CHAPTER 3
EXPERIMENT

3.1 Simulation Section

3.1.1 Program

All computer programs were written in MatLab program (Maths Works Inc.). The programs have been adapted to operate on number of the personal computer (PC). The programs developed for this work are shown in Appendix A.

3.1.2 Data Preparation

3.1.2.1 The Simulated Spectra of Pure Component

The simulated spectra of pure component were generated by peak generating program using Gaussian function. The Gaussian peaks have various widths and intensities. All spectra are in the range of 1 to 100 cm^{-1}. The simulated spectral data were stored in ASCII format files and managed into matrix form called original data matrix.

3.1.2.2 The Mixture Spectra

The spectra of mixture were generated by multiplication of original data matrix and concentration matrix. The concentration matrix contains the volume fraction of all components in the mixture. The mixture matrix was separated into two parts, the calibration set and validation set. The calibration set contains the data of calibration samples of the mixtures at various proportions included the data of pure component that consisting in all mixtures. The calibration set must be developed and tested until the accuracy was satisfied. The validation set contains the data of testing sample.

3.1.3   Factor Analysis

Factor analysis process was initiated by loading the mixture data to factor analysis program. First, the number of component in mixture was rechecked by PCA. PCA is the first process of factor analysis and it is employed as the tool for preliminary qualitative analysis. PCA calculates the total numbers of components, which are in the mixture by using smaller number of mixture samples than that in EFA. In PCA, the number of the sample must be greater than the number of component in the mixture. When the number of component was known, it is easier to prepare the enough number of calibration samples for EFA.

The result from factor analysis program is the matrix of concentration. The number of column of the matrix equals to the number of the mixture, only column of testing sample will be considered. The reproduced spectra from factor analysis program showed the position of each volume fraction of pure component in result matrix.

3.1.4   Ability of Factor Analysis in Multicomponent Analysis

The simulated spectra of five pure components were generated. The spectra of mixture consists of three, four, and five components were constructed from the multiplication of the original data matrix and the concentration matrix. The result from matrix multiplication was loaded to factor analysis program. The ability of factor analysis when handling multicomponent mixture was investigated.

3.1.5   Effect of Random Error or Noise in Simulated Spectra

The simulated spectra of mixture consists of three components were generated. Relative three levels of random noise were added to the testing spectra. The data of testing spectra with noise added were loaded to factor analysis program. The ability of factor analysis when handling the data with added noise was investigated.

3.1.6 Effect of Peak Overlap in Simulated Spectra

The simulated spectra of mixture, which consists of three components were generated to demonstrate the effect of overlapping peak in simulated spectra, the three spectra have similar peak widths and peak positions. The spectra of the mixture of three components were generated. The spectra data was loaded to factor analysis program. The result from program was investigated.

3.1.7 Effect of Nonlinearlity in Simulated Spectra

The simulated spectra of mixture consists of three components were generated. The nonlinearlity was added to one component in the mixture. The difference in the response between a perfectly linear and nonlinear was investigated. The testing samples were selected at the proportion of that component, at which presents the much more difference from original is presented. The mixture data was loaded to factor analysis program. The result from factor analysis program was investigated.

3.1.8 The Improvement of Prediction Quality in Factor Analysis

The three rules of number of calibration sample in calibration set were tested with the simulated spectra of the mixture consists of three components. The three calibration sets were constructed by using the rule of Three, rule of Five, and rule of Ten. By using the same testing sample, the results from three calibration sets were compared.

3.2 Experimental Section

3.2.1 Determination of *o*-, *m*-, and *p*-xylene in Ternary Xylene Mixture

3.2.1.1 Materials and Equipment

1. ZnSe ATR prism (Liquid Cell)
2. Multiple attenuated total reflection accessory (Spectra Tech, USA)
3. Bruker Vector 33 FT-IR spectrometer
4. Pure three xylene isomers (The specification are shown in Appendix B)
5. *n*-Hexane
6. Micropipette 100 μl (Eppendorf Model 3130)

3.2.1.2 FT-IR Spectrometer Operating Conditions

Bruker Vector 33 FT-IR Spectrometer
Experimental Setup

Mode	ATR
Resolution	4.0 cm^{-1}
Number of Scans	16
Result Spectrum	Absorbance

Optic Parameters

Source Setting	Globar (MIR)
Detector Setting	DTGS
Beam Splitter Setting	KBr

3.2.1.3 Procedure

Twenty samples of ternary xylene were prepared from *o*-, *m*-, and *p*-xylene at designed proportions. The infrared spectra were measured by ATR FT-IR spectroscopy in the region of mid-infrared (500–4000 cm^{-1}). The samples were contained in a zinc selenide (ZnSe) crystal liquid cell. Only base line correction was applied to the measured spectra before performing factor analysis. In order to suppress spectra noise and reduce experimental error, averaging spectrum from three spectra with the same composition was obtained. The data was loaded to FA program. The volume fraction and reproduced spectra obtained by factor analysis were compared with the original experimental data.

3.2.2 Determination of *o*-, *m*-, and *p*-xylene in Real Mixed Xylene

3.2.2.1 Materials and Equipment

1. ZnSe ATR prism (Liquid Cell)
2. Multiple attenuated total reflectance accessory (Spectra Tech, USA)
3. Bruker Vector 33 FT-IR spectrometer
4. Pure three xylene isomers (The specification are shown in Appendix B)
5. *n*-Hexane
6. Micropipette 100 µl (Eppendorf Model 3130)

3.2.2.2 FT-IR Spectrometer Operating Conditions

Bruker Vector 33 FT-IR Spectrometer
Experimental Setup

Mode	ATR
Resolution	4.0 cm^{-1}

Number of Scans	16
Result Spectrum	Absorbance

Optic Parameters

Source Setting	Globar (MIR)
Detector Setting	DTGS
Beam Splitter Setting	KBr

3.2.2.3 Procedure

Fifteen samples of mixed xylene were prepared by adding o-, m-, and p-xylene into unknown mixed xylene for constructing the calibration set. The original mixed xylene was separated into 2 parts, one was sent to reliable lab for performing quantitative analysis by GC technique, the other one was kept as the sample for factor analysis evaluation. The infrared spectra of all mixed xylene were measured by ATR FT-IR spectroscopy in the mid-infrared region (500–4000 cm^{-1}). The samples were contained in a zinc selenide (ZnSe) crystal liquid cell. Only base line correction was applied to the measured spectra before performing factor analysis. The data was loaded to factor analysis program. The volume fraction and reproduced spectra obtained by factor analysis were compared with those obtained by GC technique.