Transactions of the Materials Research Society of Japan 30[4] 1127-1130 (2005)

Mesoporous Recycled Ceramics for Carrier of Bio-enzyme used in the Water Purification

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The preparation of well-ordered mesoporous structures over the porous recycled ceramic materials with pore size 100-200 μm made from waste materials such as waste porcelain, earthen ware, and sludge taken from the bottom of lake, is reported. The recycled porous ceramic material was characterized by XRD, N_2 adsorption analysis, mercury intrusion porosimetry, SEM and TEM. The results clearly showed the formation of mesopores on/in the macroporous ceramics. The immobilization of microorganisms on that porous recycled ceramics is governed by the surface charge and pore size of the ceramics. The treatment of waste water using ceramics with much amount of microorganisms showed good results. The size of mesopores can be precisely controlled by changing the mixing ratio of additives. The pores 100-200 μm in size are suitable for microorganism immobilization, and the mesopores were found to enhance the enzyme encapsulation ability. The enzymes derived from the microorganisms in polluted sludge were effective in reduction of trace harmful chemicals such as an endocrine disrupter, and these encapsulated bio-enzymes in the mesopores over the porous recycled ceramics can have enhanced ability to reduce pollutant and purify polluted water.
Key words : porous materials, water purification, carrier, microorganism, enzyme

1. Introduction

Increment of polluted water and large amount of waste have been a serious problem. Therefore it is important to utilize non-reused local materials for purifying polluted water and recycling the resources. Ceramic materials are useful as recycling materials for preparation of porous carriers for supporting microbial adhesion and growth, as to its hydrophilic characteristics [1]. Therefore, the effects of surface characteristics of porous ceramics on adhesion of microorganisms and biological purification abilities are the subject to be investigated. Design and synthesis of porous ceramic materials, from recyclable waste materials, as carriers of microorganisms are also to be investigated for enhancement of microbial functions.

Recent much attention was paid to the periodic mesoporous inorganic materials because of their potential use for carrier, membrane and other applications. Inorganic membranes are to be permselective in order to allow only some components of one phase to diffuse into the other, whose transport properties depend on their microstructure, especially the pore size distribution and connectivity [2]. The synthesis of MCM-41-type oriented mesoporous films [3], or the preparation of mesoporous films prepared on dense substrates have been reported [4]. Immobilization and encapsulation of enzymes on inorganic supports have been investigated due to its potential use with biocatalysts and biosensors. Mesoporous materials with uniform pore diameters of 1.5-30 nm have been intensively reported, and the diameters in this range are close to the diameters of enzyme molecules. Uniformity of the pore size of mesoporous materials allows control of molecular adsorption based on size. Enclosure of the enzyme in a well-defined space may help to prevent denaturation. Moreover, the pore size of mesoporous materials can be changed precisely, and enzymes with various molecular volume may be applied.

The oriented materials such as MCM-41,which exhibits a hexagonal honeycomb porous framework, appear to be hardly suitable for membrane preparation because the porous network tends to align parallel to the support [5]. These coatings also limit the applications for enzyme encapsulation. However, compounds with a 3D porosity such as MCM-48 silica [6], or MSU-*X* silica prepared with nonionic poly(ethylene oxide)-based (PEO) surfactants [7], would be suitable for this purpose.

The aim of this research is to develop in situ water treatment systems in rivers and lakes using recycled local materials to reduce pollution of the water supply. Microorganisms including aerobes and anaerobes are practically used in the purification of water supplies. Also mesoporous and 3-D mesoporous silica, with pore size > 5nm were coated on the porous recycled ceramic material. The mesoporous coatings on the ceramic surface is expected to enhance their enzyme encapsulation ability. These mesoporous coated ceramics with enzyme encapsulated in the mesoporous are suitable for application in purification of water reservoirs.

2. Experimental

The porous ceramic material was prepared from waste materials such as waste porcelain, earthenware, sludge taken from the bottom of lake, and lime stone. They were broken into small species or pulverized, and dried. The porous ceramic materials were prepared by heating a mixture of waste ceramics and additives, such as burnt waste ash from incineration plants, waste materials from food processing industry, at 750-1100°C. The pore size of obtained ceramics was measured by mercury intrusion procimetry. The surface potential of ceramics was estimated by zeta potential meter.

The mesoporous materials were coated on the porous recycled ceramic material. The coating was conducted using triblock copolymer or sol-gel solution. The triblock copolymer, $EO_{20}PO_{70}EO_{20}$ (Pluronic 123, Aldrich) dissolved in HCl solution and tetraethylorthosilicate (TEOS) was mixed to solution, in which the porous recycled ceramic materials were immersed. They were hydrothermally treated at 100°C after incubation at 40°C. The obtained materials were dried and calcined at 500-600°C in air. The sol-gel solution was prepared as follows. Tetraethyl ammonium hydroxide was added to a mixture of TEOS, triethanolamine and deionized water. The porous recycled ceramic materials were dip-coated in the solution, aged at room temperature for a certain period, and dried. The dried materials were heat-treated at 600°C in air.

Artificial waste water was prepared by mixing glucose, polypeptide, corn steep liquor, $N_{24}Cl$, $KHPO_4$, $Na_2HPO_4.2H_2O$, $MgSO_4.7H_2O$ and water at pH value of 6-7. The ceramic carriers, and artificial waste water was sterilized in autoclave at 120°C for 20 min. 25 g of ceramic carrier was put into 100 mL test tube, to which the artificial waste water containing microorganism was added. The microorganisms were immobilized in the porous ceramics by immersing the ceramics into a seed culture broth. Then, some fresh waste water was supplied for assessing the performance of water treatment. Microorganisms were cultured by feeding fresh waste water every 6 hours, and the overflow was taken, and the concentration of glucose,.protein, and ammonium in treated water was measured using UV spectrometry. Phosphorous concentration was measured by ICP instrument. The immobility of microorganisms on ceramic carrier was evaluated by DNA measurement of microorganisms. Microorganisms onto ceramic carrier was dissolved in 0.1 N NaOH solution containing EDTA-Na, and then homogenized prior to the determination of the DNA quantities using fluorescent spectrometer. The morphologies of the microorganisms onto ceramic carrier were observed by scanning electron microscopy. The ceramic carrier on which microorganisms immobilized were immersed in 0.5% glutaraldehyde solution and dried by supercritical point drying method. All micro-FTIR spectra were recorded using samples encased in a transparent KBr matrix.

The immobilization of enzymes on mesoporous recycled ceramics was evaluated. The enzymes were dissolved in phosphate buffer solution, and undissolved materials were removed by centrifugation of the solution. The mesoporous recycled ceramics was immersed in enzyme solution overnight at 4°C. The supernatant was separated by centrifugation, and the catalytic activity of resultant material was measured.

3. Results and Discussion

3.1 Porous recycled ceramic carrier for microorganism

Fig.1 shows the microorganisms on the porous recycled ceramics made from waste porcelain. Some microorganisms were there on the surface of materials for 1 day cultivation, where macro pores of several dozens micrometers in size were clearly observed. After cultivation of microorganisms for a week, although many rod like microorganisms ranged at their ends, and the bundles were piled up on the surface of porous ceramics, they completely covered over the porous recycled ceramics and the surface of materials was

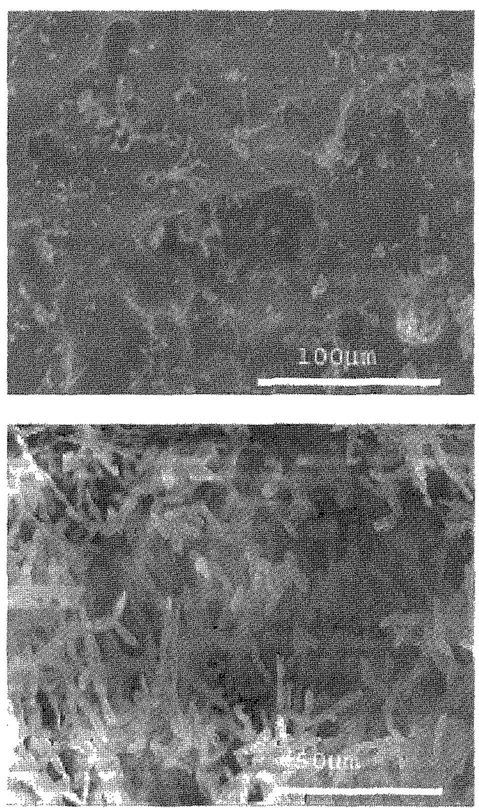

Fig.1. SEM photos of microorganisms immobilized on porous recycled ceramics prepared from waste porcelain, cultivated for 1 day (above) and 1 week (below).

scarcely seen. Therefore porous ceramics prepared from waste materials should have suitable surfaces for the immobilization of microorganisms.

DNA measurement clearly showed that the amount of microorganisms onto ceramic carriers increased as a function of culture time as shown in Fig.2. With an increase of culture time, the DNA quantity of microorganisms on the porous recycled ceramics carriers increased. The glucose concentration in treated artificial waste water decreased as a function of culture time. Consequently, the treatment ratio of glucose reached to be 90% after 130 hours of culture time. While the protein concentration in treated water jumped sharply after 130 hours of culture time, the activities of microorganisms may decline due to lack of nutrition in water. In this system, the microorganisms were cultured by feeding fresh waste water every 6 hours, while the treatment ratio of glucose, protein, ammonium seemed to be saturated by feeding every 12 hours.

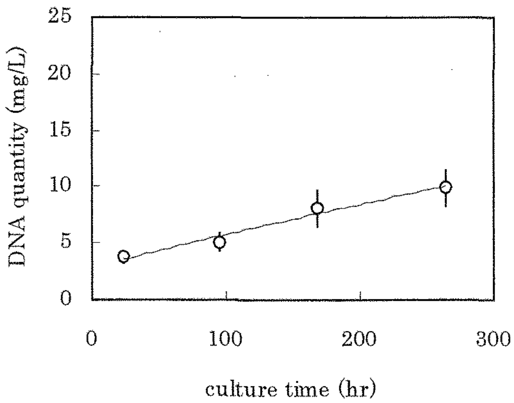

Fig 2. DNA quantities of microorganisms immobilized onto ceramic carrier prepared by heating a mixture of clay and silica, and limestone at 1050°C.

The amount of micro-organisms changed depending on the materials. After treatment of artificial waste water for a week, the purification performance using earthenware was better than that using porcelain. The higher purification performance was achieved by using the ceramics, whose surface potentials were negatively charged, which were effective for adhesion of microorganisms to the ceramics. The surface potential of ceramic carriers prepared from porcelain or earthenware was – 18 mV, -34 mV respectively, at the pH value of 7. Surface zeta potential was found to be a factor governing adhesion of microorganism.

The ceramic carrier prepared from earthenware and burnt waste ash from incineration plants was –24 mV. The treatment ratio of glucose or phosphorus content in water using this ceramics showed good results, due to many macro pores and large surface area of this ceramics. On the ceramics with pores over several dozens micrometers in size, much amount of microorganisms adsorbed onto the ceramics. These size pores might be effective as support for microorganisms over 10 micrometers in size as shown in Fig.1. The immobilization of microorganisms on that porous recycled ceramics is governed by the surface charge and pore size of the ceramics.

3.2 Mesoporous recycled ceramic carrier for enzyme

Fig.3 shows the SEM image of porous recycled ceramics prepared from the sludge taken from the bottom of lake. Many macro pores in sizes of dozens micrometers can be also observed, also were there numerous small pores in the size of several micrometers or below in the Fig. 3 (above). When this porous recycled ceramics was coated with triblock copolymer type mesoporous precursor gel, the parent ceramics is completely coated with mesoporous layer as observed Fig. 3 (below). The small pores in the parent ceramics could not be seen after coating with triblock copolymer type mesoporous materials. The roughness of the parent ceramics, although can be observed, indicating complete coverage of parent ceramic surface along their roughness and macro pores.

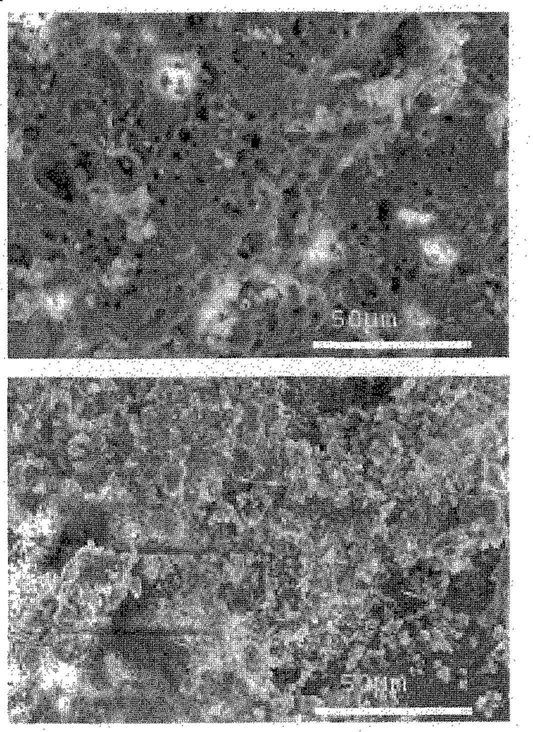

Fig.3. SEM photos of the porous recycled ceramics (above) prepared by heating the sludge taken from the bottom of lake over 1000 °C, and mesoporous coatings on the porous recycled ceramics using triblock copolymer (below).

From the TEM image of coating layer on the parent porous recycled ceramics, the clear formation of a mesoporous layer over the parent ceramic materials was seen. The formed mesoporous layer has a disordered arrangement.

The pore size distribution curve for the mesoporous coated recycled ceramics shows the average pore size of mesoporous layer on the porous recycled ceramics as shown in Fig. 4. From the results of powder X-ray diffractions, the wall thickness of the mesoporous layer is estimated to be around 1-2 nm. The hysteresis loop of the N_2 adsorption isotherm is attributed to the presence of mesoporous structure.

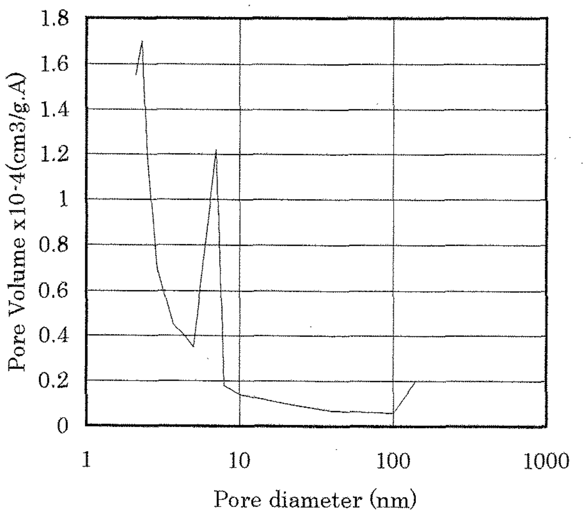

Fig.4. Pore size distribution curve for mesoporous material coated recycled ceramic materials. The parent porous recycled ceramics was prepared by heating the sludge taken from the bottom of lake over 1000°C, and mesoporous coatings on the porous recycled ceramics using triblock copolymer

The coating layer on the porous recycled ceramics is the periodic mesoporous material with uniform pore diameters, and diameters in this range are close to the diameter of enzyme molecules. Results of lipase adsorption on the mesoporous materials, lipase immobilized in mesoporous materials is tightly adsorbed in the mesopore space and is not present on the outer surface of the mesoporous materials [8]. The results of catalytic evaluation of immobilized enzymes on mesoporous coated-ceramics indicate that such mesoporous-coated ceramics can be very effective for preparation of recyclable enzyme catalysts. These encapsulated enzymes in the mesoporous coating over the ceramics can be expected to have enhanced ability to purify waste water, and to be of potential in construction of the water purification system in minimum size and energy.

4. Conclusion

The immobilization of microorganisms on that porous recycled ceramics is governed by the surface charge and pore size of the ceramics. The treatment of wastewater using ceramics with much amount of microorganisms showed good results. This study has demonstrated that porous recycled ceramic materials prepared from waste materials such as porcelain, earthenware, sludge have been found to be useful as carrier of microorganism used in the purification of water.

In conclusion we have been able to prepare a uniform coating of about 10 nm pore size mesoporous layer on the surface of macro porous ceramic materials. These large mesopores are especially suitable for encapsulation of enzymes.

References

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(Received December 23, 2004; Accepted September 15, 2005)