

Supplementary Tables to:

High glucosylceramides and low anandamide contribute to sensory loss and pain in Parkinson's Disease

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Abstract

BACKGROUND Parkinson's disease (PD) causes chronic pain in two-third of patients, in part originating from sensory neuropathies. **METHODS** The present observational study assessed associations of quantitative sensory tests (QST), pain ratings and questionnaires with plasma levels of multiple bioactive lipid species using untargeted and targeted lipidomic analyses. The study comprised two sets of patients and healthy controls (HC): the first 128 Israeli PD and 224 German young HC for exploration, the second 50/50 German PD and matched HC for deeper analyses. **RESULTS** The data show a 70% prevalence of PD pain and sensory neuropathies with a predominant phenotype of thermal sensory loss plus mechanical hypersensitivity. Multivariate analyses of lipids revealed major differences between PD and HC, mainly originating from glucosylceramides and endocannabinoids. GlcCer's were increased, whereas anandamide and lysophosphatidic acid LPA20:4 were reduced, stronger in patients with ongoing pain and with a linear relationship with pain intensity and sensory losses, particularly for GlcCer24:1 and GlcCer18:1. **CONCLUSIONS** GlcCer metabolism is mediated by glucocerebrosidase, GBA1, whose mutations are associated with PD and aggravate the toxicity of mutant synuclein. Our data suggest that PD-associated sensory neuropathies and PD-pain are in part caused by accumulations of glucosylceramides, raising the intriguing possibility to reduce PD pain and sensory loss by GBA1 refolding treatments.

Suppl. Table 1a

Demographic data German PD population: Pain VAS scores

	Healthy controls (HC)						Parkinson's Disease (PD)					
	Mean	female SD	Count	Mean	male SD	Count	Mean	female SD	Count	Mean	male SD	Count
Age (years)	60.56	7.10		65.28	9.00		64.06	8.64		68.15	7.75	
BMI	26.51	4.95		27.84	4.43		25.61	4.68		26.61	3.93	
Disease Duration (years)							6.00	5.55		8.85	7.48	
VAS Avg. pain	2.24	2.18		0.95	1.80		3.89	1.95		3.37	2.73	
VAS Pain now	1.06	1.75		0.36	1.15		1.75	1.81		0.94	1.80	
VAS maximum pain last	3.96	3.84		1.76	3.32		6.34	2.48		4.58	3.66	
VAS Quality of life impairment	1.44	2.07		0.48	0.98		3.25	2.84		2.26	2.43	
PD-associated Pain												
no pain			11			19			1			11
muscle			7			6			6			10
combined			5			0			1			5
neupath			2			0			0			2
dystonic			0			0			8			6
LDopa influence												
no			0			0			11			25
yes			0			0			5			8
does not apply			25			25			0			1

Suppl. Table 1b

Demographic data German PD population: Medication and Hoehn & Yahr

		Healthy controls (HC)						Parkinson's Disease (PD)						Sum HC Counts	Sum PD Counts
		female			male			female			male				
		Age Class			Age Class			Age Class			Age Class				
		<= 60 Count	61 - 70 Count	70+ Count	<= 60 Count	61 - 70 Count	70+ Count	<= 60 Count	61 - 70 Count	70+ Count	<= 60 Count	61 - 70 Count	70+ Count		
Levodopa	no	13	9	3	9	7	9	2	0	1	2	1	4	50	10
Carbidopa	yes							4	5	4	6	8	13		40
DA agonists	no	13	9	3	9	7	9	2	2	3	1	2	6	50	16
	yes							4	3	2	6	7	10		32
NMDA	no	13	9	3	9	7	9	6	4	5	7	7	11	50	40
antagonist	yes							0	1	0	0	2	5		8
MAOB	no	13	9	3	9	7	9	1	4	2	4	5	9	50	25
Inhibitors	yes							5	1	3	3	4	7		23
COMT	no	13	9	3	9	7	9	4	4	5	5	6	10	50	34
Inhibitors	yes							2	1	0	2	3	6		14
Anticholinergics	no	13	9	3	9	7	9	6	5	5	7	7	16	50	46
	yes							0	0	0	0	2	0		2
HoehnYahr	0.0	13	9	3	9	7	9	0	0	0	0	1	0	50	1
	1.0							2	2	1	0	0	2		7
	2.0							3	0	1	3	2	4		13
	2.5							0	1	2	0	1	4		8
	3.0							1	2	1	4	3	4		15
	4.0							0	0	0	1	2	3		6
Pain	no	7	3	1	7	6	6	0	0	1	4	4	4	30	13
	yes	6	6	2	2	1	3	6	5	4	4	5	13	20	37
Pain medication	no	11	6	1	7	4	4	3	3	4	6	5	9	33	30
	yes	2	3	2	2	2	5	2	2	1	2	4	8	16	19

Suppl. Table 1C

Demographic data German PD population: Pain VAS, QST class (based on summed z-scores), analgesic treatment

Counts		male		female		Sum HC	Sum PD	Percentages	
		HC	PD	HC	PD			Sum HC	Sum PD
Pain Questionnaire	no	19	12	11	1	30	13	60	26
	yes	6	22	14	15	20	37	40	74
QST Z-sum	no	17	15	14	9	31	24	62	48
PAIN	yes	8	19	11	7	19	26	38	52
QST sensory loss	no	13	9	19	7	32	16	64	32
	yes	12	25	6	9	18	34	36	68
Sensory loss and/or Pain	no pain, no sensory loss	9	1	11	3	20	4	40	8
	either loss or pain	12	22	11	10	23	32	46	64
	pain plus sensory loss	4	11	3	3	7	14	14	28
PHS hand	0	24	25	23	14	47	39	94	78
	1	1	3	0	1	1	4	2	8
	2	0	2	1	0	1	2	2	4
	3	0	4	1	1	1	5	2	10
PHS foot	0	17	12	18	5	35	17	70	34
	1	2	6	0	2	2	8	4	16
	2	0	5	2	4	2	9	4	18
	3	6	10	5	5	11	15	22	30
Analgesic	No	21	18	21	9	42	27	84	54
	Opioid	0	3	1	1	1	4	2	8
	NSAID or Metamizole	4	6	3	4	7	10	14	20
	Pregabalin or	0	2	0	0	0	2	0	4
	Antidepressants	0	4	0	2	0	6	0	12
	Benzodiazepines	0	1	0	0	0	1	0	2
Sensory Loss only						11	20	22	40
Sensory loss plus pain						7	14	14	28
Percentage receiving analgesic of those with pain according to questionnaire								40	62

Demographic data, Israel PD population versus young healthy controls

		Young Healthy controls (HC)						Parkinson's Disease (PD)					
		female			male			female			male		
		Mean	SD	Count	Mean	SD	Count	Mean	SD	Count	Mean	SD	Count
Age (years)		27	8	152	27	7	72	69	9	85	69	8	43
BMI		22.4	3.4		22.3	4.1		25.6	4.2		24.9	4.4	
Disease Years								8	5		9	6	
UPDRS								25	12		24	12	
Paintype	no pain			152			72			28			12
	muscle									37			23
	neupath									20			8
Analgesic	no			152			72			53			26
	cannabis									7			5
	opioid									3			1
	nsaid									2			1
	pregabalin									5			3
	antidepress									6			4
	BDZ									4			1
	anti leg cramps									5			2
	Levodopa	no									19		
Carbidopa	yes									66			33
DA Agonist	no									69			31
	yes									16			12
MAOB inhibitor	no									41			25
	yes									44			18
Amantadine	no									64			31
	yes									21			12
Anticholinergic	no									59			36
	yes									26			7

Suppl. Table 2

Descriptive statistics of raw QST data, German PD population

	Healthy controls (HC)										Parkinson's Disease (PD)									
	female					male					female					male				
	Mean	Standard Deviation	Median	Percentile 25	Percentile 75	Mean	Standard Deviation	Median	Percentile 25	Percentile 75	Mean	Standard Deviation	Median	Percentile 25	Percentile 75	Mean	Standard Deviation	Median	Percentile 25	Percentile 75
CDT hand (°C)	29.50	2.72	30.53	29.43	30.83	29.32	3.30	30.30	29.20	30.77	29.82	1.32	30.12	28.73	30.87	27.93	5.31	29.62	28.27	30.27
WDT hand (°C)	35.39	2.28	34.77	33.73	36.30	37.88	3.79	36.63	34.93	39.37	36.61	4.55	34.70	33.70	37.14	39.50	4.86	38.43	35.20	42.90
CPT hand (°C)	19.46	8.44	23.97	15.77	25.57	16.70	9.01	18.60	7.60	24.50	18.96	9.78	23.70	8.95	27.17	15.41	9.42	19.18	6.67	22.57
HPT hand (°C)	41.60	3.87	41.23	38.33	44.53	44.34	3.78	44.13	40.90	47.30	41.74	4.51	41.20	38.69	45.62	45.74	4.34	47.20	43.40	49.40
TSL hand (°C)	6.44	5.15	4.90	3.30	8.10	8.33	4.65	7.37	5.13	8.57	8.36	5.69	6.35	3.74	11.00	13.69	10.95	9.00	6.63	18.57
MDT hand (mN)	0.73	0.73	0.44	0.25	0.89	1.06	1.02	0.71	0.33	1.32	2.60	5.56	0.98	0.26	1.90	2.58	3.37	1.32	0.57	2.59
MPT hand (mN)	104.62	123.57	55.72	25.99	111.43	217.25	234.58	84.45	68.59	315.17	177.11	227.32	66.62	17.18	350.86	159.80	219.78	68.76	13.93	194.01
VDT hand (1/8 Hz)	7.67	0.58	8.00	7.67	8.00	7.25	0.83	7.67	6.33	8.00	7.17	1.16	7.75	6.84	8.00	6.71	1.22	7.00	5.67	8.00
PPT hand (kPa)	31.20	9.35	30.03	25.07	39.70	46.54	15.94	44.97	37.63	51.90	34.60	9.44	36.70	26.65	39.47	39.78	10.20	38.12	34.43	46.77
CDT foot (°C)	28.46	3.25	29.43	27.87	30.30	24.00	8.22	27.20	24.83	29.43	26.73	6.78	29.45	26.17	29.99	20.13	9.90	23.67	11.40	27.83
WDT foot (°C)	38.26	3.52	37.40	35.90	39.93	41.31	4.10	41.93	38.23	44.47	41.37	4.73	42.42	37.64	45.49	43.89	3.71	44.32	41.57	46.00
CPT foot (°C)	17.79	9.44	23.53	11.07	24.63	13.26	9.51	15.40	5.30	20.43	15.22	10.81	18.68	3.15	25.20	11.65	9.02	12.67	1.00	18.73
HPT foot (°C)	44.49	2.91	44.37	43.03	46.97	46.37	2.61	46.63	44.53	48.70	46.07	3.32	46.77	44.99	48.10	47.72	2.72	48.14	46.40	49.63
TSL foot (°C)	12.17	5.76	11.80	8.00	15.40	18.69	11.82	15.43	11.57	20.03	19.37	7.44	20.29	12.23	22.89	25.55	11.62	21.67	16.82	28.90
MDT foot (mN)	1.56	1.43	1.15	0.47	1.76	2.18	2.24	1.62	0.50	2.83	2.89	2.46	1.94	1.11	4.40	5.11	3.92	4.40	2.46	6.50
MPT foot (mN)	92.97	87.01	68.59	27.86	111.43	119.70	132.88	78.79	55.72	128.00	104.95	176.51	36.85	12.44	132.60	99.58	137.13	55.85	10.56	101.30
VDT foot (1/8 Hz)	7.01	1.33	7.67	6.00	8.00	6.15	1.52	6.33	5.00	7.83	5.15	2.60	5.50	3.83	7.34	4.89	2.18	5.00	3.83	6.00
PPT foot (kPa)	45.20	8.00	45.67	42.00	50.04	54.88	14.09	50.04	46.00	65.67	44.64	9.79	46.06	36.00	51.34	46.73	9.78	46.06	43.67	48.67

Suppl. Table 4

Mean lipid ratios of German PD patients versus healthy controls, categorized according to medication No consistent effect of any medication

	Levodopa/Carbidopa		DA agonists		NMDA antagonist		MAOB Inhibitors		COMTInhibitors		PainMedication		Number of PD drugs	Number of patients				Mean of Controls set to 1
	no	yes	no	yes	no	yes	no	yes	no	yes	no	yes		1	2	3	4-5	
Biopt	1.03	1.05	1.03	1.04	1.03	1.09	1.04	1.04	1.01	1.11	1.04	1.04	10	1.94	2.20	2.64	3.25	1.0
Neopt	0.97	1.08	1.08	1.05	1.04	1.13	1.08	1.03	1.02	1.15	1.05	1.08	15	1.04	1.03	0.99	1.08	1.0
Pterin	1.00	1.06	1.05	1.04	1.04	1.11	1.06	1.03	1.01	1.13	1.05	1.06	14	1	1.02	1.1	1.1	1.0
AEA	0.84	0.82	0.85	0.81	0.83	0.76	0.83	0.82	0.84	0.78	0.82	0.84	9	1.02	1.02	1.05	1.09	1.0
OEA	0.95	0.86	0.96	0.85	0.88	0.88	0.86	0.91	0.88	0.89	0.83	0.96	1	0.88	0.83	0.85	0.72	1.0
PEA	0.91	0.92	1.05	0.85	0.93	0.86	0.90	0.94	0.91	0.94	0.88	0.97	1	0.9	0.89	0.89	0.83	1.0
AG12	0.60	0.80	0.87	0.70	0.75	0.82	0.69	0.83	0.74	0.81	0.80	0.73	1	0.97	0.94	0.92	0.84	1.0
S1P	0.98	0.96	1.03	0.93	0.98	0.89	0.92	1.01	0.96	0.97	0.97	0.94	1	0.88	0.88	0.88	0.88	1.0
SA1P	0.94	1.00	1.02	0.98	1.02	0.86	0.95	1.04	0.98	1.01	1.02	0.93	1	0.99	0.99	0.96	1.05	1.0
C16Sphingarin	0.87	0.93	1.00	0.88	0.93	0.90	0.91	0.93	0.91	0.96	0.89	0.96	1	0.99	0.99	0.96	1.05	1.0
C24Sphingarin	0.81	0.99	1.12	0.87	1.00	0.71	0.89	1.03	0.96	0.94	1.01	0.88	1	0.93	0.92	0.93	0.91	1.0
C241Sphingarin	0.86	1.22	1.48	0.98	1.18	0.99	1.07	1.24	1.13	1.19	1.14	1.18	1	0.89	0.89	0.84	0.79	1.0
C16Cer	1.10	1.11	1.29	1.02	1.11	1.12	1.02	1.21	1.09	1.17	1.07	1.16	1	0.96	0.96	0.96	0.96	1.0
C18Cer	1.06	1.04	1.28	0.92	1.05	1.00	0.96	1.13	1.05	1.03	0.99	1.13	1	0.97	0.97	0.99	1.13	1.0
C20Cer	0.95	1.23	1.52	0.99	1.18	1.10	1.08	1.26	1.06	1.42	1.08	1.34	1	1.07	1.07	1.14	1.11	1.0
C22Cer	0.80	1.04	1.16	0.90	1.00	0.97	0.94	1.05	0.91	1.20	0.98	1.02	1	1.14	1.14	1.14	1.11	1.0
C24Cer	0.83	1.06	1.19	0.91	1.00	1.00	0.96	1.05	0.90	1.24	0.99	1.05	1	1.13	1.07	1.14	1.11	1.0
C241Cer	0.94	1.17	1.32	1.00	1.13	0.99	1.08	1.13	1.03	1.28	1.06	1.22	1	1.14	1.14	1.14	1.12	1.0
C16GlcCer	1.11	1.38	1.65	1.16	1.32	1.35	1.30	1.36	1.17	1.71	1.19	1.56	1	1.06	1.06	1.14	1.12	1.0
C18GlcCer	1.19	1.38	1.78	1.13	1.36	1.29	1.28	1.42	1.21	1.68	1.21	1.56	1	1.17	1.17	1.17	1.44	1.0
C181GlcCer	1.30	1.50	1.72	1.33	1.45	1.52	1.52	1.41	1.30	1.86	1.33	1.69	1	1.27	1.4	1.23	1.4	1.0
C241GlcCer	1.24	1.45	1.68	1.27	1.41	1.39	1.41	1.41	1.30	1.67	1.25	1.65	1	1.27	1.4	1.23	1.4	1.0
C16LacCer	1.00	1.14	1.31	1.00	1.12	1.04	1.16	1.04	1.06	1.21	1.10	1.22	1	1.39	1.37	1.4	1.74	1.0
C18LacCer	0.96	1.19	1.52	0.96	1.17	1.00	1.14	1.14	1.11	1.22	1.03	1.21	1	1.17	1.31	1.33	1.44	1.0
C181LacCer	0.91	1.15	1.31	1.01	1.13	1.01	1.16	1.06	1.05	1.26	0.93	1.35	1	1.27	1.4	1.23	1.4	1.0
C24LacCer	0.85	0.99	1.18	0.87	1.01	0.81	0.94	1.00	0.97	0.99	0.94	1.00	1	1.06	1.23	1.01	1.05	1.0
DHET14_15	0.91	0.87	0.88	0.83	0.87	0.74	0.89	0.80	0.89	0.73	0.89	0.85	1	1.2	1.23	1.04	1.06	1.0
DHET5_6	1.03	0.80	0.92	0.80	0.85	0.76	0.79	0.89	0.84	0.84	0.84	0.86	1	0.95	1.13	1.29	0.96	1.0
DHET8_9	0.91	0.79	0.82	0.79	0.82	0.70	0.80	0.80	0.82	0.74	0.82	0.80	1	0.96	1.09	0.94	0.85	1.0
LPA16_0	1.01	0.95	0.97	0.95	0.97	0.89	0.94	0.97	0.95	0.96	0.97	0.94	1	1.017	0.857	0.791	0.793	1.0
LPA18_0	0.99	0.97	0.98	0.96	0.98	0.90	0.94	0.99	0.95	1.00	0.99	0.95	1	0.881	0.897	0.799	0.83	1.0
LPA18_1	1.10	1.05	1.05	1.08	1.08	1.02	1.04	1.10	1.05	1.11	1.06	1.05	1	0.859	0.793	0.771	0.794	1.0
LPA18_2	0.93	0.93	0.91	0.95	0.95	0.88	0.92	0.97	0.96	0.90	0.97	0.86	1	0.96	0.95	0.95	0.93	1.0
LPA20_4	0.86	0.83	0.86	0.84	0.85	0.81	0.80	0.89	0.84	0.86	0.82	0.85	1	0.97	1.07	1.06	1.11	1.0
EpOME12_13	1.64	1.37	0.73	1.76	1.51	0.94	1.40	1.43	1.71	0.70	1.85	0.80	1	0.9	1.01	0.87	0.93	1.0
DiHOME9_10	0.82	0.78	0.54	0.92	0.82	0.71	0.68	0.92	0.85	0.67	0.90	0.63	1	0.78	0.84	0.9	0.86	1.0
EpOME9_10	1.64	0.90	0.87	1.16	1.12	0.76	0.80	1.35	1.20	0.73	1.14	0.96	1	1.007	2.173	1.271	1.054	1.0
DiHOME12_13	1.15	1.13	0.67	1.38	1.15	1.10	0.98	1.32	1.21	0.99	1.32	0.87	1	0.51	1.139	0.715	0.901	1.0
HODE13	1.10	1.45	0.79	1.69	1.35	1.59	1.32	1.46	1.53	1.05	1.66	0.97	1	0.919	1.318	1.118	0.745	1.0
HODE9	0.87	1.12	0.72	1.23	1.03	1.24	0.98	1.15	1.09	1.00	1.14	0.97	1	0.783	1.319	1.066	1.608	1.0
HETE5	0.81	0.71	0.86	0.68	0.77	0.58	0.67	0.81	0.74	0.74	0.68	0.80	1	0.917	1.683	1.307	1.79	1.0
HETE12	0.97	1.19	1.08	1.20	1.22	0.86	1.06	1.27	1.28	0.88	1.23	1.01	1	0.87	1.061	1.079	1.474	1.0
HETE15	0.99	0.80	0.87	0.81	0.85	0.76	0.83	0.83	0.85	0.79	0.84	0.81	1	0.828	0.774	0.804	0.603	1.0
													1	1.024	1.277	1.385	0.753	1.1
													1	0.99	0.881	0.706	0.743	1.0

Abbreviations

General abbreviations

BMI	Body mass index
PD	Parkinson's Disease
GBA1	Glucocerebrosidase

QST parameter

CDT, WDT	Cold and warm detection threshold
CPT, HPT	Cold and heat pain threshold
MDT, MPT	Mechanical detection and pain threshold
VDT	Vibration detection threshold
PPT	Pressure pain threshold
TSL	Thermal sensory limen

Key lipids

Cer	Ceramide
GlcCer	Glucosylceramide
LacCer	Lactosylceramide
AEA	Anandamide
LPA	Lysophosphatidic acid

Statistics

ANOVA	Analysis of variance
CanDisc	Canonical discriminant analysis
CI	Confidence interval

Abbreviations of analytes

Biopt		Bioperin
Neopt		Neopterin
AEA		Anandamide
OEA		Oleoylethanolamide
OEAVEA		Oleoylethanolamide -Vaccenic acid ethanolamide
PEA		Palmitoylethanolamide
AG12		1/2-Arachidonoylglycerol
Sphosin		Sphingosin
S1P		Sphingosin-1-Phosphate
SA1P		Sphingarin-1-Phosphate
C16Cer	Cer16	Ceramide with 16 C-atoms
C18Cer	Cer18	Ceramide with 18 C-atoms
C20Cer	Cer20	Ceramide with 20 C-atoms
C22Cer	Cer22	Ceramide with 22 C-atoms
C24Cer	Cer24	Ceramide with 24 C-atoms
C24:1Cer	Cer24:1	Ceramide with 24 C-atoms, one unsaturated bond
C16GlcCer	GlcCer16	Glucosylceramide, 16C
C18GlcCer	GlcCer18	Glucosylceramide, 18C
C24:1GlcCer	GlcCer24:1	Glucosylceramide, 24C, one unsaturated bond
C16LacCer	LacCer16	Lactosylceramide, 16C
C18LacCer	LacCer18	Lactosylceramide, 18C
C24LacCer	LacCer24	Lactosylceramide, 24C
C24:1LacCer	LacCer24:1	Lactosylceramide, 24C, one unsaturated bond
C18:1LacCer	LacCer18:1	Lactosylceramide, 18C, one unsaturated bond
LPA16:0		Lysophosphatidic acid 16C
LPA18:0		Lysophosphatidic acid 18C
LPA18:1		Lysophosphatidic acid 18C, one unsaturated bond
LPA18:2		Lysophosphatidic acid 18C, two unsaturated bonds
LPA20:4		Lysophosphatidic acid 20C, four unsaturated bonds
DHET		Dihydroxyeicosatrienoic acid
EpOME		Epoxy-octadecenoic acid
HODE		Hydroxyoctadeca-dienoic acid
HETE		Hydroxyeicosatetraenoic acid