Chapter 1
Introduction
1.1. Introduction

The mucopolysaccharidoses (MPS) make up a group of 11 inherited metabolic diseases caused by a deficiency of lysosomal enzymes required to catalyze glycosaminoglycans (GAGs). An accumulation of these complex carbohydrates leads to the damage of cells, tissues and organ systems. Morquio syndrome is a condition belonging to this group and is further classified into mucopolysaccharidosis IVA and mucopolysaccharidosis IVB.

Mucopolysaccharidosis IVA (MPS IVA, Morquio A syndrome, OMIM 253000) is an autosomal recessive disorder caused by the deficiency of the enzyme activity of N-acetylgalactosamine-6-sulfatase (GALNS, OMIM 612222, EC 3.1.6.4), due to mutations in the GALNS gene (Tomatsu et al., 2005b). GALNS is essential for the degradation of GAGs, keratan sulfate (KS) (Glossl and Kresse, 1982; Tohru et al., 1982) and chondroitin-6-sulfate (C6S) (Matalon et al., 1974; Singh et al., 1976). This deficiency of GALNS leads to the accumulation of these substrates in lysosomes, which in turn disrupts metabolism and cell function, thereby causing tissue and organ dysfunction (Neufeld and Muenzer, 2001).

The MPS class has a specific distribution pattern of GAGs in tissues, which determine the clinical manifestation of the disease. The accumulation of KS and C6S results in clinical manifestation of disease in multiple organ systems including the skeletal and connective tissues (Montano et al., 2007b; Harmatz et al., 2013a). Patients diagnosed with MPS IVA have a wide spectrum of clinical manifestations including short trunk dwarfism, genu valgum, pectus carinatum, odontoid hypoplasia, kyphoscoliosis, platyspondyly, and hypermobility. In addition, patients may have hearing loss, corneal clouding, widely spaced teeth, a dysmorphic face, pulmonary dysfunction, cardiovascular abnormalities and hepatomegaly (Montano et al., 2007b; Hendriksz et al., 2013b). The diagnosis of MPS IVA is performed by clinical examination, skeletal radiographs, measuring GALNS activity in leucocytes or fibroblasts and the identification of pathogenic variations in the GALNS gene (Montano et al., 2007b; Tomatsu et al., 2011). Accurate and early diagnosis of MPS IVA is important for improving the quality of life for individuals living with this condition.

GM1 gangliosidosis (OMIM 230500) is a neurodegenerative disease inherited in an autosomal recessive manner and is caused by the deficiency of lysosomal enzyme β-
galactosidase (β-GAL, E.C.3.2.1.23). This enzyme catalyzes the hydrolysis of terminal β-galactose residues from substrates like glycoproteins, sphingolipids, and keratan sulfate (Okada and O'Brien, 1968). Pathogenic variations in the GLB1 gene coding for β-GAL cause the deficiency of this enzyme, which in turn leads to the accumulation of substrates in cells (Oshima et al., 1991).

Based on the age of onset and severity of the phenotype, GM1 gangliosidosis is classified into three clinical forms: type I, type II and type III (Brunetti-Pierri and Scaglia, 2008). Patients with type I, or the infantile form (OMIM 230500), are likely to have severe central nervous system (CNS) degeneration, cherry red spots, hepatosplenomegaly, and facial and skeletal abnormalities. Phenotype of the infantile form manifest from birth to six months of age, and death usually occurs before the age of two years. Patients with the infantile form of this disease also have cardiac involvement, which includes deficiencies in both β-GAL and β-GAL-related proteins. These proteins, also referred to as elastin binding protein (EBP), result from the alternative splicing of GLB1 (Privitera et al., 1998; Morrone et al., 2000; Caciotti et al., 2005b). The type II, or late infantile or juvenile form (OMIM 23060), is a slowly progressive neurological disorder characterized by early motor problems, muscle weakness and seizures, manifesting between seven months to three years of age. The type III, or adult form (OMIM 23650), usually starts between 3 to 30 years of age and is characterized by dystonia, cerebellar dysfunction, slurred speech, short stature and mild vertebral abnormalities (Brunetti-Pierri and Scaglia, 2008; Sperb et al., 2013).

The deficiency of β-GAL is also responsible for Morquio B syndrome (MPS IVB, OMIM 253010) which is characterized by short stature, skeletal abnormalities, and elevated urinary excretion of keratan sulfate without the involvement of the CNS. MPS IVB patients seem to show reduced catalytic activity for keratan sulfate but normal catalytic activity for GM1 ganglioside (Okumiya et al., 2003). However, the precise molecular mechanisms by which GM1 gangliosidosis and MPS IVB cause disease remain unclear (Brunetti-Pierri and Scaglia, 2008). MPS IVB is rare compared with MPS IVA and patients appear to have milder phenotypes when compared to those with MPS IVA (Arbisser et al., 1977).
The mutation spectrum of Morquio syndrome and GM1 gangliosidosis has been generated in different ethnic population and allelic heterogeneity has been observed in them. However, mutational studies are not available in Indian patients with Morquio syndrome and GM1 gangliosidosis, even though India has the second largest population in the world. For this reason, we aimed to study the mutation spectrum in GALNS and GLB1 in Indian patients with Morquio syndrome and GM1 gangliosidosis respectively using Sanger sequencing. We also employed systematic bioinformatics analysis of all the novel mutations identified in this study to evaluate their impact on protein function.