

Introducing a multipurpose sample collection and transport system compatible with lab automation

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Introduction

In our laboratory, the automated inoculation system, InoquIA, (Becton Dickinson, BD) was purchased while in the molecular department the Roche FLOW system, is employed. Optimal use of these automated systems requires an uniform multipurpose sample collection and transport buffer/kit. Additional benefit is that sampling once is less troublesome for the patient. Liquid Amies, is described as one of the best transport media for culture. Among the commercially available liquid Amies media are media/kits from Copan (ESwab), BD (BD ESwab), Puritan (Liquid Amies) and Medical Wire (Σ transwab).

The aim of this study: To test the suitability of these different sample collection kits for culture of bacteria/yeasts and molecular diagnostics from the same sample by analysing stability and different storage temperatures.

Methods

For STD testing, dilutions were made, in 1 x TE, of positive clinical samples (2 SP) for *Chlamydia trachomatis* (CT) and *Trichomonas vaginalis* (TVG) and added to the different liquid Amies media. For *Neisseria gonorrhoeae* (NG) different concentrations of culture suspensions were made. Nucleid acid extraction (MagnaPure 96, Roche) and PCR testing were performed after 0, 24, 48 hours and after one week at roomtemperature. The *Neisseria gonorrhoeae* samples were also cultured. For culture testing dilutions of *Streptococcus pneumoniae, Haemophilus influenza, Streptococcus pyogenes, Escherichia coli, Pseudomonas aeruginosa, Bacteroides fragilis* and *Candida albicans* were made. Using the flocked swab an amount of this dilution was added to the Liquid Amies and stored at room temperature or at 4 degrees. Culture and plate counting were performed after 0, 24 and 48 hours.

Results

Table 1: STD stabillity testing

	Crossing point LC480 (Roche)						
Sample	0 H	24 H	48 H	168 H			
CT Copan	31,87	32,29	32,13	33,08			
CT MW	33,01	33,18	32,84	33,01			
CT BD	32,00	32,21	31,81	31,94			
CT Puritan	32,13	31,73	31,84	31,76			
CT 2SP	32,33	32,60	32,69	32,44			
NG Copan	29,58	29,74	29,38	29,96			
NG MW	29,13	28,69	29,13	30,58			
NG BD	28,74	29,02	28,99	29,56			
NG Puritan	29,02	29,00	29,03	29,26			
NG 2SP	29,29	29,04	28,82	28,89			
TVG Copan	32,25	32,44	32,42	33,71			
TVG MW	32,03	31,83	31,84	32,34			
TVG BD	32,92	32,74	32,67	32,58			
TVG Puritan	31,10	31,15	31,31	31,50			
TVG 2SP	31,80	31,81	31,87	31,88			

Table 2: Viability testing of Neisseria gonorrhoeae

	CFU after 0 H	CFU after 24 H	CFU after 48 H
Copan 50000 CFU/ml	84,5	1,5	0,0
MW 50000 CFU/ml	27,5	2,0	0,0
BD 50000 CFU/ml	82,0	0,5	0,0
Puritan 50000 CFU/ml	53,0	0,0	0,0
2SP 50000 CFU/ml	9,5	0,5	0,0

For CT, NG and TVG no significant differences in Cp values were observed after 0, 24, 48 hours of storage at room temperature, when the different Amies media were compared. Room temperature incubation of NG in different liquid amies media revealed a dramatic decrease of viability of the gonococci.

Table 3: Culture suitability of BD ESwab

		ESwab BD 4-8 °C			ESwab BD roomtemperature		
	0 H	24 H	48 H	0 H	24 H	48 H	
Straiı	าร	CFU's/ESwab			CFU's/ESwab		
S.pneumonia	<i>ne</i> 770	523	550	1045	3795	conf	
H.influenzo	<i>ae</i> 3740	1705	1100	5390	633	275	
S.pyogen	<i>es</i> 1595	2530	2365	3025	conf	conf	
E.co	oli 2145	1128	3355	3025	conf	conf	
P.aeruginos	sa 1155	1430	1650	440	conf	conf	
B.fragi	<i>lis</i> 9075	3465	2750	6875	4675	3740	
C.albica	<i>ns</i> 2035	2585	1540	2365	conf	conf	
MRS	SA 1100	4538	3025	1265	conf	conf	

As the ESwab from Copan and BD turned out to be identical, further culture-suitability was only tested for the BD ESwab. Results of culture and platecount show degredation in time of *Heamophilus influenza* and *Bacteroides fragilis* when the sample was stored at roomtemperature as well as at 4 degrees. When stored at roomtemperature *Streptococcus pneumoniae, Streptococcus pyogenes, Escherichia coli* and *Pseudomonas aeruginosa* were still replicating resulting in overgrowth of the culture-plate. When stored at 4°C, the latter organisms appeared to be stable and viable, resulting in non-overgrown plates.

Conclusion

- Culture and molecular testing for diagnostics of infectious diseases can be performed reliably, out of one tube.
 For molecular STD testing all tested commercially available Amies media were stable for at least one week at room temperature without sample degradation when diluted target organisms were used, whereas culture of *Neisseria gonorrhoeae* must take place within 48 hours.
- For general culture the samples were stable for 48 hours, only when stored at 4 degrees. Therefore it is important to place the sample at 4 degrees at the sampling-location if transport to the lab takes more than 24 hours.