DEVELOPMENT OF NEUROPATHOLOGY IN MURINE MPS IIIA AND MPS VII AND THE EFFECT OF *N*-BUTYLDEOXYNOJIRIMYCIN TREATMENT ON MPS IIIA MICE

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Abstract

The mucopolysaccharidoses (MPSs) are a family of heritable diseases caused by deficiencies in glycosaminoglycan (GAG) degrading lysosomal enzymes. GAGs accumulate in a range of tissues, resulting in diverse pathology that includes brain degeneration. The secondary accumulation of glycosphingolipids, specifically G_{M2} and G_{M3} gangliosides, occurs in the MPS brain. In MPS IIIA and MPS VII mouse models GAGs and gangliosides began to accumulate prior to the onset of behavioural changes. G_{M2} levels began to rise early, following the trend of GAG accumulation, and increased to 548% and 219% of normal levels in MPS IIIA and MPS VII respectively. G_{M3} levels began to rise later, reaching a peak of 484% and 313% of normal in MPS IIIA and MPS VII respectively.

Given that brain G_{M2} and G_{M3} accumulation precedes behavioural deficits, it is possible that these gangliosides contribute to brain degeneration. Thus, gangliosides may be a target for the treatment of MPS brain disease. *N*-butyldeoxynojirimycin (*N*B-DNJ) is an iminosugar capable of crossing the blood brain barrier and reducing brain ganglioside synthesis, consequently deceasing overall brain G_{M2} and G_{M3} levels. *N*B-DNJ treatment of MPS IIIA mice decreased brain G_{M2} and G_{M3} levels in the short but not in the long term. Despite this, the innate fear response was restored and learning ability was equivalent to normal with both lengths of treatment.

MPS IIIA mice treated with *N*B-DNJ also had a reduction in cytokine gene expression, astroglial activation and oxidation of inflammatory lipids. Whether MPS IIIA behavioural improvements were due to a delay in ganglioside accumulation with *N*B-DNJ treatment, or due to an anti-inflammatory function of *N*B-DNJ is not known. However, this thesis

demonstrates that *N*B-DNJ can improve MPS brain dysfunction in the MPS IIIA mouse model and may be a potential therapy for CNS disease for children with MPS.

Declaration

This work contains no material which has been accepted for the award of any other degree or diploma in any university or other tertiary institution to Xenia Kaidonis and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where due reference has been made in the text.

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Abbreviations

12(s)-HETE	12(s)-hydroxyeicosatetraenoic acid
13(s)-HODE	13(s)-hydroxyoctadecadienoic acid
15(s)-HETE	15(s)-hydroxyeicosatetraenoic acid
6-keto-PGF1α	6-keto-prostogmandin F1 alpha
α Syn	alpha-synuclein
AA	arachidonic acid
AAV	adeno-associated virus
BBB	blood brain barrier
BLAST	basic local alignment search tool
BMT	bone marrow transplant
CB	cerebellum
Ccl3	chemokine (C-C motif) ligand 3
Cer	ceramide
CH	cerebral hemisphere
CL	cardiolipin
CNS	central nervous system
CS	chondroitin sulphate
CSF	cerebrospinal fluid
Ctsb	cathepsin B
Cypa	cyclophilin A
d_3-G_{M1}	N-octadecanoyl-d_3-monosialoganglioside 1
DAPI	4',6-diamidino-2-phenylindole
DHA	docosahexaenoic acid
DEAE	diethylaminoethyl

DS	dermatan sulphate
EDTA	sodium ethylenediaminetetraacetic acid
EPA	eicosapentaenoic acid
ERT	enzyme replacement therapy
FITC	fluorescein isothiocyanate
GAG	glycosaminoglycan
Gal	galactose
GalNAc	*N*-acetylgalactosamine
GalNAc T	*N*-acetylgalactosamine transferase
GalT I	galactosyltransferase I
GalT II	galactosyltransferase II
GFAP/ *Gfap*	glial fibrillary acidic protein
Glc	glucose
GlcA	glucuronic acid
GlcT	glucosyltransferase
G_M	monosialoganglioside
G_{M2}-AP	monosialoganglioside 2 activator protein
GSL	glycosphingolipid
GT	gene therapy
GUSB	β-glucuronidase
HA	hyaluronan
Hex A/B	hexosaminidase A/B
HIV	human immunodeficiency virus
HS	heparan sulphate
HSD	highly significant difference
IdoA	iduronic acid
Ifnγ	interferon gamma

Il1β	interleukin 1 beta
iso-PGF2α	iso-prostoglandin F2 alpha
KS	keratan sulphate
LC-ESI MS/MS	liquid chromatography- electrospray ionisation tandem mass spectrometry
LSD	lysosomal storage disorder
MCB	membranous cytoplasmic bodies
Mip1a	chemokine (C-C motif) ligand 3
MPA/B	mobile phases A/B
MPS	mucopolysaccharidosis
MRM	multiple reaction monitoring
MS/MS	tandem mass spectrometry
*N*B-DGJ	*N*-butyldeoxygalactonojirimycin
*N*B-DNJ	*N*-butyldeoxynojirimycin
NEB	New England Biolabs
NPC	Niemann Pick C
NSAID	non-steroidal anti-inflammatory drug
OCT	optimal cutting temperature
PA	phosphatidic acid
PBS	phosphate buffered saline
PC	phosphatidylcholine
PCR	polymerase chain reaction
PE	phosphatidylethanolamine
PGD2	prostoglandin D2
PGE2	prostoglandin E2
PGF2α	prostoglandin F2 alpha
PI	phosphatidylinositol

PUFA	polyunsaturated fatty acid
RAPC	repeated acquisition and performance chamber
ROS	reactive oxygen species
RVD1α	resolvin D1 alpha
SA	sialic acid
SAT I	sialotransferase I
SDS	sodium dodecyl sulphate
SDT	substrate deprivation therapy
SHIRPA	SmithKline Beecham, Harwell, Imperial College, Royal London Hospital, phenotype assessment
SM	sphingomyelin
SOD	superoxide dismutase
TAE	tris-acetate/ sodium ethylenediaminetetraacetic acid
TFA	trifluoroacetic acid
Tgfβ1	transforming growth factor beta 1
TLC	thin layer chromatography
Tnfα	tumor necrosis factor alpha
Tnfrs1α	tumor necrosis factor family receptor superfamily member 1a
TXB2	thromboxane B2
UA	uronic acid
UDP	uridine diphosphate