Interactions among Urogenital, Intestinal, Skin, and Oral Infections in Pregnant and Lactating Panamanian Ngäbe Women: A Neglected Public Health Challenge

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Abstract. Interrelationships among bacteria, protozoa, helminths, and ectoparasites were explored in a cross-sectional survey of 213 pregnant and 99 lactating indigenous women. Prevalences in pregnancy and lactation, respectively, were: vaginitis (89.2%; 46.8%), vaginal trichomoniasis (75.3%; 91.1%), bacterial vaginosis (BV; 60.6%; 63.3%), hookworm (56.6%; 47.8%), asymptomatic bacteriuria/urinary tract infection (AB/UTI; 56.2%; 36.2%), cervicitis (33.3%; 6.3%), vaginal yeast (24.9%; 11.4%), *Ascaris* (32.5%; 17.4%), vaginal diplococci (20.4%; 31.6%), caries (19.7%; 18.2%), scabies (17.4%; 8.1%), and *Trichuris* (12.5%; 8.7%). Multiple regressions revealed positive associations during pregnancy (trichomoniasis and AB/UTI; diplococci and *Ascaris*) and lactation (yeast and scabies). Negative associations were detected in pregnancy (BV and trichomoniasis; hookworm and diplococci) and lactation (BV and yeast). Vaginal *Lactobacillus* reduced odds of diplococci in pregnancy and lactation, but increased *Ascaris* eggs per gram (epg) and odds of trichomoniasis in pregnancy and yeast in lactation. These associations raised a concern that treatment of one condition may increase the risk of another.

INTRODUCTION

In remote, extremely impoverished communities in developing countries, there is little scientific information on the range of infections during pregnancy and lactation. In addition to malaria, toxoplasmosis, tuberculosis, human immunodeficiency virus (HIV), and other viral infections, pregnant women may be infected with a wide variety of bacterial, fungal, and parasitic infections including bacterial vaginosis (BV), vaginal candidiasis, trichomoniasis,^{1,2} asymptomatic bacteriuria/urinary tract infections (AB/UTI) most often caused by *Escherichia coli*,³ intestinal nematode infections (*Trichuris*, *Ascaris*, and hookworm),⁴ dental and periodontal disease,^{5,6} and skin infections (scabies, bacterial and fungal infections), which are common under conditions of poor hygiene, poor nutritional status, and overcrowding.^{7,8}

Compared with pregnancy, BV is reportedly more prevalent during lactation,⁹ whereas periodontal disease (but not periodontitis) and associated microflora are less common in lactating women.¹⁰ Information on other infections during lactation is largely limited to those transmitted during breast-feeding such as HIV^{11,12} and cytomegalovirus,¹³ mammary infections produced by *Staphylococcus* spp. and *Candida* spp.,^{14,15} UTI associated with institutional vaginal delivery,¹⁶ and puerperal sepsis mostly associated with vaginal infections.¹⁷

In conditions of poverty where access to health care is limited, coinfection is likely common. Yet research on coinfections during pregnancy and lactation has primarily focused on the associations of HIV and malaria with other pathogens including intestinal nematodes^{18–20} or associations with other infections of the intestine²¹ or the reproductive tract.^{22–24} For example, intestinal nematode infections are often positively associated with one another²⁵ and BV is often positively associated with other common vaginal pathogens.^{26–28} Much less is understood about interactions among vaginal, intestinal, urinary

tract, skin, and oral infections. Nematode parasites have been reported to influence the progression of microparasite infections.^{29,30} For example, pregnant women infected with *Ascaris* had increased risk of malaria in both Gabon¹⁹ and Ethiopia,³¹ but those with higher intensity of *Ascaris* had reduced risk of malaria perhaps as a result of the strong anti-inflammatory response induced by *Ascaris*.^{32,33} Given that host defense mechanisms (both innate and adaptive) are increasingly understood to be sensitive to the presence of coexisting pathogens through the systemic response that they elicit,^{33–36} the impact of one infection is likely to extend to other regions of the body.

The objectives of this observational cross-sectional survey were 1) to characterize the range of infections (oral, skin, respiratory, urinary, vaginal, and intestinal) during pregnancy and the first 6 months of lactation in Ngäbe women using a clinical exam and locally available laboratory tests; 2) to determine whether prevalence of infections differed among trimesters or between pregnancy and lactation; 3) to explore associations between the presence or severity of one pathogen and the community of other pathogens; and 4) to determine whether such associations were similar in pregnant and lactating women. The study was conducted in a remote, rural region of western Panamá within the *Comarca*, an administrative area inhabited by the Ngäbe-Buglé, the largest indigenous population of Panamá, and comprising a territory between Bocas del Toro, Chiriquí, and Veraguas Provinces where over 60% of households live in extreme poverty.³⁷ Routine prenatal care is limited to basic clinical measurements (vital signs, fundal height, fetal cardiac rate, fetal movements, and presence of edema) performed by an auxiliary nurse at the local health center. Depending on the distance from the nearest hospital, a physician may provide more detailed pregnancy follow-up from once a week to a few times a year. Most deliveries occur at home and are attended by traditional midwives or relatives.

MATERIALS AND METHODS

Ethical considerations. Ethical approval was obtained from McGill University in Canada, the Gorgas Memorial Institute

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Ethics Board in Panamá, the Panamanian Ministry of Health, provincial and local health authorities, and indigenous authorities. All mothers were informed of their right to refuse to participate and to withdraw at any time. Participants signed a consent form. Those who were not able to write marked the consent form with their fingerprint, in the presence of a witness who signed the consent form. Participants received no direct financial compensation but they received a complete medical evaluation and verbal referral when follow-up was needed.

Study design. An observational cross-sectional survey was conducted with indigenous Ngäbe pregnant women and lactating women with infants < 6 months old. For the sample of pregnant women, the three inclusion criteria were 1) pregnancy as detected either by a positive pregnancy test for women with amenorrhea > 5 weeks or a physical exam by staff at the health centers, community health workers, or traditional midwives; 2) homes within a 2-hour walk of one of the 14 health centers; and 3) mothers well enough to walk to the health centers. The only exclusion criteria were twin pregnancy or abnormal pregnancy (ectopic pregnancy or hydatidiform mole). Of the 217 pregnant women approached between August and October 2010, 214 agreed to participate. One participant was excluded because of abnormal pregnancy (hydatidiform mole), resulting in a participant population of 213. No cases of ectopic pregnancy were found.

For the sample of lactating women, the three inclusion criteria were 1) women who had a singleton delivery within the last 6 months and whose babies were living with them; 2) homes within a 2-hour walk of one of the 14 health centers; and 3) mothers well enough to walk to the health centers. Of the 99 women approached, all agreed to participate. Only one woman recruited in the sample of lactating women had also been recruited into the pregnancy sample.

A clinical exam was used to diagnose skin (scabies, impetigo, and dermatomycosis), oral (caries, gingivitis), respiratory (cold, bronchitis), and vaginal tract (vaginitis, cervicitis) infections. Laboratory examinations of urine and stool samples and vaginal smears were used to diagnose AB/UTI, BV including *Bacteroides/Gardnerella*, *Mobiluncus* and *Lactobacillus*, vaginal trichomoniasis, vaginal diplococcal infection, vaginal yeast infection, intestinal parasites (*Ascaris*, *Trichuris*, hookworm, *Giardia*, and *Entamoeba coli*). Comparisons were made among trimesters and between pregnancy and lactation and associations among organisms were explored.

Participant recruitment. We identified 14 health centers located in the districts of Besikó (Soloy, Emplanada de Chorchá and Quebrada de Hacha), Nole Duima (Hato Chamí, Oma, Lajero and Quebrada Guabo), Müna (Alto Caballero and Chichica), and Mironó (Hato July, Kuerima, Hato Pilón, Quebrada de Loro and Hato Corotú) with year-round road access to the regional hospital in San Felix. Meetings were held at the health centers with all staff and associated community health workers and traditional midwives to explain the study and to provide training on pregnancy follow-up and recognition of complicated pregnancies. The staff, community health workers, and midwives then contacted all pregnant women and lactating mothers with infants up to 6 months old within their respective catchment areas, explained the research project and invited them to attend an information session at the local health center where objectives, procedures, and confidentiality were explained and fully informed consent was obtained. Pregnant women staying at the tempo-

rary home near the San Felix hospital were also invited to participate. We estimated that about 90% of eligible women in the catchment area met with the research team during our visit to their local health center.

Medical history, clinical exam, and health questionnaire.

All women were seen at their local health center or at the temporary home where they received a complete clinical evaluation. This included a questionnaire on obstetric history (age, parity, and gestational age [GA] calculated according to reported date of last menstrual period) and a questionnaire on symptoms of infection. The physical examination included anthropometry (measured with SECA® balances [SECA Deutschland, Hamburg, Germany] with height rod, available at the health centers) and blood pressure (BP), measured with an Omron HEM-790IT® automatic BP monitor (OMRON Healthcare Europe B.V., Hoofddorp, The Netherlands) while seated and after a minimum 15 minutes resting period. Body mass index (BMI) measurements for pregnant women were calculated ($[\text{maternal} - \text{fetal weight in kilograms}] / [\text{maternal height in meter}]^2$) where fetal weight was estimated using Johnson's formula ($\text{fetal weight [g]} = \text{symphysis fundal height [cm]} - 13 \times 155$),³⁸ and women were classified as underweight, normal, or overweight for GA compared against Pan American Health Organization (PAHO) standards for GA.³⁹ BMI for lactating women was classified as for the general population: underweight (BMI < 19.8 kg/m²), normal (19.8–26.0 kg/m²), overweight (26.1–29.0 kg/m²), or obese (> 29 kg/m²).⁴⁰ Presence of hypertension was defined as blood pressure > 140/90.⁴¹

A medical exam conducted by a physician was used to diagnose oral, respiratory, skin, and vaginal infections. Oral infections were detected through direct visual exam for presence or absence of dental cavities (caries) or inflammation of the gum (gingivitis).⁴² Symptoms of mild upper respiratory tract infection (sneezing, runny or stuffy nose, sore throat), lower respiratory tract symptoms (cough associated with auscultator findings such as bronchial rales or crepitus),⁴³ and complicated upper UTI (fever, costovertebral angle tenderness, flank pain, nausea, or vomiting)³ were recorded as present or absent. Among skin infections, lesions typical of scabies (small erythematous symmetrical papulovesicular rash),⁴⁴ superficial dermatomycosis (pruritic erythematous macules with superficial scales), and impetigo (confluent macules with honey-colored crust)⁴⁵ were recorded as present or absent. A small part of the room in the health center was temporarily screened off for the genital exam. After an explanation about the genital exam, women were asked to lie in the gynecological position and a disposable speculum was placed into the vagina. Direct visual exam allowed the clinical diagnosis of vaginitis based on vaginal discharge and/or irritation of the vaginal wall, and of cervicitis based on erythematous, edematous, or easily friable appearance.⁴⁶ Both vaginitis and cervicitis severity were graded as mild, moderate, or severe depending on the intensity and extent of the symptoms.^{47,48}

Vaginal smears. During the genital examination, a sterile swab was used to take a sample of vaginal secretion or discharge from all participants. The swab was placed in a tube with 1 mL sterile saline solution, immediately refrigerated and kept on ice during transport to the laboratory. Direct microscopic examination of wet mounts was done by an experienced laboratory technician for mobile forms of *Trichomonas vaginalis* (sensitivity of 60%)⁴⁹ and budding yeast cells and/or hyphae of

vaginal yeast infection (sensitivity 65–85%).⁴⁸ Severity of vaginal yeast was scored according to the number per high-power field (hpf) as 0 (absent), 1+ (1/hpf), 2+ (2/hpf), 3+ (3/hpf), or 4+ (4 or more/hpf).

Vaginal smears were gram-stained to detect bacterial and protozoan pathogens. Quantity of *Lactobacillus* spp. and severity of *Bacteroides* spp./*Gardnerella* spp. and *Mobiluncus* spp. were scored as 0 (absent), 1+ (1–5/hpf), 2+ (6–10/hpf), 3+ (11–20/hpf), and 4+ (> 20/hpf) by an experienced laboratory technician and confirmed by a trained clinician. Diagnosis of BV was based on the Nugent score, calculated according to the methods described by Nugent and others⁵⁰: *Bacteroides*/*Gardnerella* score + (4 – *Lactobacillus* score) + (*Mobiluncus* score/2). A Nugent score of 0–3 reflected normal vaginal microflora, 4–6 corresponded to intermediate vaginal microflora, and 7–10 was diagnostic for BV, with a sensitivity of 97%.⁵¹ Severity of *Trichomonas* was scored, according to the number of organisms/hpf on the gram stain, as 0 (absent), 1+ (1/hpf), 2+ (2/hpf), 3+ (3/hpf), or 4+ (4 or more/hpf). Results from the gram-stained slides for trichomoniasis were used for statistical analysis given the higher estimates of prevalence detected in the gram stain compared with our wet mounts. Given the technical difficulty in distinguishing extracellular diplococci from similar pathologic bacteria, presence and severity of diplococcal infection was scored according to the number of leukocytes/hpf that contained diplococci as 0 (absent), 1+ (1/hpf), 2+ (2/hpf), 3+ (3/hpf), or 4+ (4 or more/hpf). No culture or molecular diagnostic techniques were available in this research setting.

As part of the routine assessment of pregnant women in the San Felix Hospital coverage area, results from the Venereal Disease Research Laboratory (VDRL) test (indicator of syphilis) using the unheated serum reagin antibody test from Wiener[®] laboratories (Rosario, Argentina) were also available for 172 pregnant participants. Also, the provincial HIV program manages an active screening of pregnant women across the *Comarca* using Advanced Quality Rapid Anti-HIV (1&2) test (InTec PRODUCTS, INC., Xiamen, China), a rapid immunochromatographic assay for the qualitative detection of antibodies to HIV in whole blood, serum, or plasma.

Urine samples. Urine samples were analyzed by an experienced laboratory technician using dipstick URISCAN[®] strips (YD Diagnostics, Kyunggi-Do, Korea) on a Miditron-M semiautomated reflectance photometer (Roche Diagnostics GmbH, Mannheim, Germany) with reagents for semiquantitative measurement of UTI (leukocyte esterase, nitrites, hemoglobin [Hb], and urinary pH), proteinuria as an indicator of renal pathology or preeclampsia, and glucose as an indicator of gestational diabetes. In addition, fresh urine was centrifuged at 3,000 rpm for 5 minutes, the supernatant was decanted and the sediment resuspended in the remaining liquid. A drop of the sediment was examined using low- and high-power magnification from which leukocytes, bacteria, and red blood cells were recorded and their co-occurrence with mucus and epithelial cells were noted as controls for vaginal contamination. Presence of AB/UTI was diagnosed based on bacteria ≥ 1 /hpf in microscopic examination of urine (sensitivity of 46–58%, specificity of 89–94%) combined with either leukocytes ≥ 5 /hpf (sensitivity alone of 90–96%, specificity of 47–50%) or red blood cells ≥ 5 /hpf (sensitivity alone of 18–44%, specificity of 88–89%), or URISCAN results with leukocyte esterase $\geq 2+$ on a scale

of 0–3+ (sensitivity alone of 72–97%, specificity 41–86%), presence of nitrites (sensitivity alone 19–48%, specificity 92–100%), or Hb $\geq 1+$ on a scale of 0–4+ (sensitivity of 68–92%, specificity of 42–46%).^{52,53} AB/UTI was ruled out in samples when high amounts of mucus and/or epithelial cells were observed under the microscope. Urine culture was not possible at the study location. Complicated UTI was ruled out based on absence of symptoms.⁵⁴

Stool samples. All stool samples were examined using a direct smear for protozoan infections by an experienced laboratory technician trained in distinguishing *Entamoeba coli* from *Entamoeba histolytica*. When a sample of sufficient volume and appropriate consistency was available, nematode intensity (eggs per gram [epg]) was recorded using Kato-Katz⁵⁵ and FLOTAC⁵⁶ techniques. A positive result from any of the three methods was used to calculate the prevalence of nematode infections, and FLOTAC results were used to determine the epg, given its higher sensitivity for *Trichuris* and hookworm when compared with Kato-Katz technique.⁵⁷

Data analysis. All data were analyzed using STATA 10 (StataCorp LP, College Station, TX). Results are presented as raw mean $\pm$ standard error of the mean (SEM) or prevalence (%). The level of significance was set at $P < 0.05$ unless otherwise indicated.

Prevalence of infection was compared among trimesters and between pregnancy and lactation using χ^2 tests. The Kruskal–Wallis nonparametric analysis of variance was used to determine if the severity of symptoms of BV differed by trimester or between pregnant and lactating women.

Analyses of coinfections were performed first using χ^2 tests to determine whether infections in pregnant women or in lactating women occurred together more or less frequently than expected by chance. In cases where expected values were < 5 , the Fisher's exact test was used. Given that an average of 10 tests were conducted for each infection within pregnancy and within lactation samples, we applied the Bonferroni correction to obtain a more conservative critical P value of 0.005 but have also reported actual P values for all analyses with $P < 0.05$. In addition, multiple regression analyses were used to explore the interrelationships among infections, while controlling for GA in models for pregnant women or weeks after delivery in models for lactating women. Models were generated only for those infections where laboratory analyses revealed a prevalence $> 10\%$ in at least one trimester or during lactation, and where the infection was considered pathogenic. Multiple logistic regression was used to generate a model for the presence of AB/UTI. Multiple ordered logistic regression was used to generate models for the severity of BV, vaginal trichomoniasis, vaginal yeast, and vaginal diplococcal infection. Given that the Nugent score incorporates information on two sets of pathogens (*Bacteroides*/*Gardnerella* and *Mobiluncus*) and one competitive bacteria (*Lactobacillus*), models were generated using either the Nugent score for BV or the individual bacteria in an effort to better understand interrelationships among the vaginal microflora. Stepwise regression was used to generate models of *Ascaris*, *Trichuris*, and hookworm epg (square root transformed [sqrt]).

In all cases, the independent variables included in each multiple regression model are listed as a footnote to the table. The ratio of number of factors to sample size was < 0.10 , indicating that logistic regression was robust to the effect of shared risk factors.⁵⁸ In all models, collinearity

among variables in each model was tested to confirm that the variance inflation factor (VIF) was < 2.5 . Final models report only those variables that entered with a $P < 0.15$.

RESULTS

Study response rate. Data from clinical exams and vaginal wet mounts were available for all women. Cervicovaginal exams and vaginal smears were done on 211 pregnant women and 79 lactating women that did not have vaginal bleeding at the time of the exam. Urine samples were collected from 208 pregnant and 79 lactating women. Of the stool samples received (120 pregnant and 23 lactating women), Kato-Katz technique was used on 105 pregnant and 17 lactating women, and FLOTAC was used on 74 pregnant and 9 lactating women.

Characteristics of study participants. Pregnant women. Pregnant women were 24 ± 0.5 years old (13–44 years), 11.3% were in the first trimester, 37.6% in the second trimester, and 51.2% in the third trimester. Among the women, 29.1% were adolescents (≤ 19 year) and 13.1% were ≥ 35 year; 28.1% were primiparous and 32% were in their fifth or more pregnancy. Based on BMI corrected for GA, 9.8% were underweight and 23% were overweight. Mean systolic BP was 103 ± 11 mmHg and mean diastolic BP was 62 ± 9 mmHg; none were positive for hypertension but protein was found in 11 urine samples (trace in 5 samples, 1+ in 6 urine samples). None of the pregnant women were positive for gestational diabetes based on presence of glucosuria.

Lactating mothers. Mothers of infants < 6 months old were 25.1 ± 0.7 years old (14–42 years). All were breast-feeding and only 15% had started giving complementary food to their babies. For 20.2% of women, this was their first child; 25.2% of mothers had five or more children. Based on BMI, 24.4% were overweight and an additional 15.1% were obese; only one woman was underweight. Mean systolic BP was 109.4 ± 1.4 mmHg and mean diastolic BP was 68.4 ± 1.1 mmHg.

Prevalence and severity of clinically identified infections. Clinical examination revealed respiratory, skin, oral, and vaginal infections (Table 1). The prevalence of respiratory infections including both upper respiratory tract infection and bronchitis was low (6.1% in pregnancy, 5% in lactating women), as was the prevalence of dermatomycosis, impetigo, and gingivitis. Scabies was more prevalent in pregnant (17.4%) compared with lactating women (8.1%) (Table 1). Caries was equally

prevalent during pregnancy (19.7%) and lactation (18.2%). Although few women complained of genital discomfort, direct visualization of the vaginal tract and cervix allowed the detection of different degrees and characteristics of vaginal discharge (thick, foaming, mucoid adherent, or chunky) or cervicitis in 92% pregnant women and 50% lactating women. Both vaginitis and cervicitis were more prevalent in pregnant compared with lactating women (Table 1) and of greater severity (data not shown). Gingivitis was only detected in the third trimester (8.2%), but prevalence of all other infections was similar among trimesters (data not shown).

Prevalence and severity of laboratory diagnosed infections. Laboratory analyses identified a total of 12 bacteria, protozoa, and helminths (Table 2). Participants lived in a non-endemic area for malaria, and no HIV-positive women were identified.

Urinary tract infections. The prevalence of AB/UTI was higher in pregnant (56.2%) than in lactating (36.2%) women (Table 2); no symptoms of complicated UTI were reported by any of the participants. AB/UTI prevalence did not differ among trimesters (data not shown).

Vaginal infections. Among the vaginal infections, 97.2% of pregnant women and 97.4% of lactating women were positive for at least one of BV, vaginal trichomoniasis, yeast, and diplococcal infection. No differences in severity of the vaginal microflora were detected among trimesters (data not shown).

BV diagnosed using the Nugent score was of similar prevalence in pregnancy and lactation (60.6 and 63.3%, respectively) (Table 2). Commensal *Lactobacillus* was more common, and numbers were higher in pregnant compared with lactating women whereas severity of *Mobiluncus* was higher in lactating women; the severity of *Bacteroides/Gardnerella* did not differ between pregnant and lactating women (Table 2, Figure 1). Vaginal trichomoniasis was more prevalent in lactating women (91.1%) than in pregnant women (75.3%) and of greater severity (Figure 1). Diplococcal infection was also more prevalent during lactation (31.6% versus 20.4% during pregnancy), but severity was higher among pregnant women (Figure 1). On the other hand, vaginal yeast infection was more frequent (24.9%) and more severe (Figure 1) in pregnant than in lactating women (11.4%).

TABLE 1

Comparison of the prevalence of clinically diagnosed infections between pregnant ($N = 213$) and lactating ($N = 99$) Ngäbe women from western Panamá*

	Prevalence (%)		$P > \chi^2$
	Pregnancy	Lactation	
Cold, sinusitis	3.7	3.0	ns
Bronchitis	4.0	2.0	ns
Scabies	17.4	8.1	0.030
Dermatomycosis	1.8	3.0	ns
Impetigo	1.8	5.0	ns
Caries	19.7	18.2	ns
Gingivitis	4.2	5.0	ns
Vaginitis†	89.2	46.8	< 0.0001
Cervicitis†	33.3	6.3	< 0.0001

ns = nonsignificant ($P > 0.05$).

* χ^2 or Fisher's exact test.

† $N = 79$ lactating women.

TABLE 2

Comparison of the prevalence of laboratory-diagnosed infections between pregnant and lactating Ngäbe women from western Panamá*

	Pregnancy		Lactation		$P > \chi^2$
	n	Prevalence (%)	n	Prevalence (%)	
AB/UTI	208	56.2	94	36.2	0.001
Bacterial vaginosis†	211	60.6	79	63.3	ns
<i>Lactobacillus</i> ‡	211	53.5	79	26.6	< 0.0001
<i>Bacteroides/Gardnerella</i>	211	93.8	79	97.5	ns
<i>Mobiluncus</i>	211	82.4	79	87.3	ns
Vaginal trichomoniasis	211	75.3	79	91.1	0.003
Vaginal yeast	213	24.9	79	11.4	0.012
Vaginal diplococcal infection	211	20.4	79	31.6	0.044
<i>Ascaris lumbricoides</i>	120	32.5	23	17.4	ns
Hookworm	120	56.6	23	47.8	ns
<i>Trichuris trichiura</i>	120	12.5	23	8.7	ns
<i>Entamoeba coli</i> ‡	120	2.5	23	4.3	ns
<i>Giardia</i> spp.	120	10.8	23	4.3	ns

AB/UTI = asymptomatic bacteriuria/urinary tract infection; ns = nonsignificant ($P > 0.05$).

* χ^2 test or Fisher's exact test.

†Diagnosed on the basis of the Nugent score.

‡Nonpathogenic.

FIGURE 1. Relative quantity (scored as 1+, 2+, 3+, or 4+) of vaginal organisms in pregnant (A) and lactating (B) Ngäbe women from western Panamá who were positive for *Lactobacillus*, *Bacteroides/Gardnerella*, *Mobiluncus*, trichomoniasis, yeast, and diplococcal infection. * Indicates higher severity in pregnancy or lactation.

Among the 92% (196) of pregnant and 50% (40) of lactating women clinically diagnosed with cervico-vaginitis, only 5 (2.6%) pregnant women and 1 (2.0%) lactating woman had no pathological microorganisms. Among those without clinical signs of vaginitis or cervicitis, all 17 pregnant women and all but one lactating woman were positive for at least one of the vaginal pathogens.

VDRL tests for syphilis were reactive in 5 (2.9%) of the 172 pregnant women tested (2 at 1:1 dilution and 1 each at 1:4, 1:8, and 1:32 dilutions). Four of the VDRL reactive women had clinical vaginitis and the fifth had dermal lesions; all mentioned partner promiscuity. Even though no confirmatory tests were available, local physicians treated these five women for syphilis.

Intestinal infections. None of the women mentioned gastrointestinal symptoms, but laboratory analyses revealed that 67.5% pregnant and 52.2% lactating women were infected with at least one intestinal pathogen (*Ascaris*, *Trichuris*, hookworm, and *Giardia*) (Table 2). Prevalence of *Ascaris* and *Trichuris* did not differ among trimesters or between pregnant and lactating women (Table 2), but hookworm prevalence was higher in the second and third trimesters (58% and 62%,

TABLE 3

Co-occurrence of infections during pregnancy and lactation in Ngäbe women from western Panamá*

	Observed % vs. Expected %	P
Pregnancy		
<i>Bacteroides/Gardnerella</i> + <i>Mobiluncus</i>	80.1 > 77.3	< 0.0001
<i>Lactobacillus</i> + <i>Bacteroides/Gardnerella</i>	47.7 < 50.2	0.003
<i>Lactobacillus</i> + <i>Mobiluncus</i>	40.7 < 44.1	0.009
BV + vaginal diplococcal infection	19.4 > 12.3	< 0.0001
BV + vaginal trichomoniasis	39.3 < 45.6	< 0.0001
<i>Lactobacillus</i> + vaginal diplococcal infection	2.8 < 15.3	< 0.0001
<i>Lactobacillus</i> + vaginal trichomoniasis	47.4 > 40.2	< 0.0001
<i>Bacteroides/Gardnerella</i> + vaginal trichomoniasis	69.2 < 70.6	0.041
<i>Mobiluncus</i> + vaginal diplococcal infection	19.4 > 16.8	0.012
Hookworm + <i>Trichuris</i>	11.7 > 7.1	0.002
Hookworm + <i>Ascaris</i>	23.3 > 18.4	0.020
Hookworm + vaginal diplococcal infection	6.7 < 11.4	0.011
Lactation		
<i>Lactobacillus</i> + vaginal diplococcal infection	0 < 8.4	< 0.0001
BV + vaginal yeast infection	2.5 < 7.2	0.010
<i>Lactobacillus</i> + vaginal yeast infection	7.6 > 3.0	0.009
Scabies + impetigo	3.0 > 0.4	0.003
Gingivitis + dermatomycosis	2.0 > 0.1	0.006
<i>Ascaris</i> + vaginal diplococcal infection	18.7 > 4.3	0.007

BV = bacterial vaginosis.

*Significance set at $P < 0.005$ based on Bonferroni correction for multiple comparisons.

respectively) compared with the first trimester (18%) ($P = 0.024$). *Giardia* spp. was detected in 10.8% pregnant and 4.3% lactating women. Nonpathogenic *Entamoeba coli* was detected in 2.5% pregnant and 4.3% lactating women.

Concurrent infections. Based on bivariate analyses and using a stringent cutoff of $P < 0.005$, several pathogens occurred together more frequently than expected based on their prevalence in the population whereas others co-occurred less than expected. During pregnancy (Table 3), the BV pathogens *Bacteroides/Gardnerella* and *Mobiluncus* co-occurred more than expected whereas *Bacteroides/Gardnerella* was less common in the mothers with the competitive *Lactobacillus*. Mothers diagnosed with BV by the Nugent score were more likely to have vaginal diplococcal infection but less likely to have vaginal trichomoniasis. *Lactobacillus* and diplococcal infection co-occurred less frequently than expected whereas *Lactobacillus* co-occurred more than expected with trichomoniasis. Finally, hookworm co-occurred with *Trichuris* more than expected by chance.

During lactation (Table 3), *Lactobacillus* co-occurred with diplococcal infection less than expected, whereas scabies and impetigo co-occurred more frequently than expected.

Presence of AB/UTI. The odds of AB/UTI were higher in mothers with more severe vaginal trichomoniasis (odds ratio [OR] = 1.33, 95% confidence interval [CI] = 1.03–1.80) and with more severe BV based on the Nugent score (OR = 2.03, 95% CI = 1.01–3.74) (model not shown). When the Nugent score was replaced with data on individual bacteria (Table 4), the likelihood of AB/UTI was also higher in mothers with more severe vaginal trichomoniasis (OR = 1.38, 95% CI = 1.04–1.84), and the quantity of commensal lactobacilli was negatively associated with AB/UTI (OR = 0.78, 95% CI = 0.63–0.95). During lactation (Table 4), the odds of AB/UTI

TABLE 4

Final multiple logistic regression model for AB/UTI in pregnant and lactating Ngäbe women from western Panamá*

	OR ± SE	95% CI	P
Pregnancy†			
Vaginal trichomoniasis, severity‡	1.38 ± 0.20	1.04, 1.84	0.025
<i>Lactobacillus</i> , quantity§	0.78 ± 0.08	0.63, 0.95	0.017
Lactation			
<i>Mobiluncus</i> , severity§	1.72 ± 0.43	1.05, 2.81	0.029

AB/UTI = asymptomatic bacteriuria/urinary tract infection; CI = confidence interval; hpf = high-power field; SE = standard error; OR = odds ratio; VIF = variance inflation factor.
 *AB/UTI coded as 0 = absent; 1 = present.

†Variables entered in model: gestational age; presence of caries and scabies; quantity of *Lactobacillus*; severity of *Bacteroides/Gardnerella*, *Mobiluncus*, vaginal trichomoniasis, yeast, and diplococcal infection. Overall model $P = 0.017$; $N = 207$; VIF = 1.92.

‡Coded as 0 = absent; 1+ = 1/hpf; 2+ = 2/hpf; 3+ = 3/hpf; 4+ = 4 or more/hpf.

§Coded as 0 = absent; 1+ = 1–5/hpf; 2+ = 6–10/hpf; 3+ = 11–20/hpf; 4+ = > 20/hpf.

||Variables entered in model: weeks after delivery; presence of caries and scabies; quantity of *Lactobacillus*; severity of *Bacteroides/Gardnerella*, *Mobiluncus*, vaginal trichomoniasis, yeast, and diplococcal infection. Overall model: $P = 0.022$; $N = 74$; VIF = 1.00.

were higher in women with higher severity of *Mobiluncus* (OR = 1.72, 95% CI = 1.05–2.81).

Severity of BV. Among pregnant women, the odds of more severe BV were lower in pregnant women with more severe vaginal trichomoniasis (OR = 0.54, 95% CI = 0.44–0.68), but higher in mothers with more severe diplococcal infection (OR = 1.80, 95% CI = 1.43–2.25) (Table 5). During lactation (Table 6), the odds of more severe BV were lower in mothers with more severe vaginal yeast infection (OR = 0.30, 95% CI = 0.10–0.87).

TABLE 5

Multiple ordered logistic regression models for severity of laboratory-diagnosed cervicovaginal infections in 207 pregnant Ngäbe women from western Panamá

	OR ± SE	95% CI	P
Bacterial vaginosis severity*†			
Vaginal trichomoniasis, severity‡	0.54 ± 0.06	0.44, 0.68	< 0.0001
Vaginal diplococcal infection, severity‡	1.80 ± 0.21	1.43, 2.25	< 0.0001
Vaginal yeast infection, severity‡	1.31 ± 0.22	0.94, 1.83	0.110
Vaginal trichomoniasis severity‡§			
Vaginal diplococcal infection, severity‡	1.33 ± 0.74	1.02, 1.72	0.030
<i>Lactobacillus</i> , quantity	1.97 ± 0.21	1.60, 2.42	< 0.0001
<i>Mobiluncus</i> , severity	1.23 ± 0.14	0.95, 1.58	0.110
Vaginal yeast infection severity‡**			
GA, week	0.97 ± 0.01	0.94, 1.00	0.122
Vaginal diplococcal infection, severity‡	1.36 ± 0.19	1.02, 1.80	0.032
<i>Bacteroides/Gardnerella</i> , severity	1.79 ± 0.48	1.05, 3.05	0.032
<i>Lactobacillus</i> , quantity	1.48 ± 0.31	0.98, 2.25	0.064
Vaginal diplococcal infection severity‡††			
GA, week	1.03 ± 0.02	0.99, 1.07	0.105
<i>Lactobacillus</i> , quantity	0.26 ± 0.08	0.14, 0.47	< 0.0001
Vaginal yeast infection, severity‡	1.46 ± 0.33	0.93, 2.30	0.098
Vaginal trichomoniasis, severity‡	1.30 ± 0.21	0.94, 1.81	0.114

CI = confidence interval; GA = gestational age; hpf = high-power field; OR = odds ratio; SE = standard error; VIF = variance inflation factor.

*Bacterial vaginosis severity according to Nugent score.

†Variables entered in model: GA; presence of caries and scabies; severity of vaginal trichomoniasis, yeast, and diplococcal infection. Overall model: $P < 0.001$; VIF = 1.00.

‡Coded as 0 = absent; 1+ = 1/hpf; 2+ = 2/hpf; 3+ = 3/hpf; 4+ = 4 or more/hpf.

§Variables entered in model: GA; presence of caries and scabies; quantity of *Lactobacillus*; severity of *Bacteroides/Gardnerella*, *Mobiluncus*, vaginal yeast, and diplococcal infection. Overall model $P < 0.0001$; VIF = 1.19.

||Coded as 0 = absent; 1+ = 1–5/hpf; 2+ = 6–10/hpf; 3+ = 11–20/hpf; 4+ = > 20/hpf.

**Variables entered in model: GA; presence of caries and scabies; quantity of *Lactobacillus*; severity of *Bacteroides/Gardnerella*, *Mobiluncus*, vaginal trichomoniasis, and diplococcal infection. Overall model $P = 0.015$; VIF = 1.08.

††Variables entered in model: GA; presence of caries and scabies; quantity of *Lactobacillus*; severity of *Bacteroides/Gardnerella*, *Mobiluncus*, vaginal trichomoniasis, and yeast infection. Overall model $P < 0.0001$; VIF = 1.11.

TABLE 6

Multiple ordered logistic regression models for severity of laboratory-diagnosed cervicovaginal infections in 79 lactating Ngäbe women from western Panamá

	OR ± SE	95% CI	P
Bacterial vaginosis severity*†			
Vaginal yeast infection, severity‡	0.30 ± 0.16	0.10, 0.87	0.027
Vaginal trichomoniasis severity‡§			
<i>Bacteroides/Gardnerella</i> , severity	0.60 ± 0.11	0.41, 0.88	0.008
<i>Mobiluncus</i> , severity	1.42 ± 0.30	0.94, 2.14	0.090
Vaginal diplococcal infection, severity‡	1.40 ± 0.30	0.91, 2.14	0.121
Vaginal yeast infection severity‡**			
Scabies, presence††	8.37 ± 7.53	1.43, 48.8	0.018
<i>Lactobacillus</i> , quantity	1.86 ± 0.52	1.07, 3.23	0.027
Vaginal diplococcal infection severity‡ ‡‡			
<i>Bacteroides/Gardnerella</i> , severity	0.62 ± 0.12	0.41, 0.92	0.019
<i>Lactobacillus</i> , quantity	0.18 ± 0.15	0.04, 0.91	0.039
Time after delivery, week	0.92 ± 0.04	0.85, 1.00	0.055
Vaginal trichomoniasis, severity‡	1.61 ± 0.47	0.91, 2.87	0.102

BV = bacterial vaginosis; CI = confidence interval; hpf = high-power field; OR = odds ratio; SE = standard error; VIF = variance inflation factor.

*Severity according to the Nugent score.

†Variables entered in model: weeks after delivery; presence of caries and scabies; severity of vaginal trichomoniasis, yeast, and diplococcal infection. Overall model $P = 0.023$; VIF = 1.00.

‡Coded as 0 = absent; 1+ = 1/hpf; 2+ = 2/hpf; 3+ = 3/hpf; 4+ = 4 or more/hpf.

§Variables entered in model: weeks after delivery; presence of caries and scabies; quantity of *Lactobacillus*; severity of *Bacteroides/Gardnerella*, *Mobiluncus*, vaginal yeast, and diplococcal infection. Overall model $P = 0.0043$; VIF = 1.13.

||Coded as 0 = absent; 1+ = 1–5/hpf; 2+ = 6–10/hpf; 3+ = 11–20/hpf; 4+ = > 20/hpf.

**Variables entered in model: weeks after delivery; presence of caries and scabies; quantity of *Lactobacillus*; severity of *Bacteroides/Gardnerella*, *Mobiluncus*, vaginal trichomoniasis, and diplococcal infection. Overall model $P = 0.009$; VIF = 1.00.

††Coded as 0 = absent; 1 = present.

‡‡Variables entered in model: weeks after delivery; presence of caries and scabies; quantity of *Lactobacillus*; severity of *Bacteroides/Gardnerella*, *Mobiluncus*, vaginal trichomoniasis, and yeast infection. Overall model $P < 0.0001$; VIF = 1.05.

|||A second model for vaginal diplococcal infection including the Nugent score revealed that severity of vaginal diplococcal infection was influenced by weeks after delivery (OR = 0.92 ± 0.03, 95% CI = 0.85, 0.99; $P = 0.036$); presence of caries (nonsignificant [ns]) and scabies (ns); Nugent score for BV (ns); severity of vaginal trichomoniasis (OR = 1.91 ± 0.51; 95% CI = 1.13, 3.23; $P = 0.015$) and yeast infection (ns). Overall model $P = 0.005$, VIF = 1.00.

Severity of vaginal trichomoniasis. Among pregnant women, the severity of vaginal trichomoniasis was lowered by BV diagnosed by the Nugent score (OR = 0.24, 95% CI = 0.14–0.42) (model not shown). When individual bacteria that contribute to the Nugent score were used (Table 5), the odds of more severe trichomoniasis were increased with more severe diplococcal infection (OR = 1.33, 95% CI = 1.02–1.72) and surprisingly higher quantity of *Lactobacillus* also increased the odds of more severe trichomoniasis (OR = 1.97, 95% CI = 1.60–2.42). During lactation (Table 6), the odds of more severe vaginal trichomoniasis were lower in mothers with more severe *Bacteroides/Gardnerella* (OR = 0.60, 95% CI = 0.41–0.88). No variables emerged as significant in the model using the Nugent score (model not shown).

Severity of vaginal yeast infection. The odds of more severe vaginal yeast infection were higher in those pregnant women with more severe vaginal diplococcal infection both in the model using the Nugent score (OR = 1.34, 95% CI = 1.04–1.73) (model not shown) and in the model using individual BV bacteria (OR = 1.36, 95% CI = 1.02–1.80) where severity of *Bacteroides/Gardnerella* also increased the odds of more severe vaginal yeast infection (OR = 1.79, 95% CI = 1.05–3.05) (Table 5). During lactation, a different picture emerged. The odds of more severe vaginal yeast infection was higher in mothers with scabies (OR = 6.45, 95% CI = 1.11–37.3) but lower in those with a higher Nugent score (OR = 0.72, 95% CI = 0.52–0.99) (model not shown). When reanalyzed with the individual BV bacteria (Table 6), presence of scabies continued to increase the likelihood of more severe yeast infection

(OR = 8.37, 95% CI = 1.43–48.8) and amount of *Lactobacillus* also increased the likelihood of more severe yeast infection (OR = 1.86, 95% CI = 1.07–3.23).

Severity of vaginal diplococcal infection. During pregnancy, the risk of more severe diplococcal infection was associated with a higher Nugent score (OR = 20.3, 95% CI = 4.73–87.04) (model not shown). Replacement of the Nugent score with individual BV-associated bacteria (Table 5) revealed a lower odds of more severe diplococcal infection in mothers with more *Lactobacillus* (OR = 0.26, 95% CI = 0.14–0.47). During lactation (Table 6), more severe diplococcal infection was more likely in mothers with more severe trichomoniasis (OR = 1.91, 95% CI = 1.13–3.23) in a model including the Nugent score (model not shown). However, when the data were reanalyzed with the individual BV-associated bacteria (Table 6), trichomoniasis severity was no longer significant, but both severity of *Bacteroides/Gardnerella* (OR = 0.62, 95% CI = 0.41–0.92) and amount of *Lactobacillus* (OR = 0.18, 95% CI = 0.04–0.91) lowered the likelihood of more severe diplococcal infection.

Intestinal nematode epg. During pregnancy, the multiple linear regression model using the Nugent score revealed that *Ascaris* epg was higher in pregnant women with AB/UTI ($P = 0.031$) and with more severe vaginal diplococcal infection ($P = 0.011$) (model not shown). When BV-associated bacteria were included in the model (Table 7), AB/UTI and diplococcal infection remained significant and the quantity of lactobacilli also emerged as positively associated with *Ascaris* epg. The positive relationship between hookworm and *Trichuris* was evident in models for both nematodes (Table 7), and neither model differed when the Nugent score was replaced with BV-associated bacteria. No regression analyses were done for

TABLE 7

Final multiple linear regression models on FLOTAC epg for gastrointestinal nematode infections from 72 pregnant Ngäbe women from western Panamá

	β coefficient $\pm$ SE	P
<i>Ascaris</i> , sqrt epg*		
AB/UTI, presence†	17.54 $\pm$ 6.52	0.009
Vaginal diplococcal infection, severity‡	11.16 $\pm$ 3.64	0.003
<i>Lactobacillus</i> , quantity§	8.91 $\pm$ 3.15	0.006
<i>Bacteroides/Gardnerella</i> , severity§	6.06 $\pm$ 3.84	0.12
Constant	–28.9 $\pm$ 15.9	0.07
Hookworm, sqrt epg		
GA, week	0.42 $\pm$ 0.23	0.067
<i>Trichuris</i> , epg	0.01 $\pm$ 0.002	< 0.0001
<i>Mobiluncus</i> , severity§	3.52 $\pm$ 1.96	0.077
Constant	–1.26 $\pm$ 7.16	0.861
<i>Trichuris</i> , sqrt epg**		
Hookworm, epg	0.004 $\pm$ 0.0004	< 0.001
Vaginal trichomoniasis, severity‡	2.02 $\pm$ 1.16	0.085
Constant	–1.70 $\pm$ 1.66	0.309

AB/UTI = asymptomatic bacteriuria/urinary tract infection; GA = gestational age; hpf = high-power field; sqrt epg = square root transformed eggs per gram; VIF = variance inflation factor.

*Variables entered in model: GA, presence of caries, scabies, and AB/UTI; amount of *Lactobacillus*; severity of *Bacteroides/Gardnerella*, *Mobiluncus*, vaginal trichomoniasis, yeast and diplococcal infection; hookworm and *Trichuris* epg. Overall model $F_{4, 67} = 5.56$; $P = 0.0006$; adjusted $R^2 = 0.20$; VIF = 1.58.

†Coded as 0 = absent, 1 = present.

‡Coded as 0 = absent; 1+ = 1/hpf; 2+ = 2/hpf; 3+ = 3/hpf; 4+ = 4 or more/hpf.

§Coded as 0 = absent; 1+ = 1–5/hpf; 2+ = 6–10/hpf; 3+ = 11–20/hpf; 4+ = ≥ 20 /hpf.

||Variables entered in model: GA, presence of caries, scabies, and AB/UTI; amount of *Lactobacillus*; severity of *Bacteroides/Gardnerella*, *Mobiluncus*, vaginal trichomoniasis, yeast, and diplococcal infection; *Ascaris* and *Trichuris* epg. Overall model $F_{3, 68} = 16.58$; $P < 0.0001$; adjusted $R^2 = 0.39$; VIF = 1.01.

**Variables entered in model: GA, presence of caries, scabies, and AB/UTI; amount of *Lactobacillus*; severity of *Bacteroides/Gardnerella*, *Mobiluncus*, vaginal trichomoniasis, yeast, and diplococcal infection; *Ascaris* and hookworm epg. Overall model $F_{2, 69} = 49.45$; $P < 0.0001$; adjusted $R^2 = 0.58$; VIF = 1.00.

the nematode infections during lactation because of the small number of fecal samples.

A composite picture. Within the vaginal tract, positive associations were detected among severity of vaginal yeast, diplococcal infection, and trichomoniasis (Figure 2). A complexity emerged as we explored the role of bacteria morphotypes included in the Nugent score diagnosis of BV. During pregnancy, the amount of *Lactobacillus* was associated with less severe diplococcal infection but with more severe vaginal trichomoniasis (Figure 2A). During lactation, the amount of *Lactobacillus* lowered the odds of more severe diplococcal infection but increased the odds of more severe vaginal yeast infection (Figure 2B). The presence of *Bacteroides/Gardnerella* increased the odds of more severe vaginal yeast infection during pregnancy (Figure 2A) but reduced the odds of more severe trichomoniasis and diplococcal infection during lactation (Figure 2B).

FIGURE 2. Relationships among infections of the vaginal tract (Nugent score for bacterial vaginosis as well as component bacteria shown in dashed boxes [*Bacteroides/Gardnerella*, *Mobiluncus*, and *Lactobacillus*], trichomoniasis, yeast, diplococcal), urinary tract (AB/UTI), intestine (hookworm, *Trichuris*, *Ascaris*), and skin (scabies, impetigo) in pregnant (A) and lactating (B) Ngäbe women from western Panamá. Associations based on multiple regression analysis ($P < 0.05$) are indicated as positive ($\longrightarrow$) or negative ($\dashrightarrow$). Associations emerging from χ^2 analysis are indicated as more ($\cdots\longrightarrow$) or less ($\cdots\dashrightarrow$) than expected by chance ($P < 0.005$).

The vaginal microflora was also associated with infections beyond the vaginal tract. In pregnancy, the quantity of *Lactobacillus* and severity of diplococcal infection were positively associated with *Ascaris* epg, AB/UTI was associated with more severe trichomoniasis but lower quantity of *Lactobacillus* (Figure 2A); and during lactation, the odds of AB/UTI were higher in women with higher quantity of *Mobiluncus* (Figure 2B).

DISCUSSION

Constraints on primary health care for vulnerable pregnant and lactating women living in remote areas of developing countries typically limit the possibility of screening for many infections. This study of pregnant and lactating women revealed the wide diversity of neglected infections in this extremely impoverished, rural, and indigenous population. Of particular note were the high prevalences of *Trichomonas vaginalis* (> 70%), BV (~60%), AB/UTI (> 35%), and hookworm (> 45%) in pregnancy and lactation as well as caries, scabies, vaginal yeast, and vaginal diplococcal infection at prevalences between 10% and 30%. Vaginal yeast infection was more common in pregnancy than lactation whereas vaginal diplococcal infection was less common but more severe in pregnancy. This preliminary descriptive study reveals several potential associations, both positive and negative, among pathogens that should be explored further. If confirmed, the findings highlight the need for appropriate management, in particular, of vaginal infections.

In comparing frequency and severity of infections between pregnant and lactating women, our observations were consistent with known physiological and hormonal differences. During pregnancy, higher levels of estradiol and higher glycogen content in vaginal secretions have been associated with vulvovaginal yeast,⁵⁹ and may similarly explain our finding that vaginal yeast was more prevalent during pregnancy than lactation. Elevated progesterone has been shown to facilitate diplococcal replication,⁶⁰ which could account for our finding of a more severe, though less prevalent, diplococcal infection in pregnancy compared with lactation. During normal pregnancy, the vaginal bacterial flora has been shown to be dominated by *Lactobacillus*⁶¹ whereas, during lactation, disruption of this balance has been associated with decrease in the number of *Lactobacillus*.⁶² This was consistent with our finding that *Lactobacillus* was more common in pregnancy than lactation. To our knowledge, ours is the first report of a higher prevalence of scabies during pregnancy than lactation.

With regard to associations among vaginal infections, BV has been reported to co-occur with *Trichomonas* in nonpregnant women.^{63–66} However, we observed that a higher severity of trichomoniasis was associated with lower severity of BV (but higher quantity of *Lactobacillus*) during pregnancy. To our knowledge, this has not been previously reported, and suggests that *Lactobacillus* may not be protective against trichomoniasis during pregnancy.

Furthermore, our results suggest both positive and negative associations of *Lactobacillus* with local as well as systemic infections, as higher quantities of vaginal lactobacilli were associated with reduced odds of diplococcal infection in both pregnant and lactating women and reduced the odds of AB/UTI in pregnancy, but with higher intestinal *Ascaris* epg during pregnancy and more severe vaginal yeast infection during lactation. Orally administered lactobacilli have been shown to

have immunomodulatory and oxidative properties in the urinary tract, and to decrease vaginal pH.⁶⁷ *Lactobacillus* has also been shown to inhibit pathogen adherence.⁶⁸ On the other hand, *Lactobacillus* was reported to be dominant within the vaginal microbiome of nonpregnant Chinese women who had vulvovaginal yeast infection,⁶⁹ suggesting that its presence increased the risk of yeast infection. Furthermore, *Lactobacillus casei* increased the susceptibility of rodents to intestinal nematodes,⁷⁰ whereas *Lactobacillus rhamnosus* reduced the duration of nematode infection.⁷¹ Our findings suggest that vaginal *Lactobacillus* may increase susceptibility to *Ascaris* or prolong duration of infection, an observation not previously reported.

Soil-transmitted nematodes are known not only to co-occur in pregnant women⁷² and women of reproductive age⁷³ but also to co-occur more frequently than expected by chance in age-adjusted data from a rural community in Brazil.⁷⁴ This co-occurrence is typically explained by common exposure routes, common household factors, common immune response mechanisms, common host genetics, or facilitated establishment in the presence of the other pathogen.^{75,76} Our data similarly revealed co-occurrence of *Trichuris* with hookworm, but *Ascaris* was not associated with hookworm or *Trichuris* in either χ^2 tests or in the multiple regression models of epg. This is consistent with our recent finding that high-prevalence spatial clusters of *Ascaris* did not overlap with those of *Trichuris* or hookworm in Panamanian preschool children.⁷⁷ Interestingly, *Ascaris* epg was positively associated the presence of AB/UTI, the quantity of vaginal lactobacilli, and the severity of vaginal diplococcal infection. To our knowledge, these associations have not been previously reported.

This study had several strengths. Despite the remote location and the limited health facilities, a range of infections was detected using a clinical approach together with basic laboratory assays. The absence of malaria and HIV in this population allowed us to explore relationships among the more neglected vaginal infections, skin and oral infections, as well as intestinal nematodes. By using multiple diagnostic methods for the intestinal nematodes and for AB/UTI, we reduced the likelihood that misclassification error resulted in spurious associations.⁷⁸

We also acknowledge several limitations. Our cross-sectional design precluded examination of causation. Although we were able to recruit approximately 90% of eligible women, without a full census of the catchment area, we cannot rule out selection bias. It was difficult to obtain fecal samples from the women who visited the health center only once. It is more difficult to detect negative associations than positive associations using standard statistical approaches.⁷⁹ Urine culture would have allowed us to establish the etiology of AB/UTI. Nucleic acid amplification tests would have allowed us to confirm if intracellular diplococci were *Neisseria gonorrhoeae* and to determine if *Chlamydia* was prevalent in this population (suspected based on our clinical findings, morphology of epithelial cells, and the presence of abundant leukocytes in the gram-stained vaginal smears⁸⁰).

Our findings have several implications for clinical care of this vulnerable population. Presumably healthy women with no evident physical distress had a multiplicity of conditions that together may have serious implications for maternal, fetal, and infant health. Efforts to prevent infection including improvements in hygiene, water and sanitation are needed. Sexual transmission may account for many of the vaginal

infections and possibly the skin and oral infections. With awareness of vaginal infections, women may be more receptive to discussions about reproductive health. Given the very high occurrence of vaginal infections, screening should be a routine part of prenatal care. To this end, health centers need to provide privacy for cervicovaginal exams and provide pathogen-specific differential diagnosis rather than clinically based management to prevent inappropriate or partial treatment of underlying pathologies. Until this is possible, new diagnostic protocols based on local epidemiology are urgently needed for the treatment of infectious diseases in pregnant and lactating women of the Ngäbe community. Finally, further research is needed to confirm both positive and negative associations reported here, to ensure appropriate management of co-occurring infections.

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