (1). Although type 1 diabetes mellitus (T1DM) is the most frequent form of diabetes among young people, various reports have indicated that the incidence of childhood type 2 diabetes mellitus (T2DM) has increased and the number of affected individuals continuous to increase (2,3). The growing incidence of T2DM reflects the epidemic of childhood obesity and is becoming a grave public health concern. Because T2DM can be asymptomatic at diagnosis and requires tight glycemic control to delay the onset of chronic vascular complications, screening for T2DM in children was approved by the American Diabetes Association (ADA) and the American Academy of Pediatrics (4,5).

Since 1974, several countries, including Korea, have performed routine urine screening at school for early detection of chronic renal disease and DM. Although urinalysis screening is non-invasive and inexpensive, it is a poor screening test for chronic kidney disease (6). However, recent Japanese studies have indicated a high incidence of childhood T2DM detected by a school urine glucose screening program (7,8). A study based on the Chinese Foundation of Health that was conducted from 1993 to 1999 in Taiwan reported that the incidence of childhood DM of all types increased in puberty and was higher in girls than in boys (9). In Korea, large-scale urine screening for asymptomatic glucosuria concomitant with asymptomatic hematuria and proteinuria has been conducted in schoolchildren since 1998 (10). More than 4 million students have participated in this annual screening program, and glucosuria was detected in approximately 0.072% of children who were screened between 2007 and 2011 (11). However, studies that analyze data about schoolchildren with DM detected through the urine glucose screening program in Korea have not yet been conducted.

The aim of this study was to evaluate the prevalence of glucosuria from 2010 to 2013 in schoolchildren and adolescents in Jeonbuk province and the characteristics of DM as detected using the school urine glucose screening program.
MATERIALS AND METHODS

Subjects and methods

Between March 2010 and September 2013, 707,238 schoolchildren participated in a large-scale school urine screening program in the Jeonbuk province area (Table 1). Each year, second, third, fifth, and sixth grade students in elementary schools, and second and third grade students in middle and high schools participated in the school screening program regardless of known diabetes or renal glucosuria. A urinalysis of morning spot urine specimens was performed using glucose oxidase tape (UriScan; YD Diagnostics Corp., Yongin, Korea). If the result of the first urine test was positive (urine glucose level > 5.6 mmol/L [100 mg/dL]), a second urine test was required within 2 weeks. If the result of the second test was also positive for glucosuria, the student was advised to visit Chonbuk National University Children’s Hospital for further evaluation. Subjects with known DM or previous renal glucosuria diagnoses were excluded from this study.

Data that included anthropometric measures and medical history were collected at the patient’s first hospital visit. Height and weight measurements were converted to body mass index (BMI) percentiles according to the 2007 Korean National Growth Charts (12). Overweight was defined as an age- and sex-specific BMI ≥ 85th and < 95th percentiles, respectively, and obesity was similarly defined as a BMI ≥ 95th percentile. Blood and urine samples were obtained in the morning after 12 hours of fasting, and an oral glucose tolerance test (OGTT) was performed to confirm DM. For the OGTT, 1.75 g glucose/kg (maximum, 75 g) were used, and the World Health Organization (WHO) criteria for the diagnosis of glucose intolerance were followed. According to the WHO criteria based on the primary OGTT (13), DM was defined as a 2-hour post-load glucose level ≥ 11.1 mmol/L (200 mg/dL) or fasting glucose level ≥ 7.0 mmol/L (126 mg/dL), impaired glucose tolerance (IGT) was defined as a 2-hour post-load glucose level of 7.8–11.0 mmol/L (140–199 mg/dL), and normal glucose tolerance (NGT) was defined as a 2-hour post-load glucose level < 7.8 mmol/L (< 140 mg/dL). Fasting blood samples were drawn from the cubital vein. Fasting plasma glucose (FPG), total and high-density lipoprotein (HDL) cholesterol, and triglycerides were measured using enzymatic methods (Roche Diagnostics, Mannheim, Germany). Serum insulin and C-peptide levels were measured using immunoradiometric assay (IRIMA) with commercial kits (DIAsource Immuno-ASSay S.A., Louvain-La-Neuve, Belgium for insulin; Institute of Isotopes Co., Ltd., Budapest, Hungary for C-peptide). Glicated hemoglobin (HbA1c) was measured via high-performance liquid chromatography (Bio-Rad Variant II, Richmond, CA, USA). The serum anti-glutamic acid decarboxylase antibody (GADA) concentration was measured by IRIMA (RSR Ltd., Cardiff, UK) and GADA positivity was defined as serum GADA concentration ≥ 1 U/mL, in accordance with the manufacturer’s reference range (> 2 standard deviation [SD] among healthy patients). If a child exhibited GADA positivity, the islet cell antibody (ICA) was measured by using radioimmunoassay with an IA-2 antibody Cosmic kit (RSR Ltd.). Insulin sensitivity was estimated using the previously validated homeostasis model assessment of insulin resistance (HOMA-IR) index (14). Childhood T2DM is defined based on negative test results for diabetes-related autoantibodies, a substantial residual insulin secretory capacity, and evidence of insulin resistance.

Table 1. Annual numbers of students who underwent urine glucose screening and newly identified cases of diabetes

| Characteristics          | Students examined | Glucosuria on second urine test | OGTT performed | DM by OGTT |
|--------------------------|------------------|-------------------------------|----------------|------------|
| Year                     |                  |                               |                |            |
| 2010                     | 183,039          | 132 (0.072)                   | 39             | 16         |
| 2011                     | 181,223          | 141 (0.077)                   | 27             | 5          |
| 2012                     | 176,888          | 135 (0.076)                   | 17             | 11         |
| 2013                     | 166,088          | 137 (0.082)                   | 28             | 8          |
| **Total**                | **707,238**      | **545 (0.077)**               | **110 (0.015)**| **40 (0.0056)** |
| **Age, yr**              |                  |                               |                |            |
| -                        |                  | 15.10 ± 2.32                 | 14.60 ± 2.88   | 14.52 ± 2.52 |
| **BMI, kg/m²**           |                  | 23.78 ± 4.92                 | 23.41 ± 5.11   | 25.26 ± 4.88 |
| **Obesity (BMI > 95th percentile)** | | 123 (22.6) | 34 (31.5) | 16 (40.0) |

| School level            |                  |                               |                |            |
|-------------------------|------------------|-------------------------------|----------------|------------|
| Elementary              | 313,911          | 98 (0.031)                    | 29 (0.009)     | 11 (0.0035) |
| Middle                  | 195,430          | 163 (0.083)                   | 29 (0.014)     | 13 (0.0066) |
| High                    | 197,897          | 284 (0.143)                   | 52 (0.026)     | 16 (0.0081) |

Values are presented as number (%).
OGTT = oral glucose tolerance test, DM = diabetes mellitus, BMI = body mass index.
Statistical analysis
All variables were expressed as mean ± SD values. Statistical analyses were performed using SPSS software (version 12.0, SPSS Inc., Chicago, IL, USA). Statistical significance of differences variance among 3 groups (NGT, IGT, and DM) was tested using one-way analysis of variance (ANOVA) analysis. Multivariate logistic regression models calculated adjusted odds ratio (OR) and 95% confidence interval (CI) between sex, school level, family history of T2DM, and serum lipid profiles in DM group. Statistical significance was defined as a P value < 0.05 for all clinical and laboratory data.

Ethics statement
This study was approved by the Institutional Review Board at Chonbuk National University Hospital (IRB No. 2016-09-023-001), and informed consent was exempted by the board.

RESULTS

Glucosuria prevalence
From 2010 to 2013, a total of 707,238 schoolchildren (66.4%; 313,911 elementary school students, 195,430 middle school students, and 197,897 high school students) from a school population of 1,064,999 were screened for glucosuria (Table 1). Nine-hundred twenty-seven children (0.131%) were found to exhibit glucosuria on the first urine test. Of these, 919 children (99.2%) underwent a second urine test, and 545 (0.077%) exhibited glucosuria.

The positive rate of glucosuria was higher among middle and high school students than among elementary students (0.083%, 0.143%, and 0.031%, respectively). In addition, the glucosuria prevalence was higher among girls than among boys (0.087% vs. 0.068%).

OGTT dysglycemia prevalence
Of the 545 students who tested positive for glucosuria on the second urine test, 398 were excluded because of known DM (n = 367) or renal glucosuria (n = 31) diagnoses prior to this study (11). Among the remaining 147 students, 37 (25.2%; man-to-woman ratio, 1.0:1.5; mean ± SD age, 14.2 ± 3.3 years old; BMI, 23.2 ± 4.9 kg/m²) did not visit the hospital for unknown reasons and 110 (74.8%; man-to-woman ratio, 1.0:1.3; age, 14.6 ± 3.1 years old; BMI, 23.2 ± 4.9 kg/m²) underwent a second OGTT test. Of these, 32 (35.4%; man-to-woman ratio, 1.0:1.1) exhibited OGTT dysglycemia.

Table 2. Characteristics of the 110 school children and adolescents to whom the OGTT was administered

| Characteristics                  | Total      | Sub-total | T2DM       | SPIDDM     | P value |
|----------------------------------|------------|-----------|------------|------------|---------|
| No. of subjects                  | 110        | 49 (44.5) | 21 (19.1)  | 40 (36.4)  | 0.095   |
| Age, yr                          | 14.60 ± 2.88 | 15.06 ± 2.97 | 13.64 ± 3.13 | 14.52 ± 2.52 | 14.61 ± 2.49 | 11.04 ± 2.14 | 0.164 |
| School level                     |            |           |            |            |         |
| Elementary                       | 29 (26.4)  | 9 (18.4)  | 9 (42.9)   | 11 (27.5)  | 10       |
| Middle                           | 29 (26.4)  | 11 (22.4) | 5 (23.8)   | 13 (32.5)  | 13       | 0       |
| High                             | 52 (47.3)  | 29 (59.2) | 7 (33.3)   | 16 (40.0)  | 16       | 0       |
| Sex (man:woman)                  | 47:63      | 27:22     | 7:14       | 13:27      | 1:0      | 0.030 |
| BMI, kg/m²                       | 23.41 ± 5.11 | 21.45 ± 4.27 | 24.62 ± 6.27 | 25.26 ± 4.88* | 25.27 ± 4.94 | 24.87 ± 4.94 | 0.001 |
| Family history of diabetes in 1st degree relatives | 47 (42.7) | 17 (34.7) | 8 (38.1) | 22 (55.0) | 21 (53.8) | 1 (100.0) | 0.058 |
| Obesity (BMI ≥ 95th percentile)  | 34 (31.5)  | 7 (14.6)  | 11 (52.4)  | 16 (40.0)  | 15       | 0.006 |
| Cholesterol, mg/dL               | 169.63 ± 32.50 | 159.12 ± 25.21* | 166.95 ± 27.96* | 183.48 ± 37.81* | 183.62 ± 38.29 | 178.00 ± 42.86 | 0.001 |
| HDL, mg/dL                       | 49.31 ± 12.82 | 53.51 ± 13.17* | 47.38 ± 12.14* | 45.10 ± 11.43* | 45.13 ± 11.58 | 44.00 ± 11.38 | 0.006 |
| LDL, mg/dL                       | 98.75 ± 32.08 | 87.20 ± 24.66* | 103.62 ± 29.31* | 111.20 ± 36.02* | 111.50 ± 36.46 | 119.00 ± 36.66 | 0.002 |
| Triglyceride, mg/dL              | 112.50 ± 66.38 | 87.72 ± 50.57* | 111.14 ± 65.32* | 136.05 ± 72.09* | 137.69 ± 72.27 | 72.00 ± 72.27 | 0.001 |
| HOMA-IR index                    | 5.29 ± 7.04  | 2.30 ± 1.51* | 4.41 ± 4.15* | 10.17 ± 10.22* | 10.43 ± 10.35 | 3.06 ± 10.35 | 0.000 |
| HbA1c                            | 0.000       |           |           |            |         |
| nmol/mol                         | 52 ± 25     | 36 ± 3*   | 44 ± 12*   | 76 ± 260   | 77 ± 26   | 63       |
| %                                | 6.95 ± 2.27 | 5.43 ± 0.26* | 6.21 ± 1.13* | 9.12 ± 2.34* | 9.15 ± 2.26 | 7.90     |
| Fasting glucose, mmol/L          | 7.04 ± 3.60 | 4.98 ± 0.40* | 5.44 ± 0.93* | 10.43 ± 4.21* | 10.56 ± 4.19 | 5.14 ± 4.06 | 0.000 |
| Two-hour OGTT, mmol/L            | 11.67 ± 7.22 | 6.26 ± 0.88* | 9.07 ± 1.29* | 19.64 ± 6.27* | 19.69 ± 6.35 | 17.93 ± 6.07 | 0.003 |
| Fasting glucosuria (+)           | 37 (33.9)  | 9 (18.4)  | 3 (14.3)   | 26 (52.9)  | 25 (52.9) | 0 (0%)   | 0.000 |
| GADA (+)                         | 14 (12.7)  | 6 (12.2)  | 3 (14.3)   | 13 (25.9)  | 2 (4.1)   | 1 (100.0) | 0.662 |
| C-peptide, ng/mL                 | 1.28 ± 0.29 | 0.39 ± 0.57* | 0.28 ± 0.32 | 2.75 ± 13.26 | 0.30 ± 0.56 | 72.91 ± 46.97 | 0.469 |
| Inulin, ml/mL                    | 15.80 ± 15.36 | 10.43 ± 6.97* | 17.73 ± 13.28* | 22.43 ± 29.05* | 22.69 ± 29.59 | 13.41 ± 0.328 |
| Triglyceride, mg/dL              | 2.52 ± 1.46 | 1.98 ± 0.75* | 2.64 ± 1.39* | 3.14 ± 1.85* | 3.14 ± 1.87 | 2.97 ± 1.01 |

The data are presented as mean ± SD values or number (%). Statistical significance of differences variance among 3 groups (NGT, IGT, and DM) was tested using one-way ANOVA analysis.

OGTT = oral glucose tolerance test, NGT = normal glucose tolerance, IGT = impaired glucose tolerance, T2DM = type 2 diabetes mellitus, SPIDDM = slowly progressive insulin dependent diabetes mellitus, BMI = body mass index, HDL = high-density lipoprotein, LDL = low-density lipoprotein, HOMA-IR = homeostasis model assessment of insulin resistance, HbA1c = glycated hemoglobin, GADA = anti-glutamic acid decarboxylase antibody, SD = standard deviation, DM = diabetes mellitus, ANOVA = analysis of variance.

* † Identical superscript letters indicate a non-significant difference between the groups based on the Tukey multiple comparison test.

https://doi.org/10.3346/jkms.2017.32.6.985
However, there were no differences in age, sex, family history of T2DM, serum cholesterol, HbA1c, insulin, fasting glucose, and fasting glucosuria between these 2 groups.

The determinants for DM in the schoolchildren and adolescents who exhibited glucosuria were woman (OR, 2.549; P = 0.033), high BMI (OR, 4.075; P = 0.005) and total cholesterol (OR, 4.267; P = 0.015), LDL cholesterol (OR, 5.8169; P = 0.006), and triglyceride (OR, 10.594; P < 0.001) levels.

**DISCUSSION**

Through the school urine glucose screening program conducted in the Jeonbuk province, we identified 40 new cases of asymptomatic DM (97.5% were classified as T2DM). This is the first study to evaluate students with persistent glucosuria detected through large-scale urine screening of Korean schoolchildren and adolescents.

The main purpose of urine glucose screening is to identify children and adolescents with DM, especially T2DM, in the early stages of the condition. Since 1974, several countries have performed urine glucose screening at schools. In Japan, if the result of a second urine test is glucosuria-positive, OGTT is performed to confirm a DM diagnosis. According to Japanese studies (7), the incidence of T2DM in children has increased over the last 3 decades and is estimated at 2.63 per 100,000 children during the period 1974–2002. These studies also observed that most of the DM cases detected by the screening program were eventually diagnosed as T2DM and that > 80% of children with T2DM were obese (7,8,16). According to a study by Wei et al. (9), the annual incidence of T2DM detected through mass urine screening of Taiwanese children from 1993 to 1999 was 8.3 per 100,000 children among boys and 12.0 per 100,000 children among girls. In Korea, more than 4 million schoolchildren have participated in the annual urine screening program since 1998, and approximately 3,200 students are found to have glucosuria annually (11). In contrast to Japan and Taiwan, in Korea, if the result of a second urine test is positive for glucosuria, a follow-up evaluation for DM is advised but not mandated. Therefore, no data regarding DM detected through the school urine glucose screening program are currently available.

From 2010 to 2013, a total of 707,238 schoolchildren and adolescents completed urine glucose tests in the Jeonbuk province area, and the mean positive rate of glucosuria of the second urine test was 0.077%. In this study, morning spot urine was used to test glucosuria. Notably, first morning urine may better reflect a glycemic condition during night time than would morning spot urine. However, to obtain first morning urine sample, children collect urine at home (before breakfast) and bring it to school in refrigerated container. For convenience, in this study, students collected morning spot mid-stream urine at school and was tested within 1 hour after collection. The positive rate of glucosuria
was similar to that reported from a nationwide Korean schoolchildren screening study (11). In addition, the rate in this study was rather higher than a Japanese study (0.05%) and Taiwanese study (0.03%) (9,16).

Of the 147 students who did not have known DM or a renal glucosuria diagnosis prior to this study, 110 (74.8%) underwent the OGTT, of whom 40 (36.4%) were newly diagnosed with DM. Compared to Japanese study (7), the OGTT recapture rate in this study was low (74.8%) and the low recapture rate may have influenced the incidence of DM in Korean schoolchildren. However, there were no significant differences in anthropometric characteristics between schoolchildren who did and did not participate in the OGTT. This finding suggests that the incidence rates of DM in children who did and did not undergo the OGTT might be similar. Of the 40 children with DM, 39 were classified as having T2DM. Meanwhile, only 1 child in this study was classified as having SPIDDM. None of children exhibited symptoms of severe hyperglycemia or ketosis at the time of diagnosis. These children were more likely to be obese and to have high serum C-peptide levels and HOMA-IR index scores. Even though no significant difference in the family history of T2DM was observed among the 3 groups, the frequency of a family history of T2DM was higher in the DM group than in the NGT group (55.0 vs. 34.7%; P < 0.05). Several Japanese studies have reported the detection of small numbers of children with T1DM through the urine glucose screening program (7,16). These children exhibited a SPIDDM with minimal or no symptoms of DM and maintained enough β-cell capacities until 2 years after diagnosis, and most were positive for pancreatic autoantibodies (7,16,17).

The presence of GADA is a sensitive and specific marker of the future need for insulin therapy in patients with T2DM and SPIDDM (18). Compared with Western countries, the prevalence of pancreatic autoantibodies is lower in Korean patients with T1DM and non-insulin dependent diabetes mellitus (NIDDM) (19,20). In this study, we examined only GADA and only 3 children exhibited GADA positivity.

Based on the results of our study, the mean incidence of T2DM among schoolchildren in the Jeonbuk province was 5.5 per 100,000 schoolchildren and adolescents. Considering the low recapture rate (74.8%) for OGTT, the incidence of T2DM in Korean children and adolescents may be higher than estimated by our study and may be similar to the incidence of DM among Taiwanese children (9). In contrast, our estimated incidence is much higher than that among schoolchildren in Japan (16). Epidemiological studies in Korea have shown that the incidence of T1DM is approximately 1.5 per 100,000 for the population younger than 15 years (21). However, no previous studies have reported the incidence or prevalence of T2DM among Korean youths.

Prediabetes, defined as IGT or impaired fasting glucose (IFG), is regarded a risk factor for later DM. Children and adults with IGT have an increased risk of developing cardiovascular problems prior to the progression of IGT to DM (22). In this study, 21 children (19.1%) were diagnosed with IGT and had higher BMI, HbA1c, insulin, C-peptide, and HOMA-IR index values than did children with NGT. Several patients were lost to follow-up, and only 8 subjects were followed up for more than 1 year; of these children, 2 developed T2DM. The frequency of IGT in this study was higher than that reported in previous Japanese studies (17).

Although previously considered a condition that primarily affects the middle-aged or elderly, T2D is increasingly being reported among children and adolescents. This is likely due to the emerging epidemic of youth obesity (2). Evidence demonstrates that the number of children with T2DM has increased in recent years and continues to increase in the United States (2,4). Several studies have shown an increased risk of microvascular complications in youth with T2DM relative to those with T1DM (4,5). This increment in obesity is also apparent in Korea, where the incidence of obesity has increased significantly over the last decade (23,24). According to a recent report from the Jeonbuk Office of Education, Korea, prevalence rates of obesity among schoolchildren of the Jeonbuk province in 2013 was 9.6% for elementary school, 12.3% for middle school, and 17.2% for high school (25). In this study, we found that prevalence of obesity (22.6%) among children with glucosuria was higher than the mean prevalence among Jeonbuk schoolchildren, but lower than that of children who underwent the OGTT and those with DM. This finding suggests that urinary glucose screening is important for the detection of asymptomatic DM in obese schoolchildren.

Random urinalysis is not satisfactorily sensitive to detect most cases of DM, and the sensitivity and specificity of urinalysis screening for DM vary according to the study (26,27). In addition, some children with DM may have received medical treatment. Therefore, the urine screening program described herein might underestimate the actual prevalence and incidence of DM in schoolchildren. Nevertheless, our data suggest that the school urine glucose screening program is an effective, simple and easy method for the early detection of DM in asymptomatic children and adolescents, especially among woman and obese Korean students. To improve low OGTT performance response rates such as that observed in our study, school urine glucose screening programs in Korea should be adapted to incorporate standards from other countries.

This study was limited by several factors. First, we used morning spot urine instead of first morning urine samples. First morning urine may better reflect a glycemic condition during night time after dinner. Second, only 8 of the 12 grades in the whole school year were eligible. Third, this screening program had a recapture rate of 74.8%. This low recapture rate might have caused underestimation of the prevalence. Finally, 398 children with glucosuria during the second urine test were excluded from the analysis because of known DM or renal glucosuria. This exclu-
detection asymptomatic DM, especially T2DM in children and adolescents.

DISCLOSURE

The authors have no potential conflicts of interest to disclose.

AUTHOR CONTRIBUTION

Investigation: Kim MS, Lee DY. Writing - original draft: Kim MS, Lee DY. Writing - review & editing: Kim MS, Lee DY. Investigation: Kim MS, Lee DY. Writing - original draft: Kim MS, Lee DY. Writing - review & editing: Kim MS, Lee DY. The authors have no potential conflicts of interest to disclose.

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