Academic Journal of Engineering and Technology Science
ISSN 2616-5667 Vol.2, Issue 4: 53-60, DOI: 10.25236/AJETS.2019.020408

Preparation of Mitomycin-Modified Chitosan Sustained-Release Membrane Based on Grey Theory

Chao Lin, Daguan Ke*

Wenzhou Medical University, Wenzhou, Zhejiang, 325035, China
**Corresponding author*

ABSTRACT. *Chitosan is a kind of natural macromolecular polysaccharide with positive charge on glycosyl. Chitin is usually extracted from crustacean shell and formed by deacetylation of chitin. Chitosan is easier to be absorbed and utilized than chitin because it has removed the phthalide group from chitin through chemical treatment. Mitomycin-chitosan drug-loaded sustained-release membrane has good flexibility, permeability and hydrophilicity, and has certain drug sustained-release capability in vitro. Although the biological properties of chitosan are improved compared with chitin, chitosan can only be dissolved in weak acid solution, which greatly limits its application. In order to avoid propylation of the amino group of chitosan, chitin can be first reacted with epichlorohydrin and then deproteinized. This paper studies the properties of mitomycin-chitosan drug-loaded sustained-release membrane, evaluates its inhibitory effect on fibroblasts, biocompatibility and biodegradability in vitro experiments, and evaluates the feasibility of its application in the preparation of sustained-release membrane.*

KEYWORDS: *Chitosan, Mitomycin, Diaphragm, Sustained release*

1. Introduction

Chitin, also known as chitin, is a common polysaccharide and widely distributed on the earth. Chitin has been mainly composed of crustacean shells in nature, cell wall components of some plants and some fungi, which have high application value [1]. Chitosan is widely used in medicine, engineering and construction industries due to its excellent biocompatibility, environmental friendliness, degradability and component controllability [2]. Chitosan is mainly used to prepare sustained-release microspheres, film agents, tablets, etc. Chitosan is insoluble in water but only soluble in inorganic acid and some organic acid solutions, which limits its application [3]. The poor water solubility of chitosan may be due to the crystal structure of its molecule and the strong hydrogen bond between amino group and

light group. Compared with chitin, chitosan has milder physical properties and decomposes at about 185℃. As deacetylation destroys the regularity of chitin molecular internal structure, chitosan solubility is greatly improved and its chemical properties are more active [4]. Chitin is degraded and deacetylated under natural conditions to gradually form chitosan with different molecular weights and deacetylation degrees. Mainly exists in the laboratory, usually chitin is treated under strong alkaline conditions to hydrolyze it. The study of chitosan sustained-release system has improved the release time of the loaded reagents, especially in the field of medicine. Drug release and targeted delivery can be controlled through chitosan sustained-release membrane [5].

Chitosan is a polysaccharide composed of acetyl glucose and glucosamine. Its basic structural unit is glucosamine, which is a substance that is abundant in human body and plays a great physiological role. Chitosan and its derivatives have certain curative effects, and are called the sixth vital element necessary for human health after protein, fat, sugar, vitamins and minerals [7]. Under different reaction conditions, the fusization reaction can occur on the amino and hydroxyl groups of chitosan molecules to prepare derivatives with good water solubility. Chitosan molecule has -NH2 group and active hydroxyl group, and the substituent group can enter these two positions during the reaction to obtain the corresponding derivative [8]. Through the preparation of derivatives and optimization of controllable systems, the adsorption performance, degradability and slow release performance of chitosan are improved, and its application field and scope are widened [9]. Chitosan's strong hydrophilicity makes it unable to effectively control drug release, and can only improve drug release by increasing its dosage. Therefore, chitosan is rarely used alone as a sustained and controlled release matrix material. In order to avoid propylation of the amino group of chitosan, chitin can be first reacted with epichlorohydrin and then deproteinized [10]. There are some relations between the degree of substitution of light propyl chitosan and its biodegradability. When the degree of substitution of propyl chitosan increases, the biodegradation speed will also increase.

2. Application of Chitosan and Its Derivatives in Ophthalmology

Chitosan sustained-release microspheres are round in appearance, smooth in surface, well dispersed, and have good sustained-release effect. They are one of the research hotspots of sustained-release systems. Drug sustained-release carriers play an important role in maintaining blood drug concentration level, reducing drug administration times, reducing drug toxicity and improving drug efficacy. Chitosan and its derivatives have good biocompatibility, biodegradability and non-toxicity. Its metabolites are harmless to human body and can be absorbed by the body. Medical

ISSN 2616-5667 Vol.2, Issue 4: 53-60, DOI: 10.25236/AJETS.2019.020408

chitosan has no irritant reaction to cornea and conjunctiva, and can be used to make contact lens and contact lens cleaning solution, artificial tears, etc. Chitosan sustained-release gel refers to a three-dimensional network structure of chitosan that swells in water, retains a large amount of water but does not dissolve. Chitosan, as a natural biodegradable polymer carrier, has relatively complicated drug release process. While the drug is released from the chitosan skeleton by diffusion, the chitosan skeleton undergoes continuous molecular degradation [11]. Chitosan and its derivatives are ideal materials that can meet the above requirements. Chitosan can be crosslinked by light radiation or aldehyde crosslinking agents to make contact lenses. Changes in molecular structure directly affect the properties of molecules, so accurate determination of the degree of substitution is very important for analysis of product properties.

Chitosan is dissolved in acetic acid mixed solvent and evaporated on polyethylene film to obtain soft film, which can be used to make soft contact lens. The synthetic route of completely acylated chitosan is shown in Figure. 1.

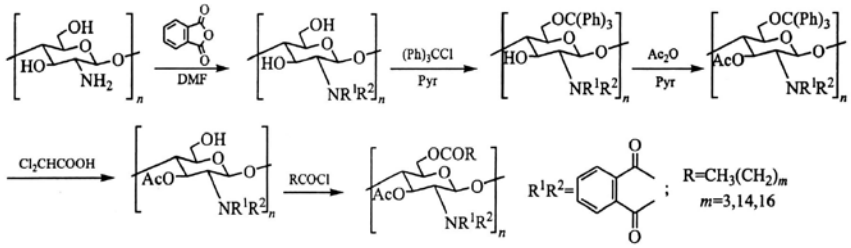

Figure. 1 Completely acylated chitosan synthesis route

The drug release rate of chitosan carrier system is related to the degradation rate. If the degradation mode of chitosan is bulk degradation, the drug release rate is independent of the surface volume ratio of the system. When the degradation of chitosan or drug diffusion are rate-limiting steps respectively, the drug release mechanisms of the drug-loaded system are the slow release mechanism of chitosan degradation and the slow release mechanism of drug-loaded diffusion respectively. Chitosan contact lens can also cure the wound on eyeball. It can be expected that the effect of chitosan contact lens on healing eyeball wound will be applied in practice when cornea is burned by radiation. The drug release rate is slow at the beginning of degradation, and increases with the dissolution of chitosan [12]. Chitosan, chitosan derivatives and their degradation products oligosaccharide and monosaccharide have different functions. As natural non-toxic molecules, different molecular weights can give them different physical and chemical properties and functions. Chitosan has good water absorption and film forming properties, and can form viscous macromolecular colloid solution when dissolved in water, thus prolonging the retention time of drugs in conjunctival sac and prolonging the curative effect. If the degradation mode of chitosan is surface degradation, the initial drug release rate is

affected by the ratio of surface area to volume of the system and the shape of the chitosan drug delivery system. Modification of some groups on glycosyl can obtain chitosan derivatives with good water solubility and excellent biological properties, greatly expanding the application range of such substances.

3. Preparation and Properties of Mitomycin-Chitosan Sustained Release Membrane

Chitosan is not soluble in water, but only in weak acids and some organic solvents, which greatly limits its application in production and life. As an important derivative of chitosan, light propyl chitosan not only has excellent properties such as biocompatibility, complete decomposability, multifunctional reactivity, stereo structure and chirality, reproducibility, etc., but also has many good special properties such as moisture absorption, moisture retention, foaming, emulsifying and film forming properties, physiological activity and functional properties. There are some differences in the actual slow release process due to the different forms of chitosan carriers. The water outside the chitosan particle contacts with the surface of the chitosan particle. Due to the hydrophilicity of the surface and the interior of the chitosan particle, the water gradually permeates into the interior of the particle from the surface of the chitosan drug-carrying particle, thus swelling the chitosan substrate. In vitro and in vivo degradation studies show that light propyl chitosan films with different degrees of substitution have better biodegradability and biocompatibility [13]. The poor water solubility of chitosan may be due to its crystal structure and relatively stable hydrogen bonds. The polymer substrate swells due to absorption of a large amount of water, and the free volume of the material changes, changing from a glassy state to a rubbery state, and the drug is inside the swollen rubbery substrate. By substituting two groups on chitosan molecules with different substitutes, various chitosan materials with different structures and functions can be obtained.

By strictly controlling the operation and environmental conditions during synthesis and purification, HEC and N-CMC with higher purification level were obtained. The quality index determination results of the samples are shown in Table 1.

Table 1 Sample quality index measurement results

Quality index	Ash content (%)	Rotational viscosity (mPa·s)	Computer rate(μs/cm)
N-CMC	7.312	72	387.7
HEC	3.719	15	68.3

Activation of important signal transduction pathways within the cell regulates changes in cellular function, which in turn triggers a signal cascade within the cell. Activation of mitogen-activated protein kinases and the like results in activation of nuclear transcription factors and ultimately promotes apoptosis. In order to make the calculation convenient, the logarithm of the above formula becomes:

$$W = \alpha(\beta(\frac{E^2_{i-current}}{E^2_{i-init}}) + (1-\beta)\frac{d_i}{d_{\max}})) \tag{1}$$

According to the characteristics of the chitosan sustained-release membrane itself and the assumption of independent and identical distribution, the above formula becomes:

$$E_{ch} = lE_{elec}(\frac{N}{k} - 1) + lE_{DA}\frac{N}{k} + lE_{elec} + l\xi_{amp}d^4_{toBS} \tag{2}$$

The mutual information that describes the correlation between two random variables is calculated by the following formula:

$$E[d^2_{toCH}] = \iint (x^2 + y^2)\rho(x,y)dxdy = \iint r^2\rho(r,\theta)drd\theta \tag{3}$$

The drug is controlled by the membrane and conforms to the diffusion mechanism of the drug from the swollen elastic substrate. Table 2 shows the inhibition rate of L929 fibroblast growth by two membranes.

Table 2 Inhibition rate of L929 fibroblast growth by two membranes

Time	24h		48h	
	OD492	Inhibition rate (%)	OD492	Inhibition rate (%)
Contrast	0.478	-	0.371	-
HEC	0.456	7.21	0.342	11.17
N-CMC	0.433	11.49	0.316	16.45

The output of a neuron is represented by a function, and the following functional expressions are generally used to represent the nonlinear characteristics of the network:

$$\mathrm{K_f^{\Phi}} = \mathrm{K_p^{\Phi}} = \frac{\mathrm{p_G^g p_H^h}}{\mathrm{p_D^d p_E^e}}\left(\mathrm{p^{\Phi}}\right)^{-\sum_{\mathrm{B}}\gamma_{\mathrm{B}}} \tag{4}$$

Wire splitting algorithm based on kinematic parameters can be adopted, and the wire splitting force of the method is expressed by the following formula:

$$K_b = \frac{R\left(T_b^*\right)^2}{\Delta_{vap}H_m\left(A\right)} \bullet M_A \tag{5}$$

Where P represents the force of the contraction unit and q represents the force of the parallel elastic unit, which is calculated by:

$$BH(p,q) = \sum_{u=1}^{n} \sqrt{p_u(f)q_u} \tag{6}$$

Where m is the intrinsic constant of mitosis, p and k are the length of the cleavage and the rate of contraction, respectively, and the iso-tension ej is calculated by:

$$e_j = -k\sum_{i=1}^{m}(p_{ij}\ln p_{ij}) \tag{7}$$

Drug absorption in tissues is largely determined by drug concentration, surface area, permeability and drug residence time. The chitosan sustained-release membrane treated by coagulation bath will not be dissolved by body fluid or swelled by water to form gel in vivo. Hydroxyethyl chitosan can be obtained by hydroxyethylation reaction between basic chitosan and ethylene oxide, while the same hydroxypropyl chitosan is prepared from basic chitosan and epichlorohydrin under the condition of strong alkali. It can directly react with ethylene oxide or propylene oxide in alkaline solution or in some organic solvents to obtain nitrogen and oxygen substituted chitosan derivatives. Chitosan itself is an alkaline polysaccharide with positive charge, which is conducive to its combination with cells and the structural reconstruction of membrane proteins, allowing drugs to pass through the cell bypass pathway. After alkalization, the normal crystalline region of chitosan is destroyed. When propane comes into contact with the free charge on the hydroxyl group, nucleophilic substitution reaction occurs to generate hydroxylated chitosan [. The way and route of metabolism of fusimethyl chitin in eyes are similar to that of sodium hyaluronate, and because it can return to normal in a short period of time, there is no obvious damage to the intraocular structure. Chitosan can prolong the residence time of drugs on the surface of eyeball and keep a high drug concentration on the corneal surface, thus improving the effective utilization rate of drugs.

4. Conclusions

The study of chitosan derivative sustained-release system is one of the research hotspots in related fields. With the research and development of new chitosan derivative sustained-release systems and modification methods, the application of chitosan derivative sustained-release systems will be more and more extensive. In this experiment, mitomycin-modified chitosan derivatives were used to make

membranes. By comparing their physical and chemical properties and biological properties, a more suitable scaffold material for drug sustained-release system was found. In order to overcome the shortcomings of poor water solubility of chitosan, chitosan is degraded into chitosan which has a small molecular weight and can be completely dissolved in water, thereby developing a chitosan carrier having a desired release effect. The membrane has different degrees of growth inhibition on fibroblasts, and the degree of inhibition increases with the increase of drug release rate. The membrane has good permeability and hydrophilicity, and has certain mechanical strength and good flexibility. The surface of the chitosan carrier is modified to have the desired selectivity for target cells, target tissues, and target organs. It has potential application prospects for the embedding and release of bioactive macromolecular drugs such as polypeptides, proteins, nucleic acids and vaccines.

References

[1] Tanaka N, Matsumoto I, Suzuki M, et al.(2015). Chitosan tubes can restore the function of resected phrenic nerves. Interactive CardioVascular and Thoracic Surgery, vol. 21, no.1, pp.8-13.

[2] Debarros M, Martin M(2015). Traumatic Diaphragm Injuries: A Comprehensive Review. Current Respiratory Medicine Reviews, vol.11, no.1, pp.55-64.

[3] Layachi L, Georges M, Gonzalez-Bermejo, Jésus, et al.(2015). Diaphragm pacing failure secondary to deteriorated chest wall mechanics: When a good diaphragm does not suffice to take a good breath in. Respiratory Medicine Case Reports, no. 15:20-23.

[4] Heunks L M A, Doorduin J, Van d H J G(2015). Monitoring and preventing diaphragm injury. Current Opinion in Critical Care, vol.21, no.1, pp.34-41.

[5] Tomaszewski M, Stepien K M, Tomaszewski A(2014). Diaphragm fibrillation - an incidental finding detected by transoesophageal echocardiography. Acta Cardiologica, vol.69, no.3, pp.338-340.

[6] Gao J, Kong J, Ding X M, et al.(2015). Laparoscopic vs computerized tomography-guided radiofrequency ablation for large hepatic hemangiomas abutting the diaphragm. World Journal of Gastroenterology, vol.21, no.19, pp.5941-5949.

[7] Liu B, Zhou H, Liu L, et al.(2018). An Optical Fiber Fabry-Perot Microphone Based on Corrugated Silver Diaphragm. IEEE Transactions on Instrumentation and Measurement, vol.8, no.8, pp. 1994 - 2000.

[8] Ewan C. GoligherFranco LaghiMichael E. Detsky, et al. (2015). Measuring diaphragm thickness with ultrasound in mechanically ventilated patients: feasibility, reproducibility and validity. Intensive Care Medicine, vol.41, no.4, pp.642-649.

[9] Vieira S P, Elena P, Albuquerque André Luis Pereira, et al.(2016). Identifying decreased diaphragmatic mobility and diaphragm thickening in interstitial lung disease: the utility of ultrasound imaging. Jornal Brasileiro de Pneumologia, vol.42, no.2, pp.88-94.

[10] Zhou H, Hu Z(2014). Man With Air Below the Right Diaphragm. Annals of Emergency Medicine, vol.63, no.1, pp.13-47.
[11] Shortt C M, Fredsted A, Chow H B, et al.(2014). Reactive oxygen species mediated diaphragm fatigue in a rat model of chronic intermittent hypoxia. Experimental Physiology, vol. 99, no.4, pp.688-700.
[12] Baria M R, Shahgholi L, Sorenson E J, et al.(2014). B-Mode Ultrasound Assessment of Diaphragm Structure and Function in Patients With COPD. Chest, vol.146, no.3, pp.680-685.
[13] Onders R P, Elmo M J, Kaplan C, et al.(2014). Extended use of diaphragm pacing in patients with unilateral or bilateral diaphragm dysfunction: A new therapeutic option. Surgery, vol. 156, no.4, pp.776-786.