A Strong Secular Trend in Serum Gamma-Glutamyltransferase from 1996 to 2003 among South Korean Men

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Serum gamma-glutamyltransferase (GGT) level, within its normal range, has dose-response associations with many cardiovascular disease risk factors, components of the metabolic syndrome, and the future risk of developing diabetes mellitus, stroke, cardiac mortality, and nonfatal myocardial infarction (1–8). Very recently, serum GGT level, within its normal range, has been proposed as a sensitive and reliable marker of oxidative stress (9).

Serum gamma-glutamyltransferase (GGT) concentration, within its normal range, has recently been proposed as a reliable marker of oxidative stress. Oxidative stress plays a central pathogenic role in many metabolic and/or cardiovascular diseases, incidences of which have recently increased in South Korea. Since serum GGT has strong associations with these diseases and their risk factors, the authors hypothesized a corresponding secular trend of increasing serum GGT levels in South Korea. Study subjects were 8,072 male workers at a large steel company who were aged 24–44 years at baseline and had received annual physical examinations from 1996 to 2003. The secular trend was a 0.1066-units/liter increase in ln(GGT) level per calendar year (a 180% increase during the 7-year follow-up period) (p < 0.01). Adjustment for body mass index, alcohol consumption, smoking, exercise, and cholesterol level as time-dependent covariates did not change the results. Although cholesterol is commonly used as a marker of epidemiologic transition, there was a less dramatic secular trend in ln(serum cholesterol) level, and it disappeared after adjustment for the secular trend in serum GGT. These findings suggest that serum GGT concentration can be used as a sensitive marker of epidemiologic transition, and they portend a continuing rise in incidences of metabolic and/or cardiovascular diseases in this population in the coming years.

Abbreviations: ALT, alanine aminotransferase; GGT, gamma-glutamyltransferase; Korea; oxidative stress; time

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to cardiovascular disease and its risk factors, one would expect to observe a secular trend of increasing serum GGT levels in South Korea.

We performed this study to evaluate the hypothesis that there has been a secular trend of increasing serum GGT levels in South Korea. We further compared the secular trend in serum GGT with secular trends in two related variables: cholesterol and alanine aminotransferase (ALT). Serum total cholesterol is an important cardiovascular disease risk factor which has typically shown an increasing secular trend (13, 14) during the epidemiologic transition in Asia. The trend in total cholesterol would be expected to parallel the trend in GGT if the trend in GGT reflected primarily that trend. Serum ALT is a more liver-specific enzyme whose pattern would be expected to parallel that of GGT if the trend in GGT reflected primarily worsening liver function.

MATERIALS AND METHODS

Study population

The study population was male workers at a large steel company in South Korea. Workers who had received annual physical examinations from 1996 and 2003 and had no missing data for any year were eligible for this analysis. A total of 11,234 men received a health examination in 1996. After exclusion of 2,298 men who retired before 2003 (mostly because of reaching retirement age (≥55 years)) and 864 men who had missed any examination during the follow-up period, 8,072 men were included in the final analysis. (Inclusion of the 2,298 retirees and the 862 men with any missing examinations in the repeated-measures regression analysis did not materially change the results.) Besides the health examinations, this company conducted an active health promotion campaign during 1999–2001, focusing on smoking cessation and exercise.

Measurements

The annual health check-ups were performed at the company health-care center, located on the premises, between 9:00 a.m. and noon after an overnight fast. Information on lifestyle factors such as cigarette smoking, alcohol consumption, and exercise was obtained primarily by self-reported questionnaire. All workers were asked to fill out the same questionnaire, or a slightly modified one, each year. Body weight was measured with the employee standing in light clothing, and body height was measured without shoes. Body mass index was calculated as weight (kg) divided by height squared (m²). Venous blood samples were obtained from an antecubital vein after a 12-hour overnight fast. The serum samples were kept at 4°C and analyzed within 48 hours. Serum GGT and ALT concentrations were measured in the same analytic run at 37°C with an automatic analyzer, and total cholesterol level was measured using enzymatic methods (Shimadzu CL-7000; Shimadzu Corporation, Kyoto, Japan (1996–1997) and Hitachi 7170, Hitachi Ltd., Tokyo, Japan (1998–2003)). We saw no differences in the sample collection methods and analytic techniques used between 1996 and 2003. Although we did not have available quantitative records of assay variability, quality control is performed annually by the Korean Association of Quality Assurance for Clinical Laboratories, which sends standard serum samples to each laboratory in South Korea four times per year. This association officially determined that the laboratory had results within the range of prespecified values for each of the analytes discussed in this paper throughout the time period of this investigation.

Statistical analysis

The data were analyzed by means of a mixed-effects model, which is a type of statistical analysis commonly used for repeated measurements. We used statistical models defined by Jacobs et al. (15) for the estimation of beta coefficients for an age effect, period effect, or cohort effect. There are numerous ways to conceptualize these effects; it is well-known that in cross-sectional data, the separate effects are nonidentifiable. Nonidentifiability also applies to the baseline of a cohort study, such as this one.

However, Jacobs et al. (15) pointed out that during the follow-up period of a cohort study, the three effects are identifiable. They defined the age effect as variation observed as an individual ages (independent of calendar time) and period effects as population-wide changes over calendar time (independent of age). In this framework, a birth cohort effect is variation as an individual ages specific to a particular calendar time—that is, the interaction between the age and period effects. Modeling single years of age and single years of calendar time, this model is an exact fit to the age- and calendar-year mean levels of the dependent variable, which would produce the exact mean values shown in figure 1, for example, if there were no missing follow-up data. To resolve the baseline nonidentifiability problem, we assumed no historical birth cohort effect—that is, that the data as they existed at baseline were negligibly influenced by the birth cohort of each individual. The interaction between age and period effects during follow-up is called the “evolving birth cohort effect” and is directly observable. A small evolving birth cohort effect in this study is consistent with, but does not prove, the absence of a historical cohort effect. Some confusion may arise because the term “birth cohort effect” may refer to the totality of what happens to a particular birth cohort or only to the interaction between age and period effects. In this paper, following the method of Jacobs et al. (15), we mean the interaction when we refer to birth cohort effect; then the total experience for an individual is the sum of the age, period, and cohort effects, which are nonidentifiable at the baseline observation but identifiable during follow-up (15).

In this framework, we computed the coefficients for age at examination and calendar time in regression analyses using serum GGT as the dependent variable, also considering the interaction between age and calendar time. The age coefficient estimates between-person differences in serum GGT per year of age, which we call the serial cross-sectional age slope. The time coefficient estimates the age-matched time trend, namely the change in serum GGT level after the removal of between-person differences in age; this coefficient
includes purely time-related effects, such as a secular trend and methodologic artifacts (which might include, among other things, year-to-year laboratory variation). If we can exclude methodologic artifacts from the age-matched time trend, the age-matched time trend can be interpreted as a secular trend.

Under this model, the analysis does find a significant interaction between age and calendar time, suggesting the presence of a birth cohort effect that has been evolving since the initial observation in 1996. Because the evolving birth cohort effect was very small compared with the age-matched time trend and because reporting the evolving cohort effect would have added greatly to the complexity of this report, in most analyses we suppressed the interaction term, but we comment here on the nature of the interaction. We further fitted both age and time as linear effects as a simple device for reporting. Rather than fit complex nonlinear models—for example, with each year of age and calendar time modeled as an indicator variable—we simply comment on obvious nonlinearities that are seen graphically. We repeated these analyses first with serum cholesterol and then with serum ALT as the dependent variable. To examine whether changes in health behavior during follow-up could explain the age-matched time trend, we adjusted for body mass index, alcohol consumption, smoking, exercise, and serum total cholesterol level as time-dependent covariates.

RESULTS

The average age of the study subjects at baseline was 36.7 years (range, 24–44 years). As the first step in the analyses, we examined geometric mean values for serum GGT, ALT, cholesterol, body mass index, alcohol drinking, smoking, and regular exercise on a yearly basis from 1996 through 2003 (table 1). Serum GGT level increased by 180 percent during the 7-year follow-up period; geometric mean values for serum GGT were 10.5, 14.5, 18.6, 20.2, 21.4, 22.1, 26.6, and 29.2 units/liter in successive examination years. During the period of the company’s health promotion program, from 1999 to 2001, the trend of increasing serum GGT levels slowed a little but continued. Moreover, strong secular trends were similarly observed among nondrinkers, non-smokers, subjects with a body mass index less than 25, subjects with an ALT level less than 35 units/liter, subjects who were negative for hepatitis B surface antigen, and office workers (who would be less likely to be exposed to any possible toxins in the steel factory). In parallel, serum cholesterol level also increased by 8 percent—apparently a less dramatic change than that for GGT. In contrast, serum ALT level actually decreased by 0.9 percent during the follow-up period. During the same period, subjects began to smoke less, exercise more, drink more alcohol, and gain more body fat. Especially during the period of health promotion, from 1999 to 2001, the proportion of smokers dramatically decreased. The age-adjusted prevalences of diabetes at the start and end of follow-up were 1.65 percent and 2.64 percent, respectively. The corresponding figures for hypertension were 16.7 percent and 19.2 percent, respectively.

Figure 1 shows the age-specific mean levels of serum GGT from 1996 to 2003, by examination year. Serum GGT levels increased approximately linearly across age, similarly in all examination years. The vertical distance from 1996 to 2003 for persons of a certain age was similarly observed across all age ranges, indicating an age-matched time trend (period effect) of approximately 15 units/liter across the 7-year period; this period effect was fairly uniform for each calendar year but was nevertheless somewhat nonlinear, slowing during the period 1999–2001. On the other hand, longitudinal changes in serum GGT levels by birth cohort are shown in figure 2. The longitudinal changes in serum GGT were also consistently observed for all birth cohorts, the notch in each
curve reflecting the decelerated increase from 1999 to 2001. The total longitudinal change over 7 years is the combination of the aging trend and the age-matched time trend ( 18 units/liter), suggesting that 7 years of aging accounts for an increase in serum GGT of approximately 3 units/liter. For any given age, serum GGT level is higher in younger (more recent) cohorts than in older cohorts.

In parallel with GGT level, serum total cholesterol level also showed calendar-time-related increases for all ages, with the vertical distance from 1996 to 2003 for a certain age being approximately 8 mg/dl (figure 3). The total longitudinal changes in serum cholesterol level were also consistently observed in all birth cohorts, amounting to approximately 15 mg/dl; this suggests that 7 years of aging corresponded to an increase in serum cholesterol of approximately 7 mg/dl (figure 4). For any given age, serum cholesterol concentration was higher in younger workers than in older workers. Contrary to serum GGT, serum ALT did not increase with age in any examination year and did not show any longitudinal changes by birth cohort, suggesting no age, period, or cohort effect in serum ALT levels (data not shown).

We applied the mixed-effects model for serially measured data to estimate more quantitatively the age-matched time trend and age-related changes in serum GGT. The interaction term for cross-sectional age effect and age-matched time trend was statistically significant, suggesting the presence of an evolving cohort effect. However, compared with the cross-sectional age effect or the age-matched time trend, the absolute value of this cohort effect appeared to be trivial, as can be seen visually by observing the nearly parallel patterns in figures 1 and 2. Therefore, we ignored the evolving cohort effect in the following analyses.

<table>
<thead>
<tr>
<th>Examination year</th>
<th>Mean values</th>
<th>% change (2003 – 1996)</th>
</tr>
</thead>
</table>

### TABLE 1. Distribution of data on serum gamma-glutamyltransferase, alanine aminotransferase, serum total cholesterol, and health behavior variables among 8,072 men aged 24–44 years at baseline, Pohang, South Korea, 1996–2003

<table>
<thead>
<tr>
<th>Examination year</th>
<th>Gamma-glutamyltransferase level (units/liter)*</th>
<th>Cholesterol level (mg/dl)*</th>
<th>Body mass index (kg/m^2)*</th>
<th>Alcohol consumption (g/week)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1996</td>
<td>10.5</td>
<td>27.6</td>
<td>22.9</td>
<td>100</td>
</tr>
<tr>
<td>1997</td>
<td>14.5</td>
<td>26.9</td>
<td>23.0</td>
<td>99</td>
</tr>
<tr>
<td>1998</td>
<td>18.6</td>
<td>28.1</td>
<td>23.1</td>
<td>103</td>
</tr>
<tr>
<td>1999</td>
<td>20.2</td>
<td>27.3</td>
<td>23.3</td>
<td>107</td>
</tr>
<tr>
<td>2000</td>
<td>21.4</td>
<td>25.6</td>
<td>23.6</td>
<td>109</td>
</tr>
<tr>
<td>2001</td>
<td>22.1</td>
<td>25.9</td>
<td>23.6</td>
<td>110</td>
</tr>
<tr>
<td>2002</td>
<td>26.6</td>
<td>25.9</td>
<td>23.6</td>
<td>117</td>
</tr>
<tr>
<td>2003</td>
<td>29.2</td>
<td>27.0</td>
<td>23.7</td>
<td>120</td>
</tr>
</tbody>
</table>

* Geometric mean.
† Subjects who reported not drinking, not smoking, or having had a body mass index less than 25, respectively, every year from 1996 to 2003.
‡ Weight (kg)/height (m)^2.
§ ALT, alanine aminotransferase.
¶ The definition of incident diabetes was a serum fasting glucose level ≥126 mg/dl or use of diabetes medication.
# The definition of hypertension was systolic blood pressure ≥140 mmHg or diastolic blood pressure ≥90 mmHg or use of antihypertensive medication.
In the models, \( \ln(\text{GGT}) \) increased by 0.1066 units/liter per calendar year and by 0.0221 units/liter per year of age (approximately 11 percent and 2 percent per year on the natural GGT scale; both \( p \)'s < 0.01) (table 2). After we adjusted for body mass index as a time-dependent covariate, the coefficient of age-matched time trend for serum GGT was not materially changed, implying that increasing fatness in the sample could not explain the age-matched time trend of serum GGT. Additional adjustment for alcohol consumption, smoking, exercise, and serum cholesterol level as time-dependent covariates did not change the results either. The serial cross-sectional age effect of serum GGT substantially decreased after adjustment for body mass index, but it remained highly statistically significant. The coefficients for body mass index, alcohol, exercise, and smoking were all highly statistically significant, suggesting that these factors and their changes were each closely related to GGT and its changes.

On the other hand, \( \ln(\text{cholesterol}) \) increased by 0.0061 mg/dl per calendar year and by 0.0059 mg/dl per year of age (both \( p \)'s < 0.01) (table 3). Compared with serum GGT, the increasing slopes of serum cholesterol level were much weaker in terms of both age-matched time trend and serial cross-sectional age effect. However, after adjustment...
for serum GGT, the age-matched time trend in serum cholesterol disappeared (model 6 in table 3).

**DISCUSSION**

In this study, a striking increase in serum GGT concentration was observed during 1996–2003 among South Korean men aged 24–44 years at baseline. The age-matched time trend—that is, the period effect—was quite dramatic, particularly given that most ages in the study sample were present in all calendar years. A period effect can be a secular change (a systematic increase in GGT in the whole population over 7 years) or a methodologic artifact (e.g., a change in analytic or blood handling methods from year to year) (15). Although laboratory equipment was changed once in 1998, it did not appear to explain our observation, because the laboratory met a quality control standard each year and the age-matched time trends were fairly consistent throughout the 7-year follow-up period (allowing that, as would be expected, the trend slowed somewhat during the health promotion campaign of 1999–2001). In addition, serum ALT, measured in the same analytic run as GGT, did not show any period effect.

**FIGURE 4.** Age-specific geometric mean values for serum total cholesterol level among 8,072 men aged 24–44 years at baseline, by birth cohort, Pohang, South Korea, 1996–2003.

**TABLE 2.** Beta coefficients* for the regression of cross-sectional age effect and age-matched time trend on natural log-transformed serum gamma-glutamyltransferase level (units/liter)

<table>
<thead>
<tr>
<th>Variable</th>
<th>Age (per year)</th>
<th>Time trend (per year)</th>
<th>Body mass index† (per kg/m²)</th>
<th>Alcohol drinking (per category‡)</th>
<th>Smoking (per category§)</th>
<th>Exercise (per category¶)</th>
<th>Total cholesterol level (per mg/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Model 1</td>
<td>0.0221</td>
<td>0.1066</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Model 2</td>
<td>0.0145</td>
<td>0.0986</td>
<td>0.1213</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Model 3</td>
<td>0.0145</td>
<td>0.0973</td>
<td>0.1191</td>
<td>0.0470</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Model 4</td>
<td>0.0147</td>
<td>0.1019</td>
<td>0.1206</td>
<td>0.0461</td>
<td>0.0518</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Model 5</td>
<td>0.0150</td>
<td>0.1035</td>
<td>0.1199</td>
<td>0.0462</td>
<td>0.0501</td>
<td>−0.0366</td>
<td></td>
</tr>
<tr>
<td>Model 6</td>
<td>0.0116</td>
<td>0.0996</td>
<td>0.1041</td>
<td>0.0458</td>
<td>0.0482</td>
<td>−0.0329</td>
<td>0.7414</td>
</tr>
</tbody>
</table>

* All coefficients were significant at p < 0.001.
† Weight (kg)/height (m)².
‡ Alcohol drinking was introduced into the model as a continuous variable with categories 1–5 (0, 1–90, 91–180, 181–270, and >270 g/week).
§ Smoking was introduced into the model as a continuous variable with categories 0–1 (nonsmoker vs. smoker).
¶ Exercise was introduced into the model as a continuous variable with categories 1–3 (never, <3 times/week, and ≥3 times/week).
Although the laboratory equipment used to assay serum ALT changed at the same time as that used to assay serum GGT. Any laboratory variation that remained unaccounted for would have been random and would likely have attenuated the estimated regression coefficients. Therefore, we interpret the period effect shown in this study as a secular trend.

Many cross-sectional studies have reported a positive association between age and serum GGT that is similar to ours (1–4). A cross-sectional relation between age and GGT may reflect several phenomena: physiologic changes due to aging itself, period effects confounded with aging, cohort effects, or various mixtures of these. Therefore, it is common that simple estimates of the age effect based on the cross-sectional age slopes give pictures different from those of longitudinal analyses that separate out period effects and consider birth cohort effects. In this study, compared with longitudinal analyses that separate out period effects and sectional age slopes give pictures different from those of that simple estimates of the age effect based on the cross-sectional finding.

Comparison of serum GGT with cholesterol is of particular interest, because serum cholesterol has been widely studied as one of the important cardiovascular disease risk factors, showing clear secular trends as countries have undergone epidemiologic transition (13, 14). In this study, serum cholesterol levels also increased in South Korean men, but only by 8 percent. The increase of only 8 percent might explain the secular trend in serum GGT, although the corresponding tendencies towards smoking and a sedentary lifestyle were inconsistent with the increase in serum GGT. However, even after adjustment for changes in these factors as time-dependent covariates, there was still a clear secular trend in serum GGT. Moreover, similar trends were observed among nondrinkers, subjects with a body mass index less than 25, subjects with an ALT level less than 35 units/liter, and subjects negative for hepatitis B surface antigen.

There may be concern as to whether an unspecified environmental hazard in the steel factory could explain the secular trend in serum GGT levels. However, the trend was observed at all worksites (data not shown); each worksite had environmental hazards specific to its work process. Moreover, a similar trend was observed even among office workers, who are unexposed to most environmental hazards. Thus, unspecified environmental hazards do not appear to explain the secular trend in serum GGT levels, enhancing the likelihood that the observed secular trend may be applied to the general population in South Korea.

Comparison of serum GGT with cholesterol is of particular interest, because serum cholesterol has been widely studied as one of the important cardiovascular disease risk factors, showing clear secular trends as countries have undergone epidemiologic transition (13, 14). In this study, serum cholesterol levels also increased in South Korean men, but only by 8 percent. The increase of only 8 percent might be slightly misleading, because the base value for serum cholesterol (177 mg/dl in 1996) was substantially higher than that for GGT (10.5 units/liter in 1996). A 1-percent increase in serum cholesterol may be interpreted as translating to a 1–2 percent increased risk of coronary heart disease (16, 17); an 8 percent increase in this risk is substantial. Nevertheless, the increase in GGT appeared to be
surprisingly greater than the increase in cholesterol, and the secular trend in serum cholesterol was statistically explained by adjustment for serum GGT. These observations would appear to indicate that GGT is an even more sensitive marker of epidemiologic transition than is serum cholesterol.

We did not collect dietary information and therefore could not evaluate the impact of dietary changes on GGT within this study. Nevertheless, we suspect that changes in the South Korean diet may be a key factor in the secular increase in serum GGT levels reported here. In recent decades, South Korea has undergone rapid economic growth. Socioeconomic development influences many factors that affect health, especially diet and nutrition. The traditional Korean diet is low in fat, cholesterol, animal protein, and sugar, high in total carbohydrate, and adequate in total protein. More foods are derived from vegetables than in comparable Western countries (18). Current changes in the Korean diet include a decreasing proportion of plant food, such as cereals and grains, and increasing proportions of animal food, such as meat, poultry, and meat products (10, 19–21). The nutritional transition in South Korea began to appear in the 1970s. It presented as a gradual pattern until around 1985, and after that it accelerated greatly (19). The nutritional transition paralleled changes in disease patterns (19). For example, age-adjusted rates of mortality from ischemic heart disease in 1999 were 3.8 and 3.6 times higher than the rates in 1984 among men and women, respectively (10). More recent data show that the mean intake per capita per day of meat, poultry, and meat products has continued to increase, from 47.3 g in 1990 to 56.0 g in 1995 and 91.7 g in 2001, while the decrease in cereal and grain intake has slowed down, from 344.0 g in 1990 to 308.9 g in 1995 and 310.5 g in 2001 (19, 21).

Supporting our speculation that GGT changes relate to a dietary transition, previous studies (22, 23) have shown that subsequent levels of serum GGT are greatly influenced by dietary factors; serum GGT decreased with a higher consumption of fruit or various plant foods, while serum GGT increased with a higher consumption of meat. Interestingly, when nutrients in plant foods and meat were examined, dietary constituents related to an oxidative stress mechanism were associated with serum GGT levels. For example, dietary antioxidants such as vitamin C and beta-carotene showed inverse associations with serum GGT level, and dietary heme iron was positively associated with serum GGT level. Free iron is a critical catalyst in generating oxidative stress (24), although it is uncertain whether a higher intake of heme iron is directly related to the presence of free iron.

Contrary to the trend in serum GGT, serum ALT levels appeared to decrease during the follow-up period. If the increase in serum GGT in this study were reflective of liver damage, ALT levels ought to have increased as well. However, such was not the case, implying that the secular trend in serum GGT does not primarily reflect a population-wide deterioration in liver function.

In summary, we found a strong and consistent increase in serum GGT level per calendar year in these South Korean men. Although we could not clearly explain why serum GGT levels dramatically increased during the 7-year follow-up period, on the basis of findings from previous studies, we suspect that an increase in oxidative stress due to dietary changes in this population might be a key factor. In any case, whatever the cause of this strong secular trend in serum GGT, the public health implication is a likely increase in the incidence of cardiovascular diseases and metabolic syndrome in the coming years, given that serum GGT has strongly predicted these diseases in previous studies.

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Conflict of interest: none declared.

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