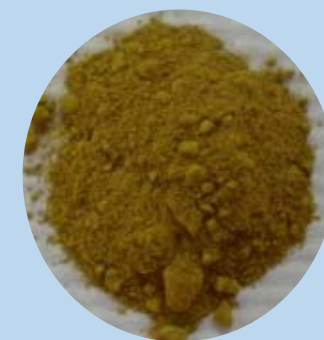




Organic Antimicrobial products from Sea shells and Mangosteen peels extract



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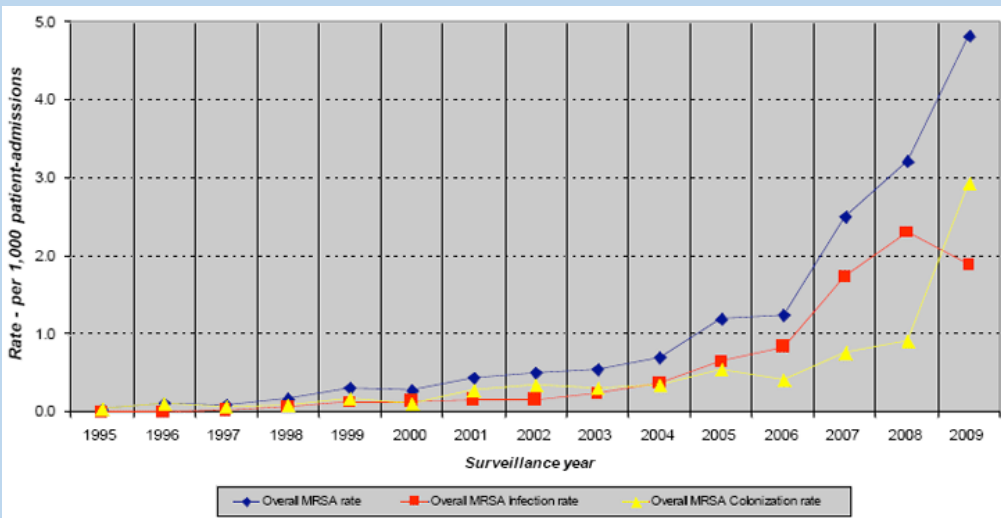
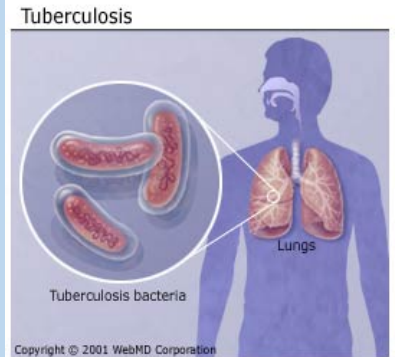


Airborne Microbes; *M. tuberculosis* (MTB) cause of tuberculosis (TB)

- 1 / 3 of world population have infected TB (WHO)
- Dead 1.6 million / year
- Increasing of Multidrug resistant TB (MDR-TB), XDR-TB cases
- Inhale only 2 MTB can cause disease

Harmful Microbes

Directed contact; Methicillin resistant *S. aureus* (MRSA)



Dead 700,000 (2014)

10 Million (2050)

www.wired.com/2014

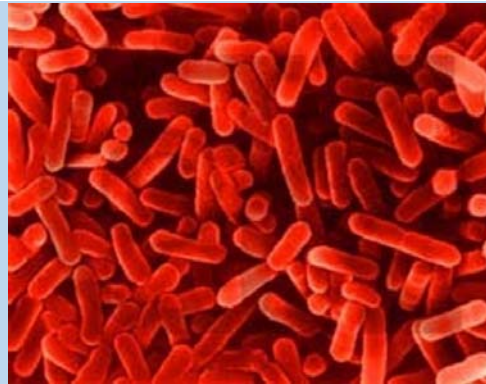


Chemical antimicrobial is available .but....

- Irritation to eye, skin.
- Can not use for body in everyday life.



- Non irritant organic antimicrobial is needed.



Garcinia Mangostana Peel (GMP)

GMP Antimicrobial products



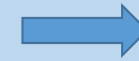
Garcinia Mangostana Peel (GMP)

GMP was use as Thai traditional medicine for long time.

Rich of xanthone derivatives



GMP powder



Researches reported that GMP extract has antimicrobial effects against following pathogens;

1. *Mycobacterium tuberculosis (MTB)* (Suksumran et al, 2006; Tipkrua et al, 2012)
2. Multi-drug resistant tuberculosis (MDR-TB) (Sudta et al, 2013)
3. Methicillin resistant *S. aureus* (MRSA) (Voravuthi Kunchai et al, 2005; Koh et al 2013)
4. Human Immuno-deficiency virus (HIV-1) (Chen et al, 1996)
5. Hepatitis C virus (Choi et al, 2014)
6. Vancomycin resistant Enterococcus (Sakagami et al, 2005)
7. Bacteria in mouth & dental carried (Somprasit et al, 2015)
8. Bacteria in chronic wound (Temrangsee et al, 2011)
9. Bacteria that cause atopic dermatitis (Higuchi et al, 2013)
10. Acne caused bacteria (Pothitirat et al, 2009; Chomnaunny et al, 2005)
11. Candida (Kaomongkolgit et al, 2009)

GMP coated pre-filter

The pre-filer was coated with GMP. and tested for;

- Mycobactericidal activity against MTB,
- MTB viability at difference exposure time.



Jeab Natthakan

MTB inhibition testing of GMP coat pre-filter

Uncoated pre-filter



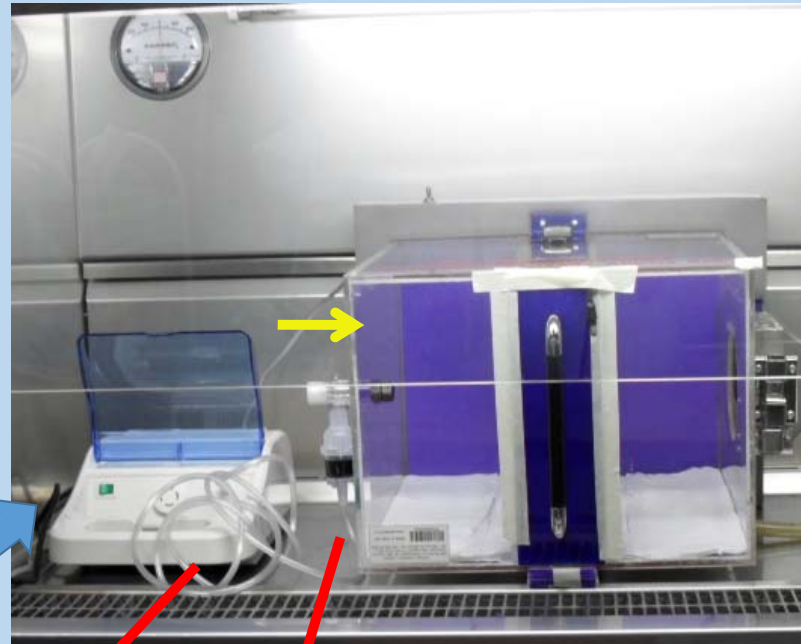
GMP coated pre-filter



Filter holder

Nebulization
for 30 min.

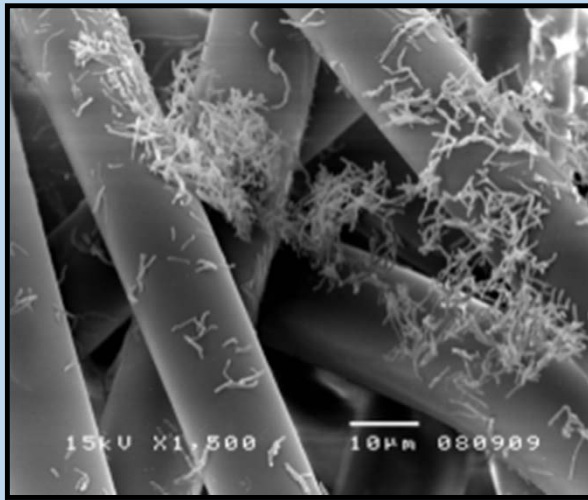
5ml of 10^5 cells / ml
(*M. smegmatis*)



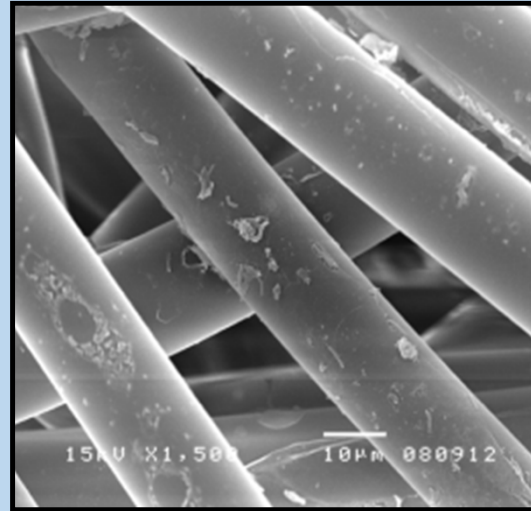
Determine
bacterial number
by Electron
Microscope

MTB viability on pre-filter After sprayed

Uncoated
pre-filter



GMP coated
pre-filter



GMP can inhibit growth of MTB on pre-filter

Type of pre-filter	Average colonies count per 5 cm ² (n = 24)	SD
Un-coated	210	36.3
GMP-coated	4	2.2

the average colony count between coated and uncoated pre-filter was significantly difference (p-value < 0.0001)

Core or recent technology for solving problems

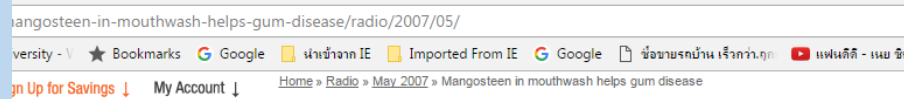
- Organic waste
- Low cost (0.5 - 70 Thai B / liter)



GMP extract



- Alternative Anti-TB drug; Drug resistant MTB
- Antimicrobial Spray
- Mouthwash



Mangosteen in mouthwash helps gum disease

May 10, 2007

You may want to add a Mangosteen Extract to your mouth rinse. In this study researchers gave 60 patients either a mouthwash that included Mangosteen or a look alike placebo mouthwash. At two weeks the patients were examined and polished. Vs. placebo, the Mangosteen decreased odor released in the mouth, decreased plaque and gingivitis; all significantly decreased. The study is published in the January 2007 issue of the Journal of Periodontology.



GMP coated pre-filter

- Antimicrobial filter
- Air condition
- Air filtration
- Anti TB Net

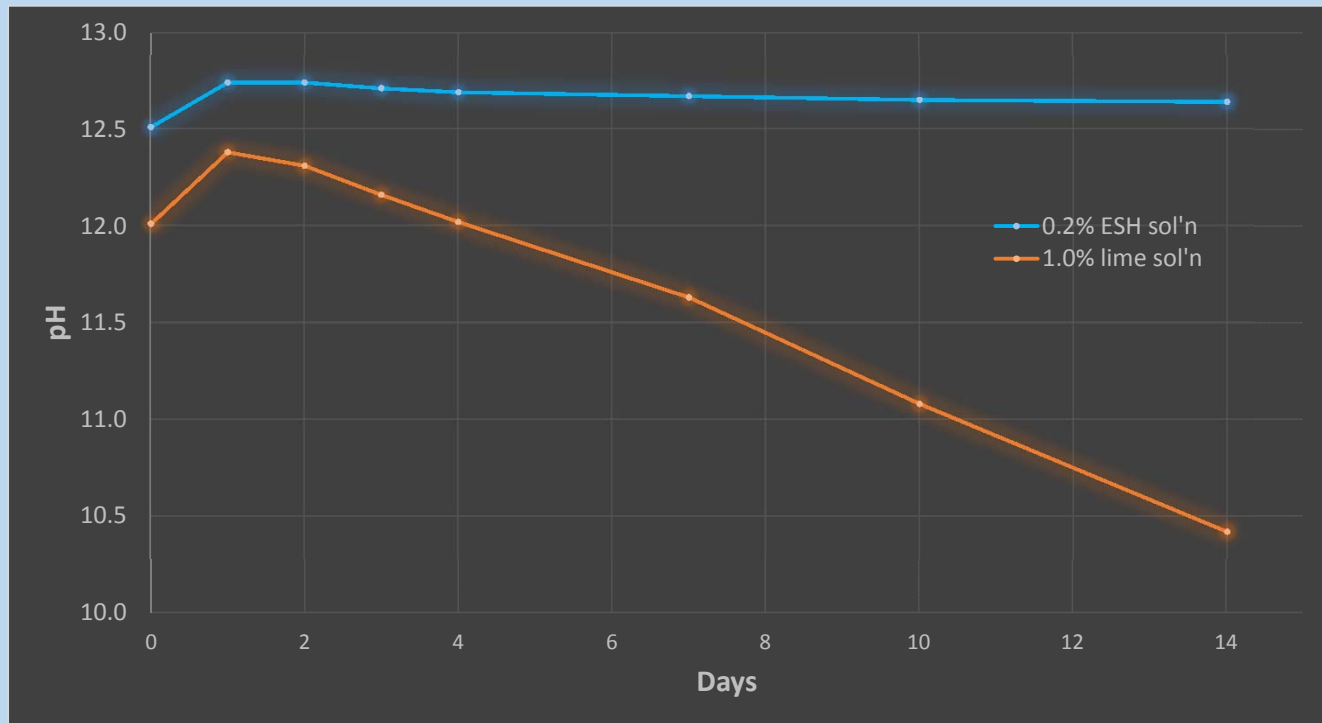
EcoShellHybrid (ESH) is Calcium dihydroxide



Biomedical Research Institute (BMRI),
Chiba, Japan

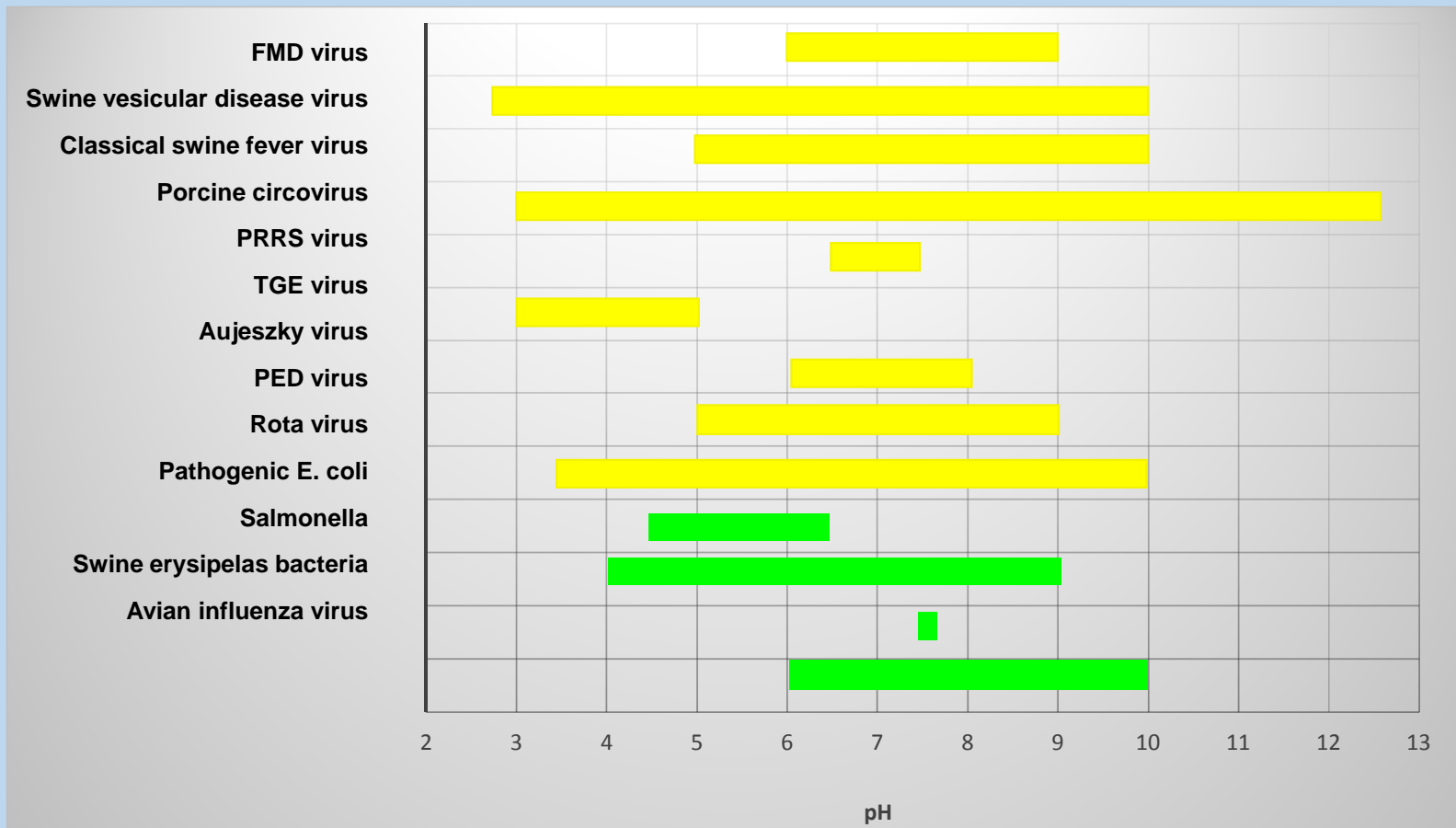
- Also other synonym is Hydrated lime; Slaked Lime; Calcium Oxide, hydrated.
- Purity of lime usually about 65% but purity of ESH is more than 97%.
- ESH is permitted as food additives in Japan (very safe to human).
- ESH is very low cost (made from waste)
- Dissolved ESH with water can make solution with PH 12-13.

pH change in ESH and normal lime Solutions (BMRI)



- 0.2% ESH solution can keep pH more than 12.0 over 2wks. pH of the solution doesn't drop down much even if 16wks later.
- On the other hand, pH of 1.0% normal lime solution drops to less than 12.0 after 4 days.

pH Surviving range of some organisms (BMRI)



Inactivation of virus with ESH solution (BMRI)

+0.2% ESH solution	PFU/ml of virus Log10	Inactivation time needed
None Envelope virus		
Feline Calicivirus	6.0	<10 pfu/ml 60sec.
Murine Norovirus	5.2	<10 pfu/ml 60sec
Poliovirus	6.6	<10 pfu/ml 60sec
Envelope virus		
Influenza virus	6.2	<10 pfu/ml 10min
Porcine Epidemic Diarrhea	5.6	<10 pfu/ml 30min

Antibacterial effect of ESH solution (BMRI)

+0.2% ESH solution	No. of bacteria/ml (Log ₁₀)	Inactivation time needed		
Gram negative				
<i>Campylobacter jejuni</i>	6.70	<10	30sec	
<i>Escherichia coli</i>	6.28	<10 ²	30min	<10 60min
Pathogenic <i>E. coli</i> O-157	5.80	<10 ²	30min	<10 60min
<i>Salmonella enterica</i>	6.40	<10 ⁴	30min	<10 60min
<i>Vibrio parahaemolyticus</i>	6.23	<10 ²	30min	<10 60min
<i>Legionella pneumophila</i>	6.60	<10 ³	30min	<100 60min <10 120min
Gram positive				
<i>Staphylococcus aureus</i>	7.17	<10 ⁵	2h	<100 4h
MRSA	6.94	<10 ³	2h	<100 4h

To pass the Thai FDA as antimicrobial solution; Contact bacteria inhibition test of ESH is needed

1. *Staphylococcus aureas*
2. *Pseudomonas aeruginosa*
3. *Salmonella Spp.*

1.5×10^8 CFU/ ml of each bacteria was used

ESH

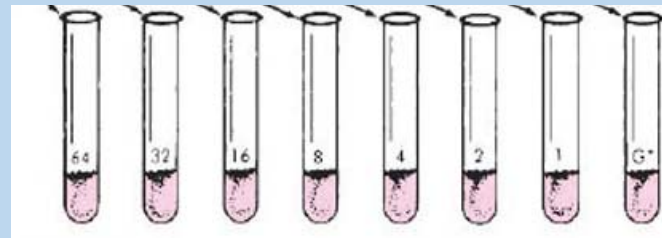
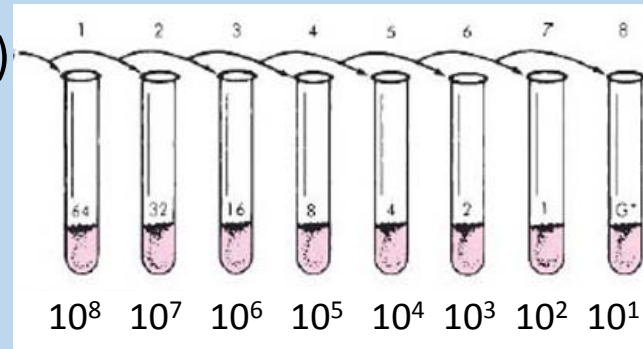
0.2 % ESH (0.2 gram ESH + 1000 ml Sterile mineral water)



Broth Dilution Method

1. 1.5×10^8 CFU/ ml (0.5 Mcfarland)
2. 1.5×10^7 CFU/ ml
3. 1.5×10^6 CFU/ ml
4. 1.5×10^5 CFU/ ml
5. 1.5×10^4 CFU/ ml
6. 1.5×10^3 CFU/ ml
7. 1.5×10^2 CFU/ ml
8. 1.5×10^1 CFU/ ml

1 ml



0.2 % ESH 1 ml

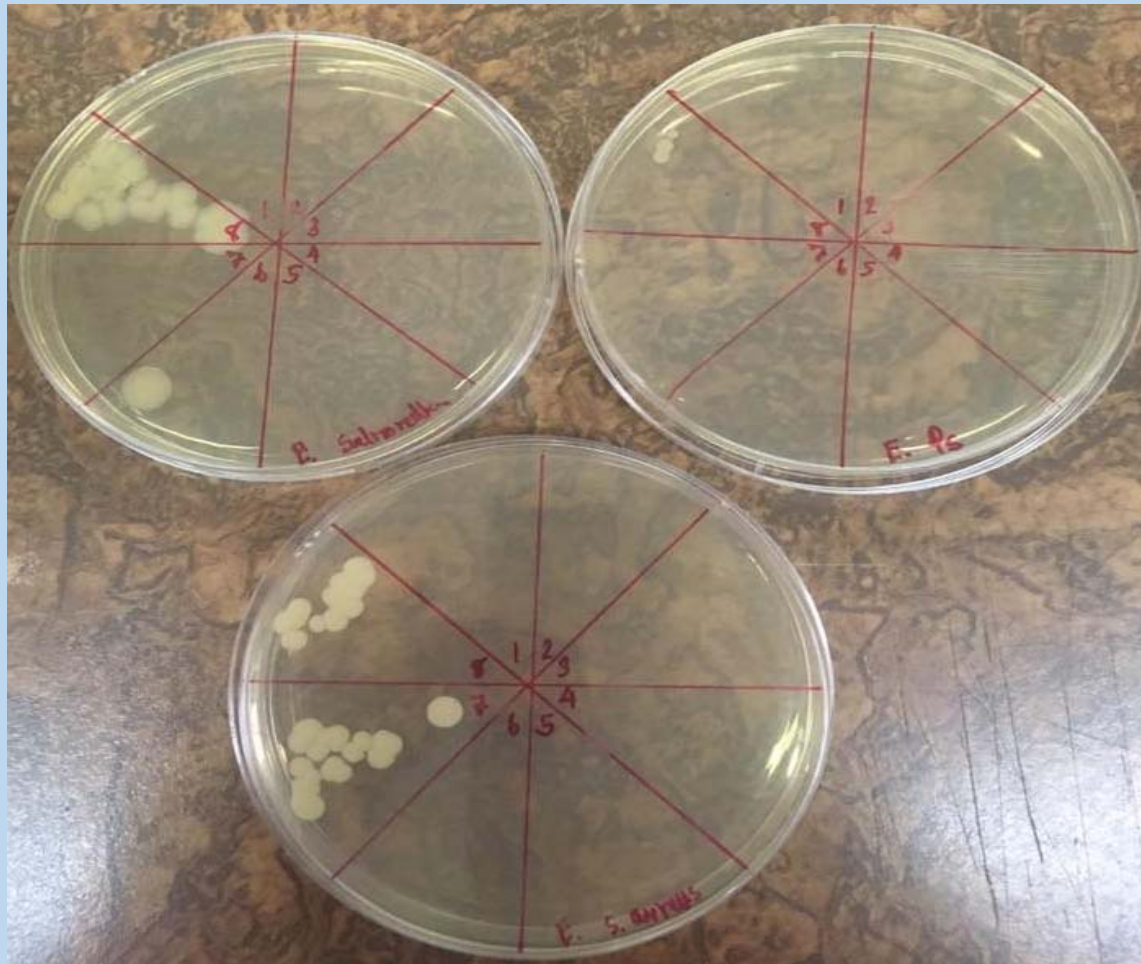
Triplicate experiment were performed



Incubated at 35 C 24 hr

0.2 % ESH Inhibition of 3 bacteria

Salmonella Spp.
from
 $10 \cdot 10^7$ CFU/ ml



Pseudomonas
aeruginosa
from
 $10 \cdot 10^7$ CFU/ ml

Staphylococcus aureus
From $10 \cdot 10^6$ CFU/ ml

To pass the Thai FDA as antimicrobial spray; Aerosol Bacterial inhibition test of ESH is needed

1. *Staphylococcus aureus*
2. *Pseudomonas aeruginosa*
3. *Klebsiella pneumoniae*

1.5×10^8 CFU/ ml of each bacteria was used

- **ESH**

0.2 % ESH (0.2 gram ESH + 1000 ml Sterile mineral water)



Aerosol chamber for
ESH spray tested

1 cm thick acrylic

Length
1.04 M

Width 0.79M



High
0.63M

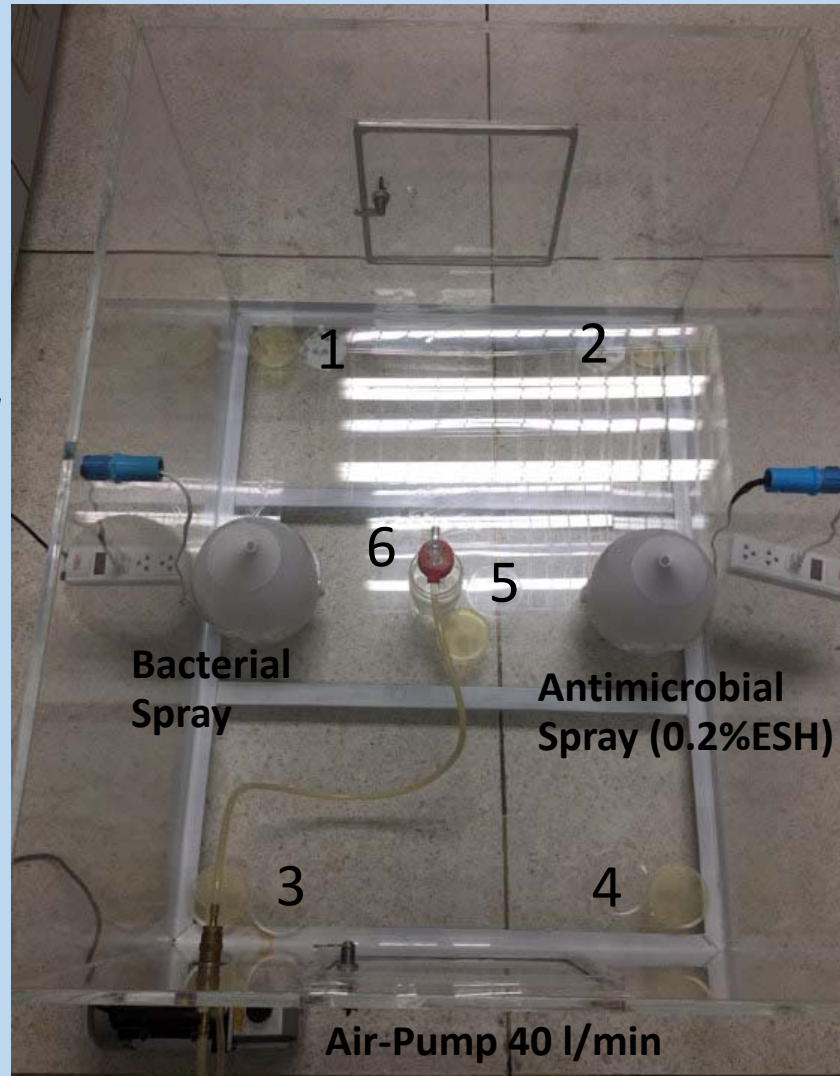
$$\begin{aligned} \text{Area} &= W \times L \times H \\ &= 0.79 \times 1.04 \times 0.63 \\ &= 0.52 \text{ Cubic M} \end{aligned}$$

Aerosol chamber for ESH spray tested

Bacterial spray

1.5×10^5 CFU/ 150 ml
of each bacteria

1. *Staphylococcus aureus*
2. *Pseudomonas aeruginosa*
3. *Klebsiella pneumonia*



ESH spray 150 ml

- 0.2 % ESH (0.2 gram ESH + 1000 ml Sterile mineral water)



- Triplicate experiment
- Cleaned chamber & equipment with 70% alcohol & Clorox

Aerosol chamber for ESH spray tested

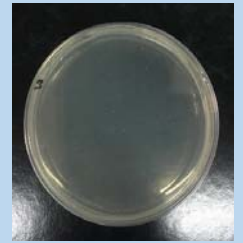
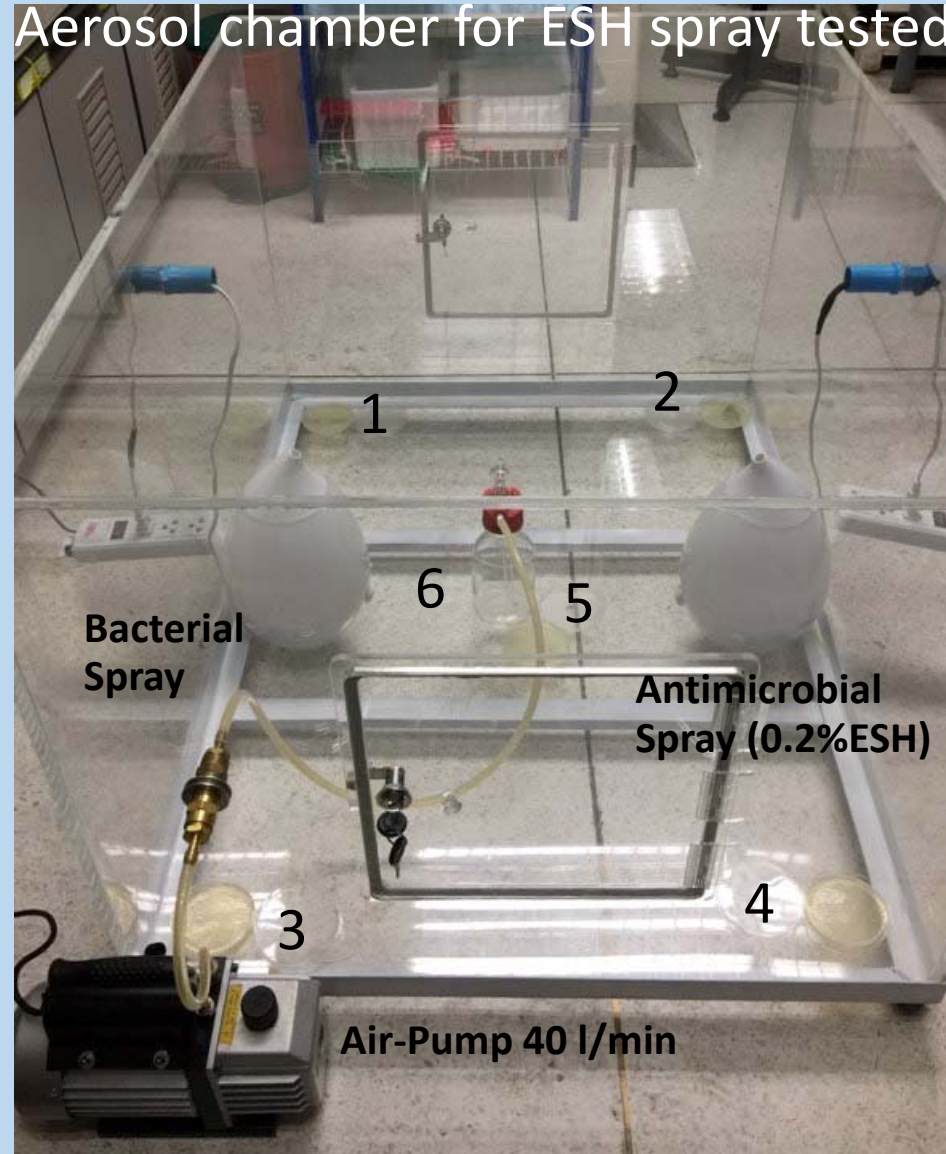
Bacterial spray

1.5×10^5 CFU/ 150 ml
of each bacteria

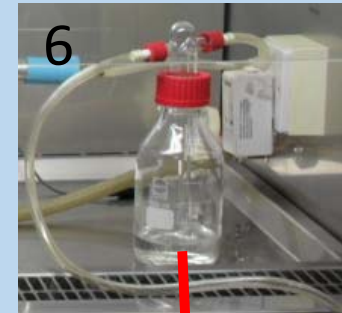
1. *Staphylococcus aureus*
2. *Pseudomonas aeruginosa*
3. *Klebsiella pneumoniae*



0.2 % ESH spray
150 ml



1, 2, 3, 4, 5



**Determine
bacterial number
by culture**

- Triplicate experiment
- Cleaned chamber & equipment with 70% alcohol & Clorox

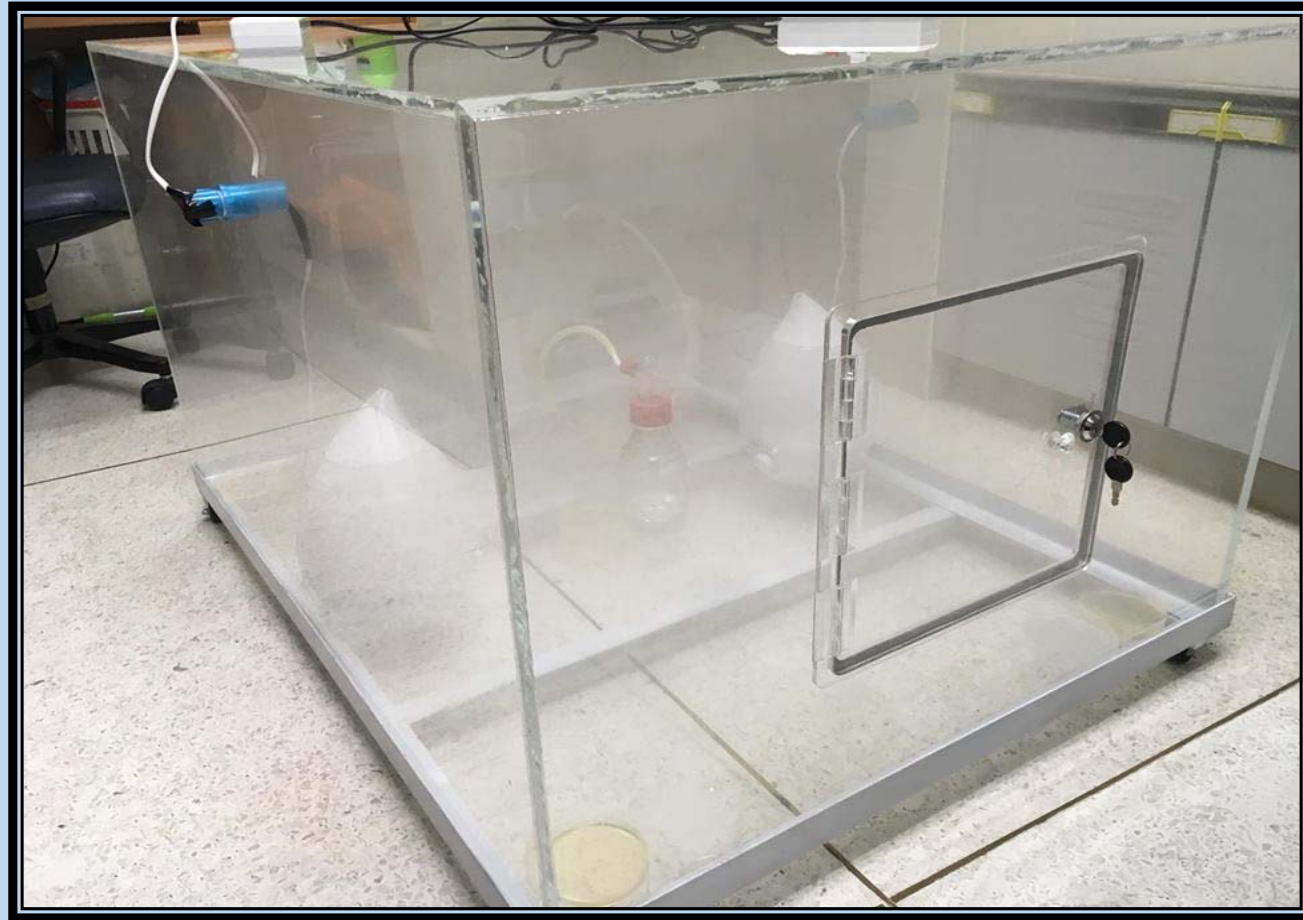


Bacterial spray

1.5×10^5 CFU/ 150 ml
of each bacteria

1. *Staphylococcus aureus*
2. *Pseudomonas aeruginosa*
3. *Klebsiella pneumonia*

- Triplicate experiment
- Cleaned chamber & equipments with 70% alcohol & Clorox

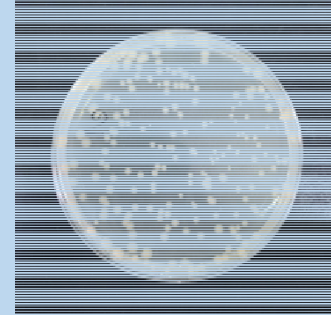


1



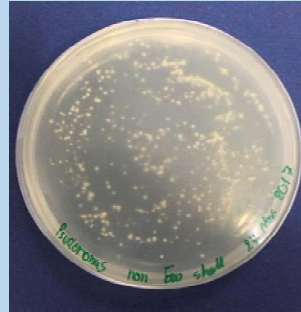
187 CFU.

2



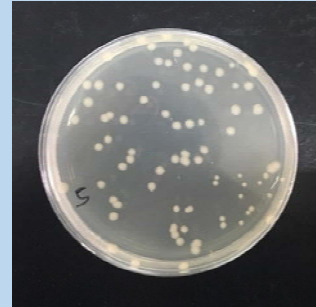
212 CFU.

6



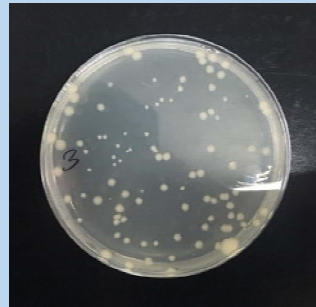
313 CFU.

5



100 CFU.

3



86 CFU.

4



69 CFU.

Bacterial spray

1.5×10^5 CFU/ 150 ml
of each bacteria

1. *S. aureus*
2. *P. aeruginosa*
3. *K. pneumonia*



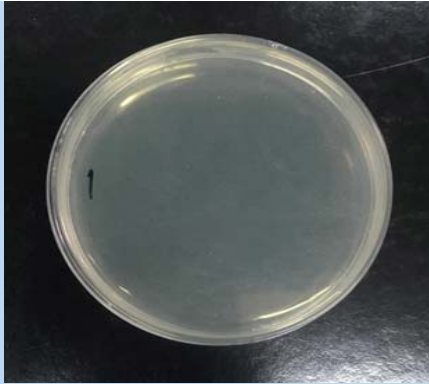
Antimicrobial spray

0.2 % ESH
150 ml

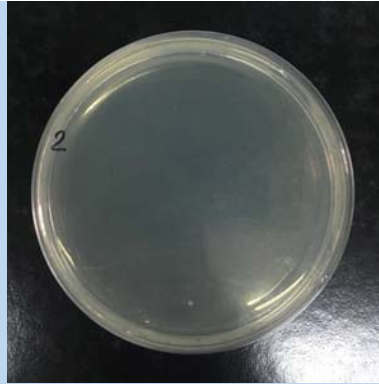


- Triplicate experiment
- Cleaned chamber & equipment with 70% alcohol & Clorox

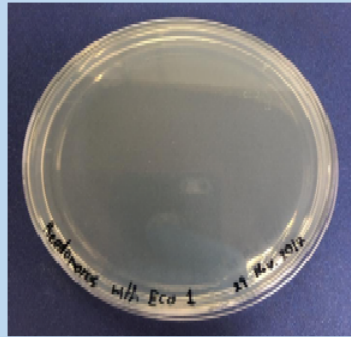
1



2



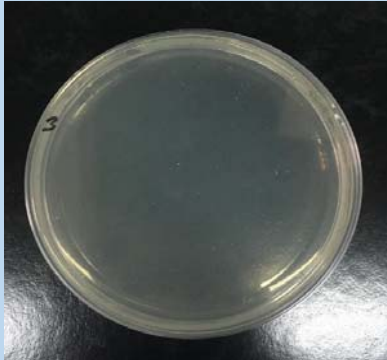
6



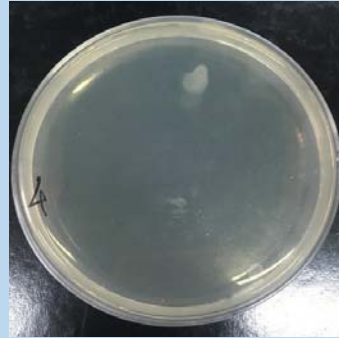
5



3



4





1st model with pressure gas



2nd model without gas (full 300 cc ESH)



www.babyconnectionshop.com/a_36816_18829_Store-Location.htm

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Available in the market as "Air spray"



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Site Home | Store Location

Store Location

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16 branches

- โรงพยาบาลพญาไท 3
- โรงพยาบาลป่าจุงราษฎร์
- โรงพยาบาลสมิติเวช สุขุมวิท
- โรงพยาบาลเวชธานี
- โรงพยาบาลนครธน
- โรงพยาบาลกรุงเทพ พัทยา
- โรงพยาบาลกรุงเทพ ราชสีมา
- โรงพยาบาลเอกอุดร
- โรงพยาบาลเสรีรักษ์
- โรงพยาบาลสินแพทย์
- โรงพยาบาลขอนแก่นราม
- โรงพยาบาลวิภาวดี
- โรงพยาบาลกรุงเทพหาดใหญ่
- โรงพยาบาลกรุงเทพพระยอง
- โรงพยาบาลพญาไท 2
- โรงพยาบาลสมิติเวชชลบุรี



Approved
by
Thai FDA



8,000 branches



Thank you

