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Non-Standardized Test Report For:

Survivor Filter, a division of Zakaib Holdings Limited

Model: Survivor Filter ™ Triple Absolute Filtration to 0.05 Microns, and
Survivor Filter ™ PRO 0.01 Micron Water Purifier
Project No. G102092758

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Objective

The primary objective of this evaluation is to provide test data showing the amount of bacteria, virus, and heavy metals (Lead, Mercury and Cadmium) removed from a water matrix by the Survivor Filter and Survivor Filter Pro.

Overview

A challenge suspension of known concentration will be prepared and introduced into the system. The suspension will be run through the filter and then analyzed for remaining concentrations.

Separate and new filters are to be used for the bacteria, virus, and heavy metals.

Detection will be as follows:

I. Bacteria
   a. Collection and enumeration technique derived from the Standard Total Coliform Membrane Filter Procedure in accordance with APHA's *Standard Methods for the Examination of Water and Wastewater.* This procedure is commonly used for NSF/ANSI certification standards for food equipment.

II. Virus
   a. Collection and enumeration technique derived from the Standard Total Coliform Membrane Filter Procedure in accordance with APHA's *Standard Methods for the Examination of Water and Wastewater.* This procedure is commonly used for NSF/ANSI certification standards for food equipment.

III. Heavy Metals
   a. The Elemental analysis of the Lead, Mercury and Cadmium will follow the IEC 62321 Elemental analysis procedure. This procedure is commonly used for RoHS analysis of various materials.

Procedural

I. Bacterial Analysis

a. Challenge Suspension Preparation
   1. *E. coli*, (ATCC 11229) and *S. aureus* (ATCC 6538) bacteria obtained from ATCC
   2. Stock cultures rehydrated with Tryptic Soy Broth (TSB) and incubated at 36 ± 1 °C (97 ± 1 °F) for approximately 24 hours
   3. The suspension will be diluted with phosphate buffer solution (PBS) to yield a concentration of 1 to $5x10^6$ cfu/mL of each organism

b. Test
   1. 100 mL of the challenge suspension is filtered through the Survivor unit
   2. Samples will be serial diluted and will be plated on both MacConkey's agar and Mannitol Salts agar, inverted, and incubated at 36 ± 1 °C (97 ± 1 °F) for 24 h
      i. Serial dilution plating will be performed in duplicate
   3. Plates are to be enumerated after the incubation period and results expressed as the number of CFU/mL
      i. Only plates within the range of 20-300 cfu will be reported
   4. Control Samples to be performed as follows:
      i. Negative control: PBS will be filtered through unit and enumerated

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ii. Positive control: Serial dilution of the bacterial suspension will be performed using PBS and aseptically processed using the membrane filter technique

5. Steps (b)1-3 are repeated for Survivor Filter Pro Model

II. Viral Analysis

a. Challenge Suspension Preparation
   1. Phi X174 (#124425) to be obtained from Carolina Biosciences
   2. Sample to be enumerated via serial dilution to confirm concentration
   3. A dilution of the stock suspension will be made so that the density of viral particles is approximately 1 to $5x10^6$ plaque forming units (PFU) per mL

b. Test Procedure
   1. 100 mL of the challenge suspension is filtered through the Survivor unit
   2. Samples will be serial diluted will be plated on nutrient agar utilizing the agar overlay technique, inverted, and incubated at 36 ± 1 °C (97 ± 1 °F) for 24 h
      i. Serial dilution plating will be performed in duplicate
   3. Plates are to be enumerated after the incubation period and results expressed as the number of PFU/mL
      i. Only plates within the range of 20-300 PFU will be reported
   4. Control Samples to be performed as follows:
      i. Negative control: PBS will be filtered through unit and enumerated
      ii. Positive control: Serial dilution of the viral suspension will be performed using PBS and aseptically processed using the membrane filter technique
   5. Steps (b)1-3 are repeated for Survivor Filter Pro Model

III. Chemical Analysis

a. Challenge Suspension Preparation
   1. Plasma Water is obtained from Fisher Scientific (W9-2)
   2. Pb/Hg/Cd standards at 50,000ppm shall be used.
   3. 20mL of plasma water shall be doped with 10mL of Each Standard totaling 10000ppm Pb/Hg/Cd in 50ml H20

b. Test Procedure
   1. 50 mL of the challenge suspension is filtered through the Survivor unit
   2. The filtered solution shall be analyzed via ICP-OES analysis as specified in IEC 62321
   3. Control Samples will be performed as follows:
      i. Negative Control: Sterile Plasma will be filtered through the unit prior to testing and collected. This filtered plasma water will serve as a blank and subtracted out of any testing results.
      ii. Positive Control: The elemental suspension created in V will be diluted and ran through the ICP-OES analysis to confirm concentration.

Parameters

A digital hygrometer will be used to ensure the temperature range stayed within that specified in the procedure above.

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Hypothesis

The hypothesis for the microbiological portion is that the percent reduction is measurable outside the tolerance of the method, which is 8%. This tolerance is based on the within laboratory precision of the standard plate count test that accounts for a within-analyst variation of 8% (AATCC 100-2012). The hypothesis for the chemical portion of this test is that the percent reduction is measurable outside the tolerance of the method, which is 40% (IEC 62321).

Sample Acquisition

Sample will be submitted by the client for evaluation.

Sample #	Description	Serial #	Model	Date	Condition
COL1505291606-001	Survivor Filter	X000M99LWN	Survivor Filter TM Triple Absolute Filtration to 0.05 Microns	4/29/2015	New
Sample #	**Description**	**Serial #**	**Model**	**Date**	**Condition**
COL1505291606-002	Survivor Filter Pro	X000P3MQJV	Survivor Filter TM PRO 0.01 Micron Water Purifier	4/29/2015	New

Equipment List

Item	Equipment Type	Intertek Asset No.	Calibration Due
1	Micropipette	CE 1141	03/11/16
2	Thermofisher Heracell Incubator	CE 2381	For Reference Only
3	Balance	CE 1182	11/3/15
4	Autoclave	CE 2376	Verify before use
5	Graduated Cylinder	CE 2264	Initial Calibration Only
6	Digital Hygrometer	E226	6/12/15
7	Centrifuge	CE 2382	For Reference Only
8	Filtration System	CE 2031	Initial Calibration Only
9	Refrigerator	CE 1157	05/05/16
10	10-100 µL Pipette	CE2315	12/01/15
11	100-1000 µL Pipette	CE2234	03/30/16
12	1-5 ml Pipette	CE2228	03/30/16
13	Analytical Balance	CE2235	09/16/15
14	ICP	CE2100	Verify Before Use
15	Mercury Standard 50,000ppm	S140619010	03/01/16
16	Lead standard 50,000 ppm	1060190	11/03/15
17	Cadmium standard 50,000 ppm	S150407007	04/01/17
18	Custom Standard 600-653-108	S140709010	07/01/15

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Testing to be conducted at: Intertek Testing Laboratory
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Raw Microbiological Test Data

Test Parameter		Result	Units
Organism	Bacterial Species	Escherichia coli	--
	ATCC No.	11229	--
	Challenge Concentration	4.45×10^6	CFU/mL
Survivor Filter ™ Triple Absolute Filtration to 0.05 Microns	Serial Dilution Replicate 1	<1	CFU/mL
	Serial Dilution Replicate 2	<1	CFU/mL
	Average	**<1**	**CFU/mL**
Survivor Filter ™ PRO 0.01 Micron Water Purifier	Serial Dilution Replicate 1	<1	CFU/mL
	Serial Dilution Replicate 2	<1	CFU/mL
	Average	**<1**	**CFU/mL**

Test Parameter		Result	Units
Organism	Bacterial Species	Staphylococcus aureus	--
	ATCC No.	ATCC 6538	--
	Challenge Concentration	4.96×10^6	CFU/mL
Survivor Filter ™ Triple Absolute Filtration to 0.05 Microns	Serial Dilution Replicate 1	<1	CFU/mL
	Serial Dilution Replicate 2	<1	CFU/mL
	Average	**<1**	**CFU/mL**
Survivor Filter ™ PRO 0.01 Micron Water Purifier	Serial Dilution Replicate 1	<1	CFU/mL
	Serial Dilution Replicate 2	<1	CFU/mL
	Average	**<1**	**CFU/mL**

Test Parameter		Result	Units
Organism	Viral Species	Phi X174	--
	ATCC No.	13706-B1	--
	Challenge Concentration	5.0×10^6	PFU/mL
Survivor Filter ™ Triple Absolute Filtration to 0.05 Microns	Serial Dilution Replicate 1	740	PFU/mL
	Serial Dilution Replicate 2	810	PFU/mL
	Average	**775**	**PFU/mL**
Survivor Filter ™ PRO 0.01 Micron Water Purifier	Serial Dilution Replicate 1	<1	PFU/mL
	Serial Dilution Replicate 2	<1	PFU/mL
	Average	**<1**	**PFU/mL**

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Raw Chemical Test Data

Test Parameter		Result			Units
Chemical Standards	Chemical Contaminant	Mercury	Cadmium	Lead	--
	Challenge Concentration	8111	8358	8846	PPM
Survivor Filter ™ Triple Absolute Filtration to 0.05 Microns	Analysis Replicate 1	882	1054	1040	PPM
	Analysis Replicate 2	841	1006	993	PPM
	Average	**862**	**1030**	**1017**	PPM
Survivor Filter ™ PRO 0.01 Micron Water Purifier	Analysis Replicate 1	40	1080	596	PPM
	Analysis Replicate 2	39	1056	584	PPM
	Average	**40**	**1068**	**590**	PPM

Percent Reduction Calculations

Percent reduction calculated as follows-

$$\text{Percent Reduction} = \frac{(A-B) \times 100}{A}$$

Where:
A is the number organism/chemical concentration before running through the filter.
B is the number organism/chemical concentration after running through the filter.

Percent Reduction Values -Microbiological

	Bacterial Challenge		Viral Challenge
Unit Type	**E.coli**	**S.aureus**	**Phi-X174**
Survivor Filter ™ Triple Absolute Filtration to 0.05 Microns	**>99.9%**	**>99.9%**	**99.9%**

	Bacterial Challenge		Viral Challenge
Unit Type	**E.coli**	**S.aureus**	**Phi-X174**
Survivor Filter ™ PRO 0.01 Micron Water Purifier	**>99.9%**	**>99.9%**	**>99.9%**

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Percent Reduction Values -Chemical

Chemical Challenge			
Unit Type	**Mercury**	**Cadmium**	**Lead**
Survivor Filter ™ PRO 0.01 Micron Water Purifier	99.5%	87%	93%

Chemical Challenge			
Unit Type	**Mercury**	**Cadmium**	**Lead**
Survivor Filter ™ Triple Absolute Filtration to 0.05 Microns	89%	88%	89%

Conclusion

The microbiological hypothesis has been accepted since the plate count values were well outside the 8% measurement of uncertainty of the microbiological test method.

The chemical hypothesis has been accepted since the percent reduction is well outside the 40% uncertainty of the chemical test method.