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Pleurodesic Effect of Bioglass in Experimental Rabbits

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Abstract

Background: Pleurodesis has been a widely used treatment option for recurrent and persistent pleural effusions and air leaks. However, an ideal pleurodesing agent has not been found yet. The purpose of this study was to investigate the dose dependent effects of bioglass on pleurodesis.

Methods: Fifty six male, New Zealand rabbits weighing 3000-3500 gr were used in this study. After right chest tube insertion, 35, 70, 150, and 400 mg/kg bioglass; and 70 and 400 mg/kg talc in saline solution were administered through the chest tube into the pleural cavity. One ml/kg isotonic saline solution was administered in the control group. After 4 weeks, the rabbits were sacrificed for pleurodesis evaluation.

Results: Bioglass 400 and Talc 400 had a higher pleurodesic effect than the other doses with no statistically significant difference. Local inflammation, fibrosis and particle dissemination were significantly higher in Bioglass 400 and Talc 400 than in the controls. Talc 400 caused more inflammation and more particle accumulation than those by bioglass 400.

Conclusions: Bioglass may be a valuable pleurodesic agent. However, further studies are needed for more definitive results.

Keywords: Pleura; Pleurodesis;Talc; Bioglass; Rabbit

Introduction

Pleurodesis has been a widely used treatment option for recurrent and persistent pleural effusions and air leaks. However, an ideal pleurodesing agent has not been found yet. Several chemical and biological agents have been experienced with some advantages and disadvantages. Bioglass is a silica based bioactive ceramic composed of sodium and calcium salts, phosphates, and silicon dioxide.[1] It is hemostatic,[2] radiopaque,[3] biocompatible and absorbable,[1] and also has antibacterial activity against some bacteria[4] in the bone tissue. It has been widely used experimentally and clinically for filling bone defects and for bone augmentation,[1] in drug delivery systems,[5] as a scaffold in tissue engineering,[6] and for covering the surface of implant materials.[7]

We showed in the previous study that bioglass had pleurodesic effect as well as talc in rabbits.[8] In this study, we purposed to confirm our previous study and to determine the dose dependent pleurodesic effects of bioglass.

Materials and Methods

Fifty six male, New Zealand rabbits weighing 3000-3500 gr were enrolled. The study was approved by local Ethics Commitee, and financially supported by the Scientific Research Board of our university. All the animals received humane care in compliance with the "Guide for the Care and Use of Laboratory Animals," prepared by the Institute of Laboratory Animal Resources.

Bioglass particulate (Perioglass USBiomaterials Corporation, Florida, USA) and talc (Steritalc, Novatech, France) were commercially supplied.

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Perioglass, with particle size ranging from 90 to 700 microns, was pestled in a ceramic pot and sifted with a 100-micron filter (Emperor Aqautics, USA). Thus, bioglass powder was obtained with particle size smaller than 100 microns. Both materials were balanced, packaged and sterilized with dry autoclave before using for pleurodesis.

Rabbits were randomly divided into 7 equal groups (n=8). They were anesthetized with 35 mg/kg of ketamine (Ketasol, Richterpharma, Austria) and 5 mg/kg xylazine (Rompun, Bayer, Germany). The right hemithorax was shaved, and cleaned with povidone iodine solution. After right chest tube insertion, 35, 70, 150, and 400 mg/kg of bioglass; and 70 or 400 mg/kg of talc in saline solution were administered through the chest tube into the pleural cavity in each group. One ml/kg isotonic saline solution was administered in the control group. Chest tubes were immidiately removed after the instillation. A dose of 2 gr/l of paracetamol (Parol, Atabay, Turkey) was administered in their drinking water after pleurodesis to induce analgesia. At the 28th day, the rabbits were sacrificed with a high dose of penthothal (Pental sodyum, I.E. Ulagay, Turkey) intravenously. Local pleurodesic, inflammatory and fibrotic effects, and systemic inflammatory response were evaluated as follows:

For pleurodesis, The thoracic cavity was opened after sacrification. Gross pleurodesis was graded according to the following scheme: 1=no adhesions; 2=rare adhesions with no symphysis; 3=a few scattered adhesions with no symphysis; 4=many adhesions with no symphysis; 5=many adhesions with symphysis involving less than 5% of the thoracic cavity; 6=many adhesions with symphysis involving 5% to 25% of the thoracic cavity; 7=many adhesions with symphysis involving 25% to 50% of the thoracic cavity and 8=many adhesions with symphysis involving more than 50% of the thoracic cavity.[9]

Local inflammation and fibrosis were graded histopathologically as; 1=absent; 2=mild; 3=moderate and 4=severe.

To determine the systemic inflammatory response, the blood samples were received at the 6th and 24th hours of pleurodesis. Lactic dehydrogenase (LDH) and interleukin-8 (IL-8) were measured to evaluate systemic inflammatory response.

To measure particle accumulation, lung parenchyma and bronchoalveolar lavage fluid was received immediately after sacrification. Existance of the particle was graded under polarized light microscope as: 1=absent; 2=mild; 3=moderate and 4=severe.

SPSS 10.0 statistics programme was used for statistical analysis. The data were expressed as mean±standard deviation (SD). Kruskall Wallis and Mann Whitney U tests were performed for nonparametric data. A p value less than 0.05 was described as significant.

Results

Bioglass 400 and Talc 400 had a higher pleurodesic effect than the other doses (p<001), with no significant difference between them (p=0.130). No difference was demontrated among the other groups. Table 1 shows the evaluation of pleurodesis among the groups.

Local inflammation, fibrosis and particle dissemination were significantly higher in bioglass 400 and talc 400 than the control group (p<0.05). Talc 400 caused more inflammation and more particle accumulation than bioglass 400 (p<0.05). Evaluation of local effects are shown in Table 2.

Systemic effects of the pleurodesic agents are shown in Table 3. At the 6th hour of pleurodesis, in talc 400 group, LDH level was significantly higher than that in bioglass 400 (p=0.038). Interleukin-8 level was higher in bioglass 400 and talc 400 than in the control group, but no significant difference was noticed between the bioglass and talc groups.

Table 1: The statistically evaluation of pleurodesis

Parameter (mean±SD)	Control	Bio35	Bio70	Bio150	Talc70	Bio400	Talc400	P value
Pleurodesis	1.0±0.0	1.2±0.4	1.3±0.5	1.5± 0.5	1.5± 0.5	3.5± 0.6	3.8±0.5	<0.001*

Comparison of control group vs Bio400, p<0.001*; control vs talc400, p<0.001*; bio35 vs Bio400, p=0.001*; Bio35 vs Talc400, p=0.003*; Bio70 vs Bio400, p=0.001*; Bio70 vs Talc 400, p=0.007*; Bio150 vs Bio400, p=0.002*; Bio150 vs Talc400, p=0.021*; Talc70 vs Bio400, p=0.002*; Talc70 vs Talc400, p= 0.021*. (*) Statistically significant.

Table 2: Evaluation of local inflammatory, fibrosis effect and particle dissemination of the groups

Local response (mean±SD)	Control	Bio400	Talc400	P value
Inflammation	1.0±0	1.6±0.3	2.6±0.5	<0.001*
Fibrosis	1.0±0	1.6±0.6	2.2±0.4	<0.001*
Particle in lung	1.0±0	2.1±0.4	2.5±0.9	0.002*
Particle in BAL	1.0±0	1.0±0	2.0±0.3	0.004*

Multiple comparisons: Inflammation: Control vs Bio400, p<0.001*; Control vs Talc400, p<0.001*; Bio400 vs Talc400, p<0.001*; Fibrosis: Control vs Bio400, p<0.001*; Control vs Talc400, p<0.001*; Bio400 vs Talc400, p=0.083; Particle in lung: Control vs Bio400, p=0.010*; Control vs Talc400, p=0.002*, Bio400 vs Talc400, p=0.382; Particle in BAL: Control vs Biog400, p=0.002*; Control vs Talc400, p=0.105; Bio400 vs Talc400, p=0.279, (*) Statistically significant

Table 3: Evaluation of systemic inflammatory effect

Parameter (mean±SD)	Control		Bio400		Talc400		P value	
	6h	24h	6h	24h	6h	24h	6h	24h
LDH (u/L)	684±56	864±55	981±111	1501±216	2400±53	1346±210	0.003*	0.029*
IL-8 (pg/ml)	160±32	304±38	312±31	630±81	297±33	736±68	0.001*	0.002*

Multiple comparisons: LDH 6h and 24h: Control vs Bio400, p=0.002* and 0.028*; Control vs Talc400, p=0.028* and 0.015*; Bio400 vs Talc400, p= 0.038* and 0.721; IL-8 6h and 24h: Control vs Bio400, p=0.001* and 0.003*; Control vs Talc400, p=0.001* and 0.001*; Bio400 vs Talc400, p= 0.878 and 0.382. (*) Statistically significant.

Discussion

This study showed that a high dose of (400 mg/kg) bioglass and talc produced a similar pleurodesic effect. In the current study, we found that doses of 35, 70 and 150 mg/kg of bioglass and talc caused a weak pleurodesis in contrast to a previous study.[8] We think that this opposity might have resulted from using a different pleurodesis grading methodology. In the previous study, as in the literature, we kept the thoracic case block in 10% formalin solution for at least 48 hours before pleurodesis grading. We noticed that formalin exposure made the tissues firm and shrinked, making the evaluation of adhesions and symphis difficult. Therefore, in the current study, we graded pleurodesis immediately after sacrification, while the tissues were fresh and soft. Thus, we tried to minimize the probability of making mistakes in pleurodesis grading.

Miller et al.[10] and Light et al.[11] reported that doses of 50-70 mg/kg talc created a weak pleurodesis; however, a dose of 400 mg/kg talc produced an effective pleurodesis.[11,12] We found that a dose of 400 mg/kg of talc resulted into a moderate degree of pleurodesis (mean=3.8±0.5). We never noticed strong adhesions and symphisis in any group. It might have been due to the difference in the grading methodology mentioned above, but we could not find any report supporting us.

The effect of bioglass may be related to its silicon dioxide content. Silicon dioxide is a crystalline type of silica. Silica exposure may cause pleural inflammation. Another possible inflammatory effect is due to its particular structure. Particles of a size smaller than 1 mm, when phagocytized by cells, are held to be responsible for the cytokine response. Those particles that are greater in size may not be phagocytized. However, they may induce inflammatory mediator release.[13] Hence, Bioglass is capable of releasing ions, which may affect cellular responses and change intracellular ions, resulting into an increases in pH, calcium, potassium, small decreases in sodium, and increases in lactate production and ATP generation by stimulation of glycolysis.[14] As a result, collagen and cytokine release from the cells mentioned above are stimulated by bioglass. The current study showed that bioglass created a local and systemic inflammation when applied intrapleurally. While its local inflammatory effect was lesser than that of talc, systemic inflammation was not significantly different.

Talc is a hydrated magnesium silicate, $Mg_3Si_4O_{10}(OH)_2$, and is one of the most popular pleurodesing agents because of its high effectiveness, low cost, and wide availability. However, talc use is associated with adult respiratory distress syndrome

(ARDS) in 3% to 9% of cases after intrapleural administration.[15] Other reported side effects include fever, pain, infection, hypotension, arrhythmia, arterial desaturation syndrome, and sclerosis.[16] Systemic distribution and progressive deposition of talc particles after intrapleural administration have been shown in animal studies.[15] In addition, talc may cause carcinogenesis in mice.[17]

Bioglass may be a better candidate agent for pleurodesis. It has some advantages over talc: (i) bioglass is biocompatible and nontoxic; (ii) it is easily available; (iii) it causes less pleural fibrosis; (iv) it has an antibacterial activity, and (v) there is no evidence that it has carcinogenic properties.[18] In addition, because it produces less inflammation and fibrosis, it may cause less pain.

There were some limitations in this study. First, there was not a commercial form of bioglass for pleurodesis. So, we had to make its size smaller ourselves. We could obtain particles smaller than 100 micron, but they were not homogenous in size and the range was so wide. Probably, if we could achieve smaller particles in size, we would provide different outcomes. Second, pleurodesis model performed in rabbits does not entirely fit the clinic pleurodesis performed for human beings. The agent intrapleurally was given as 1 ml/kg in saline solution to remain in the thoracic cavity. This solution should be resorbed first, and then pleurodesis begins. This reabsorption may not be similar in all rabbits. So, experimental models may not exactly reflect the clinical outcomes. Lastly, this is an animal study, and we have not had an adequate knowledge about the safety of application of large amounts of bioglass in human cavities. Therefore, further studies are needed for more reliable results.

In conclusion, bioglass may be a good new candidate agent for pleurodesis. A dose of 400 mg/kg bioglass has similar pleurodesic effect as the same dose of talc in rabbits. Bioglass causes less inflammation and fibrosis. This condition may make bioglass a more valuable pleurodesic agent than talc. However, further studies are needed for more definite results.

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Conflict of interest: None declared.

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