Studies on the role of glycosphingolipids in metabolism

Langeveld, M.

Citation for published version (APA):
Chapter 6

OVERWEIGHT, INSULIN RESISTANCE AND TYPE II DIABETES IN TYPE I GAUCHER DISEASE PATIENTS IN RELATION TO ENZYME REPLACEMENT THERAPY

M. Langeveld, M. de Fost, J.M. Aerts, H.P. Sauerwein and C.E.M. Hollak

Blood Cells Molecules and Diseases 2008; 40:428-432
Abstract

Objectives and background
Type I Gaucher disease, a lysosomal storage disorder, is associated with metabolic abnormalities such as high resting energy expenditure, low circulating adiponectin concentrations and peripheral insulin resistance. Treatment with enzyme replacement therapy (enzyme therapy) leads to a decrease in resting energy expenditure, but its influence on weight and risk of development of type II diabetes is unknown.

Methods
We studied the BMI, prevalence of overweight, insulin resistance and type II diabetes in untreated and enzyme therapy treated Gaucher patients before and after several years of follow up and compared this to data on healthy subjects from literature.

Results
We established that in untreated Gaucher patients the prevalence of overweight is lower than in the general population. Long term treatment with enzyme therapy induces a larger than average weight gain leading to a similar prevalence of overweight in enzyme therapy treated patients and the general population. The prevalence of type II diabetes increases significantly during treatment with enzyme therapy, resulting in a comparable prevalence of type II diabetes in enzyme therapy treated patients and the general population.
Introduction

Type I Gaucher disease, a lysosomal storage disorder, can be successfully treated by enzyme replacement therapy (enzyme therapy). A recombinant form of the deficient enzyme, glucocerebrosidase, is administered intravenously, resulting in clearance of the storage material (glucocerebroside) from Gaucher macrophages. This leads to reversal or improvement of most symptoms related to the presence of these macrophages in liver, spleen and bone marrow, including hepatosplenomegaly, cytopenia and skeletal complications\textsuperscript{1,2}.

Apart from these classical symptoms, type I Gaucher disease is associated with several abnormalities in whole body metabolism. In untreated type I Gaucher disease the resting energy expenditure is increased up to 24-44\%\textsuperscript{3,4}. Children with type I Gaucher disease suffer from growth retardation and treatment with enzyme therapy leads to catch up growth in both length and weight\textsuperscript{5,6}. In adults enzyme therapy leads to partial correction of the hypermetabolism\textsuperscript{7}. Levels of plasma adiponectin, a cytokine produced by adipose tissue that is involved in regulating glucose homeostasis are lower in Gaucher patients than in matched control subjects\textsuperscript{8}. Low adiponectin levels are correlated with impaired insulin sensitivity\textsuperscript{9}. A recent hyperinsulinemic, euglycaemic clamp study showed Gaucher patients to be relatively insulin resistant compared to healthy control subjects\textsuperscript{10}.

In our experience (treating a cohort of 62 adult type I patients) long term treatment with enzyme therapy leads to an increase in body weight, resulting in overweight in many patients. In theory, the risk for developing type II diabetes of type I Gaucher patients that are treated with enzyme therapy is high, based on both the intrinsic insulin resistance and the induction of overweight by treatment with enzyme therapy. We therefore investigated the weight gain and the prevalence of overweight, insulin resistance and type II diabetes in our type I Gaucher patient cohort in relation to enzyme therapy.

Patients and methods

Cross sectional study
We performed a cross sectional study recording the prevalence of overweight, insulin resistance (as measured by homeostasis model assessment of insulin resistance) and type
II diabetes in Gaucher patients without a history of diabetes mellitus. In 2005 a total of 62 adult type 1 patients (49 treated with enzyme therapy) were seen at regular intervals at our outpatient clinic at the Academic Medical Center in Amsterdam. Of these 62 patients, 12 were not considered eligible for this part of the study: two because they are treated with substrate reduction therapy that may influence insulin sensitivity, four patients with severe co-morbidities (malignancies, severe cardiac disease) which hampered their participation, one patient with type I diabetes and five patients with type II diabetes mellitus. The latter five patients are included in the retrospective part of the study. Of the 50 eligible patients, 42 (7 untreated, 35 treated with enzyme therapy) agreed to participate. A fasting blood sample (fasting duration was approximately twelve hours) was obtained from each patient, followed by a study visit during which body weight and length were determined. Two patients chose to have their fasting blood glucose levels determined in a local hospital. Other laboratory values (e.g. insulin) are not available for these two patients. One patient agreed to participate but did not want to be weighed. The study protocol was approved by our hospital’s ethics committee. Informed consent was obtained from all participants.

Retrospective study
For the time period between 1991 and 2005, the number of type II diabetes patients in our cohort was recorded. Data on the time at which they were diagnosed with diabetes, the kind of treatment they received and their weight change were retrieved. Of the 35 enzyme therapy and 7 untreated patients that took part in the cross sectional study we collected historical data on their weight, Severity Scoring Index (SSI) and chitotriosidase before start of treatment.

Methods

Laboratory analysis
Chitotriosidase was determined by the standard enzyme activity assay with 4 MU-chitotriosidase (4-methylumbelliferyl b-d-N, N, N, N-tetraacetylchitotriose; Sigma, St Louis, MO) as substrate as previously described11. Chitotriosidase activity was doubled in carriers for the common chitotriosidase mutation (n=9)12. Plasma glucose was measured using
the HK/G-6PD method (Roche Diagnostics, Almere, the Netherlands). Plasma insulin concentration was determined by radioimmunoassay (RIA) (Insulin RIA 100, Pharmacia Diagnostic AB, Uppsala, Sweden): intra-assay coefficient of variation (CV) 3% to 5%, inter-assay CV 6% to 9%, detection limit 15 pmol/L. Homeostasis model assessment of insulin resistance ((fasting insulin* fasting glucose/22.5), with fasting insulin expressed in µU/ml and fasting glucose expressed in mmol/l) was used for detecting insulin resistant individuals. In a large study (2,321 individuals) comparing homeostasis model assessment of insulin resistance and hyperinsulinemic euglycaemic clamp (golden standard for determining insulin resistance) a cut-off value for the homeostasis model assessment of insulin resistance of 4.65 corresponded with insulin resistance (defined as an insulin stimulated whole body glucose disposal per kg lean body mass at an insulin infusion rate of 40 mU/min*m^2 of 28 µmol/min) with a sensitivity 84.9 % of and specificity of 78.7%\textsuperscript{13}.

Patients
Gaucher disease severity was assessed using the severity scoring index (SSI)\textsuperscript{14}. Change in weight over time, the incidence and prevalence of overweight (BMI>25 kg/m\textsuperscript{2}) and insulin resistance and type II diabetes were compared to data from cohort studies in healthy controls, comparable with respect to age and duration of follow-up. Obese subjects (BMI>30 kg/m\textsuperscript{2}) were not separately analyzed, but included in the overweight group since the prevalence of obesity in the Gaucher cohort was too low (n=2 in the cross sectional study). Weight loss and weight gain were defined as a consistent change in weight of more that 1 kg.

Statistical analysis
Results are given as median and range. For comparison between paired data (measurements before and after therapy), a Wilcoxon signed rank test was used. For comparison between groups (treated and untreated) a Mann-Whitney test was used. Correlations were calculated by using rank correlation (Spearman). In all cases, a p-value less than 0.05 was considered statistically significant. Comparison of incidences and prevalences in our cohort and data from population studies was done by calculating an absolute risk reduction (ARR).
Results

Cross sectional study
The median age of the enzyme therapy treated patients at the time of the cross sectional study was 49 (range 18 to 75 years, n=35). The median treatment duration in the enzyme therapy group was 11 years (range 1 to 14 years). The median BMI was 25.7 (range 19.0 to 30.4) and the prevalence of overweight 56%. The median fasting plasma glucose was 4.9 mmol/L (range 4.0 to 6.4) and the median plasma insulin concentration 50 pmol/L (range 20 to 141). The prevalence of insulin resistance (homeostasis model assessment of insulin resistance >4.65) was 6% (table 1). The median age in the untreated group at the time of the cross sectional study was 52 (range 35 to 57 years, n=7), with a median period of follow up of 8.2 years (range 1-13 years). The prevalence of overweight was 57%. The median fasting blood glucose level in this group was 5.2 mmol/L (range 4.3 to 6.0) and the median insulin level 53 pmol/L (range 17 to 111 pmol/L). The prevalence of insulin resistance was 0% in the untreated group. No new cases of type II diabetes were detected during this cross sectional analysis in both groups. There was no significant difference in fasting glucose and insulin levels between the treated and the untreated group (p=0.9 and 0.8).

Retrospective study
Before start of treatment, the disease severity was assessed as moderate to severe (SSI 13-17) in the enzyme therapy group (n= 35, median age 37 range 13 to 66 years) and the median chitotriosidase activity was 23,922 nmol/ml.hour (range 6,973-82,062). The median BMI before treatment in the enzyme therapy group was 23.3 (range 16.1-28.1). The BMI's of two patients were not included in this calculation because they were 13 and 15 years old when treatment was started. The prevalence of overweight before treatment was 16% (table 1). In the untreated group, the disease severity at the beginning of the follow up period was assessed as mild (SSI 1-6) and the median chitotriosidase activity was 6,459 (1,790-14,703). The median BMI at the beginning of follow up was 22.8 (range 20.7-26.3) kg/m². The prevalence of overweight at the beginning of follow up was 14%.
Table 1  Prevalence of overweight, insulin resistance and type II diabetes before and after enzyme therapy

<table>
<thead>
<tr>
<th>Patient nr</th>
<th>Age at diagnosis of type II diabetes</th>
<th>Years of enzyme therapy at diagnosis with type II diabetes</th>
<th>BMI (kg/m²)* (BMI at start enzyme therapy)</th>
<th>Weight gain during enzyme therapy (kg)*</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>51</td>
<td>no enzyme therapy</td>
<td>not known</td>
<td>not applicable</td>
</tr>
<tr>
<td>2</td>
<td>68</td>
<td>1.3</td>
<td>26.4 (25.4)</td>
<td>3</td>
</tr>
<tr>
<td>3</td>
<td>41</td>
<td>8.1</td>
<td>29.2 (23.7)</td>
<td>19</td>
</tr>
<tr>
<td>4</td>
<td>50</td>
<td>7.0</td>
<td>25.0 (22.5)</td>
<td>8</td>
</tr>
<tr>
<td>5</td>
<td>65</td>
<td>15.3</td>
<td>28.9 (23.8)</td>
<td>14</td>
</tr>
</tbody>
</table>

Table 2  Characteristics of type I Gaucher patients with type II diabetes mellitus. * Weight gain and BMI are measured at time of diagnosis with type II diabetes

Changes in weight and the development of overweight during follow up

Type I Gaucher patients that were treated with enzyme therapy gained an average 6 kg (range -8 to 29 kg) or 2.4 kg/m² (figure 1) in weight during 11 years of treatment, increasing the prevalence of overweight significantly from 16% to 56% (ARR 0.40, 95% CI 0.19-0.60) (table 1). In the enzyme therapy group, 19% remained stable in weight or lost weight during follow up. Of the five patients that lost weight (-1.3 to -8 kg), three did so as a result of a calorie restricted diet and one because of strenuous exercise following recovery form skeletal complications. In one patient no obvious cause for the weight loss could be found . Weight gain was not correlated to the duration of therapy (r=0.04, p=0.8), nor to response to therapy as measured by the relative decrease in chitotriosidase (r=0.12, p=0.5)(data not shown).

In the untreated group the median weight gain during 8 years of follow up was 3.8 kg (range -5 to 14 kg) or 2.2 kg/m². Two patients gained weight (12 and 14 kg) after starting treatment with antidepressive medication and one patient gained 10 kg in weight in six months after cholecystectomy because of improved appetite. Because of the small sample, no statistical test was performed.
Patients with diabetes mellitus

Five patients in our complete adult type I Gaucher patient cohort (n=62) were diagnosed with type II diabetes. One patient had mild Gaucher disease without therapy indication. Four patients developed diabetes during treatment with enzyme therapy, increasing the prevalence in the enzyme therapy treated patients significantly from 0% before treatment to 8.2% (4 out of 49) after a median eleven years of therapy (ARR 0.082, 95% CI 0.005-0.158) (table 1). The characteristics of these patients are described in table 2. The prevalence of type II diabetes at the time of the cross sectional study in our complete type I Gaucher patient cohort is 8.1% (5 out of 62). The 11 year cumulative incidence of type II diabetes in the complete cohort was 6.6% (4 out of 61) and in the enzyme therapy treated group 8.2% (4 out of 49).

Discussion

The assumption that bodyweight in Gaucher disease patients was lower before treatment and showed a steeper increase in response to enzyme therapy was further substantiated by comparing the outcome of this study with data from the literature and a national database. In a random sample of inhabitants of Doetinchem15, data on weight gain during a follow up period of eleven years were available for 4,810 individuals aged 20-59 years at inclusion (first
measurement in 1987 to 1991). The median weight gain in that study was 4.5 kg increasing the mean BMI from 24.6 at the first measurement to 26.2 kg/m² after follow up, resulting in an increase in prevalence of overweight (from 41% to 59%). The prevalence of overweight in the Gaucher population at baseline was significantly lower (ARR: 0.25, 95% CI 0.13-0.38), but after a median duration of 11 years of enzyme therapy, this difference disappeared (ARR 0.03, 95% CI -0.14-0.20). In 20,000 randomly selected individuals aged 20 years and older, retrieved from the Central Bureau of Statistics in the Netherlands (CBS), the prevalence of overweight was 45%, which is also not significantly different from the Gaucher patients after long term enzyme therapy (ARR 0.11, 95% CI -0.06-0.28).

Data from untreated type I Gaucher disease patients indicate that the prevalence of overweight at baseline as well as during follow up is not different from the treated population, but the number of patients followed does not allow a definite conclusion: there is also no significant difference at baseline with the control population. The median increase in weight was less than in the treated group, although again, this was not statistically significant. Since the untreated patients were less severely ill, presumably their weight gain follows the course of the control population. Thus, long term treatment with enzyme therapy induces a larger than average weight gain leading to a similar prevalence of overweight in enzyme therapy treated patients as compared to the general population. The actual gain in fat mass during enzyme therapy is probably an underestimation of the true increase, because excess liver and spleen volume (which consist in most untreated patients of several kilograms of storage cells) disappears as a result of treatment. This weight gain could be related to the decrease in resting energy expenditure during therapy⁷, speculating that patients do not adjust their caloric intake to their decreased energy expenditure.

Before treatment initiation, none of the patients in the enzyme therapy group were diagnosed with type II diabetes and no new cases of type II diabetes developed during follow up in the untreated group. During treatment with enzyme therapy, the prevalence of type II diabetes increased significantly. The prevalence of type II diabetes was further analyzed by comparison of our data to findings from the Hoorn study, a random sample of the Dutch population (50-74 years old, n = 2,484) that was screened for the presence of type II diabetes. The prevalence of type II diabetes was similar to that found in the Gaucher cohort (8.3%). However, in the Hoorn study the mean age was higher and the growing prevalence of type II diabetes with increasing age may have led to an under appreciation of the prevalence of
type II diabetes in the Gaucher cohort. Indeed, the prevalence of type II diabetes as found in the previously mentioned CBS prevalence report was lower: 4.3% in individuals aged 25 to 64 and 4.8% in 1663 individuals aged 40 to 55, followed for a decade starting in 1990\textsuperscript{17}. In addition, the 12 year cumulative incidence of type II diabetes in a large study in the UK (24,714 individuals aged 40 to 74) was even lower: 3.3%\textsuperscript{16}. The period in which the follow up was done in that study is more similar to the follow up period of the treated Gaucher cohort. Although no statistically significant differences can be established between the cumulative incidence of type II diabetes in the complete Gaucher cohort (or the enzyme therapy treated group) compared to these control populations, it is possible that this is due to the relatively small cohort in our study and larger studies are needed to clarify this issue.

Hypothetically, treatment with enzyme therapy could influence insulin sensitivity and therefore the likelihood of developing type II diabetes in the following manner. Untreated Gaucher patients are relatively insulin resistant\textsuperscript{10}. This might be related to the enhanced synthesis of the glycosphingolipid GM3, for which glucosylceramide is the precursor. GM3 concentrations are elevated in plasma of type I Gaucher patients (Ghauharali et al, chapter 4). GM3 has been shown to induce insulin resistance in vitro and in mouse models\textsuperscript{18,19} and inhibition of glycosphingolipid synthesis leads to normalization of insulin signaling\textsuperscript{20}. Treatment with enzyme therapy results in clearance of storage material and could therefore also lower GM3 production, leading to improvement of insulin sensitivity. The weight gain during treatment with enzyme therapy however induces overweight in a large number of patients, which once more induces insulin resistance. However, it is also possible that insulin resistance in obesity involves similar pathways as is hypothesized for untreated Gaucher patients. Although, the exact mechanism via which obesity induces insulin resistance is not known\textsuperscript{21} specific lipid metabolites (e.g. ceramides, GM3 ganglioside) may act as intermediates, providing a link between excess nutrients (i.e. saturated fatty acids), inflammatory cytokines and the induction of insulin resistance\textsuperscript{22}. Future research is needed to support or refute this possibility.

In conclusion, type I Gaucher patients that start treatment with enzyme therapy should be informed about the potential weight gain and be advised to adjust their diet when small weight changes start to occur so that occurrence of overweight and possibly also type II diabetes can be prevented.
References


