



Assessment of Human Milk Intake by Breastfed Infants Using Deuterium Oxide Dose-to-Mother Technique in “*Tumbuh Kembang Anak*” Cohort, Indonesia

*Webinar serial 16 Application of nuclear
techniques for the assessment of nutritional
status and breastfeeding practices*

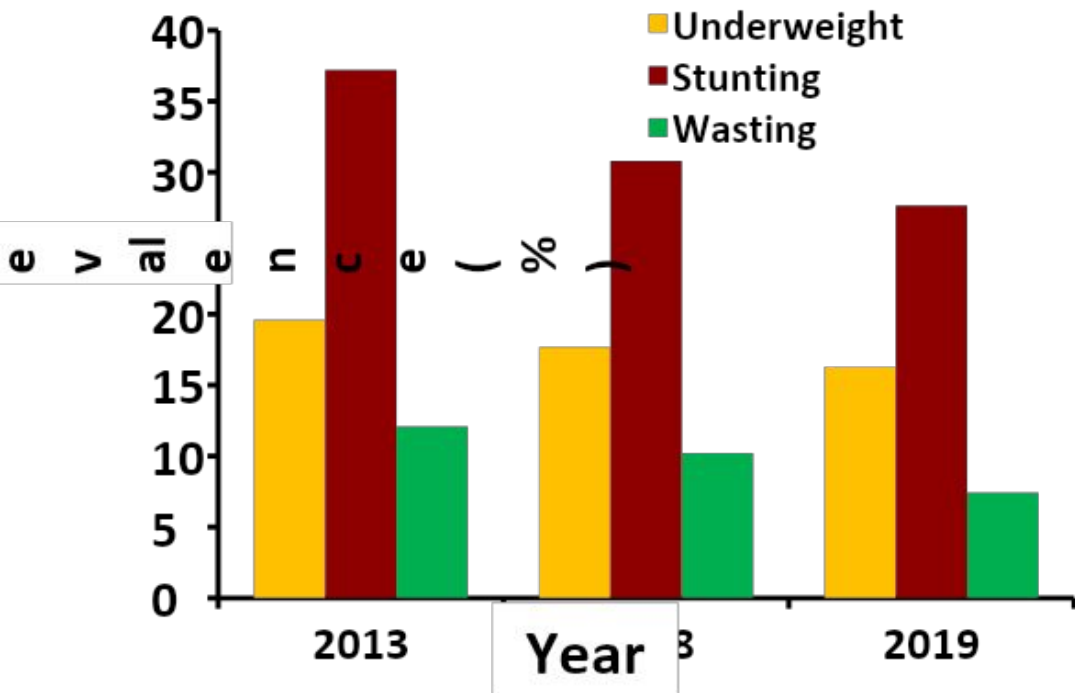
Friday, 20 November 2020

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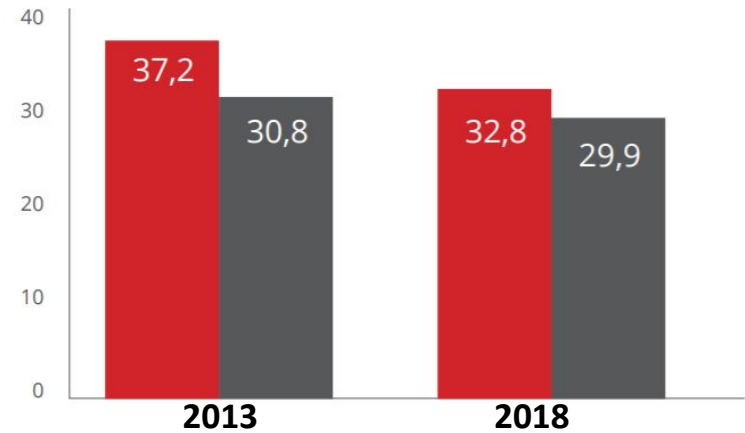
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Centre for Research and Development
of Health Resources and Services



Prevalence of children nutrition status in Indonesia (< 5 years age)



Stunting prevalence of children < 5 years and < 2 years



Sumber: Riskesdas 2013 dan 2018, diolah Balitbangkes

Exclusive Breastfed rates by Provinces in 2017

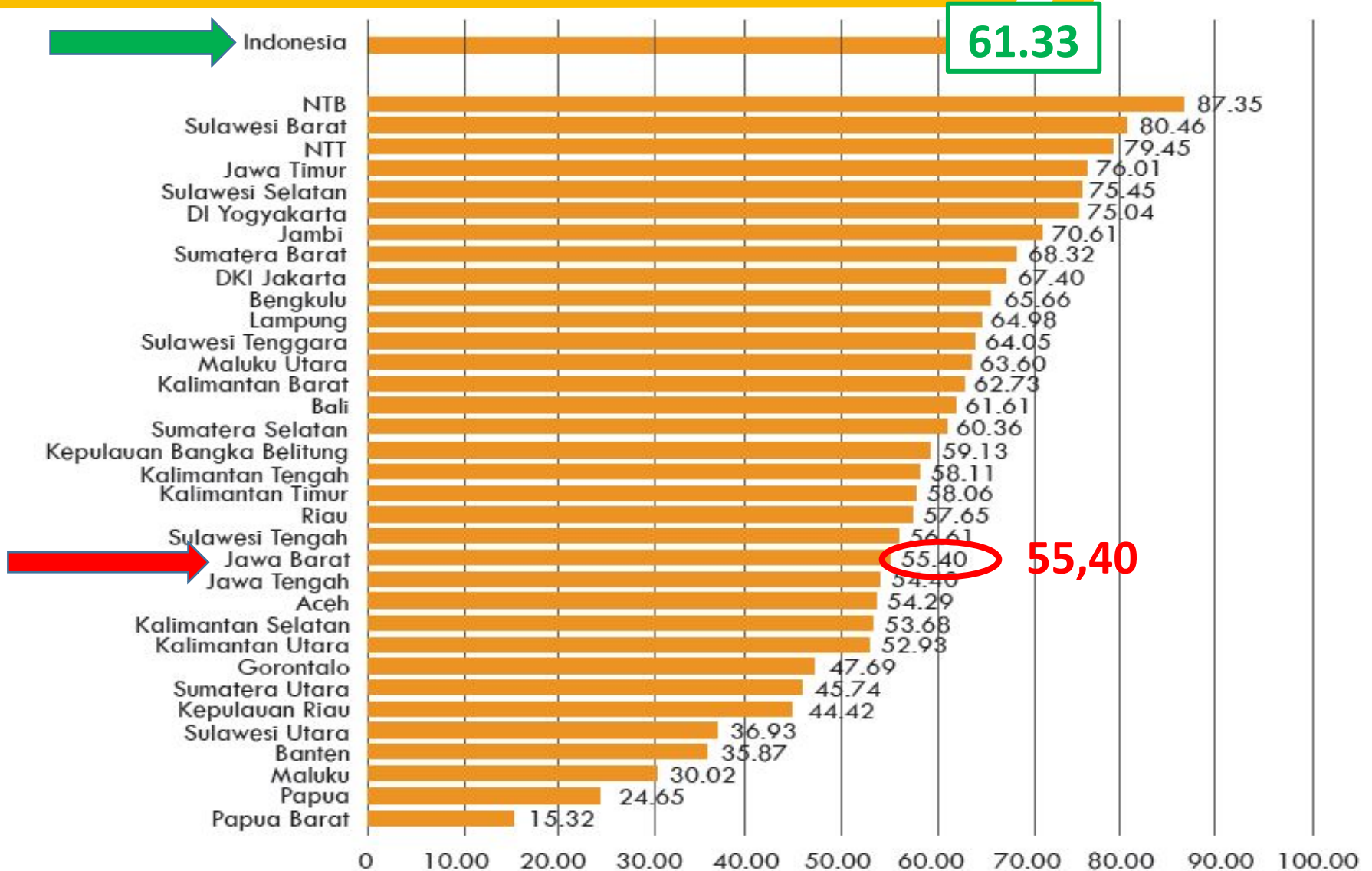


TABLE 1 Countries and characteristics of study included in the Human Milk Intake Analysis

Country	Reference	Type of study	Infants, <i>n</i>	Age range or age at follow-up, <i>mo</i>	Measurements, <i>n</i>
UK	Laskey et al., 1991 (22)	Cross-sectional	30	1–2	30
UK	Jones, 2003 (23)	Cross-sectional	50	1–2	50
UK	YWP study (M. A. Laskey, unpublished)	Cross-sectional	33	1–2	33
USA	Butte et al., 1988 (20)	Cross-sectional	9	1–6	9
Senegal	Cissé et al., 2002 (18)	Cross-sectional	129	2–3	129
Bangladesh	Moore et al., 2007 (10)	Cross-sectional	94	2–3	94
Kenya	Ettyang et al., 2005 (14)	Cross-sectional	10	2–5	10
Brazil	Haisma et al., 2003 (11)	Cross-sectional	70	3–4	70
Brazil	Haisma et al., 2006 (12)	Cross-sectional	74	7–8	74
Zambia	Owino et al., 2007 (24)	Cross-sectional	52	9	52
Mexico	Balaños et al., 2000 (16)	Longitudinal	52	0.5, 3	104
UK	Goldberg et al., 1991 (21)	Longitudinal	10	1,2,3	29
Chile	Alvear et al., 2004 (13)	Longitudinal	29	1,3,6	78
Gambia	Jarjou et al., 2006 (19)	Longitudinal	30	4, 12	60
Malawi	Golpin et al., 2007 (15)	Longitudinal	42	6–9	79
PNG	Orr-Ewing et al., 1986 (17)	Longitudinal	23	0.7, 2, 3, 5, 7, 9, 12, 14, 17, 20, 24	214
Total			737		1115

Country	Maternal BMI (kg·m ⁻²)	Maternal % body fat	Infant Age (months)	Feeding Pattern	Breast Milk Intake* (g·day ⁻¹)
Bangladesh [22]	20.7 ± 1.7		3.5	EBF	911 ± 168
Brazil [13]	24.8 ± 3.5	34 ± 6	4	EBF	881
Ethiopia [20]	21.2 ± 2.1	27 ± 6	1.5	EBF	880 ± 120
Kenya [8]	18.5 ± 1.0	17 ± 6	2 - 4	EBF	569 ± 23
		22 ± 6	5	EBF	955 ± 189
Malawi [16]		21 ± 9	5.5	EBF	921 ± 185
		25 ± 7	5.5	EBF	839 ± 109
Mexico [15]	23.8 ± 3.2	24 ± 6	4	EBF	885 ± 146
		23.3 ± 3.1	6	EBF	869 ± 150
Papua New Guinea [19]			4 - 6	PBF	863 ± 157
The Gambia [10]			4	EBF	820 ± 38
Pakistan (this work-subset defined by Figure 4)	22.9 ± 6.1	31 ± 13	5 - 7	EBF	757 ± 249

Breast milk consumed at 3 mo : 700 g/day and at 6 mo : >750 g/day
Christine Slater *et al.* (2019)

- | | |
|---------------|-------------|
| 1. Bangladesh | 5. Pakistan |
| 2. Indonesia | 6. Srilanka |
| 3. Malaysia | 7. Vietnam |
| 4. Mongolia | 8. Thailand |

1. Center for Isotopes and Radiation Application-BATAN, Jakarta,
2. Center for Applied Health Technology & Clinical Epidemiology (PPTK&EK) - National Institute of Health Research and Development (NIHRD), Bogor

supported by International Atomic Energy Agency (IAEA), Vienna



Objectives

- to measure human milk intake of Indonesian infants during first 6 months
- to assess the breastfeeding practices of mothers

Study Design

- human milk intake of infants at 3 and 6 months according to mother's report of infant feeding
- Exclusive breastfeeding (EBF), Predominant breast feeding (PBF), Partial breastfeeding (ParBF)
- The study was approved by the Ethical Review Committee of the Ministry of Health, Jakarta, Indonesia

Inclusion criteria

- mother who exclusive breastfeeding
- parity no more than 3
- infant aged 3 months \pm 7 days
- nutrition status of infants WHZ more than -2

Exclusion criteria

- twin baby or more,
- low birth weight (<2.5 kg)
- baby with oedema,
- mother with fluid retention disease
- mother with smoking and drinking alcohol

- Respondents were selected from the maternal and child “*Tumbuh Kembang Anak*” cohort of National Institute of Health Research and Development, Bogor, West Java, Indonesia. This Cohort was established in 2011.
- Socialization to respondents: The objective of the research, benefit, and procedures.
- Anthropometry: mother and infant weight and height/length
- Selection and the respondents had been given the informed consent the mothers after explaining about the research.
- There were 30 mother and infant pairs aged 3-6 months who agreed to participate in this research (from 50 respondents).
- Interview and mothers answer had been recorded in the questionnaire.



1. Mother drinks $^2\text{H}_2\text{O}$
2. Baby consumes $^2\text{H}_2\text{O}$ in human milk
3. Saliva in both mother and baby is enriched with $^2\text{H}_2\text{O}$

Fig. 1. Dose-to-mother technique for assessing human milk intake

MATERIAL & EQUIPMENT *for saliva samplina*



Schedule:

- Baseline
- 1st day
- 2nd day
- 3rd day
- 4th day
- 13th day
- 14th day

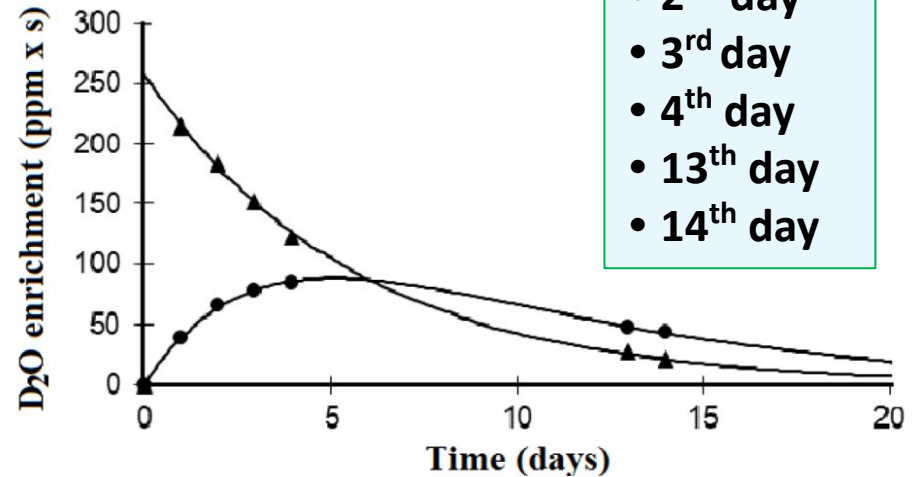
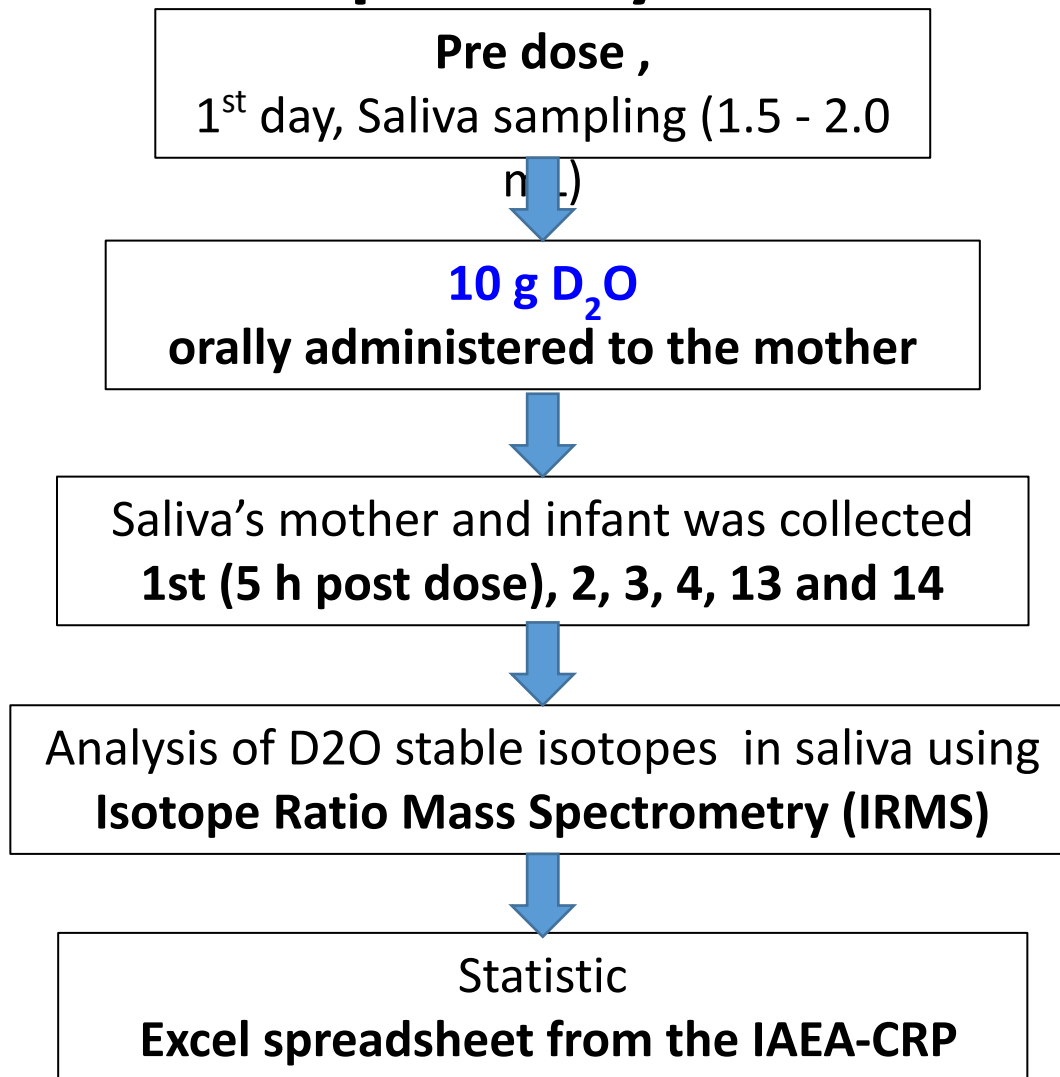


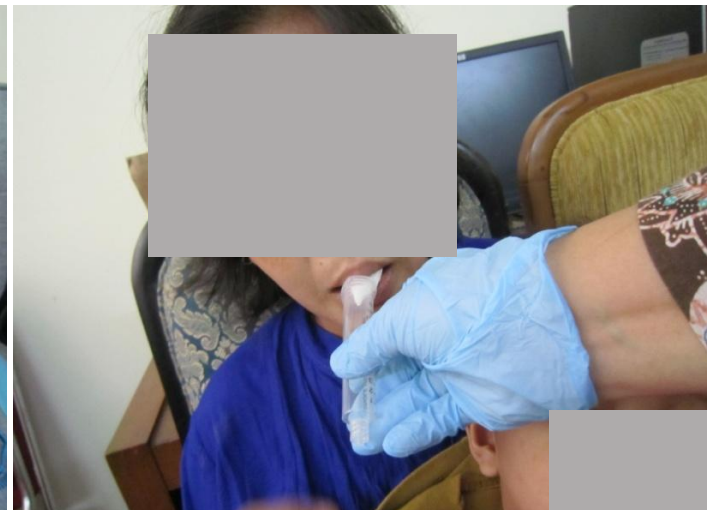
Fig. 2. Kinetic of D₂O concentration in mother's (-▲-) and baby's saliva (-●-) at days of observation.

Disappearance of deuterium from the body water of the mother and appearance in her baby

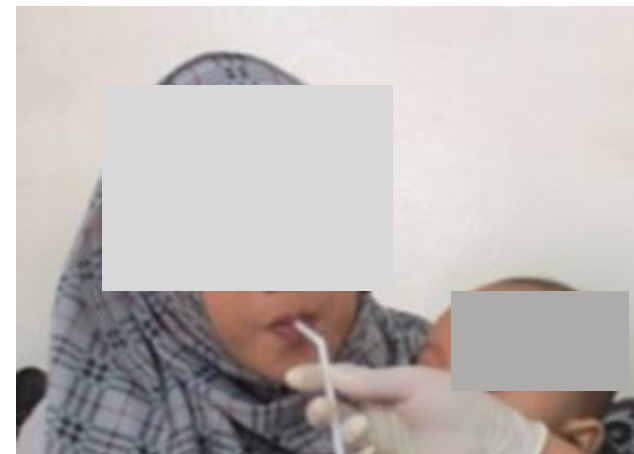
Saliva collecting and isotopic analysis



Saliva collection and D₂O administration



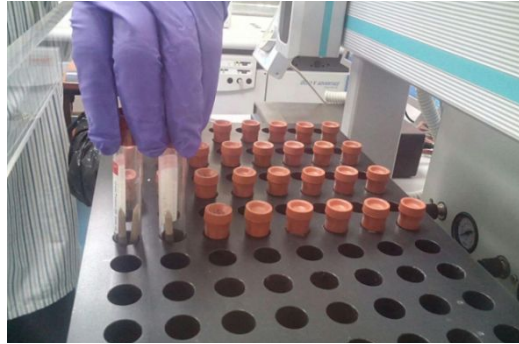
Wet ball cotton was removed into syringe



Saliva sampling from the infant using the cotton stick

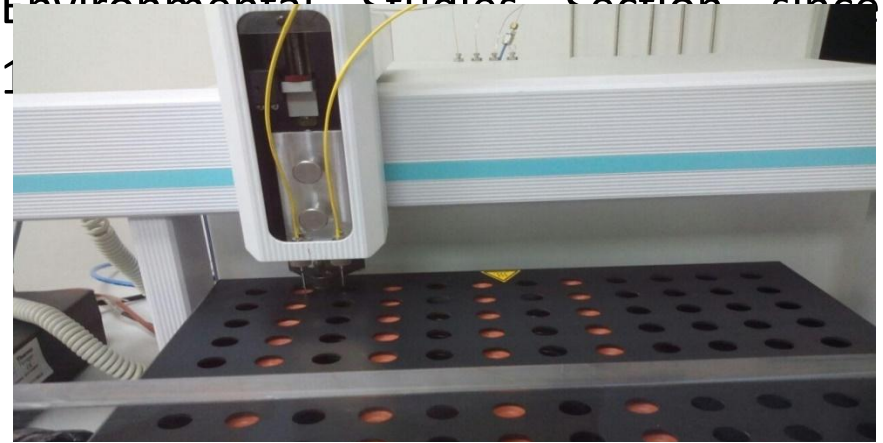
10 g D₂O, rinsed with 20 ml of water 2x

D₂O analysis using IRMS



At St. John's Research Institute (SJRI), Bangalore, India;

The SJRI has been working with the IAEA's Nutrition and Health-Related Environmental Studies Section since



Analysis of deuterium enrichment

The enrichment of deuterium oxide in saliva samples were reduced to hydrogen gas using Pt and the hydrogen produced was transferred into Thermo Delta Advantage continuous-flow IRMS system, SJRI, Bangalore, India.

Table 1. Mother characteristics.

Characteristic	Value
<i>Weight, kg</i>	
3 months	59.2±10.9
6 months	59.1±11.4
<i>Height, cm</i>	
3 months	151.9±5.3
6 months	152.3±5.2
<i>BMI, kg.m²</i>	
3 months	25.6±4.1
6 months	25.4±4.2
<i>Age, y, %</i>	
20 - 25 y	5 (16.7)
25.5 - 30 y	10 (33.3)
30.5 - 40 y	13 (43.3)
> 40 y	2 (6.7)
<i>Parity, n,%</i>	
1	4 (13.3)
2	12 (40.0)
≥3	14 (46.7)
<i>Education, n,%</i>	
Primary School	4 (13.3)
Junior High School	8 (26.7)
High School	15 (50.0)
University	3 (10.0)
<i>Employment, n,%</i>	
Unemployed	29 (96.7)
Employed	1 (3.3)

RESULT

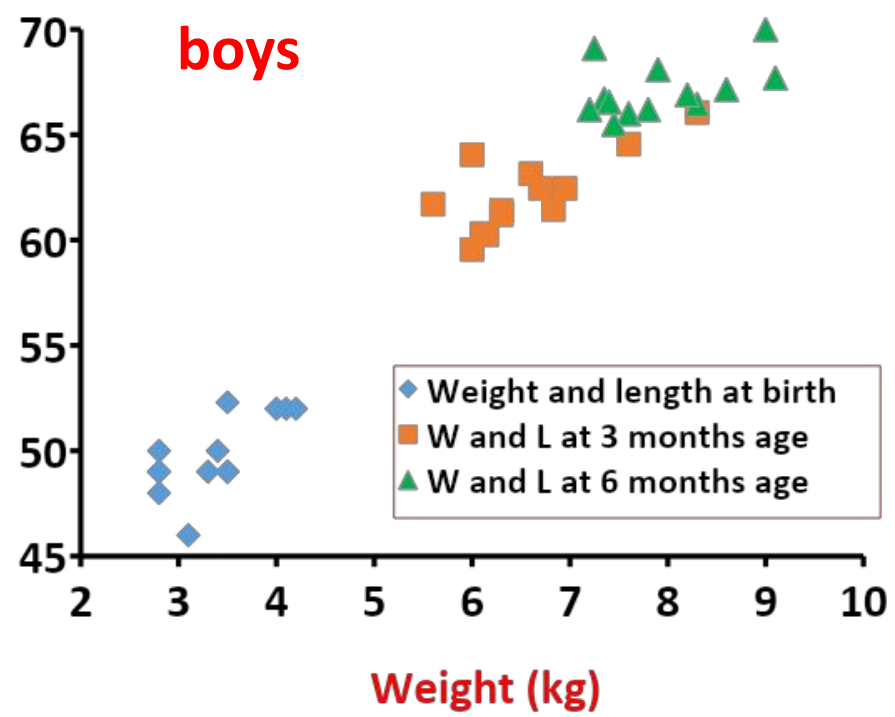
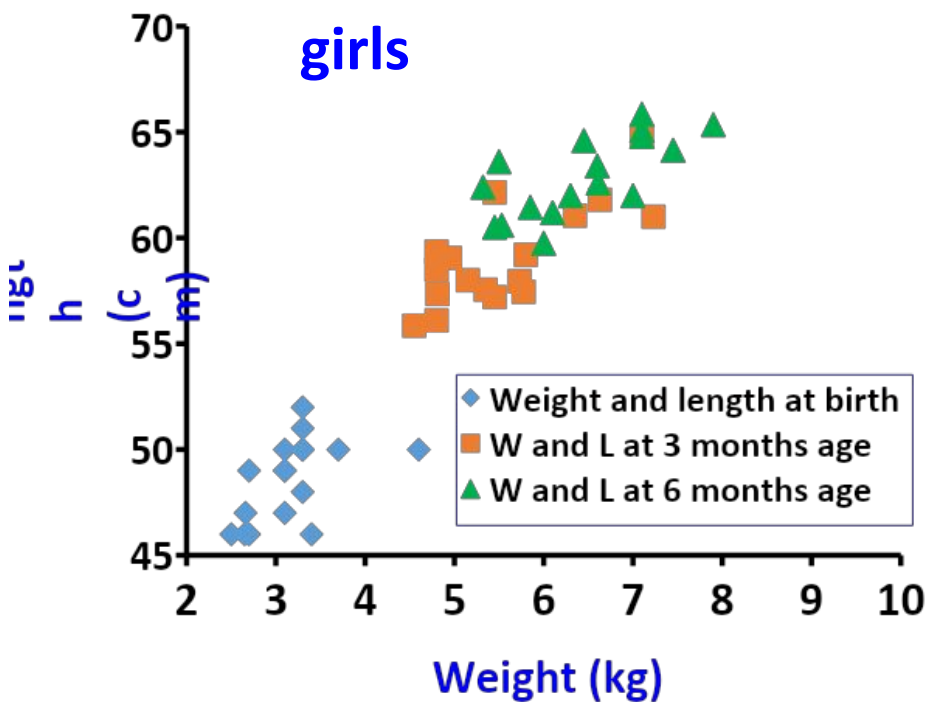


Table 2. Infant Characteristics.

Characteristic	Value	
	Boys	Girls
<i>Weight, kg</i>		
3 months	6.57 ± 0.7	5.50 ± 0.7
6 months	7.93 ± 0.7	6.48 ± 0.8
<i>Length, cm</i>		
3 months	62.20 ± 1.9	59.08 ± 2.4
6 months	66.08 ± 2.6	63.89 ± 2.6
<i>BMI, kg.m²</i>		
3 months	16.6 ± 1.2	15.8 ± 1.6
6 months	17.9 ± 2.1	15.6 ± 2.0
<i>Birth weight, kg</i>	3.4 ± 0.5	3.2 ± 0.5
<i>Birth length, cm</i>	49.8 ± 1.9	48.6 ± 1.69

***E.K. Winarno, H. Winarno, Susanto, T. Fajarwati, and T. Thomas
Atom Indonesia Vol. 45 No. 1 (2019) 51 - 58 .***

Weight and length of infants at birth, 3 months, and 6 months age (girls and boys)





Participant Study ID	0401R1			
MOTHER'S DATA			BABY'S DATA	
Date of Birth	6-Apr-81		Date of Birth	25-Dec-14
age	33 years		sex	Female
weight	52.80 kg		age	3.2 months
			start weight	6.15 kg
			final weight	6.35 kg
DOSE DATA				
Bottle number	1			
Weight D ₂ O (g) in bottle	10.0681			
Date dose taken by mother	31-Mar-15			
Time dose taken by mother	9:38			

Human milk intake (M)	0.820	kg.day ⁻¹	820	g.day ⁻¹
Water input from milk (F _m)	0.788	kg.day ⁻¹		
Water used in growth (F _g)	0.007	kg.day ⁻¹		
Total water output (F _{ob})	0.946	kg.day ⁻¹		
Non-oral water intake (F _a)	0.060	kg.day ⁻¹		
Non-milk oral intake (F _s)	0.104	kg.day ⁻¹	104	g.day ⁻¹
Square root MSE (mg/kg)	6.98	Total error		

(1) Data for mothers saliva

Date	time	time since dose (days)	Deuterium abundance	
			ppm	ppm
31-Mar-15	Baseline	0	142.08	142.88
1-Apr-15	9:31	1.00	370.90	370.32
2-Apr-15	9:20	1.99	342.91	338.61
3-Apr-15	9:21	2.99	313.23	311.87
4-Apr-15	9:24	3.99	285.39	284.92
13-Apr-15	0:55	12.64	181.84	180.30
14-Apr-15	9:40	14.00	177.73	180.09

(2) Data for baby's saliva

Date	time	time since dose (days)	Deuterium abundance	
			ppm	ppm
31-Mar-15	Baseline	0	146.3	145.6
1-Apr-15	9:28	0.99	189.2	192.0
2-Apr-15	9:25	1.99	213.2	211.7
3-Apr-15	9:25	2.99	229.9	228.7
4-Apr-15	9:24	3.99	235.7	237.0
13-Apr-15	0:50	12.63	198.4	200.7
14-Apr-15	9:32	14.00	192.4	197.9

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²H Kinetics

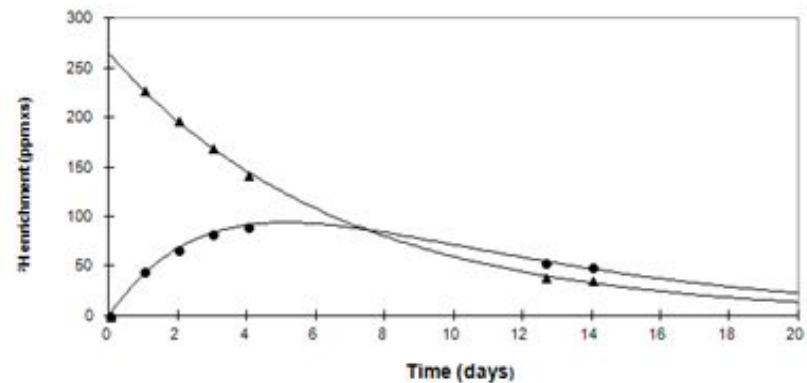


Table 3. Kinetic data of the mothers and the infants based on the deuterium oxide-to mother model.*

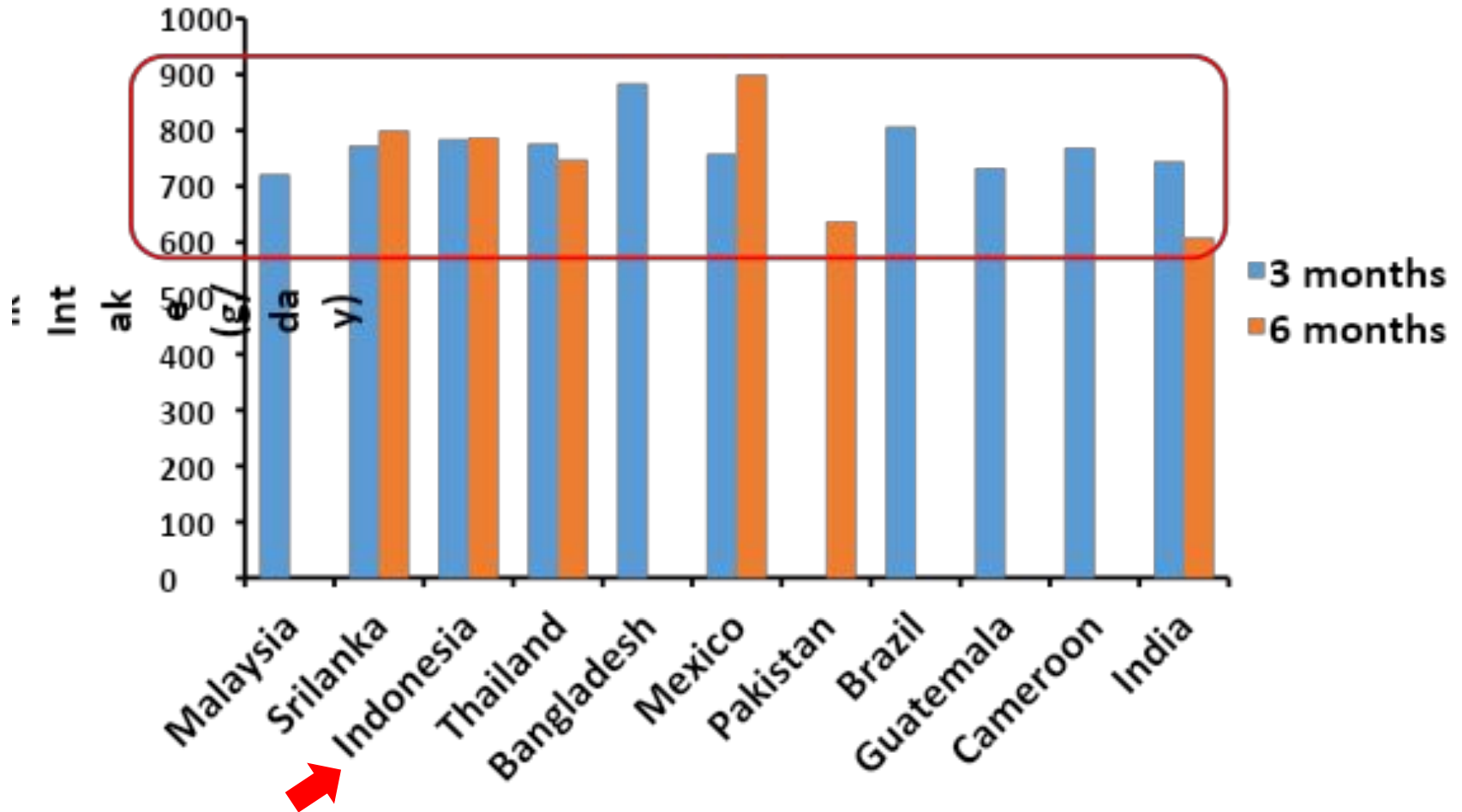
Index	3 months	6 months
<i>Kinetic data of the mother</i>		
D space (V_m), kg	36.0 ± 5.2 (26.2 - 46.2)	37.4 ± 5.0 (27.8 - 49.0)
TBW, kg	34.6 ± 5.0 (25.1 - 44.4)	35.9 ± 4.8 (26.7 - 47.0)
Fat-Free Mass, kg	47.3 ± 6.8 (34.4 - 60.6)	49.1 ± 6.6 (36.4 - 64.2)
Fat Mass, kg	11.3 ± 6.2 (2.4 - 25.8)	10.0 ± 7.9 (-7.6 - 26.6)
Fat Mass, %	18.4 ± 8.1 (4.0 - 32.6)	15.5 ± 11.2 (-15.4 - 32.7)
Water intake, kg.d ⁻¹	5.0 ± 0.9 (3.0 - 7.0)	5.1 ± 1.0 (3.3 - 8.5)
D ₂ O concentration in mother's body water at time 0, mg.kg ⁻¹	284 ± 42 (217 - 382)	270 ± 35 (205 - 362)
<i>Kinetic data of the infants</i>		
Human milk intake, g.d ⁻¹	784 ± 117 (525 - 1105)	787 ± 180 (320 - 1246)
Water input from milk, g.d ⁻¹	753 ± 112 (505 - 1062)	756 ± 173 (308 - 1198)
Water used in growth, g.d ⁻¹	12 ± 18 (-6 - 101)	1 ± 10 (-47 - 10)
Total water output, g.d ⁻¹	841 ± 128 (606 - 1193)	938 ± 168 (645 - 1386)
Non-oral water intake, g.d ⁻¹	54 ± 8 (39 - 79)	65 ± 29 (41 - 210)
Non-milk oral intake, g.d ⁻¹	43 ± 48 (-63 - 142)	140 ± 144 (-93 - 648)
Square root MSE, mg.kg ⁻¹	11 ± 8 (4 - 49)	20 ± 26 (4 - 115)

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Table 4. Human milk intake and growth of the boy and girl infants.

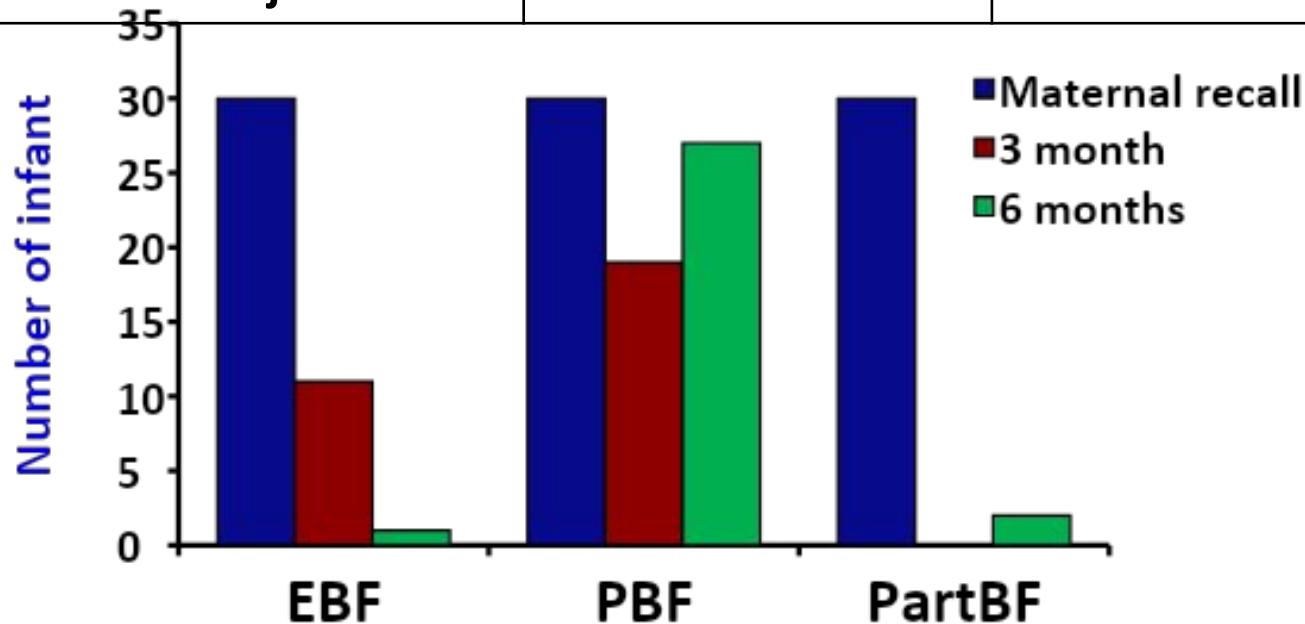
Variables	3 months		6 months	
	Boy (<i>n</i> = 13)	Girl (<i>n</i> = 17)	Boys (<i>n</i> = 13)	Girls (<i>n</i> = 17)
Human milk intake, g.kg ⁻¹ .d ⁻¹	131 ± 23	136 ± 19	111 ± 23	113 ± 28
Weight gain, g.d ⁻¹	23 ± 9	14 ± 12	5 ± 12	7 ± 8
Weight-for-age z score	0.26 ± 0.98	-0.53 ± 0.98	0.13 ± 0.73	-0.84 ± 1.02
Lenght-for-age z score	0.43 ± 0.95	-0.31 ± 1.03	0.07 ± 0.59	-0.84 ± 0.88
Ponderal index ² , g.cm ³	2.73 ± 0.19	2.67 ± 0.28	2.62 ± 0.2	2.58 ± 0.22

Human milk intake's infant from some countries



Mothers were categorized as practicing exclusive breastfeeding

Classification	Non-milk oral intake	3 months age	6 months age
Exclusive Breast Feeding	≤ 25 g/d	11	1
Predominant Breast Feeding	$> 25 - 220$ g/d	19	27
Partial Breast Feeding	> 220 g/d	0	2
Total of subjects		30	30

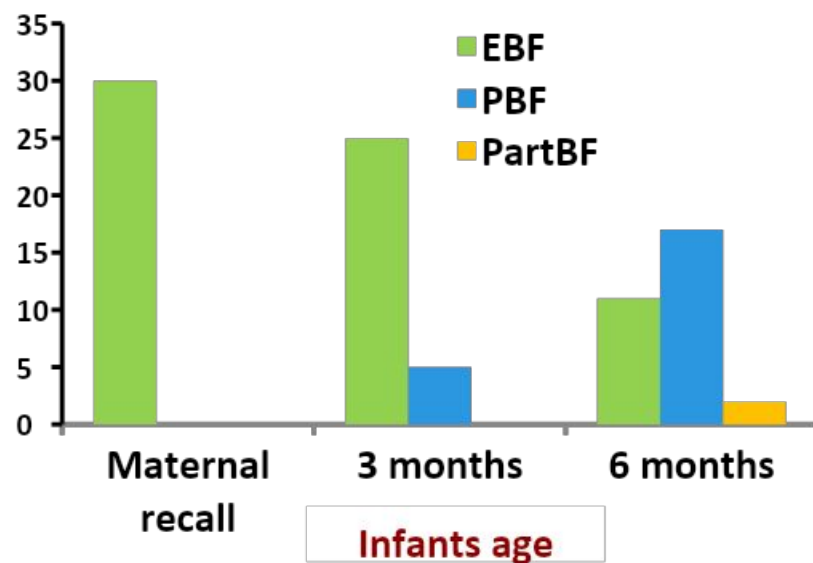
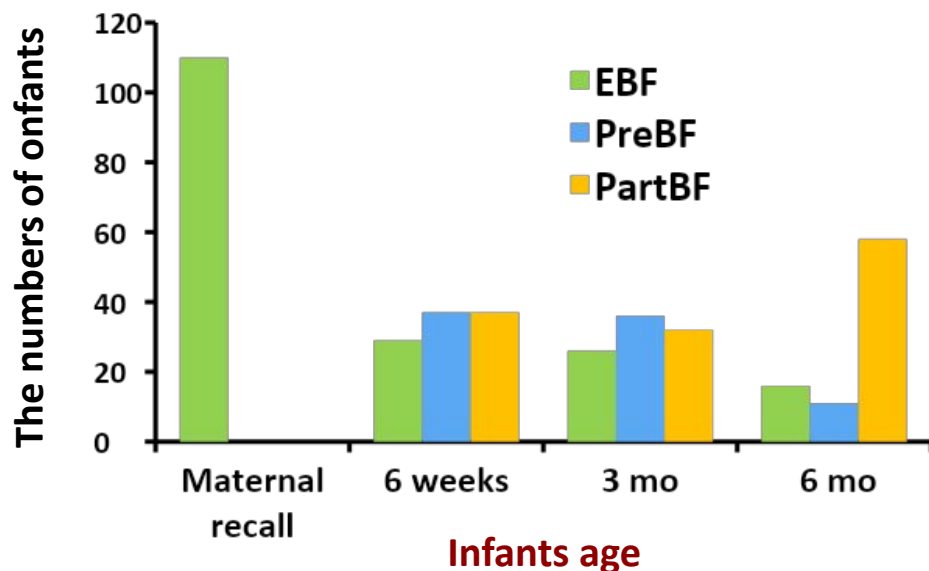


The cutoff of NMOI = 86.6 g/day (Liu Z., et al. 2019) 121 pairs mother and infant in Sumedang, West Java



Thailand	Maternal recall	6 weeks	3 mo	6 mo
EBF	110	29	26	16
PreBF		37	36	11
PartBF		37	32	58

Indonesia	Maternal recall	3 months	6 months
EBF	30	25	11
PBF		5	17
PartBF		0	2



- Wanabhorn Tongchom et al., Human Milk Intake of Thai Breastfed Infants During the First 6 Months Using the Dose-to-Mother Deuterium Dilution Method. Food and Nutrition Bulletin. 2020; 1-12.
- The new cutoff is 86.6 g/day. This value is the maximum water intake that is recommended for an infant to be classified as EBF.

- 1) The milk intake of Indonesian infants during the first 6 months of their life could be classified **sufficient (76.7 %, > 650 g/day)**.
- 2) Using D₂O to the mother technique, it was found that **36.7% of infants were categorized as exclusive breastfed infants at 3 months age and only 3.3% at 6 months age (cutoff 25 g/day)**.
- 3) The % fat mass (18,4 at 3 mo and 15.5 at 6 mo) of Indonesian lactating mothers were lower than that reported in Sri Lanka (24.8% at 6 mo) and Brazil (24.7% at 4 mo) using the same methodology.
- 4) Exclusive breastfeed practices of 30 pairs of mother-infant still showed **that it had not been properly adhered to by mothers who deliver babies**.
- 5) It is recommended that 'mother and baby health officer' in Indonesia should be **increasingly promoting** in order mothers in urban and rural areas to breastfeed exclusively to their infants up to 6 months age.

