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Performance evaluation of a urine collection tube with a new preservative : VACUETTE 10,5 ml URINE CCM (16X100) REF. 455052R

Introduction :

The relevance of the pre analytical management of urine samples is becoming more important for several reasons :

Laboratory agglomeration involves long distances, specimens shipment and longer storage of samples before the analytical testing.

New bacteriological guidelines require the use of lower cut-offs for the clinical interpretations

To accomplish these two needs, manufacturers are taking efforts to develop new devices able to ensure a good stability of the specimens (also if stored at room temperature) minimizing the effects of the preservatives on the quality of the laboratory test.

Aim of the study :

The aim of this study was the evaluation of performances of a new collection tube for urine with a new preservative : VACUETTE 10,5 ml URINE CCM (16X100) REF. 455052R manufactured by Greiner Bio-One GmbH – 4550 Kremsmunster- Austria

Study description :

501 native urine samples collected in a Public Hospital of North East of Italy in the period (February – June 2012) have been processed with and without use of preservative at different times and by different manual and automated technologies (traditional Petri dish culture and Alfred 60 instrument).

The sample collection was done by a urine beaker ; then different aliquots of specimens have been obtained by filling the vacuum urine tubes with or without preservative.

The urine tubes without preservatives have been tested first at the TIME 1(by Petri dish and Alfred 60) to record the results obtained with the native specimens. This data has been considered as a reference for the performance evaluation of the preservative.

The same urine specimens filled in tubes with the preservative have been re-tested (by Petri dish and Alfred 60) respectively after 3h and 18h storage at room temperature.

The purpose of the study was to evaluate eventual discrepancies of the analytical results after the exposure of the biological specimen to preservatives and after different times of storage at room temperatures taking the results obtained with a fresh unpreserved samples as reference.

The results of the evaluation will particularly consider the increase of false negative results due to potential toxic effects of the preservative and the statistical incidence of sample contamination occurred after the storage of the urine samples.

Materials and methods :

For native urine sample collection : VACUETTE URINE BEAKER (100 ml) REF 724310

For unpreserved urines : filling with VACUETTE 3 mL Z No Additive (13x75) Ref.454241

For preserved specimens: filling with VACUETTE 10,5 mL Urine CCM (16x100) Ref. 455052R,

For Petri dish culture : ChromID CPS Agar Ref. 43821 (Biomerieux ; 69289 Marcy L'Etoile-France).

For Alfred testing : Instrument Alfred 60 (Sire Analytical Systems , 33100 Udine – Italy)
SW configuration settings :

- Native specimens : Mode Standard / Incubation time 5 hours - Cut off : < 50 CFU/ml
- Preserved specimens : Mode Boric Acid / Incubation time 5 hours Cut off : < 50 CFU/ml

Reagents : Uro Quick Screening KIT SI 390 .900 (Sire Analytical Systems , 33100 Udine – Italy)

Results:

Validation of Alfred results for specimens collected in CCM urine collection tube with preservative after 3hours from withdrawal.

Samples : 501

CCM			
CN	**CP**	**FN**	**FP**
259	**170**	**62**	**10**

CCM collection:
Results in agreement : 429/ 501 (85.62%)
FN vs. plate : 62

- Not significant isolates : 46
- Significant isolates : 16

FP vs. plate : 10

List of FN significant with CCM collection but positive with unpreserved collection tube :

N	Isolate	N.	Count T1 (Alf)
1	E. coli	8	1.000.000
2	P. aeruginosa	40	1.000 *
3	E. coli	48	1.000*
4	C albicans	72	700.000
5	E. coli	95	1.000.000
6	S. epidermidis / E. faecalis	151	400.000

7	S. agalactiae	163	20.000
8	S. agalactiae	250	1.000*
9	S. aureus	255	400.000
10	P. mirabilis	312	7.000*
11	P. aereuginosa	361	800*
12	E. faecalis	414	9.000*
13	Corynebacterium spp.	421	1.000.000
14	E. coli	440	1.000.000
15	E. coli	460	1.000.000
16	E. coli	487	neg *

Comments :

Considering the conventional cut-off 10^4CFU/ml and discarding the 7 results (marked with *), detected positive on native specimens by Alfred 60 at TIME 1 but counted below this threshold 9 patients were missed after 3h storage with preservation.

According to these criteria the sensitivity after the workpath with preservation and 3 h hours storage was : 94,97 %

According to these criteria the VPN after the workpath with preservation and 3 h hours storage was : 96,64 %

The specificity after the workpath with preservation and 3 h hours storage was : 96,28%

The VPP after the workpath with preservation and 3 h hours storage was : 94,44 %

Validation of Alfred results for specimens collected in CCM urine collection tubes with preservative after 18 hours from withdrawal.

Samples : 501

CCM			
CN	**CP**	**FN**	**FP**
269	170	59	3

CCM collection:
Results in agreement : 439/501 (87.82 %)
FN vs. plate : 59

- Not significant isolates : 42
- Significant isolates : 17

FP vs. plate : 3

List of FN significant with CCM collection but positive with unpreserved collection tube :

N	Isolate	N.	Count T1 (Alf)
1	P.aeruginosa	40	1000/ 1.000.000
2	E.coli	48	10.000
3	S.agalactiae	58	300.000
4	C. albicans	72	700.000
5	P.aeruginosa	135	100.000
6	S. epidermidis / E. faecalis	151	400.000
7	S.agalactiae	163	20.000
8	E.coli	200	100.000
9	P.aeruginosa	248	15.000
10	S. aureus	255	400.000
11	P. mirabilis	313	7.000 *
12	E. coli	383	Neg (800) *
13	E. faecalis	414	9.000 *
14	Corynebacterium spp.	421	1.000.000
15	E. coli	440	1.000.000
16	E. coli	460	1.000.000
17	E. coli	487	neg *

Comments :
Considering the conventional cut-off 10^4CFU/ml and discarding the 3 results (marked with *), detected positive on native specimens by Alfred 60 at TIME 1 but counted below this threshold 13 patients were missed after 18h storage with preservation.

According to these criteria the sensitivity after the workpath with preservation and 18 h hours storage was : 92.89 %

According to these criteria the VPN after the workpath with preservation and 18 h hours storage was : 95.39 %

The specificity after the workpath with preservation and 18 h hours storage was : 98.89%

The VPP after the workpath with preservation and 18 h hours storage was : 98.26 %

Conclusions :

The present study concerning the management of urine specimens in the VACUETTE 10,5 mL Urine CCM tubes (16x100) Ref. 455052R with different storage times (3h and 18h) showed acceptable performances in comparison with the tests of the correspondent fresh unpreserved specimens (both processed by Alfred 60).
However the treatment of the specimen by this preservative induce a limited lack of sensitivity after 3h (94.97 %) more evident after 18h (92.89 %).

In particular the recovery of some specific micro-organisms (ex : *S. agalactiae* , *P. aeruginosa* , *Candida spp*.) seems to be sometimes more difficult.

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On the other hand the use of the new device significantly limited the presence of contaminants induced by different other factors (Ex . vaginal contamination , collection problems):
46 contaminated native samples were preserved by CCM in the 3 h storage study
42 contaminated samples were preserved by CCM in the 18 h storage study

Concerning the specimen positive to urine culture no particular effects of VACUETTE Urine CCM have been observed in the quantification of bacterial counts (expressed in CFU/ml) detected by Alfred 60 in comparison to unpreserved samples.

Nimis , 30/05/2013

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