Neurological aspects of Gaucher and Fabry disease

Biegstraaten, M.

Citation for published version (APA):

General rights
It is not permitted to download or to forward/distribute the text or part of it without the consent of the author(s) and/or copyright holder(s), other than for strictly personal, individual use, unless the work is under an open content license (like Creative Commons).

Disclaimer/Complaints regulations
If you believe that digital publication of certain material infringes any of your rights or (privacy) interests, please let the Library know, stating your reasons. In case of a legitimate complaint, the Library will make the material inaccessible and/or remove it from the website. Please Ask the Library: http://uba.uva.nl/en/contact, or a letter to: Library of the University of Amsterdam, Secretariat, Singel 425, 1012 WP Amsterdam, The Netherlands. You will be contacted as soon as possible.
GENERAL INTRODUCTION
LYSOSOMAL STORAGE DISORDERS

The lysosomal storage disorders are a group of inherited disorders of metabolism, characterised by an accumulation of undegraded macromolecules in lysosomes. Depending on the type and nature of the accumulating substance, a wide variety of disorders can be distinguished. More than 50 diseases have now been described and several were already known long before the discovery of the lysosome by Christian de Duve in 1955\(^1\). The identification of this cellular compartment stimulated further research into its structure and function. Different lysosomal proteins were identified and were related to specific disease entities. Over the last two decades, further progress has been made in the understanding of the pathophysiology and treatment of lysosomal storage diseases. Although individually rare the lysosomal storage disorders as a group have a birth prevalence of about 14/100,000 live births\(^2\). The identification of attenuated forms with a more chronic disease course as well as the development of effective but costly therapeutic interventions make this disease group a major challenge for the health care system.

SPHINGOLIPOIDOSES

Sphingolipidoses are a subgroup of the lysosomal storage disorders. A deficiency of one of the lysosomal proteins involved in sphingolipid catabolism leads to lysosomal storage of sphingolipids and sometimes related substances. Sphingolipids are important components of the eukaryotic cell plasma membrane. They are characterised by the presence of a sphingoid base within the hydrophobic part of the molecule. In sphingomyelin and glycosphingolipids, a phosphorylcholine or a carbohydrate moiety is bound to the terminal hydroxyl group of ceramide, respectively (see Fig. 1). Glycosphingolipids display a high structural diversity and

**Figure 1** Structure of the glycosphingolipid GM3

the glycosphingolipid composition of membranes varies from one cell type to another, reflecting the diverse functions of glycosphingolipids. They are involved in cell adhesion, signal transduction and immunology, and interact with hormones, microbial toxins and other glycolipids.

The plasma membrane is continuously internalized and regenerated, thereby comprising a major endogenous source of the sphingolipids that are degraded in the lysosomes. Lipoproteins, rich in sphingolipids, constitute another source of sphingolipids for cellular degradation. Finally, in phagocytes like macrophages, phagocytic uptake of senescent/damaged cells results also in delivery of sphingolipids to lysosomes (see Fig. 2). The sphingolipid molecules are intralysosomally broken down in a highly ordered manner to ceramide, followed by degradation to sphingosine. Sphingosine is subsequently reutilized as backbone in sphingolipids or is metabolized to sphingosine-1-phoshate that may be degraded to hexadecenal and phosphoethanolamine.

The mixture of sphingolipids that is delivered to the lysosomes of a specific cell depends on the cell type and its function. Sulfatides, for instance, are almost exclusively synthesized by oligodendrocytes in the central nervous system where they comprise a major lipid component of the myelin sheath. Lysosomes of neurones are therefore exposed to large amounts of sulfatides, while lysosomes of other cell types are not.

Sphingolipids are degraded by lysosomal enzymes in the presence of sphingolipid activator proteins and other lipid transfer proteins. An inherited genetic defect of one of the lysosomal proteins involved in the breakdown of sphingolipids leads to an impaired lysosomal break down of the substrate. As a consequence, the substrate accumulates within the lysosome which results in cellular dysfunction and clinical abnormalities. The cell type that is most affected by the deficient enzyme activity and the nature of the storage material or related substances are believed to contribute importantly to the clinical phenotype of the sphingolipidosis. For instance, metachromatic leucodystrophy is caused by a

Figure 2 Delivery of sphingolipid substrate to lysosomes
mutation in the arylsulphatase A (ASA) gene. ASA is concerned with the breakdown of sulfatides. The major site of sulfatides is the myelin sheaths of central and peripheral neurons. ASA deficiency therefore leads to sulfatide accumulation in neurons resulting in severe demyelinisation and finally to death. In Gaucher disease, on the other hand, the enzyme glucocerebrosidase is deficient. This enzyme is concerned with the breakdown of glucosylceramide. In this disease, the macrophage is the cell that stores undegraded glucosylceramide after the phagocytic uptake of apoptotic blood cells which are rich in these lipids.

NEUROLOGICAL INVOLVEMENT IN THE SPHINGOLIPIDOSES

Within the sphingolipidoses, neurodegenerative disease is common. This varies from severe central nervous system involvement with a rapidly progressive course and early death as seen in type II Gaucher disease and infantile Sandhoff and Tay Sachs disease, to non-lethal peripheral nervous system involvement in Fabry disease. Niemann-Pick disease type B and Type I Gaucher disease are even considered to be non-neuronopathic, although the co-occurrence of type I Gaucher disease and parkinsonism and secondary neurological complications as a consequence of nerve root or spinal cord compression following vertebral collapse has been reported. Table 1 shows an overview of the neurological complications that are encountered in the various sphingolipidoses.

Unquestionably, the nervous system involvements in type I Gaucher disease and Fabry disease are less severe as compared to the central nervous system complications in type II Gaucher disease and infantile Sandhoff and Tay Sachs disease, but they still may be very disabling and therefore need attention. In this thesis the neurological complications that are encountered in type I Gaucher disease and Fabry disease will be discussed.

TYPE I GAUCHER DISEASE

Gaucher disease (OMIM 230800) is an autosomal recessively inherited glycosphingolipid storage disease with a prevalence of 1 in 57,000 live births. The disease is divided into three types, based upon the presence or absence and rate of progression of neurological manifestations. Type II is known as ‘acute neuronopathic’ or ‘infantile’ Gaucher disease, with infantile onset of severe central nervous system involvement leading to death usually by the age of two years. Type III is known as ‘chronic neuronopathic’ or ‘juvenile’ Gaucher disease, with an onset of central nervous system involvement in childhood, adolescence or early adulthood and a more indolent course. The distinction between type II and III Gaucher disease is made on the basis of age of onset and the rate of progression
<table>
<thead>
<tr>
<th>Disease</th>
<th>Enzymatic defect</th>
<th>Storage material</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gaucher disease</td>
<td>Glucocerebrosidase</td>
<td>glucosylceramide</td>
</tr>
<tr>
<td>GM1 gangliosidosis</td>
<td>β-Galactosidase</td>
<td>GM1</td>
</tr>
<tr>
<td>Tay-Sachs</td>
<td>β-Hexosaminidase α-subunit</td>
<td>GM2</td>
</tr>
<tr>
<td>Sandhoff</td>
<td>β-Hexosaminidase β-subunit</td>
<td>GM2</td>
</tr>
<tr>
<td>GM2 activator deficiency</td>
<td>GM2 activator protein</td>
<td>GM2</td>
</tr>
<tr>
<td>Krabbe</td>
<td>β-Galactocerebrosidase</td>
<td>galactosylceramide</td>
</tr>
<tr>
<td>Fabry disease</td>
<td>α-Galactosidase A</td>
<td>globotriaosylceramide</td>
</tr>
<tr>
<td>Metachromatic leucodystrophy</td>
<td>Arylsulfatase A</td>
<td>sulfatide</td>
</tr>
<tr>
<td>Niemann-Pick disease</td>
<td>Acid sphingomyelinase</td>
<td>sphingomyelin</td>
</tr>
<tr>
<td>Niemann-Pick disease type C</td>
<td>Cholesterol transporter</td>
<td>cholesterol, secondary accumulation of sphingomyelin and glycosphingolipids</td>
</tr>
</tbody>
</table>
### Neurological picture

<table>
<thead>
<tr>
<th>Disease</th>
<th>Enzymatic defect</th>
<th>Storage material</th>
<th>Neurological picture</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gaucher disease</td>
<td>Glucocerebrosidase</td>
<td>glucosylceramide</td>
<td>Type I: Characterised by haematological, visceral and skeletal disease, considered to be non-neuronopathic</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Type II: Onset of disease in early infancy; psychomotor delay, brainstem dysfunction, hypotonia and death in infancy</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Type III: Onset of disease in childhood, adolescence or early adulthood; severe systemic involvement and horizontal supranuclear palsy with or without psychomotor delay, hearing loss, and other brain stem deficits</td>
</tr>
<tr>
<td>GM1 gangliosidosis</td>
<td>β-Galactosidase</td>
<td>GM1</td>
<td>Infantile onset: Onset of disease in early infancy; delayed developmental milestones followed by progressive hypotonia, mental regression, seizures and death &lt; 2 yrs of age</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Juvenile onset: Onset of disease &lt; 2 yrs; ataxia and muscle weakness followed by mental regression, spasticity, myoclonic seizures and death by the age of 3-7 yrs</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Adult onset: Presents with gait and speech difficulties followed by progressive extrapyramidal disorder with prominent dystonia</td>
</tr>
<tr>
<td>Tay-Sachs</td>
<td>β-Hexosaminidase α-subunit</td>
<td>GM2</td>
<td>Infantile onset: Normal early development, presentation with hyperacusis and macular cherry red spots followed by developmental delay, seizures and blindness</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Juvenile onset: Onset of disease &gt; 2 yrs; mental regression with spasticity, dementia, blindness and seizures followed by death by the age of 10-15 yrs</td>
</tr>
<tr>
<td>Sandhoff</td>
<td>β-Hexosaminidase β-subunit</td>
<td>GM2</td>
<td>Infantile onset: Indistinguishable from Tay-Sachs disease, infantile type</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Juvenile onset: Indistinguishable from Tay-Sachs disease, juvenile type</td>
</tr>
<tr>
<td>GM2 activator deficiency</td>
<td>GM2 activator protein</td>
<td>GM2</td>
<td>Adult onset: Indistinguishable from Tay-Sachs disease, adult type</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Indistinguishable from Tay-Sachs disease, infantile type</td>
</tr>
<tr>
<td>Fabry disease</td>
<td>α-Galactosidase A</td>
<td>globotriaosylceramide</td>
<td>Infantile onset: Onset of disease in infancy; progressive psychomotor delay with irritability, seizures, optic atrophy and death &lt; 2 yrs of age</td>
</tr>
<tr>
<td>Metachromatic leucodystrophy</td>
<td>Arylsulfatase A</td>
<td>sulfatide</td>
<td>Late onset: Onset of disease in late childhood or early adolescence; vision problems, ataxia, progressive spastic tetraparesis, irritability, behavioural changes and sometimes mental regression with survival into adulthood</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Mild to severe systemic involvement with small fibre neuropathy and white matter lesions</td>
</tr>
<tr>
<td>Niemann-Pick disease type A</td>
<td>Acid sphingomyelinase</td>
<td>sphingomyelin</td>
<td>Infantile onset: Onset of disease &lt; 4 yrs; normal development until age of 12-18 months, followed by progressive hypotonia and mental regression and death in the first decade of life</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Juvenile onset: Onset of disease &gt; 4 yrs; gait disturbance followed by behavioural and cognitive decline, and ultimately by complete loss of developmental milestones by the age of 10 years.</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Adult onset: Onset of disease &gt; 16 yrs; psychiatric disturbances and loss of cognitive function followed by severe incapacity</td>
</tr>
<tr>
<td>Niemann-Pick disease type B</td>
<td>Cholesterol transporter</td>
<td>cholesterol</td>
<td>Type A: Onset of disease in early infancy; severe systemic involvement, hypotonia, muscular weakness, feeding problems and macular cherry red spots followed by mental regression and death &lt; 2 yrs of age</td>
</tr>
<tr>
<td></td>
<td></td>
<td>secondary</td>
<td>Type B: Characterised by hepatosplenomegaly and progressive pulmonary infiltration, considered to be non-neuronopathic</td>
</tr>
<tr>
<td></td>
<td></td>
<td>accumulation of</td>
<td>Onset of disease in childhood; clinical heterogeneity with systemic involvement, ataxia, dysarthria, dysphagia, vertical supranuclear palsy, gelastic cataplexy, seizures, psychomotor delay and life span related to age of onset</td>
</tr>
<tr>
<td>Niemann-Pick disease type C</td>
<td>Acid sphingomyelinase</td>
<td>sphingomyelin</td>
<td></td>
</tr>
</tbody>
</table>
of neurological manifestations. Unlike the rare type II and type III phenotypes, type I Gaucher disease, or non-neuronopathic Gaucher disease, usually has an onset in adolescence or early adulthood. The absence of nervous system involvement has been considered mandatory for a classification into this type.

Gaucher disease is caused by a genetic defect in the glucocerebrosidase (GBA) gene (EC 3.2.1.45) which has been mapped to chromosome 1q21. To date, over 250 mutations have been reported in GBA. These include missense mutations, nonsense mutations, small insertions or deletions that lead to either frameshifts or in-frame alterations, splice junction mutations, and complex alleles carrying two or more mutations in cis. Recombination events with a highly homologous pseudogene downstream of the GBA locus also have been identified, resulting from gene conversion, fusion, or duplication. The N370S and L444P mutations are the most prevalent disease-causing alleles. The phenotype-genotype correlation in Gaucher disease is far from perfect: there is significant genotypic heterogeneity among clinically similar patients, and there are vastly different phenotypes among patients with the same mutations. However, it has been recognised that the N370S mutation invariably leads to type I disease and that homozygosity for L444P is associated with central nervous system involvement.

The genetic defect leads to a deficient activity of GBA resulting in build-up of glucosylceramide almost exclusively in the lysosomes of macrophages (so called ‘Gaucher cells’). The predominance of this storage site may be explained by the extreme exposure of macrophagal lysosomes to glucosylceramide after the phagocytotic uptake of apoptotic blood cells which are rich in glycosphingolipids. The Gaucher cells are found preferentially in the spleen, liver and bone marrow. The most common manifestations include splenomegaly, hepatomegaly, anemia, thrombocytopenia, bone disease and growth retardation. In type II and III disease, there is also central nervous system involvement ranging from rapidly progressive and devastating neurological deterioration in type II disease to slowed horizontal
saccadic eye movements in type III patients. Brain pathology studies showed neuronal cell loss in specific hippocampal and cortex regions in type II and type III patients whereas patients classified as type I Gaucher disease had only astrogliosis\textsuperscript{10}. It is still unclear why some patients develop central nervous system damage where others do not, despite the same underlying defective gene, and in some instances even the same underlying genotype. The difference may be explained by an impairment of neuronal glycosphingolipid breakdown in types II and III disease: possibly, in type I Gaucher disease patients there is sufficient residual enzyme activity in the central nervous system to allow the degradation of glucosylceramide, while in the neuronopathic forms there is less residual enzyme activity leading to a greater accumulation inside the central nervous system. This would implicate that there is a threshold of enzyme activity below which the likelihood of developing neuronopathic disease is high. However, it has long been known that there is no close correlation between the residual enzyme activity or the amount of stored lipid and the patient’s phenotype\textsuperscript{11}. A second hypothesis is that a toxic metabolite is responsible for the nervous system damage in type II and III patients; glucosylsphingosine, a glycosphingolipid that is also degraded by glucocerebrosidase, is a highly cytotoxic compound that has been demonstrated to be elevated in spleen and liver samples of Gaucher patients of all types, while levels in brain samples were elevated only in those with neuronopathic forms\textsuperscript{12}. Furthermore, it has become increasingly clear that environmental factors and genetic modifiers are critical in defining the phenotype in many individuals\textsuperscript{13}.

Currently, two therapeutic approaches for the treatment of type I Gaucher disease are used: enzyme replacement therapy (ERT) and substrate reduction therapy (SRT) (see Fig. 3). In ERT treated patients, the reduced amount of enzyme is supplemented by recombinant enzyme which is administered intravenously every other week. The enzyme can reach the lysosomes from the extracellular space via endocytosis mediated by lectins on the plasma membrane, such as

<table>
<thead>
<tr>
<th>Disease</th>
<th>Enzymatic defect</th>
<th>Storage material</th>
<th>Neurological picture</th>
</tr>
</thead>
<tbody>
<tr>
<td>Farber disease</td>
<td>Acid ceramidase</td>
<td>ceramide</td>
<td>Onset of disease in early infancy; severe systemic involvement, psychomotor delay, dysphagia and death usually &lt; 2 yrs of age, but patients with milder forms can reach adulthood</td>
</tr>
<tr>
<td>Saposins 1 deficiencies</td>
<td>Sap-A</td>
<td>galactosylceramide</td>
<td>Similar to Krabbe disease, late onset</td>
</tr>
<tr>
<td></td>
<td>Sap-B</td>
<td>sulfatide, globotriaosylceramide, digalactosylceramide</td>
<td>Similar to metachromatic leucodystrophy, juvenile onset</td>
</tr>
<tr>
<td></td>
<td>Sap-C</td>
<td>glucosylceramide</td>
<td>Similar to type III Gaucher disease</td>
</tr>
<tr>
<td></td>
<td>Sap-D</td>
<td>ceramides</td>
<td>Human diseases caused by an isolated defect of Sap-D are unknown to date</td>
</tr>
<tr>
<td></td>
<td>LIMP2 deficiency</td>
<td>Lysosomal integral membrane type 2</td>
<td>Onset of disease in adolescence; progressive myoclonic epilepsy and nephrotic syndrome</td>
</tr>
</tbody>
</table>
the mannose 6-phosphate receptor and in case of Gaucher disease through the mannose receptor, resulting in degradation of the storage material. This approach has been proven to be very successful in the treatment of the visceral complications of the disease\textsuperscript{14-16}. Decreases in splenic and hepatic size and improvement in cytopenia are already apparent after 6 months of treatment\textsuperscript{16}. Currently, two different preparations have been approved by the authorities (EMA, FDA): imiglucerase, Cerezyme\textsuperscript{®}, Genzyme Corporation, Cambridge, MA, USA; and velaglucerase alfa, VPRIV\textsuperscript{®}, Shire Human Genetic Therapies, MA, USA. Drug approval applications for a third drug are in progress: taliglucerase alfa, Protalix Biotherapeutics, Carmiel, Israel. In general, these products have been tolerated very well\textsuperscript{17}. However, treatment costs of imiglucerase and velaglucerase are high with an estimate of 250,000 euro per patient (70 kg, 30U/kg every other week) per year.

While ERT increases the breakdown of the accumulated substrate, substrate reduction therapy aims at decreasing the amount of substrate by inhibiting its synthesis. The most well-known and studied compound which acts by SRT, the imunosugar N-butyldeoxynojirimycin (miglustat, Zavesca\textsuperscript{®}, Actelion Pharmaceuticals, Allschwil, Switzerland), is an inhibitor of glucosylceramide synthase, which catalyses the first step in the glucosylceramide biosynthetic pathway\textsuperscript{18}. Other forms of substrate reduction therapy are underway. Although miglustat has the advantage of oral administration and has been proven effective in the treatment of visceral aspects of Gaucher disease\textsuperscript{19}, it has not become the first choice treatment of Gaucher disease; miglustat is only indicated for the treatment of adult patients with mild to moderate type I Gaucher disease for whom enzyme replacement therapy is unsuitable. In addition, side effects such as diarrhoea, weight loss and tremors have been reported\textsuperscript{20-22}.

As mentioned in the first paragraph, type I Gaucher disease is traditionally considered to be non-neuronopathic and thus to be without nervous system

**Figure 3 Current therapies for Gaucher disease.**
involvement. However, an increasing number of reports emerged on central nervous system manifestations in patients with type I Gaucher disease, such as Parkinson disease and Lewy body dementia. Moreover, neuropathological and neuro-imaging studies also suggested the presence of central nervous system involvement in type I disease. Besides, peripheral nervous system involvement occurred in some patients: reports were published on the occurrence of polyneuropathy in type I Gaucher disease patients during a clinical trial with miglustat. A relationship to treatment was unconfirmed due to the absence of baseline neurological assessment. Following all these observations, the question arose whether the central and peripheral nervous system are involved after all in the non-neuronopathic type of Gaucher disease. The strict division in three different phenotypes became subject of debate.

To determine whether the three types of Gaucher diseases should be interpreted as a continuum rather than separate phenotypes, and to find out whether polyneuropathy is part of the natural course of type I Gaucher disease rather than a consequence of medication, studies on neurological complications in general and polyneuropathy in particular were needed. The studies in the first part of this thesis focus on this topic.

**FABRY DISEASE**

Fabry disease (OMIM 301500) is a glycosphingolipid storage disease with an estimated prevalence of 1 in 40,000 live births. The disease is caused by a genetic defect in the α-galactosidase-A gene which is located on the long arm of the X-chromosome (Xq22.1) and is transmitted in a recessive trait. Despite the X-linked inheritance pattern, significant numbers of females experience symptoms and signs possibly as a result of skewed X-chromosome inactivation or non-random lyonization. Over 200 different mutations have been identified including missense and nonsense mutations, splicing defects and small deletions. Some mutations result in a decreased enzyme activity, others in absent activity. Decreased or absent activity of α-galactosidase-A (EC 3.2.1.22) leads to the accumulation of glycosphingolipids, mainly globotriaosylceramide (Gb3), in lysosomes of various cell types, in particular endothelial and vascular smooth muscle cells, cardiomyocytes, kidney cells and sensory and autonomic ganglia of the peripheral nervous system. It has long been thought that the progressive accumulation of Gb3 in vascular endothelial cells is responsible for vascular dysfunction, and thereby for ischemic lesions in many organs. However, studies showed that the removal of accumulated glycosphingolipids from endothelial cells does not prevent progression of vascular disease in many patients. Apparently, other components contribute to the vascular damage. Histopathology of Fabry arteries showed that smooth muscle cell involvement with stored glycosphingolipids is the most prominent and probably the earliest
feature. The smooth muscle cells are hypertrophied as well. It has been shown that Fabry plasma is capable of stimulating proliferation of vascular smooth muscle cells. Further research revealed that Gb3 itself does not exert a stimulating effect, while the lyso-compound of Gb3, lyso-Gb3 (globotriaosylsphingosine), does induce smooth muscle cell proliferation in vitro. The precise mechanism by which lysoGb3 exerts its pathological effects has to be further elucidated.

It has been hypothesized that the smooth muscle cell involvement results in a decreased vascular wall compliance, which in turn may result in an upregulation of local renin-angiotensin systems. Such upregulation might lead to endothelial dysfunction and thereby to the Fabry vasculopathy.

Clinical manifestations of the disease include angiokeratomas, corneal opacities, renal failure, stroke, cardiomyopathy, cardiac rhythm disturbances, hypo- or anhidrosis, gastrointestinal complaints and painful acroparesthesias. Males are usually severely affected, while females show a more protracted course. Symptomatic treatment is offered to males as well as females, and includes neuropathic pain management with antiepileptic drugs such as carbamazepine. Although the efficacy of carbamazepine has been studied only scarcely, it has been widely used since improvement of neuropathic pain has been observed in most patients. In addition, antihistamines are used for the control of vertigo, antiplatelet drugs are used to prevent thrombotic complications, and ACE inhibitors are prescribed if microalbuminuria is present.

Two enzyme preparations are available for the treatment of Fabry disease: agalsidase alfa (Replagal®, Shire Human Genetic Therapies, Boston, MA, USA) and agalsidase beta (Fabrazyme®, Genzyme Corporation, Cambridge, MA, USA). The enzymes are both administered intravenously every other week and the presence of mannose-6-phosphate moieties in their N-linked glycans mediates uptake via the mannose-6-phosphate receptor, ubiquitously present on most cell types in various tissues. Treatment costs are about 230,000 euro per patient (70kg) per year. Effectiveness of ERT in Fabry disease is less spectacular than in Gaucher disease. Beneficial effects on neuropathic pain, renal function, cardiac pathology and bowel function have been observed. However, a number of patients suffer from recurrent complications despite treatment, ultimately leading to end organ failure or death. One of the major factors responsible for the variability in efficacy is the presence of irreversible organ damage. Also, the frequent development of neutralizing antibodies may play a role. Only male Fabry patients are prone to develop these antibodies as they often do not express any residual enzyme activity. Treating patients with a higher dose may be necessary to overcome neutralization of infused proteins. Alternatively, the institution of immuno-tolerance regimens could have a beneficial effect on the antibody formation. Methotrexate, for instance, has been shown to reduce antibodies to the infused enzyme in mice.

The cardiac rhythm disturbances, an- and hypohydrosis, gastrointestinal complaints and pain in Fabry disease are thought to be caused by small fibre
neuropathy as a result of accumulation of Gb3 in the vasa nervorum and/or accumulation in the dorsal root ganglia. To appreciate this hypothesis, it is important to know that within the peripheral nervous system several types of nerve fibres are being distinguished. In general, the peripheral nervous system comprises large and small diameter nerve fibres. Two groups of small nerve fibres are recognised: somatic small nerve fibres and autonomic small nerve fibres. Somatic small nerve fibres are divided into small thinly myelinated \( \text{A}_\delta \)-fibres carrying cold sensation, and small unmyelinated \( \text{C} \)-fibres carrying warm sensation. Both \( \text{A}_\delta \)- and \( \text{C} \)-fibres are involved in pain sensation. Autonomic nerve fibres consist of preganglionic small myelinated \( \text{B} \)-fibres and postganglionic small unmyelinated \( \text{C} \)-fibres, and are involved in the innervation and control of visceral organs, smooth muscle and secretory glands. To diagnose small fibre neuropathy and to get insight in the severity of small fibre damage, the following methods are used: autonomic function tests, quantitative sensory testing and skin biopsies. Autonomic function tests assess small nerve fibre function by determining the heart rate variability and blood pressure responses to several challenges, and quantitative sensory testing assesses small nerve fibre function by measuring the perception as well as pain thresholds for warm and cold. Skin biopsies are used as a method to quantify small nerve fibres in the epidermis; small unmyelinated nerve fibres are counted, representing both \( \text{A}_\delta \)-fibres that lose their myelin sheet before entering the dermis, and unmyelinated \( \text{C} \)-fibres.

Previous studies using quantitative sensory testing as well as intraepidermal nerve fibre counts in Fabry patients showed evidence of isolated small fibre neuropathy in this patient group. Since autonomic functions and pain are carried through small nerve fibres to the central nervous system, the cardiac rhythm disturbances, an- and hypohydrosis, gastrointestinal complaints and pain in Fabry disease have generally been attributed to small fibre neuropathy. However, the presence of autonomic neuropathy in Fabry patients has been studied only scarcely. Moreover, findings indicative of autonomic neuropathy in other diseases, such as orthostatic intolerance and male sexual dysfunction, have been infrequently reported in Fabry disease. Also, the relationship between small fibre neuropathy, pain and Fabry disease has not been firmly established so far: studies on the relation between age, disease severity, pain and severity of small nerve fibre damage have shown conflicting results. Thus, to gain further insight into the small fibre neuropathy in Fabry disease, additional investigations were needed. The studies in the second part of this thesis focus on the function and structure of small nerve fibres in Fabry patients.
AIMS OF THIS THESIS

The studies in section I deal with the neurological complications in type I Gaucher disease. The objective of this part of the thesis is to give an overview of the full neurological spectrum of the disease. For this purpose, we describe the results of a literature study on all peripheral and central nervous system complications in type I Gaucher disease (chapter 2). The results of a large multicentre study on the prevalence and incidence of polyneuropathy (chapter 3) and the cognitive profile (chapter 4) of patients with type I Gaucher disease are presented. To facilitate diagnosis and treatment, knowledge about possible neurological complications in type I Gaucher disease is important for patients, parents and the treating physicians.

The studies in section II of this thesis are aimed to gain further insight into small fibre neuropathy and its consequences in Fabry disease. Both nerve fibre function as assessed by quantitative sensory testing (chapter 6) and autonomic function tests (chapter 8), and nerve fibre structure as assessed by skin biopsies and quantification of intraepidermal nerve fibres (chapter 7), are addressed. Aside from acquiring knowledge about the clinical features of small fibre neuropathy, these studies are aimed to gain insight into the underlying pathophysiological mechanisms. Knowledge about the full neurological picture and insight into the underlying mechanism may lead to more appropriate clinical end-points in studies to the efficacy of therapy.

REFERENCE LIST

10. Wong K, Sidransky E, Verma A et al. Neuropathology provides clues to the pathophysiology of


