Pathogen screening in pooled food samples: Applications, implications, and observations from proficiency testing

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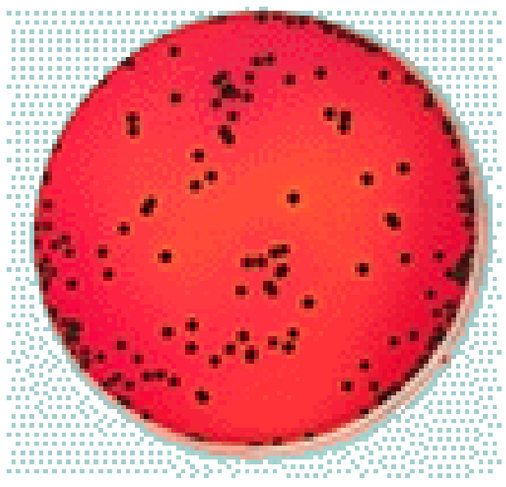
Purpose of pooled samples in microbiological testing

Laboratories testing food for the presence of pathogens generally test 25g samples, with the specification that pathogens should be absent in 25g. Fortunately most food samples tested do not contain any pathogens, but this means that a lot of negative tests are being performed daily, wasting time and consumables such as reagents and agar. A number of 25g samples (up to 15) can therefore be 'pooled' into larger volumes of 125g or 375g for testing.

If the 'pooled' sample is negative then it can be assumed that the component 25g samples were also negative. If the 'pooled' sample is positive, then samples will need to be retaken and retested to identify the source of the positive result.

The concept of pooled samples originated in WWII for testing for syphilis in US soldiers (Dorfman 1943). These days it is commonly used to screen donated blood for viruses including hepatitis and HIV, but also increasingly used in food pathogen testing. One of the most commonly tested pathogens in food is *Salmonella* .

The main advantage of pooled samples is savings in cost and time. The drawback is the need to consider whether pooling samples reduces the level of contamination to below the detection limit.



Salmonella **on XLD agar**

Pooled samples in LGC PT schemes

LGC provide a number of samples for pathogen testing, including *Salmonella*, Campylobacter, Listeria, Cronobacter, Yersinia etc usually in 25g food matrix. However demand for larger sample quantities due to increased use of 'pooled' samples led to new samples being introduced in the past 2 years;

PT-MC-40

Salmonella in 375g skimmed milk powder

PT-MC-43

Cronobacter in 375g skimmed milk powder

PT-CT-720

Salmonella in 375g cocoa powder

PT-SC-02M

E. coli O157 & STEC in 375g meat powder

Sample format

The format of each sample is a vial plus matrix, to provide a realistic sample to test whilst also ensuring stability of the organisms at low levels during transport. Participants are provided with comprehensive instructions for performing the PT. See Figure 1.

Results

The PT results for *Salmonella* in pooled samples over time are entirely consistent with results for unpooled samples, with 95% of results correctly detecting Salmonella (when present) from the pooled sample. See Figure 2.

Conclusion

Introducing pooled samples onto a PT scheme enables laboratories performing this kind of testing to assess their results and compare with peers, providing reassurance to participants that the methods used are producing the expected result despite the lower inoculum level and greater challenge with testing pooled samples.

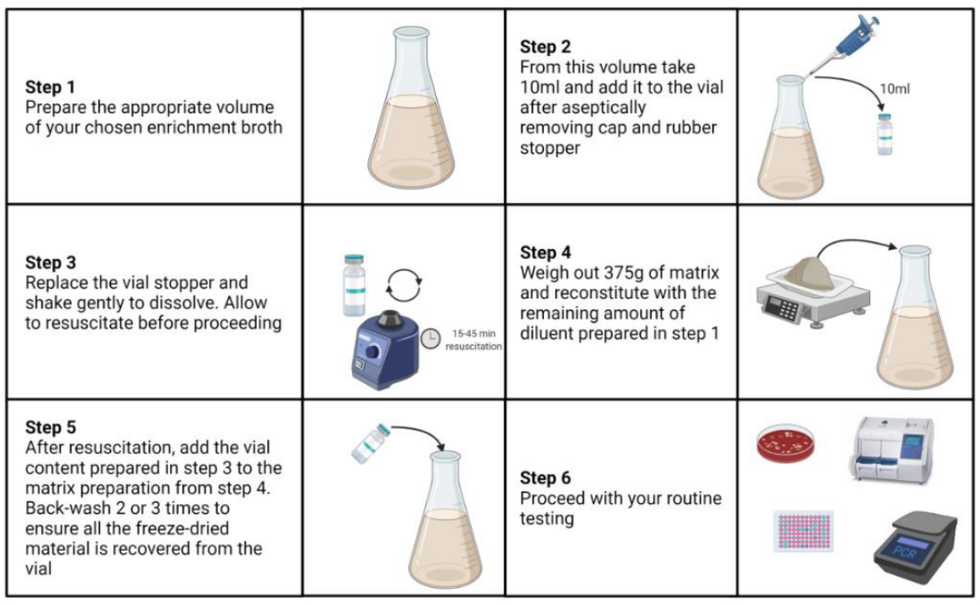


Figure 1: PT instructions

Year	Round	Sample Contents	No. of results	Inoculum level cfu/375g	% Correct
Sep-18	269	*S.* Nottingham	38	11	95.0
Mar-19	275	*S.* Manchester	23	12	94.3
Sep-19	281	*S.* Tranoroa	71	15	94.4
Mar-20	287	*S.* Bracknell	93	11	95.7
Sep-20	293	*S.* Panama, *E. coli*	65	10	87.7
Mar-21	299	*S.* Bovis morbifi-cans	110	89	99.1
Sep-21	305	*S.* Montevideo	77	79	100.0
Mar-22	311	*S.* Manchester	108	90	98.1
		Total/Average	**585**	**40**	**95.5**

Figure 2: Results table