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Effects of chlorhexidine-coated intrauterine device on the bacteriology of the uterine cavity

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Abstract

Twenty-nine women, aged 31–53 years, scheduled for an abdominal hysterectomy, participated in this study. The patients were divided into three groups. The first received a Multiload MLCu250 intrauterine device (IUD); the second group received a chlorhexidine acetate medicated Multiload MLCu250 IUD; the third group acted as a control group receiving no IUD.

Bacteriological cultures of the vagina and ectocervix were taken prior to insertion of the IUD. At hysterectomy, some 18 hours later, specimens were taken from the cervical canal, uterine cavity, and the device itself, for culture of anaerobic and aerobic organisms using a standardized previously validated technique.

In three of the nine control patients, organisms were cultured from the uterine cavity. There were no differences between the bacteriological results of the two groups with medicated and non-medicated devices, with regard to the cultures from the cervical canal, uterine cavity or the devices themselves. Nor was there any difference between the control and the IUD group.

Introduction

The use of intrauterine devices (IUDs) has been associated with some serious side effects such as bleeding, pain, accidental pregnancy, expulsion and pelvic inflammatory disease (PID) [1]; yet it is pelvic inflammatory disease that has caused the most controversy. The association between copper IUD use and PID in terms of relative risk is probably equal to, or at worst only a little greater than controls and confined to the first few months following insertion [2,3]. The bacterial contamination of the uterine cavity which occurs at the time of insertion of the device is well documented, yet seems to decline with time, reaching zero, in one study, 30 days following insertion [4].

Certainly, what risk there is of PID seems highest during the first four months after insertion [5]. After this time, the risk of PID is no greater than that for non-contraceptors [5]; any increased incidence of PID thereafter is the result of subsequently acquired sexually transmitted disease.

The role of the IUD tail remains unclear; claims of decreased infection rates in tailless devices [6] have not been confirmed [7,8], while recent device specific studies seem to indicate a lower rate of PID in relation to copper-medicated devices, when compared to non-medicated devices [2,3,9,10].

In an effort to overcome the infection risk at the time of insertion, it seems logical to produce an antiseptic-releasing IUD. Chlorhexidine is an appropriate antiseptic compound; it is active against the organisms causing PID, e.g. *Neisseria gonorrhoeae, Chlamydia trachomatis* and other potential pathogens found in the vaginal flora. It is also active against yeasts (*Candida albicans*), a range of viruses, and protozoal species (*Trichomonas vaginalis*) [11].

The present study was undertaken to determine if a medicated IUD would materially affect the bacterial contamination of the uterine cavity at the time of insertion of an IUD and thus, by implication, affect the incidence of PID in the first few months of IUD usage. Ethical approval was granted by the committees of the hospitals involved, and written informed consent was obtained from all participating volunteers.

Materials and methods

Twenty-nine parous female volunteers aged 31–53 years underwent abdominal hysterectomy for menorrhagia or small fibroids less than 2 cm in diameter. They had had no intrauterine procedure, nor had they used any antibiotics in the previous two months. These women were selected and randomized into three groups. One group received a non-medicated IUD. A second group received a medicated IUD coated with 2 mg of chlorhexidine acetate (CHA) of which 1/3 on the cross arms would be released on insertion and the remaining 2/3 on the stem would be released on removal of the insertion tube. The third group, the controls, received no IUD. All IUDs were the Multiload MLCu250.

Immediately prior to the insertion of the IUD, the cervix was exposed with a non-lubricated sterile speculum and samples for culture taken from the vaginal vault and ectocervix. The swabs taken were immediately placed in Stuart's transport medium and a viral transport medium. The vagina and cervix were then cleansed with 5% w/v chlorhexidine gluconate (Hibitane^{R}). The anterior lip of the cervix was grasped with the tenaculum and the Multiload was carefully inserted into the uterus without prior sounding of the uterine cavity. The control group was treated in the same fashion but no IUD was inserted.

No pre-operative vaginal antisepsis was used; great care being taken to ensure that no antiseptic leaked back into the vagina at catheterization. Once removed, the uterus was placed in a sterile kidney dish and transferred to a sterile towelled trolley containing the sterile sampling instruments. The trolley had been prepared using strict

aseptic technique; the scalpels and needles had been readied using a no touch technique.

The anterior uterine wall was then incised with a sterile scalpel from the fundus to the external os. Great care was taken not to contaminate the cavity with any cervical organisms. Fundal placement of the IUD was noted before the IUD was removed with sterile forceps and cut into sections, the arms being cultured separately from the stem, each cut being made with separate sterile instruments. The sections were placed in 2 ml of nutrient broth containing reducing agent, anaerobic indicator and chlorhexidine inactivator. Using a series of identical 6 mm tissue borers, with surface areas of approximately 30 mm^2, four samples were taken from the posterior wall of the cavity (UC 1, 2, 3 & 4) and two from the cervical canal (CC1 & CC2); none were taken from the junction. Sampling began at the fundus and proceeded cervically using a different set of instruments for each sample; positions of each sample were carefully documented (see Figure 1). The 'mucosa' was separated from the underlying tissue and placed in 9 ml of lecithin 'Lubrol' broth containing 10% glycerol and 0.3 g/L of sodium sulphoxylate. A swab of both cavity and cervical canal was taken and placed in viral transport medium for subsequent chlamydial culture. The samples were then transported immediately to the laboratory in a sterile container. The sampling technique described above is that of Sparks *et al.* [12].

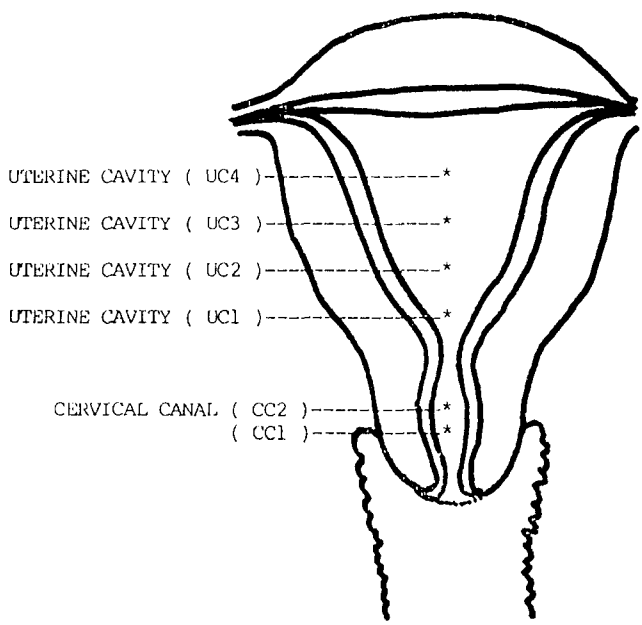

Figure 1 Site of samples from uterus and cervical canal

On receipt, each sample was homogenized, using Griffiths tubes or, if fibrous, an Ultra-Turrax (Sartorious^{R}) homogenizer. Aliquots of 500 μl, 100 μl and 10 μl were transferred to 3 separate plates of each agar medium and distributed with a glass spreader. The IUD sections in nutrient broth were vortex mixed for 1 minute and agar plates were inoculated with aliquots of 100 μl and 10 μl. All samples were cultured on the following media and incubated as indicated:

(a) Blood agar
(b) MacConkey agar
(c) MacConkey agar with colistin 1 mg/L
(d) Malt agar

All were cultured aerobically for 48 h.

(e) Lysed blood agar with vancomycin 1.2 mg/L, colistin 7.5 mg/L and trimethoprim 3 mg/L
(f) Chocolate agar
(g) Blood agar with colistin 1 mg/L and kanamycin 10 mg/L

All were incubated in increased CO_2 for 48 h.

(h) Blood agar
(i) Rogosa agar
(j) Lysed blood agar with menadione and kanamycin 70 mg/L
(k) Naladixic acid 'tween' agar

All were incubated anaerobically for 5 days

All media were incubated at 37°C except malt agar, which was incubated at 30°C. The organisms cultured were identified by standard laboratory techniques.

Results

Twenty-nine patients were included in this study. In those subjects receiving a device, a mean time of 17 h 43 min elapsed prior to hysterectomy, the range being 14:00–23:30 h. Before insertion of the IUD, bacteria were isolated from cultures of the vaginal vault and the ectocervix in all patients. The organisms most commonly found are shown in Table 1.

As one might expect, *Lactobacillus* species was isolated most frequently from the patients. Surprisingly, no chlamydial species were isolated.

Table 2 shows the abbreviations used in expressing the results of the three groups of patients; the groups comprising 10 patients each in Groups A and B, 9 patients in Group C. The results are summarized in Table 3, which gives the number of positive and negative cultures by patients in the three groups. A negative result indicates no growth on the media plates used. A positive result indicates one or more species of

bacteria grown, at one or more sites. Tables 4 and 5 give the bacterial species isolated in the control, standard IUD and medicated IUD groups.

Table 1 Organisms isolated from the vagina and ectocervix preinsertion of the IUD

Organism	*Vagina*	*Ectocervix*
Lactobacillus species	16	14
Bacteroides species	9	8
Staphylococcus albus	9	8
Streptococcus faecalis	10	8
Diphtheroid species	8	7
Bacteroides melaninogenicus	7	7
Peptostreptococcus species	6	4
Escherichia coli	6	3
Candida albicans	5	4
α-Haemolytic *Streptococcus*	4	4
Others*	16	15

*Bh (Lance), Bif, Clos, Cs, Gv, Nhs, Pros, Sa, Ye: see Table 2

Table 2 Abbreviations for bacterial species seen in Tables 3, 4 and 5

Ac	Anaerobic corynebacterium	Gv	*Gardnerella vaginalis*
Ansbb	Anaerobic spore-bearing bacilli species	Hsa	α-Haemolytic streptococcus
		Ls	*Lactobacillus*
Asbb	Aerobic spore-bearing streptococcus	Ms	Microaerophilic
		Ne	*Neisseria gonorrhoeae*
Bh (Lance)	Lancefield group B β-haemolytic streptococcus	Nhs	Non-haemolytic streptococcus
		Pros	*Proteus* species
Bif	*Bifidobacterium* species	Prs	*Proprionibacterium* species
Bm	*Bacteroides melaninogenicus*	Ps	*Peptostreptococcus* species
Bs	*Bacteroides* species	Rr	*Rhodotorula rubra*
Ca	*Candida albicans*	Rs	*Rhodotorula* species
Chlam	*Chlamydia trachomatis*	Sa	*Staphylococcus aureus*
Clos	*Clostridium* species	Sal	*Staphylococcus albus*
Cs	*Corynebacterium* species	Sarl	*Sarcina lutea*
Dh	Group D β-haemolytic streptococcus	Sars	*Sarcina* species
		Sf	*Streptococcus faecalis*
Dip	*Diphtheroid* species	Ss	*Streptococcus* species
Ec	*Escherichia coli*	Ye	Yeasts
Eus	*Eubacterium* species		

Table 3 Positive and negative culture results in all groups

Group	*Total number of patients*	*All specimens negative*	*Patients who had one or more positive samples*			
			Number of patients	*Distribution of positive culture(s)* *Cervix*[a]	*Uterus*[b]	*IUD*[c]
C (control)	9	5	4	2	2	NR
				1	0	NR
				0	1	NR
A standard IUD	10	4	6	2	2	2
				1	1	0
				1	0	0
				0	2	0
B Chlor-hexidine IUD	10	2	8	1	1	1
				2	2	0
				2	0	0
				0	1	1

NR = IUD not required in control group
[a]Two sampling sites in cervix (see text)
[b]Four sampling sites in uterus (see text)
[c]Three sampling sites on IUD (see text)

Table 4 Positive uterine culture results in three group C (control) patients after culture at all sites (UC 1,2,3 & 4)

Patient trial number	*Pre-op culture species**	*Post-op site of positive culture*	*Post-op species**
30	Dip	UC1 & 3	Sal
	Bs		
	Ps		
40	Sal	UC1, 3 & 4	Prs
	Ps		
	Ls		
	Bm		
	Bs		
	Nhs		
43	Ca	UC4	Ca

* For full name of species see Table 2 for abbreviations used

Table 5 Positive cultures Groups A and B by site and species*

Patient trial number	*Pre-operative culture*	*Positive culture species and post-op site*			
		Uterus culture site	*Species*	*IUD culture site*	*Species*
Group A					
04	Ls** Hsa Ca Clos	UC1 & 2 UC4	Ls Sa	Stem, tail Arm	Ls Hsa
14	Ls Sal Ca Dip Bm	UC1 & 2 UC3 UC4	Pros Sf Pros & Sf	Arm	Pros
23	Ls Clos	UC1 & 4	Sf	None	–
38	Sal Ls	UC2	Sal & Sars	None	–
39	Sf Sal Ps Dip Bm Ls	UC3	Sf	None	–
Group B					
11	Dip Ss	UC1, 2 & 3	Sf Dh	Stem, arm & tail	Dh
	Gv	UC2	Sf	stem	Sarl
28	Sf Bh(Lance) Bs Bif Bm Ec Ls Ps	UC3	Sf Sal	tail	Sf
09	Ec Ca Sf Ls	UC1	Sal	None	Nil
21	Ls Bm	UC3	Sal	None	Nil
34	Sal Hsa	UC2	Sal Dip	None	Nil
42	Sf Sal	UC2	Sal	None	Nil

* See Table 2 for abbreviations used

** Organisms underlined were present at more than one site

Note: In the interest of space this table summarizes the data. Those requiring the full data can obtain the transcript by sending an SAE to Professor Newton

Group C

In the control group five of the nine patients had negative cultures. In the four patients with positive cultures, three had positive cultures in the uterine cavity, all of which were considered to represent true colonization rather than contamination. The finding of *Candida* at the uterine fundus in patient 43 was unexpected in the face of negative cultures in all other postoperative samples. However, repeat culture confirmed a *Candida* isolate, and *Candida* was found pre-operatively in both the vagina and ectocervix in this case.

Groups A and B

Comparing the IUD groups, four out of the ten standard IUD users showed positive cervical cultures, compared to five out of ten in the medicated IUD group. In the uterine cavity samples, five of the ten were positive in the standard IUD group A, compared with six in the medicated IUD group B. This was statistically not significant from controls, $\chi^2 = 0.0725$, p 0.5 and $\chi^2 = 0.0259$, p 0.5 (Yates correction applied), respectively. There was also no difference between the two IUD groups. Only two of the ten devices in each group showed positive cultures (see Table 5).

Discussion

This study has once again raised the question of whether the normal uterine cavity is sterile. Our control results are contrary to the earlier work of Sparks *et al.* [12], despite the use of the same sampling methodology and the active involvement of the author in this study. There are, however, differences between the two studies. The use of 5% chlorhexidine gluconate solution preinsertion, in our study, may have *reduced* the bacterial contamination of the uterine cavity if, as has been shown with bovine mucus and chlorhexidine diacetate [13], cervical penetration of the chlorhexidine (gluconate) used in this study had taken place, but had not increased it, as was found. In addition, a fixed number of cavity samples were taken in our study (this number being greater than in previous studies). These differences alone may account for some of the discrepancy. However, a greater variety of culture media were used in the present study, compared with that of Sparks *et al.* [12], and the anaerobic media were incubated without interruption for longer. This may have allowed a small increase in the number of fastidious organisms to be isolated. In only one of the three control patients with uterine organisms, from whom *Proprionibacterium* species was isolated, could the organism be described as fastidious. The other two were *Staphylococcus albus* and *Candida albicans*, which are both easily cultured organisms. The finding in the patient with *Candida albicans* from a single uterine sample is difficult to interpret. There must be suspicion that the samples were numbered in reverse order and that this single isolate from the uterus (UC4) had originated from the cervical canal (CC1). It must also be borne in mind that the numbers were small in both studies;

however, the complex design and culture technique precluded larger numbers in each group. Yet we feel from our results that it is unlikely that the uterine cavity is in a permanent state of sterility.

Our results show, as one would expect, bacterial colonization of the endometrial cavity following insertion of an IUD. The level of colonization up to 24 h post-insertion has previously been reported at 100% [4]. Thus, it is of interest to note the level of colonization in this study is only 55%. This we attribute to the pre-insertion use of chlorhexidine. Of greater interest is the fact that we could not demonstrate any statistical difference between the standard IUD group and the group with IUDs coated with chlorhexidine. Thus, it appears that the extra 2 mg on the device does little to reduce the bacterial load introduced at the time of insertion. This finding is difficult to explain, as all the organisms cultured were sensitive to chlorhexidine and the device when tested *in vitro* was active against *S. aureus, N. gonorrhoeae, C. trachomatis* and *M. hominis*. Examination of the mean inhibitory concentrations of the organisms grown shows an excess concentration of chlorhexidine, requiring the passage of some 200 ml of uterine secretions in 24 h to produce a significant dilutional effect. Yet, the fact that only two of the devices, in the six bacteriologically positive cavities, were positive, points to some inhibiting and/or concentration effect. It is possible that local binding to the endometrial membrane surface may have taken place and effectively reduced the local concentration of chlorhexidine. A dose study could not be performed, as the 2 mg used is the maximum amount of chlorhexidine that the device can currently be coated with.

Considering the level of bacterial contamination of all the uterine cavities, we found no statistical difference between them, nor could we discern any pattern of bacterial growth in the three groups.

Conclusion

Our study demonstrates the uterine cavity not to be sterile at all times but the insertion of an IUD under the conditions of this study results in bacterial contamination within the first 24 h in the uterine cavity to a level no greater than that of controls. Thus, we would endorse the use of chlorhexidine gluconate prior to the insertion of an IUD and consider the role of medicated IUDs worthy of further clinical study.

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Resumé

Vingt-neuf femmes, âgées de 31 à 53 ans, qui devaient subir une hysterectomie, ont participé à cette étude. Ces patientes ont été divisées en trois groupes. On a demandé à celles du premier groupe de porter un dispositif intra-utérin Multiload Cu250, à celles du premier groupe de porter un dispositif intra-utérin Multiload Cu250, à celles du deuxième groupe un dispositif Multiload Cu250 traité à l'acétate de chlorhexidine, tandis que le troisième groupe devait servir de témoin et ne portait pas de DIU.

Des cultures bactériologiques du vagin et de l'ectoderme cervical ont été entreprises avant l'insertion du DIU. Au moment de l'hystérectomie, quelque 18 heures plus tard, des échantillons ont été pris dans le canal cervical, dans la cavité utérine et sur le dispositif lui-même pour culture de germes anaérobies et aérobies appliquant une technique normalisée préalablement validée.

Pour trois des neufs patientes ayant servi de témoins, des germes provenant de la cavité utérine ont été cultivés. Aucune différence n'a été constatée entre les résultats bactériologiques des deux groupes portant des dispositifs traités et non traités en ce qui concerne les cultures des échantillons provenant du canal cervical, de la cavité utérine ou des dispositifs eux-mêmes, ni aucune différence entre les groupes portant les dispositifs et le groupe témoin.

Resumen

En este estudio participaron veintinueve mujeres, de 31 a 53 años de edad, que debían ser sometidas a una histerectomía. Las pacientes fueron divididas en tres grupos. Se solicitó a las del primer grupo que tuvieran un dispositivo intrauterino Multiload Cu250 y a las del segundo grupo un dispositivo Multiload Cu250 tratado con acetato de clorhexidina; el tercer grupo servía de testigo y no tenía colocado un DIU.

Se realizaron cultivos bacteriológicos de la vagina y del ectodermo cervical antes de colocarse el DIU. En el momento de la histerectomía, unas 18 horas despúes, se tomaron muestras del canal cervical, de la cavidad uterina y del propio dispositivo a fin de realizar un cultivo de organismos anaerobios y aerobios aplicando una técnica estandarizada previamente validada.

En tres de las nueve pacientes testigo, se cultivaron organismos provenientes de la cavidad uterina. No se verificó ninguna diferencia entre los resultados bacteriológicos de los dos grupos que tenían dispositivos tratados y no tratados, en lo que respecta a los cultivos de muestras provenientes del canal cervical, de la cavidad uterina o de los propios dispositivos. Tampoco se observó ninguna diferencia entre el grupo testigo y los grupos que tenían DIU.