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APPENDICES

APPENDIX A

Reagents and Media

APPENDIX A

Reagents and Media

Lactic acid bacteria (LAB) media recipes.

1. MRS medium

1. Dissolve the following in 850 ml of distilled water.
    - 10 g Gelatin
    - 8 g Beef extract
    - 4 g Yeast extract
    - 18.5 g Glucose
    - 1 g Tween-80
    - 2 g K_2HPO_4
    - 3 g Na-acetate
    - 2 g $(NH_4)_2$ citrate
    - 0.2 g $MgSO_4.7H_2O$
    - 0.05 g $MnSO_4.H_2O$
    - 0.04 g Bromocresol purple
2. Adjust pH to between 6.2 and 6.5.
3. Bring to 1000 ml with distilled water.
4. Autoclave at 121°C for 15 min or filter sterilize.

2. GYP agar plate

- 10 g Glucose
- 10 g Yeast extract
- 10 g Peptone
- 5 ml Salt solution (see below)
- 0.04 g Bromocresol purple
- 0.2 g NaN_3
- 1000 ml Distilled water

Stock salt solution :

- 40 g $MgSO_4.7\ H_2O$
- 2 g $MnSO_4.4\ H_2O$
- 2 g $FeSO_4.7\ H_2O$
- 1000 ml Distilled water

1. Dissolve the following in distilled water.
2. Add 15 g/l agar before autoclaving.
3. Autoclave at 121°C for 20 min.
4. Let cool to ~ 55 °C and add desired antibiotics at this point. Pour into 10 cm Petri plates. Let the plates harden, then invert, and store at +4 °C.

3. Skim milk solution

   - 150 g Skim milk powder
   - 5 g Yeast extract
   - 10 g Glucose
   - 1000 ml Distilled water

   1. Dissolve the following in distilled water.
   2. Autoclave at 110°C for 10 min (twice).

Reagents recipes

1. Stock Sodium Chloride (NaCl) 0.85%
    - 0.85 g NaCl
    - 100 ml Distilled water

    1. Dissolve the following in distilled water.
    2. Autoclave at 121°C for 15 min.

2. Glycerol stock solution
    - 650 ml Glycerol
    - 12.048 g MgSO4.7H2O
    - 3.0285 g Tris-HCl
    - 1000 ml Distilled water

    Tris-HCl :

    1. To make 1 mol/L Tris-HCl dissolve 121.1 g of tris base in 700 ml of double distilled water.
    2. Bring to desired pH with concentrated HCl_{aq} (usually 7.5 or 8.0).
    3. Add double distilled water to 1 L.
    4. Filter with 0.5 µm filter.
    5. Autoclave.
    6. Store at room temperature.

APPENDIX B

Equipments

APPENDIX B

Spray dryer

Specification

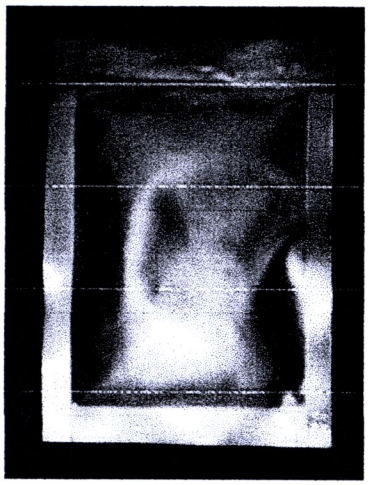
- Spray dryer manufactured by J.C. Machinery Engineering, Thailand.
- A nozzle atomizer with a length of 44 cm.
- A co-current air flow.
- A blower speed at 38 Hz.

Infrared Moisture Determination Balance FD-620

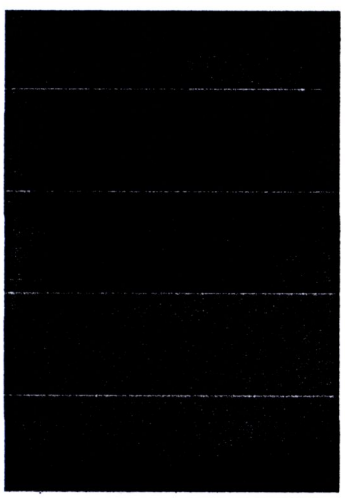
Specification

- Kett Electric Laboratory, Japan.

Aluminum bag for storage.



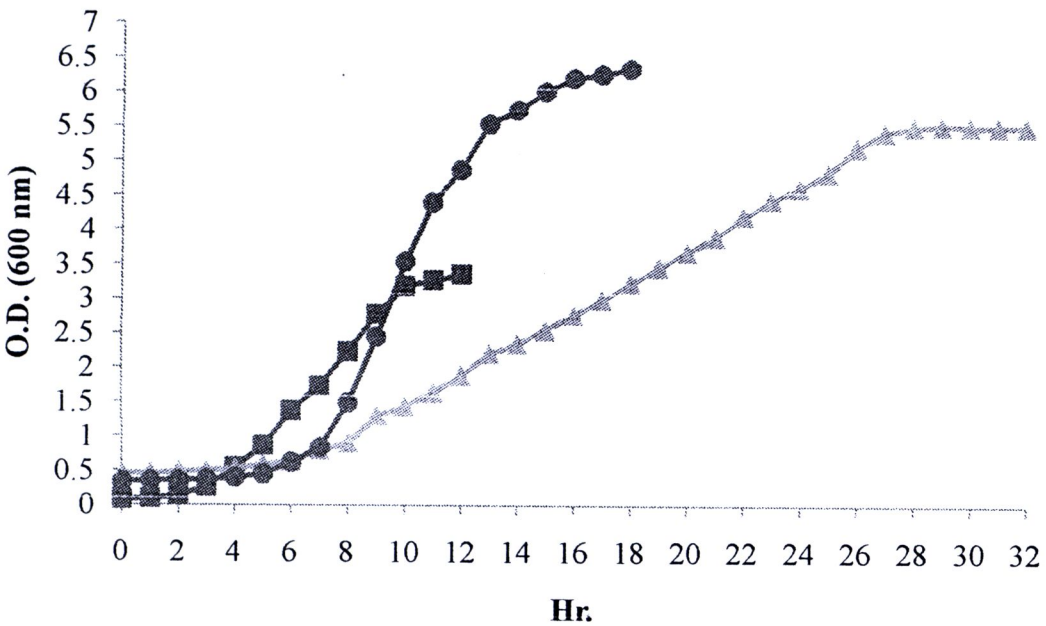
Plastic zip bag for storage.



APPENDIX D

Growth curve of probiotic microoraganisms

Growth curve of probiotic microoraganisms



Growth curve of *L. casei* subsp. *paracasei* F-19 (▲)

L. fermentum 2311M (■)

L. plantarum V-299 (●)

CURRICULUM VITAE

Name	Ms. Kanokwan Sarinyawat
Date of birth	22 August 1983
Academia background	
Bachelor Degree	Bachelor of Science (Biology), Chiangmai University, 2005