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MICROGEN BIOPRODUCTS

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PULSIFIER® The Difference is Now Clear

It is over a year since the Pulsifier® was launched by Microgen Bioproducts. In that time distributors have been appointed in all major international markets and a number of studies using the instrument have been completed or have commenced. This edition of the Microlab News will provide an update on some of the experiences of early customers during the validation of the Pulsifier® for routine sample processing tasks and the development of new / novel applications.

The instrument has been validated by key opinion leaders such as Professor Daniel Fung,Kansas State University, USA and has been used in various applications including isolation of parasites from food samples and plant pathogens from crops. In all of these studies the resulting suspensions contain less food matrix debris than samples prepared using Paddle-type blenders. Furthermore studies on the rate of processing with the Pulsifier® suggest that the detachment process is more rapid than with a Paddle-type blender.

In this issue we will summarise a number of studies including:

- **Comparison of the Pulsifier® and Stomacher™ for recovering micro-organisms in vegetables.**
- **An Evaluation on Pulsifying for Efficiency of Microbe recovery.**
- **Comparison of the Pulsifier® and Stomacher™ for the recovery of pathogenic bacteria from food samples.**

Figure 1. Pulsifier® showing pulsification ring.

Figure 2. Pulsifier® showing door release for easy cleaning.

Comparison of the Pulsifier® and the Stomacher for recovering micro-organisms in vegetables. *Wu V.C.H., Jitareerat P. & Fung Y.C.* (2003) Journal of Rapid Methods and Automation in microbiology **11**: 145-152

The treatment of 30 different vegetables by the Pusifier® for 30 seconds and the Stomacher™ for 60 seconds showed that the total viable counts and coliform counts were almost identical with less than 0.2 Log CFU/g difference and a ratio of approximately 1.00 between the two methods, Table 1. However, the diluents from the Pulsified samples were much clearer (Optical Density) of 0.052, with less total soluble solids (SS) of 0.3% and slightly higher pH (6.56) than Stomached samples, which a produced a more turbid liquid (Optical Density) of 0.237, with more total soluble solids of 0.55% and a lower pH (6.35).

Table 1. Total Viable Counts from a range of vegetables prepared using the Pulsifier® and Stomacher™

Vegetable	Log CFU/g		P/S
	Pul	Stom	Ratio
Head Lettuce	4.78	5.09	0.94
Onions Green	6.20	6.15	1.00
Fresh Carrot	5.83	5.85	1.00
Radish-radicchio	6.07	6.04	1.00
Parsley	5.96	6.16	0.97
Green Leaf Lettuce	5.85	6.13	0.95
Romaine	5.82	4.03	1.44
Red Leaf	6.12	6.12	1.00
Boston Lettuce	5.99	5.79	1.03
Spinach	5.76	6.11	0.94
Endive	6.11	4.82	1.27
Orange Pepper	4.14	4.36	0.95
Cucumber	6.03	4.25	1.42
Celery Hearts	5.04	4.39	1.15
Broccoli	5.61	4.90	1.14
Cauliflower	3.29	5.31	0.62
Snow peas	4.91	5.70	0.86
Turnips	5.41	5.58	0.97
Cilantro	6.07	6.10	1.00
Zucchini squash	6.20	6.01	1.03
Rhubarb	5.65	4.38	1.29
Parsnips	6.12	6.12	1.00
Asparagus	6.10	5.72	1.07
Cabbage	3.54	2.31	1.53
Chinese Cabbage	5.74	5.84	0.98
Green Beans	5.34	4.21	1.27
Tomato (Hot House)	2.01	2.31	0.87
Potato (Baking)	5.75	5.48	1.05
Egg Plant	5.64	5.91	0.95
Average			**1.02**

Table 2. Total Viable Counts from a range of vegetables prepared using the Pulsifier® and Stomacher™

Vegetable	OD		Total SS	
	Pul	Stom	Pul	Stom
Head Lettuce	0.01	0.073	0.2	0.2
Onions Green	0.031	0.148	0.4	0.6
Fresh Carrot	0.016	0.365	0.25	0.9
Radish-radicchio	0.021	0.037	0.1	0.4
Parsley	0.033	0.322	0.15	0.4
Green Leaf Lettuce	0.063	0.192	0.05	0.4
Romaine	0.063	0.222	0.15	0.4
Red Leaf	0.138	0.32	0.4	0.3
Boston Lettuce	0.047	0.177	0.4	0.3
Spinach	0.282	0.56	0.4	0.4
Endive	0.102	0.283	0.4	0.5
Orange Pepper	0.001	0.125	0.22	0.7
Cucumber	0.003	0.095	0.4	0.5
Celery Hearts	0.003	0.123	0.2	0.5
Broccoli	0.024	0.271	0.4	0.7
Cauliflower	0.004	0.078	0.3	0.5
Snow peas	0.012	0.227	0.5	1.0
Turnips	0.001	0.019	0.3	0.6
Cilantro	0.14	0.765	0.1	0.7
Zucchini squash	0.003	0.094	0.3	0.5
Rhubarb	0.079	0.224	0.3	0.5
Parsnips	0.284	1.158	0.5	0.8
Asparagus	0.041	0.154	0.4	0.7
Cabbage	0.001	0.034	0.2	0.4
Chinese Cabbage	0.011	0.01	0.22	0.2
Green Beans	0	0.232	0.2	0.7
Tomato (H/ House)	0.023	0.05	0.5	0.4
Potato (Baking)	0.018	0.247	0.3	0.6
Egg Plant	0.082	0.47	0.22	0.8
Average	**0.052**	**0.237**	**0.3**	**0.56**

The data in this evaluation supports the fact that the Pulsified samples produce equivalent viable cell counts to samples prepared using the Stomacher™, whilst producing clearer diluents with less total soluble solids. This is advantageous for further microbiological manipulations such as the use of pipetting, the use of Petrifilm™, Polymerase Chain Reaction, ATP evaluation and Immunological testing.

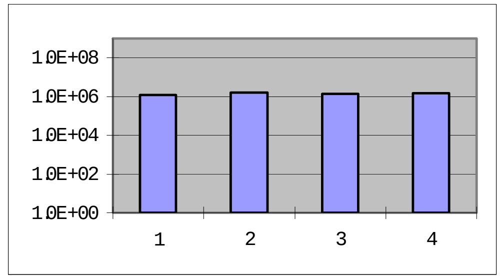
An Evaluation on Pulsifying for Efficiency of Microbe Recovery. Tokai University, Shizuoka, Japan.

Parsley, bean sprouts, spinach, raw prawn, clam and ham obtained from a local supermarket were tested. Samples were tested after 15, 30 and 60 seconds processing in the Pulsifier® and after 30 seconds in a Stomacher™ (Figure 3.). Both plain bags and filter bags were tested to assess bag splitting. In terms of total viable count (TVC) the Pulsifier™ recovered equivalent CFU after 15 seconds as the Stomacher™ after 30 seconds, and the counts did not increased after longer Pulsification times (Table 3.).

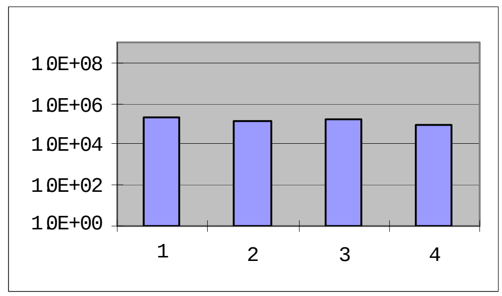
In terms of bags splitting the Pulsifier® did not cause breakages with plain bags after 30 seconds with the range of food samples tested, including hard samples such as Spinach stems and broken clam shells. The Stomacher™ produced some leakage from plain bags containing Spinach stems and clams after 30 seconds. In the case of filter bags no leakage were seen after 15 seconds, however, leakage were seen in some filter bags after 30 seconds of Pulsification(Table 4.).

It is clear from these studies that the Pulsifier® is an efficient instrument for the recovery of bacteria from a range of food samples. This is emphasised by the ability of the instrument to process the samples in 15 seconds compared to the advised 30 second processing time for the Stomacher™ (Table 3.). There is some evidence that the Pulsifier® will produce leakage in filter bags but apparently only after 30 seconds so a processing time of 15 seconds should deliver results as expected. In any case the cleaner samples produced by the Pulsifier should minimise the numbers of samples that need to be processed using filter bags.

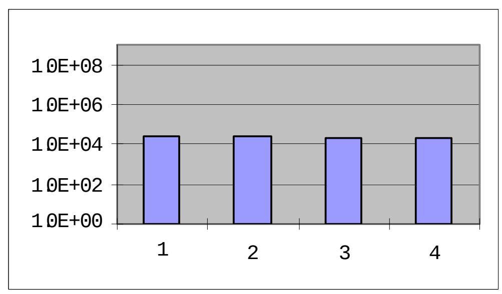
Figure 3. Total Viable Counts from a range of food types Pulsified for varying times or stomached for 30 seconds.



1. Spinach



2. Uncooked Prawn



3. Clam

LEGEND:1=Pulsifier 15 seconds, 2=Pulsifier 30 seconds, 3=Pulsifier 60 seconds, 4=Stomacher 30 seconds

Table 3. Average Total Viable Counts from a range of food types Pulsified for 15 seconds stomached for 30 seconds.

Sample	Pulsifier® (cfu/g)	Stomacher (cfu/g)
Bean Sprout	8.2×10^6	7.6×10^6
Parsley	3.1×10^6	6.3×10^6
Spinach	1.6×10^6	1.5×10^6
Raw Prawn	1.4×10^5	9.6×10^4
Clam	2.3×10^4	1.9×10^4

Table 4. Percentage of bag breakages by Pulsifier® and Stomacher™ .

Food	Bag	Pulsifier® 15s	Pulsifier® 30s	Stomacher 30s
Parsley	Filter	0%	20%	0%
Bean Sprouts	Filter	0%	60%	0%
Spinach	Plain	0%	0%	60%
Ham	Plain	0%	0%	0%
Raw Prawn	Plain	0%	0%	0%
Clam	Plain	0%	0%	60%

Table 5 : *Salmonella enteritidis*

Sample	Method	Log $_{10}$cfu/gm
Frozen	Pulsifier®	7.58
Peas	Stomacher™	7.17
Minced	Pulsifier®	9.48
Beef	Stomacher™	9.63
Potato	Pulsifier®	9.67
Powder	Stomacher™	9.68

Table 6 : *Listeria monocytogenes*

Sample	Method	Log $_{10}$cfu/gm
Frozen	Pulsifier®	3.54
Peas	Stomacher™	3.68
Minced	Pulsifier®	3.79
Beef	Stomacher™	3.76
Potato	Pulsifier®	3.55
Powder	Stomacher™	3.45

Table 7 : *E.coli* 0157

Sample	Method	Log $_{10}$cfu/gm
Frozen	Pulsifier®	3.54
Peas	Stomacher™	3.68
Minced	Pulsifier®	3.79
Beef	Stomacher™	3.76
Potato	Pulsifier®	3.55
Powder	Stomacher™	3.45

Comparison of the Pulsifier® and Stomacher for the recovery of pathogenic bacteria from food samples. A summary of a commissioned study by the Central Science Laboratory, Norwich, UK.

The ability of the Pulsifier®, in comparison to the Stomacher, to recover pathogenic bacteria from different food types was investigated using strains of ***Salmonella enteritidis***, ***Listeria monocytogenes*** and ***E. coli*** 0157. The food samples spiked for these studies were frozen peas, minced beef and potato powder purchased from a local supermarket.

Analysis of this data (Table 5,6 and 7) demonstrates no significant difference in the counts achieved by Pulsification or Stomaching samples. This data confirms the observations of other workers that the Pulsifier® produces recoveries of bacteria from a wide variety of food samples.

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FOR MORE INFORMATION ON THE PULSIFIER® OR ANY OF THE OTHER MICROGEN PRODUCTS FEATURED, PLEASE CONTACT YOUR LOCAL MICROGEN DISTRIBUTOR OR COMPLETE THE INFORMATION REQUEST FORM ENCLOSED WITH THE RELEVANT RESPONSE CODE AND RETURN IT TO MICROGEN BIOPRODUCTS

MICROGEN BIOPRODUCTS LTD
1 Admiralty Way
Camberley
Surrey
United Kingdom GU15 3DT
Phone: +44 (0)1276 600081
Fax: +44 (0)1276 600151
E-:mail sales@microgenbioproducts.com
Web: www.microgenbioproducts.com

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