INCIDENCE OF GARDNERELLA VAGINALE IN NON SPECIFIC VAGINITIS

By

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SUMMARY

The present study was undertaken to see the incidence of Gardnerella vaginalis in clinically diagnosed cases (N 770) of non-specific vaginitis and age matched controls (N 380). Semi-quantitative assessment of G. vaginalis with severity of disease was also made along with the antimicrobial sensitivity pattern of the isolates.

High vaginal swabs of 770 women clinically diagnosed as non-specific vaginitis and 380 age matched healthy women were investigated by microscopy, examination of pH of vaginal secretion, Amine Test and Culture examination. The isolates were identified and tested for the antibiogram. G. vaginalis was isolated in 180 (23.4%) study cases as a pure/predominant organism and from 12 (3.2%) in control group. The difference was statistically significant ($p < 0.05$).

In the culture positive cases an acidic vaginal pH was observed in 91.0%, presence of small gram negative bacilli in 81.0%, clue cells in 61.0% and a positive Amine Test in 31.0%. The antimicrobial agent found to be consistently most effective (in vitro) against G. vaginalis was chloramphenicol.

Introduction

The commonest cause of non-trichomonal, non-monilial sexually transmitted vaginitis is Gardnerella vaginale, a pleomorphic Gram negative bacillus. Gardnerella vaginale has been widely described as a common cause of infectious leukorrhoea in women (Akerlund and Mardh, 1974; Gardner and Dukes, 1955). Despite numerous studies the role played by G. vaginale is still uncertain. This present prospective study was undertaken to re-evaluate the relation of G.vaginale in leukorrhoea and in an age matched control group. The in vitro susceptibility of G.vaginale to antibiotic is also considered.

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Material and Methods

A total of 770 women in the reproductive age group, clinically diagnosed as cases of non-specific vaginitis (NSV) and 380 age matched controls attending OPD for complaints other than leukorrhoea,

participated in the study. None received any antibiotics or vaginal medication during preceding month. The diagnosis of non-specific vaginitis was made when the secretion did meet at least two of the following criteria (i) thin grey discharge (ii) pH of discharge > 4.5, (iii) positive amine test (iv) presence of clue cells (Spiegel *et al*, 1980).

High vaginal swabs were collected. The pH was measured by the bedside and some secretion was collected for microscopic and bacteriological study. The wet coverslip and Gram stain preparations were studied for the presence of clue cells, bacterial clumps, leukocytes and other significant findings. Amine test (Balsdon *et al*, 1980) was carried out by adding a drop of 10% KOH to the wet mount preparation and immediate emittence of fishy odour was noted.

Vaginal swabs were innoculated onto vaginalis agar (v-agar) as recommended by Greenwood *et al* (1976), sheep blood agar and MacConkey's medium. The culture plates were incubated at 37°C in the presence of 5-10% CO_2 for 48 hours.

G.vaginalis was identified on the basis of colonial morphology on V-agar plates, failure to produce catalase and oxidase, ability to acidify glucose, maltose and starch within 4 hours in Buffered single substate (BSS) test, ability to hydrolyse hippurate and characteristic morphology on Gram's staining (Greenwood *et al* 1976; Hwang and Ederer 1975).

Quantitation of the relative numbers of the G.vaginalis in both patient and control was done assigning the cultures to one of four classes viz., class I—pure cultures; Class II—predominant organism; Class III—many organisms and Class IV—few organisms (Greenwood *et al*, 1976).

The other associated organism when isolated as pure or predominant flora were identified by standard procedures (Cruickshank *et al*, 1975).

Results

The percentage incidence of G.vaginalis found in each group are shown in Tables I and II.

Of the 770 cases of vaginitis 44 (57%) yielded Candida albicans, 12 (1.6%) showed T.vaginalis and in 2 more G.vaginalis was associated with T.vaginalis.

TABLE I

Causative Organisms in Non Specific Vaginitis (n 770) and Matched Control (n 380)

Organism isolated Microscopy	Cases Study group (%)	Controls (%)
G. vaginalis only	180 (23.4)	12 (3.2)
Candida albicans	44 (5.7)	17 (4.5)
T. vaginalis	12 (1.6)	5 (1.3)
T. vaginalis (Microscopy)	2 —	— —
Candida albicans and T. vaginalis (Microscopy)	2 —	— —
G. vaginalis + Group B Streptococcus	1 —	— —
Staph. aureus	4 (0.5)	— —
Esch. coli	9 (1.2)	3 (0.8)
Group B Streptococcus	6 (0.9)	— —
Klebsiella sp.	2 —	2 (0.5)
Proteus sp.	1 —	— —

TABLE II

Types of cases	No.	Incidence of G. vaginalis
Women with N.S. vaginitis	770	180 (23.4)
Pregnancy	180	6
IUCD	65	2
On pills	60	2
Undergone tubectomy	55	1
Infertility/sterility group	20	1

Figures in parenthesis shows percentage.

Thus 58 cases were excluded from the present study as they do not fall into the category of NSV. Rest were investigated further. In the study group commonest organism isolated was G.vaginalis (23.4%), followed by Esch.coli (1.2%) and group B streptococcus (0.8%) and Staph aureus (0.5%). G.vaginalis was isolated in pure culture (class I) from 152 (84.4%) patients and as predominant flora (class II) in 28 (15.5%) patients. Amongst the control group who attended Gynaec outpatients for reasons other than vaginitis, 12 (3.2%) women harboured G.vaginalis. Three of these isolates fell in Class II and remaining 9 isolates were recovered in insignificant numbers (Class IV).

Results of the present study showed that clue cells were present 110/180 (61.1%). These are also observed in 9 (1.9%) other instances where bacteria other than G.vaginalis were isolated. Clue cells were also observed in 2 specimen from control group. An acidic PH (4.5-5.5) of the vaginal secretion was noted amongst 91.5% of culture positive cases, comparing well with the results of Gardner and Dukes (1955). Characteristic gram negative 'bacilli/cocobacilli were observed in 80% of samples in the study group.

Results of the in vitro antimicrobial susceptibility testing of G. vaginalis isolates shows that the organism is most sensitive to chloramphenicol followed by Gentamycin, Cotrimoxazole and Kanamycin. A limited number of isolates were tested against 2-hydroxymetronidazole and is found to be sensitive in 89% of isolates as against the parent Metronidazole (50.5%) compound (Table III).

Discussion

G. vaginalis has held a controversial position ever since its detection by Leopold in 1953. The available vast literature leaves no doubt that it causes bacterial vaginosis. But there is still some uncertainty as to what extent it contributes or is associated with non-specific vaginitis. The most important controversy surrounding G. vaginale vaginitis has to do with a possible etiological role of frequently associated anaerobic bacteria (Pheifer *et al*, 1978; Spieget *et al*, 1980). Recent reports of isolating G. vaginalis from a significant percentage of women exhibiting no evidence of infection have stirred yet another controversy (Homes *et al*, 1981). According to Gardner (1983) whenever the organism is found in the undouched, untreated patient it is always predominant in a very high ratio over every other

TABLE III

Antimicrobial Spectrum of G. Vaginale Isolated from Patients (n 180) and Control (n 12)

Antimicrobial agent	U/ug/disc	Number of strains sensitive	
		Cases (n 180)	Controls (n 12)
Ampicillin	10	120 (66.6)	6 (50.0)
Chloramphenicol	30	167 (92.7)	9 (75.0)
Cephalexin	30	120 (66.6)	9 (75.0)
Co-trimoxazole	25	128 (71.1)	7 (58.2)
Gentamicin	10	130 (72.2)	8 (66.6)
Kanamycin	30	128 (71.1)	7 (58.2)
Penicillin	10	110 (61.1)	6 (50.0)
Streptomycin	10	121 (67.2)	4 (33.3)
Tetracyclin	30	130 (72.2)	9 (75.0)
Metronidazole	5	91 (50.5)	5 (41.5)
Hydroxymetronidazole Tested against 58 strains	5	48 (83.0)	Not done

Figures in parenthesis shows percentage.

bacterial agent and that clinical features will present or develop.

The results of the present study demonstrate that G. vaginalis was much more commonly isolated and in higher concentration from patients with non-specific vaginitis than from other clinical group. Other aerobic organisms were also present in cases of nonspecific vaginitis, but in none of the cases were they isolated with G. vaginalis. The isolation rate in asymptomatic patient is not only low than in the patients with NSV but the concentration of the organisms isolated is much lower in control. The difference is statistically significant ($p<0.05$).

According to Gardner (1980) a reliable test in the absence of culture is provided by presence of clue cell and an accuracy rate of 90% is not unusual. However other investigators did not find such correlation (Bhujwala *et al*, 1985; Chattopadhya and Teli, 1984). Results of this study showed that 110/180 (61.1%) of culture positive specimens had clue cells. Amine test was not found useful, being positive in 31.0% only. An acidic pH (4.5-5.5) of the vaginal secretion was noted amongst 91.5% of culture positive cases in the present study, comparing well with the results of Gardner and Dukes (1955). The results thus suggest isolation of the organism in appropriate medium is necessary to ascertain the etiological significance.

Antibiotic sensitivity pattern was studied and chloramphenicol was found to be the drug of choice. The efficacy of 2-hydroxymetronidazole was tried with good results (Ralph, 1983).

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