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·基础研究·

硫酸钙基乳牙根管封闭剂的性能研究

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【摘要】　目的　探讨含半水硫酸钙、硫酸钡、醋酸氯己定和聚乙二醇（polyethylene glycol 400，PEG 400）的新型硫酸钙基乳牙根管封闭剂的理化性能和生物学性能。**方法**　本研究已通过单位伦理委员会审查批准。将不同质量分数半水硫酸钙和硫酸钡的混合粉末按照3∶1粉液比与PEG 400混合，醋酸氯己定依照PEG 400的体积以0.2 mg/mL加入。将上述材料以250 r/min的转速球磨24 h得到硫酸钙基乳牙根管封闭剂。根据各组分质量分数的不同分组，通过固化时间、流动性、阻射性能实验结果优化出合适的组分质量分数，再用最优质量分数组对封闭剂的pH值、体外质量丧失、显微形态进行评价。通过细菌-材料共培法检测封闭剂的抗菌性，通过CCK-8实验和细胞形态染色评价其细胞相容性，通过皮下组织埋置实验评价其生物相容性。**结果**　优化后的封闭剂，半水硫酸钙质量分数为80 wt%，硫酸钡质量分数为20 wt%。封闭剂的流动性、辐射不透明度均符合国际标准。封闭剂在去离子水组和磷酸盐缓冲液（phosphate buffer solution，PBS）组的pH值稳定在6～7，第7天和第14天时封闭剂在去离子水组中的pH值显著高于PBS组（$P<0.001$），随后二组的pH值逐渐接近（$P>0.05$）。体外降解实验发现，前期封闭剂的质量丧失率约15.17%，3周后质量丧失率仅约8.33%。X射线衍射和扫描电子显微镜检测发现水化后的封闭剂主要成分是二水硫酸钙。细菌学实验和细胞学检测中，封闭剂抑制粪肠球菌生长的作用明显（$P<0.001$）；第1天和第4天时，1∶10和1∶20封闭剂浸提液稀释组的骨髓间充质细胞活力明显低于对照组（$P<0.05$），至第7天时，1∶20封闭剂浸提稀释液对细胞增殖能力无明显影响（$P>0.05$）。组织切片观察封闭剂组与对照组（Vitapex糊剂及ZOE糊剂）均为轻度炎症反应。**结论**　本实验研制了一种新型硫酸钙基乳牙根管封闭剂，其具有良好的理化性能和较强的抗菌性，可以满足生物相容性的要求，为乳牙根管封闭剂的研发提供了新思路。

【关键词】　硫酸钙；　氯己定；　根管封闭；　乳牙；　可降解；　生物相容性；　抗菌性；　组织切片
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Performance of calcium sulfate-based root canal sealers for deciduous teeth

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【Abstract】　Objective　To investigate the physicochemical and biological properties of a new calcium sulfate-based root canal sealer for deciduous teeth containing calcium sulfate hemihydrate, barium sulfate, chlorhexidine acetate, and polyethylene glycol 400 (PEG 400). **Methods**　This study was reviewed and approved by the Ethics Committee. The calcium sulfate hemihydrate and barium sulfate powders with different mass percentages were mixed with liquid PEG 400 at a powder-to-liquid ratio of 3∶1, and chlorhexidine acetate was added to a concentration of 0.2 mg/mL according to the volume of PEG 400. The above materials were mechanically ground at 250 r/min for 24 h to obtain a calcium sulfate-based root canal sealer for deciduous teeth. The sealer was classified into different groups according to mass percentages of components. The mass percentages of components were optimized by performing time, fluidity, and radi-

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opacity experiments, and then the pH, mass loss *in vitro*, and microscopic morphology of the optimal sealer were evaluated. The antimicrobial properties of the sealer were evaluated by a bacterial-material cocultivation method. The cytocompatibility of the sealer was evaluated by a CCK-8 assay and cytomorphological staining, and its biocompatibility was evaluated by a subcutaneous tissue embedding assay. **Results** After optimization, mass percentage of calcium sulfate hemihydrate was 80 wt%, and the mass percentage of barium sulfate was 20 wt%. The flowability and radiopacity of the sealer were in accordance with international standards. The pH stabilized between 6-7. On the 7th and 14th days, the pH in the water group was significantly greater than that in the PBS group ($P < 0.001$), although the pH in both groups gradually increased ($P > 0.05$). *In vitro* degradation experiments, the mass loss of the sealer was approximately 15.17% during the preimmersion period, and rate of mass loss decreased after 3 weeks, reaching only approximately 8.33%. X-ray diffraction (XRD) and scanning electron microscopy (SEM) revealed that the main component of the sealer after hydration was calcium sulfate dehydrate. In bacterial growth assays and cytological tests, the sealer showed significant inhibition of the growth of *E. faecalis* ($P < 0.001$). After 1 and 4 days of culture, the cell viability in the 1∶10 and 1∶20 sealer extract dilution group was lower than that in the control group ($P < 0.05$). On the 7th day, the 1∶20 sealer extract dilution had no significant effect on cell proliferation ($P > 0.05$). Both the sealer group and the control group (Vitapex and zinc oxide eugenol) caused mild inflammatory reactions in tissue sections. **Conclusion** In this study, a new type of root canal sealer for deciduous teeth was designed based on calcium sulfate, which has good physicochemical properties and strong antibacterial properties and meets biocompatibility requirements. This study provides an idea for the development of a new type of root canal sealer for deciduous teeth.

【Key words】 calcium sulfate; chlorhexidine; root canal sealing; deciduous teeth; degradable; biocompatible; antibacterial properties; tissue sectioning

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乳牙的牙髓和根尖周病变若没有得到有效治疗，可导致乳牙过早脱落，进而影响患儿的生活[1]，根管治疗术是其最有效的治疗方法[2]，简单的机械和化学清理不能完全清创[3]，因此选择合适的根管封闭剂非常重要[4]。乳牙根管封闭剂的理想特性如下[5]：①与乳牙牙根吸收速度基本一致；②不会对根尖周组织和恒牙胚造成损害；③超充的材料易吸收；④具有防腐性能并能抑制微生物的生长[6]。

氧化锌丁香酚（zinc oxide eugenol，ZOE）因其易于使用已被广泛用于乳牙根管充填[7-8]，但其缺乏生物相容性，严重可导致根尖周组织坏死[9-11]；还可能会影响后续恒牙的萌出。目前临床上最常用的是氢氧化钙碘仿糊剂（vitapex）。然而，其过早吸收会使根管失去抗感染性能，导致再次感染[12-13]。硫酸钙来源丰富，在体内可被完全吸收[14]；硫酸钡具有优异的显影能力，常用作造影剂[15]；氯己定是一种广谱杀菌剂，具有良好的抗菌性和生物相容性，在口腔疾病的防治中应用广泛[16-17]；聚乙二醇400（polyethylene glycol 400，PEG 400）稳定，经FDA（食品药品监督管理局）批准可用于医药用途，例如药物制剂、眼药水和化妆品的赋形剂[18]。本研究拟研制含半水硫酸钙、硫酸钡、醋酸氯己定和PEG 400的硫酸钙基乳牙根管封闭剂，探究其理化性能及生物学性能，以期为乳牙根管封闭剂提供一种新思路。

1　材料和方法

1.1　主要试剂和仪器

半水硫酸钙（C885057，麦克林，中国），硫酸钡（10004460，沪试，中国），PEG 400（P103737，阿拉丁，中国），醋酸氯己定（C107054，阿拉丁，中国），磷酸盐缓冲液（phosphate buffer solution，PBS）（P1010，索莱宝，中国），DMEM（11965092，Gibco，美国）、胎牛血清（A5669701，Gibco，美国），青链霉素混合液（C0222，碧云天，中国），脑心浸液肉汤（HB8297-1，海博，中国），罗丹明异硫氰酸酯（TRITC）鬼笔环肽（CA1610，索莱宝，中国），DAPI（C0060，索莱宝，中国）。球磨机（MITR YXQM-0.4L，米琪，中国），pH计（Seven Compact™ S210，Mettler Toledo，瑞士），牙科X线射线机（Discovery XR650，GE，中国），X线射线机（X Pert Pro MPD，

Malvern Panalytical，荷兰），水纯化系统（RiOs8，Millipore，美国），微型高速离心机（MINISPIN PLUS，Eppendorf，德国），厌氧培养箱（DG250，广州华粤行，中国），酶标仪（Enspire，PerkinElmer，新加坡），场发射扫描电子显微镜（Gemini SEM 300，Zeiss，德国）。

1.2　材料的分组与制备

如表1所示，根据半水硫酸钙和硫酸钡的不同质量分数分为G1、G2、G3、G4组。按照粉液比为3∶1与PEG 400混合，醋酸氯己定依照PEG 400的体积以0.2 mg/mL加入。将上述材料在球磨机中均匀混合24 h得到硫酸钙基乳牙根管封闭剂。

表1　各组硫酸钙基乳牙根管封闭剂各成分质量分数

Table 1　Mass fraction of each component of calcium sulfate-based root canal sealer for deciduous teeth in different group

Group	CSH (wt%)	$BaSO_4$ (wt%)	CHX (mg/mL)
G1	95	5	0.2
G2	90	10	0.2
G3	85	15	0.2
G4	80	20	0.2

CSH: calcium sulfate hemihydrate. BaSO4: barium sulfate. CHX: chlorhexidine

1.3　材料理化性质检测

1.3.1　固化时间和流动性检测　根据ISO 6876-2012[19]标准检测。从封闭剂注入模具计算，直到Gilmore针的针尖无法在其水平表面留下压痕为止，该时间为封闭剂最终固化的时间，重复3次。

流动性检测：将0.05 mL封闭剂注于玻璃的中心，再用第二块玻璃板覆盖封闭剂，并在其上放置一个砝码，总重量为（120 ± 2）g。10 min后，取下砝码并记录封闭剂的最大和最小直径。如果相差在1 mm以内，则取平均值。如果相差大于1 mm，则重复测试，每组3个样本。

1.3.2　辐射不透明度检测　按照ISO 6876-2012[19]进行测试。将封闭剂与楔状铝阶放置于咬合片上。使用牙科X射线机平行投影，具体参数为：70 kV，8 mA，投影距离30 cm，曝光时间0.12 s。使用Adobe Photoshop CC处理图像，根据图像的灰度将封闭剂的辐射不透明度转换为铝板的厚度，每组3个样本。

1.3.3　pH值和体外降解测定　封闭剂固化后分别浸入10 mL去离子水和PBS中。在1、7、14、21、28 d测量pH值，每组设置5个平行样本。记录封闭剂的初始质量，浸入10 mL PBS溶液中。于1、7、14、21、28 d从恒温摇床中（37 ℃，80 r/min）取出，PBS漂洗3次后放入烘箱干燥，称重并记录，每组取5个样本，质量丧失率=（初始质量−期末质量）/初始质量×100%。

1.3.4　封闭剂的结构特征　用扫描电子显微镜和X线射线机观察封闭剂结构形貌，进行X射线衍射（X-ray diffraction，XRD）分析。

1.3.5　体外生物学性能　抗菌性能检测：以新鲜脑心浸出液肉汤培养基作为对照，封闭剂浸入脑心浸出液肉汤液体培养基中24 h，收集上清液将粪肠球菌的菌落（*E. faecalis*; ATCC 29212）调整为1 × 10^6 CFU/mL（实验组）。取200 μL菌液接种至96孔板中，每组设5个复孔，于1、3、5、7、9、12、24 h在酶标仪中读取A600处的OD值，绘制细菌生长曲线。

细胞相容性检测：本研究获得安徽医科大学实验动物伦理委员会（批号：LLSC-2023-2002）批准，使用雌性SD大鼠的骨髓间充质细胞检测封闭剂的细胞相容性。经由颈椎脱位将3周龄SD大鼠处死，75%乙醇浸泡后分离股骨和胫骨表面的肌肉和肌腱组织，PBS冲洗干净后放入完全培养基。裁剪股骨、胫骨，用注射器冲出骨髓，收集至15 mL离心管中反复吹打至无明显细胞团后离心去上清，重新加入2 mL的完全培养基重悬后培养在37 ℃，95%湿度，5% CO_2环境中。经过消化，离心，再悬浮，细胞培养到第3代进行后续细胞实验。

封闭剂溶解后的颗粒会影响细胞贴壁，所以采用其浸提液培养细胞。封闭剂浸入1 mL DMEM培养基24 h，得到提取液，用培养基按1∶10和1∶20的比例稀释提取液（实验组），并以新鲜培养基为对照组，在1、4、7 d检测细胞增殖情况。

细胞荧光免疫染色：1、4、7 d分别取出对照组和1∶20组的细胞爬片，用2.5%戊二醛固定24 h后加入罗丹明标记的鬼笔环肽和含防荧光淬灭的DAPI，荧光显微镜下观察细胞骨架形态。

1.3.6　体内生物学性能　使用戊巴比妥钠对6周龄，体重200 ~ 250 g的SD雄性大鼠（动物合格证号：SCXK（辽）2020-0001）行腹腔麻醉。将Vitapex、ZOE（对照组）和封闭剂（实验组）植入大鼠背部皮下，于2、4周后处死大鼠，收集材料植入处的皮下组织并用4%多聚甲醛固定，固定标本处理后用石蜡包埋，苏木精-伊红染色后镜下观察。

1.4　统计学方法

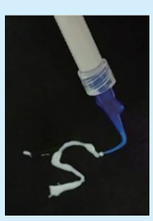
本研究采用SPSS 27.0统计软件进行分析，计量资料以均数 ± 标准差表示。用单因素方差分析

（ANOVA）对数据进行检验，$P < 0.05$表示差异有统计学意义。

2　结　果

2.1　封闭剂的制备

硫酸钙基乳牙根管封闭剂是一种颜色均匀的乳白色糊剂，可通过注射器推出，具有可注射性（图1）。



The sealer was a milky white paste of uniform color with injectability

Figure 1　Morphology of calcium sulfate-based root canal sealer for deciduous teeth

图1　硫酸钙基乳牙根管封闭剂的形态

2.2　封闭剂的理化性能

2.2.1　固化时间和流动性　结果如表2所示，各组封闭剂的流动性均超过17 mm，符合ISO标准（6876-2012）。封闭剂的凝固时间约为9～10 h，此范围适用于临床上根管充填。国际标准组织仅对生产商提供的固化时间提出要求，并未对封闭剂固化时间做出具体规定，因此表2仅分析各组流动性结果与ISO标准之间的关系。

2.2.2　辐射不透明度　结果如图2所示，硫酸钡含量为10 wt%时，达到ISO标准（3 mm Al）。除G2组外，各组与ISO标准之间的差异均有统计学意义（$P < 0.001$）。美国牙科协会第57号[20]规定，以铝板的阻射厚度作为参考，根管封闭剂的阻射厚度要比牙本质或者骨质至少大2 mm。因为牙本质的X线阻射值接近于3 mm铝板，只有G4组符合上述规定，而且G4组的凝固时间相较于其他组缩短，便于临床应用。

表2　各组硫酸钙基乳牙根管封闭剂的流动性和凝固时间

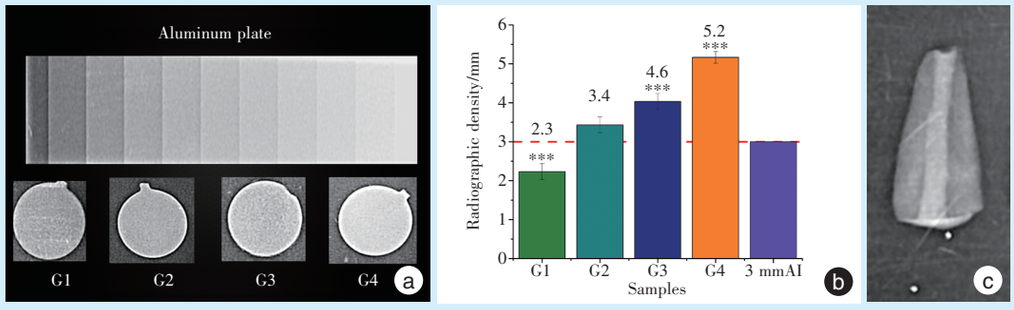
Table 2　Flowability and setting time of calcium sulfate-based root canal sealer for deciduous teeth in different group

Group	Setting time/h	Flow/mm	*t*-test (flow)*	
			t	*P*
G1	10.3 ± 0.737	26.2 ± 0.346	17.705	0.003
G2	9.8 ± 1.044	24.4 ± 0.709	18.147	0.003
G3	9.2 ± 0.872	22.9 ± 0.600	17.032	0.003
G4	8.6 ± 0.608	21.3 ± 0.625	11.926	0.007

*: *vs* ISO (17 mm). The setting time of sealer in Group G4 was shorter than in other groups, which was convenient for clinical application. Groups G1-G4 were shown in Table 1. CSH: calcium sulfate hemihydrate. $BaSO_4$: barium sulfate

2.2.3　pH值和体外降解　图3a显示第7天和第14天，去离子水中组封闭剂的pH值显著高于PBS组（$P < 0.001$）。随后两组差异逐渐缩小，第21天和28天时，封闭剂在PBS组和去离子水组中的pH值无显著差异（$P = 0.690$，$P = 1.000$）。图3b显示了封闭剂浸入PBS后质量随时间的变化情况，封闭剂初始质量为（0.579 ± 0.050）g，前3周，质量丧失率约为15.17%，随后，质量丧失率下降至8.33%。封闭剂的质量丧失情况表明其具有可降解性，适用于乳牙根管充填。

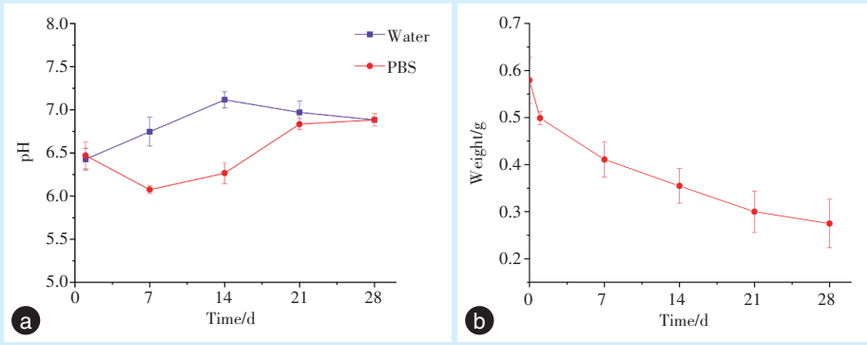
2.2.4　结构特征　SEM结果如图4a、4b所示，水化48 h后的样品能够观察到针状晶体，溶解30 d后，断裂表面出现了更厚的多面晶体，这些晶体在形态学上与二水硫酸钙晶体相同[21-22]。封闭剂粉末通过XRD观察（图4c），2θ在23.56°、25.84°、28.746°、31.52°、41.6°、42.9°、43.98°、48.9°处的衍射



a: representative X-ray images of the different sealers and aluminum step wedges in 1 mm increments from 1 to 10 mm. b: the relative radiation density of each group was quantified compared to that of a 3 mm aluminum step wedge. *** $P < 0.001$. c: radiograph of the Group G4 sealer in an isolated tooth. Groups G1-G4 were shown in Table 1.The radiopacity of sealer in Group G4 was better than in other groups, with the blocking thickness 2 mm thicker than dentin, which was in line with ISO

Figure 2　Images of calcium sulfate-based root canal sealers for deciduous teeth from different groups

图2　各组硫酸钙基乳牙根管封闭剂的影像学结果



a: change in pH of the sealer in PBS and deionized water over time. From Day 1 to Day 28, the pH of the sealer in PBS and deionized water was stable between 6 and 7. b: change in the mass of the sealer in PBS over time. The change in the mass of sealer indicated that it was degradable and suitable for filling deciduous teeth root canals; the initial mass was (0.579 ± 0.050) g. Group G4: 80 wt% calcium sulfate hemihydrate; 20 wt% $BaSO_4$

Figure 3　Changes in the pH and mass of the Group G4 calcium sulfate-based root canal sealer for deciduous teeth in PBS and deionized water

图3　G4组硫酸钙基乳牙根管封闭剂在PBS和去离子水中的pH值和质量的变化

峰可以与硫酸钡的标准衍射峰很好匹配。2θ在29.1°、35.96°、43.43°、48.84°、50.32°、55.82°、65.82°、66.68°的衍射峰与二水硫酸钙的标准图谱衍射峰基本一致。

2.3　体外评估封闭剂的生物特性

2.3.1　抑菌实验　图5a显示了封闭剂对粪肠球菌生长的抑制作用。实验组和对照组培养基中细菌生长曲线的形态及趋势基本一致，同一个时间点实验组的细菌生长均低于对照组，两组之间的差异具有统计学意义（$P<0.001$）。

2.3.2　体外细胞实验　图5b为大鼠骨髓基质细胞形态学图片，经过5 d培养，镜下观察到细胞呈集落贴壁生长，形态呈梭形或多边形。培养7 d后，细胞数量增多，重叠生长。

在不同浓度浸提液中培养的大鼠骨髓基质细胞增殖情况如图5c所示。第1天，实验组（1∶10和1∶20封闭剂浸提液稀释组）与对照组的细胞活性之间均有显著差异（$P<0.001$）；第4天，1∶10和1∶20浸提液稀释组细胞活性与对照组之间差异明显（$P<0.001$，$P=0.018$）；第7天，1∶10浸提液稀释组细胞活性与对照组之间差异明显（$P=0.002$），1∶20浸提液稀释组与对照组之间无明显差异（$P=0.461$）。

选取1∶20浸提液稀释组观察细胞骨架形态，如图5d所示。第1天，爬片上的细胞呈梭形附着；第4天，细胞数量明显增加；第7天时，细胞形态更显著。

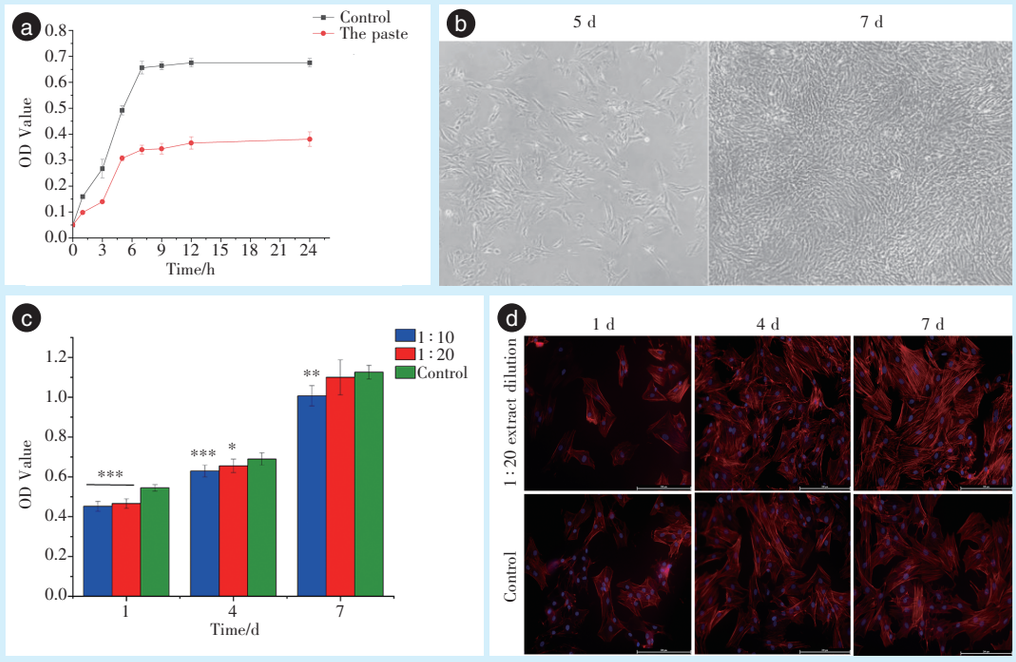
2.4　生物相容性

皮下炎症组织切片如图6所示，封闭剂组、Vitapex组和ZOE组在2周时均观察到轻中度的炎症反应。4周时，封闭剂组和对照组的炎症反应均减轻，仅存在少量的炎症细胞。相较于Vitapex组和ZOE组，封闭剂组并未出现更加严重的炎症反应。

a: the SEM image of the sealer after 48 h of hydration showing needle-like crystals; scale bar = 5 μm; b: the SEM image of the cured sealer after dissolution for 30 d showing short rod-like crystals; scale bar = 5 μm; c: X-ray diffraction image of the sealer after 48 h hydration. The main components of the sealer were CSD (the hydrated CSH) and $BaSO_4$. CSD: calcium sulfate dehydrate. CSH: calcium sulfate hemihydrate. Group G4: 80 wt% CSH; 20 wt% $BaSO_4$

Figure 4　Structural charateristics of Group G4 calcisum sulfate-based root canal sealer for deciduous teeth

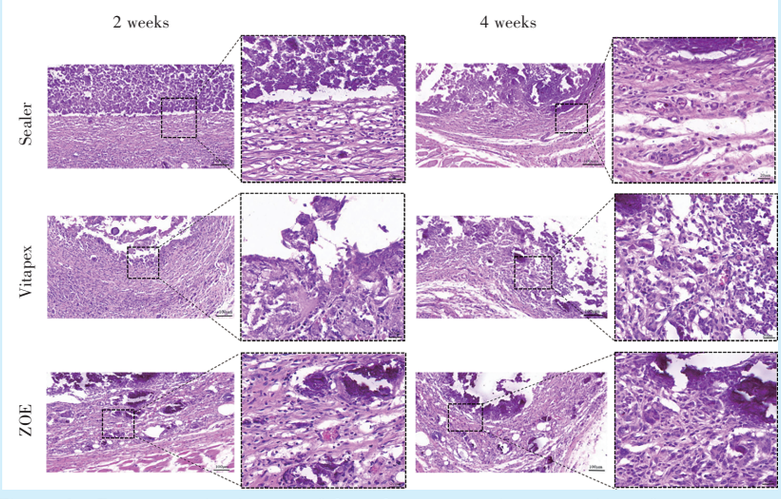
图4　G4组硫酸钙基乳牙根管封闭剂的结构特征



a: curves showing the effect of the sealer on the growth of *E. faecalis*. There was less bacterial growth in the experimental (G4) group than that in the control group, indicating that the sealer had good antibacterial properties. The difference between the two groups was significant ($P < 0.001$); b: morphology of SD-BMSCs in DMEM (the cells were shuttle-shaped at 5 d, and this morphology was more pronounced at 7 d, at which time the cells increased in number; ×200); SD-BMSCs: SD rat bone marrow mesenchymal stem cells; c: effect of the sealer (Group G4) on the proliferation of SD-BMSCs: 1:10 extract dilution and 1:20 extract dilution. The extracts were obtained after the sealer was immersed in DMEM for 24 h, after which the extract was diluted with medium at ratios of 1: 10 and 1: 20. *vs*. control group, *: $P < 0.05$, **: $P < 0.01$, ***: $P < 0.001$; d: cytoskeletal morphologies of SD-BMSCs. There were no significant differences in cytoskeletal morphology between the sealer and control groups. Control group: DMEM without material extract. Nuclei are shown in blue, and actin is shown in red (bar = 200 μm). BMSCs: bone marrow mesenchymal stem cells. Group G4: 80 wt% calcium sulfate hemihydrate; 20 wt% $BaSO_4$

Figure 5　*In vitro* evaluation of the biological characteristics of Group G4 calcium sulfate-based root canal sealer for deciduous teeth

图5　体外评估G4组硫酸钙基乳牙根管封闭剂的生物特性



At 2 weeks, the sealer group (G4), Vitapex group and ZOE group exhibited mild to moderate inflammatory responses. At 4 weeks, the inflammatory responses were reduced and only a few inflammatory cells remained in the sealer group, Vitapex group and ZOE group. The sealer group (G4) did not show more severe inflammatory responses than the clinically used sealers Vitapex and ZOE, suggesting that the sealer had good biocompatibility. Scale bar=100 μm or 20 μm. Vitapex: calcium hydroxide iodoform paste; ZOE: zinc oxide eugenol. Group G4: 80 wt% calcium sulfate hemihydrate; 20 wt% $BaSO_4$

Figure 6　Representative images of the subcutaneous tissue reactions after treatment with the Group G4 calcium sulfate-based root canal sealer for deciduous teeth, Vitapex and ZOE

图6　G4组硫酸钙基乳牙根管封闭剂、Vitapex和ZOE的皮下组织反应代表性图像

3　讨　论

本研究研制出一种硫酸钙基乳牙根管封闭剂，具有良好的理化性能和生物学性能。在材料成分方面，采用体内几乎可被完全吸收的硫酸钙为主体[23]，其来源丰富，获取容易，作为乳牙根管封闭剂的成分被研发应用，具有巨大的社会效益和经济效益。采用硫酸钡作为封闭剂的X线显影成分，阻射结果显示，硫酸钡（20 wt%）显影能力优异，可以满足临床放射学检查要求。氯己定是一类广谱抗菌剂，对革兰阳性和革兰阴性细菌以及细菌孢子、亲脂性病毒、酵母菌和皮肤真菌等均有抗菌效果，并且具有低浓度抑菌高浓度杀菌的作用[24]。加入氯己定的封闭剂对粪肠球菌生长趋势的影响显著，表明其抑菌活性良好，可提高乳牙根管治疗的成功率。临床上长时间使用氯己定漱口水会造成牙齿染色[25]，这主要是因为氯己定属阳离子物质，会与食物、饮料中的阴离子物质（如变形蛋白质）形成硫化亚铁沉积，而本实验研制的封闭剂为乳白色糊剂，且被严密充填在根管内，不与外界接触，所以不会导致牙冠变色。

硫酸钙的临床应用历史较长，吸收率可以调整[26-27]，不会引起显著的炎症反应[28]。半水硫酸钙通过吸收周围环境中的水进行水化反应，这也是封闭剂固化的原因。当半水硫酸钙与水混合时，二水合物伴随一个温和的放热反应形成[29]：$CaSO_4 \cdot 1/2H_2O + H_2O \rightarrow CaSO_4 \cdot 2H_2O + Heat$。水化反应时，在二水硫酸钙晶体长大的过程中，针状晶体互相挤压、支撑，使结构中存在孔隙，体积膨胀。这一过程有利于避免根充材料与牙本质壁之间产生缝隙，可形成“自我封闭”，保证了严密充填，也降低了根管发生再次感染的风险。

根管封闭剂应具有抗菌性和良好的生物相容性[30-31]，否则从材料中释放出的有毒物质可能会影响细胞形态和附着[32]。实验中加入醋酸氯己定，封闭剂表现出较强的抑菌作用，同时又在CCK-8试验中表现出良好的生物相容性。250 μg/mL的氯己定已被证明具有一定的细胞毒性[33]，而在本实验中，封闭剂依然表现出较好的生物活性，考虑为氯己定的阳离子基团与硫酸基团产生静电作用[34]，吸附到硫酸钙上从而降低了细胞毒性。

综上所述，硫酸钙基乳牙根管封闭剂具有良好的理化性能、生物相容性和抗菌性，为新型乳牙根管封闭剂的研发提供了思路。但是作为一种新型乳牙根管充填材料，其在乳牙内的情况需进一步评价，吸收速率与硫酸钙含量间关系也需在后续实验中进一步探究，以期临床转化应用。

【Author contributions】 Li XX performed the experiments and wrote the article. Li XF analyzed the data and revised the article. Li QL designed the study, conceptualized and reviewed the article. All authors read and approved the final manuscript as submitted.

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