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Specificity of Two Commercial Acid Phosphatase Determination Kits with Respect to Feminine Hygiene Products and Vaginal Contraceptives

In the analysis of substances removed from the vagina of a victim, the criminalist relies upon histological and chemical tests that must possess the highest degree of accuracy and specificity to afford no margin of error. The identification of intact spermatozoa by proper histological techniques is the best proof for the demonstration of seminal fluid. There are many situations in which morphologically complete spermatozoa cannot be isolated, including old specimens, aspermia, azoospermia, heavy menstrual flow, or cleansing by the victim. In these cases, reliable chemical tests are an absolute necessity.

It has been known for some time that secretions from the prostate gland, which comprise a portion of seminal fluid, are very rich in acid phosphatase activity [*1*]. Comparably high values have been found only in the prostatic secretions of man and monkey [*2*]. Because of this high activity, interference with the enzymatic assay for prostatic acid phosphatase is not produced by various body fluids or food stains [*3*]. Demonstration of high amounts of this particular enzyme is positive proof of seminal fluid.

There are several commercial acid phosphatase determination kits on the market that claim particular specificity for prostatic acid phosphatase. Since many criminal investigation laboratories use these convenient kits, it was decided to test the specificities of two of them with respect to feminine hygiene products, vaginal contraceptives, vaginal antibiotics, and a variety of miscellaneous items. Since suspected seminal stains on assorted types of materials are encountered quite often, a representative sample of detergents was also analyzed. Several carbonated soft drinks were analyzed because of their impromptu use as douches.

Materials and Methods

Ninety-six different products were tested, including cleansing douches, creams and foams for vaginal infections, contraceptive creams and foams, deodorant sprays and powders, lubricants for diaphragms and vaginal surgical instruments, a variety of detergents, and several miscellaneous items. The majority of these products were purchased from stores in the Denver area, and the remainder were obtained as free samples from pharmaceutical companies.

Solutions of each product were prepared according to manufacturer's specifications using physiological saline (0.9 percent NaCl in distilled water) instead of water, since

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vaginal fluids include 0.9 percent NaCl. With the creams and foams, a specific amount of each was mixed with 1.5 ml saline. When the product was a suppository, approximately ⅔ of it was placed into 1.5 ml saline. To avoid contamination, all of the solutions were prepared in test tubes that had been washed with soap once, rinsed with distilled water three times, and dried between each washing or rinsing. The rubber stoppers were treated in the same manner. Each solution was tested by the two methods immediately after being prepared, and a representative sample of the solutions (approximately 50 percent) was retested after 1–2 days.

The two commercial acid phosphatase determination kits included Acid Phosphatase manufactured by American Monitor[3] and Phosphatabs-Acid manufactured by Warner-Chilcott.[4] Each of the assays were performed according to directions supplied by the manufacturer, except for one modification. With the American Monitor kit, 0.1 ml of prepared solution was analyzed, instead of 0.2 ml. This modification is used routinely by both laboratories in the analysis of suspected rape cases because it increases the specificity of this test.

Results

The results of both acid phosphatase assay methods are recorded in Table 1. There were no changes in the results if the assays were performed immediately after preparation of the solutions or one to two days later.

All of the five products which gave positive results with one of the assay methods were tested further. Clorox solution was the only product with which positive results were obtained with the American Monitor Acid Phosphatase kit (Table 1). However, a piece of cloth, which was washed with the dilute Clorox solution and tested after drying with or without rinsing, gave negative results (Table 2).

The other four products, including two douche powders, a contraceptive jelly, and a jelly utilized in the treatment of vaginal infections, gave positive results with the Warner-Chilcott Phosphatabs-Acid kit (Table 1). In the directions for this kit, it is suggested that all borderline or elevated results be repeated quantitatively. Solutions of each of these products were, therefore, reanalyzed using the Warner-Chilcott Phosphatabs-Acid, Quantitative kit, according to directions supplied by the manufacturer. The results of each of these analyses are recorded in Table 2.

None of the products tested in this study gave positive results with both assay methods.

Discussion

The only product tested that produced positive results with the American Monitor Acid Phosphatase kit was dilute Clorox solution. However, material soaked in Clorox solution and allowed to dry gave negative results (Table 2). Therefore, with respect to the products tested in this study, the specificity of the American Monitor kit for prostatic acid phosphatase as stated has been supported. Approximately 14 percent of the products analyzed produced varying amounts of color with the American Monitor kit. With all such products, the same amount of color was produced in the respective blanks, therefore resulting in an optical density of zero when each was read against its blank at 650 μm using an ultraviolet-visible spectrophotometer. The extreme importance of running a blank with each sample cannot be overemphasized. If a blank is not run, interference may result from any of the tested products that produced color.

Four products tested gave positive results with the Warner-Chilcott Phosphatabs-Acid kit. Although this particular kit has been reported to be specific for prostatic acid phos-

[3] American Monitor Corporation, Indianapolis, Ind.
[4] Warner-Chilcott Laboraotries, Morris Plains, N. J.

TABLE 1—*Results of products analyzed.*

Type of Product	Product Name and Manufacturer	American Monitor, Optical Density	Warner-Chilcott, + or −
Cleansing Douches	Aqua Fresh Hypo-Allergenic Douche Tablets, Abbott Laboratories	0	−
	Betadine Douche Liquid, (also for vaginal infections), The Purdue Frederick Co.	0	−
	Bo-Car-Al Douche Powder, Calgon Corp.	0	−
	Certane Douche Powder, Vogarell Products, Inc.	0	−
	Cupid's Quiver Liquid Douche, Tawn Limited	0	−
	Demure Douche Concentrate, Roycemore, Inc.	0	−
	Femicare Vaginal Douche Powder, The Femicare Co.	0	−
	Feminique Douche Liquid Concentrate, Intec Laboratories	0	−
	Hexol (also germicide), Hexol, Inc.	0	−
	Hy-Geen Douche Powder, The Pfeiffer Co.	0	−
	Jeneen Liquid Douche, The Norwich Pharmacal Co.	0	−
	Koromex Douche Powder, Holland-Rantos Co., Inc.	0	−
	Kotique Douche Powder, Kimberly-Clark, Corp.	0[a]	−
	Lanteen Douche Powder, Esta Medical Laboratories, Inc.	0	−
	Lysette Liquid Douche, Sterling Drug Co.	0	−
	Massengill Douche Powder, S. E. Massengill Co.	0	−
	Massengill Douche Powder (Floral Scent), S. E. Massengill Co.	0	−
	Norforms Suppositories, The Norwich Pharmacal Co.	0	−
	Nylmerate Antiseptic Solution Concentrate (also effective against pathogenic organisms), Holland-Rantos Co., Inc.	0	−
	Pamprin Concentrated Douche Powder, Chattem Drug and Chemical Co.	0	−
	StomAseptine Douche Powder, Cooper Laboratories, Inc.	0	−
	Summer's Eve Douche Liquid, Personal Laboratories	0	−
	Takara Douche Powder, Ladeo Laboratories	0	−
	Tannette Feminine Hygiene Powder, Walgreen Laboratories, Inc.	0	−
	Trichotine Vaginal Douche Powder, Reed and Carnrick	0	−
	Triva Douche Powder (also a spermicide and used for vaginitis), Boyle and Co. Dist.	0	+
	Tyree's Douche Powder, J. S. Tyree	0	−
	V.A. Douche Powder, Norcliff Laboratories Inc.	0	+
	Vagisec Liquid Douche Concentrate, Julius Schmid, Inc.	0	−
	Veen Douche Powder, The Norwich Pharmacal Co.	0	−
	Verazeptol Douche Powder, Newman Pharmacal Co.	0	−

(*Continued*)

TABLE 1—Continued

Type of Product	Product Name and Manufacturer	American Monitor, Optical Density	Warner-Chilcott, + or −
	Zeptin Vaginal Douche Powder, Larre Laboratories Inc.	0	−
	Zonite Liquid Douche, Chemway Corp.	0	−
	Zonitors Suppositories, Chemway Corp.	0	−
Deodorants	Amolin Deodorant Powder, Norwich Pharmacal Corp.	0	−
	Assure, Avon Co.	0	−
	Bidette Towelettes, Youngs Drug Products Corp.	0	−
	Easy Day Feminine Hygiene Spray Deodorant, Carter-Wallace, Inc.	0	−
	FDS Feminine Hygiene Deodorant Spray, Alberto-Culver Co.	0	−
	FDS Towelettes, Alberto-Culver Co.	0	−
	Feminique Personal Hygiene Towelettes, Intec Laboratories	0	−
	Koro Sanitary Napkin Deodorant Spray, Holland-Rantos Co., Inc.	0	−
	Kotique Feminine Deodorant Spray, Kimberly-Clark	0	−
	Midol Deodorant Spray Mist, Glenbrook Laboratories	0	−
	My Own Hygiene Deodorant Spray, Emko Co.	0	−
	Perifoam (also effective against infections), Holland-Rantos Co., Inc.	0	−
	Pristeen Femine Hygiene Deodorant Spray, Warner-Lambert Co., Dist.	0	−
	Quest Deodorant Powder, Vick Chemical Co.	0	−
	Rantex Personal Cloth Wipes, Holland-Rantos Co., Inc.	0	−
	Vespre Feminine Hygiene Deodorant Spray Mist, Personal Products Co.	0	−
Contraceptives (Spermicides)	Conceptrol Birth Control Cream, Ortho Pharmaceutical Corp.	0	−
	Delfen Contraceptive Foam, Ortho Pharmaceutical Corp.	0	−
	Emko Vaginal Foam, Emko Co.	0	−
	Immolin Contraceptive Vaginal Cream-Jel, Julius Schmid, Inc.	0	−
	Koromex—A Vaginal Jelly, Holland-Rantos Co., Inc.	0	−
	Lanteen Vaginal Jelly, Esta Medical Laboratories, Inc.	0	+
	Lorophyn Jelly, Eaton Laboratories	0	−
	Lorophyn Suppositories, Eaton Laboratories	0	−
	Milex Crescent Vaginal Jelly, Milex Products	0	−
	Ortho-Creme Contraceptive Cream, Ortho Pharmaceutical Corp.	0	−
	Ortho-Gynol Contraceptive Jelly, Ortho Pharmaceutical Corp.	0	−
	Preceptin Vaginal Gel, Ortho Pharmaceutical Corp.	0	−
	Ramses Contraceptive Vaginal Jelly, Julius Schmid, Inc.	0	−

(*Continued*)

TABLE 1—Continued

Type of Product	Product Name and Manufacturer	American Monitor, Optical Density	Warner-Chilcott, + or −
Cures for Vaginal Infections	Aci-jel Therapeutic Vaginal Cream, Ortho Pharmaceutical Corp.	0[a]	+
	AVC Cream, The National Drugs Co.	0	−
	Dienestrol Cream, Ortho Pharmaceutical Corp.	0[a]	−
	Sufamal Vaginal Cream, Milex Products, Inc.	0	−
	Sultrin Triple Sulfa Cream, Ortho Pharmaceutical Corp.	0[a]	−
	Sporostacin Cream, Ortho Pharmaceutical Corp.	0[a]	−
	Tricofuron Vaginal Powder, Eaton Laboratories	0	−
	Tricofuron Vaginal Suppositories, Eaton Laboratories	0	−
	Vagitrol Cream, Syntex Laboratories	0	−
Lubricants	Alphosyl Lubricating Cream, Reed and Carnrick	0	−
	H-R Sterile Lubricating Jelly, Holland-Rantos Co., Inc.	0	−
	K-Y Sterile Lubricating Jelly, Johnson & Johnson	0	−
	Koromex Vaginal Cream, Holland-Rantos Co., Inc.	0	−
	Koromex Vaginal Jelly, Holland-Rantos Co., Inc.	0	−
	Lubafax Surgical Lubricant, Burroughs-Wellcome and Co.	0	−
	Lubasporin Urethral Antibacterial Lubricant, Burroughs-Wellcome and Co.	0	−
Detergents	Biz, Procter and Gamble	0[a]	−
	Breeze, Lever Brothers Co.	0[a]	−
	Burst, Colgate-Palmolive Co.	0[a]	−
	Clorox, The Clorox Co.	200+	−
	Cold-Power, Colgate-Palmolive Co.	0[a]	−
	Gain, Proctor and Gamble	0[a]	−
	Par, Safeways Stores, Inc.	0[a]	−
	Sta-Puf Fabric Softener, A. E. Staley Mfg. Co.	0	−
	Tide, Procter and Gamble	0[a]	−
	Woolite, Boyle-Midway Inc.	0	−
Miscellaneous	Pro-ception (sperm nutrient douche), Milex Products	0	−
	Sheik Lubricated Prophylactic, Julius Schmid, Inc.	0	−
	Alum	0	−
	Baking Soda	0	−
	Pepsi Cola	0[a]	−
	Seven-Up	0	−
	Vinegar	0	−

[a] Products that produced varying amounts of color (see **Discussion**).

TABLE 2—*Results of products reanalyzed.*

Product Name	American Monitor Optical Density	Warner-Chilcott Quantitative Optical Density
Clorox		
Cloth, not rinsed	0	...
Cloth, rinsed	0	...
Triva Douche Powder	...	0
V.A. Douche Powder	...	0
Lanteen Vaginal Jelly	...	0
Aci-jel Therapeutic Vaginal Cream	...	0

phatase [*4*,*5*], the results of this study do not support this specificity. When the Warner-Chilcott Phosphatabs-Acid, Quantitative kit was used to reanalyze these four products, the resultant optical densities were all zero when each was read against its blank at 530 μm (Table 2). However, an undesirable precipitate was produced with this assay method. Therefore, the specificity of the latter kit for prostatic acid phosphatase as stated by Warner-Chilcott has been shown with respect to the products tested in this study. However, our experience with the quantitative kit has been that high values typical of fairly fresh seminal fluid obtained from vaginal aspirants are not reliably quantitated.

Summary

Reliable chemical tests are an absolute necessity in the analysis of suspected rape cases, especially in the absence of spermatozoa. Cleansing by the victim or use of contraceptives or antibiotics will affect the chemistry of the vagina and may interfere with the chemical tests for seminal fluid. Solutions of cleansing douches, creams and foams for vaginal infections, contraceptive creams and foams, deodorant sprays and powders, lubricants, detergents, and several miscellaneous items were analyzed using the American Monitor Acid Phosphatase kit and the Warner-Chilcott Phosphatabs-Acid kit. Clorox solution produced positive results with the American Monitor kit, and four other products gave positive results with the Warner-Chilcott kit. However, specific retesting of these products each resulted in optical densities of zero when compared to the respective blank.

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