

Supplementary Appendix 1

Gut microbiome diversity measures for metabolic conditions: A systematic scoping review

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Supplementary File 1. Full search strategy

Set #	PubMed	Results
1	"gastrointestinal microbiome"[MeSH Terms] OR ((gut[tiab] OR intestin*[tiab] OR gastrointestinal*[tiab] OR colon*[tiab] OR rectal[tiab] OR rectum[tiab] OR stool[tiab] OR feces[tiab] OR faeces[tiab] OR fecal[tiab] OR faecal[tiab]) AND ("microbiota"[MeSH Terms] OR "mycobiome"[MeSH Terms] OR microbiome*[tiab] OR microbiota*[tiab] OR microbial[tiab] OR microbe*[tiab] OR microflora*[tiab] OR flora*[tiab] OR microorganism*[tiab] OR pathobiont*[tiab] OR mycobiome*[tiab] OR mycobiota*[tiab] OR virome*[tiab] OR phylotype*[tiab])) OR enterotype*[tiab]	89194
2	diversit*[tiab] OR abundan*[tiab] OR richness*[tiab] OR evenness*[tiab] OR dissimilarit*[tiab]	489439
3	#1 AND #2	16344
4	animals[MeSH Terms] NOT humans[MeSH Terms]	4682770
5	#3 NOT #4	11807
6	English[lang]	26086457
7	#5 AND #6	11634
8	"2019/01/01"[PDAT] : "2019/12/31"[PDAT]	1402266
9	#7 AND #8	3111

Set #	Embase	Results
1	'intestine flora'/exp OR (('gut':ti,ab OR 'intestin*':ti,ab OR 'gastrointestin*':ti,ab OR 'colon*':ti,ab OR 'rectal':ti,ab OR 'rectum':ti,ab OR 'stool':ti,ab OR 'feces':ti,ab OR 'faeces':ti,ab OR 'fecal':ti,ab OR 'faecal':ti,ab) AND ('microbiome'/exp OR 'microflora'/exp OR 'mycobiome'/exp OR 'microbiome*':ti,ab OR 'microbiota*':ti,ab OR 'microbial':ti,ab OR 'microbe*':ti,ab OR 'microflora*':ti,ab OR 'flora*':ti,ab OR 'microorganism*':ti,ab OR 'pathobiont*':ti,ab OR 'mycobiome*':ti,ab OR 'mycobiota*':ti,ab OR 'virome*':ti,ab OR 'phylotype*':ti,ab)) OR 'enterotype*':ti,ab	126127
2	'diversit*':ti,ab OR 'abundan*':ti,ab OR 'richness*':ti,ab OR 'evenness*':ti,ab OR 'dissimilarit*':ti,ab	549572
3	#1 AND #2	21632
4	[animals]/lim NOT [humans]/lim	5765837
5	#3 NOT #4	13783
6	english:la	31021053
7	#5 AND #6	13542
8	[2019]/py	1585050
9	#7 AND #8	2536

Set #	CENTRAL	Results
1 Gut Microbiome	[mh "gastrointestinal microbiome"] OR ((gut:ti,ab,kw OR intestin*:ti,ab,kw OR gastrointestinal*:ti,ab,kw OR colon*:ti,ab,kw OR rectal:ti,ab,kw OR rectum:ti,ab,kw OR stool:ti,ab,kw OR feces:ti,ab,kw OR faeces:ti,ab,kw OR fecal:ti,ab,kw OR faecal:ti,ab,kw) AND ([mh microbiota] OR [mh mycobiome] OR microbiome*:ti,ab,kw OR microbiota*:ti,ab,kw OR microbial:ti,ab,kw OR microbe*:ti,ab,kw OR microflora*:ti,ab,kw OR flora*:ti,ab,kw OR microorganism*:ti,ab,kw OR pathobiont*:ti,ab,kw OR mycobiome*:ti,ab,kw OR mycobiota*:ti,ab,kw OR virome*:ti,ab,kw OR phylotype*:ti,ab,kw)) OR enterotype*:ti,ab,kw	8915
2 Diversity	diversit*:ti,ab,kw OR abundan*:ti,ab,kw OR richness*:ti,ab,kw OR evenness*:ti,ab,kw OR dissimilarit*:ti,ab,kw	4452
3	#1 AND #2	1067
4	[mh animals] NOT [mh humans]	6950
5	#3 NOT #4	1051
6	#5 restrict publication year to 2019	282

Supplementary table 1. Characteristics of Included Studies

Author	Country	Income Level	WHO Regions	Study design	n	Population	Mean age (SD)
Robinson[1]	Australia	HICs	Western Pacific	Observational	22	22 Obesity/Overweight	with Ketonuria Med 33 (IQR 29-38), without Med 32 (IQR 29-33)
Harbison[2]	Australia	HICs	Western Pacific	Observational	88	47 T2DM, 41 Control	Med 10.9 (Range 4.8-26.7)
Horvath[3]	Austria	HICs	European	RCT	26	26 T2DM with Obesity	60 (NR)
Liu T[4]	China	UMICs	Western Pacific	Observational	6627	495 IFG, 304 T2DM, 5828 Control	51.8 (NR)
Lv[5]	China	UMICs	Western Pacific	Observational	28	9 Overweight/Obesity, 10 Normal, 9 Lean	22.03 (1.32)
Mushtaq[6]	China	UMICs	Western Pacific	Observational	80	50 HT, 30 Control	61.8 (10.6)
Nuli a[7]	China	UMICs	Western Pacific	Observational	60	20 T2DM, 20 Impaired glucose regulation, 20 Control	NR
Nuli b[8]	China	UMICs	Western Pacific	Observational	561	145 T2DM, 138 IFG, 278 Control	47.08 (10.43)
Qiu[9]	China	UMICs	Western Pacific	Observational	55	28 Obesity, 27 Control	37.75 (12.54)
Tao[10]	China	UMICs	Western Pacific	Observational	56	14 DN, 14 T2DM without DN, 28 Control	50.84 (12.30)
Wang[11]	China	UMICs	Western Pacific	Observational	46	26 Obesity 20 Healthy	34.2 (10.1)
Wu[12]	China	UMICs	Western Pacific	Observational	49	23 GDM, 26 Non-GDM	GDM Mean 36 (IQR 32-38.5), Non-GDM Mean 32.5 (IQR 30-35)
Xu[13]	China	UMICs	Western Pacific	Observational	61	30 GDM, 31 Control	33.0 (4.5)
Zhang[14]	China	UMICs	Western Pacific	Observational	180	130 T2DM, 50 Healthy	58.7 (9.4)
Zhao[15]	China	UMICs	Western Pacific	Observational	58	25 Obesity with NAFLD, 18 Obesity only, 15 Healthy	13.7 (1.8)
Zheng[16]	China	UMICs	Western Pacific	Observational	99	36 DLP, 63 Healthy	36 (NR)
Zhou[17]	China	UMICs	Western Pacific	Quasi-exp	30	30 Obesity or Overweight	47.9 (5.4)
Zuo b[18]	China	UMICs	Western Pacific	Observational	49	34 HT, 15 Healthy	HT Med 54.5 (IQR 49.25, 57.75), Healthy Med 58 (IQR 52, 60)
Cuesta-Zuluaga[19]	Colombia	UMICs	Americas	Observational	441	30% Obesity (132), 41% Central Obesity (180), 59% HT (259)	41 (1)
Kern[20]	Denmark	HICs	European	RCT	88	88 Overweight or Obesity	Med 36 (IQR 30, 41)
Salah[21]	Egypt	LMICs	Eastern Mediterranean	Observational	60	25 Obesity with T2DM, 25 Obesity only, 5 T2DM only, 5 Control	43.9 (13.3)
Olsson[22]	France	HICs	European	Observational	58	34 Obesity, 24 non-Obesity	42.3 (16.3)
Seck[23]	France	HICs	European	Observational	56	17 Obesity, 39 Lean	NR
Frost[24]	Germany	HICs	European	Quasi-exp	12	12 T2DM with Obesity	57.2 (NR)
Kellerer[25]	Germany	HICs	European	Quasi-exp	17	17 Morbid Obesity	41.8 (9.1)
Ejtahed[26]	Iran	UMICs	Eastern Mediterranean	RCT	36	36 Non-Diabetic Obese Woman	36 (7)
Conterno[27]	Italy	HICs	European	RCT	62	62 Hypercholesterolemia	48.5 (9.0)
Ponzo[28]	Italy	HICs	European	Observational	29	29 GDM Offspring	3-5 Day of Life
Uemura[29]	Japan	HICs	Western Pacific	RCT	44	44 Obesity	62.7 (8.8)
Vera[30]	Mexico	UMICs	Americas	RCT	81	81 T2DM	Range 30-60
Bakker[31]	Netherlands	HICs	European	Quasi-exp	20	10 Obesity with Metabolic Syndrome, 10 Lean	43.6 (17.2)
vanBommel[32]	Netherlands	HICs	European	RCT	44	44 T2DM	NR
Belkova[33]	Russia	UMICs	European	Observational	30	12 Obesity, 18 Normal weight	14.6 (1.6)
Koo[34]	Singapore	HICs	Western Pacific	Observational	35	5 Metabolic Syndrome, 9 Obesity, 2 T2DM, 22 Central Obesity	Med 39 (Range 22-70)
Velikonja[35]	Slovenia	HICs	European	RCT	43	43 Metabolic syndrome/High risk for Metabolic Syndrome	52.07 (7.71)
Kim[36]	South Korea	HICs	Western Pacific	Observational	766	313 NAFLD, 453 Control	44.9 (8.1)
Romo-Vaquero[37]	Spain	HICs	European	Quasi-exp	249	49 Obesity/Overweight, 200 Healthy	42.6 (11.5)
Lin[38]	Taiwan	HICs	Western Pacific	Observational	20	20 Obesity	37.11 (10.0)
Astbury[39]	UK	HICs	European	Observational	141	65 Biopsy-proven NASH, 76 Healthy	64.19 (2.96)
Faucher[40]	USA	HICs	Americas	Observational	21	15 Obesity, 6 Normal	29.7 (5.1)
Fei[41]	USA	HICs	Americas	Observational	655	229 Obesity, 149 Overweight, 277 Normal	34.9 (6.4)
Karvonen[42]	USA	HICs	Americas	RCT	502	149 Obesity, 353 Normal	3 (NR)

Maskarinec[43]	USA	HICs	Americas	Observational	1735	30.3% Obesity, 40.4% Overweight, 29.3% Normal	69 (NR)
Schwimmer[44]	USA	HICs	Americas	Observational	124	87 Biopsy-proven NAFLD, 37 Obesity without NAFLD	NAFLD Med 12 (IQR 10,14), Obesity without NAFLD Med 13 (IQR 11,14)
Shen[45]	USA	HICs	Americas	Observational	26	26 Severe Obesity	NR
Stanislawski[46]	USA	HICs	Americas	Observational	152	103 Obesity, 49 Control	34.0 (12.2)
Sun[47]	USA	HICs	Americas	Observational	529	35.1% HT	55.3 (3.4)

DLP, dyslipidemia; DN, diabetes nephropathy; GDM, gestational diabetes mellitus; HICs, high-income countries; HT, hypertension; IFG, impaired fasting glucose; IQR, interquartile range; LMICs, lower-middle-income countries; Med, median; NAFLD, nonalcoholic fatty liver disease; NR, not reported; Quasi-exp, quasi-experimental; RCT, randomized controlled trial; SD, standard deviation; T1DM, type I diabetes mellitus; T2DM, type II diabetes mellitus; UMICs, upper-middle-income countries; WHO, World Health Organization.

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Supplementary Table 2 The difference between each measure for alpha-diversity estimators and beta-diversity estimators

	Diversity index	Inventor	Established year
Alpha-diversity	ACE index	Chao, A. and Lee, S.M.	1992
	ASV richness	N/A	N/A
	Chao index	Anne Chao	1984
	Faith's Phylogenetic diversity	Faith, D.P.	1992
	Fisher's index	Fisher, R.A	1943
	Good's coverage	Good, I.J	1953
	Inverse simpson	Edward Simpson	1949
	OTU richness	Robert R. Sokal and Peter H. A. Sneath	1963
	Pielou's evenness	Pielou, E.	1966
	Shannon	Claude Shannon	1948
	Simpson	Edward H. Simpson	1949
Beta-diversity	UniFrac distances	Catherine Lozupone	2005
	unweighted UniFrac distances	Catherine Lozupone	2005
	weighted UniFrac Distance	Catherine Lozupone	2005
	Bray-Curtis dissimilarity	J. Roger Bray and John T. Curtis	1957

	Diversity index	Description
Alpha-diversity	ACE index	Estimate diversity from abundance data
	ASV richness	The number of unique bacterial taxa measured by ASV methods
	Chao index	Estimate diversity from abundance data
	Faith's Phylogenetic diversity	Measures of biodiversity that incorporates phylogenetic difference between species
	Fisher's index	Relationship between the number of species and the abundance of each species
	Good's coverage	Estimates the percent of an entire species that is represented in a sample
	Inverse simpson	The metric tells us the sum of squares of all abundance ratios
	OTU richness	The count of different species for the sample
	Pielou's evenness	Measure of relative evenness of species richness
	Shannon	Summarizes the diversity in the population while assuming all species are represented in a sample and they are randomly sampled. The Shannon index increases as both the richness and evenness of the community increase. Influenced by rare OTUs
	Simpson	Calculate a measure of diversity taking into account the number of taxa as well as the abundance. The simpson index gives more weight to common or dominant species which means a few rare species with only a few representatives will not affect the diversity of the sample
Beta-diversity	UniFrac distances	Estimates differences between samples or groups based on phylogenetic distance.
	unweighted UniFrac distances	Purely based on sequence distances (does not include abundance information)
	weighted UniFrac Distance	Branch lengths are weighted by relative abundances (includes both sequence and abundance information)
	Bray-Curtis dissimilarity	Measure used to quantify the compositional dissimilarity between two samples or groups. Its value ranges from 0 to 1, where 0 means that the two samples or groups share all species, and 1 means that they do not share any.

	Diversity index	Pros	Cons
Alpha-diversity	ACE index	Useful for rare OTUs/ very accurately and efficiently estimated as long as the sample size is reasonably large	Underestimate if species rich and highly heterogenous assemblages
	ASV richness	Outperformed the adopted OTU method concerning community diversity, especially for fungus-related sequences	Need high sequencing depth to capture the community complexity
	Chao index	Useful for rare OTUs/ useful when there are many undetectable species in a highly diverse assemblage	Underestimate if species rich and highly heterogenous assemblages
	Faith's Phylogenetic diversity	Provide a comparable, evolutionary measure of biodiversity not possible with species count and supported functional diversity in explaining species richness ecosystem function connections.	Lack of sequence data for vast majority of species, require a molecular lab with basic facilities and sequencing cost can be high
	Fisher's index	Useful for examining the diversity within a group of individuals, rather than pooling together all samples in the group	Not particularly useful for evaluating the dominance or evenness of species
	Good's coverage	Unbiased algorithmic technique for rarefying samples to a fixed sample coverage instead of a fixed size	Does not tell you whether there is evenness in the species abundance
	Inverse simpson	Do not tend to be as affected by sampling effort as the shannon index	Heavily weighted toward the most abundant species in the sample.less sensitive to species richness
	OTU richness	Simple, Easy to calculate	Sensitive to sample size
	Pielou's evenness	More thorough description of a community's structure can be interpreted	Evenness index that generally varied less in amplitude
	Shannon	Combines richness and evenness, widely used, easy to calculate, useful index for the community which two or three species were dominant	Insensitive measure of the character of the species abundance distribution (evenness)
	Simpson	Combines richness and evenness, widely used, easy to calculate	Heavily weighted toward the most abundant species in the sample.less sensitive to species richness
Beta-diversity	UniFrac distances	Could be used to determine whether communities were significantly different	Should not be used as a distance metric for multivariate statistical analyses.
	unweighted UniFrac distances	Sensitive for detecting richness changes in rare species	Ignores the abundance information in the computation.
	weighted UniFrac Distance	Incorporates the abundance information and reduces the contribution of rare species	N/A
	Bray-Curtis dissimilarity	Gives more weight to common species	not a real distance