CHAPTER IV

EXPERIMENTAL

4.1 Introduction

Chapter 4 provides the information on the experimental methods used for all the experiments carried out in the present work. Furthermore, this chapter explains the experimental detail in an order of raw materials, reactors and equipments, experimental conditions, experimental procedures, and product characterizations (divided into those for liquid and solid products). In raw materials section, we used different classes of carbohydrates such as native corn starch, modified starch, amylopectin, amylose and glucose to synthesize carbon microspheres. The reactors and equipments for hydrothermal process and carbonization process consisted of a Teflon-lined stainless autoclave, a horizontal tube furnace reactor, a vacuum flask set, and hot air oven. In the experimental conditions section, we described about initial concentration of carbon precursor, reaction temperature, and reaction time for hydrothermal process. In carbonization process, we addressed the carbonization conditions such as flowrate of nitrogen gas, heating rate, target temperature, and holding time. In the experimental procedures, we described the procedures in hydrothermal process and carbonization process in detail including carbon precursor preparation, reaction time, reaction temperature, product collecting (filtration and rinsing), drying of carbon microspheres, and storage of carbon microspheres in desiccators for preventing them from moisture. Finally, in the product characterization, we have divided the product characterizations into two parts. In solid characterization part, the CMS particles were characterized by many techniques to reveal their particularly properties both before and after the carbonization process. In liquid product characterization part, we have chosen main intermediates which formed during hydrothermal process including glucose, fructose, 5-HMF, furfural, and TOC compounds. The concentrations of intermediates were used to fit model for carbon microspheres formation which was separately described in Chapter 8.

4.2 Raw materials

The specifications of raw materials were used in the hydrothermal process and the carbonization process were shown in Table 4.1

Table 4.1 List of raw materials were used in this research

Raw materials	Used for	Manufacturers/grades
Native starch -Corn starch -Tapioca starch -Rice starch -Sticky rice starch -Wheat starch	Synthesis of carbon microspheres	General commercial source
Modified starch -HI-CAP®100 -CAPSUL®	Synthesis of carbon microspheres	National Starch and Chemical Ltd, (Bangplee, Thailand)/food grade
- Amylopectin - Amylose - Glucose	Synthesis of carbon microspheres	Sigma Aldrich
- Hydroxymethyl furfuraldehyde (5-HMF) - Furfural	Standard for HPLC analysis	Sigma Aldrich
- De-mineralized water	Synthesis of carbon microspheres	Production from MilliQ apparatus (Millipore, Bedford, MA).
-Ethanol	Rinsing carbon microspheres	Merck/analytical grade
-Nitrogen (N_2)	Carbonization process	TIG/purity 99.999%

4.3 Reactors and equipments

4.3.1 Teflon-lined stainless autoclave reactor

Figure 4.1 shows a Teflon-lined stainless autoclave reactor, of which containing of Teflon tube inside and stainless steel outside, for hydrothermal process. Figure 4.2 shows and a horizontal tube furnace reactor for carbonization process.

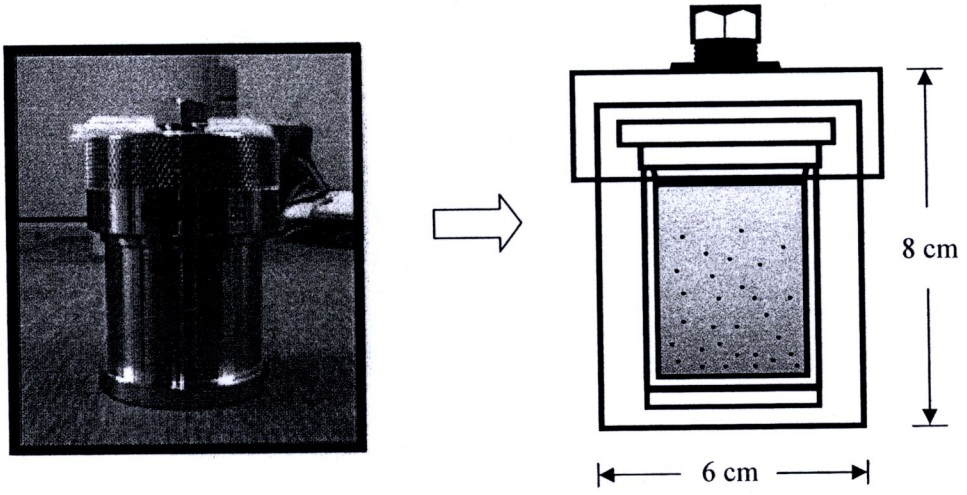

Figure 4.1 Illustration of a Teflon-lined stainless autoclave

4.3.2 Horizontal furnace reactor for carbonization process

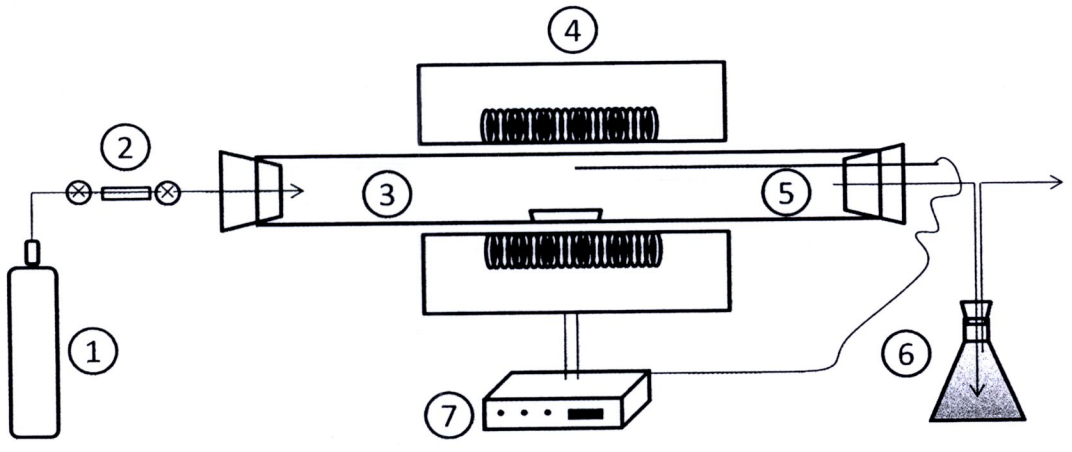

Figure 4.2 Schematic diagram of the quartz tube furnace reactor used in this work, it is composed of (1) N_2 gas container, (2) N_2 gas rotameter, (3) cylindrical quartz tube, (4) furnace, (5) thermocouple, (6) flask containing alcohol for residual trap, and (7) furnace controller

4.3.3 Vacuum suction flask set

Products from hydrothermal process were separated into solid products and liquid products. Figure 4.3 shows a vacuum suction flask, of which including flask, and vacuum pump for separating the solid products from the liquid products.

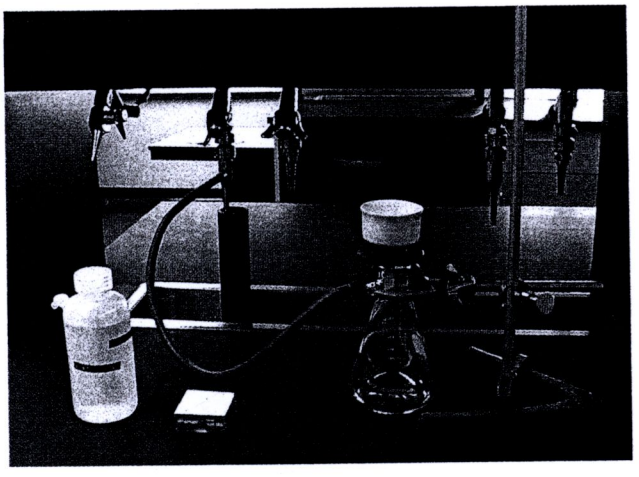

Figure 4.3 A vacuum suction flask for separation of solid samples from liquid samples

4.3.4 PVDF membranes for filtration

The solid products were the particles suspended in the liquid products or precipitated under bottom of storage bottles. The former was obtained by filtering the liquid product through polyvinylidene fluoride membranes (0.45 μm pore size) as shown in Figure 4.4.

Figure 4.4 Polyvinylidene fluoride membranes for filtration

4.3.5 A hot air oven for drying of CMS particles

The CMS particles after removing residual byproducts, were dried in a hot air oven (as shown in Figure 4.5) at 100°C until constant weight. After a drying step, the CMS particles were cooled down to room temperature and collected in a desiccator for preventing them from moisture.

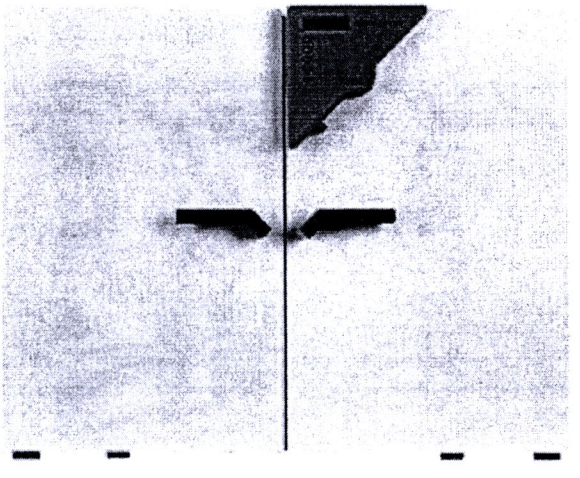

Figure 4.5 The hot air oven for drying of CMS particles

4.3.6 A desiccators for carbon microspheres storage

After the drying step, the CMS particles were dried and collected in the desiccator again, and weighed to a constant weight as shown in Figure 4.6.

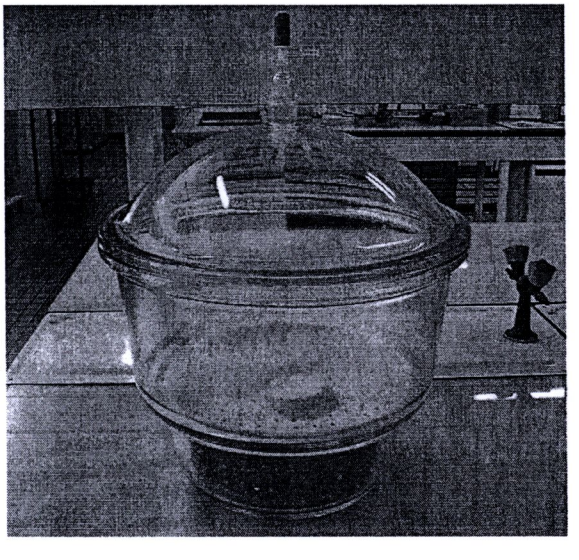

Figure 4.6 The desiccator for storage of CMS particles

4.4 Experimental conditions

The experiments were carried out under the hot compressed water (140-220°C) conditions so that the difference in the reaction mechanisms between each temperature condition could be systematically compared. There are four chapters with the results and discussion in this study: comparison between native corn starch and modified starch (Chapters 5), pure amylopectin and pure amylose (Chapter 6), pure glucose (Chapter 7), and a carbon microspheres formation and a kinetic model (Chapter 8). The typical experimental conditions for the hydrothermal process and the carbonization process were shown in Table 4.1

Table 4.1 Experimental conditions for hydrothermal and carbonization process

Hydrothermal Process	
Temperature (°C)	140, 180 and 220
Pressure	autogenously
Starch initial concentration (wt%)	1,5,10,15,20
Fill rate in reactor (%v/v)	80
Reaction time (min)	0, 30, 60, 120, 150, 180, 240, 360, 540, 720, 900, 1080, 1260 and 1440
Carbonization Process	
Heating rate (°C/min)	1
Target temperature (°C)	600
Holding time (h)	3
Flowrate of N_2 gas (mL/min)	100

4.5 Experimental procedures

4.5.1 Hydrothermal process of CMSs step

4.5.1.1 Hydrothermal process of CMSs

A carbon precursor (native corn starch, modified starch, amylopectin, amylase and glucose) was mixed with de-mineralized water (generally 10wt%), and then the suspension was stirred at room temperature for 15 minutes. After complete dissolution of the carbon precursor, the mixture was filled in a 50 mL Teflon-lined stainless autoclave with 80 %v/v fill rate. Subsequently, the autoclave was put into a hot air oven, which was heated to set point temperature (180°C or 220°C). After reaction time reached, the autoclave was finally cooled to room temperature naturally.

4.5.1.2 Filtration and rinsing of CMSs

Liquid products were filtrated through a 0.45 μm pore size syringe filter and kept in a refrigerator at 5°C for further TOC and HPLC analysis. Dark precipitate solids were collected and rinsed with de-mineralized water and ethanol several times to remove an organic residual.

4.5.1.3 Drying of CMSs

The obtained powders were dried in an oven at 100°C until constant weight of CMSs. Finally, the solid products were kept in a desiccator in order to prevent them from humidity.

4.5.1.4 Storage of CMSs

The CMS particles after drying until constant weight were stored in a desiccator with fresh silica gel for preventing the CMS particles from moisture. The CMS particle were characterized in order to reveal their many particularly properties.

4.5.2 Carbonization of CMSs step

The obtained powders were carbonized in a horizontal tube furnace reactor under nitrogen (N_2) atmosphere. The N_2 flow rate, target temperature, heating rate of the furnace, and holding time will be 100 mL/min, 600°C, 1°C/min, and 3 hours, respectively (see Figure 4.7). In brief, A 1.0 gram of the CMS particle after drying step was placed in alumina boat. The alumina boat was taken in the tube furnace reactor which subsequently set to the operating conditions. After the horizontal tube furnace reactor cooled to room temperature naturally, the porous CMS particles were collected and weighted for further characterization.

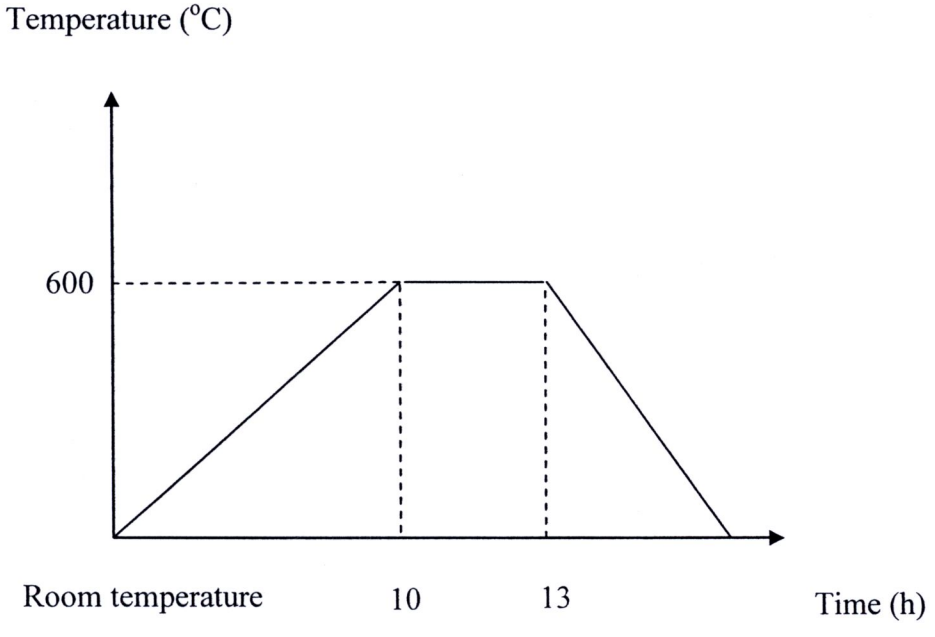

Figure 4.7 Schematic diagram of the carbonization condition of CMSs

The porous CMS particles were characterized by X-ray diffraction method (XRD) to reveal their crystalline properties. They were determined their specific surface area both before and after carbonization process to reveal the development of porous structure using adsorption – desorption of nitrogen or the Brunauer, Emmett, Teller method (BET method). Moreover, elemental components of the porous CMS particles were determined by energy dispersive X-ray method (EDX) to demonstrate carbon content in their structure. The transmission electron microscopy (TEM) was also used to reveal their internal structure.

4.6 Product characterizations

Figure 4.8 gives an overview of the product analyses used in this study. The reaction products are divided into liquid and solid, as follows. In the solid product characterizations, the CMS particles were characterized by CHNS/O analyzer to reveal elemental components in their structure. They were also analyzed by FT-IR technique (determined functional groups), SEM (demonstrated morphology), EDX technique (determined carbon content in porous CMS particles) BET technique (determined their surface area), and XRD technique (revealed their crystalline structure). In the liquid products characterization, the liquid products were analyzed by HPLC (determined concentrations of intermediates) and TOC analyzer (determined concentrations of total carbon compound in liquid products).

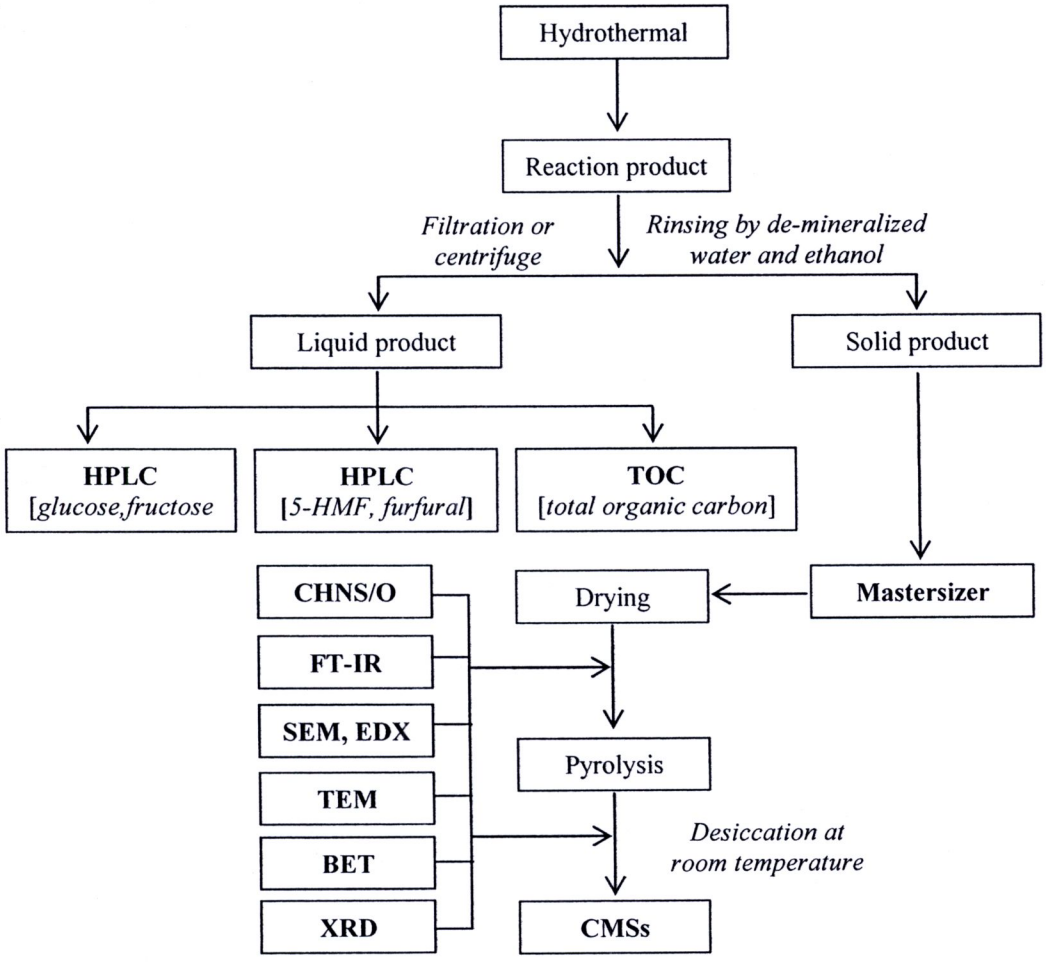

Figure 4.8 Schematics of an overview of product characterizations

4.6.1 Liquid product characterizations

4.6.1.1 HPLC analysis

First of all, the liquid products were filtered to obtain a clear liquid product. Then, 5-HMF, and furfural were quantitatively analyzed by HPLC (high-performance liquid chromatography) with an RSpak DE-413L column (Shodex). The following conditions were used for the analysis: flowrate 0.4 mL/min; eluent 0.005 M of H_3PO_4 aqueous solution; oven temperature 60°C; UV/vis detector_1 = 284 nm, detector_2 = 274 nm, detector_3 = 280 nm, detector_4 = 240 nm and detector_5 = 210 nm. The sample injection volume was 20 μL. Figure 4.9 is an example of an HPLC graph plotting the peaks of identified (5-HMF and furfural) and unknown liquid components. The 5-HMF and furfural peaks were observed at about 52 min and 83 min, respectively. Glucose and fructose were also analyzed by HPLC using a Lichrocart amino-NH_2 250x4 mm ID, packing 5 μm (Shimadzu LC-3A, LDC 4100) with a condition; 89% acetonitrile and 11% H_2O, 1.5 mL/min, 25°C of detector 20 μL sampling.

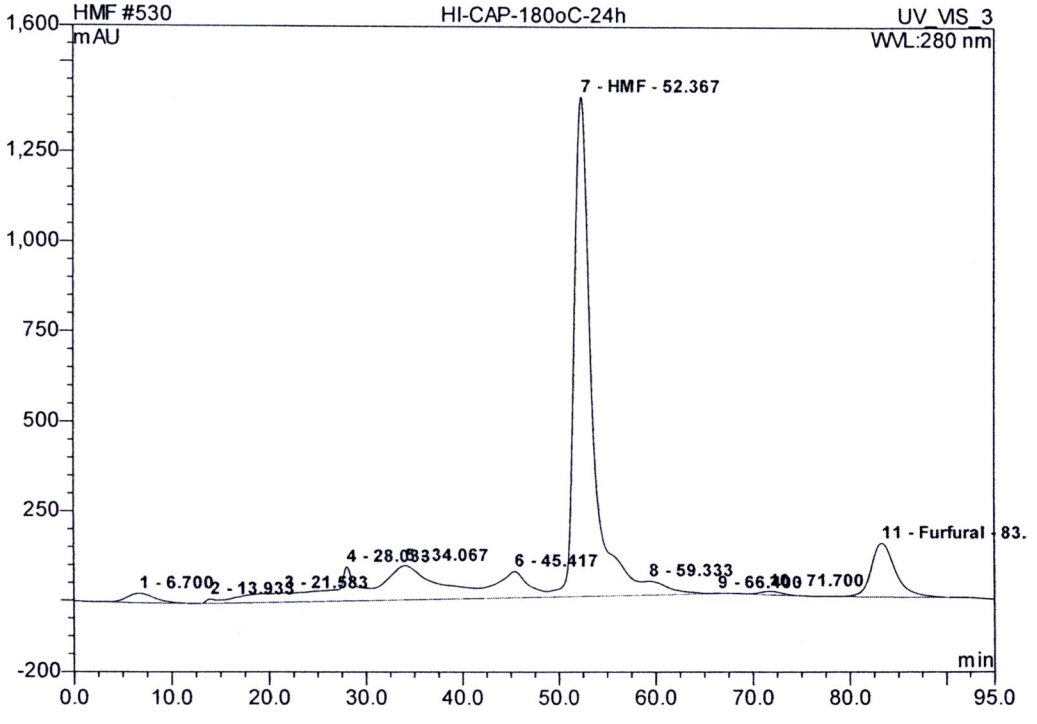

Figure 4.9 An HPLC graph plotting of 5-HMF and furfural components

4.6.1.2 TOC analysis

The liquid products were also analyzed by a total organic carbon (TOC) analyzer (Shimadzu TOC-V CHP) to quantify the amounts of carbon in the liquid products (non-purgeable organic carbon or NPOC) and in the dissolved gas product (inorganic carbon or IC). Standards were standard total carbon (STD TC) and standard inorganic carbon (STD IC). Potassium hydrogen phthalate (KHP) which was dried at 105°C for 1 hr, was dissolved with ultra pure water to obtain STD TC (10-1000 ppm). A mixture of sodium bicarbonate which was calcined at 280°C for 1 hr and sodium carbonate which was desiccated for 2 hr at 1.60 mole ratio, was dissolved to obtain STD IC (10-1000 ppm). A 2 Molar of Hydrochloric acid and 25 %v/v of phosphoric acid were used for nonpurgeable organic carbon analysis. An air zero gas was used to decompose organic carbon to CO_2 gas for detecting. The following conditions were used for the analysis: carrier gas flowrate and pressure were 150 mL/min and 200 kPa, respectively; the oven temperature 686-700°C.

4.6.2 Solid product characterization

4.6.2.1 Filtration

The solid products were the particles suspended in the liquid products or precipitated under bottom of storage bottles. The former was obtained by filtering the liquid product through polyvinylidene fluoride (PVDF membranes) membranes (0.45 μm pore size) with a vacuum suction. Before the filtration, the filter membrane and porcelain crucible were dried overnight in the desiccator at room temperature and weighed. De-ionized water was added several times to ensure that no trace of liquid products were left on the membrane. After the filtration, the membrane with the filtered solids was placed onto a porcelain crucible, dried overnight in the desiccator again, and weighed to a constant weight.

4.6.2.2 Particle size analysis

Particle size distributions and uniformity of CMS particles, were determined by laser scattering analyzer (*Mastersizer 2000: Malvern, United Kingdom*) as shown in Figure 4.10. In brief, 1.0 gram of the CMS particles after rinsing was filled in a sample vessel which had water as dispersant. The sonication was performed in order to disperse the CMS particles in the dispersant before measuring. The reflexive index of the CMS particle was set to 2.40 and the reflexive index of water was 1.33. The measurement was performed with three cycles repeating in each sample.

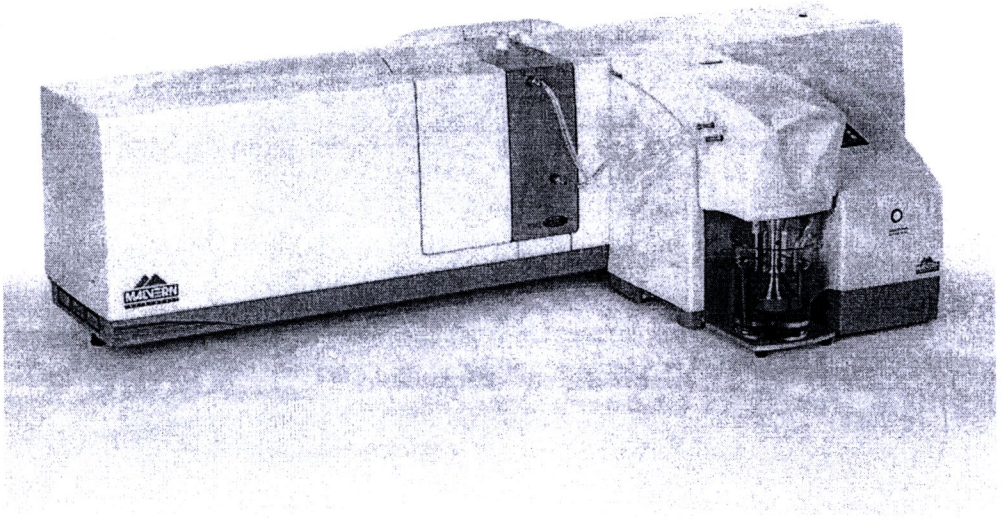

Figure 4.10 Laser scattering analyzer (*Mastersizer 2000: Malvern, United Kingdom*)

After the measurement, raw data was analyzed to obtain geometric mean particle size (d_g) using equation (4.1) [47]. Geometric standard deviation (σ_g) was calculated to demonstrate size distribution of the CMS particles using equation (4.2) [47]. Moreover, geometric coefficient of variance (CV_g) was also calculated to indicate uniformity of the CMS particles using equation (4.3) [48].

4.6.2.2.1 Particle size distribution, geometric mean and standard deviation

1. Particle size distributions were shown in Log-normal distributions.

2. Geometric mean particle size was

$$d_g = \exp[\frac{\sum n_i \ln d_i}{\sum n_i}] \quad (4.1)$$

where, d_g = geometric mean particle size [μm]

d_i = midpoint size for a size interval [μm]

n_i = mass fraction [-]

$$\sigma_g = \exp[\frac{\sum n_i (\ln d_g - \ln d_i)}{N-1}] \quad (4.2)$$

where, σ_g = geometric standard deviation [-]

d_g = geometric mean particle size [μm]

d_i = midpoint size for a size interval [μm]

n_i = mass fraction [-]

N = total mass fraction [-]

4.6.2.2.2 Uniformity of CMS particles was indicated by geometric coefficient of variance.

The geometric coefficient of variance (CV_g) is a dimensionless.

$$CV_g = [\exp(\sigma_g{}^2) - 1]^{1/2} \quad (4.3)$$

where, CV_g = geometric coefficient of variance [-]

σ_g = geometric standard deviation [-]

4.6.2.3 SEM and EDX analysis

Scanning electron microscope (SEM) was used to visually observe CMS particles by placing them onto conductive carbon tape. Morphologies and shapes of the CMS particles, therefore, were characterized by Scanning Electron Microscope (*SEM, JEOL: JSM-5410LV, Japan*) as shown in Figure 4.11.

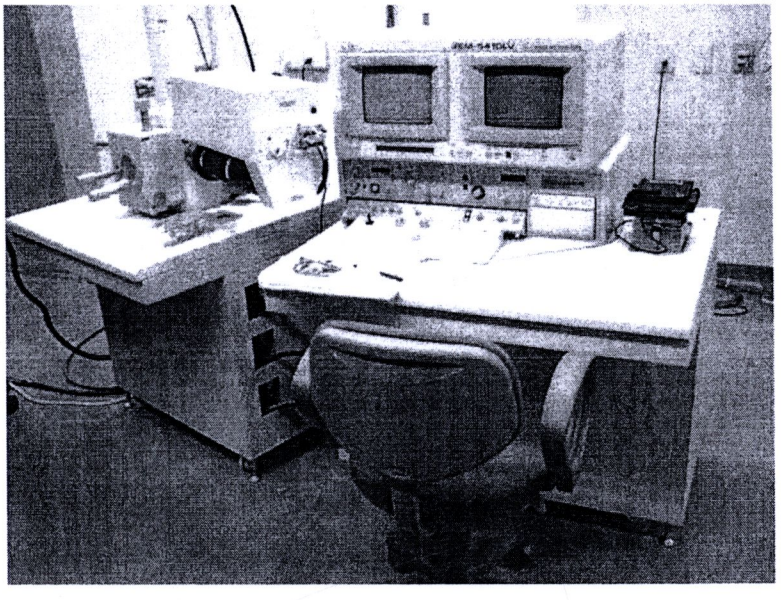

Figure 4.11 Scanning electron microscope (*SEM, JEOL: JSM-5410LV, Japan*)

Porous CMS particles were analyzed by energy-dispersive X-ray spectra (EDX) to show elemental component in the porous CMS particles after carbonization process. The elemental analysis by EDS is achieved by monitoring and analyzing X-rays emitted by matter when bombarded with charged particles. Because of its unique atomic structure, each element radiates x-rays at characteristic wavelengths which allow its identification. The JEOL JSM 5410 SEM is equipped with an Oxford Link Isis - Energy Dispersive X-ray Spectrometer (EDS), which serves as an elemental analyzer. The solid state Si(Li) detector, permits the detection of X-rays and identification of the elements responsible for the emission in a microscopic location.

4.6.2.4 FT-IR analysis

Functional groups on surface of CMS particles were analyzed by FT-IR (Fourier transform-infrared spectroscopy) with an IR Prestige-21 spectrometer (*Shimadzu*) using KBr pellets as shown in Figure 4.12. The scanning was commenced at wavenumbers ranging from 4000 to 450 cm^{-1} at 4.0 cm^{-1} resolution and number of scan 16.

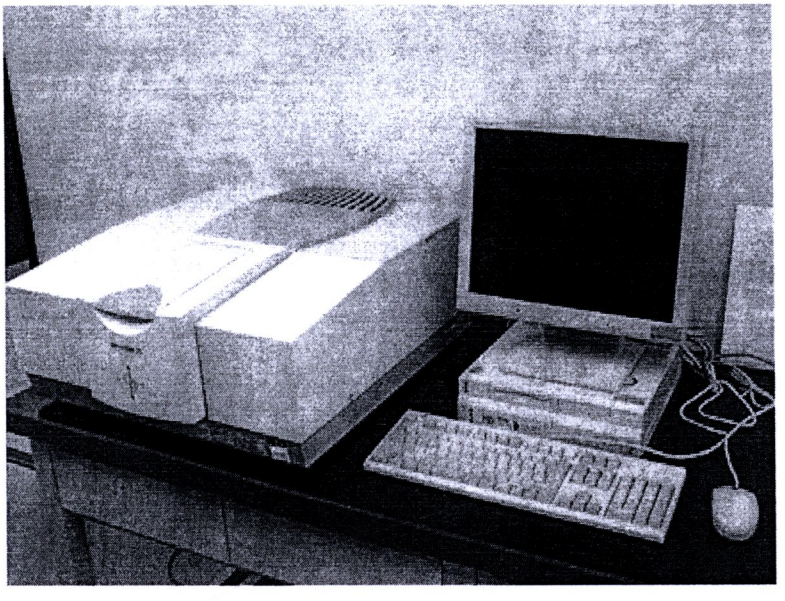

Figure 4.12 Fourier transform-infrared spectroscopy (*Shimadzu*)

4.6.2.5 Elemental analysis

Elemental compositions of carbon microspheres were analyzed by CHNS/O analyzer (*Perkin Elmer PE2400 Series II*) as shown in Figure 4.13. In this analysis, gaseous products freed by pyrolysis in high-purity oxygen and were chromatographically separated by frontal analysis with quantitatively detected by thermal conductivity detector. In this analysis, nitrogen and sulfur in products were neglected because of small quantity. This work used this value for the calculation for carbon balance.

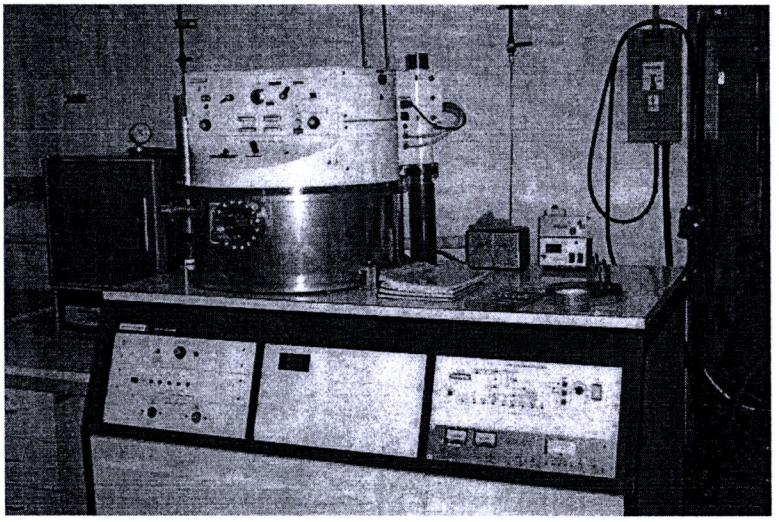

Figure 4.13 CHNS/O analyzer (*Perkin Elmer PE2400 Series II*)

4.6.2.6 Porosity analysis

Porous structure of porous CMS particles after carbonization process was characterized by nitrogen adsorption – desorption at –196°C (*BEL: BELSORP-mini, Japan*) as shown in Figure 4.14. In brief, the porous CMS sample (~0.8g) was pretreated at 150°C under vacuum for 3 hours in order to remove moisture and gaseous residual. BET surface area (S_{BET}) was determined by BET equation. Micropore volume (V_{mic}) was determined by t-plot method.

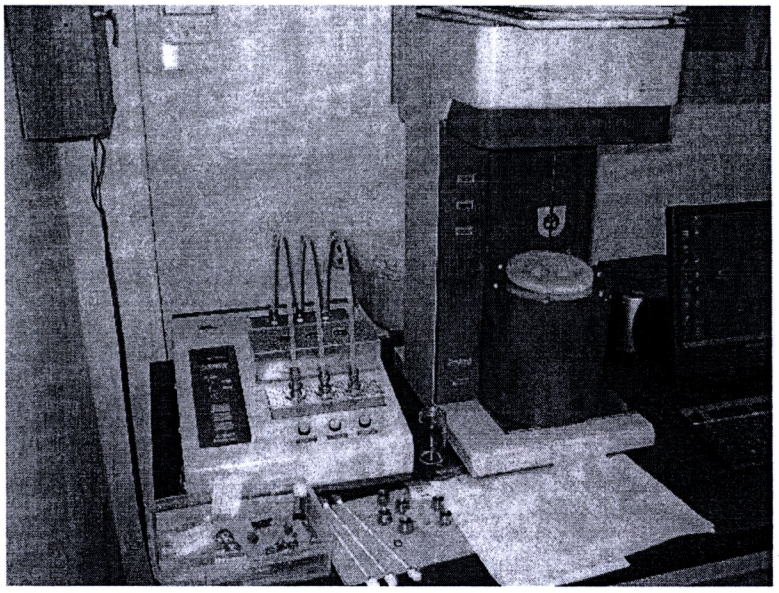

Figure 4.14 N_2 adsorption – desorption analyzer (*BEL: BELSORP-mini, Japan*)

4.6.2.7 Structure and crystallinity analysis

Structure and crystallinity of porous CMS particles after carbonization process was determined by X-ray diffraction analysis. The porous CMS samples were characterized by X-ray powder diffraction (*XRD, SIEMENS D 5000, Japan*) as shown in Figure 4.15 using CuKα radiation with Ni filter in the 2q range of 20-80 degrees resolution 0.04°. The crystallite size was calculated from Scherrer's equation.

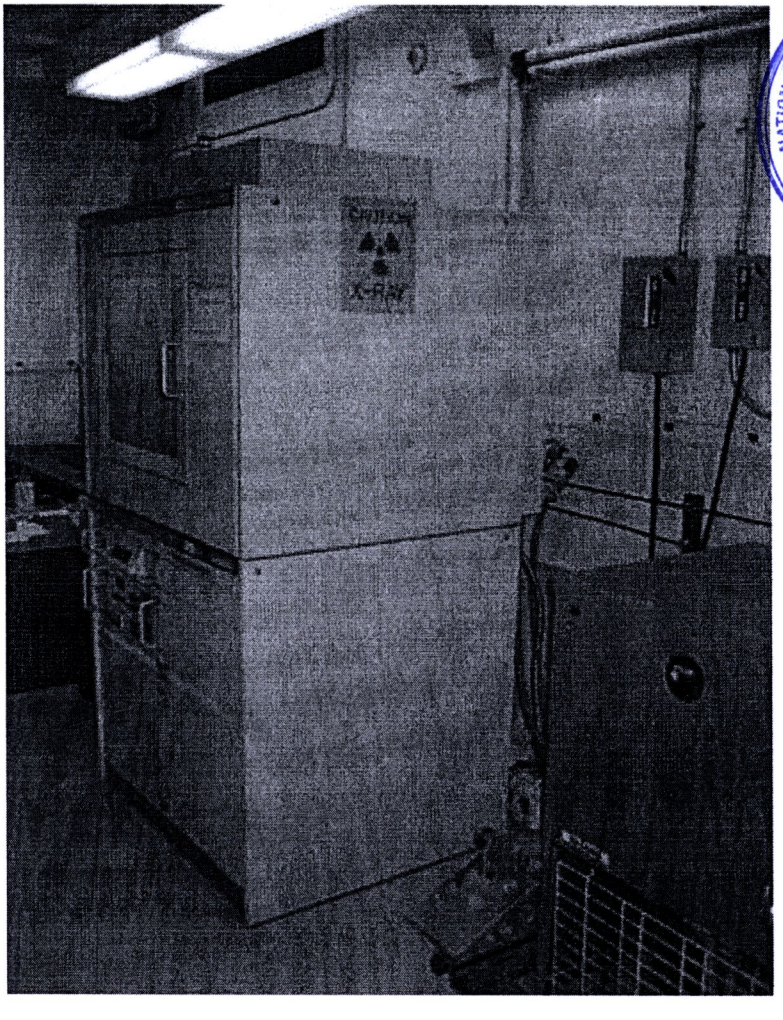

Figure 4.15 X-ray diffraction analyzer (*XRD, SIEMENS D 5000, Japan*)

4.6.2.8 Thermogravimetric analysis

The CMS particles before carbonization process were analyzed by thermogravimetric analysis (TGA) to show their thermal decomposition. The CMS sample was analyzed by pyrolysis under nitrogen gas with heating rate of 1°C/min. Thermal gravimetric analysis (TGA) and differential thermal analysis (DTA) were performed using an SDT Analyzer Model Q600 from TA Instruments, USA as shown in Figure 4.16.

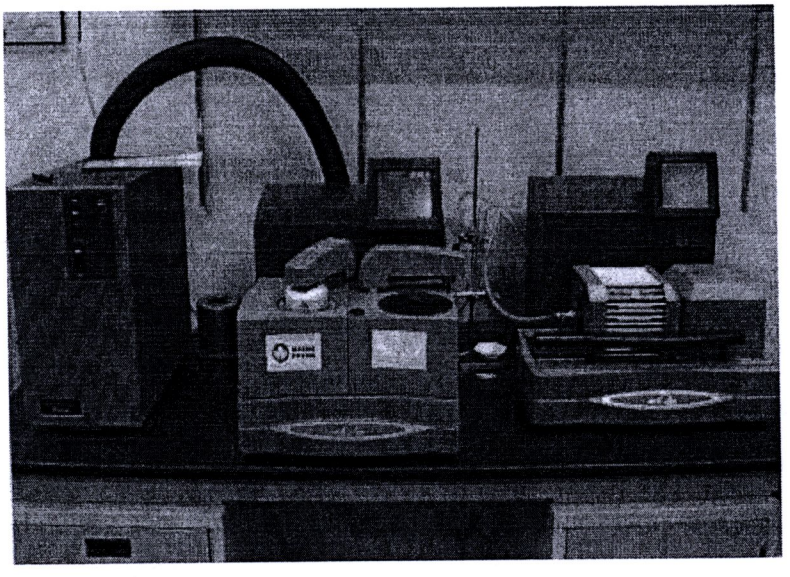

Figure 4.16 SDT analyzer (*Model Q600 from TA Instruments, USA)*