Performance Summary

RAPID'*B.cereus* Method

Introduction

RAPID'*B.cereus* Agar is a chromogenic medium for the detection and enumeration of the *Bacillus cereus* group in food samples. The method is based on a chromogenic reaction and phospholipase activity. Typical *B. cereus* colonies develop a characteristic red color, generally surrounded with an opaque halo. The medium's selective mixture prevents the growth of interfering flora and enables the analysis of a broad range of foods. The sensitivity of the medium has been specially optimized to allow growth of all pathogenic *B. cereus* strains, even those that are difficult to culture, such as *B. cytotoxicus*. The interpretation of RAPID'*B.cereus* plates is further facilitated by the high level of contrast between the color of the colonies and the agar. The RAPID'*B.cereus* method can be used with pour-plate or spread-plate inoculation. The RAPID'*B.cereus* method has been rigorously tested and validated by an internationally recognized validation agency (Table 1).

Table 1. Validations for the RAPID'*B.cereus* method.

Validation	Certificate Number
NF Validation	BRD 07/26–03/19

Inclusivity/Exclusivity Testing

Inclusivity testing is performed to verify that the method can detect the *B. cereus* group (strains listed below), while exclusivity studies test non–*B. cereus* group strains to ensure there is no cross-reactivity. Inclusivity and exclusivity strains were cultured in Brain Heart Infusion Broth for 24 ± 3 hr at both 30 and 37°C. Decimal dilutions were made and cultured onto RAPID'*B.cereus* Agar and incubated at 30 ± 1°C for 24 ± 3 hr. All colonies were confirmed regardless of morphology. Results are shown in Table 2.

The following strains were tested in the inclusivity study:

B. cereus
B. cytotoxicus
B. mycoides
B. pseudomycoides
B. thuringiensis
B. weihenstephanensis

Table 2. Results of inclusivity/exclusivity testing.

Strains Tested	Positives Detected	Results
50 *B. cereus* group strains tested	50/50	100% inclusivity
30 non–*B. cereus* group strains tested (including other 15 *Bacillus* strains)	0/30	100% exclusivity

Method Comparison: Relative Trueness

Relative trueness is the degree of agreement between the results obtained by the reference method and the results obtained by the RAPID'*B.cereus* method on identical food samples. The following matrices have been tested (Table 3) and statistically analyzed (Table 4). The statistical analysis of the relative trueness study did not show any difference between enumeration performed with the RAPID'*B.cereus* method and the reference method.

Table 3. Matrices tested with RAPID'*B.cereus* method.

Category	Matrices Tested
Composite foods RTRH and RTE, pastries and egg products	**RTRH and RTE**: Pasta salad with beans, beef ravioli with onions, rice salad, tabbouleh, pasta salad with tomato and mozzarella, ravioli with ham, fish pâté, rice with curry, lasagna bolognese meal, rice-and-bean meal, chicken sandwich, mousse with red fruits, arancini, gratin potatoes, rice noodle meal, tuna-and-rice salad, pâté, shepherd's pie, black wheat crepe, egg blini, stew, stewed vegetables, paella **Pastries and egg products**: Strawberry tart, vanilla éclair, raspberry pastry
Dairy products Cheeses	**Cheeses**: Fresh sheep milk cheese, fresh goat milk cheese, fresh cow milk cheese, sheep milk cheese with rosemary, cow milk cheese with cumin
Egg products Unprocessed, processed	**Unprocessed**: Liquid egg yolk, liquid whole egg **Processed**: Fresh egg pasta, egg white powder, egg yolk powder, whole egg powder
Fishery products	Tuna sushi, tuna sashimi, raw salmon, salmon sushi, salmon sashimi, tuna maki, salmon maki with avocado, crunchy California roll, salmon tartare, haddock fillet, cod fillet, salmon terrine, trout terrine, shrimp, cod with dressing, surimi
Produce	Frozen leeks, frozen peas, frozen carrots
Dried cereals, fruits, nuts, seeds, and vegetables	Oregano, cinnamon, turmeric, cumin, ginger, coriander, Colombo spice mix (coriander, chili, garlic, and turmeric), dehydrated violet petals, dehydrated vegetable soup, dehydrated onion soup, dehydrated leek soup, dehydrated bean soup, dehydrated mashed potatoes, flaked almonds, muesli, oatmeal, corn flour, dried apricots, dried golden raisins, pea flour, white rice flour, dry pasta, dry cake mix, quinoa flour, protein powder, hemp protein, lupin flour, barley flour, raw bread flour, soybean flour, insect powder, cricket flour, mealworm flour, wheat flour
Infant formula and cereals	Infant formula, infant formula with probiotics, whey powder, calcium carbonate, lactose protein starch, maltodextrin, caseinate

RTE, ready to eat; RTRH, ready to reheat.

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Table 4. Statistical analysis of relative trueness.

Category	Samples Tested	$\bar{D}$	Standard Deviation	95% Lower Limit	95% Upper Limit
Spread-plate	51	–0.06	0.20	–0.45	0.33
Pour-plate	51	–0.14	0.25	–0.64	0.36

$\bar{D}$, average difference between enumeration of colony forming units (CFU) on RAPID'*B.cereus* and the reference method in log value.

Method Comparison: Accuracy Profile

The accuracy profile study is a comparative study between the results obtained by the reference method and the results of the RAPID'*B.cereus* method. This study is conducted using artificially contaminated samples in three levels (low, medium, and high) in each category. Examples of accuracy profiles are shown in Figure 1. All accuracy profiles were generated within acceptability limits for both spread- and pour-plate RAPID'*B.cereus* methods and all matrices tested.

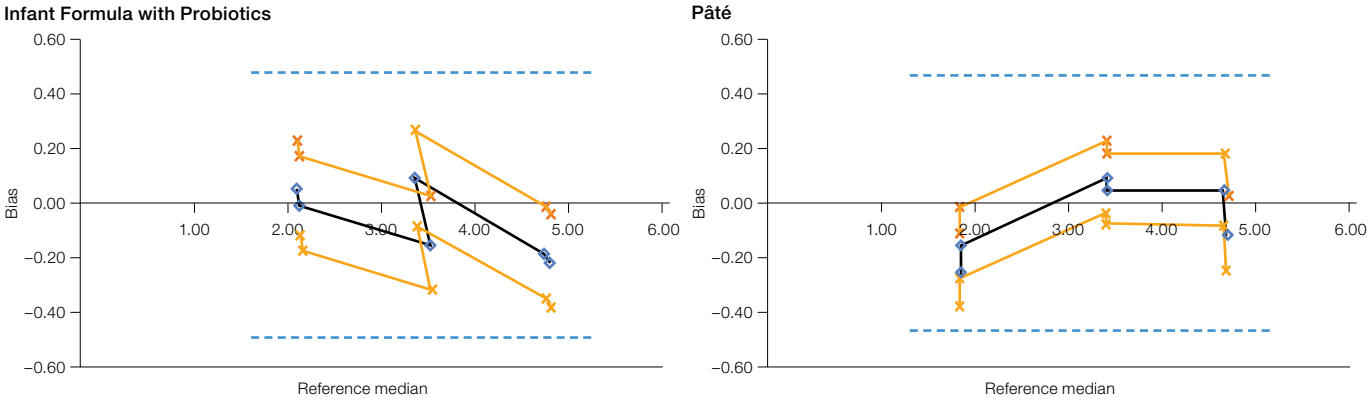

Fig. 1. Example of accuracy profiles for the RAPID'*B.cereus* spread-plate method on a dairy and RTE matrix. Bias ( ), β-ETI, expectation tolerance interval, set at 80% ( ); AL, acceptability limit ±0.5 log ( ).

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