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The impact of daily soap use in rural areas of Senegal on respiratory infectious diseases, fevers and skin microbiota

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ABSTRACT

Objectives: Children aged <5 years are the group most affected by infectious diseases, more specifically in underdeveloped countries. A study was performed to assess the effects of daily soap use on the incidence of diarrhoea, fever, respiratory infection, and the prevalence of pathogenic bacteria on the skin.
Methods: Soap was distributed to the population of the village of Ndiop (test) for use in their daily hygiene but not to the population of the village of Dielmo (control). Fieldworkers daily recorded the clinical events in the two villages and encouraged the use of soap in Ndiop.
Results: A total of 638 people participated in the study. The incidence rates of cough, runny nose and fever significantly decreased in 2016 compared with 2015, unlike that of diarrhoea. In 2016, significant reductions in the incidence rates of cough, runny nose and fever were observed in children aged <15 years in Ndiop. The prevalence of *Streptococcus pneumoniae*, *Staphylococcus aureus* and *Streptococcus pyogenes* in the palms of the hands significantly dropped in Ndiop.
Conclusion: Using soap reduces the incidence of respiratory infections, fevers and the prevalence of pathogenic bacteria on the skin. However, for diarrhoea, additional strategies are needed to improve outcomes.

Introduction

Diarrhoea and pneumonia are the leading causes of death in young children. Of the 6.3 million deaths among children aged < 5 years that occurred in 2013, pneumonia accounted for 15% and diarrhoea accounted for 9% (UNICEF et al., 2014; Liu et al., 2015; Qazi et al., 2015). Together, these two diseases account for 29% of all deaths in children aged <5 years, all causes combined, and cause the loss of 2 million young children each year (Qazi et al., 2015). In Africa, infectious diseases account for 48% of the global deaths of children aged <5 years (UNICEF et al., 2014). The seriousness of the situation and the lack of coordination in the most affected countries led the World Health Organization (WHO) and UNICEF to set up the integrated Global Action Plan for the Prevention and Control of Pneumonia and Diarrhoea (GAPPD) in 2015 (Qazi et al., 2015).

Handwashing has been proposed as a preventive and inexpensive action for preventing diarrhoea and pneumonia (Qazi et al., 2015). The WHO recommends handwashing with soap and water as the most appropriate measure for reducing diarrhoea and pulmonary disease incidences. The incidence of *Shigella* infections in Bangladesh has been significantly reduced as a result of using soap for handwashing (Shahid et al., 1996). A randomized control trial of handwashing performed in Karachi, Pakistan, resulted in a 50% reduction in the incidence of pneumonia compared with the control in children aged <5 years (Luby et al., 2005) and a reduction of 53% of the incidence of diarrhoea in children aged <15 years (Luby et al., 2005). A systematic review concluded that the promotion of handwashing can reduce diarrhoea by 47% (Curtis and Cairncross, 2003). Based on current evidence, handwashing with soap can reduce the risk of diarrhoeal diseases by 42–47% (Curtis and Cairncross, 2003). Approximately half of the world's deaths from pneumonia and diarrhoea are recorded in five countries, most of which are poor and densely populated, namely:

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India, Nigeria, the Democratic Republic of Congo, Pakistan, and Ethiopia (UNICEF, 2012).

In Senegal, diarrhoeal diseases are ranked as the third leading cause of death among children aged <5 years. Their prevalence in 2016 was very high (28%) among children aged 6–23 months (ANSD, 2016). The rate of acute respiratory infections was 3% among children aged <5 years and 5% among those aged 12–23 months (ANSD, 2016). In Sub-Saharan Africa, socioeconomic and cultural challenges undermine efforts to improve hygiene conditions in the general population (Akpabio and Takara, 2014). The improvement of health practices in a community is influenced by the knowledge of good hygiene practices, customs and the economic level (Hoque et al., 1995a; Hoque et al., 1995b). However, handwashing with soap may not be enough to permanently reduce the incidence of diarrhoea and pneumonia in a population. Socioeconomic and cultural factors must be taken into account in the implementation of control strategies (Bulled et al., 2017).

The purpose of this study was to assess the effects of daily soap use in rural areas, compared with a control village, on the incidence of diarrhoeal, febrile and respiratory diseases. It also aimed to assess the prevalence of pathogenic bacteria on the skin that had already been documented in these villages such as: *Streptococcus pneumoniae* (*S. pneumoniae*), *Staphylococcus aureus* (*S. aureus*), *Rickettsia felis* (*R. felis*), *Borrelia crocidurae* (*B. crocidurae*), *Tropheryma whipplei* (*T. whipplei*), *Bartonella quintana* (*B. quintana*), *Streptococcus pyogenes* (*S. pyogenes*), and *Coxiella burnetii* (*C. burnetii*).

Materials and methods

Study area

Dielmo and Ndiop are two Muslim villages in the Fatick region of Senegal. Dielmo (13°43′N; 16°24′W) is located 280 km southeast of Dakar and 15 km from the northern border of the Republic of The Gambia, both of which belong to the central region of the Sudanese savannah area. Dielmo is located on the banks of a freshwater river, the Néma. The village of Ndiop (13°41′N; 16°22′W) is 5 km from Dielmo and is located in a dry area. They are populated by farmers and ranchers. Provision of power to villages has brought about a profound change in the way of life of their populations. Commercial activities are expanding, and the mode of travel has also been revolutionised by the adoption of Jakarta motorbikes (Lagier et al., 2017). Water consumption from wells is widespread, despite the existence of fountains.

This study used Dielmo as the control village and Ndiop as the test village. In the control village, no intervention was undertaken. Preparatory meetings with the inhabitants of the two villages were organised. The populations were informed and made aware of the objectives of the study and the importance of respecting the instructions. In particular, the inhabitants of the control village were invited to maintain their health and social habits. On the other hand, for the test village, the population was invited to use soap every day for washing hands and the body.

This study was divided into five parts that were intended to allow capture of social, clinical and health events, bacteriological

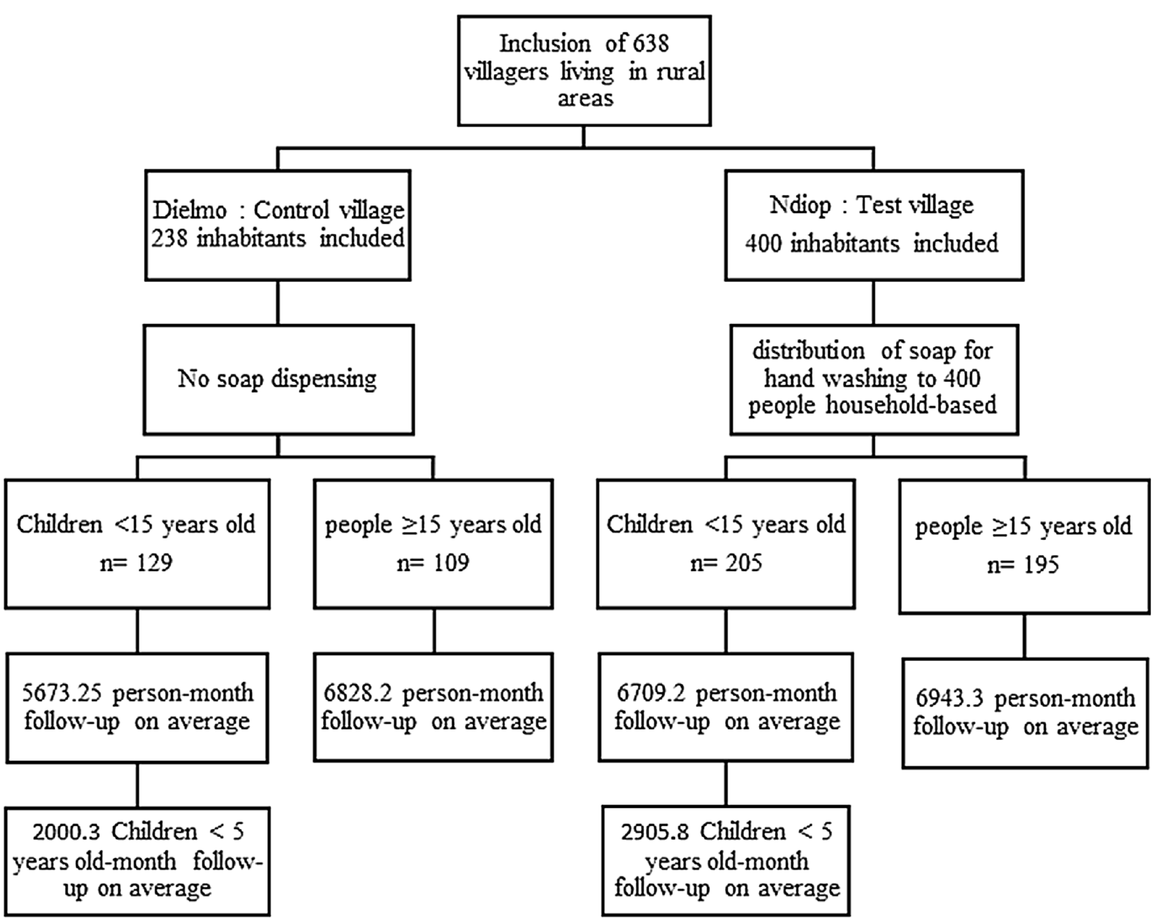

Figure 1. The study design.

diagnoses and soap use during the study period. Social data and biological samples were obtained through a quarterly survey questionnaire followed by skin swabs taken from the palms of the hands. This activity was carried out by investigators with good levels of experience. Clinical data were collected every day throughout the study by nurses at the health posts in the two villages. The soap supply was the responsibility of a team member. Finally, the isolation of pathogenic bacteria was ensured by a laboratory technician.

Data collection

Participants and procedures

From January to December 2016, the study was conducted in the villages of Dielmo and Ndiop. The aim was to study the impact of the daily use of an unscented soap (Marseille-type soap) on fevers and respiratory and diarrhoeal diseases. Data were collected through daily active and passive monitoring in both villages during the study period. The study design is summarised in Figure 1, including different stages of the study, with details of the numbers of children aged <5 and <15 years.

Participants. Residents of all ages and of both sexes from both villages were targeted to participate in the study. In Ndiop, the participants had to commit to complying with the soap use instructions. They also had to agree to use only the soap given to them by the project. In Dielmo, the participants had to commit to not changing their lifestyle or being influenced by the distribution of soap in Ndiop.

Clinical surveys. From January to December 2016, the field workers (two per village) met daily with each participant to record their temperature and check their health. Whenever the sought symptoms were detected, the patient was referred to the village health post for a consultation with the nurses. Data on the participant's health status were recorded on the registration documents kept by the investigators, and data from the consultations were recorded on pathological episode forms.

Soap distribution. In each village, concessions were divided into households headed by a wife. Ordinary solid Marseille soap type, based on sodium hydroxide, was used. The soap was not supplemented with a specific antibacterial agent. During the

Table 1
Target sequences, primers and probes used in the study.

Targeted organism	Targeted gene	Name	Sequences	References
All *Rickettsia* except *R. typhi* and *R. prowazekii*	RC0338	1029-F1	GAM AAA TGA ATT ATA TAC GCC GCA AA	Ndiaye et al. (2018)
		1029-R1	ATT ATT KCC AAA TAT TCG TCC TGT AC	
		Rick1029_MBP	6FAM- CGG CAG GTA AGK ATG CTA CTC AAG ATA A-TAMRA	
Rickettsia felis	Biotin synthase	R_fel0527_F	ATG TTC GGG CTT CCG GTA TG	Ndiaye et al. (2018)
		R_fel0527_R	CCG ATT CAG CAG GTT CTT CAA	
		R_fel0527_P	6FAM- GCT GCG GCG GTA TTT TAG GAA TGG G -TAMRA	
All *Bartonella*	ITS	Barto_ ITS3_ F	GAT GCC GGG GAA GGT TTT C	Ndiaye et al. (2018)
		Barto_ ITS3_ R	GCC TGG GAG GAC TTG AA CCT	
		Barto _ITS3 _P	6FAM- GCG CGC GCT TGA TAA GCG TG -TAMRA	
All *Borrelia*	16S	Bor_16S_3_F	AGC CTT TAA AGC TTC GCT TGT AG	Ndiaye et al. (2018)
		Bor_16S_3_R	GCC TCC CGT AGG AGT CTG G	
		Bor_16S_3_P	6FAM-CCG GCC TGA GAG GGT GAA CGG-TAMRA	
Borrelia crocidurae	glpQ	Borcro_glpQ_F	CCT TGG ATA CCC CAA ATC ATC	Ndiaye et al. (2018)
		Borcro_glpQ_R	GGC AAT GCA TCA ATT CTA AAC	
		Borcro_glpQ_MGB_P	6FAM-ATG GAC AAA TGA CAG GTC TTAC-MGB	
Coxiella burnetii	IS1111A	Coxbur_IS1111_0706_F	CAA GAA ACG TAT CGC TGT GGC	Ndiaye et al. (2018)
		Coxbur_IS1111_0706_R	CAC AGA GCC ACC GTA TGA ATC	
		Coxbur_IS1111_0706_P	6FAM-CCG AGT TCG AAA CAA TGA GGG CTG-TAMRA	
	Hyp. Protein	Coxbur_IS30A_3_F	CGC TGA CCT ACA GAA ATA TGT CC	Ndiaye et al. (2018)
		Coxbur_IS30A_3_R	GGG GTA AGT AAA TAA TAC CTT CTG G	
		Coxbur_IS30A_3_P	6-FAM- CAT GAA GCG ATT TAT CAA TAC GTG TAT GC-TAMRA	
Staphylococcus aureus	NucA	Saur__NucA__F	TTG ATA CGC CAG AAA CGG TG	Ndiaye et al. (2018)
		Saur__NucA__R	TGA TGC TTC TTT GCC AAA TGG	
		Saur__NucA_MGB_P	6FAM- AAC CGA ATA CGC CTG TAC -MGB	
	Amidohydrolase	Saur_F	CCT CGA CAG GTA ACG CAT CA	Ndiaye et al. (2018)
		Saur_R	AAA CTC CTA TCG GCC GCA AT	
		Saur_P	6FAM-TGC AAT GGT AGG TCC TGT GCC CA	
Streptococcus pyogenes	Hypothetical	Spyo_hypp_F	ACA GGA ACT AAT ACT GAT TGG AAA GG	Ndiaye et al. (2018)
		Spyo_hypp_R	TGT AAA GTG AAA ATA GCA GCT CTA GCA	
		Spyo_hypp_P	6FAM- AAAATGTTGTGTTTTAGGCACTGGCGG-TAMRA	
	MipB	Spyo_mipB_F	CCA TAC GGT TAT AGT AAG GAG CCA AA	Ndiaye et al. (2018)
		Spyo_mipB_R	GGC TAT CAC ATC ACA GCA ACC	
		Spyo_mipB_P	6FAM- TCAGCGCCAGCTTCAATGGC- TAMRA	
Streptococcus pneumoniae	plyN	Pneumo_plyN_F	GCG ATA GCT TTC TCC AAG TGG	Ndiaye et al. (2018)
		Pneumo_plyN_R	TTA GCC AAC AAA TCG TTT ACC G	
		Pneumo_plyN_P	6FAM-CCC AGC AAT TCA AGT GTT CGC CGA-TAMRA	
	lyt A	Pneumo_lytA_F	CCT GTA GCC ATT TCG CCT GA	Ndiaye et al. (2018)
		Pneumo_lytA_R	GAC CGC TGG AGG AAG CAC A	
		Pneumo_lytA_P	6-FAM- AGA CGG CAA CTG GTA CTG GTT CGA CAA-TAMRA	
Tropheryma whipplei	WiSP family protein (WHI2)	T_whi2_F	TGA GGA TGT ATC TGT GTA TGG GAC A	Ndiaye et al. (2018)
		T_whi2_R	TCC TGT TAC AAG CAG TAC AAA ACA AA	
		T_whi2_P	6FAM- GAG AGA TGG GGT GCA GGA CAG GG-TAMRA	
	WiSP family protein (WHI3)	T_whi3_F	TTG TGT ATT TGG TAT TAG ATG AAA CAG	Ndiaye et al. (2018)
		T_whi3_R	CCC TAC AAT ATG AAA CAG CCT TTG	
		T_whi3_P	6FAM- GGG ATA GAG CAG GAG GTG TCT GTC TGG-TAMRA	

distribution of soap in the test village, concession chiefs, wives and older children received soap for their personal use, while smaller children were organised into groups of three individuals who shared the same soap. The soaps were first weighed before being given to the participants. After 7 days of use, each participant had to return the soap. The soaps were weighed again, and another soap was given to the participant. Thus, the soaps were renewed on a weekly basis. This process of soap delivery and recovery continued throughout the study.

Swabbing and treatment. Swabs were performed on the hands after moistening the swab with sterile physiological serum. The swabs were then immerged in an individual tube containing 600 μ L 1x phosphate buffered saline (PBS) manufactured by Oxoid Limited (Hampshire, England). Once saturated, the swabs were agitated and pressed against the edges of the tube to release the sample. Then, two 200-μL aliquots of the swab suspension were prepared, one for DNA extraction using the Cetyl Trimethyl Ammonium Bromide (CTAB) 2% method and one for stock.

DNA extraction. Approximately 200 μL of the swab bacterial suspension was used for DNA extraction following the CTAB 2% method (Bi et al., 1996). At the end of the extraction, the DNA solution was stored for 24 h in the refrigerator at a temperature of 8 °C, as DNA cannot deteriorate after 24 h of storage. After qPCR, the DNA was stored at −20 °C.

DNA amplification. Each reaction was performed at a final volume of 20 μL, containing 10 μL Takyon polymerase, 1 μL of each primer, 1 μL probe, 2 μL RNase-free water, and 5 μL DNA. A positive and negative control was included in each run. DNA extracted from the swabbing of a healthy person in Dakar was used as a negative control. The positive control consisted of a suspension made from the swabbing of a healthy person, to which bacterial cultures were added. The strains used as positive controls are available on the Rickettsia Unit Strains Collection (CSUR, WDCM 875) under the following accession numbers: S. pneumoniae CSURP5700, *S. aureus* CSURP2200 and *S. pyogenes* CSURP6897 (Ndiaye et al., 2018). For each species, approximately ten colonies were suspended in 200 μL PBS, and DNA was extracted (Ndiaye et al., 2018). The primers and probes used for pathogenic identification are listed in Table 1. The quality of the DNA handling and the extraction of the swab suspensions was verified by quantitative PCR for a housekeeping gene coding for β-actin, as previously described (Ndiaye et al., 2018; Mediannikov et al., 2010). Samples were considered to be positive if both specific qPCR reactions were positive using a cycle number at the threshold level for a log-based fluorescence (Ct) value <35. This number corresponds to the ability to reveal 10–20 copies of bacterial DNA (Mediannikov et al., 2010).

Statistical analysis. Infectious diseases were assessed by estimating the incidence rate. For each village, the annual incidence rate of a clinical manifestation was calculated as the ratio of the number of registered clinical cases divided by the number of person-times in the year. Monthly incidences were calculated by dividing the number of monthly cases recorded by the number of person-times in the month. The mean incidences of the years 2015 (the year before the intervention) and 2016 (the year of the intervention) were compared using the Fisher exact test to determine the overall impact of soap use on clinical events such as fever, cough, diarrhoea, and nasal discharge. The differences in incidence rates were analysed using Epi Info software, version 3.4.1 (Centers for Disease Control and Prevention, Atlanta, GA, USA). Nonparametric values were compared using a χ^2 test. Statistical significance was defined as $p < 0.05$.

Results

The study carried out 372,288 observations, including 174,021 in the control village and 198,267 in the test village. These observations were made on 638 participants: 238 in Dielmo and 400 in Ndiop. Participants aged 0–15 years were in the majority in both villages, with 51.25% (205/400) in the test village and 54.2% (129/238) in the control village (Figure 2). In the two villages, 87 concessions (50 in Dielmo and 37 in Ndiop) and 158 households (72 in Dielmo and 86 in Ndiop) were counted. The average number of people per household was 3.3 in the control village and 4.65 in the test village; thus, the number of people per household was significantly higher in the test village (χ^2 = 5.7, df = 1, p = 0.02).

A total of 2,707 (1,308 in Ndiop and 1,399 in Dielmo) clinical events was observed in both villages. Events related to respiratory infections were predominant. Households participating in the study received an average of three to four pieces of soap per week. Each household member used an average of 4 g of soap per day. Incidence rates were highest in the year prior to the intervention. Except for diarrhoea in the test village, there was a significant decrease in the annual incidence rate of the targeted clinical events in 2016 in both villages (Table 2). Participants aged <15 years who received soap presented fewer incidences of cough, nasal discharge and fever, and the difference versus the control was significant (Table 3). The difference versus the control was more significant with respiratory problems than with fevers. No significant differences were observed in participants aged ≥15 years in either village. Concerning diarrhoea, there was a significant increase in the annual incidence during the period of intervention in Ndiop.

A total of 649 swabs were collected during the study. The PCR β-actin was positive for 638 samples, for a prevalence of 98.30% (638/649). The DNA of *B. crocidurae*, *C. burnetii*, *S. pneumoniae*, *S. aureus*, and *S. pyogenes* was detected in 273 participants, representing a prevalence of 42.79% (273/638). *S. pneumoniae* was the predominant bacteria, with a prevalence of 31.81% (203/638), followed by *S. aureus* at 25.39% (162/638) and *S. pyogenes* at 1.88% (12/638). The prevalence of bacteria before intervention (T_0) was higher than that obtained during the intervention (Table 3). Before the intervention, the difference in the prevalence of *S. aureus* between the villages test and control was not significant. However, the differences concerning *S. aureus* and *S. pneumoniae* were significant during the period of intervention (Table 3). *S. pyogenes* was absent in Ndiop before the distribution of soap. During the first two quarters of the intervention, this bacteria was completely absent in the two villages; however, during the last quarter, a resurgence of *S. pyogenes* was noted in both villages, with a significantly higher prevalence in Dielmo (Table 4). During the period before the intervention, the prevalence of *S. aureus* on the palms of the hands was significantly higher in Dielmo (χ^2 = 9.24, df = 1, p = 0.002) and that of *S. pneumoniae* was also significant (χ^2 = 40.02, df = 1, p < 0.001); however, the difference between the prevalence of *S. pyogenes* in the test and control villages could not be verified because one of the values was <5 (Figure 3). The prevalence of *S.*

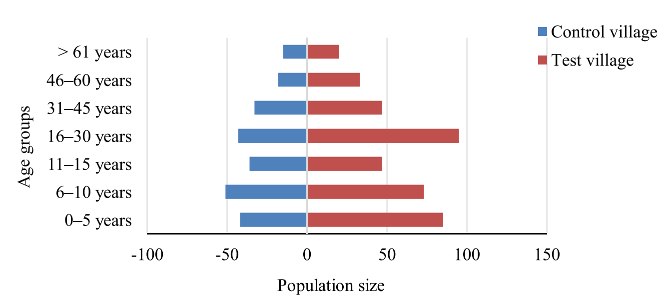

Figure 2. Age pyramid of test and control villages.

Table 2
Comparison of annual incidence rates for the years 2015 and 2016.

	Cough			Nasal discharge			Diarrhea (≥4 stool/day)			Fever		
	Annual incidence rate	*p* (95% CI)	Annual incidence rate	*p* (95% CI)	Annual incidence rate	*p* (95% CI)	Annual incidence rate	*p* (95% CI)	Annual incidence rate 2015	*p* (95% CI)	Annual incidence rate	*p* (95% CI) 2015
Years	2015	2016		2015	2016		2015	2016		2016		
Dielmo	14.84	12.51	$p = 0.005$	12.41	10.1	$p = 0.005$	3.4	1.7	14.84	12.51	$p = 0.005$	12.41
Ndiop	12.72	9.59	$p < 0.001$	10.12	8.16	$p = 0.003$	4.31	3.68	$p = 0.16$	10.4	7.0	$p < 0.001$

Table 3
Comparison of incidence rates.

	Participant aged <5 years			Participant aged <15 years			Participant aged ≥15 years		
	Annual incidence rate		*p* (95% CI)	Annual incidence rate		*p* (95% CI)	Annual incidence rate		*p* (95% CI)
	Test (n = 31,964)	Control (n = 24,004)		Test (n = 73,801)	Control (n = 68,079)		Test (n = 76,376)	Control (n = 81,938)	
Cough	1.08	2.52	$p < 0.001$	1.86	3.22	$p < 0.001$	0.94	0.82	$p = 0.18$
Nasal discharge	1.12	3.16	$p < 0.001$	0.89	1.45	$p < 0.001$	0.72	0.63	$p = 0.26$
Diarrhoea (≥4 stool/day)	0.87	0.87	$p = >0.99$	0.11	0.09	$p = 0.41$	0.41	0.25	$p = 0.004$
Fever	1.16	1.90	$p < 0.001$	0.91	1.31	$p = 0.004$	0.46	0.40	$p = 0.36$

n, number of observations in the year.

Table 4
Comparison of the prevalence of bacteria between villages test and control before and after the intervention.

	Bacteria	Control	Test	*p*
T_0	*S. pneumoniae*	49% (116/238)	21.75% (87/400)	$p < 0.001$
	S. pyogenes	5% (12/238)	0%	
	S. aureus	30% (71/238)	22.75% (91/400)	$p = 0.05829$
T_{01}	*S. pneumoniae*	5.5% (13/238)	1.75% (7/400)	$p = 0.01792$
	S. pyogenes	0%	0%	
	S. aureus	6.30% (15/238)	0.75% (3/400)	$p < 0.001$
T_{02}	*S. pneumoniae*	7.56% (18/238)	1.0% (4/400)	$p < 0.001$
	S. pyogenes	0%	0%	
	S. aureus	3.0% (7/238)	0.75% (3/400)	$p = 0.07$
T_{03}	*S. pneumoniae*	10.0% (23/238)	3.0% (12/400)	$p < 0.001$
	S. pyogenes	3.0% (7/238)	0.5% (2/400)	$p = 0.02915$
	S. aureus	10.0% (23/238)	3.0% (11/400)	$p < 0.001$

S. pneumoniae, *Streptococcus pneumonaie*; *S. pyogenes*, *Streptococcus pyogenes*; *S. aureus*, *Staphylococcus aureus*.

aureus in children aged <15 years before soap distribution (T_0) was significantly higher than the average prevalence during the study period ($\chi^2 = 4.51$, df = 1, $p = 0.04$); this outcome was also found in *S. pneumoniae* ($\chi^2 = 45.91$, df = 1, $p < 0.001$).

Before the intervention, the prevalence of *S. aureus* on the palms of the hands was significantly higher in Dielmo ($\chi^2 = 9.24$, df = 1, $p = 0.002$) and that of *S. pneumoniae* was also significant ($\chi^2 = 40.02$, df = 1, $p < 0.001$). *S. pyogenes* was absent in Ndiop before the distribution of soap (Figure 4). The prevalence of *S. aureus* in children aged <5 years before soap distribution (T0) was significantly higher than the average prevalence during the study period ($\chi^2 = 4.51$, df = 1, $p < 0.001$); this result was also found for *S. pneumoniae* ($\chi^2 = 45.91$, df = 1, $p < 0.001$). Among participants aged >15 years, the prevalence of different pathogens before soap distribution was significantly higher in Dielmo. Values were obtained for *S. aureus* ($\chi^2 = 14.96$, df = 1, $p < 0.001$) and *S. pneumoniae* ($\chi^2 = 9.997$, df = 1, $p < 0.001$). However, the significance could not be measured for *S. pyogenes* because no contamination was detected in Ndiop during this period (Table 5). The number of people infected with *S. aureus* during the intervention in Ndiop significantly decreased during the period of soap use. The number of people infected with *S. pneumoniae* also decreased, but the *p*-value could not be measured because the total contamination was <5. However, no contamination for *S. pyogenes* was detected before or after the distribution of soap in Ndiop (Table 5).

Most control strategies aimed at eliminating skin pathogens have targeted those pathogens responsible for diarrhoea in children or healthy persons. Swabs were taken either from the palms of the hands or from specific parts of the body (Table 6). Previous studies that have assessed the impact of soap on the occurrence of respiratory infections, fevers and diarrhoea have yielded mixed results. While the majority have agreed that handwashing with soap and water helps to reduce the incidence of these diseases, some studies have not been as successful (Table 7).

Discussion

This work, which was carried out in rural Senegal, presents the beneficial effects of soap use on the incidence of respiratory diseases and fevers. The population generally adhered well to the project, with a very high participation rate. The soap was correctly used by the participants for their hygiene.

In both villages, the incidence of fevers and respiratory diseases significantly decreased between 2015 and 2016. However, the number of people with diarrhoea did not significantly decrease. Diarrhoea is a disease with multiple causes (bacterial, parasitic, functional, etc.) and the use of soap may not be sufficient to reduce its incidence rate. The pathogens are usually transmitted through the faecal-oral route (Curtis et al., 2000) by contaminated drinking water or the consumption of contaminated food.

The use of soap in households was not controlled, and the participants were aware of the benefits and importance of soap in the fight against infectious diseases. However, unlike previous studies that have focused on the promotion of handwashing to fight nosocomial infectious diseases (Bodena et al., 2019; Oyapero and Oyapero, 2018; Jemal, 2018) in poor neighbourhoods (Luby

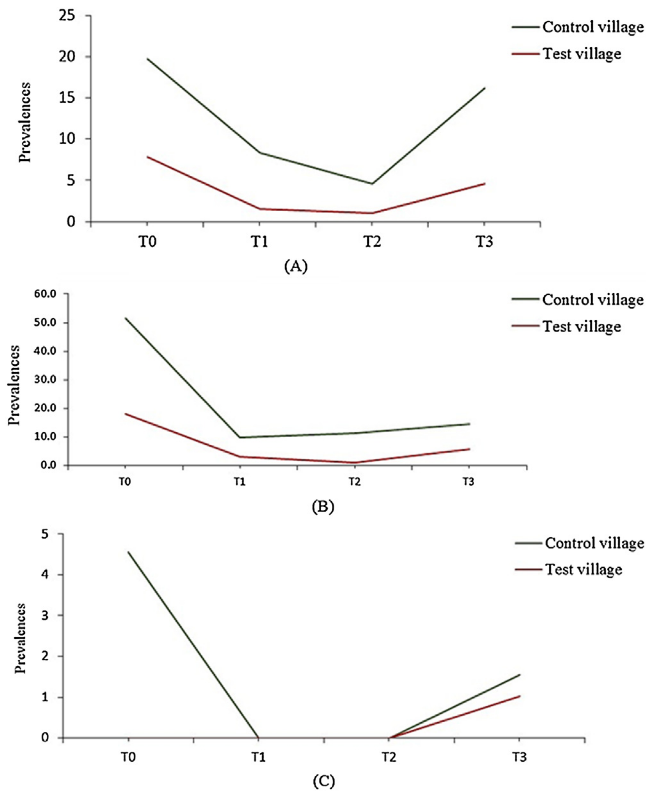

Figure 3. Evolution of bacterial prevalence on the palm of the hands of children aged <15 years.
(A) Evolution of the prevalence of *Staphylococcus aureus* in children aged <15 years. T_0 refers to before the distribution of soap, and T_1 to T_3 refer to periods during soap use.
(B) Evolution of the prevalence of *Streptococcus pneumoniae* and (C) that of *Streptococcus pyogenes*.

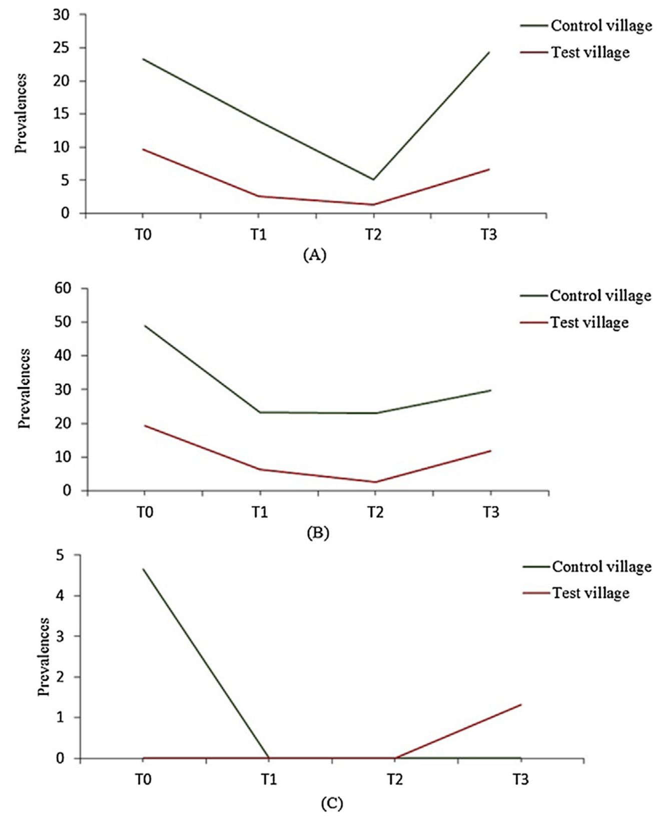

Figure 4. Evolution of bacterial prevalence on the palm of the hands of children aged <5 years.
(A) Evolution of the prevalence of *Staphylococcus aureus* in children aged <5 years. T_0 refers to before the distribution of soap, and T_1 to T_3 refer to periods during soap use.
(B) Evolution of the prevalence of *Streptococcus pneumoniae* and (C) that of *Streptococcus pyogenes.*

et al., 2005), the soap use in the current study was unsupervised. Despite this lack of control, the annual incidence rate of respiratory diseases and fevers significantly decreased among the participants aged <15 years residing in the test village. These results show that the participants took ownership of the study objectives and that soap use was beneficial for this segment of the population. The adoption of basic hygiene rules such as hand and body washing with soap has had a positive impact on the occurrence of fevers and respiratory infections (Luby et al., 2005; Rabie and Curtis, 2006; Pickering et al., 2019) (Table 6). It has also had a negative impact on the development and multiplication of pathogenic bacteria living on the skin (de Aceituno et al., 2015; Chen, 2015; Jensen et al., 2017; Pérez-Garza et al., 2017; Raza and Avan, 2013; Yu et al., 2018) (Table 5). Studies have shown that in some areas, ethnic, religious and social factors must be taken into account to ensure the success of such a strategy (Bulled et al., 2017). The current results suggest that it is mainly adults who are the problem. In this segment of the population, no significant differences were recorded for all targeted diseases. The largest decreases were recorded among children aged <5 years. This is a segment of the population that is generally under the care of their mothers. This could explain the approximate 2-point decrease in the incidence rate. During the year of intervention, no significant differences were recorded in the occurrence of diarrhoea.

The qPCRs performed on the swabs showed that bacteria may have caused the clinical manifestations recorded during the study period that were present on the palms of the hands. Other bacteria, such as *S. pneumoniae*, *S. pyogenes* and *S. aureus*, are involved in respiratory and skin infections, specifically *S. pyogenes* and *S. aureus*. This is why their presence on the palms of the hands is an important factor in the spread of respiratory infections in this area. The prevalence of *S. pneumoniae*, *S. pyogenes* and *S. aureus* was much lower in Ndiop. This result is probably due to the use of soap in the personal hygiene routines of the Ndiop population. It is suspected that the population was slackening, in terms of their compliance with the study rules outlined at the beginning of the study, in the third trimester. This situation could explain the presence of *S. pyogenes* in children aged <5 years in the third trimester after soap distribution began. Among the participants aged >15 years, a decrease in the prevalence of bacteria was also observed. Previous studies (Vishwanath et al., 2019; Ghimire et al., 2015) have shown that the palms of the hands are home to many pathogenic bacteria that could spread through skin contact. *S. pneumoniae*, *S. pyogenes* and *S. aureus* are involved in respiratory tract infections; in children, 15% of these respiratory tract infections are pneumonia (Liu et al., 2016).

Some limitations should be noted in this study, notably the impossibility of measuring the real effect of soap on the reduction of targeted pathologies. It would have been necessary to deprive the population of the control village of soap for their daily hygiene (which would be ethically unacceptable). The soap that was used was based on sodium hydroxide and it was not supplemented with an antibacterial agent.

The identification of these bacteria on the palms of the participants' hands and the significant reduction in their presence after soap use proves that promoting personal hygiene through simple and inexpensive actions such as handwashing and body washing with soap can substantially reduce the incidence of infectious diseases.

Table 5
Prevalence of bacteria on the palms of the hands in participants ≥15 years.

	T_0		T_1		T_2		T_3	
	Control	Test	Control	Test	Control	Test	Control	Test
S. aureus	31% (34/111)	12% (24/199)	4% (4/111)	0	1% (1/111)	0.5% (1/205)	2% (2/113)	1% (2/206)
S. pneumoniae	43% (48/111)	25% (50/199)	0	0.5% (1/202)	3% (3/111)	1% (2/205)	3% (4/113)	0.5% (1/206)
S. pyogenes	5% (6/111)	0	0	0	0	0	4% (5/113)	0

S. pneumoniae, *Streptococcus pneumonaie*; *S. pyogenes*, *Streptococcus pyogenes*; *S. aureus*, *Staphylococcus aureus*.

Table 6
Effect of interventions on pathogenic bacteria living on the human skin.

Study	Study population	Intervention component	Pathogens
Chen et al. (2015)	Neonates (preterm) Born <33 weeks' gestational age (mean age = 2 days)	Hand hygiene education	Invasive Candida infections
Raza and Avan (2013)	Neonates (ages not reported)	Clean care delivery kits (soap, spirit for clean cord care, sterile equipment)	Neonatal tetanus infection
Pérez-Garza et al. (2017)	Ages not reported	Hand decontaminated	*Escherichia coli* and *Enterococcus faecalis*
Jensen et al. (2017)	24.5 ± 3.9 years (mean ± SD)	Hand washing	*Escherichia coli* ATCC 11229
de Aceituno et al. (2015)	Ages not reported	Hands washing	*Escherichia coli* and *Enterococcus* spp.
Yu et al. (2018)	18–75 years	Four skin sites (right ankle, right medial forearm, right outer hand, right armpit)	Skin microbiota
Barker et al. (2017)	≥8 years	Hand hygiene	*Clostridium difficile*

Table 7
Effect of interventions on the occurrence of pathological episodes.

Study	Study population	Estimator	Diagnosed diseases	Difference vs control effect estimate (95% CI)	Reduction observed
Larson et al. (2004)	Children <5 years	Risk	Cough Nasal discharge Diarrhoea Fever	0.97 (0.79–1.18) 1.03 (0.81–1.32) 0.90 (0.54–1.50) 0.84 (0.63–1.12)	No statistically significant differences between intervention and control
Luby et al. (2005)	Children <15 years	Mean incidence	Cough Diarrhoea	−50% (−65% to −36%) −50% (−64% to −37%)	Statistically significant differences between intervention and control were observed
Rabie and Curtis (2006)	Ages not reported	Relative risk	Acute respiratory infection	16% (11%–21%)	Statistically significant differences between intervention and control were observed
Kim et al. (2018)	Adults	Incidence rate	Acute respiratory illness	13.0% (10.6%–15.9%)	Statistically significant differences between intervention and control were observed
Pickering et al. (2019)	18–24 months	Prevalence	Diarrhoea	27% in the control group SHINE trials (10% in the control group)	Reduction was observed in Bangladesh but not in Kenya or Zimbabwe
Slayton (2019)	<2 years	Rates per 100 person-visits	Diarrhoea	-	No statistically significant differences between intervention and control
Humphrey et al. (2019)	18 month	Prevalence ratio	Diarrhoea Acute respiratory infection	1.18 (0.87–1.61) 1.75 (0.62–4.90)	No statistically significant differences between intervention and control
Chard et al. (2019)	Pupils in schools	Risk	Diarrhoea Respiratory infections	0.80 (0.51–1.26) 1.08 (0.95–1.23)	Reduction was not observed

Conclusion

This study shows that a simple and inexpensive strategy based on hand and body washing with soap and water could significantly reduce the incidence of respiratory infections, febrile diseases and pathogenic bacteria development. The efficacy of handwashing with soap on diarrhoea was not demonstrated in this study. Additional measures – such as good public awareness, care for malnourished children, increased drinking water supplies, etc. – must be combined with handwashing to reduce incidence rates. Based on these results, it appears necessary to undertake a study evaluating the sustainability of behavioural change in handwashing promotion, and it is also important to assess cost-effectiveness. Finally, Senegal's health authorities should take these results into account and promote handwashing with soap and water in their infectious disease prevention programs.

Conflict of interest

No conflict of interest was declared by the authors.

Ethical approval

This study was authorised by the National Ethics Committee for Health Research (CNERS), No. 00053/Ministry of Health and Social Action (MSAS)/ Direction of Planning, Research and Statistics (DPRS)/CNERS.

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