以不同方法製備可注射式水膠

應用於軟骨組織工程

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一、前言

二、以光交聯製備可光聚合幾丁聚醣/玻尿酸水膠

三、以共價交聯製備羧甲基幾丁聚醣/硫酸軟骨素水膠

四、以梔子素交聯製備緊密堆疊之幾丁聚醣微膠

五、以酵素交聯製備酚類幾丁聚醣水膠

六、結論

摘要

骨關節炎是經由軟骨逐漸變異而形成的一種肌肉與骨骼之間的缺陷，流行病學研究指出 55-78 歲成年人中有 70%曾受骨關節炎之苦，人體軟骨由於無血管且修復細胞少，因此自我修復的能力有限。可注射式水膠是一種新穎的智慧型材料，在組織工程方面可應用為生物支架材料，利用注射的方法將支架材料打入人體，達到減少手術傷口大小而降低患者術後其他併發症的可能性，而可注射式水膠主要功能為提供適合細胞生長的環境，若要達到軟骨修復的目的，必須藉由包覆生長因子或幹細胞來促進軟骨細胞生長。幾丁聚醣為具生物相容性且結構類似軟骨裡黏多醣的天然多醣，因此是一種引人注目且可以應用於軟骨修復的生醫材料。在此會以光交聯、共價交聯、梔子素交聯、酵素交聯製備出可注射式水膠，光交聯的最適條件為 6 μM RF、照光 300 秒；羧甲基幾丁聚醣/硫酸軟骨素溶液經由 Schiff base 反應成功形成水膠，而加入 20 mg/ml 微粒能提升水膠的機械性質，另外裝載有 BSA 的微粒可提升水膠之生物活性；梔子素含量較低的 GP-5 微膠由於有較好的剪切稀化性質且可包覆不同電荷的血管內皮生長因子(Vascular endothelial growth factor, VEGF)，因此適合當作可注射式水膠；藉由消耗 H_2O_2 與利用辣根過氧化酶(Horseradish peroxidase, HRP)催化交聯反應成功形成水膠，最適條件為 0.5 mM H_2O_2、1 unit/ml HRP 且在室溫下即可成膠。

参考文獻

Atsumi, T., Iwakura, I., Fujisawa, S., Ueha, T. (2001). The production of reactive oxygen species by irradiated camphorquinone-related photosensitizers and their effect on cytotoxicity. *Archives of Oral Biology*, 46, 391-401.

Cheng, N. C., Estes, B. Young, T. H. Guilak, F. (2013). Genipin-crosslinked cartilage derived matrix as a scaffold for human adipose-derived stem cell chondrogenesis. *Tissue Engineering Part A*, 19, 484-496.

DeFail, A. J., Chu, C. R., Izzo, N., Marra, K. G. (2006). Controlled release of bioactive TGF-β 1 from microspheres embedded within biodegradable hydrogels. *Biomaterials*, 27, 1579-1585.

Di Martino, A., Sittinge, M., Risbud, M. V. (2005). Chitosan: a versatile biopolymer for orthopaedic tissue-engineering. *Biomaterials*, 26, 5983-90.

Dodane, V., Vilivalam, V. D. (1998). Pharmaceutical applications of chitosan. *Pharmaceutical Science & Technology Today*, 1, 246-53.

Fan, M., Ma, Y., Tan, H., Jia, Y., Zou, S., Guo, S., Zhao, M., Huang, H., Ling, Z., Chen, Y., Hu, X. (2017). Covalent and injectable chitosan-chondroitin sulfate hydrogels embedded with chitosan microspheres for drug delivery and tissue engineering. *Materials Science and Engineering: C*, 71, 67-74.

Fedorovich, N. E., Oudshoorn, M. H., van Geemen, D., Hennink, W. E., Alblas, J., Dhert, W. J. A. (2009). The effect of photopolymerization on stem cells embedded in hydrogels. *Biomaterials*, 30, 344-53.

Galeska, I., Kim, T. K., Patil, S. D., Bhardwaj, U., Chatttopadhyay, D., Papadimitrakopoulos, F., Burgess, D. J. (2005). Controlled release of dexamethasone from PLGA microspheres embedded within polyacid-containing PVA hydrogels. *AAPS Journal*, 7, E231-E240.

García Cruz, D. M., Escobar Ivirico, J. L., Gomes, M. M., Gómez Ribelles, J. L., Sánchez,M. S., Reis, R. L., et al. (2008). Chitosan microparticles as injectable scaffolds fortissue engineering. *Journal of Tissue Engineering and Regenerative Medicine*, 2(6), 378-380.

Griffin, D. R., Weaver, W. M., Scumpia, P. O., Di Carlo, D., Segura, T. (2015). Accelerated wound healing by injectable microporous gel scaffolds assembled from annealed building blocks. *Nature Materials*, 14(7), 737-744.

Guo, Z., Xing, R., Liu, S. Yu, H., Wang, P., Li, C., Li, P. (2005). The synthesis and antioxidant activity of the Schiff bases of chitosan and carboxymethyl chitosan. *Bioorganic & Medicinal Chemistry Letters*, 15, 4600-4603.

Hunziker, E. (2002). Articular cartilage repair: basic science and clinical progress. A review of the current status and prospects, Osteoarthritis Cartilage, 10, 432-463.

Jiang, Y., Chen, J., Deng, C., Suuronen, E.J., Zhong, Z. (2014). Click hydrogels, microgels and nanogels: emerging platforms for drug delivery and tissue engineering. *Biomaterials*, 35, 4969-4985.

Ketz, R. J., Prud'homme, R. K., Graessley, W. W. (1988). Rheology of concentrated microgel solutions. *Rheologica Acta*, 27(5), 531-539.

Kheir, E., Stapleton, T., Shaw, D., Jin, Z., Fisher, J., Ingham, E. (2011). Development and characterization of an acellular porcine cartilage bone matrix for use in tissue engineering. *Journal of Biomedical Materials Research Part A*, 99, 283-294.

Kim, I. L., Mauck, R. L., Burdick, J. A. (2011). Hydrogel design for cartilage tissue engineering: a case study with hyaluronic acid. *Biomaterials*, 32, 8771-82.

Kim, S. E., Park, J. H., Cho, Y. W., Chung, H., Jeong, S. Y., Lee, E. B., Kwon, I. C. (2003). Porous chitosan scaffold containing microspheres loaded with transforming growth factor-β1: implications for cartilage tissue engineering. *Journal of Controlled Release*, 91, 365-374.

Kittur, F. S., Prashanth, K. V. H., Sankar, K. U., Tharanathan, R. N. (2002). Characterization of chitin, chitosan and their carboxymethyl derivatives by differential scanning calorimetry. *Carbohydrate Polymers*, 49, 185-193.

Koo, H. J., Song, Y. S., Kim, H. J., Lee, Y. H., Hong, S. M., Kim, S. J., Kim, B.C., Jin, C., Lim, C. J., Park, E. H. (2004). Anti-inflammatory effects of genipin, an active principle of gardenia. *European Journal of Pharmacology*, 495(2-3), 201-208.

Kumari, S., Chauhan, G. S. (2014). New Cellulose–Lysine Schiff-Base-Based Sensor–Adsorbent for Mercury Ions. *ACS Applied Materials & Interfaces*, 6, 5908-5917.

Lee, K. Y., Ha, W. S., Park, W. H. (1995). Blood compatibility and biodegradability of partially N-acylated chitosan derivatives. *Biomaterials*, 16, 1211-6.

Lee, M., Siu, R. K., Ting, K., Wu, B. M. (2010). Effect of Nell-1 delivery on chondrocyte proliferation and cartilaginous extracellular matrix deposition. *Tissue Engineering Part A*, 16, 1791-800.

Li, Y., Liu, C., Tan, Y., Xu, K., Lu, C., Wang, P. (2014). In situ hydrogel constructed by starch-based nanoparticles via a Schiff base reaction. *Carbohydrate Polymers*, 110, 87-94.

Lin, C. C., Anseth, K. (2009). PEG hydrogels for the controlled release of biomolecules in regenerative medicine. *Pharmaceutical Research*, 26(3), 631-643.

Lin, Y. H., Liang, H. F., Chung, C. K., Chen, M. C., Sung, H. W. (2005). Physically crosslinked alginate/N, O-carboxymethyl chitosan hydrogels with calcium for oral delivery of protein drugs. *Biomaterials*, 26, 2105-2113.

Lu, X. W., Liu, W., Wu, Z. Q., Xiong, X. H., Liu, Q., Zhan, W. J., Chen, H. (2016). Substrate-independent, Schiff base interactions to fabricate lysinefunctionalized surfaces with fibrinolytic activity. *Journal of Materials Chemistry B*, 4, 1458-1465.

Madeira, C., Santhagunam, A., Salgueiro, J. B., Cabral, J. M. (2015). Advanced cell therapies for articular cartilage regeneration, *Trends in Biotechnology*, 33, 35-42.

Murphy, L., Helmick, C. G. (2012). The impact of osteoarthritis in the United States: a population-health perspective. *American Journal of Nursing*, 112, S13.

Nandi, S. K., Kundu, B., Basu, D. (2013) Protein growth factors loaded highly porous chitosan scaffold: A comparison of bone healing properties, *Materials Science and Engineering: C*. 33, 1267-1275.

Park, H., Choi, B., Hu, J., Lee, M. (2013). Injectable chitosan hyaluronic acid hydrogels for cartilage tissue engineering. *Acta Biomaterialia*, 9, 4779-4786.

Riederer, M. S., Requist, B. D., Payne, K. A., Way, J. D., Krebs, M. D. (2016). Injectable and microporous scaffold of densely-packed, growthfactor-encapsulating chitosan microgels. *Carbohydrate Polymers*, 152, 792-801.

Rinaudo, M. (2006). Chitin and chitosan: properties and applications. *Progress in Polymer Science*, 31, 603-632.

Sakai, S., Yamada, Y., Zenke, T., Kawakami, K. (2009). Novel chitosan derivative soluble at neutral pH and in-situ gellable via peroxidase-catalyzed enzymatic reaction. *Journal of Materials Chemistry*, 19, 230-235.

Schatz, C., Pichot, C., Delair, T., Viton, C., Domard, A. (2003). Static light scattering studies on chitosan solutions: From macromolecular chains to colloidal dispersions. *Langmuir*, 19(23), 9896-9903.

Shi, J., Guobao, W., Chen, H., Xing, M. (2014). Schiff based injectable hydrogel for in situ pH-triggered delivery of doxorubicin for breast tumor treatment. *Polymer Chemistry*, 5, 6180-6189.

Sikkema, J., de Bont, J. A., Poolman, B. (1994). Interactions of cyclic hydrocarbons with biological membranes. *Journal of Biological Chemistry*, 269(11), 8022-8028.

Siu, R. K., Zara, J. N., Hou, Y., James, A. W., Kwak, J., Zhang, X. (2012). NELL-1 promotes cartilage rengeneration in an in vivo rabbit model. *Tissue Engineering Part A*, 18, 252-61.

Solorio, L., Zwolinski, C., Lund, A. W., Farrell, M. J., Stegemann, J. P. (2010). Gelatin microspheres crosslinked with genipin for local delivery of growth factors. *Journal of Tissue Engineering and Regenerative Medicine*, 4(7), 514-523.

Spiller, K. L., Maher, S. A., Lowman, A. M. (2011). Hydrogels for the repair of articular cartilage defects. *Tissue Engineering Part B: Reviews*, 17, 281-99.

Sridharan, B., Sharma, B., Detamore, M.S. (2015). A roadmap to commercialization of cartilage therapy in the United States of America. *Tissue engineering*. 22(1), 15-33.

Tomihata, K., Ikada, Y. (1997). In vitro and in vivo degradation of films of chitin and its deacetylated derivatives. *Biomaterials*, 18, 567-75.

Xu, Q., Mao, C., Liu, N. N., Zhu, J. J., Sheng, J. (2006).Direct electrochemistry of horseradish peroxidase based on biocompatible carboxymethyl chitosan-gold nanoparticle nanocomposite. *Biosensors and Bioelectronics*, 22, 768-773.

Yuan, Y., Chesnutt, B. M., Utturkar, G., Haggard, W. O., Yang, Y., Ong, J. L., Bumgardner, J. D. (2007). The effect of cross-linking of chitosan microspheres with genipin on protein release. *Carbohydrate Polymers*, 68(3), 561-567.

Zhang, Y., Zhu, W., Wang, B., Ding, J. (2005). A novel microgel and associated post-fabrication encapsulation technique of proteins. *Journal of Controlled Release*, 105(3), 260-268.

Zhu, J., Marchant, R. E. (2011). Design properties of hydrogel tissue-engineering scaffolds. *Expert Review of Medical Devices*, 8(5), 607-626.