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ORIGINAL ARTICLE

Improvement of abnormal vaginal flora in Ugandan women by self-testing and short use of intravaginal antimicrobials

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Abstract The vaginal composition of African women is more often lactobacillus-deficient compared to that of women from other areas around the world. Lactobacillus-deficient microflora is a known risk factor for serious health problems, such as preterm birth, cervix cancer, and entrapment of human immunodeficiency virus (HIV) and other sexually transmitted infections (STIs). The aim of this study was to assess the effect of local vaginal antibiotic or antiseptic treatment on abnormal vaginal flora (AVF), aerobic vaginitis (AV), and bacterial vaginosis (BV) among women in rural, semi-urban, and urban areas in Uganda, as compared to placebo. In a double-blind, placebo-controlled, randomized trial, 300 women presenting for outpatient routine, follow-up, or medical care at Mulago Hospital in Kampala, Uganda, were enrolled to receive 6 days of treatment with vaginal rifaximin (RFX), dequalinium chloride (DQC), or placebo if they had an increased vaginal pH of >4.5 as determined by self-testing. At initial visit and at control visit after 4 weeks, a smear was taken for blinded wet mount microscopy to determine AVF, BV, AV, and *Candida* severity scores. As compared to placebo, both RFX or DQC treatments dramatically diminished BV prevalence and severity from the initial to follow-up visit: the BV score declined from 2.5 to 1.6 ($p < 0.0001$) and from 2.5 to 1.9 ($p < 0.0001$), respectively. Similarly, strong improvements in the AV score were seen in both treatment regimens: moderate and severe AV declined from AV scores of 6.3 to 3.6 ($p = 0.003$) and from 6.6 to 4.1 ($p < 0.004$), respectively. Also, women with AVF (deceased or absent lactobacilli) showed similar improvements when compared with placebo. Women with normal flora and *Candida* at the initial visit showed less *Candida* after 4 weeks in the group treated with DQC ($p = 0.014$). Even after a short duration of intravaginal treatment with local non-absorbable antiseptics or antibiotics produced significant, lasting improvements in the vaginal

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microbiome composition of women with disturbed vaginal microflora. As African women have high prevalences of BV, AV, and AVF, this approach could improve their odds to prevent health-compromising complications. Further studies assessing direct health outcomes are needed to substantiate this.

Introduction

The vaginal composition of African women is more often lactobacillus-deficient compared to that of women from other areas around the world. Lactobacillus-deficient microflora is a known risk factor for serious health problems, such as preterm birth, cervix cancer, and entrapment of human immunodeficiency virus (HIV) and other sexually transmitted infections (STIs). Due to the multiple side effects, questionable efficiency, and cost price of oral antibiotics, local vaginal application of antiseptics, antibiotics, and probiotics are preferred, but, to our knowledge, their efficiency has never been properly investigated in asymptomatic African women with abnormal vaginal microbiota.

Materials and methods

Aim

The aim of this study was to assess the intermediate term efficacy of a single course of two local non-absorbable, intravaginal antimicrobial products on different subtypes of abnormal vaginal flora (AVF) in a placebo-controlled randomized trial.

Regulatory and ethical considerations

Prior to the initiation of a study site, Femicare obtained approval from the appropriate regulatory agency and ethical committee of Kampala University Hospital to conduct the study in accordance with applicable country-specific regulatory requirements. The study was conducted in full accordance with Good Clinical Practice (GCP), all applicable subject privacy requirements, and the guiding principles of the Declaration of Helsinki.

Subjects

Three hundred asymptomatic non-pregnant women between 18 and 50 years of age, living in rural, semi-urban, or urban areas, and presenting at the routine gynecological outpatient clinic of Kampala University Hospital were invited to participate if they had an increased vaginal pH on self-testing. All women signed an informed consent before entering the study.

The women were asked to use a sterile glove and examine themselves by introducing one finger into the vagina and spread the obtained vaginal fluid on a glass slide, which is tipped with a pH strip (Macherey-Nagel, pH range 3.6–7.0) [1]. The glass slide was air-dried and kept for later transportation to Femicare in Belgium. Before discarding the glove, the finger was rinsed in a vial of CytoRich fluid, which was closed and numbered accordingly for later testing.

Consenting women with increased pH as assessed by themselves (red or orange color of the pH strip) were randomized to receive either rifaximin tablets ($n = 100$), dequalinium chloride ($n = 100$), or placebo ($n = 100$) intravaginally for 6 days. Patients and doctors were discouraged from adding other anti-infectious treatments, except when deemed necessary on clinical grounds. All women received treatment for 6 days and were asked to return for a control visit 4 weeks after the end of therapy. A number of questions, concerning age, medical history, obstetrical history, and sexual health were asked by a registered nurse.

Treatment

Rifaximin (RFX, Alfa Wassermann, Bologna, Italy) is a non-absorbable antibiotic that is indicated for infectious diarrhea. Currently, the drug is studied for vaginal application in women with bacterial vaginosis (BV), where it was found to be efficient and safe in doses ranging from 25 to 100 mg a day for 5 days [2]. Furthermore, it was proven to introduce profound changes in the microbiome of these women [3–5]. Dequalinium chloride (DQC, Medinova, Zürich, Switzerland) is an antiseptic product that has been marketed for women with AVF in some European countries for the past several decades. Only recently was it studied in a double-blind randomized way and proven to be as efficient as clindamycin in the treatment of women with BV [6]. Neither of the drugs have ever been formally tested for AV.

The manufacturers of RFX and DQC were each asked to produce their product and placebo in similar, undistinguishable tablets in a 2:1 product:placebo ratio, to treat 150 women in each group, respectively.

Randomization procedure

A computerized randomization list was created with Research Randomizer to allocate a unique patient number that was linked with product A, product B, or placebo in a 1:1:1 ratio. So, in total, 300 patients were randomized to receive the next number of the study medication in the order of their appearance at consultation. During the full duration of the study, the patients, investigators, or lab personnel had no knowledge about the type of study drug that was used by the subjects.

Table 1 Composition of aerobic vaginitis (AV) score according to Donders et al. [9]

AV score	LBG	No. of leukocytes	Proportion of toxic leukocytes	Background flora	Proportion of parabasal cells
0	I and IIa	≤10/hpf	None or sporadic	Unremarkable or cytolysis	None or <1% of epitheliocytes
1	IIb	>10/hpf and ≤10/epithelial cell	≤50% of leukocytes	Small coliform bacilli	≤10% of epitheliocytes
2	III	>10/epithelial cell	>50% of leukocytes	Cocci or chains	>10% of epitheliocytes

LBG Lactobacillary grade; *HPF* high-power field (400× microscopy)

Laboratory testing

The slides were numbered, air-dried, and kept in a plastic holder in a dry environment at room temperature [7], to be collected and sent to Femicare, Tienen, Belgium, for microscopy reading and scoring of the microbial abnormalities according a standardized scheme published elsewhere [8]. After reading, the results were linked to the unblinded original clinical database created on-site.

The slides were classified as follows: BV score: BV is diagnosed as the presence of *Gardnerella* or *Mobiluncus* morphotypes and/or clue cells, in combination with the absence of lactobacilli (lactobacillary grade III). A distinction is made between full-blown BV and partial, patchy granular BV. Absence of BV was scored 0, partial BV (<20% clue cells) scored 1, and full-blown BV scored 2.The latter diagnosis of full-blown BV equates to a Nugent score >6 on Gram stain. AV score: to be comparable, aerobic vaginitis (AV) is also purely based on microscopy (Table 1). Lactobacillary grades are the basis for a composite AV score to which any of the following four variables are added: number of leukocytes proportional to the epitheliocytes, proportional presence of toxic leukocytes to the total number of leukocytes, proportional presence of parabasal cells vs. intermediate/superficial cells, and characteristics of the background flora [9]. A composite score <3 represents normal flora, a score of 3 to 4 corresponds to light AV, 5 to 6 to moderate AV, and a score above 6 (with a maximum of 10) to severe AV. The latter is identical to the so-called "desquamative inflammatory vaginitis (DIV)", so that such a diagnosis can be seen as the most extreme form of AV [10]. *Candida* was diagnosed as the unequivocal presence of blastospores, pseudohyphae of hyphae.

The CytoRich vial with the glove-rinsing fluid was collected in adequate tubes, numbered, and frozen at −20 °C until transport to Femicare, and, there, was kept frozen at −80 °C until it was required for further

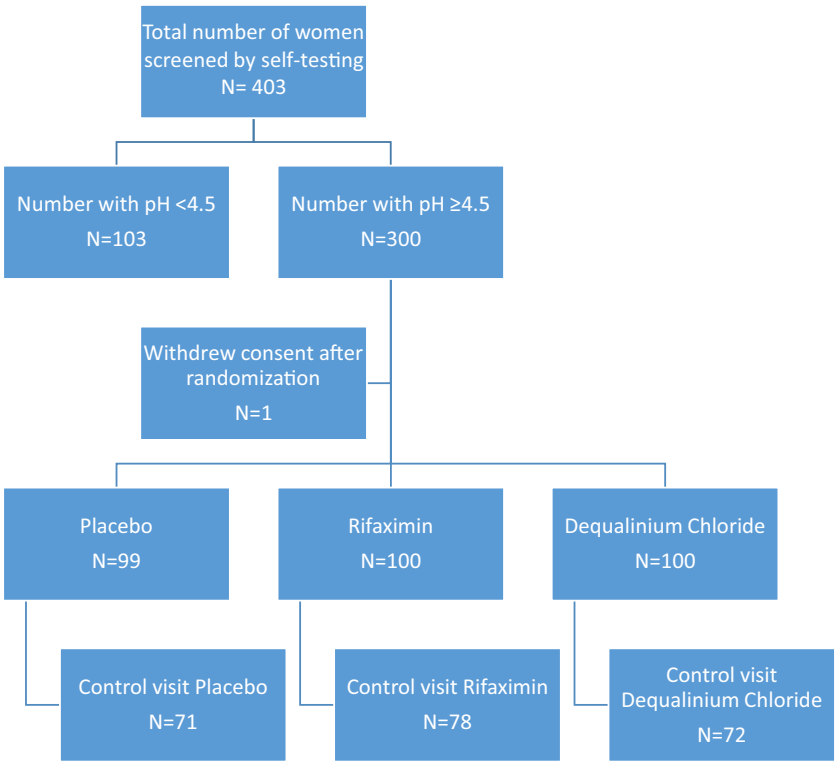

Fig. 1 CONSORT flow chart of the included study patients

microbiome testing. As these analyses are not within the scope of this contribution, they are not discussed here.

Data analysis

After database lock and unblinding of the results, comparisons within groups were made by Chi [2], or Fisher's T if appropriate, for discontinuous variables. After checking for normal distribution, Student's T was used for continuous variables.

Results

Of the 403 women who performed self-testing, 300 were randomized because of a self-assessed vaginal pH of 4.5 or above (Fig. 1). Of them, 299 enrolled in the study; one patient declined after signing the informed consent, before starting the medication, without giving a reason. Living area and demographics, including age, parity, degree of education, medical history, and sexual habits, were evenly

Table 2 Demographics of the three treatment groups. There were no significant differences between groups

		Placebo, $n = 99$	Rifaximin, $n = 100$	Dequalinium chloride, $n = 100$
Age (years, mean ± SD)		31.34 ± 7.51[a]	29.88 ± 7.07	30.66 ± 7.52
Number of children (mean ± SD)		1.98 ± 1.84[a]	1.84 ± 1.73[a]	1.61 ± 1.68
Number of pregnancies (mean ± SD)		2.47 ± 1.99	2.21 ± 1.91[b]	2.09 ± 1.83
Age when had first child ($n = 73$ para)		19.38 ± 4.3	20.11 ± 3.85	20.63 ± 3.93
Schooling	None	3	6	1
	<6 years	5	7	4
	6–12 years	26	20	20
	12–18 years	23	25	24
	>18 years	39	41	40
Region	Rural	10 (10.7%)	4 (4.2%)	11 (11.7%)
	Semi-urban	17 (18.3%)	30 (31.3%)	22 (23.4%)
	Urban	66 (71.0%)	62 (64.6%)	61 (64.9%)
Marital status	Married	71	72	70
	Living together	2	3	6
	Single	25	24	23
Visited ambulatory care		51	52	59[b]
Visited local healthcare center		41[c]	39[b]	47
Visited traditional healer		13	10	14[a]
HIV-positive		8 (8.6%)[d]	13 (14.4%)[e]	12 (12.9%)[d]
Medical history of genital infections				
Vaginitis (yeast)		35[a]	39[b]	40[b]
Genital herpes		4[a]	10[c]	2[a]
Syphilis		11[a]	25[b]	17[a]
Contraception	None, no partner	11 (11.6%)	9 (9.7%)	8 (8.4%)
	None, pregnancy wish	32 (33.7%)	27 (29.0%)	27 (28.4%)
	None, other reason	20 (21.1%)	16 (17.2%)	20 (21.1%)
	Calendar/temperature	3 (3.2%)	2 (2.2%)	3 (3.2%)
	Coitus interruptus	2 (2.1%)	0 (0%)	0 (0%)
	Oral oestroprogestins	6 (6.3%)	6 (6.5%)	6 (6.3%)
	Condoms	4 (4.2%)	9 (9.7%)	6 (6.3%)
	Copper IUD	5 (5.3%)[f]	6 (6.5%)	10 (10.5%)
	DMP injection	8 (8.4%)	14 (15.1%)	13 (13.7%)
	Sterilization	3 (3.2%)	2 (2.2%)	1 (1.1%)
	Other	0 (0%)	2 (2.2%)	1 (1.1%)
Vaginal douching	>1/day	84 (85.7%)	77 (77%)	74 (74%)
	1/day	3 (3.1%)	3 (3%)	3 (3%)
	Few times/week	3 (3.1%)	2 (2%)	1 (1%)
	1/week	0 (0%)	0 (0%)	5 (5%)
	Now and then[g]	1 (1.0%)	3 (3%)	1 (1%)
	Never	7 (7.1%)	15 (15%)	16 (16%)
Vaginal douching fluid	Plain water	63 (68.5%)	64 (72.7%)	56 (65.1%)
	Soap	29 (31.5%)	23 (26.1%)	30 (34.9%)
	Disinfectant	0 (0%)	1 (1.1%)	0 (0%)
Last sexual intercourse	<24 h	14 (14.7%)	17 (17.2%)	16 (17.0%)
	1–3 days	14 (14.7%)	16 (16.2%)	12 (12.8%)
	>3 days	67 (70.5%)	66 (66.7%)	66 (70.2%)
Condom used at last intercourse (mean ± SD)		14 (15.1%)[d]	17 (17.7%)[h]	18 (19.1%)[i]
Age at first sex (mean ± SD)		17.34 ± 2.99[a]	17.78 ± 3.07[a]	18.04 ± 3.26[a]
Number of life sex partners (mean ± SD)		2.36 ± 1.05[c]	2.27 ± 1.16[b]	2.35 ± 1.10[c]
Number of sex partners in the last 2 months (mean ± SD)		0.89 ± 0.35[c]	0.91 ± 0.36[c]	0.90 ± 0.42[c]

Table 3 Influence of treatment with vaginal placebo, rifaximin, or dequalinium chloride tablets on different markers of disturbed vaginal bacterial microflora in Ugandan women with increased vaginal pH

	Placebo			Rifaximin			Dequalinium chloride		
	Pretreatment, $n = 99$	One month posttreatment, $n = 71$	OR (95% CI), p-value	Pretreatment, $n = 100$	One month posttreatment, $n = 78$	OR (95% CI), p-value	Pretreatment, $n = 100$	One month posttreatment, $n = 72$	OR (95% CI), p-value
LBG (nl flora)	1.56 ± 0.98 (18)	2.75 ± 1.34 (16)	1.2 (0.4, 2.0), $p = 0.007$	1.39 ± 0.98 (18)	2.00 ± 1.29 (13)		1.22 ± 0.58 (27)	1.68 ± 1.06 (19)	
BV (p and f)	2.45 ± 0.50 (51)	2.12 ± 0.84 (34)		2.48 ± 0.50 (48)	1.57 ± 0.88 (35)	−0.9 (−1.2, −0.6), $p < 0.0001$	2.51 ± 0.51 (49)	1.87 ± 0.84 (38)	−0.6 (−0.9, −0.3), $p < 0.0001$
Light AV	3.51 ± 0.52 (16)	3.09 ± 1.70 (11)		3.50 ± 0.51 (18)	2.47 ± 1.73 (15)	−1.0 (−2.0, 0.04) $p = 0.040$	3.69 ± 0.48 (13)	1.86 ± 1.68 (7)	−1.8 (−3.4, −0.3) $p = 0.029$
Moderate/severe AV	6.00 ± 0.87 (9)	4.67 ± 4.51 (3)		6.33 ± 1.30 (12)	3.64 ± 2.25 (11)	−2.7 (−4.3, −1.0), $p = 0.003$	6.64 ± 1.86 (11)	4.11 ± 1.54 (9)	−2.5 (−4.1, −0.9), $p = 0.004$
AVF (LBG IIb, III)	3.66 ± 0.48 (67)	3.26 ± 1.03 (47)	−0.4 (−0.7, −0.1), $p = 0.016$	3.84 ± 0.37 (67)	3.28 ± 1.05 (54)	−0.6 (−0.8, −0.3), $p = 0.0005$	3.78 ± 0.42 (63)	3.17 ± 1.15 (47)	−0.6 (−1.0, −0.2), $p = 00016$
Absence of LB	4.00 ± 0.0 (46)	3.44 ± 0.96 (34)	−0.6 (−0.9, −0.2), $p = 0.002$	4.00 ± 0.0 (58)	3.42 ± 0.97 (45)	−0.6 (−0.9, −0.3), $p = 0.0002$	4.00 ± 0.0 (50)	3.29 ± 1.09 (38)	−0.7 (−1.1, −0.4), $p = 0.0003$
pH >4.7 (red)	5.22 ± 0.38 (53)	5.17 ± 0.61 (31)		5.27 ± 0.34 (46)	5.02 ± 0.55 (37)	−0.3 (−0.5, 0.04), $p = 0.019$	5.25 ± 0.36 (38)	5.04 ± 0.58 (26)	

Values are given as mean ± standard deviation. The numbers in any given category are given in parentheses

BV Bacterial vaginosis (*p* partial, *f* full-blown); *AV* aerobic vaginitis; *AVF* abnormal vaginal flora; *LBG* lactobacillary grade; *OR* odds ratio; *CI* confidence interval

spread amongst the treatment groups (Table 2). Of all the women, 72, 78, and 72% returned for the second visit in the placebo, RFX, and DQC groups, respectively, and had analyzable data from both visits. According to the inquiry, no women had used rescue medication in the 4-week period.

The influence of a single course of 6 days of intravaginal treatment on the vaginal bacterial microflora, as assessed after 4 weeks, is shown in Table 3. In women receiving placebo, women with normal flora slightly deteriorated [the mean lactobacillary grade (LBG) increased from 1.6 to 2.8, diff 1.2, 95% confidence interval (CI) 0.4, 4.2, $p = 0.007$], while on the other hand, some women with abnormal flora at inclusion had a better flora (fewer LBG II and III) after 4 weeks (mean LBG from 3.7 at inclusion to 3.3 after 4 weeks, diff −0.4, 95% CI −0.7, −0.1, $p = 0.016$) (Fig. 2). The prevalence and severity of AV and BV did not change after 4 weeks of placebo treatment.

Women on RFX or DQC treatment, however, dramatically diminished their BV score from the initial visit to the follow-up visit: BV score declined from 2.5 to 1.6 [odds ratio (OR) −0.9, 95% CI −1.2, −0.6, $p < 0.0001$] and from 2.5 to 1.9 (OR −0.6, 95% CI −0.9, −0.3, $p < 0.0001$), respectively. Similarly, strong improvements in AV score was seen in both women starting with light AV [AV score declined from 3.5 to 2.5 (OR −1.0, 95% CI −2.0, 0.04, $p = 0.04$) and from 3.7 to 1.9 (OR −1.8, 95% CI −3.4, −0.3, $p = 0.029$)] and, to a greater extent, with moderate and severe AV [AV score declined from 6.3 to 3.6 (diff −2.7, 95% CI −4.3, −1.0, $p = 0.003$) and from 6.6 to 4.1 (diff −2.5, 95% CI −4.1, −0.9, $p < 0.004$)]. Also, women with AVF (decreased or absent lactobacilli) showed similar improvements of the LBGs 4 weeks after treatment in both the RFX and DQC groups ($p = 0.0005$ and $p = 0.00016$, respectively) when compared with placebo. Although some decreasing trends of pH measurements were seen, these improvements did not reach significance.

The influence of treatment on the presence of vaginal *Candida* after 4 weeks is shown in Table 4. It can be seen that there is no overall change in *Candida* carriage during the second visit of women treated with placebo, RFX, or DQC, respectively. Also, in cases with disrupted flora, like BV or AV, the presence of *Candida* did not change in the three treatment groups, except in the group of *Candida* combined with AV treated with RFX, where an increase in the *Candida* severity score was seen (diff 0.73, 95% CI 1.0, 0.5, $p = 0.0001$). On the contrary, women with normal flora and *Candida* at the initial visit showed less *Candida* after 4 weeks [severity score dropped from 1.32 to 1.05 (diff −0.35, 95% CI −0.5, −0.1, $p = 0.014$) in the group treated with DQC].

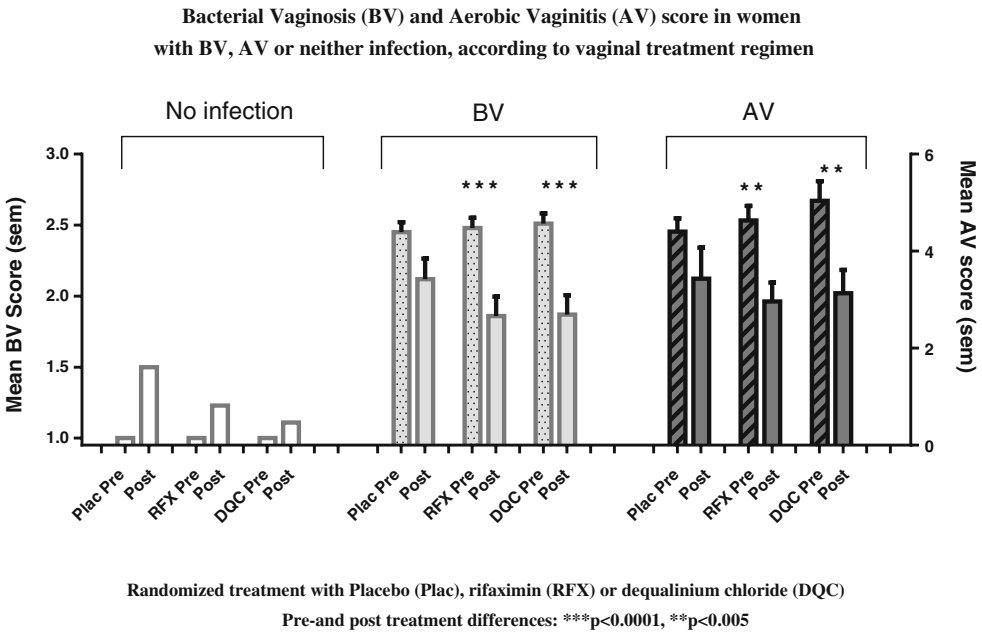

Fig. 2 Evolution of bacterial vaginosis (BV) score and aerobic vaginitis (AV) score before and 4 weeks after 6 days of intravaginal treatment with placebo (*Plac*), dequalinium chloride (*DQC*), or rifaximin (*RFX*) in 300 Ugandan women with a vaginal pH of 4.5 or more

Discussion

To perform a study in asymptomatic women in Central Africa has inherent limitations. Motivation to return for control visits is not obvious if no direct health benefits can be promised, as in the current study. Payment other than transport costs were not permitted, and the journal journey to the hospital is not easy due to poor basic infrastructure and public transport, especially for those who come from the rural areas around the city of Kampala. Due to the study setup with placebo tablets that were indistinguishable from products and the computer-generated randomization list, the process of random allocation to treatment groups seemed to be unaffected by any known bias.

With persistence and the help of regular phone calls and phone messages, 72–78% of women were reached to return for control visits after 4 weeks, with an equilibrated return rate within the three treatment groups (placebo, rifaximin, and dequalinium chloride groups). Women did not use rescue medication during the study period. Besides being asymptomatic, this can be explained by the poor availability of over-the-counter medication, distance, and constraints to see another healthcare provider within the short period of time. Also, as women were not included based on symptoms but were consulting for reasons other than vulvovaginitis, this decreased the risk for needing extra medication.

In former studies, we demonstrated that Ugandan women accept vaginal self-testing and understand the procedure and

Table 4 Influence of treatment with vaginal placebo, rifaximin, or dequalinium chloride tablets on the microscopic presence of vaginal *Candida* (Ca, *Candida* severity score) in different settings of vaginal microflora in 300 Ugandan women with increased vaginal pH

	Placebo			Rifaximin			Dequalinium chloride		
	Pretreatment	One month posttreatment	OR (95% CI), *p*-value	Pretreatment	One month posttreatment	OR (95% CI), *p*-value	Pretreatment	One month posttreatment	OR (95% CI), *p*-value
All women	1.24 ± 0.64 (99)	1.21 ± 0.56 (71)		1.32 ± 0.7 (100)	1.24 ± 0.65 (78)		1.39 ± 0.78 (100)	1.31 ± 0.80 (72)	
Ca + normal flora	1.15 ± 0.83 (20)	1.19 ± 0.40 (16)		1.45 ± 0.69 (20)	1.38 ± 0.65 (18)		1.32 ± 0.48 (28)	1.05 ± 0.23 (19)	−0.35(−0.5, −0.1), $p = 0.014$
Ca + msAV	1.44 ± 0.92 (25)	1.14 ± 0.53 (14)		1.10 ± 0.40 (30)	1.83 ± 0.59 (26)	0.73 (1.0, 0.5), $p = 0.0001$	1.38 ± 0.88 (24)	1.88 ± 1.36 (16)	
Ca + BV	1.24 ± 0.59 (51)	1.25 ± 0.70 (34)		1.23 ± 0.69 (48)	1.26 ± 0.78 (35)		1.41 ± 0.85 (63)	1.42 ± 0.95 (35)	

Values are given as mean ± standard deviation. The numbers in any given category are given in parentheses

BV Bacterial vaginosis (*p* partial, *f* full-blown); *msAV* moderate to severe aerobic vaginitis; *OR* odds ratio; *CI* confidence interval

interpretation of the results well [7]. Furthermore, we concluded in that study that AVF is frequent in these women, and is not related only to BV, but also in a large proportion to AV, which was unrecognized in former African studies relating to this subject [11, 12].

It is of major importance for women to maintain a healthy, lactobacillary-dominant vaginal microflora [13], in order to be protected against the acquisition [14] and female-to male transmission [15] of HIV, *Trichomonas vaginalis* vaginitis [16], herpes simplex 2 [17], chlamydia [18, 19], and other adverse outcomes, such as preterm birth [20–22] or enhanced development of human papilloma virus (HPV)-induced cervical cancer [23, 24]. It is clear that not only BV but also AV and its associated bacteria play a crucial role in the prevention of HIV [14, 25] and the other above-mentioned health impairments [19, 21, 24, 26]. In order to improve the health odds in women with AVF, vaginal disinfectants, probiotics, and local antibiotics could be tried. For this purpose, and given the wide variety of aerobic and aerobic microbial conditions associated with AVF in African women, we decided not to opt for testing narrow-spectrum vaginal products that are active against BV (e.g., metronidazole, tinidazole), but, rather, tested non-absorbed novel vaginal products with a broad antimicrobial spectrum. Dequalinium chloride is a broad-spectrum antiseptic that was favorably tested against clindamycin in BV patients [6], but was never tested for AV. Similarly, another locally active, broad-spectrum antimicrobial, rifaximin, was tested extensively in BV patients [2–5], but not in AV patients.

In a double-blind, randomized, placebo-controlled trial, we were able to demonstrate that both products were able to improve the vaginal flora significantly up to 4 weeks after a single treatment phase of 6 days. This was unexpected, as many products only provide short-lived effects in asymptomatic women and as it was not known what the effect could be in a society where vaginal douching at least once a day is routine [7]. Also, participants were allowed to have unprotected sexual intercourse as soon as the 6-day therapy was ended, so sexual intercourse did not influence the results. Of importance, not only was BV microbiota ameliorated significantly, but also the prevalence of AV decreased dramatically.

As an antibiotic, RFX seems to have a more negative effect on the likelihood to develop *Candida* colonization, especially in women with a coinfection with AV, whereas the antiseptic DQC decreased *Candida* prevalence, at least in women with a normal microflora. Similar findings surfaced when the effect of DQC on BV was studied: there was a non-significant trend towards less *Candida* colonization when compared to intravaginal RFX treatment [6, 27].

As a single treatment is efficient for causing improvement of the vaginal microbiome that endures for weeks, it can be discussed whether repetitive, intermittent treatment for longer periods of time would not enable further improvement of the health threats that these women face. For this, studies measuring the long-term effects of vaginal disinfectant treatment over 3–6 months would need to be designed. More studies using long-term prophylactic treatment to assess health outcomes are needed. Due to its anti-*Candida* effect, with similar positive effects on the bacterial microflora and low likelihood to develop antimicrobial resistance, DQC could be seen as a preferred product for this purpose.

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Compliance with ethical standards

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Conflict of interest None of the authors has any competing interests to disclose. Prof. G. Donders was a scientific consultant for both Alfa Wasserman, Bologna, Italy, and Medinova, Zürich, Switzerland.

Ethical approval Protocol revised and approved by Kampala University Hospital Ethical Committee.

Informed consent Every patient received information and signed written informed consent in full accordance with Good Clinical Practice (GCP), all applicable subject privacy requirements, and the guiding principles of the Declaration of Helsinki.

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