

# TEST REPORT

## INTERLABORATORY COMPARISON ON THE DETECTION OF *SALMONELLA* spp. IN FOOD organised by CRL-*Salmonella* FOOD STUDY I - 2006

Laboratory code	
Laboratory name	
Address	
Country	
Date of arrival of the parcels	..... - ..... – 2006
Start time of storage at - 20°C (capsules)	Date:..... Time:.....
Start time of storage at 5°C (meat)	Date:..... Time:.....
Parcels damaged?	YES                      NO
Starting date testing	..... - ..... – 2006

Is your laboratory accredited or certified for the determination of <i>Salmonella</i> . If yes, according to which system (e.g. ISO 17025) ? If no, are you planning to become accredited or certified in the near future ?	
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**PRE-ENRICHMENT – Buffered Peptone Water (BPW) (I)****Medium information BPW**

What did you use to prepare the BPW?

Individual ingredients

Dehydrated medium

Ready-to-use medium

**In case of dehydrated or ready-to-use medium , give information on the manufacturer of BPW**

Name

Code number

Batch number

Expire date

**Specific data of composition of BPW medium. What is the concentration of the following compounds in 1000 ml water:**

Enzymatic digest of casein

Sodium chloride

Disodium hydrogen phosphate dodecahydrate  
( $\text{Na}_2\text{HPO}_4 \cdot 12\text{H}_2\text{O}$ )Potassium dihydrogen phosphate ( $\text{KH}_2\text{PO}_4$ )**Preparation of BPW**

Date of preparation

..... - ..... - 2006

pH after preparation

....., measured at ..... °C

pH at the day of use

....., measured at ..... °C

Did you perform quality control of BPW?

yes

no

**PRE-ENRICHMENT – Buffered Peptone Water (BPW) (II)****Prewarming time and temperature of the BPW**

At the start	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the end	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C

**Incubation time and temperature for dissolving the capsules**

At the start	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the end	time: ..... h ..... min temperature incubator: ..... °C

**Incubation time and temperature for pre-enrichment ( 18 ± 2 ) hrs  
after adding the meat**

At the start	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the end	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C

**SELECTIVE ENRICHMENT - Rappaport Vassiliadis Soya medium (RVS) (I)****Medium information RVS**

What did you use to prepare the RVS?

Individual ingredients

Dehydrated medium

Ready-to-use medium

**In case of dehydrated or ready-to-use medium , give information on the manufacturer of RVS**

Name

Code number

Batch number

Expire date

**Specific data of composition of RVS medium. What is the concentration of the following compounds in 1000 ml water:**

Enzymatic digest of Soya

Sodium chloride (NaCl)

Potassium dihydrogen phosphate ( $\text{KH}_2\text{PO}_4$ )Dipotassium hydrogen phosphate ( $\text{K}_2\text{HPO}_4$ )Magnesium chloride anhydrous ( $\text{MgCl}_2$ )Magnesium chloride hexahydrate  
( $\text{MgCl}_2 \cdot 6\text{H}_2\text{O}$ )

Malachite green oxalate

**Preparation of RVS**

Date of preparation

..... - ..... - 2006

pH after preparation

....., measured at ..... °C

pH at the day of use

....., measured at ..... °C

Did you perform quality control of RVS?

yes

no

**SELECTIVE ENRICHMENT - Rappaport Vassiliadis Soya medium (RVS) (II)**

<b>Incubation time and temperature for selective enrichment</b>	
At the start of the first period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the end of the first period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the start of the second period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the end of the second period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C

<b>SELECTIVE ENRICHMENT - Muller Kauffmann Tetra Thionate + novobiocin (MKTTn) (I)</b>
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<b>Medium information MKTTn</b>
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What did you use to prepare the MKTTn?
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Individual ingredients
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Dehydrated medium
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Ready-to-use medium
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<b>In case of dehydrated or ready-to-use medium , give information on the Manufacturer of MKTTn</b>
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Name	
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Code number	
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Batch number	
--------------	--

Expire date	
-------------	--

<b>Specific data of composition of MKTTn medium. What is the concentration of the following compounds in 1000 ml water:</b>
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Meat extract	
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Enzymatic digest of casein	
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Sodium chloride (NaCl)	
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Calcium carbonate (CaCO <sub>3</sub> )	
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Sodium thiosulfate pentahydrate (Na <sub>2</sub> S <sub>2</sub> O <sub>3</sub> ·5H <sub>2</sub> O)	
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Ox bile for bacteriological use	
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Brilliant green	
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Iodine	
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Potassium iodide (KI)	
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Novobiocin	
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<b>SELECTIVE ENRICHMENT - Muller Kauffmann Tetra Thionate + novobiocin (MKTTn) (II)</b>
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<b>Preparation of MKTTn</b>	
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Date of preparation	..... - ..... - 2006
pH after preparation	....., measured at ..... °C
pH at the day of use	....., measured at ..... °C
Did you perform quality control of MKTTn?	yes                  no

<b>Incubation time and temperature for selective enrichment</b>	
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At the start of the first period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the end of the first period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the start of the second period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the end of the second period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C

<b>SELECTIVE ENRICHMENT - Modified Semi solid Rappaport Vassiliadis medium (MSRV) (I)</b>
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<b>Medium information MSRV</b>
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What did you use to prepare the MSRV?
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Individual ingredients
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Dehydrated medium
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Ready-to-use medium
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<b>In case of dehydrated or ready-to-use medium , give information on the manufacturer of MSRV</b>
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Name	
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Code number	
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Batch number	
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Expire date	
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<b>Specific data of composition of MSRV medium. What is the concentration of the following compounds in 1000 ml water:</b>
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Enzymatic digest of casein	
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Acid hydrolysate of casein	
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Sodium chloride (NaCl)	
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Potassium dihydrogen phosphate (KH <sub>2</sub> PO <sub>4</sub> )	
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Magnesium chloride anhydrous (MgCl <sub>2</sub> )	
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Malachite green oxalate	
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Agar	
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Novobiocin	
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<b>Preparation of MSRV</b>
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Date of preparation	..... - ..... - 2006
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pH after preparation	....., measured at ..... °C
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pH at the day of use	....., measured at ..... °C
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Did you perform quality control of MSRV?	yes	no
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<b>SELECTIVE ENRICHMENT - Modified Semi solid Rappaport Vassiliadis medium (MSRV) (II)</b>
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<b>Incubation time and temperature for selective enrichment</b>	
At the start of the first period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the end of the first period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the start of the second period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the end of the second period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C

**OWN SELECTIVE ENRICHMENT - Selective medium, routinely used in your laboratory (I)**

**If you use more selective media, please write these on an annex.**

Medium:

**Medium information**

What did you use to prepare the medium?

Individual ingredients

Dehydrated medium

Ready-to-use medium

**In case of dehydrated or ready-to-use medium , give information on the manufacturer of the medium**

Name

Code number

Batch number

Expire date

**Specific data of composition of the medium. What is the concentration of the compounds in 1000 ml water:**


**Preparation of the medium**

Date of preparation

..... - ..... - 2006

pH after preparation

....., measured at ..... °C

pH at the day of use

....., measured at ..... °C

Did you perform quality control of the medium?

yes                  no

<b>OWN SELECTIVE ENRICHMENT - Selective medium, routinely used in your laboratory (II)</b>
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<b>Further details concerning the medium</b>	
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Volume of the medium per jar/tube in ml	
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Inoculation volume of BPW	
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Prescribed incubation temperature in °C	
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<b>Incubation time and temperature for own selective enrichment</b>	
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At the start of the first period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
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At the end of the first period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
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At the start of the second period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
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At the end of the second period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
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**FIRST AND SECOND ISOLATION - Xylose Lysine Desoxycholate medium (XLD) (I)****Medium information XLD**

What did you use to prepare the XLD ?

Individual ingredients

Dehydrated medium

Ready-to-use medium

**In case of dehydrated or ready-to-use medium , give information on the manufacturer of XLD**

Name

Code number

Batch number

Expire date

**Specific data of composition of XLD medium. What is the concentration of the following compounds in 1000 ml water:**

Xylose

L-lysine hydrochloride

Lactose

Sucrose

Sodium chloride (NaCl)

Yeast extract powder

Phenol red

Agar

Sodium desoxycholate

Sodium thiosulfate

Iron(III) ammonium citrate

**FIRST AND SECOND ISOLATION - Xylose Lysine Desoxycholate medium (XLD) (II)****Preparation of XLD**

Date of preparation	..... - ..... - 2006
pH after preparation	....., measured at ..... °C
pH at the day of use	....., measured at ..... °C
Did you perform quality control of XLD ?	yes                      no

**Incubation time and temperature for isolation**

At the start of the first period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the end of the first period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the start of the second period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the end of the second period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C

**FIRST AND SECOND ISOLATION – Second Isolation medium. (I)****Give information on the second isolation medium.**

Name of the medium

Prescribed incubation temperature in °C

**Medium information of the second isolation medium**

What did you use to prepare the second isolation medium?

Individual ingredients

Dehydrated medium

Ready-to-use medium

**In case of dehydrated or ready-to-use medium , give information on the manufacturer of the second isolation medium**

Name

Code number

Batch number

Expire date

**Specific data of composition of the second isolation medium. What is the concentration of the compounds in 1000 ml water:**


**FIRST AND SECOND ISOLATION – Second Isolation medium. (II)****Preparation of the second isolation medium**

Date of preparation	..... - ..... - 2006
pH after preparation	....., measured at ..... °C
pH at the day of use	....., measured at ..... °C
Did you perform quality control ?	yes                      no

**Incubation time and temperature for isolation**

At the start of the first period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the end of the first period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the start of the second period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the end of the second period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C

<b>FIRST AND SECOND ISOLATION – Own Isolation medium routinely used in your lab. (I)</b>
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<b>If you use more selective media, please write these on an annex.</b>
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Name of the medium	
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Prescribed incubation temperature in °C	
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<b>Medium information of your own medium</b>
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What did you use to prepare your own medium ?
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Individual ingredients
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Dehydrated medium
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Ready-to-use medium
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<b>In case of dehydrated or ready-to-use medium , give information on the manufacturer of your own medium</b>
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Name	
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Code number	
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Batch number	
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Expire date	
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<b>Specific data of composition of your own medium. What is the concentration of the compounds in 1000 ml water:</b>
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<b>FIRST AND SECOND ISOLATION – Own Isolation medium routinely used in your lab. (II)</b>
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<b>Preparation of your own medium</b>	
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Date of preparation	..... - ..... - 2006
pH after preparation	....., measured at ..... °C
pH at the day of use	....., measured at ..... °C
Did you perform quality control ?	yes                      no

<b>Incubation time and temperature for isolation</b>	
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At the start of the first period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the end of the first period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the start of the second period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C
At the end of the second period	Date: ..... - ..... - 2006 time: ..... h ..... min temperature incubator: ..... °C

**CONFIRMATION – Nutrient agar (I)**

**Did you streak the colonies on Nutrient agar before starting confirmation?**

yes                                  no    If yes give further information on nutrient agar below

**Medium information Nutrient agar**

What did you use to prepare the nutrient agar ?

Individual ingredients

Dehydrated medium

Ready-to-use medium

**In case of dehydrated or ready-to-use medium , give information on the manufacturer of the nutrient agar**

Name

Code number

Batch number

Expire date

**Specific data of composition of nutrient agar medium. What is the concentration of the compounds in 1000 ml water:**


**Preparation of the nutrient agar**

Date of preparation                  ..... - ..... - 2006

pH after preparation                  ....., measured at ..... °C

pH at the day of use                  ....., measured at ..... °C

Did you perform quality control of agar ?    yes                          no

**BIOCHEMICAL CONFIRMATION****Manufacturer of the TSI agar**

Name	
Code number	
Batch number	
Expire date	
pH of the medium:.....	Measured at.....°C Date ..... - ..... 2006

**Manufacturer of the urea agar**

Name	
Code number	
Batch number	
Expire date	
pH of the medium:.....	Measured at.....°C Date ..... - ..... 2006

**Manufacturer of the l-Lysine decarboxylation medium**

Name	
Code number	
Batch number	
Expire date	
pH of the broth:.....	Measured at.....°C Date ..... - ..... 2006

**Manufacturer of other confirmation tests - .....**

Name	
Code number	
Batch number	
Expire date	
pH of the medium :.....	Measured at.....°C Date ..... - ..... 2006

**DETECTION BY PCR**

**General questions**

Is the PCR used commercially available ?	Yes No
If yes, name of PCR, manufacturer and batch used in the study:	
Is the PCR validated ? If yes, for which matrix/matrices ?	Yes Matrix/Matrices :..... No
How much samples did you test for <i>Salmonella</i> using this PCR in 2005 ?	
At what moment did you start with the extraction/detection?	before pre-enrichment in BPW after pre-enrichment in BPW
Volume of pre-enrichment broth used for extraction	.....
Volume of DNA-sample obtained from extraction	.....
Volume of DNA-sample added to PCR-mixture	.....

**Composition of PCR-mixture**

Compound	Volume per sample	Manufacturer and batch of specific compound

Name of thermocycler	
Write down the cycles	
What kind of detection system is used	

Table 1: Results of isolation using **RVS** (dish numbers 1-25)

sample no.	RVS 24 hours						RVS 48 hours					
	XLD		Second isolation medium		Own isolation medium		XLD		Second isolation medium		Own isolation medium	
	Col <sup>a</sup>	Sal <sup>b</sup>	Col	Sal	Col	Sal	Col	Sal	Col	Sal	Col	Sal
1												
2												
3												
4												
5												
6												
7												
8												
9												
10												
11												
12												
13												
14												
15												
16												
17												
18												
19												
20												
21												
22												
23												
24												
25												

Col<sup>a</sup> = number of colonies used for confirmation  
 Sal<sup>b</sup> = number of colonies confirmed as *Salmonella*

Table 1 (continued): Results of isolation using **RVS** (dish numbers C1-C12)

sample no.	RVS 24 hours						RVS 48 hours					
	XLD		Second isolation medium		Own isolation medium		XLD		Second isolation medium		Own isolation medium	
	Col <sup>a</sup>	Sal <sup>b</sup>	Col	Sal	Col	Sal	Col	Sal	Col	Sal	Col	Sal
C1												
C2												
C3												
C4												
C5												
C6												
C7												
C8												
C9												
C10												
C11												
C12												

Col<sup>a</sup> = number of colonies used for confirmationSal<sup>b</sup> = number of colonies confirmed as *Salmonella*

Table 2: Results of isolation using MKTTn (dish numbers 1-25)

sample no.	MKTTn 24 hours						MKTTn 48 hours					
	XLD		Second isolation medium		Own isolation medium		XLD		Second isolation medium		Own isolation medium	
	Col <sup>a</sup>	Sal <sup>b</sup>	Col	Sal	Col	Sal	Col	Sal	Col	Sal	Col	Sal
1												
2												
3												
4												
5												
6												
7												
8												
9												
10												
11												
12												
13												
14												
15												
16												
17												
18												
19												
20												
21												
22												
23												
24												
25												

Col<sup>a</sup> = number of colonies used for confirmation  
 Sal<sup>b</sup> = number of colonies confirmed as *Salmonella*

Table 2 (continued): Results of isolation using **MKTTn** (dish numbers C1-C12)

sample no.	MKTTn 24 hours						MKTTn 48 hours					
	XLD		Second isolation medium		Own isolation medium		XLD		Second isolation medium		Own isolation medium	
	Col <sup>a</sup>	Sal <sup>b</sup>	Col	Sal	Col	Sal	Col	Sal	Col	Sal	Col	Sal
C1												
C2												
C3												
C4												
C5												
C6												
C7												
C8												
C9												
C10												
C11												
C12												

Col<sup>a</sup> = number of colonies used for confirmationSal<sup>b</sup> = number of colonies confirmed as *Salmonella*



Table 3: Results of isolation using **MSRV** (dish numbers 1-25)

sample no.	MSRV 24 hours						MSRV 48 hours					
	XLD		Second isolation medium		Own isolation medium		XLD		Second isolation medium		Own isolation medium	
	Col <sup>a</sup>	Sal <sup>b</sup>	Col	Sal	Col	Sal	Col	Sal	Col	Sal	Col	Sal
1												
2												
3												
4												
5												
6												
7												
8												
9												
10												
11												
12												
13												
14												
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16												
17												
18												
19												
20												
21												
22												
23												
24												
25												

Col<sup>a</sup> = number of colonies used for confirmation  
 Sal<sup>b</sup> = number of colonies confirmed as *Salmonella*

Table 3 (continued): Results of isolation using **MSRV** (dish numbers C1-C12)

sample no.	MSRV 24 hours						MSRV 48 hours					
	XLD		Second isolation medium		Own isolation medium		XLD		Second isolation medium		Own isolation medium	
	Col <sup>a</sup>	Sal <sup>b</sup>	Col	Sal	Col	Sal	Col	Sal	Col	Sal	Col	Sal
C1												
C2												
C3												
C4												
C5												
C6												
C7												
C8												
C9												
C10												
C11												
C12												

Col<sup>a</sup> = number of colonies used for confirmationSal<sup>b</sup> = number of colonies confirmed as *Salmonella*

Table 4: Results of isolation using own selective enrichment (dish numbers 1-25)

sample no.	Own selective enrichment 24 hours						Own selective enrichment 48 hours					
	Col <sup>a</sup>	Sal <sup>b</sup>	Col	Sal	Col	Sal	Col	Sal	Col	Sal	Col	Sal
1												
2												
3												
4												
5												
6												
7												
8												
9												
10												
11												
12												
13												
14												
15												
16												
17												
18												
19												
20												
21												
22												
23												
24												
25												

Col<sup>a</sup> = number of colonies used for confirmation  
 Sal<sup>b</sup> = number of colonies confirmed as *Salmonella*

Table 4 (continued): Results of isolation using own selective enrichment (dish numbers C1-C12)

sample no.	Own selective enrichment 24 hours						Own selective enrichment 48 hours					
	Col <sup>a</sup>	Sal <sup>b</sup>	Col	Sal	Col	Sal	Col	Sal	Col	Sal	Col	Sal
C1												
C2												
C3												
C4												
C5												
C6												
C7												
C8												
C9												
C10												
C11												
C12												

Col<sup>a</sup> = number of colonies used for confirmationSal<sup>b</sup> = number of colonies confirmed as *Salmonella*

Table 5: Results of detection using PCR (dish numbers 1-25)

sample no.	PCR + or -	
		no.
1		C1
2		C2
3		C3
4		C4
5		C5
6		C6
7		C7
8		C8
9		C9
10		C10
11		C11
12		C12
13		
14		
15		
16		
17		
18		
19		
20		
21		
22		
23		
24		
25		

Comment(s) on operational details that might influence the test results:

Name of person (s) carrying out the first Food interlaboratory Comparison study (2006)	
Date and signature	

Name of person in charge	
Date and signature	