Concise report

Gout and Type 2 diabetes have a mutual inter-dependent effect on genetic risk factors and higher incidences

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Abstract

Objective. To explore the causal relationship between gout and Type 2 diabetes based on genetic evidence and national outpatient database.

Methods. Twenty male gout patients with early-onset, gout family history, without a habit of alcohol consumption or obesity before the first attack of gout were selected from hospital in 2010; and 42 unrelated male Chinese subjects were selected from HapMap as controls for genome-wide analysis study (GWAS). The comorbid diseases with gout were revealed by applying the significant single nucleotide polymorphisms (SNPs) to MetaCore platform, and the comorbid relationship was analysed by standardized incidence ratio (SIR) from outpatient database.

Results. A total of 334 SNPs were significantly related to gout in GWAS ($P < 10^{-7}$), and Type 2 diabetes was the most significantly associated disease with gout as recognized by 36 gene symbols correspondent to the above significant SNPs. The analysis of national outpatient database showed that the overall incident Type 2 diabetes was 1.50 cases per 1000 person-months among gout patients, which was higher than the overall incident gout (1.06 cases) among Type 2 diabetes. The age-adjusted SIR of incident Type 2 diabetes among gout was 2.59 (95% CI 2.42, 2.78), whereas the age-adjusted SIR for incident gout among Type 2 diabetes was 1.61 (95% CI 1.48, 1.74).

Conclusion. After excluding obesity and alcohol consumption behaviour, this study showed that patients with gout and Type 2 diabetes shared the common genetic factors most, and that there existed a mutual inter-dependent effect on higher incidences.

Key words: gout, Type 2 diabetes, early onset, genome-wide analysis study, incidence, genetic risk factor, genetic epidemiology, national health insurance, outpatient records, male.

Introduction

Gout is frequently associated with various kinds of comorbidities, such as hypertension, chronic kidney disease and hyperlipidaemia, which complicate management and affect long-term prognosis [1, 2]. In addition, insulin resistance and Type 2 diabetes have also been noted to be associated with gout. Yoo et al. [3] reported that the incidence of insulin resistance in gout patients increased by as much as 35% over individuals without gout, and Suppiah et al. [4] demonstrated a high prevalence of gout in patients with Type 2 diabetes, whereas Rodriguez et al. [5] suggested that patients with diabetes were at a lower future risk of gout independent of other...
risk factors. Furthermore, some studies have even shown strong associations between gout and metabolic syndrome [6–10] as well as cardiovascular disease [11, 12]. Whether the onset of Type 2 diabetes exacerbates the future risk for developing gout or vice versa has not been well studied.

The metabolic syndrome, obesity, alcohol consumption, lead exposure as well as age and male gender have been shown to associate with gout [13–16]. Meanwhile, the genetic component findings are related to hyperuricaemia [17–21] as well as function of pro-inflammatory cytokines, such as TNF-α gene [22], cyclic GMP-dependent protein kinase II gene [23] and TGF-β1 gene [24]. The contributions of the associated genetic factors to gout are various, and some diseases may share the common genetic components with gout. Thus, it is important to clarify the relationship between comorbid diseases and gout based on genetic evidence.

The purpose of our study was to explore the diseases that shared the most genetic markers with gout by a genome-wide association study (GWAS), and conducted an analysis of standardized incidence ratio (SIR) from the outpatient database to reveal the incidence risks between gout and comorbid diseases in the general population.

Materials and methods

Study design

We designed this study in three stages: (i) screening genetic risk markers for gout by a GWAS; (ii) exploring the comorbid diseases with gout based on genetic component by applying the genetic risk markers to MetaCore platform; and (iii) confirming the associations by analysis with the national outpatient records released from National Health Research Institute (NHRI) in Taiwan.

GWAS

The participants included in GWAS were divided into case and control groups. The case group was 20 male gout patients, who were enrolled under the following criteria: with primary gout, non-Taiwan indigenous ancestry, at least father or brother with gout history, age < 40 years at first gout attack, without a habit of alcohol consumption behaviour and BMI < 27 kg/m² before first gout attack. The gout patients were diagnosed by a rheumatologist in the hospital, and were enrolled after giving their informed consent to Kaohsiung Chang-Gung Memorial Hospital in 2010. The study was approved by the Institute Review Board of Kaohsiung Chang-Gung Memorial Hospital (IRB No. 98-2622B). The control group contained 42 unrelated male subjects of Chinese ancestry (Beijing) selected from HapMap web site (http://www.hapmap.org) to have their genetic data (Phase 3) serve as control for GWAS, since both cases and controls were of Han Chinese ethnic origin.

Genomic DNA was applied to the Illumina Human660W-Duad BeadChip (Illumina, Inc., San Diego, CA, USA) for whole-genome genotyping. Single nucleotide polymorphisms (SNPs) were excluded if: (i) the SNP was not matched with the controls; (ii) the SNP had missing data from any one of the cases; and (iii) there was significant distortion from Hardy–Weinberg equilibrium in the controls ($P < 10^{-7}$). The chip for GWAS revealed 592 652 SNPs, and a total of 479 403 SNPs were effective for further analysis after excluding those that did not meet the above criteria.

National outpatient database

The outpatient database was released from the NHRI in Taiwan, which provided us with one million random subjects covering the years 1997–2008 for the study. The data sets contain outpatient records and prescription details, including the gender, three diagnosed codes (CD-9-CM) and each prescription. We also obtained the approval of the Institute Review Board of Kaohsiung Medical University for analysis of the database for research (KMUH-IRB-20110130).

The cumulative incidence of Type 2 diabetes or gout were revealed by analysis of the outpatient database. The authors defined those male patients with or without gout diagnosed during the period from 1997 to 2000 as the first baseline population, whose age was > 20 years, and who had no diagnosis of outcome disease; and then we analysed the records to find incident Type 2 diabetes from 2001 to 2008. Inversely, second baseline population was selected to reveal an incident gout among Type 2 diabetes patients during the same time period. At enrolment, no patients were diagnosed with cancer. The controls were selected by matching the age groups of cases on a ratio of 5:1. Gout was identified by diagnosis with ICD-9 code 274 and prescription of any of the following drugs more than three times: colchicine, allopurinol, benz bromarone, probenecid and sulphinpyrazone. Type 2 diabetes was diagnosed by ICD-9 code 250 more than three times after excluding Type 1 diabetes codes (250.x1, 250.x3), whereas cancer was diagnosed by the ICD-9 code from 140 to 208, and those not diagnosed by ICD-9 codes 274 and 250 were classified as gout- and diabetes-free patients, respectively.

Statistics

The association analysis in GWAS was carried out by chi-square test to compare allele frequency and genotype distribution between cases and controls using four single-point methods for each SNP: genotype, allele, dominant and recessive models. The most significant statistical result was chosen from the four models. The alpha level was $10^{-7}$ after adjustment of Bonferroni correction. SNPs with $P$-values less than alpha level were considered to be significantly associated with the traits, and these were then applied to MetaCore platform (version 6.6; http://portal.genego.com) to explore the comorbid diseases with gout. The incidence of comorbid disease per 1000 person-months was estimated among those with or without gout (or Type 2 diabetes), and age-adjusted SIR and 95% CI were estimated to show the rate ratio of incident disease among these two
groups [25]. We performed the statistical analysis for GWAS by SAS program (v9.2), and the national outpatient records by PERL (v5.8) program.

Results

Twenty male gout patients were selected for GWAS, the participants’ mean (s.d.) ages at enrolment and at gout onset were 41.75 (7.79) and 26.70 (7.32) years, respectively. A total of 334 SNPs were found to be significantly related to gout by GWAS (all $P < 10^{-7}$), and then these SNPs were applied to MetaCore platform. The results displayed the most significantly related disease was Type 2 diabetes as recognized by 36 gene symbols (Table 1). Even when we increased the alpha level to $10^{-3}$, a total of 921 SNPs were found to have significant associations between gout patients and controls, and the result also showed that Type 2 diabetes was the most significant disease related to gout as recognized by 74 gene symbols (data not shown).

The first baseline population included 9022 male gout patients and 45,110 male controls aged >20 years and without diagnosis of Type 2 diabetes or cancer. The mean (s.d.) age of the gout patients was 51.75 (14.60) years and the controls was 51.64 (15.03) years (Table 2), while the means of follow-up were 7.53 and 8.38 years for gout patients and controls, respectively. The overall incidence Type 2 diabetes were 1.50 and 0.57 cases per 1000 person-months among those diagnosed with and without gout, respectively. The SIR was 2.59 (95% CI 2.42, 2.78) after adjustment of age (Table 2). Concerning the second baseline population, the mean (s.d.) age of those with Type 2 diabetes was 57.66 (11.55) years ($n=8078$) and the controls was 57.47 (12.07) years ($n=40,390$), while the means of follow-up were 7.92 years for Type 2 diabetes and 7.86 years for controls. The overall incident gout was 1.06 and 0.64 cases per 1000 person-months among those diagnosed with and without Type 2 diabetes, respectively. The age-adjusted SIR was estimated to be 1.61 (95% CI 1.48, 1.74; Table 2).

Discussion

Our study suggests that gout and Type 2 diabetes share the common genetic markers identified by the GWAS most, and that there existed a mutually inter-dependent effect on increased incidences confirmed by analysis of the national outpatient clinical records. Although the incident association between gout and Type 2 diabetes has not been defined clearly before, here we suggest, based on genetic and clinical evidence, that gout showed a most significant association with incident Type 2 diabetes, and vice versa. The gout patients who participated in the GWAS tended to have genetic causal factors such as those with gout family history, age <40 years as well as no alcohol consumption habit or obesity at the time of first gout attack. The above criteria ensure this study explored the genetic factors related to gout and that they would not have been confounded by environmental factors such as alcohol consumption and obesity.

Although Type 2 diabetes showed an association with gout, obesity and insulin resistance may represent important links between them. Some studies even

### Table 1 SNPs and gene symbols shared by gout and Type 2 diabetes

<table>
<thead>
<tr>
<th>No.</th>
<th>SNP ID</th>
<th>Gene symbol</th>
<th>Chromosome location</th>
<th>No.</th>
<th>SNP ID</th>
<th>Gene symbol</th>
<th>Chromosome location</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>rs2343519</td>
<td>A2BP1</td>
<td>16p13.3</td>
<td>19</td>
<td>rs10225163</td>
<td>JAZF1</td>
<td>7p15.2-p15.1</td>
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<tr>
<td>2</td>
<td>rs4149327</td>
<td>ABCA1</td>
<td>9q31.1</td>
<td>20</td>
<td>rs2465065</td>
<td>LHFP1</td>
<td>7q22.1</td>
</tr>
<tr>
<td>3</td>
<td>rs2077654</td>
<td>ABCC8</td>
<td>11p15.1</td>
<td>21</td>
<td>rs187775</td>
<td>LIPC</td>
<td>15q21-q23</td>
</tr>
<tr>
<td>4</td>
<td>rs4148613</td>
<td>ABCC8</td>
<td>11p15.1</td>
<td>22</td>
<td>rs10239506</td>
<td>PDE1C</td>
<td>7p14.3</td>
</tr>
<tr>
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<td>rs4148660</td>
<td>ABCC9</td>
<td>12p12.1</td>
<td>23</td>
<td>rs9900205</td>
<td>PRKCA</td>
<td>17q22-q23.2</td>
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<td>6</td>
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<td>ABCG1</td>
<td>21q22.3</td>
<td>24</td>
<td>rs3798343</td>
<td>PPARD</td>
<td>6p21.2</td>
</tr>
<tr>
<td>7</td>
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<td>ALX4</td>
<td>11p11.2</td>
<td>25</td>
<td>rs12501032</td>
<td>PPARGC1</td>
<td>4p15.1</td>
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<td>ATP1A4</td>
<td>1q21-q23</td>
<td>26</td>
<td>rs10815925</td>
<td>PTPRD</td>
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<td>RARB</td>
<td>3p24</td>
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<td>2p21.1</td>
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<td>rs847058</td>
<td>SLC15A3</td>
<td>20q12-q13.1</td>
</tr>
<tr>
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<td>Carboxyl peptidase E</td>
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<td>CFFR</td>
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<td>SLC22A18AS</td>
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<td>rs4149458</td>
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</tr>
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<td>10q24.3-pter</td>
<td>32</td>
<td>rs1499614</td>
<td>TPST1</td>
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</tr>
<tr>
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<td>8q13</td>
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<td>FSTL4</td>
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<td>35</td>
<td>rs4766398</td>
<td>WNT5B</td>
<td>12p13.3</td>
</tr>
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<td>HGF</td>
<td>7q21.1</td>
<td>36</td>
<td>rs10773971</td>
<td>WNT5B</td>
<td>12p13.3</td>
</tr>
</tbody>
</table>

A total of 334 SNPs were found to be significantly associated with gout in the GWAS; after applying the SNPs to MetaCore platform, it was found that the listed 36 genes were shared by Type 2 diabetes.
showed that increased body weight is associated with higher serum urate levels and an increased risk of gout [26]. Obesity also leads to insulin resistance, which in turn results in enhanced risk for the development of Type 2 diabetes [29]. However, all the patients included in our study in the GWAS had not been obese before the first gout attack, so we do not consider obesity to be the risk factor for the association of Type 2 diabetes in this study. The authors found that Type 2 diabetes was comorbid with gout on the basis of genetic evidence instead of environmental evidence; the following results also showed a similar effect, i.e. that there was a higher incidence of Type 2 diabetes among gout patients than the incidence of gout among Type 2 diabetes patients. The molecular aetiologies of Type 2 diabetes are diverse, which results in reduced beta-cell mass, disruptions of beta-cell function as well as lipid profile metabolism. Our study showed 36 genes for the risk of both gout and Type 2 diabetes, including carboxypeptidase E (CPE), which is a major link between hyperlipidaemia and beta-cell death pathways in diabetes [30]; and its variants resulting in altered CPE activity have been reported in patients with early-onset Type 2 diabetes [31]. Other variants in ATP-binding cassette (ABCC8) genes were shown to be associated with the regulation of insulin secretion, and they were suggested to be diabetes susceptibility genes [32, 33]. Genetic variation in the solute carrier (SLC22A1) gene has found a glucose-lowering effect by reducing glycated haemoglobin (HbA1c) level in diabetes patients [34]. Alterations in Wnt signalling might also be involved in the pathogenesis of diabetes, which influences endocrine pancreas development and modulates mature beta-cell functions including insulin production. Variants in the WNT5B gene were also found to be strongly associated with Type 2 diabetes [35, 36].

Moreover, epidemiological studies showed that hyperlipoproteinaemia is significantly associated with gout and Type 2 diabetes [37]. In our study, the ATP-binding cassette (ABCA1 and ABCG1) genes, which are involved in the cellular lipid removal pathway, were found to be associated with gout and Type 2 diabetes [38]. Early studies also showed that low density lipoprotein (LDL) cholesterol, when low density lipoprotein (LDL) cholesterol and other cholesterol pathways also were independent risk factors for developing Type 2 diabetes [39, 40]. In conclusion, this study suggests that obesity and alcohol consumption are important factors in the development of Type 2 diabetes. Epidemiological studies confirmed this finding, and based on genetic evidence, the function and studies of the ATP-binding cassette and ABCG1 genes have been shown to be associated with gout and Type 2 diabetes [41]. The function and studies of these genes are consistent with the hypothesis that cholesterol regulates the function and studies of the ATP-binding cassette and ABCG1 genes, which are involved in the cellular lipid removal pathway. In conclusion, this study suggests that obesity and alcohol consumption are important factors in the development of Type 2 diabetes.
Rheumatology key messages

- Gout and Type 2 diabetes share common genetic risk markers.
- A mutual inter-dependent effect on higher incidences exists between gout and Type 2 diabetes.

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References


