Evaluation of different analytical methods for detection of monophasic variants of *S.* Typhimurium during process and shelf-life of dried sausages.

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Monophasic variants of Salmonella Typhimurium (4,12:i:- and 4,5,12:i:-) have been increasingly reported in France and in Europe in the past few years. These variants had been identified in humans, animals and different foodstuffs, especially in pork products. During the foodborne outbreaks involving these variants in 2010 and 2011, despite the absence of detection in food samples, the epidemiological and food investigations suggested dried pork sausages as the source of the outbreak. The aims of this study was to evaluate the detection capacity of various analytical methods for 2 monophasic variants of S. Typhimurium and 2 control strains (Salmonella Derby and Salmonella Typhimurium) in dried sausages.

Materials and Methods

Detection during dried sausages process

- One-strain contaminated batches
- Four strains : 2 monophasic variants of *S.* Typhimurium (4,12:i:- and 4,5,12:i:-), 2 control strains: *S.* Derby, *S.* Typhimurium.
- Detections on the same sausage by 11 alternative validated methods and the reference method (NF EN ISO 6579)
- During ripening, drying and shelf-life of dried sausages.

Complementary protocol

- Derived from the standard method
- From enriched broth (BPW or specific media)

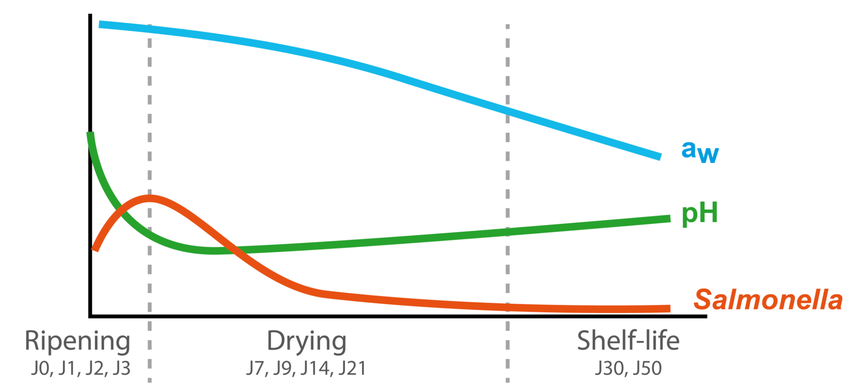

Figure 1 – Physico-chemical and *Salmonella* evolution during dried sausages process and sampling date.

Results

Detectability of the strains

- The four strains were detected by 73% of the alternative validated methods during process and shelf-life.
- The complementary protocol used to confirm samples status help to avoid false-negative results of standard and alternative validated methods.
- For cultural methods, the complementary protocol was less efficient because of selective agents in the enrichment broth.

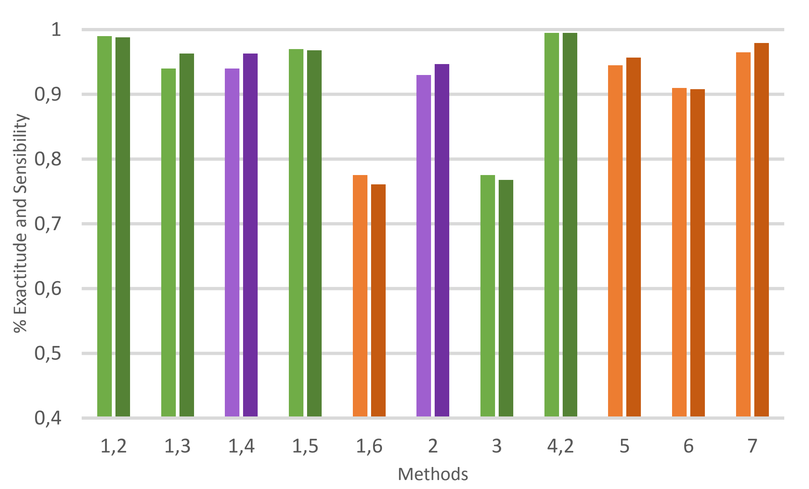

Figure 2 – Exactitude and sensitivity of the different methods tested on the four strains (green : molecular methods, violet : immune enzymatic methods and orange : cultural methods/ light: exactitude and dark: sensibility).

Table 1 – Alternative validated methods tested which allow detection of *S.* Typhimurium monophasic variants on dried sausages.

	Methods (suppliers)*
Molecular methods	IQ-Check Salmonella (Biorad) Microseq Salmonella spp detection (Life Technologies) Transia Plate *Salmonella* Gold (Bio Control)
Immune enzymatic methods	Assurance GDS *Salmonella* (Bio Control) Vidas Up *Salmonella* (Biomérieux)
Cultural methods	IRIS *Salmonella* (Biokar) Rapid *Salmonella* (Biorad) *Salmonella* Precis (Oxoid)

*****All of the alternative validated methods by AFNOR at the date of the project were not tested. The above list contains only methods tested and which give good detection results.

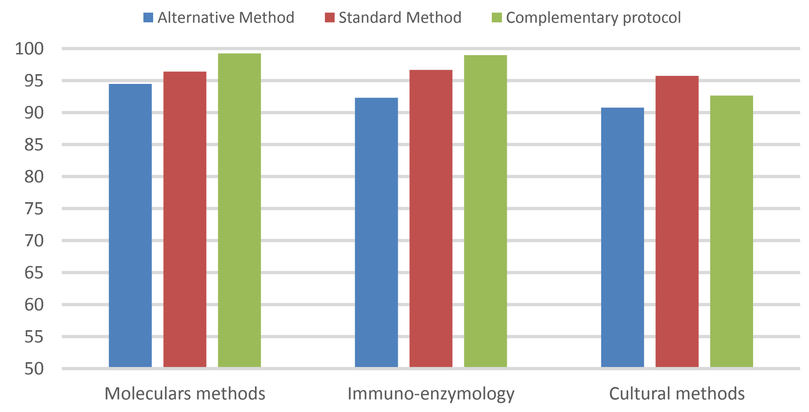

Figure 3 – Results of the detections of the 4 *Salmonella* strain by the different methods on dried sausages samples during process and shelf-life.

Conclusion

- **Detection of *Salmonella* stressed cells in dried sausages and especially of *S.* Typhimurium monophasic variants is not efficient following the strains and the sampling dates**
- **Complementary protocol allows to detect stressed *Salmonella* cells in negative samples analyzed by either alternative or reference methods**
- **Strains and matrices variability have to be taken into account to evaluate the use of detection methods.**

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