

Increased vaginal pH in Ugandan women: what does it indicate?

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Abstract Abnormal vaginal flora (AVF), indicative of bacterial vaginosis (BV) and/or aerobic vaginitis (AV), amongst other abnormalities, is a risk factor for multiple complications in pregnant as well as non-pregnant women. Screening for such conditions could help prevent these complications. Can self-testing for increased vaginal pH reliably detect BV and other high-risk microflora types, and is this more accurate than performing Gram stain-based Nugent score when screening for high-risk microflora? A total of 344 women presenting at different outpatient clinics in Mulago Hospital and Mbuikwe Outpatient clinics in Kampala, Uganda, were asked to test themselves by introducing a gloved finger into the vagina and smearing it on a microscopy slide, on which a pH strip was attached. Self-assessed categories of normal (pH 3.6–4.4), intermediate (4.5–4.7) or high pH (>4.7) were compared with demographic and with centralised microscopic data, both in air-dried rehydrated wet mounts (Femicare), as well as in Gram-stained specimens (Nugent). AVF was present in 38 %,

BV in 25 % and AV in 11 % of patients. High pH and AVF is correlated with human immunodeficiency virus (HIV), infertility, frequent sex, but not vaginal douching. Screening for raised pH detects 90 % of AVF cases, but would require testing over half of the population. As AV and non-infectious conditions are frequent in women with AVF and high pH, Nugent score alone is an insufficient technique to screen women for a high-risk vaginal microflora, especially in infertile and HIV-infected women.

Introduction

Bacterial vaginosis (BV) is a pathologic condition of the vagina, caused by an overgrowth of anaerobic bacteria, leading to a bothersome foul smelling discharge. Increased pH is one of the Amsel criteria to diagnose the disease and is often used as a surrogate marker for the presence of BV [1–4]. However, it needs to be taken into consideration that pH can also be increased by other factors, both infectious (e.g. chlamydia, gonorrhoea, trichomoniasis, aerobic vaginitis) [5] and non-infectious (e.g. sperm, vaginal douching, use of creams etc.). Former studies suggested a high specificity of increased pH for predicting BV in a population with a low risk of infectious lower genital tract abnormalities [1] and can be used to prevent preterm birth [6], but in high-risk populations, it has never been properly addressed.

Besides BV, another abnormal infectious condition of the vagina, aerobic vaginitis (AV), is often associated with a disrupted flora, but without the typical granular microflora displayed by anaerobes in BV flora [7]. As opposed to BV, a more scanty flora of cocci or small enteric bacilli is present in AV, as well as a variable inflammatory response, as demonstrated by increased numbers of leucocytes, presence of toxic leukocytes and high concentrations of pro-inflammatory

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cytokines in the vaginal fluid [8]. The presence of an abnormal vaginal flora (AVF), AV and BV increases the risk of morbidity in both pregnant and non-pregnant women [9].

Black women seem to be more vulnerable to AVF and BV than Caucasian women [10], both in the US [11] and in African countries [12], with alleged prevalence rates of over 50 % [11, 12]. In a former paper, we reported high acceptance and feasibility of vaginal pH self-screening of Ugandan women [13]. In the present paper, we address the question as to whether such vaginal pH measurement is a surrogate marker for BV, or also relates to other important, non-BV abnormalities of the vaginal microenvironment.

Materials and methods

Aim

To determine which proportion of women with abnormal vaginal pH in Uganda has microscopical confirmed BV. How important are flora types other than full-blown BV, like AV and partial BV, in these women?

Subjects

Inclusion of 360 unselected, consecutive and consenting women between 18 and 50 years of age, living in rural areas, semi-urban areas or urban areas, and presenting at birth control, general gynaecology, infertility and prenatal clinics at Mulago/Mbukwe Hospital outpatient departments. The study and informed consent document was approved by the ethical committee of the Mulago University Hospital and all patients approved and signed an informed consent before entering the study.

After having read and signed the informed consent, women were asked to examine themselves with a sterile glove by introducing one finger into the vagina and spreading the obtained vaginal fluid on a glass slide, on which a pH strip (pH-Fix, range pH 3.6–6.1, Macherey-Nagel GmbH & Co. KG, Düren, Germany) was fixed, as described and demonstrated elsewhere [14]. After 1 min, the patients had to interpret the pH strip result as: (1) normal (yellow, corresponding to a pH range of 3.6–4.4), (2) intermediate (orange, corresponding to a pH range of 4.4–4.7) or (3) abnormal (red, corresponding to a pH range of more than 4.7). The slide was kept for later transportation to Femicare, clinical centre for research for women in Tienen, Belgium, where microscopy was performed after rehydration with normal saline by a blinded investigator [15] and for Gram staining and scoring according to Nugent [16].

With the assistance of a registered nurse, a questionnaire was filled out to collect information on age, medical history, obstetrical history and sexual health of the participating

women. Participating women were offered a small transportation fee of 5 Euros.

Laboratory testing

The slides were transported to a central microscopy laboratory (Femicare, Tienen, Belgium) for microscopy reading of rehydrated air-dried smears. A 400× magnification Leica IM1000 phase contrast light microscope (Leica, Marburg, Germany) was used, and various were recorded, as outlined in the following sections. The validated microscopic findings [lactobacillary grade (LBG), BV, AV] were linked to the three pH groups, normal, intermediate and abnormal, as described above.

LBG

Normal, grade I flora, corresponds to predominantly lactobacillary morphotypes, with very few coccoid bacteria present [care being taken not to misidentify the cellular debris from lysed epithelial cells (epitheliolysis) as coccoid bacteria]. The intermediate grade II flora corresponds to a diminished lactobacillary flora, mixed with other bacteria. We subdivided this group into slightly disturbed, fairly normal (IIa) and moderately disturbed, rather abnormal (IIb) lactobacillary flora [7, 17]. Finally, the grossly abnormal grade III flora consists of numerous other bacteria, with no lactobacilli present. LBG 0 is a state where no flora is discovered, so, also, no lactobacillary morphotypes can be seen, nor other microflora. As both LBG IIb and LBG III are considered abnormal (e.g. in pregnancy), both were considered together with LBG 0 as AVF in the definition of outcome parameters. It is important to stress that not all cases with AVF equal BV. Several other abnormalities can be associated with AVF, such as complicated candidosis, trichomoniasis and AV [7].

BV types

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). Full-blown BV is defined as a full replacement of the lactobacillary flora with typical granular, anaerobic morphotypes of bacteria, resulting in >20 % clue cells, while partial BV indicates a transition form, with patchy granular BV areas and areas with normal or AV flora in the same slide [7]. This results in sporadic clue cells, accounting for less than 20 % of epithelial cells. The former diagnosis equates with a Nugent score >6 on Gram stains; the Nugent equivalent of the latter is unknown, as their intermediate flora does not completely cover this diagnosis. Due to their particular and diverse pathogenicity, both partial BV and full-blown BV are withheld as outcome parameters in the outcome analysis.

Indeed, during pregnancy, partial BV seemed to be more frequently related to preterm delivery than full-blown BV, indicating that the presence of these non-aerobic bacterial areas may play an important role in the causation of preterm birth [18, 19].

AV score

The diagnosis of AV is also purely based on microscopy. Lactobacillary grades are the basis for a composite score (AV score) to which any of the four following variables are added: leucocytes, presence of toxic leucocytes, presence of parabasal cells and background flora [8]. A composite score ≤ 2 represents normality. A score of 3 to 4 corresponds to slight AV, a score above 6 (to a maximum of 10) to severe AV. A score of 8 to 10 is similar to the so-called ‘desquamative inflammatory vaginitis,’ so that the latter diagnosis can be seen as the most extreme form of AV. Moderate and severe AV (AV score >4) and severe AV (AV score >6) were considered clinically significant outcome parameters in the analysis.

Confounding factors

As possible confounding factors, we also searched for the presence of red blood cells and sperm on the smears. Red blood cells were not discovered, since women were not allowed to participate when they have any sign of menstrual or other bleeding. Sperm was recognised as typical ovaloid-shaped bodies, with a clear acrosomal cap in darker shade, and with or without the presence of tails [7]. Presence of sperm was marked as a non-quantitative, bimodal marker (present or absent).

Lastly, slides were also stained according to Gram’s method and scored according to the Nugent scoring system [16]. In this system, a Nugent score of 1 to 3 is considered normal, 4 to 6 intermediate and 7–10 compatible with BV.

Statistical analysis

Discrete numerical values were expressed as proportions and compared by the use of Chi-square tests or Fisher’s test in case of numbers lower than 5 expected values in any particular cell. For multiple columns, the analysis of variance (ANOVA) test was used. Continuous variables, after confirming normal distribution, were tested with Student’s t-test. *p*-Values <0.05 were considered significant and <0.01 highly significant. For sensitivity and specificity analysis of the pH as a screening test, a Nugent score of 7–10 was used as the gold standard, as the classical Amsel criteria contain pH itself as a composite criterion.

Results

Of the 360 women, the data from 344 (95.6 %) could be analysed. Of the remaining 16, the questionnaire was not properly filled out, a self-assessment of vaginal pH was not possible, the vaginal smear was lost or the slide was broken. Vaginal wet mount and Gram stain was possible for the slides of 338 women (93.9 %). The epidemiologic characteristics of the study group are given in Table 1. The mean age of the women was 28.3 ± 6.0 years, parity ranged from 0 to 7 (mean 2.0) and educational level was low: only 92 (26.7 %) attended school until the age of 18 years or above. The proportion of women with abnormal vaginal pH did not differ with the level of education. Most women lived in urban regions or in the vicinity of a big city, 87/336 (25.9 %) living in the countryside; their pH patterns did not differ according to the living area, nor to the tribes from which they originated (detailed data on file).

Of the whole group, 144 (41.9 %) women attended antenatal, 30 (8.7 %) gynaecology, 18 (5.2 %) infertility and 147 (42.7 %) family planning clinics. Most women attending either the gynaecology or the antenatal clinic had a low pH (43.3 % and 42.4 %, respectively), while 20 % of women expressed a high pH above 4.7. This was in contrast with the low proportion of women with low pH in the family planning clinic (21.8 %) or infertility clinic (5.6 %) (33/165 vs. 74/174, $p < 0.0001$). In the family planning clinic, 75/147 (51.0 %) women had an intermediate pH of 4.5–4.7, compared to 52/144 (36.1 %) in the antenatal group ($p < 0.01$) and 11/30 (36.7 %) in the gynaecology group ($p = 0.1$). In the infertility group, 10/18 (55.6 %) patients had a high vaginal pH of >4.7 , compared to 77/323 (23.8 %) in the other groups ($p = 0.0027$).

Age of sexual debut did not influence the vaginal pH, but the number of previous sexual partners did: 45/147 (30.6 %) of women with three or more partners had high vaginal pH, compared to 44/193 (22.8 %) women with less than three lifetime partners ($p = 0.039$).

Since the only *p*-values below 0.1 were found within the groups presenting on different consultations (prenatal vs. gynaecology, infertility or family planning), multivariate analysis was not considered appropriate.

As shown in Fig. 1, self-assessed vaginal pH correlated very well with the wet mount findings lactobacillary grade (AVF, scores 1–4) and the AV (1–10) and BV scores (1–3). Similarly, self-measured pH also correlated very well with the mean Nugent score on Gram-stained specimens: the Nugent score was 1.14 ± 2.1 for $\text{pH} < 4.4$, 3.5 ± 3.6 for $\text{pH} 4.5\text{--}4.7$ and 5.2 ± 3.3 for $\text{pH} > 4.7$ ($p < 0.0001$, Table 2).

The prevalence of AVF was 146/338 (43.2 %), ranging from 15.1 % in women with low pH to 45.5 % in women with intermediate pH and 77.0 % in women with high pH (Table 2). BV (Nugent 7–10) was present in 84/338 (24.9 %) women,

Table 1 Demographics of the 344 Ugandan women with normal, intermediate and elevated vaginal pH. Complete data on age and tribe region were missing for five women. The partner was present at the consultation in only four women (0.8 %)

| | pH < 4.4, n = 107 | pH 4.5–4.7, n = 145 | pH > 4.7, n = 87 | p-Value |
|--|-----------------------|-----------------------|----------------------|--------------------|
| Age ^a | 27.53 ± 5.31, n = 101 | 28.72 ± 6.13, n = 137 | 28.51 ± 6.64, n = 83 | 0.9 |
| Parity | 1.86 ± 1.65, n = 105 | 2.1 ± 1.59, n = 144 | 2.14 ± 1.73, n = 87 | 0.4 |
| Tribe region | n = 107 | n = 145 | n = 87 | |
| Central | 50 | 68 | 52 | 0.1 |
| East | 11 | 14 | 5 | 0.5 |
| West | 17 | 25 | 6 | 0.09 |
| South | 4 | 3 | 2 | 0.7 |
| North | 6 | 14 | 5 | 0.4 |
| Other | 19 | 21 | 17 | 0.8 |
| Educational level | n = 106 | n = 145 | n = 86 | |
| <6 years | 5 (4.7 %) | 3 (2.0 %) | 4 (4.6 %) | 0.5 |
| 6–12 years | 20 (18.9 %) | 34 (23.4 %) | 24 (27.9 %) | |
| 12–18 years | 50 (47.2 %) | 65 (44.8 %) | 40 (46.5 %) | |
| >18 years | 31 (29.2 %) | 43 (29.6 %) | 18 (20.9 %) | |
| Region | n = 106 | n = 144 | n = 86 | |
| Rural | 23 (21.7 %) | 40 (27.8 %) | 24 (27.9 %) | 0.8 |
| Semi-urban | 43 (40.6 %) | 51 (35.4 %) | 30 (34.9 %) | |
| Urban | 40 (37.7 %) | 53 (36.8 %) | 32 (37.2 %) | |
| HIV-positive | 9/104 (8.7 %) | 20/139 (14.4 %) | 11/84 (13.1 %) | 0.039 ^c |
| Age when having first child | 20.2 ± 4.29, n = 83 | 20.0 ± 3.83, n = 123 | 19.6 ± 4.69, n = 71 | 0.9 |
| Age when first had sex | 17.79 ± 3.13, n = 104 | 17.76 ± 2.79, n = 142 | 16.92 ± 2.91, n = 85 | 0.9 |
| Visited traditional healer | 10 (9.4 %), n = 106 | 10 (6.9 %), n = 144 | 6 (6.9 %), n = 87 | 0.5 |
| Previous lifetime sexual partners | n = 106 | n = 145 | n = 89 | |
| Less than 3 | 69 (65.1 %) | 80 (55.2 %) | 44 (49.4 %) | 0.039 ^b |
| 3 or more | 37 (34.9 %) | 65 (44.8 %) | 45 (50.5 %) | |
| Vaginal douching during last 2 months ^d | n = 106 | n = 145 | n = 87 | 0.15 ^b |
| >1/day | 76 (71.7 %) | 107 (73.8 %) | 72 (82.8 %) | 0.3 |
| Infrequent | 10 (9.4 %) | 16 (11.0 %) | 2 (2.2 %) | 0.3 |
| Never | 20 (18.9 %) | 22 (15.2 %) | 13 (14.9 %) | Ref. |
| Contraception | n = 107 | n = 145 | n = 87 | |
| No contraceptive ^e | 32 (29.9 %) | 57 (39.3 %) | 32 (36.8 %) | Ref. |
| Pill | 3 (2.8 %) | 6 (4.1 %) | 5 (1.1 %) | – |
| Condoms | 7 (6.5 %) | 10 (6.9 %) | 3 (3.4 %) | 0.3 |
| Copper IUD | 3 (2.8 %) | 2 (1.4 %) | 3 (3.4 %) | – |
| Hormonal IUD | 0 | 1 (0.7 %) | 0 | – |
| Depo-Provera injection | 5 (4.7 %) | 12 (8.3 %) | 9 (10.3 %) | 0.8 |
| Norplant | 3 (2.8 %) | 3 (2 %) | 0 | – |
| Collection group | n = 107 | n = 145 | n = 87 | |
| Antenatal | 61 (57.0 %) | 52 (35.9 %) | 31 (35.6 %) | Ref. |
| Family planning | 32 (29.9 %) | 75 (51.7 %) | 40 (46.0 %) | 0.0027 |
| Infertility | 1 (0.9 %) | 7 (4.8 %) | 10 (11.5 %) | 0.01 |
| Gynaecology | 13 (12.1 %) | 11 (7.6 %) | 6 (6.9 %) | Ref. |

^a Twenty-three women did not know their exact age or did not want to disclose

^b Chi-square for trend

^c Chi² for pH < 4.4 versus pH ≥ 4.5

^d Infrequent vaginal douching was reported as follows: once/day (n = 9); few times/week (n = 5); once/week (n = 8); now and then (n = 3); only after menses (n = 3); none, due to no partner

^e Reasons for no contraception: none, want to become pregnant; none, due to other reason; calendar method; temperature method; coitus interruptus; sterilisation woman/man

corresponding well with the number of BV women diagnosed by the wet mount technique [84/338, of which 63/338 (18.6 %) had full-blown BV and 21/338 (6.2 %) partial BV]. Full-blown BV was found in 34.5 % of women with pH above 4.7 and in 3.8 % of women with pH < 4.5 ($p < 0.001$). Partial BV was found with an almost equal likelihood in the group with pH 4.5–4.7 (7.5 %) as in the group with pH above 7.7

(8.1 %), which is higher than in the low pH (pH < 4.5) group (2.8 %, $p = 0.08$, $p_{\text{trend}} = 0.03$). This was different from the prevalence of intermediate flora on Gram stains, which was similar in all pH groups.

Compared to Nugent 7–10 as the gold standard, the sensitivity (Se) of increased pH (pH ≥ 4.5) was 95.2 % (80/84) and the specificity (Sp) was 40.6 % (103/254). The negative

Mean microscopy score according to vaginal pH

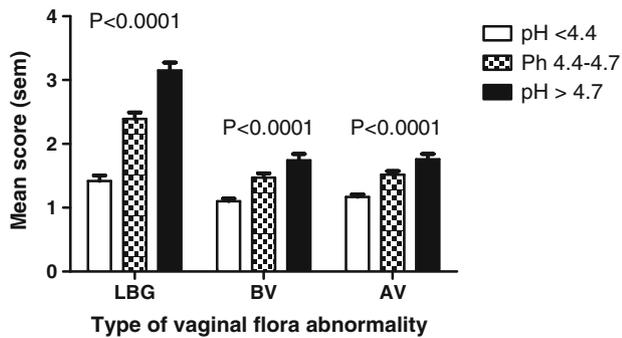


Fig. 1 Relation of self-measured vaginal pH with microscopic flora abnormalities in 338 unselected Ugandan women

(NPV) and positive (PPV) predictive values were 96.3 % (103/107) and 34.6 % (80/231), respectively, indicating a high false-positive test rate of 65.4 %. If a pH cut-off of 4.7 was chosen, the Se was 45.2 % (38/84), Sp 81.7 % (205/254), NPV 81.7 % (205/251) and PPV 43.6 % (38/87), still implicating a high false-positive rate of 56.3 %.

Moderate AV was present in 38/338 (11.2 %) and severe AV in 11/338 (3.3 %). Most cases of severe AV were present in the elevated pH group: 9.2 % in pH>4.7, 2 % in the intermediate pH group and none in the low pH group of pH<4.5 (*p*<0.001). For moderate to severe AV, a similar trend is seen with high pH: 17.2 %, 7.6 % and 0.9 % AV presence in high, moderate and low pH groups, respectively (*p*<0.001).

If pH above 4.4 was to be used as a method of selection for further screening, 87.3 % of AVF, 91.7 % of BV and 96.3 % of AV would have been discovered. On the other hand, this type of screening would require that about two-thirds of women would have to be screened (68.6 %) in order to detect about 90 % of abnormalities. Increasing the threshold to pH>4.7 would lower the sensitivity and detect only 50.7 % of AVF, 44.0 % of BV and 55.6 % of AV.

As a potential confounding factor, the presence of sperm was analysed. In the group with pH<4.5, sperm was detected in 10/106 (9.4 %) women, in the group with pH 4.5–4.7 in 20/148 (13.5 %) women and in the group with high pH above 4.7 in 16/93 (15.4 %) women. Although a trend was noted, the difference between groups was not significant (Chi-square for trend: 2.4, *p*=0.1).

Discussion

In low-resource countries with a high prevalence of all the above medical conditions, screening and treating AVF and BV could be an advantageous and cost-beneficial approach. In order to be successful, such a screening test has to be widely accessible, affordable, easy to perform, accepted by the target population and efficacious. AVF, especially BV, is a particular risk factor for adverse outcome in pregnant women, and for acquiring sexually transmitted infections (STIs) in non-pregnant women. Screening for such abnormalities is of major relevance in high-risk populations.

Because of its simplicity, accessibility and low cost, self-testing for vaginal pH indicator strip could be the most ideal substitute test for high-risk flora. Over 85 % of Ugandan women were keen on testing themselves, did not find it distressing or difficult to understand and HIV-positive patients were even more motivated [13]. In the present study, we tested the relevance of this method.

We found a strong correlation between self-measured pH and the severity score of AVF, BV and AV. The rates of AV (11.2 %) and BV (24.8 %) flora together roughly accounted for almost all cases of AVF (37.1 %). Although the vast majority of women with pH above 4.7 had AVF on microscopy, only 43 % of them had BV. If the cut-off was set at a pH of 4.5, 56 % had AVF and 37.5 % had BV. With this lower cut-off, the specificity of finding BV was also very low: only roughly 4

Table 2 Correlation of self-assessed pH with rehydrated wet mount microscopy (AVF, BV, AV) and Gram stain findings (Nugent) in 338 Ugandan women presenting at different gynaecology units in Maloo University Hospital in Kampala

| Vaginal pH by self-assessment | pH < 4.4, n = 106 | pH 4.5–4.7, n = 145 | pH > 4.7, n = 87 | <i>p</i> -Value |
|-------------------------------|-------------------|---------------------|------------------|-----------------|
| Rehydrated wet mount | | | | |
| AVF (LBG 0-IIb-III) | 16 (15.1 %) | 66 (45.5 %) | 64 (77.0 %) | <0.001 |
| Partial BV | 3 (2.8 %) | 11 (7.5 %) | 7 (8.1 %) | <0.03 |
| Full BV | 4 (3.8 %) | 29 (20 %) | 30 (34.5 %) | <0.001 |
| AV score >6 | 0 | 3 (2.0 %) | 8 (9.2 %) | <0.001 |
| AV score >4 | 1 (0.9 %) | 11 (7.6 %) | 15 (17.2 %) | <0.001 |
| Gram stain | | | | |
| Nugent 1–3 (NI) | 91 (85.0 %) | 84 (58.3 %) | 33 (37.9 %) | <0.001 |
| Nugent 4–6 (IM) | 12 (11.2 %) | 18 (12.5 %) | 16 (18.4 %) | 0.3 |
| Nugent 7–10 (BV) | 4 (3.7 %) | 42 (29.2 %) | 38 (43.7 %) | <0.001 |
| Mean Nugent score | 1.14±2.12 | 3.46±3.58 | 5.23±3.32 | |

AVF: abnormal vaginal flora; LBG: lactobacillary grades; BV: bacterial vaginosis; AV: aerobic vaginitis; NI: normal flora; IM: intermediate flora (Nugent)

out of 10 of women indeed had BV (Nugent 7–10), indicating a false-positive rate as high as 65 %. Increasing the cut-off to a pH above 4.7, this specificity rose to 82 %, but at the expense of a poor sensitivity of 45 %, and still a high false-positive rate of 56 %. In other words, in this central African society, the finding of an increased pH is indicative of BV in only less than half of the cases.

So, not only are there reasons other than AVF to cause increased vaginal pH in Ugandan women (especially in the group with borderline pH elevations), but, also, BV is clearly not the only infectious abnormality that causes AVF and increased pH. Of other abnormalities, AV contributes an important part. Like BV, AV is an important risk factor in pregnancy [18, 19]. Also, outside pregnancy, studies indicate that AV is more prevalent than originally thought [20], and may be a risk factor for worsening cervical dysplasia [21] and acquisition of STIs in sex workers [22, 23]. As awareness for this condition is increasing, new therapies are being tested [24–27] and more data on its prevalence and pathogenicity will become available over time.

The positive predictive value of $\text{pH} \geq 4.5$ to detect AVF of 53 % indicates that increased pH may point to conditions other than BV or AV, such as frequent or recent sexual intercourse. Although we did not ask for the frequency or timing of intercourse, we discovered a direct relation between the number of lifetime sexual partners and vaginal pH, and a trend was seen towards increased presence of sperm in the group with higher pH. In this population, most women perform vaginal douching, the majority even on a daily basis. Whether this practise is responsible for the high prevalence of AVF and increased vaginal pH is not certain and, in this study, we could not discover any relation between the frequency of douching and the vaginal pH. Neither was age, parity, tribal origin, area of living, use of contraception or level of education associated with increased pH.

HIV-positive women were more likely to have an increased pH, which fits with the fact that AVF and BV are risk factors for acquiring HIV, especially if women also engage in high-risk sexual behaviour [28]. Remarkably, women seeking advice in a fertility clinic or in a family planning unit were at higher risk for having abnormal vaginal pH as compared to women attending general gynaecology or antenatal clinics. As BV becomes less frequent and the pH progressively decreases as pregnancy advances [29], it may explain the higher rate of low pH found in antenatal patients. The higher pH encountered in the family planning clinic may be due to the proportionally high number of postpartum patients, who were sent there as a part of normal routine after delivery, so occult vaginal bleeding and breastfeeding could be explanatory factors. In women attending fertility clinics however, the finding of increased pH and AVF/BV could be worrisome. Implantation of the fertilised egg is less efficient in women with AVF [30], BV [31–33] and AV [34], and the presence of these

abnormalities can be a sign of genital infections leading to cervicitis, endometritis or tubal damage [35]. Hence, we would strongly recommend routine vaginal pH testing of infertile women in Africa, followed by further genital testing if the pH is increased.

To be used as a general screening tool for high-risk conditions of the vagina, pH screening by self-assessment of pH of 4.5 or more would detect 90 % of at-risk women, is cheap and easy to perform, and well accepted by Ugandan women. Furthermore, besides BV, around 40 % of other pathologies, such as AV, would be discovered in these women with AVF. Given the high prevalence of AVF conditions in these Uganda women, almost two-thirds of women would have to be further examined, unless one is willing to accept an inferior sensitivity of the screening by choosing higher pH cut-offs.

In conclusion, screening by self-testing for abnormal vaginal pH is a feasible option to detect high risk for AVF, BV and AV in Ugandan women. It is acceptable, cheap and efficient, although due to the high prevalence of AVF, more than half of all women would need more testing, in order to maintain good sensitivity of the screening test. Women with HIV or consulting for infertility require extra attention for vaginal flora abnormalities and should be tested routinely for it.

Author contributions GD, JL, JB and AG participated in the design of the study.

FD, TM, NE and AG conceived the study, participated in its design and coordination, and helped to draft the manuscript.

CM, GD and GB performed the microscopic examinations and helped to draft the manuscript.

All authors read and approved the final manuscript.

Compliance with ethical standards

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Conflict of interest None of the authors declares any conflict of interest.

Ethical approval The study was approved by the ethical committee of the Kampala University Hospital in November 2008.

Informed consent Written informed consent was signed by all patients before engaging in the study.

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