THE EFFECT OF TEMPERATURE ON THE PROCESSING AND PROPERTIES OF MACROPOROUS BIOACTIVE GLASS FOAMS

J. R. Jones[1], P. Sepulveda[2], L. L. Hench[1]

[1] Centre for Tissue Regeneration and Repair, Department of Materials, Imperial College of Science, Technology and Medicine, London SW7 2BP, United Kingdom

[2] Instituto de Pesquisas Energéticas e Nucleares, IPEN/CNEN, Divisão de Materiais Cerâmicos, Travessa R, 400 - Cidade Universitária, São Paulo, SP, CEP 05508-900 Brazil

Keywords: foam, scaffold, bioactive glass, temperature, macroporous

Abstract. Resorbable 3D macroporous bioactive scaffolds have been produced for tissue engineering applications by foaming sol-gel-derived bioactive glasses. The foams exhibit a hierarchical structure, with interconnected macro-pores (10-500μm), which provide the potential for tissue ingrowth and mesopores (2-50nm), which enhance bioactivity The temperature at which the foams are produced is one of the many factors affecting the processing and properties of these foams. Foams of the 58S composition (60% SiO_2, 36% CaO and 4% P_2O_5, in mol) were produced at 20°C, 25°C, 29°C and 35°C. The effects of temperature on the foam volume, gelling time and the resulting pore structure and surface texture were investigated.

Introduction

Certain compositions of bioactive glasses containing SiO_2-CaO-P_2O_5 bond to both soft and hard tissue without forming scar tissue. Results of *in vivo* implantation show that these compositions are non-toxic [1]. The bioactivity has been associated with the formation of a crystalline hydroxyapatite surface layer, similar to the structure of the inorganic region of bone, on contact with body fluid [1]. Recently, Xynos *et al.* [2] have shown that bioactive glass dissolution products cause rapid expression of genes that regulate osteogenesis and the production of growth factors. These discoveries have stimulated extensive investigations for using bioactive glass as scaffolds for tissue engineering. An ideal scaffold should combine these properties with a structure consisting of an interconnected network with macropores (greater than 100 μm) to enable tissue ingrowth and nutrient delivery to the centre of the regenerated tissue and mesopores (2nm<pore size< 50 nm) to promote cell adhesion. The scaffold should also resorb at controlled rates to match that of tissue repair and be made from a processing technique that can produce irregular shapes to match that of the defect in the bone of the patient. The foaming of sol-gel derived bioactive glasses provides the potential to make such a scaffold. Specific architectures can be obtained to produce specific pore size ranges and controlled rates of glass dissolution. The steps for making these foams, their characterisation and physical properties are described elsewhere [3]. This paper describes the effect of on the foam volume, gelling time and the resulting pore structure and surface texture.

Experimental Procedure

Foams were prepared using the 58S composition (60mol% SiO_2, 36mol% CaO, 4mol% P_2O_5) sol-gel-derived bioactive glass. Tetraethyl orthosilicate (TEOS) was added to a mixture of distilled water and 2N nitric acid (catalyst), and left for 1 hour to allow hydrolysis to complete. Triethyl phosphate (TEP) was then added and left for 30 minutes. Calcium nitrate was then added and the sol was left for a further hour [4]. Simultaneous hydrolysis and polycondensation reactions occur to begin formation of a silica network. Viscosity of the sol increases as the condensation reaction continues. Aliquots of 50ml of the sol were foamed by vigorous agitation at 20°C, 25°C, 29°C and 35°C, using a thermostatically controlled water bath. An acidic catalyst (0.1 wt% HF) was added to

accelerate the condensation reaction and an anionic surfactant (Teepol) was added to stabilise the bubbles during the early stages of foaming. As viscosity rapidly increased and the gelling point was approached the solution was cast into moulds. The gelation process provides permanent stabilisation for the bubbles. The samples were then aged at 60°C for 72h, dried at 130°C for 48h and thermally stabilised at 600°C for 22h, according to established procedures [4]. The effect of foaming temperature on the foamability of the sol and on the foam properties was investigated. The resulting foams were characterised using scanning electron microscopy (JEOL, JSM T220A), mercury porosimetry (PoreMaster 33, Quantachrome) and nitrogen adsorption (Autosorb AS6, QuantaChrome) to measure macro and mesopore sizes and distributions respectively. B.E.T. analysis was used to determine the specific surface area. The pore diameter distribution was calculated by the BJH method applied to the N_2 desorption curves.

Results and Discussion

The effect of temperature on the foamability of 58S sol-gel derived bioactive glass and the corresponding effect on the final structure of the foams was investigated. Figure 1 shows a graph of gelling time as a function of foaming temperature (T_f), where foaming temperature is defined as the temperature of the water bath in which the sol was foamed. The graph shows that as foaming temperature was increased from 20°C to 35°C, the gelling time decreased from 11 min 10s to 6 min 20s. All gelling times were reproducible with a 5% range. The gelling time is thought to have decreased because the condensation rate increased as a result of the increase in temperature [5]. Foam volume as a function of foaming temperature followed a similar relationship (not shown), decreasing from approximately 180ml at 20°C to 70ml at 35°C. The foam volumes were reproducible within 10ml for all temperatures, except at 20°C, where foam volumes greater than 200ml were produced. For foam volumes above 180ml, the bubbles produced are so large that the polycondensation reaction cannot stabilise them and the foams collapse.

The relationship between gelling time and foaming temperature can be fitted to a first order exponential decay ($r^2 = 0.959$). Hydrolysis is completed soon after mixing, therefore the condensation reaction is the rate-determining step for gelation [6]. The condensation reaction is complex, but if the gelling time (t_{gel}) is the time over which viscosity has increases from approximately $10^{-1}P$ to 10^4P then t_{gel} can be considered as an averaged rate of gelation [6]. Therefore the exponential decay can be fitted to an Arrhenius equation;

$$1/t_{gel} = A\exp(-E^*/RT_f)$$

Where A is an Arrhenius constant, R is the gas constant and E^* is an apparent activation energy.

Therefore, there is an apparent activation energy barrier that has to be overcome for the condensation reaction to complete and for gelling to occur. The activation energy for gelation was calculated from the slope of a plot of $\ln[t_{gel}]$ as a function of $1/T_f$ and was found to be 0.002 eV. As temperature is increased the kinetics of the condensation reaction is increased (more monomers come into contact with each other), reducing the gelling time. Activation energy for gelling is affected by the pH and composition of the sol [6].

Figure 2 shows pore distributions, attained by mercury porosimetry, representative of foams produced using foaming temperatures of 20°C, 25°C, 29°C, and 35°C. All pore distributions were wide, implying that foams produced at each of the four temperatures contained some pores greater than 200µm (limit of the mercury porosimeter). At foaming temperatures of 20°C and 25°C, the pore distributions were approximately normal, with modal pore diameters of 95µm. Above 25°C pore distributions followed a positive skew and the amount of skew increased as temperature increased, with a modal pore diameter of approximately 35µm at 29°C and 22µm at 35°C. The pore distributions are far from homogeneous, and the mode of the pore distribution is of most interest when initially characterising the foams, as the mode represents the pore size that is most frequent in the sample.

From these results it can be seen that as foaming temperature increased, gelling time decreased, which meant that agitation time decreased, and therefore foam volume and macro-pore diameter decreased.

Table 1 summarises the results from nitrogen adsorption characterisation of the foams. All foams yield a type IV of isotherm (not shown), which is indicative of mesoporous materials (2 to 50nm). The hysteresis loops between adsorption and desorption modes are typical for materials with cylindrical pores. Table 1 shows that the foams exhibited mesoporous textures with a modal pore diameter in the range 7-13nm and that a change in temperature had little effect on the mesoporosity of the foams. However, the change in foaming temperature had a marked effect on the specific surface area of the foams, with surface area increasing from 150.7 m^2g^{-1} at 20°C to 192.7 m^2g^{-1} at 25°C to 454.4 m^2g^{-1}. The increase in specific surface with increasing temperature was due to the increase in the modal macropore diameter (figure 2). However, the large increase in surface area from 25°C to 35°C was also due to the large number of cracks present in the foams produced at 35°C.

Table 1. Summary of nitrogen adsorption analysis on 58S scaffolds foamed at different temperatures.

Foaming Temperature/ °C	Specific surface area (BET)/ m^2g^{-1}	C constant (BET)	Modal mesopore diameter/ nm
20	150.7	82.7	8.3
25	192.7	194.4	12.4
35	454.4	217.9	7.8

Scaffolds foamed at 35°C cracked during the drying stage of the thermal treatment of the gel. Due to the lower porosity in the scaffold foamed at 35°C it was more difficult for liquid to escape from the centre of the scaffold, therefore capillary stress increased, causing rupture of the glass.

Figure 3 shows a SEM micrograph of a foam scaffold produced at 25°C. Figure 3 shows that the foam was crack-free and contained macropores in the range 10-600µm, some of which are connected via windows in the side of the pores. Such interconnects were approximately 100µm in diameter, which is the size required for tissue ingrowth and vasularisation [3]. Figure 4 shows a SEM mircograph of a highly cracked scaffold, produced at 35°C, implying that scaffold strength decreases with an increase in foaming temperature.

The nitrogen sorption results combined with mercury porosimetry results indicate that the bioactive foams are 3D hierarchical structures of an interconnected macropore network in a matrix containing mesopores. The temperature at which the foam is produced affects the stability and morphology of this structure. Other factors in the foaming process that affect this structure are the composition and volume of the sol, the pH of the sol (catalyst type and concentration), the surfactant type and concentration and the temperature of the thermal stabilisation process. Such factors are the sucbjest of further investigations.

Conclusions

Sol-gel derived bioactive glasses can be foamed to produce macroporous structures containing interconnected pore networks. The temperature at which the foaming process is carried out has a large effect on the gelling time and foam volume produced from 50ml of sol. Thus, the final structure of the foam is affected. As temperature increased, gelling time and therefore foam volume and final mean macropore diameter decreased. A suitable temperature for foaming 58S gel-glasses was found to be 25°C. At 25°C, gelling times were reproducible to within 20s and a modal pore size (~100µm) with potential for tissue ingrowth and vascularisation was attained. This temperature will

be used in future experiments investigating the many other factors affecting the properties of the bioactive foams.
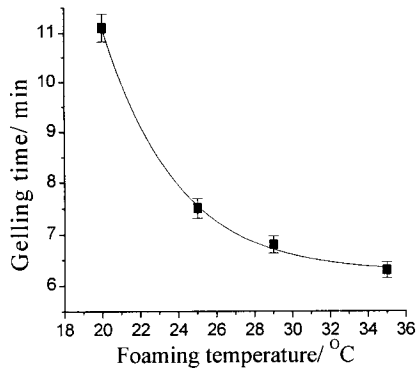
Acknowledgement

Figure 1. Graph of gelling time as a function of foaming temperature.
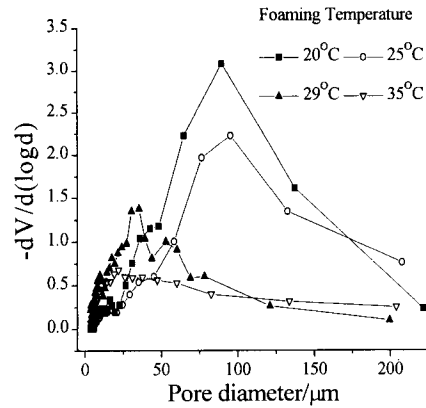

Figure 2. Porosity distributions (macropores) of foams foamed at different temperatures, from mercury intrusion porosimetry.
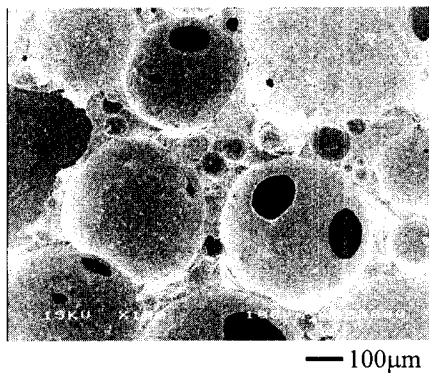

— 100μm

Figure 3. SEM micrograph of the structure of 58S sol-gel derived glass foamed at 25°C.
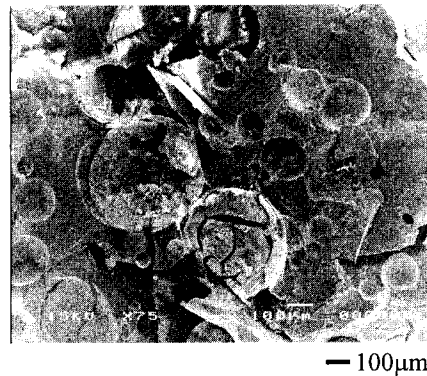

— 100μm

Figure4. SEM micrograph of the structure of 58S sol-gel derived glass foamed at 35°C.

REFERENCES

[1]. Hench, LL. Bioceramics: From concept to clinic. J. Am. Ceram. Soc, 74 [7] 1487-510 (1991)

[2]. Xynos ID, Edgar AJ, Buttery LDK, Hench LL, Polak JM. Biochem. Biophys. Res. Comm. 2000;276(2):461-465.

[3]. Sepulveda P., Jones J. R., Hench, L. L. Synthesis of sol-gel derived bioactive foams, submitted to Bioceramics 14

[4]. Li, R., Clark, A. E., Hench, L. L. J. Appl. Biomater. 2, 231-239 (1991).

[5]. Brinker, C. J. and Scherer, G. W. Sol-gel Science, pp 67-180. Academic Press, New York, 1989.

[6]. Iler, R. K. The chemistry of silica. Wiley. New York 1979 p.378.

The Effect of Temperature on the Processing and Properties of Macroporous Bioactive Glass Foams