Study on the preparation technology of Sanqi hemostatic spongia for percutaneous absorption

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Abstract. Objective: To optimize the preparation technology and matrix ratio of Sanqi hemostatic spongia. Methods: The effects of Twain 80, formaldehyde and twelve sodium alkyl sulfate on the matrix formability of the spongia were investigated by the L9 (3^3) orthogonal test, and the optimum ratio of the matrix was optimized through the water absorption, the thickness uniformity, the softness and the color degree. The orthogonal test results showed that the matrix ratio was Twain 80 1.4g, formaldehyde 1.6 mL, sodium dodecyl sulfate 1.0 g, and Sanqi hemostatic spongia was superior when the proportion of other substrates was fixed. Conclusion: Sanqi hemostatic spongia was produced by orthogonal test and matrix ratio is stable, uniform in thickness and good in quality.

1. Introduction
Traditional Chinese medicine spongia is a transdermal drug system (TDDS) [1]-[2]. It is a kind of sponge solid sterilization preparation with strong water absorption, which is made of hydrophilic colloid solution and medicine through foaming, hardening, freezing and drying [3]. It is easy to use, high safety, especially suitable for patients with massive hemorrhage. However, there is no relevant report about Sanqi hemostatic spongia, so the research group chose to study it to provide basis for clinical medication.

2. Methods and results

2.1. Extraction and separation of dencichine
50 g of crude powder of Panax notoginseng was extracted twice with 10 times of methanol at room temperature, combine the extract and store it. The residue was dried and extracted with 10 times of water for three times, then the residue was filtered to get a green extract, which was concentrated to a certain amount. The concentration of alcohol was adjusted to 75% with 95% ethanol. Filter out the precipitate, and the filtrate was extracted with equal volume n-butanol. The purified solution was obtained by combining the aqueous phase, and the purified solution was passed through cation
exchange resin. First wash the purified solution to the effluent to colorless, then eluted with 0.1mol/L ammonia water, collected in stages. The collected solution is developed under n-butanol: glacial acetic acid: anhydrous ethanol: water (4:1:1:2). Ninhydrin solution is used as the developer on the silica gel G thin-layer plate for identification, and the collected solution with only one purple red spot is combined and the same height as the control of dencichine, which proves that this fraction is dencichine extract [4].

2.2. Orthogonal test design.
On the basis of pre-test, L₉ (3³) test is used to design the level table of orthogonal factors. The levels and factors of orthogonal test were shown Table 1.

Table 1. Factors and levels of orthogonal test

| Levels | Twain 80 (g) | Formaldehyde (mL) | Sodium dodecyl sulfate (g) |
|--------|-------------|-------------------|---------------------------|
| 1      | 1.4         | 1.2               | 0.6                       |
| 2      | 1.6         | 1.4               | 0.8                       |
| 3      | 1.8         | 1.6               | 1.0                       |

2.3. Orthogonal test procedure
Take 12g gelatin, soak it in distilled water for 1h, heat it in water bath to make it completely dissolved, add a proper amount of Tween 80, formaldehyde and sodium dodecyl sulfate (according to the prescription designed in the orthogonal test design table) under high-speed stirring, continuously stir it to produce a large number of uniform and delicate bubbles, then separately pack it in the mold, put it in the refrigerator at -20 °C for 24h, take it out and dry it in the freeze dryer for 3 days, Take it out and put it into the dryer for storage [5].

2.4. Scoring standards [3] and results of orthogonal test

Table 2. Quality evaluation standard of Sanqi hemostatic spongia

| Assessment items   | Quality evaluation standard                                      | Score |
|--------------------|-----------------------------------------------------------------|-------|
| Thickness uniformity | The more uniform the thickness, the higher the score              | 25    |
| Softness           | The better the rebound after extrusion, the higher the score     | 25    |
| Water absorption   | The greater the water absorption, the higher the score            | 25    |
| Color              | The more uniform the color, the higher the score                  | 25    |
| Total score        |                                                                  | 100   |
Table 3. Results of orthogonal test

| Serial number | A  | B  | C  | Total score |
|---------------|----|----|----|-------------|
| 1             | 1.4| 1.2| 0.6| 75          |
| 2             | 1.4| 1.4| 0.8| 78          |
| 3             | 1.4| 1.6| 1  | 94          |
| 4             | 1.6| 1.2| 0.8| 70          |
| 5             | 1.6| 1.4| 1  | 88          |
| 6             | 1.6| 1.6| 0.6| 87          |
| 7             | 1.8| 1.2| 1  | 86          |
| 8             | 1.8| 1.4| 0.6| 79          |
| 9             | 1.8| 1.6| 0.8| 84          |

\[ G=741 \\
CT=61009 \]

2.5. Analysis of variance results

Table 4. Analysis of variance results

| Source of variance | Sum of squares from mean deviation | Degree of freedom | Variance | F     | P     |
|--------------------|-----------------------------------|------------------|----------|-------|-------|
| A                  | 2.67                              | 2                | 1.335    |       |       |
| B                  | 194.67                            | 2                | 97.335   | 72.91 | <0.05 |
| C                  | 234.00                            | 2                | 117.00   | 87.64 | <0.05 |
| Error(e=A)         |                                   |                  |          |       |       |

Note: \( F_{0.05}(2, 2) =19.00 \quad F_{0.01}(2, 2) =99.00 \)

The results of orthogonal test and analysis of variance show that the amount of factor B and C, that is, formaldehyde and sodium dodecyl sulfate, has a significant effect on the forming of sponge agent, and factor A, that is, the amount of Tween 80, has no significant effect on the forming of sponge agent. Based on cost saving, the level of factor A is selected, which not only saves cost but does not affect the forming of sponge agent, and coincides with the results of the third group with the highest score. To sum up, the best horizontal combination of factors is \( A_1B_3C_3 \), that is, Tween 80 1.4g, formaldehyde 1.6g mL, sodium dodecyl sulfate 1.0g.

3. Verification test

Take 12g of gelatin and soak it in distilled water for 1h, then heat it in water bath to make it completely dissolved, take 2ml of Sanqi extract and add it into gelatin solution, and add appropriate amount of auxiliary materials with the best ratio under high-speed agitation for three batches of repeated verification test. The results show that the optimal ratio is reasonable and feasible, and the thickness uniformity, softness, water absorption, color of the prepared Sanqi hemostatic spongia is the best.

4. Conclusion

Foaming is the key operation to ensure the quality of sponge agent, which directly affects the appearance, looseness, water absorption and softness of the finished product. Formaldehyde solution is
As a curing agent, in addition to its dosage should be strictly controlled during preparation, and attention should be paid to slowly add formaldehyde solution during high-speed mixing, which can make gelatin crosslinked and solidified evenly. Stirring speed and stirring time have influence on foaming performance, cooling temperature and cooling time have certain influence on curing, so it is also important to optimize stirring speed, stirring time, cooling temperature, cooling time and other process parameters. In general, the solidified foam should be cooled rapidly at about 18 °C below zero, and must be frozen thoroughly for at least 24 hours, in order to ensure that the gelatin network structure is fixed and will not be deformed during the drying process.

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References
[1] Li CJ, Xiao XC, Huang XJ, et al. Progress in percutaneous absorption preparation [J]. Technological Development of Enterprise, 2009, 28 (12): 24-25, 28.
[2] Zhang ZW. Monograph on traditional Chinese medicine pharmaceutics [M]. Beijing. People's Health Publishing House,2009:329-330.
[3] Jiang PC, Li J, Jiang FD, et al. Optimization of preparation technology of sponge [J]. China Pharmacist, 2010,13 (5): 658-659.
[4] Xie GX, Qiu MF, Zhao AH, et al. Separation, purification and structural analysis of dencichine from Panax notoginseng [J]. Nat Prod Res Dev, 2007,19 (6):1059-1061.
[5] Zhang LJ. Study on the therapeutic material basis of Rehmannia Glutinosa Libosch and the best partation technology of foam-rubber [D]. Shaanxi University of Chinese Medicine, 2011.