F-2141. Evaluation of the Copan Liquid Amies Elution Swab (ESwab) for Maintaining the Viability of Selected Fungi and *Mycobacterium* spp.

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ABSTRACT

Background: The use of swabs for obtaining specimens for culture is generally discouraged but remains a common practice. The majority of published studies have focused on the efficiency of swabs for the recovery of bacteria with little or no published data supporting their efficiency for the recovery of fungi or *Mycobacterium* spp. The ESwab consists of a flocked nylon swab for sample collection and liquid Amies transport medium. The objective of this study was to perform a quantitative evaluation of the ESwab for maintaining the viability of selected clinically relevant fungi and *Mycobacterium* spp. For comparative purposes, the BD BBL™ Culture Swab Plus transport system was tested.
Methods: The following organisms were selected for testing: *Mycobacterium chelonae, M. fortuitum, Candida albicans, C. krusei, C. parapsilosis, C. glabrata, C. neoformans, Mucor, Fusarium, Exophiala (Wangiella) dermatitidis*, and *Exophiala jeanselmei.* Quantitative viability studies were conducted in triplicate at room temperature. For each swab/organism combination, viable counts were performed at zero (0) time, 24 h, 48 h, and 72 h.
Results: Initial 0-time counts for the organisms tested were ½ to 1 log ($base_{10}$) higher for the ESwab than for the CultureSwab Plus for all organisms except *Mucor* spp. This trend was also noted for each sampling period (24 h, 48 h, and 72 h) with the percent recovery being higher with the ESwab. A relatively lower level of release was noted with the respective *Mycobacterium* representatives but the percent recovery was higher with the ESwab. A similar fold increase of mold growth was observed with both swabs.
Conclusions: The results indicate that the ESwab with liquid Amies transport medium performed better than the CultureSwab Plus with Amies gel agar. The data suggests that the ESwab is suitable for preserving the viability of pathogenic yeasts, molds, and *Mycobacterium* spp.

INTRODUCTION

The ultimate goal of the clinical microbiology laboratory is to generate clinically relevant results. Achieving this goal begins with the appropriate selection, collection, and handling of the specimen. Appropriate specimen management, or the absence of such, affects patient management in many ways. For example, good specimens are essential to accurate laboratory diagnosis that directly impacts on patient outcome, influences therapeutic decisions, affects infection control, and overall plays a major role in laboratory as well as healthcare costs. Although a number of limitations have been associated with the use of swabs (low volume, susceptible to surface and subsurface contamination), their use in the collection of specimens for microbiological culture has been and continues to be popular and widely practiced in the healthcare setting.

The Copan Liquid Amies Elution Swab (ESwab) Collection and Transport System (Copan Diagnostics Inc., Murieta, CA) consists of a flocked nylon swab for the collection of samples from the nares, throat, vagina, or wounds, and a polypropylene screw-cap containing 1 ml of Modified Liquid Amies transport medium. The specimen is absorbed by capillary action between the fibers and remains close to the surface of the fibers. Following contact with the transport medium, the system is designed for the specimen to elute with minimal entrapment of microorganisms that has been reported as a problem for other swabs (rayon, Dacron) associated with agar-based transport media.

Since the ESwab collection and transport system was initially designed to primarily maintain the viability of bacteria in clinical specimens for up to 48 h at both refrigerator and room temperature, the objective of this study was to perform a quantitative evaluation of the ESwab for maintaining the viability of selected fungi (yeasts and molds) and *Mycobacterium* spp at room temperature over a 72 h period.

MATERIALS AND METHODS

A. Transport Systems
- Copan Liquid Amies Elution Swab (ESwab) Collection and Transport System
- BD, BBL™ CultureSwab Plus Transport System with Amies Gel Medium

B. Media/Supplies
- Potato Flake Agar without antibiotics
- Middlebrook 7H10 agar
- Micropipettes (adjustable)
- 0.85% Saline

C. Challenge Organisms
1. ATCC strains of *Candida albicans* (14053), *C. krusei* (6258), *C. parapsilosis* (22019), *C. glabrata* (66032), *Cryptococcus neoformans* (14116)
2. The following clinical isolates of molds and *Mycobacterium* representatives were tested:
   - *Mucor* spp.
   - *Fusarium* spp.
   - *Exophiala (Wangiella) dermatitidis*
   - *Exophiala jeanselmei*
   - *Mycobacterium chelonae*
   - *M. fortuitum*

D. Procedure (Fig. 1)
1. All challenge isolates were cultured on Potato Flake Agar or Middlebrook 7H10 agar prior to preparing standardized inocula (0.5 McFarland Standard).
2. Conidial suspensions of molds were prepared by flooding Potato Flake agar cultures with approximately 3 ml of sterile water. The resulting mixture was withdrawn, and the heavy particles were allowed to settle for 3-5 min. The upper homogenous suspension, containing a mixture of conidia and hyphal fragments, was removed and vortexed for 15 sec. The turbidity of the suspension was measured with a MicroScan turbidity meter and adjusted to 0.5 McFarland Standard with subsequent plate counts being performed.
3. Specific colony counts were determined for each standardized inoculum
4. 100 µl of standardized inoculum was pipetted directly to each swab (3 swabs per time period). Swabs were returned to the respective transport tube and capped.
5. Swabs representing Time 0 were held at room temperature for 5-10 min before preparing serial 10-fold dilutions in sterile water.
6. 10 µl of each dilution was plated onto Potato Flake agar or Middlebrook 7H10 agar and colonies were counted following incubation.
7. All testing was performed in triplicate and viability determination was recorded for 0 time.
8. The same procedure was followed for determining colony counts representing 24, 48, and 72 h.

RESULTS

The results for ESwab and BD CultureSwab Plus are summarized in Figs 2,3, and Table 1.

1. The average initial colony counts for T0 (expressed as log_{10} CFU/ml) for the respective fungi and *Mycobacterium* spp. are shown as the first bar in the respective figures. Following inoculum standardization, initial counts ranged from 10^4 (*Mucor*) to nearly 10^7 CFU/ml (*M. fortuitum)* and served as the base-line for counts recorded at the respective sampling times.
2. Initial T0 counts were generally ½ to 1 log higher for the Eswab on all organisms except *Mucor* spp. This trend was observed for each sampling period with the percent recovery or release favoring the ESwab.
3. Release of *M. chelonae* from the initial inoculum to culture was lower for both swabs than the other organisms. The reasons for this were unclear and may warrant further investigation.
4. The overall increase in counts from T0 thru 72 h is reflective of organism growth and efficiency of release for the respective swabs.

CONCLUSIONS

1. Overall, the increased release of organisms from the ESwab combined with superior preservation of viability by the ESwab Amies Liquid offers superior performance in comparison to the BD CultureSwab Plus.
2. The data suggest that the ESwab is a suitable system for preserving the viability of pathogenic yeasts, molds, and *Mycobacterium* spp. that may be contained in a variety of specimens collected for microbiological analysis.
3. To our knowledge, this is the first study to assess the efficacy and efficiency of two commercial Swab/Transport Systems for maintaining the viability of yeasts, molds, and *Mycobacterium* spp.

Fig 2. ESwab Recovery

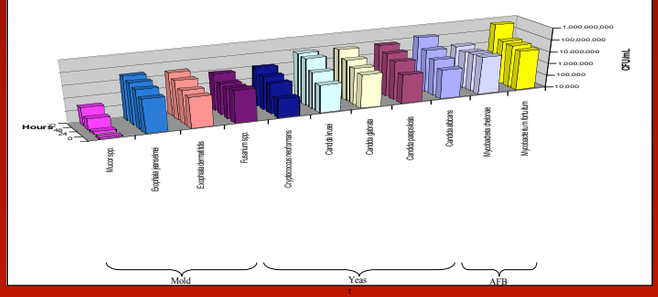

Fig 3. CultureSwab Plus Recovery

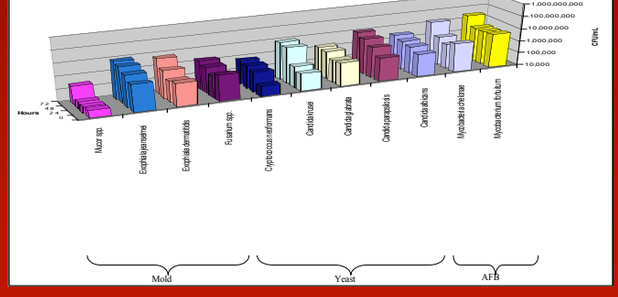

Fig. 1. Procedure for Quantitative Evaluation of the ESwab and the BD CultureSwab Plus to Maintain the Viability and Release of Selected Fungi and *Mycobacterium* spp. at Room Temperature Over a 72 h Period

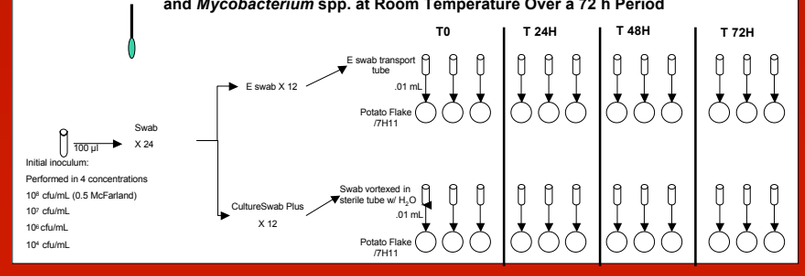

Table 1. Release Rates Expressed as Percentage of Initial Inoculum

	Time	*Mucor spp.*	*Exophiala jeanselmei*	*Exophiala dermatitidis*	*Fusarium spp.*	*Cryptococcus neoformans*	*Candida krusei*	*Candida glabrata*	*Candida parapsilosis*	*Candida albicans*	*Mycobacteria chelonae*	*Mycobacterium fortuitum*
Eswab	0	130	90	138	113	103	48	46	89	114	16	30
	24	160	213	224	74	778	243	73	545	419	14	45
	48	800	361	667	84	1417	1675	183	1323	1381	11	55
	72	2800	590	1333	255	3611	2250	692	3230	8095	18	781
CultureSwab Plus	0	370	23	32	32	18	6	6	29	32	3	8
	24	330	57	23	13	108	12	5	105	62	1	7
	48	320	120	71	29	158	178	13	261	90	2	7
	72	2600	146	281	45	208	233	14	432	148	19	42

ACKNOWLEDGMENTS

We thank Copan Diagnostics for supplying the culture media and ESwabs used in support of this study. Special thanks to the Microbiology Technical Staff for their flexibility and assistance during the final phases of this study.