

Behavior Change Communication and Breastfeeding Practices Measured by Deuterium-Oxide Turnover Method Among Infants Aged 4-5 Months in Rural Senegal

Ousmane Diongue^{1,*}, Adama Diouf¹, Pape Sitor Ndour¹, Mane Hélène Faye¹, Abdou Badiane¹, Mbeugué Thiam¹, Olouwafemi Mistourath Mama¹, El Hadji Momar Thiam², Nicole Idohou Dossou¹

¹Laboratoire de Recherche en Nutrition et Alimentation Humaine, Département de Biologie Animale, Faculté des Sciences et Techniques, Université Cheikh Anta Diop (UCAD), Dakar, Senegal

²Conseil National de Développement de la Nutrition, Rue 7 - Point E, Dakar, Senegal

Email address:

ousmane1.diongue@ucad.edu.sn (Ousmane Diongue), adama.diouf@ucad.edu.sn (Adama Diouf), papesitor2222@gmail.com (Pape Sitor Ndour), manehelene.faye@ucad.edu.sn (Mane Hélène Faye), abdou2.badiane@ucad.edu.sn (Abdou Badiane), mbeugue.thiam@ucad.edu.sn (Mbeugué Thiam), femi.mama@ucad.edu.sn (Olouwafemi Mistourath Mama), ethiam@cndn.sn (El Hadji Momar Thiam), nicole.dossou@ucad.edu.sn (Nicole Idohou Dossou)

*Corresponding author

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Abstract: Breastfeeding promotion is widely recognized as one of the most cost-effective investments in promoting optimal child health, growth, and development. Several studies have shown that breastfeeding education and counseling interventions have a significant impact on improving breastfeeding practices, including exclusive breastfeeding (EBF) rates. However, very few studies have evaluated the association between breastfeeding education and infant breast milk intake. The objective of this study was to assess the contribution of behavior change communication provided by the Nutrition Enhancement Program (NEP) on improving infant breast milk intake and breastfeeding practices. A comparative cross-sectional study was conducted in 12 Local Communities in rural Senegal, of which 6 were located in the NEP intervention area and the remaining, in the non-intervention area. Breast milk intake and EBF were measured using the deuterium dose-to-mother isotope dilution (DTM) in 140 mother-infant (4-5 mo.) pairs. Breastfeeding and complementary feeding practices were also assessed by questionnaire. Student's t-test, ANOVA, chi-square test and McNemar test were used to compare means and percentages. A mixed model linear regression was performed to identify the associated factors of breast milk intake, measured by DTM. Breast milk intake was significantly higher in infants from the NEP area (994.7 ± 197.3 g/d), compared to those from the non-NEP area (913.6 ± 222.8 g/d), $p=0.023$. Consumption of water from sources other than breast milk was not different between the groups. EBF rates measured by DTM were 37.0% and 28.4% in the NEP area and the non-NEP area, respectively. There was no difference on exclusive breastfeeding rate between the two areas regardless of evaluation method. Stunting and wasting were associated with lower milk intake of 107.1 g/d and 211.9 g/d, respectively. Mothers' participation in behavior change communication activities improved infants' breast milk intake, but not EBF rates. Lower breast milk consumption was associated with stunting and wasting.

Keywords: Breast Milk Intake, Exclusive Breastfeeding, Deuterium Dilution, Communication Intervention, Rural Senegal

1. Introduction

To improve child survival, breastfeeding is widely recognized as one of the most cost-effective investments [1]. Absence of breastfeeding has many short-term and long-term disadvantages for a child's health and development [2-4]. In developing countries, non-breastfed infants have 6 to 10 times higher risk of death during the first months of life than breastfed ones [4, 5]. Diarrhea and pneumonia, which are the leading causes of child death, are more frequent and more severe among non-breastfed infants [6]. Among mothers, breastfeeding reduces the risk of breast and ovarian cancer by 26% and 37% respectively, and the risk of type 2 diabetes by 32% [7]. On this basis, the World Health Organization (WHO) and the United Nations Children's Fund (UNICEF) has recommended early initiation of breastfeeding within one hour of birth, exclusive breastfeeding (EBF) for the first six (6) months and continued breastfeeding with diversified, adequate, safe, and age-appropriate complementary food until two years or beyond [8]. The EBF consists of giving only breast milk for the first 6 months of life, without any other food or water except medicines or syrups [9, 10]. Advantages of EBF compared to partial breastfeeding have been demonstrated for several decades by Feachem & Koblinsky, finding that the risk of death from diarrhea in partially breastfed infants aged 0-6 months was 6 to 8 times the risk for exclusively breastfed infants. For those who did not receive breast milk, the risk was 25 times higher than for those who were exclusively breastfed [11]. Recent studies have also confirmed the benefits of EBF on infants, including reduced risk of diarrheal diseases, respiratory and gastrointestinal infections and improved neurocognitive function [4, 12-14]. For the mother, EBF accelerates recovery of pre-pregnancy weight and is associated with longer duration of amenorrhea [7, 15, 16].

The World Health Assembly endorsed in 2012, through its resolution WHA65.6, the comprehensive implementation plan on maternal, infant and young child nutrition and set a series of six global nutrition targets among which to increase exclusive breastfeeding rates in the first 6 months of life to at least 50% by 2025 [17]. Despite the imminence of this deadline, EBF rates are well below 50% in most African countries [18]. In Senegal, only 41% of children under 6 months are exclusively breastfed [19]. Thus, there is an urgent need to accelerate efforts to scale up effective programs to promote exclusive breastfeeding. Several studies have shown that breastfeeding education and counseling interventions have a significant impact on improving breastfeeding practices, including early initiation of breastfeeding, EBF rates and continued breastfeeding [20-22]. The review conducted by Imdad et al. in 2011 concludes that EBF rates rose significantly as a result of educational interventions, with a greater effect observed in developing countries [22].

In Senegal, the Nutrition Enhancement Program (NEP) initiated by the *Conseil National de Développement de la Nutrition* (CNDN), formerly the *Cellule de Lutte contre la Malnutrition* (CLM) since 2002, offers a set of community nutrition services in the country. Community nutrition sites are

established in the intervention villages for the implementation of program activities. According to the last LQAS survey of 2021, EBF rates among 0 - 6 month-olds in the intervention area vary from 61% to 89% for Kaffrine and from 55% to 74% for Kaolack. However, these data do not allow for an accurate assessment of the actual level of the indicators and uptake of good breastfeeding practices. Indeed, these evaluations use the maternal recall to measure EBF in children under 6 months of age, whereas many studies have shown that this method tends to overestimate the real rates of this practice [23-25].

The deuterium isotope dilution technique (DTM) developed by Coward et al. [26] is a widely used method in human nutrition for assessing milk production and determining breastfeeding type. It allows the amount of breast milk consumed by the infant and the amount of water from sources other than breast milk to be accurately determined in order to objectively assess the type of breastfeeding. DTM has the advantage of being more accurate, less restrictive than the differential weighing method, does not interfere with infant feeding habits, and is easier to perform in the field [27-29]. In Senegal, two studies using this technique (DTM) were conducted in Dakar, the first in mother-infant pairs of 3 months to assess the impact of a food supplement on nutritional status and milk production [30] and the second in mother-infant pairs of 6 months to measure the coverage of the infant's energy needs [31].

Although the effect of counseling and education interventions on improving breastfeeding practices has been documented, their association with infant milk intake is underestimated. To our knowledge, only, the study conducted by Albernaz et al., in 2003 on a sample of 188 Brazilian infants assessed the impact of breastfeeding counselling on infant milk intake [32]. Thus, the objective of this study was to see if mother participation in behavior change communication activities improves breastfeeding practices as measured by DTM.

2. Method

2.1. Study Design

This was a comparative cross-sectional study conducted in rural areas of Senegal (Kaolack and Kaffrine). The study was conducted in 12 local communities (LCs) selected by randomly drawing 6 LCs from each area. Of the 65 LC in the two regions, the program covers 48 LC (NEP area) where the entire NEP service package is implemented. In the remaining 17 LC (non-NEP area), no intervention is conducted by the CNDN, but women receive standard messages on IYCF through the media or health workers.

2.2. Study Setting

The NEP has been implemented across the country since 2002. Phase 3 of the program was launched in 2017, covering 68%, of the country's local communities. Interventions are implemented from nutrition sites set up in villages or neighborhoods and management is entrusted to 2 community

health workers. Pregnant, lactating women and children under 5 years of age benefit from a nutrition service package including: i) growth monitoring and promotion (GMP) which targets children under 2 years of age, ii) integrated management of childhood illnesses (IMCI-C), iii) early detection and management of acute malnutrition in children aged 6-59 months and iv) behavior change communication (BCC) targeting mothers/caregivers of children under two years of age for the promotion of exclusive breastfeeding (EBF), infant and young child feeding (IYCF), good hygiene and sanitation practices (WASH) for better nutrition. This communication is carried out by community workers through monthly nutrition education sessions focusing on 16 key topics including infant and young child feeding (IYCF), good hygiene and sanitation practices (WASH). Key messages delivered on IYCF include: "Practice early initiation of breastfeeding from birth", "Breastfeed infants exclusively from birth to 6 months", "Introduce appropriate diversified complementary feeding from 6 months", "Continue breastfeeding up to 24 months". The benefits of these practices for the child and the mother are explained using picture boxes for better understanding. These sessions are coupled with growth monitoring and promotion activities targeting 0-24 months old children. The monitoring of the IYCF indicators, particularly the EBF rate is carried out by Lot Quality Assessment Sampling (LQAS) surveys every 6 months as well as from the Demographic and Health Surveys (DHS).

2.3. Participants, Recruitment and Inclusion Criteria

Participants were mother-infant (4 - 5.5 mo.) pairs living in the 12 LC targeted. For the NEP area, the mother-infant pairs were identified from the child follow-up records available at the nutrition sites. In the non-NEP area, mother-infant pairs were identified and recruited using the birth or immunization registers of the health facilities in each LC. In each LC, all mother-infant (4 - 5.5 mo.) pairs who met the inclusion criteria for participating in the study were included. Children suffering from severe acute malnutrition, diseases requiring hospitalization or congenital illnesses that do not allow the child to grow linearly (e.g. achondroplasia or Turner syndrome), as well as mothers suffering from diseases (such as mastitis) that prevent them from breastfeeding their children properly, were not included in the study.

2.4. Sampling and Sample Size

Sampling was carried out at two levels. In each area, 6 LC were randomly drawn. The sample size was calculated from the national EBF rate of 24% among children aged 4-5 months [33] with the formula of Machin *et al.*, [34] to detect a 20% difference in EBF rate between NEP area and non-NEP area using the DTM. Considering a power of 80%, a significance level of 95% with a design effect of 1.05, the required sample size is 121 mother-infant pairs (i.e. 60/area). Due to the cumbersome nature of the DTM method, a drop-out rate of 20% was considered; thus the final sample size is 75 mother-infant pairs per group, i.e. an overall sample of 150 pairs.

2.5. Data Collection

2.5.1. Measure of Breast Milk Intake and EBF Rate Using DTM

The principle consists of administering an oral dose of 30 g of deuterium with a minimum purity of 99.8% (Europa Scientific, Ltd, UK) and monitoring its disappearance in the mother and its appearance in her infant through breast milk [29, 35]. Deuterium is a stable isotope of hydrogen (non-radioactive) that exists in nature, in water and in food. It is administered orally as deuterium oxide at a dose that is not harmful to humans (max 0.1%) and has no toxicity [36]. The dose administered to the mother is homogeneously distributed in her body water and thus appears in breast milk. Thus, the breastfed infant receives deuterium through breast milk, which also enriches their body water [28, 35]. This method is very sensitive because deuterium enrichment of the infant's saliva can only come from breast milk. Saliva samples were taken with sterile cotton balls from the mother-infant pair before the dose (pre-doses) at day 0 (D0) to determine the baseline level and over a period of 14 days (D1, D2, D3, D4, D13 and D14) after the dose. Deuterium enrichment of saliva samples was measured with a Fourier Transform Infrared Spectrometer (FTIR Shimadzu IR Affinity, Kyoto, Japan). Samples were measured in duplicate with a coefficient of variation (CV) less than 1% for all enrichments retained in the analysis. A set of assumptions and equations have been developed by several authors for the calculation of human milk intake (HMI) and water from sources other than human milk intake (NMOI) by infants [29, 37, 38]. The enrichment values of the mother-infant pair were entered in an Excel spreadsheet that integrates all the equations and with the Solver function that allows to adjust the enrichment of the samples over 14 days. Classification of breastfeeding type was based on the intake of water from sources other than breast milk (non-milk oral intake) according to recommended thresholds [39]. EBF was defined as a non-milk oral intake < 86.6 g/d.

2.5.2. Anthropometry

The weight and height of the mothers and their infants were measured in duplicate on the first day (D0) and the 14th day (D14) of saliva sample collection. The weight of the mothers was measured to the nearest 100 g using an electronic scale (SECA 874 U, GmbH & Co, Hamburg, Germany). Infants were weighed without any clothing using an electronic baby scale (SECA 354, GmbH & Co, Hamburg, Germany) with a maximum capacity of 20 kg and an accuracy of 10 g. The mother's height and the length of the infant were measured at 0.1 cm with a Seca board (SECA 216, GmbH & Co, Hamburg, Germany). For the mothers, body mass index (BMI; kg/m²) was calculated and mothers were categorized as underweight (<18.5 kg/m²), normal weight (18.5 to 24.9 kg/m²), overweight (25 to 29.9 kg/m²) and obese (≥30 kg/m²) [40]. Among infants, weight-for-length z-scores (WLZ) and length-for-age z-scores (LAZ) were calculated and they were classified as wasted (WLZ < -2 z-score) or stunted (LAZ < -2 z-score) using the WHO child growth standard reference [41].

2.5.3. Infant Feeding Practices and Characteristics of Households and Mothers Assessed by Questionnaire

Information on initiation of breastfeeding, exclusive breastfeeding, use of infant formula and complementary feeding was collected by the WHO IYCF questionnaire [42, 43]. Mothers were asked whether the child was still breastfeeding; whether they had already started giving water or complementary foods to their child, and if so, at what age.

Socio-economic data, household food security, breastfeeding patterns, knowledge, attitudes and practices (KAP) of mothers and infant morbidity were collected using a questionnaire. A socio-economic status (SES) index was generated using principal component analysis based on household characteristics (household occupancy, source of drinking water, type of sanitation facilities, type of fuel used for cooking) and possession of durable goods (household equipment and livestock) [44]. Based on the index score of each household, a household SES indicator consisting of 3 categories (lowest, medium and acceptable) was obtained. Household food insecurity was assessed using the Household Food Insecurity Access Scale (HFIAS) [45]. Data on children's immunization against BCG, Polio, Pentavalent and Pneumococcus were collected from children's health records. A 15-day recall was conducted to assess common childhood illnesses such as fever, diarrhea and acute respiratory infections. Fever is defined as a body temperature higher than normal as reported by the mother. Diarrhea is defined as more than 3 loose or watery stools in one day. Acute respiratory infections are defined as diseases of the respiratory system, nasopharynx, and lungs. All questionnaires

were administered on the first day (Day 0) of the EBF measurement with the DTM.

2.6. Statistical Analysis

Data entry and processing were performed using Epi-info 3.5.1 (Centers for Disease Control and Prevention, Atlanta, USA), and Microsoft Excel 2016 (Microsoft Corporation, Redmond, USA). Statistical analysis was performed using STATA/SE 12.0 (STATA Corporation, Texas, USA). Categorical variables were expressed as percentages (%), and continuous variables were expressed as mean \pm standard deviation ($M \pm SD$), for normally distributed variables and median with interquartile range ($Me [P25; P75]$) for skewed values. Student's t-test and analysis of variance (ANOVA) were used to compare means, while the Wilcoxon Mann-Whitney test was used to compare medians between groups. Pearson's chi-square test and Fisher's exact test were used to compare percentages. McNemar test was used to compare the two methods of measuring EBF.

A mixed model linear regression was performed to determine factors associated with infant milk intake. All socio-economic and dietary variables, as well as those related to maternal and infant characteristics, which were significant at $P < 0.1$ in the binary regressions with infant milk consumption, were included in a multiple model controlling for a random group effect. The age of the child at the time of measurement (D_0) was included as a covariate, as breast milk consumption and breastfeeding status changed with age. For all statistical analyses, a P-value < 0.05 was used for significance.

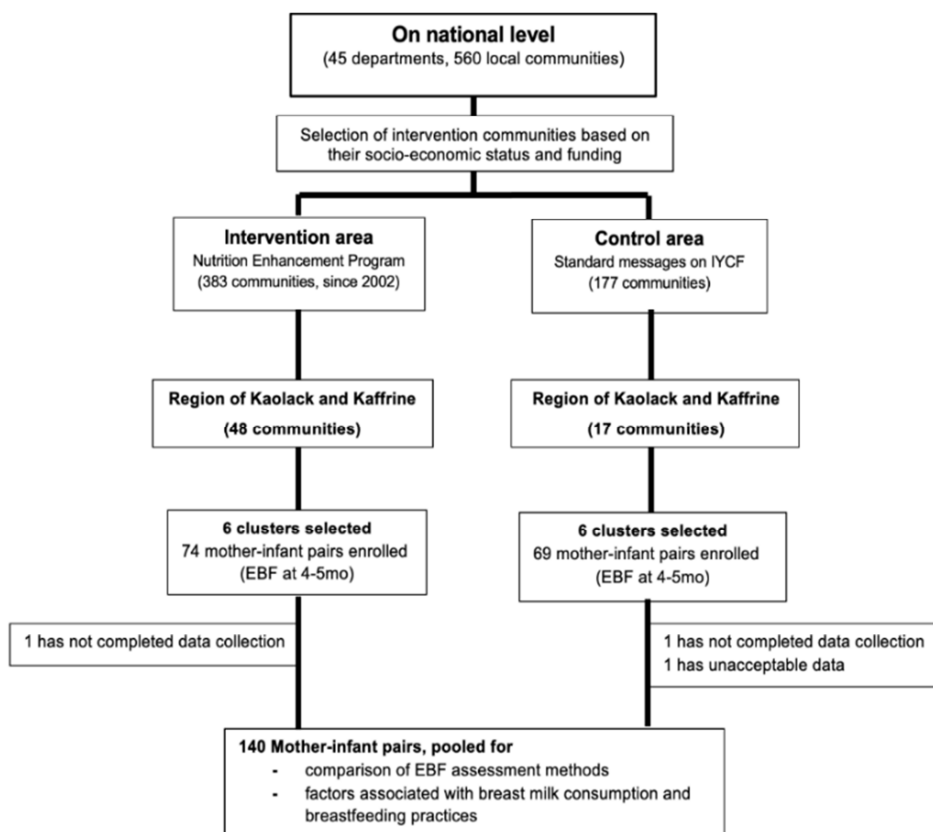


Figure 1. Study profile.

3. Results

3.1. Characteristics of the Study Population

In each LC, all mother-infant (4-5 mo.) pairs that met the inclusion criteria were included in the study. Only two cases of refusal were recorded and this was due to the absence of the heads of household who had to agree to participate in the study. A total of 143 mother-child pairs were involved in the study. Of these, 2 did not complete the collection of saliva samples in the field and 1 had physiologically unacceptable data after enrichment measurements. Thus, 140 pairs had complete data and were retained for the overall analysis (Figure 1).

Data on mother-infant pairs households are presented in Table 1. The results showed that in the NEP area all heads of household were male, while in the non-NEP area there were 4.5% female heads, with no difference between the two areas. The average number of children under 5 years of age was comparable between the two areas. Both areas had the same socio-economic characteristics, with more than 50% of households in the middle and low categories. Food insecurity affected 38% and 37% of households in the NEP and non-NEP areas, respectively. There were no significant differences in socio-economic and food characteristics between the two areas.

Table 1. Characteristics of mother-infant pair's households.

Characteristics	NEP-area (n=73)	non-NEP-area (n=67)	P value
	M ± SD or %	M ± SD or %	
Head of household gender			
Male	100 (73)	95.5 (64)	0.068
Female	-	4.5 (3)	
Number of children under 5 years	3.0 ± 1.4	3.6 ± 2.3	0.094
Socio-economics status			
Low	37.0 (27)	29.8 (20)	0.096
Medium	38.4 (28)	28.4 (19)	
Acceptable	24.7 (18)	41.8 (28)	
Household food insecurity			
Food insecure	38.4 (28)	37.3 (25)	0.733
Mildly food insecure	12.3 (9)	14.9 (10)	
Moderately food insecure	16.4 (12)	10.4 (7)	
Severely food insecure	9.6 (7)	11.9 (8)	

M ± SD mean ± standard deviation; % (n): percentage (number)

3.2. Characteristics of Mother and Infant

The characteristics of mothers and infants by group are detailed in Table 2. The mean age of the mothers (25 ± 5 years) was comparable between the two areas. Mothers were mostly multiparous (73%) and the distribution of ethnic groups was identical in both areas, with a predominance of Wolof/Lebou ethnic (68.5% NEP-area vs. 65.7% non-NEP-area). More than half (63.6%) of the mothers were illiterate and very few (3.6%) had attained middle or higher education. The proportion of mothers living in polygamous households was significantly higher in the NEP intervention area (45.2%) than in the non-NEP-area (25.4%) (p=0.014). Mother's BMI mean was

similar in the two groups (22.1 ± 4.3 kg/m²). According to the WHO classification, 21.4% of them were underweight (BMI<18.5 kg/m²) while 16.4% and 6.4% were overweight (25<BMI<30 kg/m²) and obese (BMI>30 kg/m²), respectively.

For infants, the mean age was 4.9 ± 0.5 months; 52.9% were boys, and almost all were correctly vaccinated (99.3%) with no difference between the two areas. Regarding morbidity during the last 15 days prior to the study, the prevalence of diarrhea and acute respiratory infection were significantly higher among infants in the non-NEP-area compared to those in the NEP-area (p<0.05). The mean WHZ and LAZ indices were comparable between the two groups, as were the proportions of stunting and wasting.

Table 2. Mother-infant pair characteristics.

Mothers	NEP-area (n=73)	non-NEP-area (n=67)	P value
	M ± SD or %	M ± SD or %	
Age (year)	26 ± 5	24 ± 5	0.274
Parity			
Primiparous	19.18 (14)	34.33 (23)	0.042
Multiparous	80.8 (59)	65.7 (44)	
Marital status			
Married	98.3 (72)	100 (67)	0.336
Divorced/widowed	0.7 (1)	0 (0)	
Ethnicity			
Wolof/Lebou	68.5 (50)	65.7 (44)	0.153
Serere	11.0 (8)	10.4 (7)	
Pulaar	12.3 (9)	22.4 (15)	

Mothers	NEP-area (n=73)	non-NEP-area (n=67)	P value
	M ± SD or %	M ± SD or %	
Others	8.2 (6)	1.5 (1)	
Education			
None	65.7 (48)	61.2 (41)	0.485
Primary school	26.0 (19)	23.9 (16)	
Secondary school	6.8 (5)	9.0 (6)	
High school/University	1.4 (1)	6.0 (4)	
BMI (kg/m ²)	22.1 ± 4.1	22.1 ± 4.6	0.951
Underweight	19.2 (14)	23.9 (16)	0.726
Normal weight	60.3 (44)	50.7 (34)	
Overweight	15.1 (11)	17.9 (12)	
Obese	5.5 (4)	7.5 (5)	
Infants			
Age (month)	4.9 ± 0.5	4.9 ± 0.5	
Sex			
Boys	49.3 (36)	56.7 (38)	0.381
Girls	50.7 (37)	43.3 (29)	
Immunization ^a	100 (73)	98.5 (66)	
Morbidity			
Diarrhea	30.1 (22)	58.2 (39)	0.001
Fever	15.1 (11)	17.9 (12)	0.650
Acute respiratory infection	2.7	11.9	0.035
WLZ	-0.2 ± 1.1	0.08 ± 1.0	0.154
Wasting	8.2 (6)	6.0 (4)	0.606
LAZ	-0.5 ± 1.1	-0.5 ± 1.0	0.879
Stunting	11.0 (8)	7.4 (5)	0.476

M ± SD mean ± standard deviation; % (n): percentage (number); BCC Behavior Change Communication; BMI: body mass index; WLZ: weight-for-length z-score; LAZ: length-for-age z-score; Underweight: BMI < 18.5 kg/m²; normal weight: 18.5 ≤ BMI < 25 kg/m²; overweight: 25 ≤ BMI < 29.9; obese: BMI ≥ 30 kg/m²; wasting: WLZ < -2 z-score; stunting: LAZ < -2 z-score

^aBoth BCG, Polio, Pentavalent, Hepatitis B, and Pneumococcal vaccines.

3.3. Breast Milk Intake, Non-Milk Oral Intake and Breastfeeding Practices in Both Area

Data on breastfeeding and infant feeding practices measured by DTM and questionnaire are presented in Table 3. The average breast milk intake per day was significantly higher among infants in the NEP-area (994.7 ± 197.3 g/d) compared to those of non-NEP-area (913.6 ± 222.8 g/d) p=0.023. But the daily intake of water from sources other than breast milk consumed by the infants was comparable between the two areas. According to the DTM, 37% and 28.4% of infants were exclusively breastfed in the NEP-area and non-NEP-area,

respectively. However, according to the mothers' declaration, 46.6% of the infants in the NEP-area and 44.8% of those in the non-NEP-area were exclusively breastfed. No difference was observed in exclusive breastfeeding practices between the areas, regardless of the evaluation method. Other practices such as early initiation of breastfeeding, giving water to the infant and early introduction of complementary feeding were comparable between the two groups. The comparison between the two methods of measuring breastfeeding practices showed a significant difference between the exclusive breastfeeding rate measured by the DTM and that obtained by the questionnaire (p=0.016; 95%CI [-0.28; -0.01]).

Table 3. Breast milk intake and breastfeeding practices.

	NEP-area (n=73)	non-NEP-area (n=67)	P value
Breastfeeding practices (DTM)			
Human milk intake (HMI) (g/d) M±SD	994.7 ± 197.3	913.6 ± 222.8	0.023
Non-milk oral intake (NOMI) (g/d) Me [25e – 75e]	123 [44 – 226]	155 [75 – 260]	0.105
EBF	37.0 (27)	28.4 (19)	0.278
Non-EBF	63.0 (44)	71.6 (46)	
Breastfeeding practices (questionnaire) (%)			
EBF	46.6 (39)	44.8 (37)	0.863
Non-EBF	53.4 (36)	55.2 (34)	
Infant feeding practices (%)			
early initiation of breastfeeding	74.0 (54)	70.1 (47)	0.614
Infant receiving water	37.0 (27)	49.2 (33)	0.143
eating porridge	15.1 (11)	20.9 (14)	0.369
eat the family meal	1.4 (1)	4.5 (3)	0.270

M ± SD: mean ± standard deviation; Me [25e – 75e]: median with interquartile range; % (n): percentage (number);

DTM: deuterium-oxide turnover method; g/d: gram/day; EBF: exclusively breastfeeding infants; Non-EBF: Non-exclusively breastfed

3.4. Factors Associated with Infant Breast Milk Intake

All socio-economic and dietary variables as well as those related to maternal and infant characteristics that were significant at $P < 0.1$ in the binary mixed model linear regressions with infants milk intake were entered into multiple linear regression analyses. The results in Table 4 showed that participation in behavior change communication activities was associated with a higher consumption of breast milk by infants of 87.7 g/d compared to the mothers in the Non-NEP area

($p = 0.032$). The type of breastfeeding was also associated with infant milk intake. Non-exclusively breastfed infants consumed 104.6 g/d less milk compared to exclusively breastfed infants ($p = 0.002$). Giving infants porridge reduced their breast milk intake by 156.7 g/d ($p < 0.001$). Significant negative associations were found between lower milk intake and infant nutritional status. Stunted and wasted infants had a lower breast milk intake of 107.1 g/d ($p = 0.044$) and 211.9 g/d ($p = 0.001$) respectively, compared to infants with normal nutritional status.

Table 4. Factors associated with infant's breast milk intake.

HMI (g/d)	β coefficient	95% CI	P value
NEP-area	87.7	7.5; 167.8	0.032
EBF	104.6	37.7; 171.4	0.002
Porridge consumption	-156.7	-245.5; -67.9	0.001
Stunting	-107.1	-211.5; -2.6	0.044
Wasting	-211.9	-329.2; -94.6	0.001

HMI: Human milk intake, g/d: gram/day, BCC: behavior change communication, EBF: exclusive breastfeeding

4. Discussion

This study using the isotope method to assess breast milk intake and exclusive breastfeeding practices is the first to be conducted in rural Senegal. It implies the use of precise methods in the monitoring and evaluation of programs to better assess their impact. Several studies have shown that breastfeeding counseling and education interventions improve the rate and duration of exclusive breastfeeding and the continuation of breastfeeding [20-22], however, their association with infant milk intake is under-estimated [32].

This finding showed that infants in the NEP area, whose mothers participated in breastfeeding education and counseling activities, consumed more breast milk than infants in the non-NEP zone. Indeed, a variation of 87.7 g/d was observed between infants in the two groups, and this variation was not related to the age of the children or to other potential confounding variables. This implies that mothers' participation in breastfeeding education and counselling improved their knowledge and predisposition to breastfeed their children correctly and more frequently. A similar result was observed in a recent study in Benin, where mothers participating in the breastfeeding education program were more likely to breastfeed their children more frequently and for longer [46]. However, our results differ from that of Albarnaz *et al.* who found no significant difference in breast milk intake between a control group and a group receiving breastfeeding counseling [32]. Nevertheless, a difference of 38 ml/d in favor of the latter group was found in this study despite the relatively small sample size and low statistical power of the comparisons, which may have limited the effect of breastfeeding counseling on infant breast milk intake.

This analysis showed that exclusive breastfeeding was associated with higher breast milk consumption. This result is consistent with the fact that exclusively breastfed infants received only breast milk, while non-exclusively breastfed

infants received water or other foods that decreased their breast milk intake. Indeed, infants who were given porridge had a 156.7 g/d decrease in breast milk intake. The early introduction of water and complementary foods results in a reduction in the frequency of feedings and thus a decrease in the amount of breast milk consumed by the infant [28, 31, 46]. The infant's daily intake of breast milk was also higher than WHO estimates for exclusively breastfed infants [47]. However, studies have shown that breast milk intake during this period can vary from 400 to 1200 g/d [47, 48]. The WHO estimates are based on a compilation of data usually obtained by differential weighing, whereas several studies have shown that breast milk intakes measured by the deuterium isotope dilution method are generally higher than the WHO values [49, 50]. In addition, the authors of these estimates concluded that the corresponding milk production rates represent only a small group of women and therefore do not reflect the variability in milk production and infant nutrient requirements in the population [47]. The amounts of breast milk consumed by infants in the present study are comparable to values found by other studies using the same isotopic technique in Senegalese infants less than 6 months of age [30, 31].

The association between infant milk intake and nutritional status, established in this study, has rarely been studied in the literature. Indeed, most studies in this area have focused on the relationship between the type of breastfeeding and nutritional status [51, 52]. Our results showed that stunted and wasted children had significantly lower breast milk intakes than those with normal nutritional status. Although the average breast milk intake according to the nutritional status of the infants was not presented in the study, the average among of milk consumed by stunted and wasted infants was low and below the WHO recommendations. This result reinforces the evidence for the importance of optimal breastfeeding for child growth, health, and development.

Several studies have reported that breastfeeding education and counseling interventions have positive effects on improving

breastfeeding practices on both exclusivity and duration of breastfeeding [53, 54]. However, the majority of infants in both areas of this study received water and/or other fluids or complementary foods. This showed that the intervention was not effective in changing the practice of giving water to infants under 6 months of age, which is the main problem with EBF practice in African countries [55]. But, the effectiveness of the communication interventions may depend on several factors including the approach to counseling, the qualification of the actors, and the frequency of sessions [56, 57]. The assessment showed that the number of times women had attended breastfeeding education and counseling sessions in the last 6 months prior to the study was low compared to the WHO recommendation of at least 6 sessions in the pre- and post-natal period. In addition, Mcfadden et al. showed in a recent review that breastfeeding counseling interventions were effective with at least 4 sessions in the postnatal period [53], which was not achieved in the NEP program. The WHO also recommends that counseling should be face-to-face or by telephone, whereas in the NEP program, communication mostly occur in group during growth monitoring and promotion sessions, which may alter mothers' focus on the counseling given and thus affect their knowledge and practice of breastfeeding. In addition, some head nurses in the comparison area have taken initiatives to counsel or educate mothers on breastfeeding, health and hygiene during child immunization sessions. Some mothers also in this zone may receive advice on infant and young child feeding through the media. All this has potentially improved some mothers' knowledge of breastfeeding practices in this area.

EBF rates obtained from mother's self-reporting and accurately measured by DTM were compared in this study. The results showed that the questionnaire overestimated by more than 12% the rate of exclusive breastfeeding objectively measured by DTM in mothers of children aged 4-5 months in rural Senegal. The discrepancy between these two methods found in this study can be explained by the fact that a significant number of women give water to their infants while claiming to practice exclusive breastfeeding. Whereas the isotopic method is very sensitive to the amount of non-milk water received by the infant. Furthermore, having heard at least once about the importance of EBF, mothers tend to respond positively to exclusively breastfeeding their infant with the questionnaire; even if they do not practice it. Several authors have found similar results with sometimes larger differences (40%) between these two methods, notably in Cameroon [23], Morocco [58] and Guatemala [24]. Other studies in Malaysia and Botswana also showed that 100% of mothers reported exclusively breastfeeding their infants by questionnaire, whereas only 3% and 61.2% of infants were exclusively breastfed by the isotopic method in the studies by Yong et al [25] in rural areas and Motswagole et al. [59] in urban areas, respectively.

5. Conclusion

This study, conducted for the first time in rural Senegal,

showed the benefits of behavior change communication in improving human milk intake among 4-5 months aged infants. However, the exclusive breastfeeding rate remains low in rural setting in Senegal despite the behavior change communication intervention received by the mothers through the NEP. This finding highlights the need to improve the communication strategy in accordance with the WHO guidelines on counseling mothers to better promote exclusive breastfeeding until 6 months for optimum infant growth.

Our result confirms that maternal recall questionnaire overestimated the rate of exclusive breastfeeding compared to that measured using DTM. This implies the need to use more accurate methods such as DTM to objectively monitor and assess the impact of breastfeeding promotion programs and interventions. In addition, it would be interesting to see if improved breastfeeding practices, particularly the adequacy of breast milk intake, are associated with subsequent improvements in infant growth, motor and cognitive development.

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