

Supplementation with *Bifidobacteria longum* subspecies *infantis* EVC001 for mitigation of type 1 diabetes autoimmunity - The GPPAD-SINT1A randomized controlled trial protocol

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Supplementary File 1: GPPAD-SINT1A Study: Visit-schedule (Study Flow Chart)

Visits	Intervention				
	Baseline Visit age 7 days - 6 weeks	Call age 3month s	Visit age 6 months	Call age 9 months	Visit age 12 months
Visit window	+14d	- 14d	± 14d	± 14d	+ 14d
Study visit	1		2		3
Study call		1		2	
Informed consent, Review Incl./Excl. Criteria	X				
Randomization	X				
Medical History	X				
Intervention					
Dispense supplement and compliance data sheet (<i>do not administer supplement at study site, only at home</i>)	X		X		
Local investigations & measurements					
Physical examination (height, weight)	X		X		X
Assessment of AEs and SAEs ^A		X	X	X	X
Assessment of rotavirus and MMR vaccination schedule using official records			X		X
Blood glucose ^B			X		X
HbA1c					X
Sample collection					
<200 µl capillary or venous blood for glucose			X		X
< 1ml EDTA blood for HbA1c					X
2ml blood for serum samples for central antibody measurement ^{C, D}			X		X
2ml EDTA blood for plasma samples for mechanistic studies (inflammation, metabolomics)			X		X
2 ml EDTA blood for DNA sample ^C					X

	Intervention				
	Baseline Visit age 7 days - 6 weeks	Call age 3month s	Visit age 6 months	Call age 9 months	Visit age 12 months
Visits					
Stool sample for microbiome 16S	X	X	X		X
Stool sample for colonization		x			
Stool sample for stool pH & calprotectin (in selected participants)			X		

Central measurements					
IAA; GADA; IA-2A; ZnT8RA; ZnT8WA			X		X
TGA			X		X
Stool PCR for B. infantis colonization		X			
Antibody responses to rotavirus vaccine			X		
Microbiome 16s ^E	X	X			X
Mechanistic markers (inflammation, metabolomics) ^E			X		X
Electronic questionnaires completed by families					
Questionnaire about breast-feeding and antibiotics	every 2 weeks until age 12 months				
Questionnaire about infections and vaccinations	every 2 weeks until age 12 months				
Questionnaire about allergies					X
Ancillary assessments					
Whole blood FACS ^F (Dresden and Munich only)			X		X

^A AEs/SAEs will be noted and reported as under intervention phase for 30 days after end of treatment day

^B by handmeter or haemocue

^C if there is left over material and a signed biobank consent, the left over serum and DNA will be stored in the IBBL or local biobank

^D venous or capillary blood for the AAB confirmation sample can be obtained by a local physician

^E measurements may partly be done as exploratory project after unblinding and analysis of main outcomes

^F to assess maturation of immune cell composition and response

	Follow-up (minimum 2.5 years; maximum up to 5.5 years after end of intervention)			
	Call age 18 months	Visit age 2 years	Call every 12 month (in the middle of yearly visits)	Visit every 12 months ^G
Visits				
Visit window	± 30d	± 30d	± 30d	± 30d
Study visit		4		5+
Study call	3		4+	
Local investigations and measurements				
Physical examination (height, weight)		X		X
Assessment of AEs and SAEs ^A	X			
Assessment of MMR vaccination schedule using official records		X		
Blood glucose ^B		X		X
Sample collection				
<200 µl capillary or venous blood for glucose		X		X
2ml blood for serum samples for central antibody measurement ^{C, D}		X		X
2ml EDTA blood for plasma samples for mechanistic studies (inflammation)		X		
Central measurements				
IAA; GADA; IA-2A; ZnT8RA; ZnT8WA		X		X
TGA		X		X
Antibody Responses to MMR vaccine		X		
Mechanistic markers (inflammation) ^E		X		
Electronic questionnaires completed by families				
Questionnaire about allergies	every 12 months until end of study			
Ancillary assessments				
Whole blood FACS (<i>Dresden and Munich only</i>)		X		

^A AEs/SAEs will be noted and reported as under intervention phase for 30 days after end of treatment day

^B by handmeter or haemocue

^C if there is left over material and a signed biobank consent, the left over serum and DNA will be stored in the IBBL or local biobank

^D venous or capillary blood for the AAB confirmation sample can be obtained by a local physician

^E measurements may partly be done as exploratory project after unblinding and analysis of main outcomes

^F to assess maturation of immune cell composition and response

^G Final visit must be performed within the last 6 months before last enrolled child completed 2.5 years of follow-up