

## REFERENCES

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## **APPENDIX**

**Appendix A List of chemicals and instrument with their company**

**Table 14 List of chemicals, their companies, and their grade**

Chemical/Materials	Companies	Grade
ABTS (2,2 azino-bis-(3-ethylbenz-thiazoline-6-sulfonic acid) diammonium salt	Sigma Chemical Co.	AR
Acetic acid	E. Merk	AR
Acetic acid, glacial	E. Merk	AR
Acetic acid	E. Merk	HPLC
Acetone	Labscan	AR
Acetonitrile	E. Merk	HPLC
Adenosine-5'-triphosphate	Fluka	AR
Agarose gel	Sigma Chemical Co.	AR
Ammonium iron(III) sulfate	E. Merk	AR
L-Ascorbic acid	Fluka	AR
Bathophenantholine disulfonate	Sigma Chemical Co.	AR
Boric acid	Scharlad chemical	AR
Bovine serum albumin	Sigma Chemical Co.	AR
Butan-1-ol	Labscan	AR
Bromine solution	E. Merk	AR
Calcium chloride	Spectrum Chemical	AR
Catechin	Sigma Chemical Co.	AR
(+) Catechin hydrate	Fluka	AR
Chloroform	E. Merk	AR
Dichloromethane	Lab-Scan	AR
Diethylamine	Malinckrodt	AR
Diethyl ether	Lab-scan	AR
Diethyl maleate	E. Merk	AR
Dimethyl sulfoxide(DMSO)	Sigma Chemical Co.	AR

**Table 14 (Cont.)**

<b>Chemical/Materials</b>	<b>Companies</b>	<b>Grade</b>
1,1-diphenyl-2-picryl hydrazyl(DPPH)	Fluka	AR
DMEM medium	Hyclone HyQ	AR
Epicatechin	Fluka	AR
Ethanol, absolute	E. Merk	AR
Ethyldium bromide	Vivantis Technologies	AR
Ethylenediamine tetraacetic acid(EDTA)	Scharlad Chemical	AR
Ethyl acetate	Lab-scan	AR
Ethyl formate	Sigma Chemical Co.	AR
Ferric chloride	Ucb	AR
Ferrous sulfate	Sigma Chemical Co.	AR
Fetal bovine serum	Gibco-BRL	AR
Formic acid	Lab-scan	AR
Gallic acid	Sigma Chemical Co.	AR
Gelatin	Sigma Chemical Co.	AR
D-(+) Glucose	Sigma Chemical Co.	AR
Glucose oxidase(GO)	Aurora	AR
Grape seeds extract (French paradox)	Arkopharma	Food
Glutatione assay kit	Sigma Chemical Co.	AR
HEPES sodium	Sigma Chemical Co.	AR
Hexane	Lab-Scan	AR
Hydrochloric acid	E. Merk	AR
Hydrogen peroxide	Carlo	AR
Magnesium chloride	Spectrum Chemical Co.	AR
Methanol	E. Merk	AR
Methanol	E. Merk	HPLC
MOPs	Sigma Chemical Co.	AR
Myoglobin	Sigma Chemical Co.	AR

**Table 14 (Cont.)**

<b>Chemical/Materials</b>	<b>Companies</b>	<b>Grade</b>
Octyl alcohol	E. Merk	AR
Penicillin	Sigma Chemical Co.	AR
Potassium chloride	Mallinckrodt	AR
Potassium permanganate	Carlo ERBA	AR
Potassium persulfate	Sigma Chemical Co.	AR
Potassium phosphate monobasic	Sigma Chemical Co.	AR
Potassium phosphate dibasic	Sigma Chemical Co.	AR
Polyvinylpyrrolidone (PVP)	Sigma Chemical Co.	AR
pUC 18 plasmid DNA	Vivantis Technologies	AR
Quercitin	Sigma Chemical Co.	AR
Rutin	Sigma Chemical Co.	AR
Sephadex G15	Sigma Chemical Co.	AR
Sephadex G100	Pharmacia Fine Chemicals	AR
Sephadex LH 20	Sigma Chemical Co.	AR
Siliga gel GF254	E. Merk	AR
Sodium carbonate	EM Science	AR
Sodium chloride	Mallinckrodt	AR
Sodium dihydrogen phosphate	Mallinckrodt	AR
Sodium hydroxide	E. Merk	AR
Sulfuric acid	Labscan	AR
Streptomycin	Sigma Chemical Co.	AR
Tert-butanol	Aldrich	AR
Toluene	E. Merk	AR
Trichloroacetic acid	E. Merk	AR
Trifluoroacetic acid	Sigma Chemical Co.	HPLC

Table 14 (Cont.)

Chemical/Materials	Companies	Grade
Tris(Tris[Hydroxymethyl]aminomethane	Usb	AR
Trolox (6-hydroxy-2,5,7,8-tetramethyl chlorman-2-carboxylic acid)	Sigma Chemical Co.	AR
Vanillin	Sigma Chemical Co.	AR
Zinc dust	Sigma Chemical Co.	AR

Table 15 Lists of instruments and their companies

Instruments	Companies
Centrifuge Micro22R	Hettick Zentrifugen
Microcentrifuge	Herolab
Column for chromatography	Opond Glass,
Detector HP 845x UV-VIS System	Thailand
Spectrophotometer UV-2401PC	Hillet Packard
	Shimadzu, Japan
Spectrophotometer UV-1700 Pharma Spec	Shimadzu, Japan
Synergy <sup>TM</sup> HT Multi-Detection Microplate Reader	Bio-TEK, USA
Syngene G-Box	Lab Focus, German
SCL-10AVP HPLC system consisting of a LC-10ADvp Pump, an SIL-10ADvp autosampler and SPD-10AV detector	Shimadzu, Japan
AVANCE <sup>TM</sup> NMR spectrometer	Bruker
The Agilent 1100 series HPLC system was coupled to a PE SCIEX API 4000	Applied Biosystem, Foster city, CA
510-FT-IR Spectrometer	Nicolet

Table 15 (Cont.)

Instruments	Companies
Spectrum GX FTIR Spectrometer	Perkin Elmer
Microcentrifuge (micron13)	Herolab
pH meter	Corning
Water bath, 37 oC	Lab-line
Water bath, 100 oC	Memmert
Balance	Precisa
Vacuum rotating evaporator	Buchi

**Appendix B Data showing characteristics of tested samples fractionated by HPLC using solvent system 1 and maximum absorption wavelengths of peaks from UV spectra**

**Table 16 Characteristics of TA hydrolysate fractionated by HPLC and UV absorption wavelengths of their peaks**

Peaks from HPLC				Wavelengths of peaks from UV (200-360nm)			Suggested compounds
Number	RT (min)	Height (a.u.)	Width (a.u.)	Peak 1 (nm)	Peak 2 (nm)	Peak 3 (nm)	
1	1.35	0.0287	0.073	ND	ND	ND	Standard
2	1.4	0.0027	0.05	ND	ND	ND	ND
3	1.52	0.0284	0.178	ND	ND	ND	ND
4	1.7	0.006	0.175	ND	ND	ND	ND
5	2.03	0.0051	0.175	ND	ND	ND	ND
6	2.37	0.0131	0.225	ND	ND	ND	ND
7	3.42	0.0025	0.275	ND	ND	ND	ND
8	13.98	0.0053	0.534	222.5	-	-	Non-flavonoids
9	14.6	0.0039	0.414	ND	ND	ND	ND
10	15.97	0.0044	0.294	222.5	290	-	Unknown
11	17.42	0.009	0.188	ND	ND	ND	ND
12	18.55	0.0144	0.102	222.5	274	310	Unknown
13	18.72	0.0766	0.073	222.5	274	-	Catechin
14	19.4	0.0055	0.138	222.5	275	-	Flavonoids
15	19.83	0.0048	0.124	222.5	274	-	Flavonoids
16	20.18	0.0021	0.072	ND	ND	ND	ND
17	20.8	0.0103	0.182	235	278	-	Flavonoids
18	21.38	0.0008	0.078	ND	ND	ND	ND
19	21.72	0.0084	0.236	238	274	-	Flavonoids
20	22.28	0.0012	0.233	ND	ND	ND	ND

Table 16 (Cont.)

Peaks from HPLC				Wavelengths of peaks from UV (200-360nm)			Suggested compounds
Number	RT (min)	Height (a.u.)	Width (a.u.)	Peak 1 (nm)	Peak 2 (nm)	Peak 3 (nm)	
21	22.6	0.0029	0.123	ND	ND	ND	ND
22	22.97	0.0265	0.174	226	286	-	Flavonoid
23	23.28	0.0044	0.146	ND	ND	ND	ND
24	23.73	0.0022	0.109	ND	ND	ND	ND
25	24.13	0.0525	0.14	225	260	283	Unknown
26	24.55	0.0015	0.228	ND	ND	ND	ND
27	25.12	0.0023	0.92	ND	ND	ND	ND
28	25.62	0.0015	0.133	ND	ND	ND	ND
29	25.88	0.002	0.209	240	-	-	Unknown
30	26.38	0.0017	0.113	ND	ND	ND	ND

**Note:** ND = Not determined

There were at least 29 compounds found in hydrolysate of TA (Table 16). The peak 13 at the retention time 18.72 was identified as catechin. Peaks 4,5, and 11 were also found after catechin (Sigma) injection but they were not the main compound (Table 18). Another main product of TA hydrolysate was Peak 22 (height 0.0265) which should be flavonoid.



**Table 17 Characteristics of GSE hydrolysate by HPLC and UV wavelengths of their peaks**

Peaks from HPLC				Wavelengths of peaks from UV (200-360nm)			Suggested compounds
Number	RT	Height	Width	Peak 1	Peak 2	Peak 3	
	(min)	(a.u.)	(a.u.)	(nm)	(nm)	(nm)	
4	1.85	0.028	0.202	ND	ND	ND	ND
	6.6	0.0081	4.83	-	270	-	unknown
11	17.52	0.0078	0.146	ND	ND	ND	ND
12	17.98	0.0127	0.091	ND	ND	ND	ND
13	18.68	0.0939	0.074	222.5	274	-	Catechin
15	19.7	0.0191	0.095	222.5	274	-	Flavonoids
17	20.85	0.0143	0.119	235	278	-	Flavonoids
19	21.5	0.0249	0.108	238	274	-	Flavonoids
	22.95	0.0098	0.1	238	286	-	Flavonoids
25	24.1	0.028	0.11	238	255	278	Unknown
29	25.85	0.329	0.11	240	-	-	Unknown

**Note:** ND = Not determined

At least 9 of compounds which found in TA hydrolysate were also found in GSE hydrolysate (peak 4,11,12,13,15,17,19,25 and 29 of Table 16).

**Table 18 Characteristics of catechin by HPLC and UV wavelengths of their peaks**

Peaks from HPLC				Wavelengths of peaks from UV (200-360nm)			Suggested compounds
Number	RT	Height	Width	Peak 1	Peak 2	Peak 3	
	(min)	(a.u.)	(a.u.)	(nm)	(nm)	(nm)	
1	1.35	0.0006	0.258	ND	ND	ND	Standard
4	1.67	0.0092	0.093	ND	ND	ND	ND
5	1.97	0.0367	0.167	ND	ND	ND	ND
11	17.58	0.001	0.113	ND	ND	ND	ND
13	18.42	0.161	0.133	222.5	274	-	Catechin
	33.57	0.0115	0.168	ND	ND	ND	ND

**Table 19 Characteristics of mTAS hydrolysate by HPLC and UV wavelengths of their peaks**

Peaks from HPLC				Wavelengths of peaks from UV (200-360nm)			Suggested compounds
Number	RT	Height	Width	Peak 1	Peak 2	Peak 3	
	(min)	(a.u.)	(a.u.)	(nm)	(nm)	(nm)	
3	1.52	0.0203	0.155	ND	ND	ND	Standard
	1.82	0.0275	0.258	ND	ND	ND	ND
11	17.73	0.0162	0.133	222.5	274	310	Unknown
13	18.72	0.1664	0.076	222.5	274	-	Catechin
17	20.8	0.0101	0.191	235	278	-	Flavonoids
22	23.02	0.021	0.123	226	286	-	Flavonoids
25	24.18	0.0714	0.11	225	260	283	Unknown

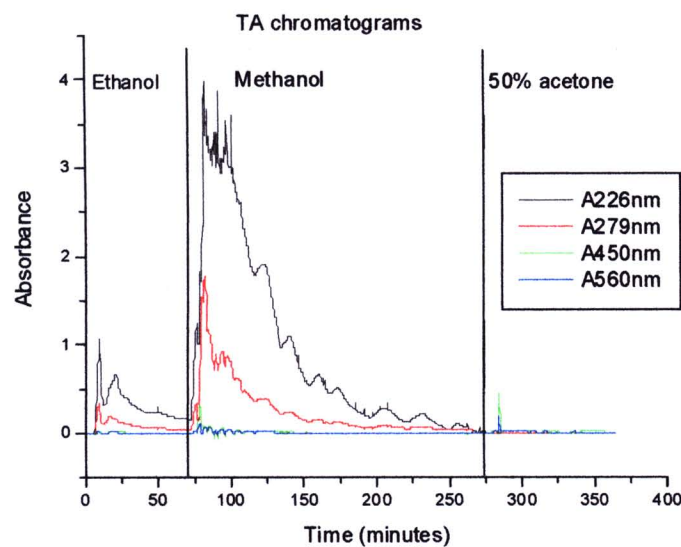
**Note:** ND = Not determined

**Table 20 Characteristics of aTAS hydrolysate by HPLC and UV wavelengths of their peaks**

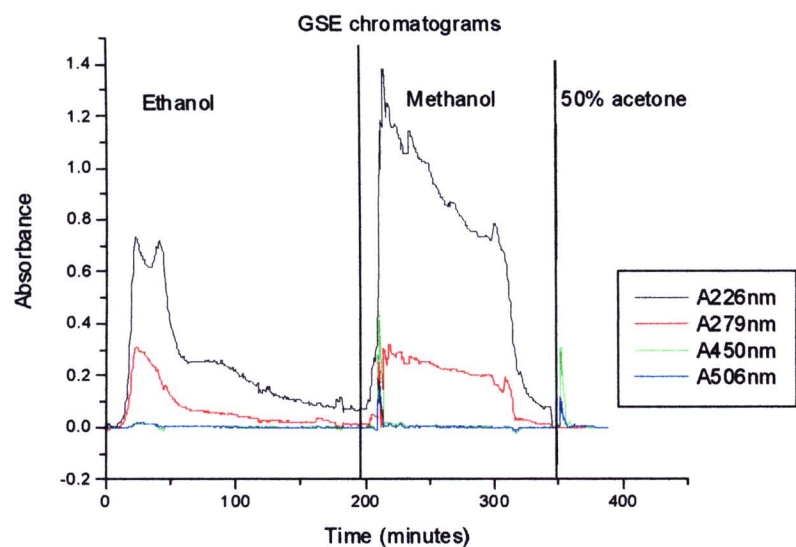
Peaks from HPLC				Wavelengths of peaks from UV (200-360nm)			Suggested compounds
Number	RT	Height	Width	Peak 1	Peak 2	Peak 3	
	(min)	(a.u.)	(a.u.)	(nm)	(nm)	(nm)	
1	1.33	0.4883	0.222	ND	ND	ND	Standard
6	2.27	0.3141	0.064	ND	ND	ND	ND
9	14	0.0054	0.696	222.5	-	-	Non-flavonoids
10	15.72	0.0085	0.368	ND	ND	ND	ND
11	17.3	0.0079	0.304	ND	ND	ND	ND
13	18.75	0.0017	0.12	222.5	274	-	Catechin
14	19.37	0.0019	0.168	222.5	275	-	Flavonoids
16	20.17	0.0094	0.153	ND	ND	ND	ND
	20.47	0.0112	0.259	222.5	275	-	Flavonoids
17	20.95	0.0104	0.246	ND	ND	ND	ND
18	21.22	0.0126	0.283	ND	ND	ND	ND
19	21.53	0.0139	0.349	ND	ND	ND	ND
21	22.5	0.0048	0.146	ND	ND	ND	ND
22	22.87	0.0373	0.133	226	286	-	Flavonoids
23	23.27	0.0195	0.782	ND	ND	ND	ND
24	23.72	0.0023	0.16	ND	ND	ND	ND
25	24.08	0.0965	0.135	225	260	283	Unknown
26	24.58	0.0031	0.117	ND	ND	ND	ND
	24.95	0.0073	0.098	ND	ND	ND	ND
27	25.13	0.0028	0.231	ND	ND	ND	ND
28	25.67	0.0025	0.117	ND	ND	ND	ND
29	25.9	0.0004	0.33	ND	ND	ND	ND
30	26.38	0.007	0.105	ND	ND	ND	ND
	27.35	0.0017	0.118	ND	ND	ND	ND

**Note:** ND = Not determined

**Appendix C Chromatogram of TA and GSE purification from sephadex LH20 column chromatography showing the absorbance value**

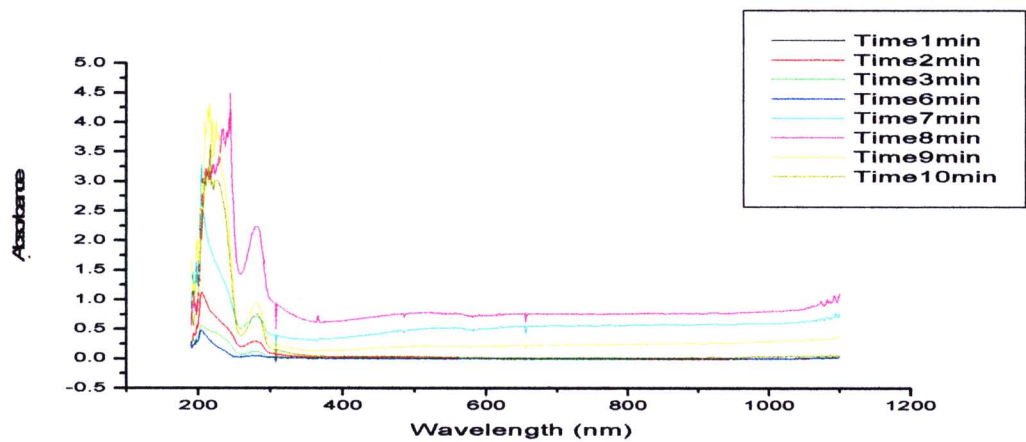


**Figure 67 Chromatogram of TA purification from sephadex LH20 column chromatography showing the absorbance value**

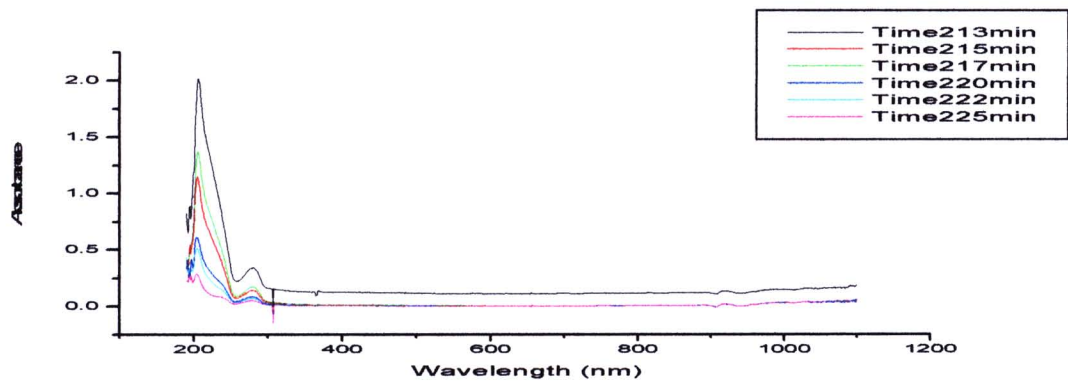


**Figure 68 Chromatogram of GSE purification from sephadex LH20 column chromatography showing the absorbance value**

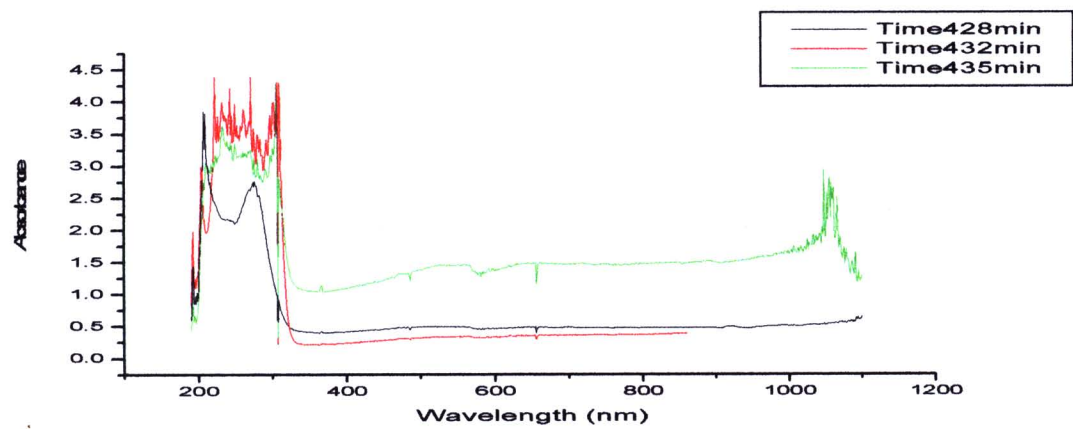
**Appendix D UV/VIS spectra of eTAS, mTAS and aTAS isolated from Sephadex  
LH20 column chromatography**



**Figure 69 UV/VIS Spectrum of ethanol fraction TA(eTAS)**



**Figure 70 Spectrum first peak of methanol fraction TA (mTAS)**



**Figure 71 Spectrum first peak of acetone fraction TA(aTAS)**

## **Appendix E Buffer preparation for neutral red assay**

### **HEPES buffer preparation for 1 liter (Neutral red assay)**

1. 125 mM NaCl (Mallinckrodt)
2. 5 mM KCl (Mallinckrodt)
3. 1.8 mM  $\text{CaCl}_2 \cdot 2 \text{H}_2\text{O}$  (Spectrum Chemical Mfg. Corp.)
4. 2 mM  $\text{MgCl}_2 \cdot 6 \text{H}_2\text{O}$  (Spectrum Chemical Mfg. Corp.)
5. 0.5mM  $\text{NaH}_2\text{PO}_4 \cdot \text{H}_2\text{O}$  (Mallinckrodt)
6. 5 mM  $\text{NaHCO}_3$  (EM Science)
7. 10mM HEPES (Sigma)

After that, the buffer solution was adjusted pH to 7.2 and was added with 10mM D(+)glucose(Sigma) before using in the experiment.

## Appendix F The reagents and their preparations used in ABTS/Metmyoglobin/H<sub>2</sub>O<sub>2</sub> experiment

### Stock phosphate buffer saline (0.15 M PBS)

NaCl	8.00 g
KCl	0.20 g
Na <sub>2</sub> HPO <sub>4</sub>	1.15 g
K <sub>2</sub> HPO <sub>4</sub>	0.20 g

Dissolved in distil water (final concentration 1,000 ml) and adjusted pH to 7.4. Stored at temperature 4 °C. The working PBS at the concentration of 5 mM was prepared.

### Myoglobin preparation

Myoglobin stock solution(400μM)

Myoglobin 0.17 g dissolved in 5 Mm PBS (final volumn 25 ml)

Potassium ferricyanide 0.1203 g dissolved in distill water (final volumn 500 ml) and then added equal volume of myoglobin solution to potassium ferricyanide solution.

Metmyoglobin was purified by gel filtration chromatography (Sephadex G100). Metmyoglobin fraction was collected. The final concentration of purified metmyoglobin was estimated by applying the Whitburn equations:

$$\begin{aligned}
 [\text{Met Mb}] &= 146A_{490} - 108A_{560} + 2.1A_{580} \\
 [\text{Ferryl Mb}] &= -62A_{490} + 242A_{560} - 123A_{580} \\
 [\text{MbO}_2] &= 2.8A_{490} - 127A_{560} + 153A_{580}
 \end{aligned}$$

Where Mb is Metmyoglobin. These equations are derived by solving simulataneous equation base on Beer's law, measuring the absorbance at 490,560 and 580nm and subtracting the reading at 700 nm to correct for background absorbance. The purity of the metmyoglobin prepared was estimated by applying all the three equations. Normally the metmyoglobin fraction is 95% of the total heme protein.

### Stock ABTS solution (5mM)



ABTS            0.06998 g

Dissolved in 5 mM PBS (final volume 25 ml). Stored at 4 °C.

Working ABTS solution was diluted 10 times from stock solution.

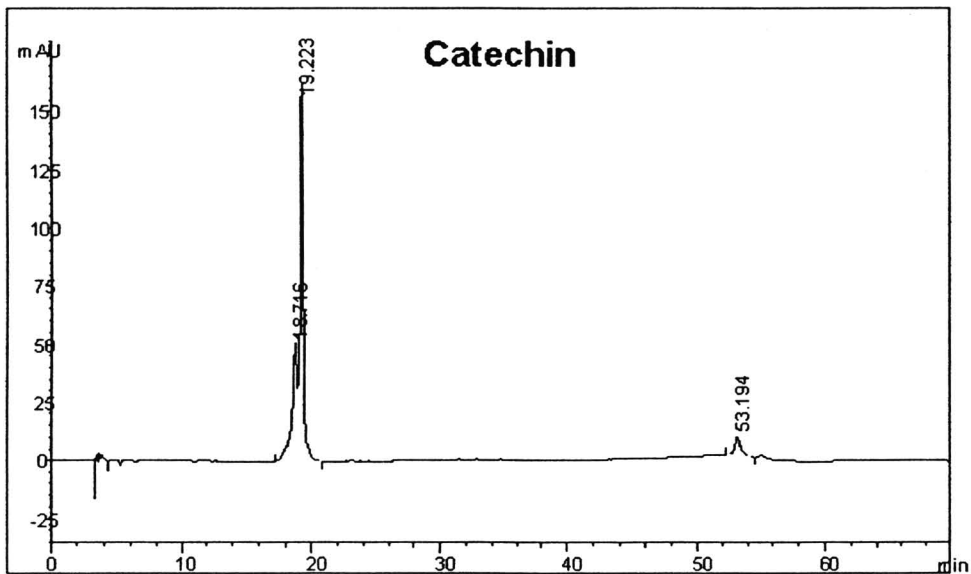
### Hydrogen peroxide preparation

Hydrogen peroxide (30-35%) 1ml dissolved in distilled water to 100 ml and titrated with potassium permanganate solution (0.02 M  $\text{KMnO}_4$ )

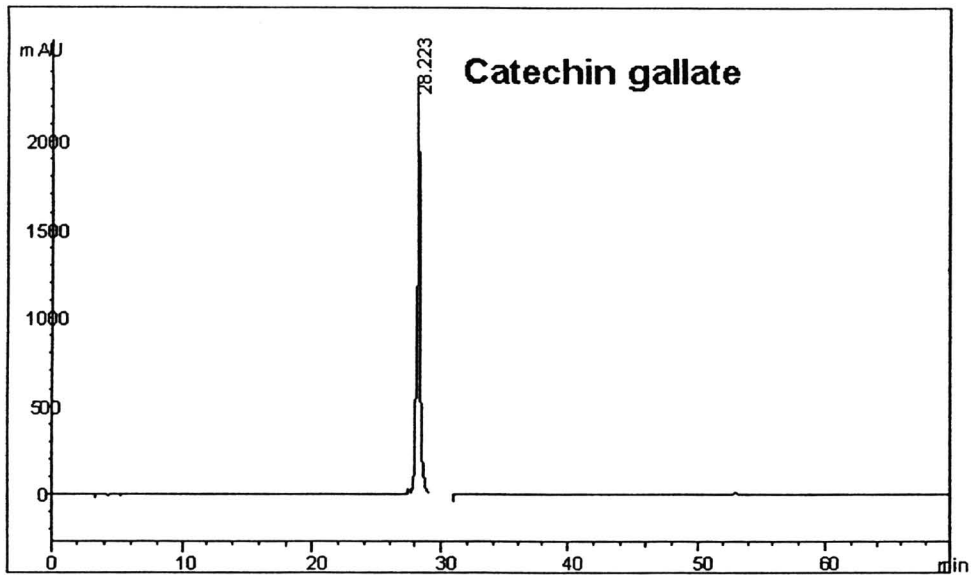
Titrant	0.1 N $\text{KMnO}_4$	
Titrand	2 N sulfuric acid	20 ml
	Hydrogen peroxide	2 ml
	Distilled water	20 ml

Calculated concentration of hydrogen peroxide solution from end point and prepared working hydrogen peroxide solution (500  $\mu\text{M}$ ).

**Appendix G HPLC profiles of the flavonoids standard and hydrolysate of TA and its fractions using eluting solvent system 2**



**Figure 72 HPLC profile of catechin using eluting solvent system 2**



**Figure 73 HPLC profile of catechin gallate using eluting solvent system 2**

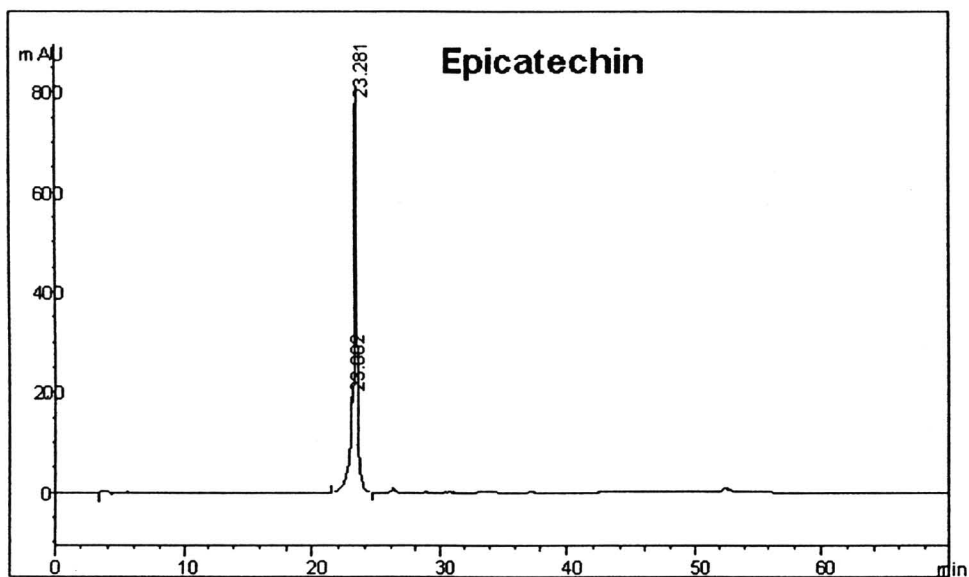


Figure 74 HPLC profile of epicatechin using eluting solvent system 2

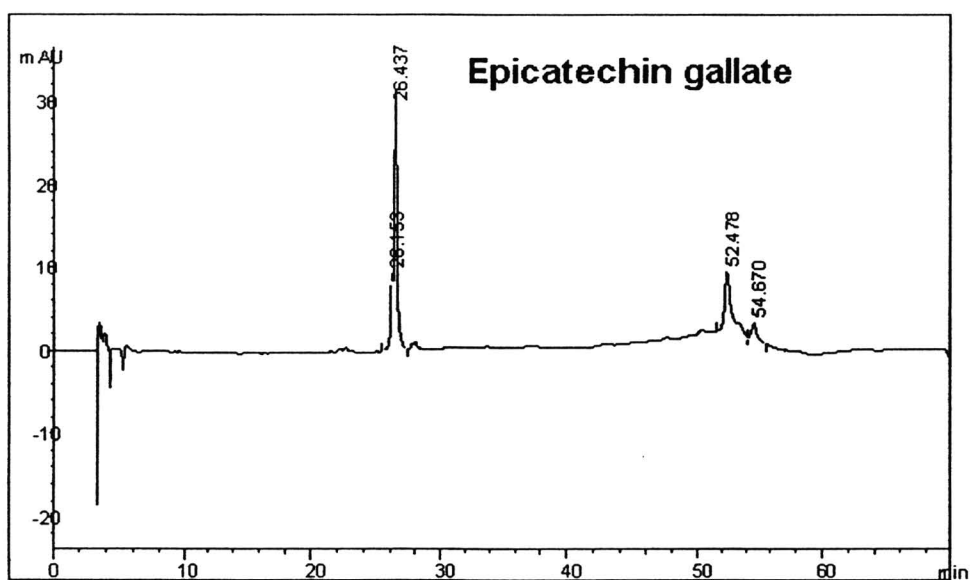


Figure 75 HPLC profile of epicatechin gallate using eluting solvent system 2

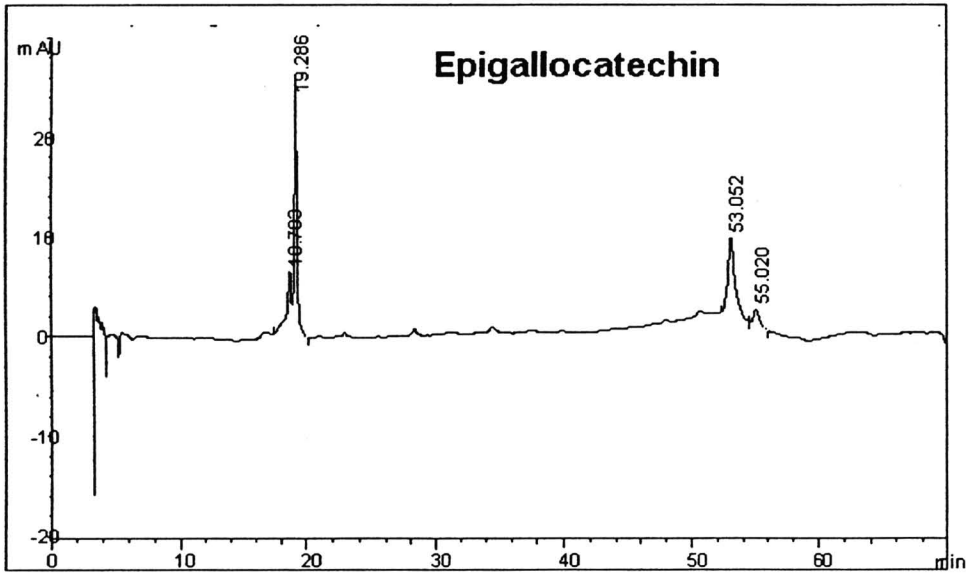


Figure 76 HPLC profile of epigallocatechin using solvent system 2

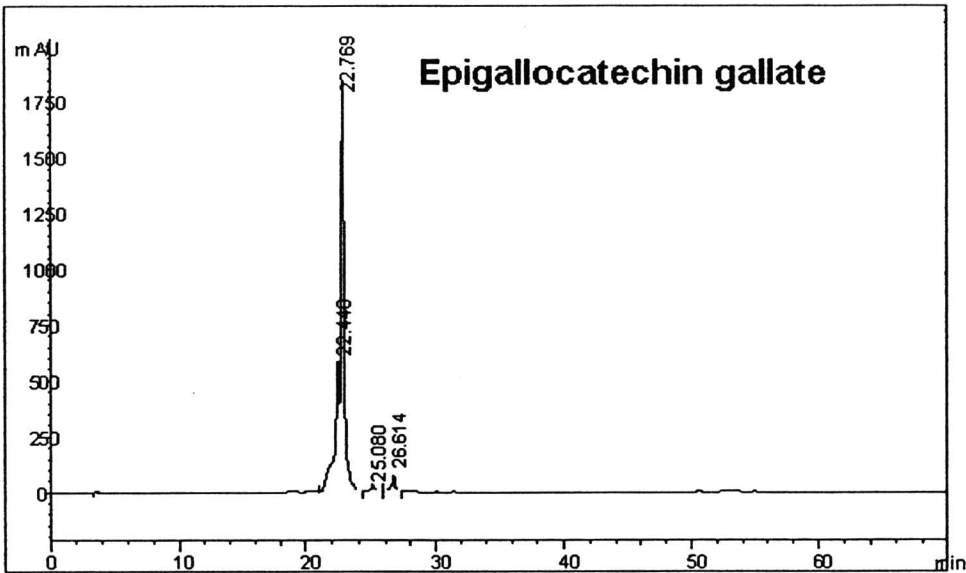


Figure 77 HPLC profile of epigallocatechin gallate using eluting solvent system 2

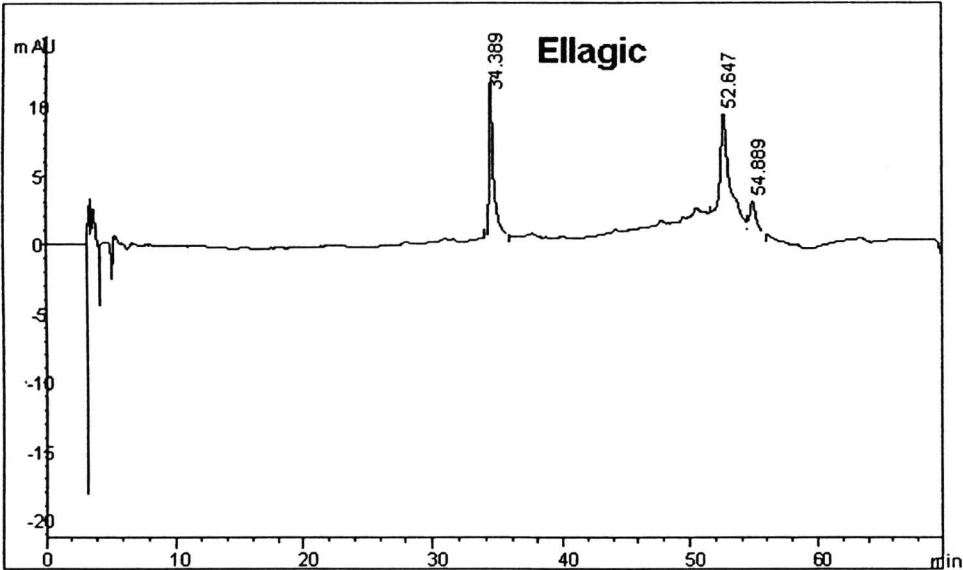


Figure 78 HPLC profile of ellagic acid using eluting solvent system 2

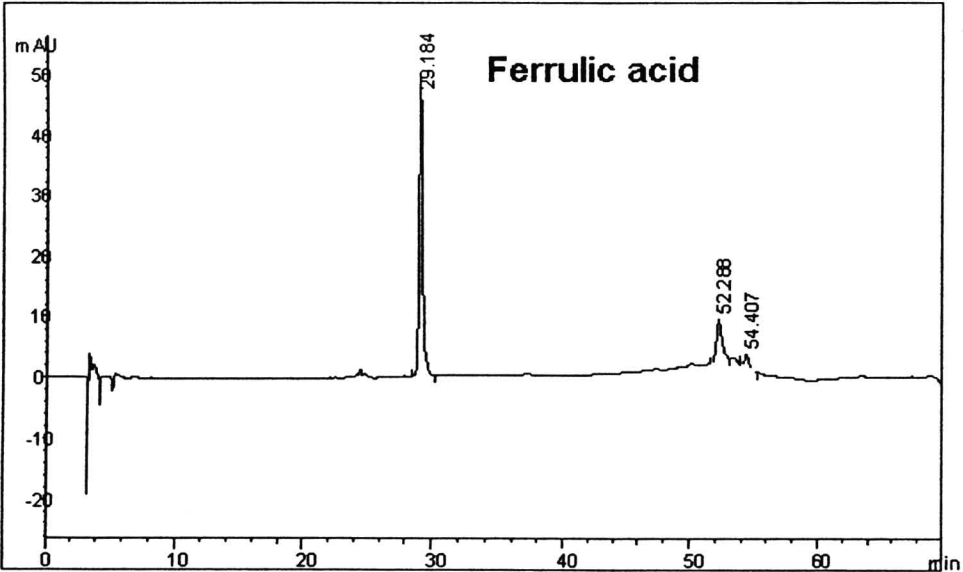


Figure 79 HPLC profile of ferrulic acid using eluting solvent system 2

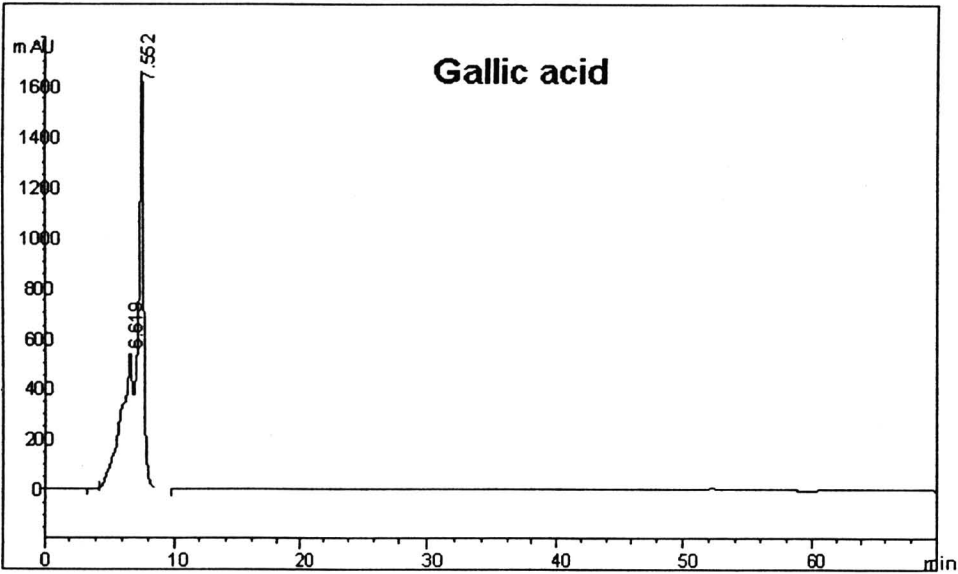


Figure 80 HPLC profile of gallic acid using eluting solvent system 2

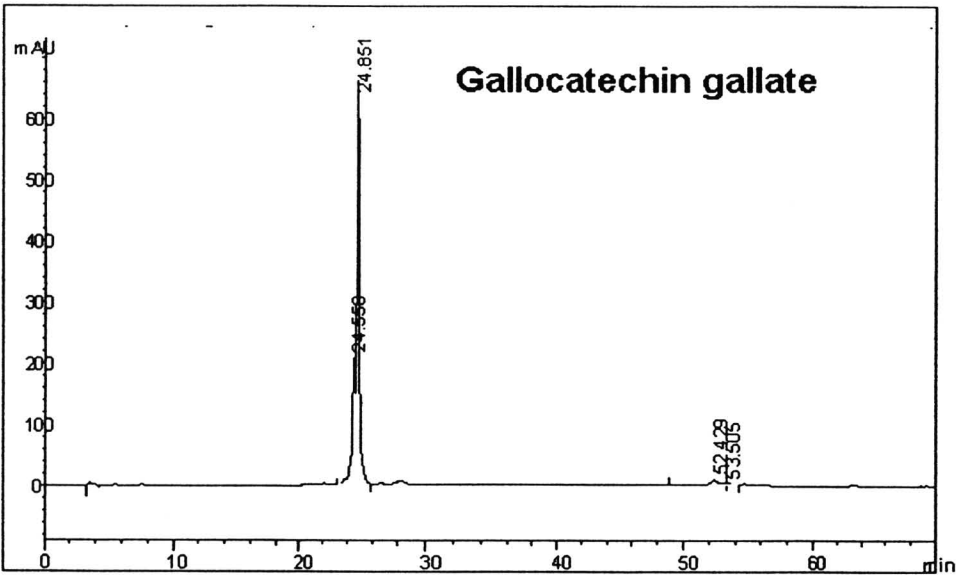


Figure 81 HPLC profile of gallocatechin gallate using eluting solvent system 2

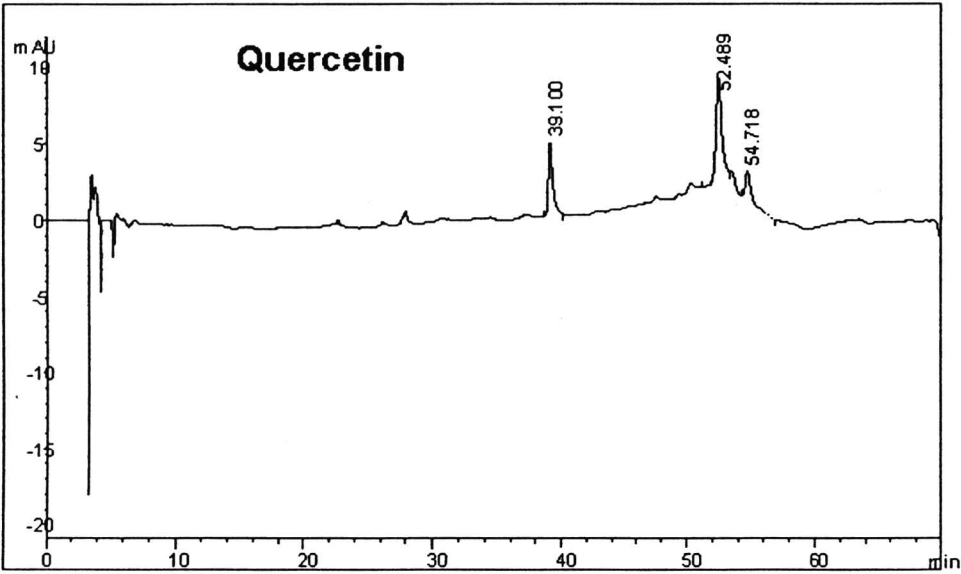


Figure 82 HPLC profile of quercetin using eluting solvent system 2

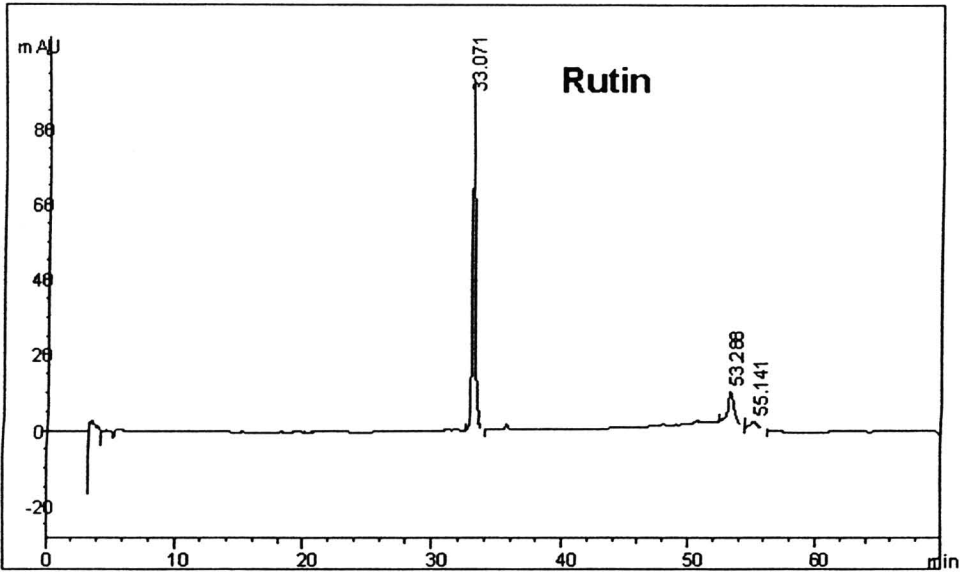


Figure 83 HPLC profile of rutin using eluting solvent system 2

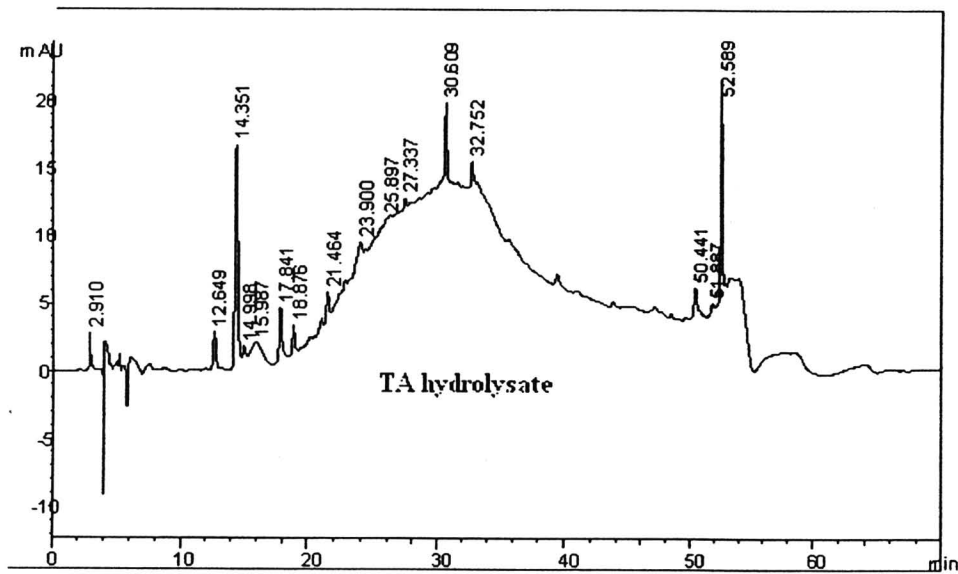


Figure 84 HPLC profile of TA hydrolysate using eluting solvent system 2

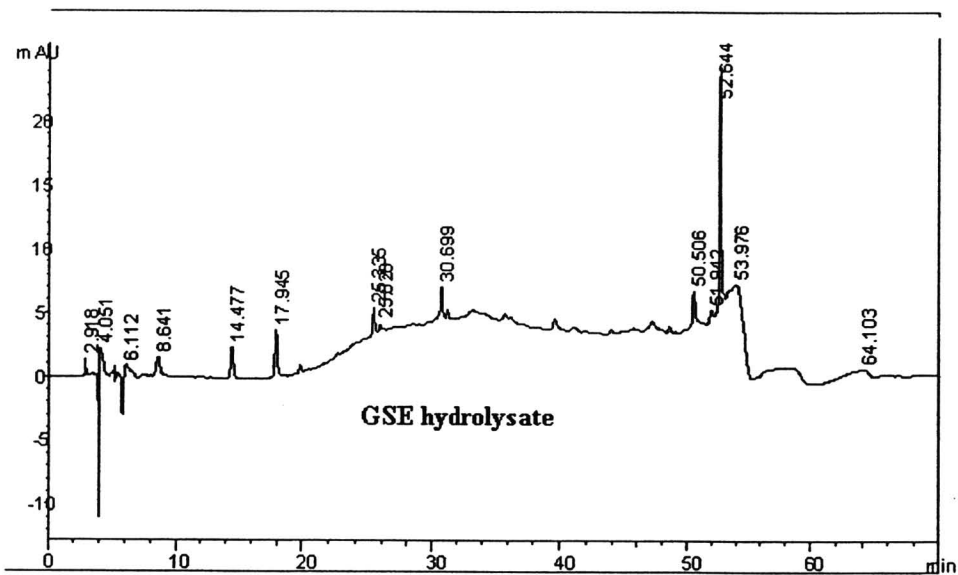


Figure 85 HPLC profile of GSE hydrolysate using solvent system 2



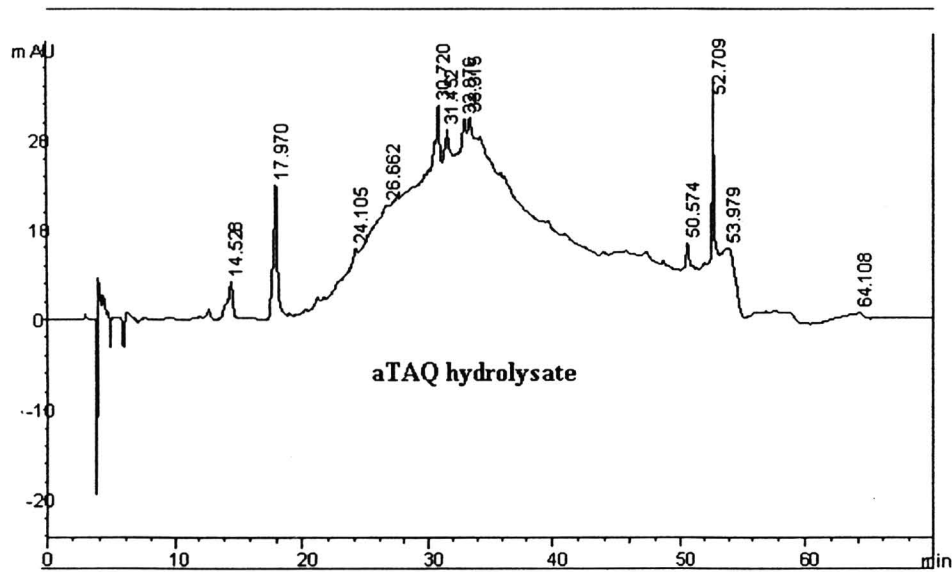


Figure 86 HPLC profile of aTAQ hydrolysate using solvent system 2

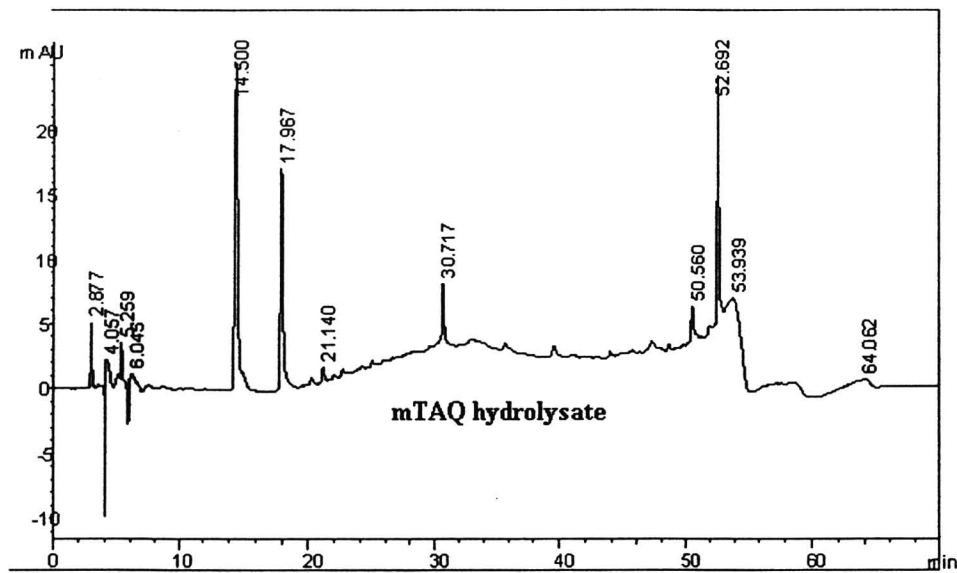
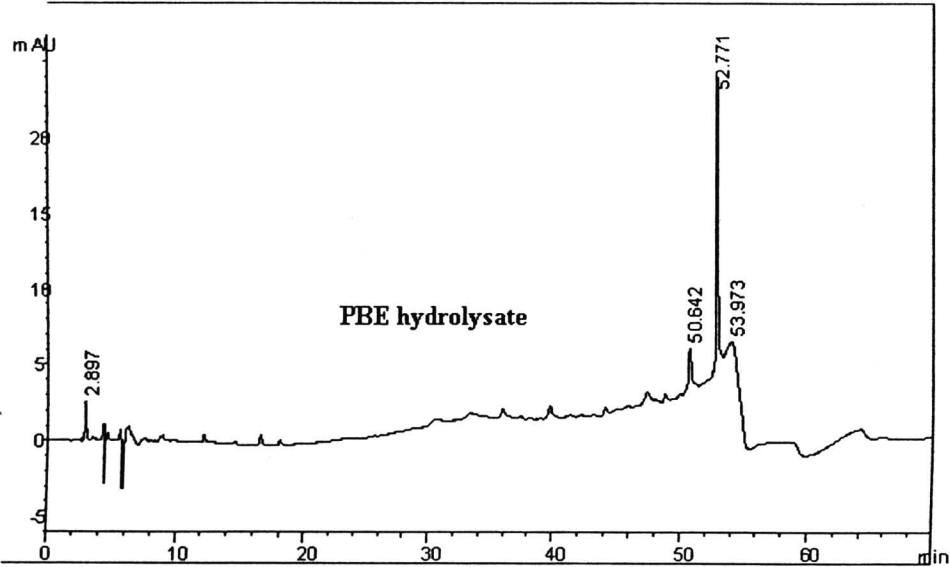


Figure 87 HPLC profile of mTAQ hydrolysate using solvent system 2



**Figure 88 HPLC profile of PBE hydrolysate using solvent system 2**

## **Appendix H Preparation of reagent from glutathione assay kit (Sigma)**

### **Precipitating stock solution**

DTNB Stock Solution (1.5 mg/ml) – Dissolve the contents of the bottle (8 mg) of DTNB with 5.33 ml of DMSO to make a 1.5 mg/ml solution. The solution may be stored in aliquots at -20°C for at least 3 months

5% 5- sulfosalicylic-acid (SSA) Solution – Dissolve the contents of the bottle of 5-sulfosalicylic acid (2.5 g) in 50 ml of water. Ensure the powder is completely dissolved. Keep at 4°C.

Glutathione (GSH) standard stock solution (10 mM) – Dissolve the contents of the vial of Glutathione Reduced, Standard in 0.1 ml of water. The solution may be stored at -20°C at least 3 months.

### **Working solution**

The volumes prepared are sufficient for 48 reactions of 200µl performed in a 96 well plate.

1× Assay Buffer (12ml) – 100mM potassium phosphate buffer, pH 7.0, with 1mM EDTA. Dilute 2.4 ml of Assay Buffer five- fold by addition of 9.6 ml of water.

Working Mixture (8ml) – To 8 ml of 1× Assay Buffer, add 228 µl of DTNB Stock Solution (1.5 mg/ml). Mix well. This solution may be kept for up to 3 hours at room temperature.

## **Appendix I Preparations of reagents for DNA damage method**

### **Preparation of 5× Tris-borate buffer (TBB)**

The Tris-borate buffer (TBB) contains Tris-base 54 g, boric acid 27.5 g and 20 ml of 0.5 M EDTA pH 8.0 in distilled water 1,000ml was prepared and keep in room temperature.

### **Preparation of agarose gel**

The agarose 1.2 g in 1× TBB 100 ml was heated until the gel was completely dissolve. Then pour the homogeneous gel in the tray, put the comb in to make wells, wait until the gel turn to solid form. The thickness of gel is 0.5 cm. to prevent Joule heating.

Appendix J Proton NMR of aTES and aGES

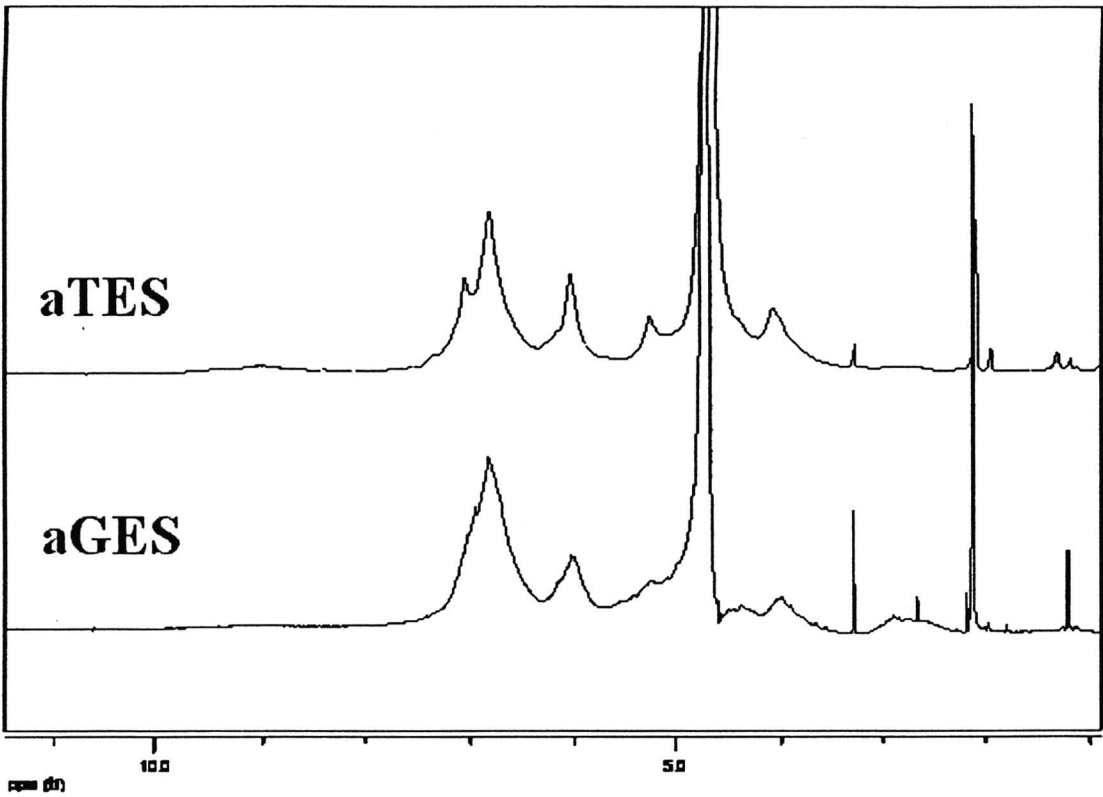
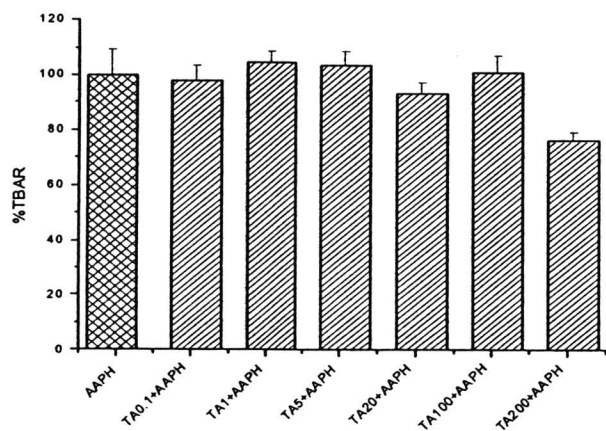


Figure 89 H-NMR spectra for polymeric PA of aTES and aGES.

**Appendix K Determination lipid peroxidation of TA by TBARS method**

This method was modified from the method of Prabhakar (2007). A solution of soybean phosphatidylcholine (247.36 mg) and cholesterol (30.92 mg) in chloroform (20 ml) was dried under vacuum in a rotary evaporator (< 50 °C) to yield a thin, homogenous film, which was placed in desiccators for 24 h. The film was then dispersed in phosphate buffered saline (PBS) solution (pH 7.2, 20 ml) in a water bath (50 °C). The mixture was sonicated to obtain a homogeneous suspension of liposome. Lipid peroxidation was initiated by adding 60 µl of AAPH (2,2'-azobis (2-amidinopropane) hydrochloride) to the mixture containing liposome (600 µl). AAPH generated peroxy radicals in the presence of oxygen. The reaction mixture was incubated at 50 °C for 24 h. After incubation, 250 µl of thiobarbituric acid (0.6% w/v), 100 µl of Triton X-100 (3% v/v) and 500 µl of BHT (20% v/v) were added to terminate the reaction. The samples were heated at 90 °C for 30 min, and then allowed to cool. The absorbance of the upper organic layer was measured by a multimode detector at 540 nm. The results shown that the TA can inhibit AAPH-induced lipid peroxidation at the concentration more than 100 µg/ml. This result was in a good agreement with Povichit (2010). He reported that *J. gossypifolia*, *T. indica*, *C. sinensis*, *T. bellerica* and *A. lakoocha* possessed high anti-lipid peroxidation, with IC<sub>50</sub> of 420, 640, 870, 1110 and 1,220 µg/ml, respectively.



**Figure 90 The effects of TA various concentration on AAPH-induced lipid peroxidation by thiobarbituric acid reactive substances (TBARS) method**

**GLOSSARY**

## GLOSSARY

Antioxidant	Any substance which inhibits a free radical reaction.
Condensed tannin	Tannin possesses an abundance of flavonoid groups. Condensed tannin is a type of tannin that, on heating with hydrochloric acid, yields phlobaphenes like phloroglucinol. (Wikipedia)
Extinction coefficient	The extinction coefficients are parameters defining how strongly a substance absorbs light at a given wavelength, per concentration.
Fenton reaction	Fenton reaction is the iron-salt-dependent decomposition of hydrogen peroxide, generating the highly reactive hydroxyl radical. $\text{Fe}^{2+} + \text{H}_2\text{O}_2 \longrightarrow \text{Fe}^{3+} + \text{OH}^\cdot + \text{OH}^\cdot$
Free radical	A free radical is any atom or molecule that has a single unpaired electron in an outer shell.
Flavan-3-ols	Flavan-3-ols (sometimes referred to as flavanols) are a class of flavonoids that use the 2-phenyl-3,4-dihydro-2H-chromen-3-ol skeleton. Flavanols are building blocks for proanthocyanidins. (Wikipedia)
Flavonoids	Flavonoids, derived from 2-phenylchromen-4-one (2-phenyl-1, 4-benzopyrone structure.
Glutathione (GSH)	Glutathione is a tripeptide that contains an unusual peptide linkage between the amine group of cysteine and the carboxyl group of the glutamate side-chain. It is an antioxidant, preventing damage to important cellular components caused by reactive oxygen species such as free radicals and peroxides. (Wikipedia)



## GLOSSARY

IC <sub>50</sub>	The half maximal inhibitory concentration of samples.
Nuclear magnetic resonance (NMR)	NMR is a property that magnetic nuclei have in a magnetic field and applied electromagnetic (EM) pulse of pulses, which cause the nuclei to absorb energy from the EM pulse and radiate this energy back out. (Wikipedia)
Oxidation	Loss of electrons by an atom or molecule
Oxidation number	The oxidation number of a central atom in a coordination compounds is the charge that it would have if all the ligands were removed along with the electron pairs that were shared with the central atom. (Wikipedia)
Phytochemicals	Phytochemicals are chemical compounds that occur naturally in plants. (Wikipedia)
Polyphenol	A polyphenol is a chemical compound belonging to a group of chemical substances found in plants, characterized by the presence of more than one phenol unit or building block per molecule. (Wikipedia)
Proanthocyanidins	The substances are based structurally on a flavan-3-ol structure which is a kind of flavonoids-like polyphenol. Proanthocyanidins are characterized by the anthocyanidins which are formed from them when they are digested with mineral acid.
Reactive nitrogen species (RNS)	Nitrogen-center radicals or nitrogen-center reactive substance (may be have a net charge of zero).
Reactive oxygen species (ROS)	Reactive oxygen species are chemically-reactive molecules containing oxygen (may be have a net charge of zero). (Wikipedia)

## GLOSSARY

Retention factor ( $R_f$ )	The retention factor ( $R_f$ ) is calculated by measuring the distance an analyze travels up a chromatographic plate divided by the distance traveled by the solvent front.
Sephadex LH20	Sephadex LH-20 is beaded, cross-linked dextran which has been hydroxypropylated from Sephadex G-25 to yield a chromatographic media with both hydrophilic and lipophilic character.
Tannin	The bitter plant polyphenolic compound that either binds and precipitates or shrinks proteins and various other organic compounds including amino acids and alkaloids. (Wikipedia)
Trolox equivalent antioxidant activity (TEAC)	The TEAC is defined as a ratio of the capacity of the substance that can inhibit the radical formation per that by one micromole of trolox (Vitamin E analog). TEAC reported is calculated by comparing with 1 mg dried residue of the test substance.

## **BIOGRAPHY**



## BIOGRAPHY

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Sinchaiyakit, P., Ezure, Y., Sriprang, S., Pongbangpho, S., Povichit, N.  
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