

NordVal Certificate

Issued for:	3M™Petrifilm™ <i>E.coli</i> / Coliform Count Plate
NordVal No:	014
First approval date:	5 May 2003
Renewal date:	1 June 2011
Valid until:	1 June 2013

3M™Petrifilm™ *E.coli* / Coliform Count Plate

Manufactured by:
3M Health Care,
Microbiology Products,
St. Paul,
Minnesota 55114-1000
USA

Supplied by:
3M MEDICA,
Hammfelddamm 11,
D-41453 Neuss,
Germany

fulfils the requirements of the NordVal validation protocol. The 3M™Petrifilm™ *E.coli* / Coliform Count Plate performance has been compared to the reference method: ISO 16649-2: Horizontal method for the enumeration of beta-glucuronidase –positive *Escherichia coli* - Part 2: Colony- count technique at 44°C using (-bromo-4-chloro-3-indoyl beta-D-glucuronate. The results document no statistical difference in the performances between the methods.


The 3M™Petrifilm™ *E.coli* / Coliform Count Plate has been fully collaboratively validated and corresponds to AOAC 991.14: Coliform and *Escherichia coli* Counts in Foods Dry Rehydratable Film.

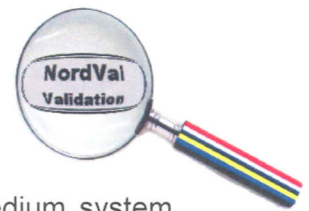
The 3M™Petrifilm™ *E.coli* / Coliform Count Plate was previously referred to as NMKL method No. 147. The NMKL method was withdrawn, when received a NordVal certificate.

Date: 01.06.2011

Yours sincerely


Sven Qvist
Chair of NordVal


Hilde Skår Norli
NMKL Secretary General



PRINCIPLE OF THE METHOD

The 3M Petrifilm *E. coli* / Coliform Count Plate is a sample-ready-culture-medium system which contains Violet red bile nutrients, a cold-water-soluble gelling agent, a tetrazolium indicator that facilitates colony enumeration and an indicator of glucuronidase activity BCIG.

E. coli: Count all blue colonies with and without gas, after 48 hours of incubation at 37°C.
Coliforms: Count red colonies with gas and all blue colonies with or without gas, after 24 hours of incubation at 37°C.

FIELD OF APPLICATION

The method has been tested on foods.

COMPARISON STUDY COMPLIANCE BETWEEN 3M PETRIFILM E.COLI / COLIFORM COUNT PLATE AND THE REFERENCE METHODS

Relative accuracy

A study carried out by ADRIA (Association pour le Développement de la Recherche Agro-Alimentaire) Quimper in 2005. The Petrifilm plate incubated at 37°C was compared against ISO 16649-2, and the following results were obtained:

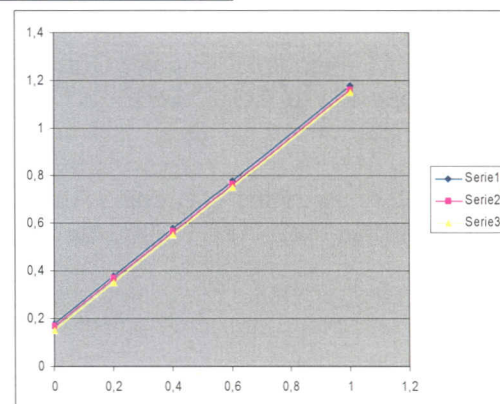
Method tested	Number of samples	Equation
Petrifilm E coli/ coliforme 37°C Diluant Peptone-salt	28	$y = 0.18 + 1.00x$
Petrifilm E coli/ coliforme 37°C Diluant Ringer 1/4	31	$y = 0.17 + 0.99x$
Petrifilm E coli/ coliforme 37°C Diluant Ringer 1/4 and Peptone salt	59	$y = 0.15 + 1.00x$

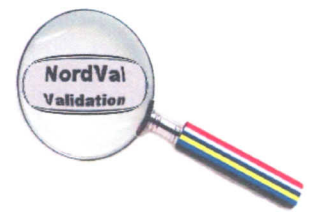
$y = \log \text{ cfu/g}$ (alternative method)
 $x = \log \text{ cfu/g}$ (reference method)

A slope close to 1.00 shows that the results obtained with Petrifilm plate incubated at 37°C are not statistically different to the results obtained by the reference method.

Selectivity

The selectivity of the method was studied by by ADRIA Normandy in 1997. 20 strains of *E. coli* were tested at 37°C, whereof 19 gave blue colonies. One strain (isolated from cheese) gave red colonies. It gave also red colony on PTX medium, and on another validated chromogenic media. 10 non *E. coli* coliform colonies were developing red colonies on the plate. Two strains of *Enterocococci* and one strain of *Staphylococcus* were not able to grow on the plate.





COLLABORATIVE STUDY RESULTS

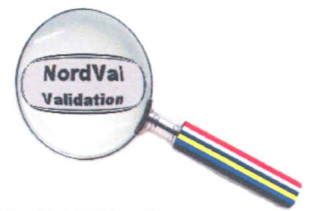
A full collaborative study has been carried out of the 3M™ Petrifilm™ *E. coli* / Coliform Count Plate, and resulted in the AOAC Official Method 991.14: Coliform and *Escherichia coli* Counts in Foods Dry Rehydratable Film. The method performance study report is published in J.AOAC **74**, 635 (1991), and a summary is given in the table below:

Total coliforms on *E. coli* count plates

Product	Level	Mean log cfu/g	sr log cfu/g	r log cfu/g	sR log cfu/g	R log cfu/g
Ground turkey	Low	1.93	0.18	0.50	0.26	0.73
	Medium	2.22	0.29	0.81	0.32	0.90
	High	2.84	0.11	0.31	0.24	0.67
Fresh mushrooms	Low	4.55	0.08	0.22	0.72	2.02
	Medium	5.28	0.60	1.68	1.00	2.80
	High	5.23	0.35	0.98	0.82	2.30
Beef with gravy	Low	1.02	0.22	0.62	0.28	0.78
	Medium	5.06	0.26	0.73	0.30	0.84
	High	2.68	0.19	0.53	0.33	0.92
Cheese	Low	0.81	0.17	0.48	0.19	0.53
	Medium	2.51	0.11	0.31	0.24	0.67
	High	3.62	0.07	0.20	0.11	0.31
Wheat flour	Low	2.64	0.24	0.67	0.88	2.46
	Medium	3.43	0.83	2.32	1.32	3.70
	High	4.06	0.93	2.60	1.58	4.42
Nut meal	Low	0.76	0.03	0.08	0.30	0.84
	Medium	1.40	0.38	1.06	0.73	2.04
	High	2.36	0.43	1.20	0.92	2.58

E. coli on *E. coli* count plates

Product	Level	Mean log cfu/g	sr log cfu/g	r log cfu/g	sR log cfu/g	R log cfu/g
Ground turkey	Low	1.42	0.32	0.90	0.32	0.90
	Medium	1.92	0.36	1.01	0.36	1.01
	High	2.61	0.10	0.28	0.23	0.64
Fresh mushrooms	Low	2.50	0.26	0.73	0.34	0.95
	Medium	3.06	0.30	0.84	0.42	1.18
	High	3.44	0.72	2.02	0.76	2.13



Product	Level	Mean log cfu/g	sr log cfu/g	r log cfu/g	sR log cfu/g	R log cfu/g
Beef with gravy	Low	0.75	0.03	0.08	0.17	0.48
	Medium	0.99	0.15	0.42	0.60	1.68
	High	1.31	0.16	0.45	0.83	2.32
Cheese	Low	0.82	0.19	0.53	0.19	0.53
	Medium	2.45	0.10	0.28	0.17	0.48
	High	3.44	0.11	0.31	0.12	0.34
Wheat flour	Low	1.76	0.52	1.46	0.74	2.07
	Medium	3.10	0.97	2.72	1.36	3.81
	High	3.55	0.82	2.30	1.44	4.03
Nut meal	Low	0.72	0.07	0.20	0.24	0.67
	Medium	1.27	0.27	0.76	0.61	1.71
	High	2.31	0.27	0.76	0.86	2.41

It might seem that wheat flour is a problematic matrix. Except for the results shadowed with grey, the results for the precision are satisfactory.

CONCLUSION

According to the comparison study no statistical differences were found between the 3M™ Petrifilm™ *E. coli* / Coliform Count Plate and the reference method ISO 16649-2. The collaborative study of the 3M™ Petrifilm™ *E. coli* / Coliform Count Plate, shows that the method is applicable for enumeration of *E. coli* and total coliforms in food. Wheat flour might however be a problematic matrix as the precision is not satisfactory.