

# Comparison Of Rapid Culture Based Methods For Detection Of Salmonella

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## Overview

**Purpose:** a comparison of rapid culture based methods for detection of Salmonella

**Methods:** Four commercially available, ISO 16140:2003 validated methods for detection of Salmonella were tested: AES Chemunex (SMS®), Bio-Rad (RAPID' Salmonella 7 hour and 24 hour methods) and the Oxoid™ Salmonella Precis™ method (Thermo Fisher Scientific).

**Results:** The AFNOR ISO 16140 validated Oxoid Salmonella Precis method proved to be a rapid, reliable and easy to use method for the detection of Salmonella from all food groups.

## Introduction

Salmonellosis remains one of the most common forms of food poisoning, caused by the ingestion of foods contaminated with bacteria of the genus *Salmonella*. Traditional culture based methods for detecting *Salmonella* are time consuming, taking up to 4 to 5 days to complete and require large numbers of sample manipulations and identification steps.

## Methods

A total of 186 food samples, of which 42 were artificially contaminated, were tested by 5 different culture based methods. Foods tested comprised raw minced beef, lettuce, raw chicken meat, unpasteurised soft cheese, liquid egg white and milk powder.

Artificially contaminated samples were spiked with serotypes of *Salmonella* typically associated with the type of food concerned. For each food type, 4 samples were artificially contaminated with <10 CFU per 25 g sample and 2 samples with between 10 and 50 CFU per 25 g sample. For liquid egg samples, 8 samples were inoculated at the low level and 4 samples at the high inoculum level. Milk powder samples (n=6) were artificially contaminated using Reference Material Capsules (IRMM) at <5 CFU per 25 g.

All spiked samples were stored at 2 to 8°C for 72 h to habituate/stress the *Salmonella* prior to testing. Samples were tested according to the ISO 6579:2002 method and the ISO 16140:2003 validated protocols; where possible pre-enrichment media were shared if common to more than one method. Presumptive positive colonies for all methods were subcultured onto Nutrient Agar prior to confirmation with the BAX® Salmonella assay (DuPont Qualicon).

**FIGURE 1. Time taken to obtain results for all methods tested**

DAY	1	2	3	4
<b>Oxoid salmonella Precis (1 broth and 1 plate)</b>	Inoculate ONE Broth Salmonella. Incubate for 18±2 h.	Inoculate Brilliance Salmonella Agar. Incubate for 24±2 h. Confirm presumptive positive results.	TOTAL: 2 days, including confirmation (Salmonella latex). Samples transfers: 1	
<b>AES SMS Method (1 broth and 2 plates)</b>	Inoculate BPW. Incubate for 18±2 h.	Inoculate SMS Agar. Incubate for 22±2 h.	Inoculate SALSA plate. Incubate for 24±2 h. Confirm presumptive positive results.	TOTAL: 3 days, including confirmation. Sample transfers: 5
<b>Bio-Rad 7 h Method (2 broths and 1 plate)</b>	Inoculate BPW. Incubate for 18±2 h.	Inoculate RVS Broth. Incubate for 7±2 h.	Inoculate RAPID' Salmonella plate. Incubate for 24±2 h. Confirm presumptive positive results.	TOTAL: 3 days, including confirmation. Sample transfers: 2
<b>Bio-Rad 24 h Method (2 broths and 1 plate)</b>	Inoculate BPW. Incubate for 18±2 h.	Inoculate RVS Broth. Incubate for 24±2 h.	Inoculate RAPID' Salmonella plate. Incubate for 24±2 h. Confirm presumptive positive results.	TOTAL: 4 days, including confirmation. Sample transfers: 2
<b>ISO 6579-2005 (3 broths and 4 plates)</b>	Inoculate BPW. Incubate for 18±2 h.	Inoculate RVS Broth and MKTTn Broth. Incubate for 24±3 h.	Inoculate each enrichment broth onto XLD and mBGA Agar. Incubate for 24±2 h. Confirm presumptive positive results.	Subculture suspect colonies onto Nutrient Agar and incubate for 24±2 h. Use ISO recommended tests to confirm presumptive positive results. TOTAL: 5-6 days, including confirmation. Samples transfers: 7

## Results

The Oxoid Salmonella Precis method performed well compared to the other commercial rapid culture based methods, and against the ISO 6579:2002 method with the range of food types tested. The recovery of *Salmonella* from egg white was only achieved with the Precis method; all other methods were unable to recover from either the low or high spike level samples.

Recovery of *Salmonella* from the 5 other food matrices was comparable between all of the methods, with the exception of the SMS method which gave four false presumptive positive results (3 with soft cheese and 1 with dried milk powder). In total, the Salmonella Precis method isolated *Salmonella* from 26 of the 42 spiked samples and one additional naturally contaminated, unspiked sample.

As Figure 1 shows, there is a considerable difference in the time required to perform each of the methods and obtain results. The Salmonella Precis method can give a confirmed positive result after 38 hours. The remaining methods all take at least 3 days to deliver a confirmed result.

The number of manipulations and sample transfers are also significantly improved with the Salmonella Precis method, with inoculation from ONE Broth-Salmonella straight onto a single plate of *Brilliance™* Salmonella Agar followed by direct confirmation by a simple latex agglutination test (Oxoid Salmonella Latex Test - FT0203A).

**TABLE 1: Results for spiked samples for each method**

	SMS	RAPID' 7 h	RAPID' 24 h	Precis	ISO
<b>Presumptive positives</b>	27	20	24	27	25
<b>Confirmed positives</b>	23	20	24	27	24

## Conclusion

The AFNOR ISO 16140 validated Oxoid Salmonella Precis method is a rapid, reliable and easy to use method for the detection of Salmonella from all food groups. The format of one broth and one plate with a simple confirmation step significantly reduces manipulations as well as time to a confirmed result.