

Breast milk cytokines and early growth in Gambian infants

Supplementary Material

Table 1: Lowest Limit of Detection (LLOD) for cytokines measured using Meso Scale methods outlined in this study (Data from Meso Scale Discovery, 2014)

Cytokine	Lowest Limit of Detection
IL-1β	0.04
IL-2	0.09
IL-4	0.02
IL-6	0.06
IL-10	0.03
IL-12p70	0.11
IL-13	0.24
TNFα	0.04
IFNγ	0.2
IGF-1	26.8
TGFβ2	265.0

All cytokine measurements are in pg/ml

Table 2: Median of cytokine concentrations in colostrum and mature breast milk

Cytokine	Colostrum median (IQR)	Total samples	Number of samples detectable (%)*	BM median (IQR)	Total samples	Number of samples detectable (%)	p- value**	p-value for proportion of detectable samples***
TNFα	21.32 (7.27-104.59)	83	83 (100)	1.04 (0.55-2.49)	90	86 (96)	p<0.001	0.12
IFNγ	13.40 (5.87-29.93)	83	82 (99)	2.50 (0.83-5.96)	90	82 (91)	p<0.001	0.04
IL10	3.46 (1.11-12.47)	83	82 (99)	0.05 (0.02-0.23)	90	57 (63)	p<0.001	p<0.001
IL12	0.85 (0.21-3.45)	83	68 (82)	0.12 (0.06-0.28)	90	48 (53)	p<0.001	p<0.001
IL13	7.77 (1.82-24.67)	83	78 (94)	1.53 (0.59-2.94)	90	74 (82)	p<0.001	0.02
IL1β	6.00 (2.20-19.92)	83	83 (100)	0.40 (0.06-1.89)	90	71 (79)	p<0.001	p<0.001
IL2	0.84 (0.35-2.70)	83	78 (94)	0.11 (0.04-0.48)	90	52 (58)	p<0.001	p<0.001
IL4	0.16 (0.05-0.57)	83	72 (87)	0.01 (0.01-0.06)	90	40 (44)	p<0.001	p<0.001
IL6	25.91 (8.43-73.08)	83	83 (100)	0.91 (0.37-3.73)	90	85 (94)	p<0.001	0.06
TGFβ2	5116.49 (3464.15- 8156.59)	42	42 (100)	7983.20 (3842.67- 19569.42)	46	46 (100)	0.11	1
IGF-1****	13.40 (13.40-13.40)	70	15 (21)	13.40 (13.4-13.4)	90	10 (11)	0.06	0.08

All cytokine measurements are in pg/ml (rounded to 2 decimal places)

*Samples were termed as detectable if concentrations measured were above the lower limits of detection shown in Table 1.

**Wilcoxon rank sum test comparing the mean concentration of each cytokine in colostrum and breast milk

*** Fisher's exact two-sided test comparing proportions of detectable samples in colostrum and breast milk

****Median and IQR of IGF-1 concentrations in colostrum and BM samples coincide because the levels were below the assay limit of detection in >75% of subjects. Given this low detection rate, data on IGF-1 has been removed from further statistical analysis.

Abbreviations: BM, breast milk; SD, standard deviation; IQR, interquartile range

Table 3: Association between cytokine concentrations in colostrum and breast milk

Cytokine	<i>r</i> *	<i>p</i> -value
TNFα	0.07	0.57
IFNγ	0.06	0.61
IL10	0.14	0.21
IL12	0.26	0.02
IL13	0.04	0.73
IL1β	0.10	0.39
IL2	-0.08	0.46
IL4	0.14	0.21
IL6	0.14	0.22
IGF-1	0.21	0.08
TGFβ2	0.11	0.50

All values are rounded to 2 decimal places

* *r* = Spearman rank correlation co-efficient