

APPENDIX A

List of the chemicals and materials used in the study

| Chemicals / Materials | Sources |
|----------------------------------|--------------------------------------|
| Absolute ethanol | E. MERCK, Germany |
| Azoxymethane | Sigma Chemical Co., U.S.A. |
| Acetic acid, glacial 100% | E. MERCK, Germany |
| Acrylamide PAGE | Pharmacia Biotech, Sweden |
| Albumin Bovine | Sigma Chemical Co., U.S.A. |
| Ammonium persulphate | Pharmacia Biotech, Sweden |
| Bromophenol blue | USB Amersham life Science, U.S.A. |
| 1-chloro-2,4-dinitrobenzene | Wako Pure Chemical Industries, Japan |
| Coomassie Brilliant Blue G250 | Fluka, Japan |
| Dimethylsulfoxide | E. MERCK, Germany |
| Glutathione | Sigma Chemical Co., U.S.A. |
| Glycerol | USB Amersham Life Science, U.S.A. |
| Glycine | E. MERCK, Germany |
| Glyoxalase I (EC 4.4.1.5) | Sigma Chemical Co., U.S.A. |
| Goat Antirabbit IgG Horseradish | Chemicon International INC., U.S.A. |
| GST Ya, Yb, Yc, YT antisera | Biotrin International Ltd., Ireland |
| Hybond C Nitrocellulose membrane | Pharmacia Biotech, Sweden |
| Hydrochloric acid | E. MERCK, Germany |
| α -ketoglutarate | Fluka |
| L-alanine | Fluka |
| L-aspartate | Fluka |
| Mercaptoethanol | Pharmacia Biotech, Sweden |
| Methanol | E. MERCK, Germany |

| | |
|-----------------------------------|-------------------------------|
| Methylenebis-acrylamind | Pharmacia Biotech, Sweden |
| Methylene blue | Sigma Chemical Co., U.S.A. |
| Methylglyoxal | Sigma Chemical Co., U.S.A. |
| MOPS | Sigma Chemical Co., U.S.A. |
| Perchloric acid | E. MERCK, Germany |
| Potassium chloride | E. MERCK, Germany |
| Potassium hydrogen phosphate | E. MERCK, Germany |
| Potassium hydroxide | E. MERCK, Germany |
| Rainbow Marker Full range | Amersham Life Science, U.S.A. |
| Sodium chloride | Sigma Chemical Co., U.S.A. |
| Sodium Dodecyl Sulfate | Sigma Chemical Co., U.S.A. |
| Sodium hydrogen phosphate | E. MERCK, Germany |
| TEMED | Pharmacia Biotech, Sweden |
| Tris | Amersham Life Science, U.S.A. |
| Tris (hydroxymethyl) aminomethane | Sigma Chemical Co., U.S.A. |
| Tween 20 | Sigma Chemical Co., U.S.A. |

APPENDIX B

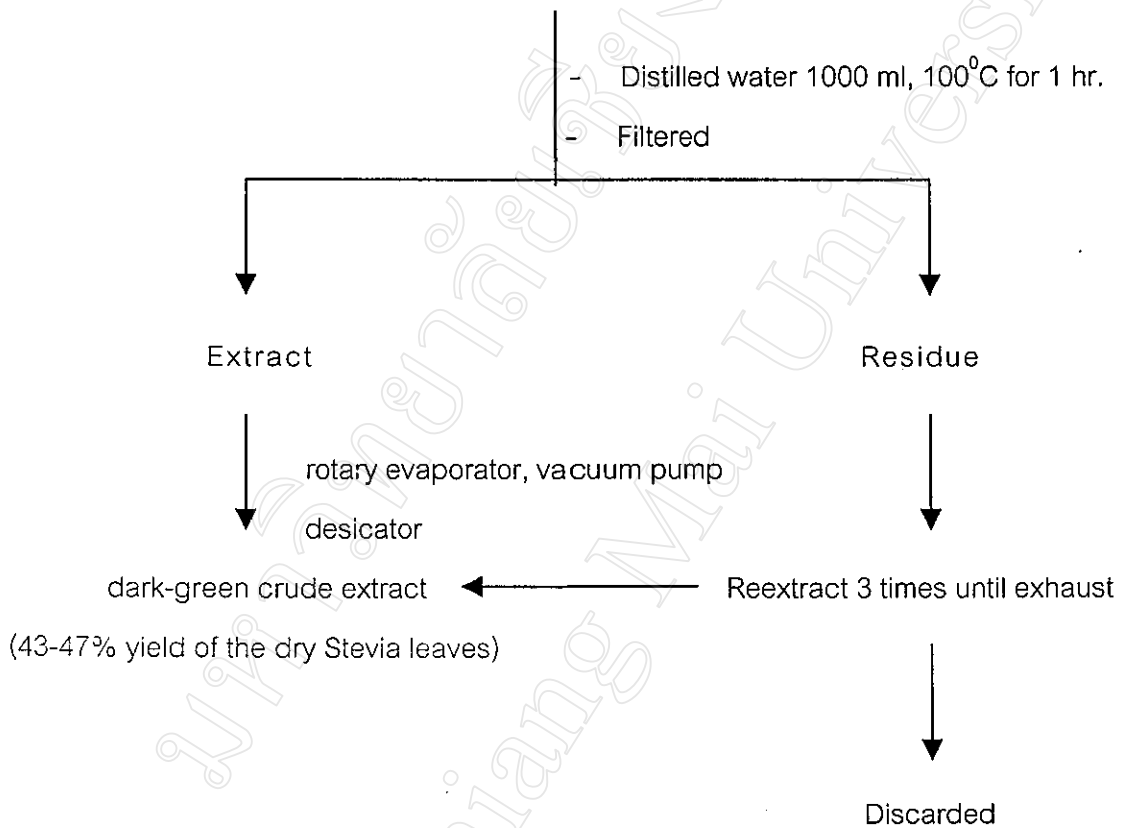
List of instruments used in the study

| Instruments | Model | Sources |
|---------------------------|----------------------|--------------------------------|
| Blotter | mini VE | Pharmacia Biotech Inc., U.S.A. |
| Densitometer | GS-700 | Bio-Rad, U.S.A. |
| Electrophoresis Apparatus | Hofer mini VE | Pharmacia Biotech Inc., U.S.A. |
| Light microscope | | Olympus, Japan |
| pH meter | | Eutech Cybematics, Singapore |
| Polytron homogenizer | GH-6010 | Kinematica, Switzerland |
| Power supply | EC570 | Apparatus Corporation, U.S.A. |
| Refrigerator (-20°C) | | Sanyo, Thailand |
| Refrigerator (-80°C) | | Forma Scientific |
| Slab Gel dryer | SGD4050 | SAVANT, U.S.A. |
| Spectrophotometer | Milton Roy 1001 plus | U.S.A. |
| Super speed centrifuge | Kokusan | Japan |
| Ultracentrifuge | Centrikon T-1180 | Kontron instrument, Italy |
| Vacuum concentrator | Speed Vac SC110 | SAVANT, U.S.A. |

APPENDIX C

Protocol for extraction of *Stevia rebaudiana*, Bertoni

100 g of dry Stevia leaves (from Aroonsri Plantation, Mae Rim, Chiang Mai)



Protocol for the preparation of Stevia extract used as sweetener in Matoom juice

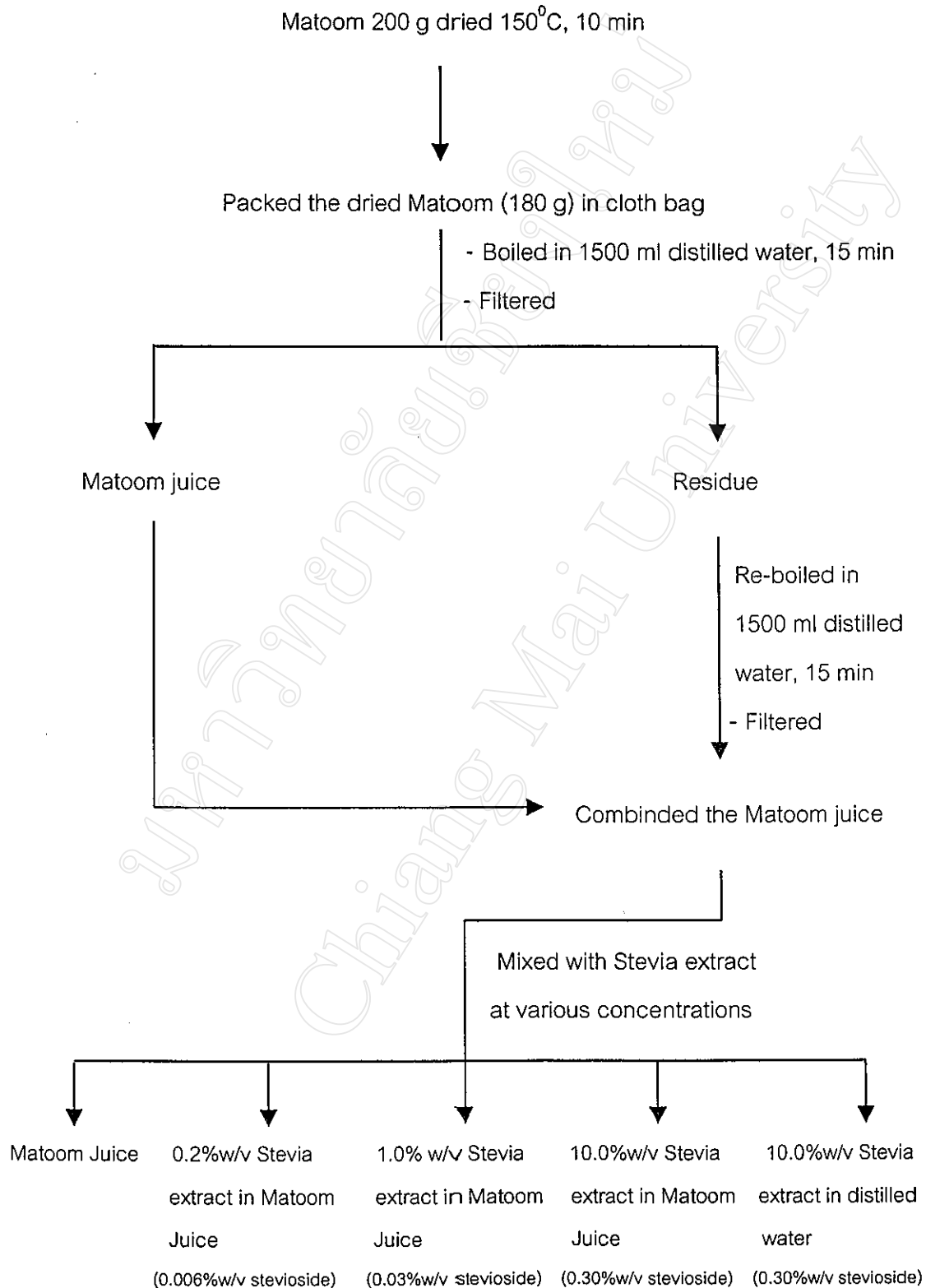




Figure 25. Matoom (*Aegle marmelos*, Corr.), Family: Rutaceae

Gel Preparation for SDS-PAGE Slab Gels

Stock Solution

A. Acrylamide/bis (30%T, 2.67%C)

| | |
|-------------------------------|----------|
| Acrylamide | 29.2 g |
| N'N'-bis-methylene-acrylamide | 0.8 g |
| Distilled water | 100.0 ml |

Filtered and stored at 4°C in the dark (30 days maximum)

B. Sample buffer (SDS reducing buffer)

| | |
|-----------------------------|--------|
| Distilled water | 4.0 ml |
| 0.5 M Tris-HCl, pH 6.8 | 1.0 ml |
| Glycerol | 0.8 ml |
| 10%(w/v) SDS | 1.6 ml |
| 2-beta-mercaptoethanol | 0.4 ml |
| 0.05%(w/v) bromophenol blue | 0.2 ml |

Sample diluted at least 1:4 with sample buffer, and heated at 95°C for 4 minutes.

C. 5x electrode (Running) buffer, pH 8.3

| | |
|-----------------|-----------|
| Tris base | 15.0 g |
| Glycine | 72.0 g |
| SDS | 5.0 g |
| Distilled water | 1000.0 ml |

Stored at 4°C Warmed to 37°C before use if precipitation

Occurred. Diluted 60 ml 5x stock with 240 ml dH₂O for one electrophoretic run.

Separating Gel Preparation

| | |
|-------------------------|---------------|
| Distilled water | 3.35 ml |
| 1.5 M Tris-HCl, pH 8.8 | 2.5 ml |
| 10% SDS (w/v) SDS stock | 100.0 μ l |
| Acrylamide/bis | 4.0 ml |
| 10% ammonium persulfate | 50.0 μ l |
| TEMED | 5.0 μ l |

Stacking Gel preparation

| | |
|-------------------------|---------------|
| Distilled water | 6.1 ml |
| 1.5 M Tris-HCl, pH 6.8 | 2.5 ml |
| 10% SDS (w/v) SDS stock | 100.0 μ l |
| Acrylamide/bis | 1.3 ml |
| 10% ammonium persulfate | 50.0 μ l |
| TEMED | 10.0 μ l |

CURRICULUM VITAE

Name : Suthasinee Sriwanna

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Education

March, 1991 Certificate of primary school, Satri Nakhonsawan school,
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March, 1994 Certificate of secondary school, Nakhonsawan school,
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May, 1998 Bachelor of Science (Biochemistry and Biochemistry
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Presentation:

1. Sriwanna S., Manosroi A., Manosroi J., Vinitketkumnue U. Effect of Stevia extract in Matoom juice on aberrant crypt foci formation in rats colon. The fourth Princess Chulabhorn International Science Congress "Chemicals in the 21st Century". November 28-December 2, 1999 Bangkok, Thailand, p.165.
 2. Sriwanna S., Manosroi A., Manosroi J., Vinitketkumnue U. Effect of Stevia extract on aberrant crypt foci formation in rats colon. Takeo Wada Cancer Research Symposium. November 30 - December 1, 2000, Faculty of Medicine, Chiang Mai University, Thailand, p.72.
 3. Sriwanna S., Manosroi A., Manosroi J., Vinitketkumnue U. Effect of
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Matoom juice with Stevia extract as sweetener on aberrant crypt foci formation in rats colon. The 8th World Congress On Clinical Nutrition "Functional Food, Nutrition and Health In the New Millennium". December 17-20, 2000, Phitsanulok, Thailand, p.123.

4. Sriwana S., Manosroi A., Manosroi J., Vinitketkumnuen U. Effect of Stevia extract in Matoom juice on aberrant crypt foci formation in rat colon. The first Annual Biochemical Research Meeting. October 18-19, 2001, Department of Biochemistry, Faculty of Medicine, Chiang Mai University, Thailand, p.36.

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