ORIGINAL RESEARCH

Interference Potential of Personal Lubricants and Vaginal Medications on ThinPrep® Pap Tests

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Background: **The effects of water-based speculum lubricants on both conventional Papanicolaou smears and liquid-based Papanicolaou tests have been well documented. However, studies are lacking concerning the effects on these tests of personal lubricants and/or vaginal medications. Lubricants containing "carbomers" or "carbopol polymers" are known to interfere with ThinPrep® Papanicolaou test processing. Many over-the-counter products that advertise effects lasting several days may contain these substances. This study tests 2 popular personal lubricants (KY Warming Liquid [Johnson & Johnson, New Brunswick, NJ] and Replens [KoRa Health Care, Swords, Ireland]) and one yeast medication (Monistat 7, McNeill-PPC, Inc., Skillman, NJ) for interference on ThinPrep® Papanicolaou tests.**

Methods: **The residual specimens from 270 ThinPrep® vials were combined to form 100 homogenous ThinPrep® Papanicolaou specimens. Nine groups of 10 vials were then contaminated with 20 μL, 100 μL, and 500 μL of each of the 3 products. One group of 10 vials served as control. Cell counts were performed after processing the specimens onto slides, and the results were recorded.**

Results: **KY Warming Liquid had no effect at any volume of contamination. Replens caused a drastic reduction in cellularity at even the lowest volume. Monistat 7 reduced cellularity incrementally as the volume increased.**

Conclusion: **The potential exists for some over-the-counter lubricants and vaginal medications to interfere with the ThinPrep® Papanicolaou test. (J Am Board Fam Med 2011;24:181–186.)**

Keywords: **Cervical Cancer, Human Sexuality, Lubricants, Pap Smears, Pap Tests, ThinPrep®, Vaginal Medications, Women's Health**

In the history of medicine, the most successful test for preventing cancer is the Papanicolaou (Pap) test. It has reduced cervical cancer mortality rates 70% to 80% among populations in whom it is administered. Elsewhere, cervical cancer is still a leading cause of cancer-related deaths among women.[1] More than 70% of the Pap tests done in the United States are ThinPrep® Pap tests, manufactured by Hologic, Inc. (Bedford, MA).[2] The specimens are collected in preservative-filled vials and sent to a laboratory for processing. During processing, a semipermeable membrane filter is used to collect cells from the vial and then transfer them to a glass slide. The ThinPrep® processor senses when the filter has enough material to transfer, but it cannot determine the nature of the material. Suction of material through the filter will cease regardless of whether it is full of epithelial cells, blood, inflammation cells, or contaminants. After the slide is prepared, a cytologist screens the specimen under a microscope for cellular abnormalities or certain infectious agents, such as yeast, trichomonas, and herpes.

In this study, cellularity is defined as the amount of cells transferred to the glass slide from the ThinPrep Pap test vial during processing. Pap tests cannot be interpreted for disease if there is inadequate cellularity (<5000 epithelial cells for ThinPrep® Pap tests) or if the cells are obscured by blood or other factors. This can occur as a result of poor sampling technique, the presence of disease, or

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interference with processing by certain contaminants.[3,4] The improper use of speculum lubricant, leading to contamination of the external cervical os and/or specimen slide, has been shown to create background artifacts, obscure cellular detail, and increase the likelihood of an unsatisfactory result in conventional Pap smears.[5,6] It has also been associated with creating artifacts and reducing cellularity in liquid-based Pap tests.[6,7] Additionally, Hologic, Inc., has indicated that lubricants that contain "carbomers" or "carbopol polymers" are known to interfere with ThinPrep® Pap test processing and recommends avoiding their use.[8] However, using an appropriate amount of speculum lubricant that does not contain these interfering substances does not seem to cause any interference with either conventional Pap smears or liquid-based Pap tests.[9–16]

Whether personal (over-the-counter) lubricants and/or vaginal medications cause interference with the processing and interpretation of Pap tests has not been well studied. These products have become increasingly prevalent during the past few years, and their impact on liquid-based Pap tests and other tests performed from the same vial (ie, testing for human papillomavirus and chlamydia/gonorrhea) is not well understood. Many of these products advertise a "tingling" and/or "warming" sensation, which suggests the epithelial surface is being irritated.[17] Burning sensations can be caused by hyperosmotic lubricants. These products can cause an increase in mucus production and in severe cases may cause tissue damage.[17] One thought was whether these in vivo effects could lead to interference during sampling or processing. However, the aim of this study was to determine whether substances in the product itself, rather than the product's effect on the epithelium, caused interference.

Other products claim to last up to 4 days or more. Recent use by the patient increases the likelihood of the ThinPrep® vial becoming contaminated. Products that advertise these increased retention times may contain the interfering substances identified by Hologic, Inc. Therefore, the purpose of this in vitro study was to determine whether these over-the-counter products have the potential to interfere with ThinPrep® Pap test processing by reducing cellularity or rendering the specimen unsatisfactory for interpretation.

Materials and Methods

To eliminate confounding variables (eg, age, being pregnant or in a postpartum phase, being postmenopausal, receiving hormone therapy, timing of the last menstrual period, and having a hysterectomy), the residual volumes of 270 ThinPrep® Pap test vials were pooled into one large container that was then used to refill 100 new vials. The residual vials were selected by a cytologist through review of the initial Pap test slides that were made. Only specimens that displayed good cellularity and clean backgrounds (ie, minimum amount of inflammation, blood, and debris) were chosen. Residual ThinPrep® Pap test specimens have a shelf life of 3 months at room temperature.[18] This was well within the range of all the samples used in this study. The 270 vials were pooled into a clean, 4-liter plastic container. The container was briefly shaken before and during the refilling of the 100 test vials to ensure that a homogenous mixture of the specimen was maintained.

Two popular personal lubricants and one vaginal yeast medication were selected for testing. These included KY Warming Liquid (Johnson & Johnson, New Brunswick, NJ), Replens (KoRa Health Care, Swords, Ireland), and Monistat 7 (McNEIL-PPC, Inc. Skillman, NJ). Ninety test vials were used to test the products (ie, 30 for each product and 10 for each concentration) and 10 test vials served as controls. The products were tested in concentrations of 20 μL, 100 μL, and 500 μL. After contamination, the vials were vortexed on a Vortex-Genie 1 (model 51–0136, Scientific Industries, Inc., Bohemia, NY) to ensure adequate mixing. The uncontaminated controls were also vortexed in this manner. The next day, slides were made from the vials on a ThinPrep® 2000 Processor (Hologic, Inc.) and were stained and coverslipped.

Cellularity was estimated by a cytologist using an Olympus BX40 compound microscope (Olympus America, Inc., Center Valley, PA). Cells were manually counted in 10 fields of view (FOV) with a 40× objective and a field no. 22 eyepiece. The FOVs were averaged and then multiplied by 1322 to arrive at the estimated number of cells.[19] Data were analyzed using the one-way analysis of variance (ANOVA) test with a significance of $P < .01$. Comparison after ANOVA used the Tukey honestly significant difference (HSD) test.

Table 1. Mean Cellularity of Samples

Sample (N = 100)	Cellularity (Mean [SE])
Control (n = 10)	61,222 (±2,200.73)
KY	
20 μL (n = 10)	60,508 (±1,745.85)
100 μL (n = 10)	58,406 (±1,418.34)
500 μL (n = 10)	59,953 (±1,592.90)
Replens	
20 μL (n = 10)	2,208 (±222.13)
100 μL (n = 10)	4,442 (±385.27)
500 μL (n = 10)	4,429 (±383.78)
Monistat	
20 μL (n = 10)	19,275 (±901.55)
100 μL (n = 10)	10,523 (±288.43)
500 μL (n = 10)	3,873 (±153.98)

The study was approved by the University of Nebraska at Kearney Institutional Review Board (IRB). IRB exempt status was granted because only residual laboratory specimens were used and no patients were identified in this study (IRB no. 100109-1).

Results

The cellularity (mean and SE) of each group of 10 specimens was calculated (see Table 1). One-way ANOVA determined that the groups displayed significantly different means ($P < .01$) when compared (see Table 2). Comparison after ANOVA using the Tukey HSD test determined that the control and KY Warming Liquid groups did not differ significantly. However, the Monistat 7 and Replens groups all displayed significantly different means compared with the control. Replens had an immediate and drastic effect on specimen cellularity at even the lowest volume (20 μL). Monistat 7 also had a significant reduction in cellularity, but the reduction occurred incrementally as the volume increased from 20 μL to 500 μL (see Table 3). All of the control slides had adequate cellularity, the cells were well-preserved, and they appeared normal in all respects.

Table 3. Tukey Honestly Significant Difference Test*

Samples Compared	$M_L - M_S$	P
$M_{Control} \cdot M_{20\ \mu L\ KY}$	714	>.05
$M_{Control} \cdot M_{100\ \mu L\ KY}$	2,816	>.05
$M_{Control} \cdot M_{500\ \mu L\ KY}$	1,269	>.05
$M_{Control} \cdot M_{20\ \mu L\ Replens}$	59,014	<.01
$M_{Control} \cdot M_{100\ \mu L\ Replens}$	56,780	<.01
$M_{Control} \cdot M_{500\ \mu L\ Replens}$	56,793	<.01
$M_{Control} \cdot M_{20\ \mu L\ Monistat}$	41,947	<.01
$M_{Control} \cdot M_{100\ \mu L\ Monistat}$	50,699	<.01
$M_{Control} \cdot M_{500\ \mu L\ Monistat}$	57,348	<.01

*Determination of significance found by comparing the means of each sample to the mean of the control ($HSD_{0.01} = 6275$; $HSD_{0.05} = 5397$).
M, mean; M_L, largest mean; M_S, smallest mean.

Discussion

This study demonstrated the potential of certain over-the-counter lubricants and vaginal medications to severely lower cellularity in the ThinPrep® Pap test. Lubricants and medications have been shown to spread efficiently throughout the vagina and to the internal os very quickly.[20–25] Many products also claim to last for several days with just one application. Therefore, it seems likely that the use of personal lubricants or vaginal medications within a few days before the examination could contaminate the Pap test specimen.

The KY Warming Liquid did not cause interference when placed directly into the vial. However, we believe further testing of this product is warranted. Warming and/or tingling sensations are often caused by hyperosmotic lubricants that irritate the epithelium and promote an increase in mucus production.[17] Excessive mucus may lead to suboptimal and/or unsatisfactory Pap tests.[26] An in vivo study involving patients who recently used this product would be needed to test this possibility.

Replens is a common lubricant used by postmenopausal women and advertised to last up to 3 days. Replens and other products that claim to last in the vagina for several days use bioadhesive polymers to adhere to the epithelium.[27,28] This study

Table 2. One-Way Analysis of Variance of Sample Means

Source	Sum of Squared Deviates	Degrees of Freedom	Mean of Squared Deviates	F ratio	P
Between groups	68,431,032,603	9	7,603,448,067	554.8	<.01
Within groups	1,233,448,955	90	13,704,988		
Total	69,664,481,558	99			

demonstrated that a very small amount (20 μL) of Replens in the ThinPrep® Pap test collection vial can lead to an unsatisfactory result. Speculum lubricants that use carbomers or carbopol polymers are not recommended by Hologic, Inc., the manufacturer of the ThinPrep® Pap test. They have identified these products as interfering substances and advise that only water or a small amount of recommended water-based lubricant be used for speculum lubrication.[8] We believe products containing these substances interfere with processing by adhering to and clogging the semipermeable membrane of the filter, thereby blocking transfer of cells to the glass slide. We infer this from the observation that very little fluid was removed from contaminated vials that produced a glass slide with reduced cellularity. This implies that filtration became blocked. Normally, fluid continues to be drawn from the vial until the processor senses the semipermeable membrane is "full" or no more fluid remains.

Monistat 7 is a miconazole nitrate cream that should be applied once a day for 7 days. It only caused unsatisfactory results when 500 μL was added to the collection vial. However, at 20 μL cellularity was 69% less than the control and at 100 μL it was 83% less than the control. Other types of Monistat also exist that do not need to be applied as often (Monistat 3 and Monistat 1). Monistat 1 advertises the use of a bioadhesive formula that lasts for the 7 days needed to clear the infection after only one application. Testing of these and other topical vaginal medications may show that these products have greater effects on reducing cellularity.

Limitations

This study used 100 homogenous specimens for testing, which eliminated any biological variables. All of the products were tested under identical conditions with identical specimens. However, the tests were conducted in vitro by placing the products directly into the vial and vortexing to adequately mix them with the specimen. In reality, the products would be on the vaginal and/or cervical epithelium and transferred to the vial with a sampling device. The actual amount of product transferred from the epithelial surface to the vial may be quite different from the amounts used in this study. The products may also be altered in some way after they are applied to the epithelium. In vivo testing should be done to determine whether these outcomes can be repeated.

The specimens combined for use in this study were all interpreted as negative for intraepithelial lesion and malignancy. Therefore, it is not known how these products might affect the transfer of abnormal cells to the glass slide. However, we believe the problem lies with the products halting filtration past the semipermeable membrane during processing. Therefore, whether or not the cells are abnormal should not make a difference to the outcome of the study.

We do not know how mixing 270 different patient samples to form one specimen might affect the test. No appropriate control exists to test this concern. The contents of the vials are fixed in preservative and contain no living organisms. The vials should only contain normal epithelial cells and vaginal flora. Additionally, all of the uncontaminated controls appeared to be well-preserved and exhibited good cellularity.

This study tested the effects of 2 over-the-counter lubricants and one vaginal medication on ThinPrep Pap tests. We do not know how or if these products would interfere with conventional Pap smears or other test methods such as SurePath (another liquid-based Pap test; BD, Franklin Lakes, NJ). There are many more products on the market than were tested in this study. We have shown that the potential exists for some of them to cause interference with Pap test processing. However, we can only confirm which specific products cause inference by directly testing each one of them.

Future Research

Future research should focus on testing more of these products on ThinPrep® and other liquid-based Pap tests. Products that advertise sensations such as tingling or warming sensations should be tested on actual patients to determine whether mucus production or epithelial tissue damage causes interference. It should also be studied whether these products interfere with other tests that are performed out of the ThinPrep® vial (ie, testing for human papillomavirus and chlamydia/gonorrhea).

Conclusion

This study has significant importance to clinicians and their patients. Unsatisfactory Pap tests are costly and frustrating for the patient, the clinician, and the laboratory. More importantly, they may

prolong the detection of disease. No determination can be made as to the presence or absence of disease when a Pap test is unsatisfactory for evaluation. Certain unsatisfactory ThinPrep® Pap tests have been shown to contain abnormal cells after reprocessing.[29] Furthermore, women with unsatisfactory Pap tests have been shown to be at an increased risk of harboring abnormal cells.[4,30] Because some personal lubricants and vaginal medications advertise lasting up to 7 days (eg, Monistat 1), it may be worth questioning whether an unsatisfactory Pap test result is caused by the patient's use of one of these products. If so, the clinician might advise the patient to avoid using the product before returning for a repeat Pap test.

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