Preparation of diopside with apatite-forming ability by sol–gel process using metal alkoxide and metal salts

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Abstract

The thermal and biological properties of diopside, $CaMgSi_2O_6$ prepared by a sol–gel process using a metal alkoxide and metal salts were investigated for its applicability as a biomaterial. Precursor wet gel was synthesized by hydrolyzing a homogeneous solution consisting of $Ca(NO_3)_2 \cdot 4H_2O$, $MgCl_2 \cdot 6H_2O$ and $Si(OC_2H_5)_4$ dissolved in ethanol. The effect of thermal treatment on crystallization of the hydrolyzed product was examined by DTA, XRD and TG–MS measurements. The dried gel powder was X-ray amorphous and crystallized into diopside single phase at 751.4 °C. This crystallization temperature was lower than that for the dried gel powder prepared by a sol–gel process using metal alkoxides reported by Nonami et al., which suggests that acidic compounds such as HNO_3 and HCl generated in the powder during the heating process promotes the crystallization. Moreover, when the sintered body of diopside was immersed in simulated body fluid, an apatite layer was formed on the surface. Thus, the diopside prepared by the sol–gel process using the metal alkoxide and the metal salts without acidic catalysts addition was found to have an apatite-forming ability.
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Keywords: Diopside; Sol–gel process; Metal alkoxide; Metal salts; Apatite-forming ability

1. Introduction

Diopside ($CaMgSi_2O_6$), one of the pyroxene minerals, is known as an excellent bioactive material and has been the subject of many studies [1–8]. According to these studies, when diopside is immersed in simulated body fluid (SBF) apatite-like calcium phosphates are formed on its surface, giving a good bioactivity [9,10]. In addition, the sintered body of diopside seems to bond to living bone tissues more rapidly than apatite [11,12]. Moreover, diopside has a fairly high mechanical strength and superb biological affinity [13–15]. Diopside is therefore considered to have a potential as a biomaterial for artificial bone and tooth.

There have been several reports on ceramic powder preparation from solution [16–18]. These reports describe that sol–gel process is effective in obtaining fine particles with a high sinterability. The process is also possible to achieve multi-component products with various shapes and forms such as dense or porous bulk-solids, fibers and films [19–23]. In addition, sol–gel process allows us to prepare highly homogeneous complex amorphous and crystalline products at comparatively low temperatures.

In sol–gel process, metal alkoxides ($M(OR)_n$, where M and R represent metal and alkyl groups, respectively) are usually employed as the raw materials. Since the rate of hydrolysis of metal alkoxides considerably differs from one species to another, it is often difficult to establish synthetic processes for reproducibly preparing expected products [24]. In addition, almost all metal alkoxides are quite unstable in the air and cannot be handled easily in the preparing process.

We expect that the synthesis of diopside by sol–gel process using not only metal alkoxides but also metal salts as the starting materials is useful to solve the problems mentioned above. In the present study, we prepared diopside by a sol–gel process using a metal alkoxide and metal salts without acidic catalysts addition and examined the effect of thermal treatment on crystallization of the dried gel powder. Furthermore, the bioactivity of the sintered body of diopside was also evaluated by means of the immersion of the diopside in SBF.

2. Experimental

2.1. Diopside preparation

At first, 0.125 mol of $Ca(NO_3)_2 \cdot 4H_2O$ and 0.125 mol of $MgCl_2 \cdot 6H_2O$ were dissolved in 150 ml of ethanol as a solvent. The starting solution was vigorously stirred with a hot-stirrer at 80 °C for 30 min. After that, 0.250 mol of $Si(OC_2H_5)_4$ was added to the solution. The homogeneous solution was slowly stirred for a few hours to yield a precursor wet gel. The wet gel was dried in an oven at 100 °C for 24 h. The dried gel powder ground with an agate mortar and pestle was calcined at 700 °C for 2 h in the air. The calcined powder was ground again and palletized at 50 MPa by a uniaxial pressing. The pellet was sintered at 1100 °C for 2 h in the air and cooled to room temperature at a rate of 10 °C/min in an electrical furnace.

2.2. Characterization of dried gel powder

The crystallization temperature was determined by differential thermal analysis (DTA, TG-8120, Rigaku Co., Tokyo, Japan). The dried gel powder, 40 mg, was placed into a platinum crucible and heated in the temperature range of 20–1100 °C at a rate of 10 °C/min in a helium gas flow. The vaporized gases from the powder during the heating process were simultaneously analyzed with a GCM spectrometer (GC-MS, GCMS-QP5050A, Shimazu Co., Kyoto, Japan) equipped with a TG–DTA instrument. The crystalline phases of heated powders were identified by powder X-ray diffraction (XRD, RINT-2000, Rigaku Co., Tokyo, Japan) with CuKα radiation in the 2θ range of 20–40°.

2.3. Evaluation of bioactivity

Simulated body fluid (SBF) was prepared based on the compositions of human blood plasma in Table 1 [25–27]. The reagents for preparing SBF given in Table 2 were dissolved in 700 ml of distilled water in a glass volumetric flask. After completely dissolution, the temperature of the solution was adjusted to 36.5 °C with a hot stirrer. The pH value of the solution was also adjusted to 7.4 by dropwise addition of 1 mol/l HCl solution. Finally, distilled water was added to modulate 1000 ml of the fluid.

Table 1
Ion concentrations in SBF and human blood plasma

Ion	Concentration (mM)	
	SBF	Blood plasma
Na^+	142.0	142.0
K^+	5.0	5.0
Mg^{2+}	1.5	1.5
Ca^{2+}	2.5	2.5
Cl^-	147.8	103.0
HCO_3^-	4.2	27.0
HPO_4^{2-}	1.0	1.0
SO_4^{2-}	0.5	0.5

Table 2
Reagents for preparing SBF

Order	Reagent	Amount
1	NaCl	8.0362 g
2	$NaHCO_3$	0.3518 g
3	KCl	0.2251 g
4	K_2HPO_4	0.1758 g
5	$MgCl_2 \cdot 6H_2O$	0.3112 g
6	1 mol/l HCl	40.0 ml
7	$CaCl_2$	0.2926 g
8	Na_2SO_4	0.0717 g
9	$(CH_2OH)_3CNH_2$	6.1182 g

A sintered diopside pellet with dimensions of $10 \times 10 \times 1.5$ mm³ was used for the evaluation of bioactivity. At least three pellets for each soaking time were immersed in 15 ml of SBF kept at 36.5 °C for the prescribed period. After being immersed in SBF, the pellets were removed from SBF, rinsed gently with distilled water for 5 min, and dried at room temperature.

2.4. Analysis of surface structure

The surface microstructure of the sintered diopside pellets before and after immersion in SBF for 3 and 7 days was analyzed by scanning electron microscopy (SEM; JSM-5300, JEOL Ltd., Tokyo, Japan) and the conventional XRD. Calcium and phosphorous element distribution maps of a polished cross-section of the pellet after immersion in SBF for 7 days were obtained with an electron probe microanalyzer (EPMA, JXA-8600, JEOL Ltd., Tokyo, Japan).

2.5. Element concentration analysis of SBF

Changes in the concentrations of calcium, magnesium, silicon and phosphorous of SBF produced by the immersion of sintered diopside pellets were measured by inductively coupled plasma atomic emission spectroscopy (ICP; SPS7000A, Seiko Instruments Inc., Chiba, Japan). For the ICP analysis, SBF was diluted 20 times with distilled water.

3. Results and discussion

3.1. Thermal properties of dried gel powder

In order to design ceramic materials with desired properties, a through understanding of the thermal behavior of raw powder during heating process is essential. Fig. 1 shows the TG–DTA curves of a gel powder dried at 100 °C for 24 h. The exothermic peak observed at 751.4 °C is presumed to be caused by crystallization of the dried gel powder into diopside. Nonami et al. synthesized diopside by a sol–gel process using metal alkoxides, i.e., $Ca(OC_2H_5)_2$, $Mg(OC_2H_4OC_2H_5)_2$ and $Si(OC_2H_5)_4$ (TEOS), which they called the alkoxide method, and reported that the crystallization temperature of the dried gel powder prepared by the alkoxide method was 840 °C [28]. This crystallization temperature reported by Nonami et al. is then considerably higher than that of the dried gel powder prepared by the sol–gel process using a metal alkoxide and metal salts in the present study. Here, the total mass loss of the dried gel powder was 52.4%. These findings indicate that the dried gel powder produced using the combined use of the metal alkoxide and the metal salts crystallizes at a temperature lower than that produced by the alkoxide method using only metal alkoxides, and that Ca and Mg salts could be handled more easily than the alkoxides at the production of precursor wet gel.

The identification of the crystalline phases formed in the thermal treatment was performed by the conventional X-ray diffraction (XRD). Fig. 2 shows the powder XRD patterns of the gel powder heated at various temperatures. The diffracted beam intensities, peak shapes and the half widths of stronger peaks around 30° suggest that the gel powder crystallized into diopside single phase at 1100 °C. Meanwhile, some of weak peaks due to impurities, akermanite ($Ca_2MgSi_2O_7$), monticellite ($CaMgSiO_4$) and other unidentified products, were observed at 700 °C, but they disappeared completely at 1100 °C. In addition, the shapes and the half widths of the diffracted peaks of diopside in Fig. 2 were greater than those of the diopside prepared by the alkoxide method reported by Nonami et al. at the same temperature [28]. This finding indicates that the resultant powder prepared by the sol–gel
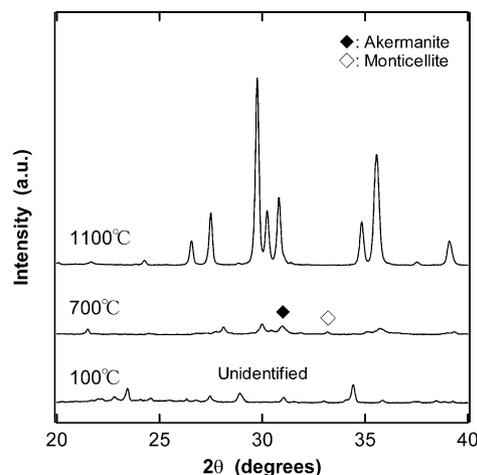
Fig. 2. Powder X-ray diffraction patterns of gel powder heated at various temperatures.

process using the metal alkoxide and the metal salts has a much higher crystallinity.

Fig. 3 shows the TG–MS curves of the gel powder dried at 100 °C for 24 h. In contrast to the TG curve of the dried gel powder produced by the alkoxide method reported by Nonami et al. [28], a drastic and gradual weight decrease was observed with the elimination of various kinds of gases from the dried gel powder. A large release of CO_2 after evaporation of H_2O suggests, moreover, that elimination and combustion of alkyl groups occur after the condensation polymerization reaction of TEOS. On the other hand, it was also found that release of NO_2^+ started at 120 °C and ceased at 610 °C. Further, the mass reduction was almost complete at 650 °C, and a slight weight loss was detected above this temperature. The result suggests, therefore, that glass networks formation was complete at about 650 °C.

A sequence of compounds released from the dried gel powder during the heating process would be related to the crystallization behavior of diopside. Acidic compounds, like hydrochloric acid, nitric acid, acetic acid and so on, are sometimes employed as acidic catalysts to promote gelation in sol–gel processes using only metal alkoxides as the starting materials. In fact, Fig. 3 illustrates that NO_2^+,
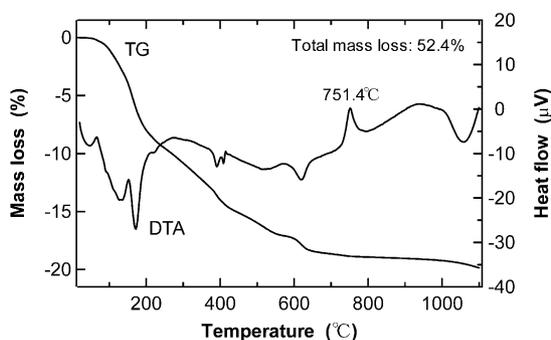
Fig. 1. TG–DTA curves of gel powder dried at 100 °C for 24 h.
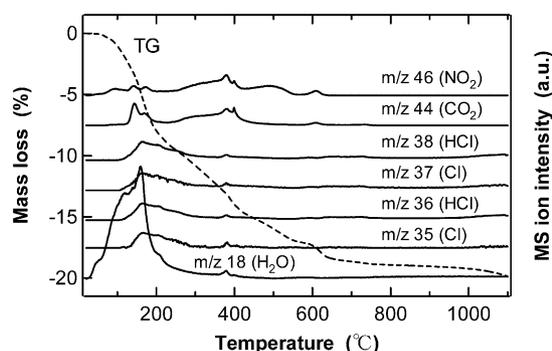
Fig. 3. TG–MS curves of gel powder dried at 100 °C for 24 h.

HCl and Cl^- were detected in the temperature range from 100–650 °C. The release of these inorganic gases from the powder was caused by decomposition of acidic compounds such as HNO_3 and HCl derived from the metal salts, $Ca(NO_3)_2$ and $MgCl_2$, during the heating process in the low temperature range. Hence, HNO_3 and HCl generated during the formation of glass networks promote the crystallization of the dried gel powder into diopside, which results in its lower crystallization temperature, as shown in Fig. 1.
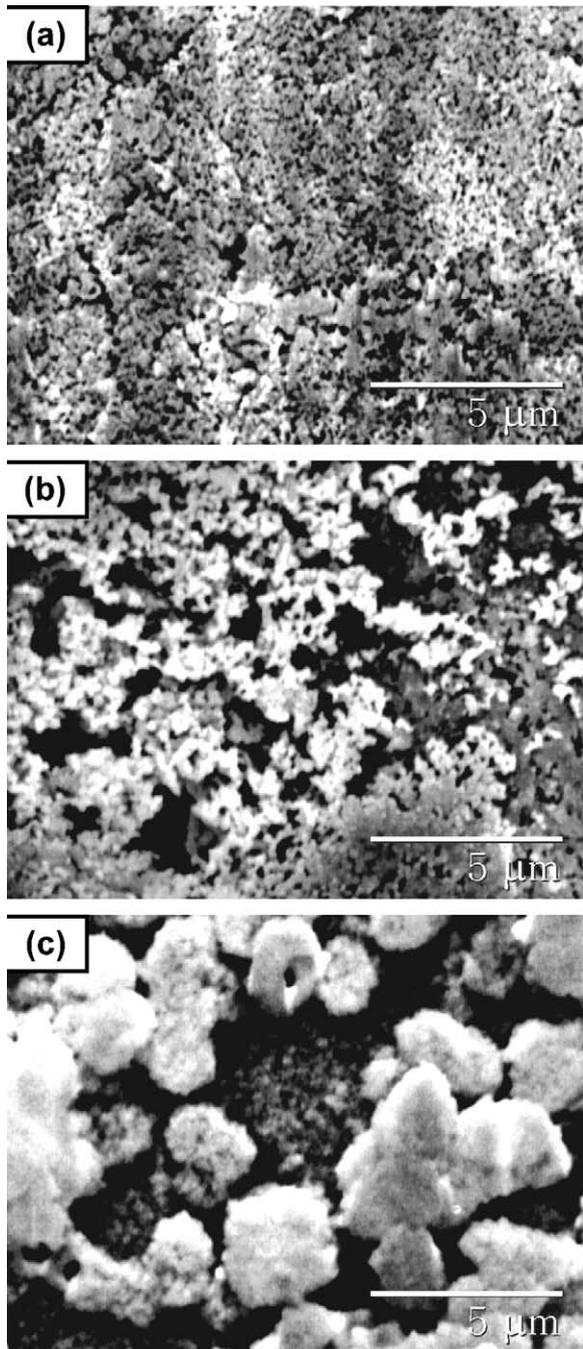
Fig. 4. SEM microphotographs of the outer surface of diopside pellets sintered at 1100 °C for 2 h before (a) and after their immersion in SBF for 3 days (b) and 7 days (c), respectively.

3.2. Formation of apatite layer on diopside surface

On the basis of the findings described above, we can easily prepare diopside by the sol–gel process using both the metal alkoxide and the metal salts at low temperatures without acidic catalysts addition. Here, if the bioactivity of the diopside can be verified, the diopside prepared by this method can be applied to a wide range of the biomedical field. It has been well known that the bonding of bioactive glasses and glass-ceramics to living bone is achieved through a bonelike apatite layer formed on the surfaces in the body environment [29–32]. It is, therefore, believed that the formation of apatite layer on the surfaces of glasses and glass-ceramics is a necessary condition to exhibit their bioactivity. The bioactivity of the sintered body of diopside was then evaluated by its immersion in simulated body fluid (SBF).

Fig. 4(a–c) shows the SEM microphotographs of the outer surface of the diopside pellets sintered at 1100 °C for 2 h before (a) and after their immersion in SBF for 3 days (b) and 7 days (c), respectively. In the morphology before soaking (Fig. 4(a)), micropores and cracks were observed on the surface of sintered pellet. After 3 days soaking (Fig. 4(b)), fine particles found on the surface. Later, the particles grew to massive particles after 7 days soaking (Fig. 4(c)). According to many previous papers, the morphology of particles on the surface of bioactive glasses and glass-ceramics formed in contact with SBF are likely to be a leaflike shape [33–35]. As is clearly shown in Fig. 4, a different morphological feature was observed on the surface of the sintered body of diopside prepared in this study.

Fig. 5 shows the XRD patterns of the outer surface of the diopside pellets sintered at 1100 °C for 2 h before and after their immersion in SBF for each soaking time. After 3 days soaking, the diffraction peaks assigned to apatite at around 26 and 32° increased with immersion time. Here, it is well known that the living bone is a microcomposite, in which
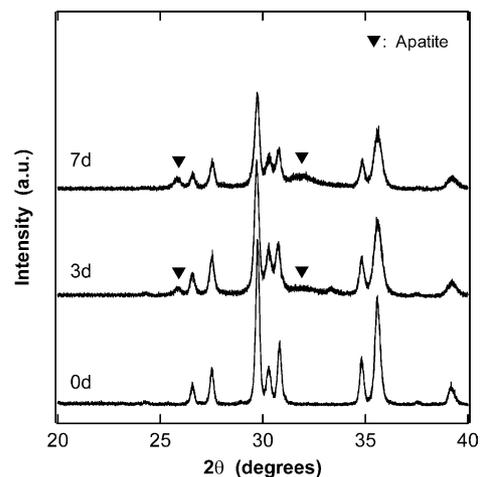
Fig. 5. X-ray diffraction patterns of the outer surface of diopside pellets sintered at 1100 °C for 2 h before and after their immersion in SBF for each soaking time.
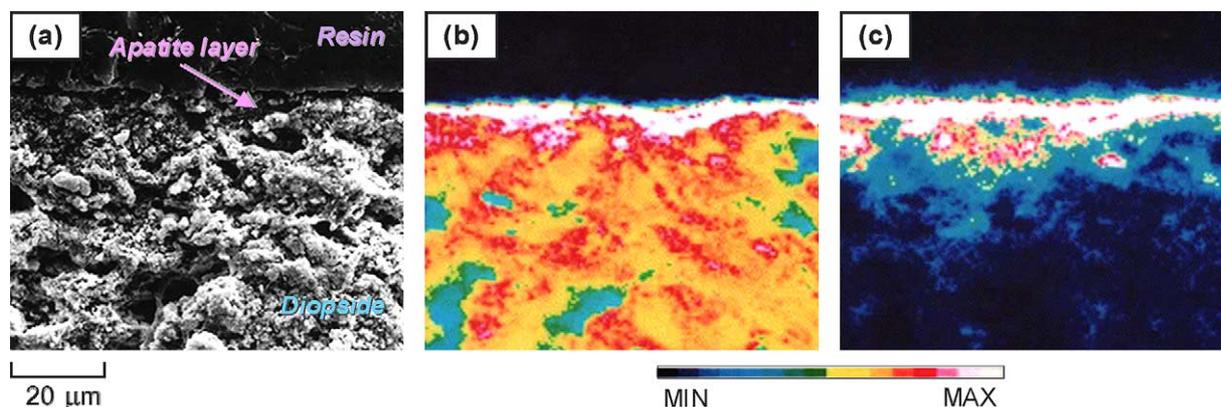
Fig. 6. EPMA images of the polished cross-section of diopside pellet sintered at 1100 °C for 2 h after its immersion in SBF for 7 days: SEM photograph (a), Ca (b) and P (c) element distribution maps.

an assembly of inorganic apatite particles is effectively re-inforced by organic collagen fibers [36,37]. Also, note here that the X-ray profiles of the apatite layer formed on the surface of the sintered body of diopside were similar to those of inorganic components in the living bone.

Fig. 6 shows the EPMA images of a polished cross-section of the diopside pellet sintered at 1100 °C for 2 h after its immersion in SBF for 7 days: SEM photograph (a), Ca (b) and P (c) element distribution maps. A thin layer rich in Ca and P was present on the surface of the sintered pellet, and was about 5 μm thick. The layer is believed to correspond to the apatite layer confirmed by the X-ray profiles in Fig. 5. These results of XRD patterns and EPMA images, therefore, indicate that a bonelike apatite layer was formed on the surface in contact with SBF and that the sintered body of diopside possesses an apatite-forming ability.

Fig. 7 shows changes in the element concentrations of SBF as a function of soaking time of diopside pellets sintered at 1100 °C for 2 h. The drastic decrease in P(V) ions in SBF is attributed to the apatite formation confirmed by X-ray analysis in Figs. 5 and 6. On the other hand, the Si content rapidly increased at the first stage and leveled off stabilized after 3 days soaking. Si(IV) ions were considered to be released from the sintered pellet because as-prepared SBF contained no Si(IV) ions. The Mg content slightly increased and remained almost unchanged with increasing soaking time, which suggests that a portion of Mg(II) ions also dissolved, but the most portions remained in the sintered pellet. It should be noted that a rapid increase in Ca(II) ions was detected at the early stage of soaking. This is because Ca(II) ions were released from the sintered pellet. The Ca(II) ions released may increase the degree of their supersaturation for apatite formation in the surrounding fluid already supersaturated with Ca(II) ions even before soaking and apatite nuclei formation easier on the surface of diopside.
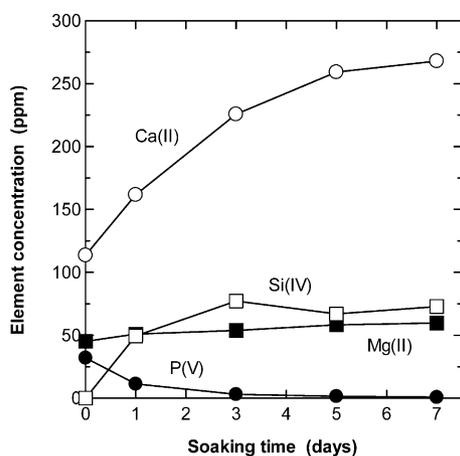
4. Conclusions

Diopside was prepared by a sol–gel process using a metal alkoxide and metal salts as the starting materials, and the effect of thermal treatment on crystallization of the dried gel powder and the bioactivity of the sintered body of diopside were examined by means of the immersion of the diopside in simulated body fluid (SBF). Referring to DTA and XRD measurements, the dried gel powder prepared by this method was suggested to crystallize into diopside single phase at 751.4 °C. The resultant powder possessed a higher crystallinity than that produced by the alkoxide method. Acidic compounds such as HNO_3 and HCl generated in the powder during the heating process promoted the crystallization in thermal treatment. In the evaluation of the bioactivity, an apatite layer was found to be formed on the surface of diopside in SBF. This result suggested that the sintered body of diopside prepared by the sol–gel process using the metal alkoxide and the metal salts without acidic catalysts addition has an apatite-forming ability and a remarkable bioactivity.

Fig. 7. Changes in element concentrations of SBF as a function of soaking time of diopside pellets sintered at 1100 °C for 2 h.

Acknowledgements

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