Determination of sodium and potassium in Dan Hong injection by atomic absorption spectrometry

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Abstract: To establish and validate the determination method of the content of sodium ion and potassium ion in Dan Hong injection by atomic absorption spectrometry (AAS). [Methods] Microwave digestion system was applied to deteriorate samples, and the content of sodium ion and potassium ion was determined by AAS. [Results] The calibration curve of sodium was liner (r=0.9998) in the range from 0.1to0.6mg/L. The average recovery was 101.1% with RSD 1.8%. The limit of detection was 5.3μg/L. The calibration curve of potassium was liner (r=0.9998) in the range from 0.1 to 0.8mg/L. The average recovery was 99.8%with RSD 2.3%. The limit of detection was 7.7μg/L. The determination results of 10 batches Dan Hong injection sample showed that the content of sodium ion was 562.3 to 734.7 mg/L and potassium ion was 206.9 to 314.4mg/L. [Conclusion] The method is simple and accurate and can be used for determination of the contents of sodiumion and potassium ion in Dan Hong injection. The accurate quantitative determination method can be promoted to quality control of Dan Hong injection

Keywords: Dan Hong injection, sodium ion, potassium ion, atomic absorption spectrometry

1. Introduction

To establish a method for determining the content of sodium (Na) and potassium (K) ions in Dan Hong injection as stipulated by the "Pharmacopoeia of the People's Republic of China" 2010 edition, which requires the limit check for K ions in the quality inspection items of intravenous injections. Sodium (Na) and potassium (K), as essential mineral elements for the human body, regulate the osmotic pressure and acid-base balance between cells and blood. Both excessively high or low levels of blood K can cause dysfunction of nerves and muscles, while abnormal levels of blood Na can lead to brain cell dehydration or edema, endangering life. Therefore, it is necessary to conduct a limit check on Na and K ions in intravenous injections. Nie Li Xing and Ning Hong Xin have used flame atomic absorption spectrophotometry to directly determine the content of K ions in Hong Hua injection and Na, K, and magnesium (Mg) ions in compound electrolyte injections. This article uses flame atomic absorption spectrophotometry to establish a method for determining the content of Na and K ions in Dan Hong injection, which is characterized by being fast, accurate, and reproducible.

2. Instruments and reagents

TAS-990 Atomic Absorption Spectrophotometer (Beijing Pu Sai General Instrument Co., Ltd.); Sodium hollow cathode lamp, Potassium hollow cathode lamp (Beijing Shu Guang Ming Electronic Light Source Instrument Co., Ltd.); Flame atomizer (Beijing Pu Sai General Instrument Co., Ltd.).

Reagents: Sodium single-element standard solution [National Center for Analysis and Testing of Nonferrous Metals and Electronic Materials, unique identifier 11042, concentration 1000 mg/L, medium 5% hydrochloric acid (HCl)]; Potassium single-element standard solution (National Center for Analysis and Testing of Nonferrous Metals and Electronic Materials, unique identifier 12303-2, concentration 1000 mg/L, medium H_2O); Cesium chloride (Tianjin Jinke Fine Chemical Research Institute, batch number 20090825);

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Ultra-pure water (self-made by Milli-Q water purification system); Nitric acid (super pure, Beijing Chemical Factory). The Dan Hong injection used in the experiment is a commercial product, with a specification of 10 mL per vial, manufactured by Heze Bu Chang Pharmaceutical Co., Ltd., with 10 batch sample numbers respectively as 14031035 (B1), 14041013 (B2), 14041035 (B3), 14051008 (B4), 14051016 (B5), 14061001 (B6), 14071047 (B7), 14081003 (B8), 14081005 (B9), 14081007 (B10).

3. Methods and results

3.1. Operating conditions of the atomic absorption spectrophotometer

Sodium (Na) Ions: Measurement wavelength: 589.0 nm, Sodium spectral band width: 2 nm, Lamp current: 2 mA, Negative high voltage (-V): 300, Sodium gas flow rate: 1.1 L/min, Burner height: 5.0 mm; using an air-acetylene flame. Potassium (K) Ions: Measurement wavelength: 766.9 nm, Potassium spectral band width: 2 nm, Lamp current: 2 mA, Negative high voltage (-V): 300, Potassium gas flow rate: 1.7 L/min, Burner height: 5.0 mm; using an air-acetylene flame.

3.2. Solution preparation

3.2.1. Preparation of blank solution

Weigh 1 gram of cesium chloride and place it into a 100 mL volumetric flask. Add water to dissolve it and then make up to the mark to serve as an electrolyte. Take 1 mL of this solution and place it into a 10 mL volumetric flask, then dilute to the mark with ultra-pure water, mix well, and you will obtain the desired solution.

3.2.2. Preparation of the test solution

Take 0.2 mL of the Dan Shen injection sample and place it in a digestion vessel. Add 4 mL of concentrated nitric acid and digest in a microwave digestion apparatus for 10 minutes at 200°C. After cooling, check the solution to ensure it is clear and transparent with no undissolved particles. Heat the digested liquid to near dryness, then add 10 mL of ultra-pure water to dilute it, transfer to a 50 mL volumetric flask, rinse the digestion vessel with ultra-pure water and gradually transfer the rinse to the volumetric flask, add dilution liquid to the mark, and mix well to serve as the test sample stock solution. Take 3.125 mL of the test sample stock solution and place it in a 50 mL volumetric flask, add 5 mL of electrolyte, then dilute to the mark with ultra-pure water, mix well, to serve as the sodium ion test sample solution. Take 2 mL of the test sample stock solution and place it in a 10 mL volumetric flask, add 1 mL of electrolyte, then dilute to the mark with ultra-pure water, mix well, to serve as the potassium ion test sample solution.

3.2.3. Preparation of the reference solution

Accurately measure 0.5 mL of the standard solutions of sodium and potassium ions (1000 mg/L) separately and place them into a 50 mL volumetric flask. Dilute to the mark with ultra-pure water and mix well, to obtain standard working solutions of sodium and potassium ions with a concentration of 10 μ g/ml.

3.3. Methodological validation

3.3.1. Construction of the standard curve

Sodium Ions: Accurately measure a certain amount of the sodium ion standard working solution mentioned in section "2.2.3" and dilute to solutions with concentrations of 0.1, 0.2, 0.4, 0.5, and 0.6 mg/L to serve as the working curve solutions. Determine according to the method, using absorbance Y as the vertical axis and concentration X (mg/L) as the horizontal axis to plot the standard curve. The regression equation obtained is: $A = 0.6243C + 0.0037$, with a correlation coefficient $r = 0.9998$, indicating that sodium ions have a good linear relationship with absorbance in the range of 0.1 to 0.6 mg/L.

Potassium Ions: Accurately measure a certain amount of the potassium ion standard working solution mentioned in section "2.2.3" and dilute to solutions with concentrations of 0.1, 0.2, 0.4, 0.6, and 0.8 mg/L to serve as the working curve solutions. Determine according to the method, using absorbance Y as the vertical axis and concentration X (mg/L) as the horizontal axis to plot the standard curve. The regression equation

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obtained is: A = 0.4348C + 0.0072, with a correlation coefficient r = 0.9998, indicating that potassium ions have a good linear relationship with absorbance in the range of 0.1 to 0.8 mg/L.

3.3.2. Determination of the limit of detection (LOD)

Sodium Ions: Take the blank solution and continuously measure the absorbance 11 times to calculate its standard deviation. Calculate the limit of detection (LOD) according to the following formula: LOD (mg/L) = 3 * SA / S, where SA is the standard deviation of the blank solution, and S is the slope of the standard working curve. The results show the absorbance values are: 0.015, 0.014, 0.013, 0.015, 0.014, 0.012, 0.012, 0.014, 0.015, 0.014, and 0.014, with a standard deviation of 0.0011. Based on the slope from the linear regression equation, the lowest detectable concentration of sodium ions is calculated to be 5.3 μ g/L.

Potassium Ions: Following the method described above, the results show the absorbance values are: 0.011, 0.010, 0.010, 0.008, 0.008, 0.008, 0.008, 0.008, 0.008, 0.008, and 0.008, with a standard deviation of 0.0011. Based on the slope of the linear regression equation, the lowest detectable concentration of potassium ions is calculated to be 7.7 μ g/L.

3.3.3. Precision experiment

Sodium Ions: Take a 0.4 mg/L sodium ion reference solution and analyze it continuously 7 times. The absorbance values measured are: 0.267, 0.273, 0.271, 0.272, 0.269, 0.266, and 0.269. The average value is 0.270, with a Relative Standard Deviation (RSD) of 1.0%.

Potassium Ions: Take a 0.2 mg/L potassium ion reference solution and directly inject it for analysis, continuously 7 times. The absorbance values measured are: 0.100, 0.101, 0.101, 0.101, 0.102, 0.102, and 0.101. The average value is 0.101, with a Relative Standard Deviation (RSD) of 0.7%.

3.3.4. Reproducibility experiment

Following the preparation method described in section "2.2.2", six batches of Dan Shen injection test solution were prepared in parallel. The content of sodium and potassium ions in the test solutions was measured separately. The average content of sodium ions was 816.7 mg/L, with a Relative Standard Deviation (RSD) of 1.0%; the average content of potassium ions was 270.2 mg/L, with an RSD of 0.7%. The results of the study indicate that the method has good reproducibility for determining the content of sodium and potassium ions in Dan Shen injection.

3.3.5. Spike recovery experiment

Precisely take a known amount of Dan Shen injection samples and accurately add a certain amount of sodium ion reference solution. Operate according to the method described in section "2.2.2" and measure. The average spike recovery rate is 101.1%, with a Relative Standard Deviation (RSD) of 1.8%. Following the same procedure, measure the average recovery rate for potassium ions, which is 99.8%, with an RSD of 2.3%. The results can be seen in Table 1.

3.3.6. Stability experiment for sodium ions:

Take one portion of the test solution and store it at room temperature. Measure its absorbance at 0, 1, 2, 4, and 6 hours, which are 0.129, 0.131, 0.131, 0.130, and 0.132, respectively. The average value is 0.131, with an RSD of 0.9%, indicating that the test solution is stable when stored at room temperature for 6 hours.

Potassium Ions: Take one portion of the test solution and store it at room temperature. Measure its absorbance at 0, 1, 2, 4, and 6 hours, which are 0.102, 0.102, 0.102, 0.102, and 0.101, respectively. The average value is 0.102, with a Relative Standard Deviation (RSD) of 0.4%, indicating that the test solution is stable when stored at room temperature for 6 hours.

3.4. Sample content determination

Using the established methods for the determination of sodium and potassium ions as described above, 10 batches of Dan Shen injection were tested. The results show the content of sodium ions (mg/L) to be: 664.0 (Batch B1), 697.9 (B2), 734.7 (B3), 562.3 (B4), 573.6 (B5), 658.4 (B6), 712.1 (B7), 618.8 (B8), 596.2 (B9), 616.0 (B10); the content of potassium ions (mg/L) is: 245.0 (B1), 270.0 (B2), 270.0 (B3), 266.9 (B4), 258.8 (B5), 271.9 (B6), 314.4 (B7), 236.3 (B8), 206.9 (B9), 221.3 (B10).

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Samples were digested using a microwave digestion system, and a method for determining the content of sodium and potassium ions in Dan Shen injection was established using flame atomic absorption spectrophotometry. A systematic methodological examination was conducted, and the content was measured for 10 batches of Dan Shen injection samples. The results of the measurements indicate that there is a certain variation in the concentration of sodium ions, which ranges from 562.3 to 734.7 mg/L, and potassium ions, which ranges from 206.9 to 314.4 mg/L, among different batches of Dan Shen injection, The reasons may be due to the impact of raw medicinal materials or the manufacturing process of the production companies on the sodium and potassium ions in Dan Shen injection, which requires further research.

Tab.1 Results of average recovery of Na^+ and K^+ in Dan Hong injection

ID	Na^+				K^+			
	Addition (μ g)	Content (μ g)	Measured (μ g)	Recovery Rate (%)	Addition (μ g)	Content (μ g)	Measured (μ g)	Recovery Rate (%)
1	80.00	81.67	160.8	98.9	25.00	27.02	51.25	96.9
2	80.00	81.67	164.0	102.9	25.00	27.02	51.25	96.9
3	80.00	81.67	163.2	101.9	25.00	27.02	52.25	100.9
4	80.00	81.67	160.8	98.9	25.00	27.02	52.50	101.9
5	80.00	81.67	164.0	102.9	25.00	27.02	52.50	101.9
6	80.00	81.67	162.4	100.9	25.00	27.02	52.00	99.9
Average (%)				101.1				99.8
RSD (%)				1.8				2.3

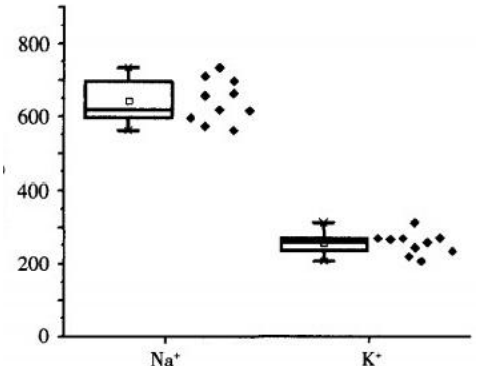

Fig.1 The contents of Na^+ and K^+ in 10 batches Dan Hong injection

Based on the average solid content of Dan Shen injection, which is approximately 30g/L, it can be calculated that the content of sodium ions accounts for 1.9% to 2.4% of the total solid content in Dan Shen injection, and the content of potassium ions accounts for 0.7% to 1.0% of the total solid content. The total amount of sodium and potassium ions accounts for 2.6% to 3.4% of the total solid content in Dan Shen injection. The detection method for sodium and potassium ions established in this experiment is simple and fast, with accurate and reliable results and good repeatability, making it suitable for the determination of sodium and potassium ion content in Dan Shen injection. It can be used for quality control research of Dan Shen injection.

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4. Discussion

4.1. Selection of masking agents and sample treatment methods

The use of cesium chloride, which has a lower ionization potential, as an ionization suppressor [9-12], eliminates ionization interference and also enhances measurement sensitivity. The lowest detectable concentrations are 5.3 μ g/L for sodium ions and 7.7 μ g/L for potassium ions. In sample processing, microwave digestion technology is employed, which has the advantages of rapid heating of samples, high temperature rise, strong digestion capability, significantly reduced dissolution time, no environmental pollution, and reduced energy waste [13-15]. Since the Dan Shen injection samples are inherently aqueous solutions, direct dilution methods for sample processing result in low recovery rates, which do not meet the requirements of methodological research. This indicates that the sample matrix interferes with the determination of sodium and potassium ions. Therefore, microwave digestion is used for sample treatment, and the matrix after digestion causes minimal interference with the absorption of sodium and potassium ions, allowing for accurate determination of their content in Dan Shen injection.

4.2. Limit requirements

According to the research requirements of the "Related Substances Test Method for Injections" in the appendix of the 2010 edition of the "Chinese Pharmacopoeia" [1], Part I: The limit check for potassium ions in the test solution is carried out using the colorimetric method. It is required that the color of the test solution should not be darker than that of the potassium ion solution (100 mg/L). Therefore, it is calculated that the potassium ion content in the injection should not exceed 1000 mg/L. Through the measurement of 10 batches of Dan Shen injection samples, the actual measured values did not exceed the specified range and complied with the relevant regulations of the pharmacopoeia. The average solid content of Dan Shen injection is about 30 g/L. It is calculated that the content of sodium ions accounts for 1.9% to 2.4% of the total solid content in Dan Shen injection, and the content of potassium ions accounts for 0.7% to 1.0% of the total solid content. The total amount of sodium and potassium ions accounts for 2.6% to 3.4% of the total solid content in Dan Shen injection, providing experimental data to guide the safe use of the drug in clinical practice.

5. Conclusion

In summary, the content of sodium (Na^+) and potassium (K^+) ions in Dan Shen injection was accurately determined using flame atomic absorption spectrophotometry, and the method was systematically examined to verify its accuracy, repeatability, stability, and applicability. The experimental results show that there are certain differences in the content of Na^+ and K^+ ions in Dan Shen injection from different batches, which may be related to the raw medicinal materials or the preparation process of the manufacturing companies. The use of microwave digestion technology to treat samples effectively eliminated the interference of the sample matrix, improving the sensitivity and accuracy of the measurements. In addition, the experiment also determined the lowest detectable concentrations of Na^+ and K^+ ions in Dan Shen injection, providing important experimental data support for the safe use of the drug in clinical practice.

6. References

[1] National Pharmacopoeia Commission, ed. Chinese Pharmacopoeia (Part I) [S]. Beijing: Chemical Industry Press, 2010: Appendix IXS.

[2] Dai Shuchun, Song Jun, Zhou Hua sheng, et al. Interference and Elimination of Potassium and Sodium in Special Medical Formula Foods Measured by Flame Atomic Absorption Method [J]. Food Industry Technology, 2014, 35(9): 303-306.

[3] Liu Xiu Shu, Du Qian, Yin Shi Qiang. Two cases of hyperkalemia caused by traditional Chinese medicine in patients with chronic renal failure [J]. Tianjin Pharmacy, 2015, 27(2): 55-56.

[4] Tong Dan Hong, Chen Quan Yong, Wu Xiaole. Comparative Analysis of Potassium and Sodium Ion Levels in Arterial and Venous Blood [J]. Chinese Rural Medicine, 2014, 21(13): 54-55.

https://doi.org/10.62852/csa/2024/62

[5] Shi Wei, L v Pengfei, Liu Ran, et al. The Impact of Traditional Chinese Medicine Compound on Serum Potassium Concentration in the Treatment of Chronic Renal Failure [J]. China Pharmacy, 2015, 24(8): 37-39.
[6] Zhu Ping Xian, Li Ming. Research Progress on the Impact of Potassium Metabolism on Cardiac Arrhythmias [J]. Cardiovascular Disease Prevention and Treatment Knowledge (Academic Edition), 2011, 4(2): 84-85.
[7] Nie Li Xing, Jin Hongyu, Lin Rui chao, et al. Determination of Potassium Content in Carthamus Tinctorius L. Injection by Atomic Absorption Spectrophotometry [J]. Journal of Pharmaceutical Analysis, 2010, 30(3): 468-470.
[8] Ning Hong Xin, Wang Wei, Yang Ying, et al. Determination of Sodium, Potassium, and Magnesium Ions in Compound Electrolyte Injection by Flame Atomic Absorption Spectrophotometry [J]. Drug Evaluation Research, 2011, 34(4): 267-269.
[9] Ren Liping, Deng Lijuan, Fan Hui Hong. Determination of Sodium and Potassium Ions in Calf Blood Protein Extract Concentrate by Flame Atomic Absorption Method [J]. Journal of Pharmaceutical Analysis, 2014, 34(4): 636-638.
[10] Tang Wei, Sun Xiaoyi. Determination of Potassium in Precipitation by Flame Atomic Absorption Method Using Ionization Suppressor [J]. Heilongjiang Environmental Bulletin, 2002, 26(4): 89-90.
[11] Jiang Yucheng, Hu Man Cheng, Gao Shi yang, et al. Atomic Absorption Analysis of Alkali Metal Impurities in High Purity Cesium Chloride [J]. Laboratory of Analysis and Test, 2004, 23(3): 77-79.
[12] Liu Dong, Zhang Chang, Yang Shi Kun. Method Study on Determination of Trace Sodium and Potassium Ions in Calcium Stearate by Atomic Absorption Method [J]. Journal of Southwest University for Nationalities (Natural Science Edition), 2002, 28(2): 234-235.
[13] Zhang Guo Yue, Song Li, Fan Bao Juan, et al. Investigation of Test Conditions for Determination of Chromium in Gelatin Capsules by Microwave Digestion-Atomic Absorption Spectrophotometry [J]. Journal of Pharmaceutical Analysis, 2012, 32(8): 1328-1330.
[14] Tan He ping, L v Hao, Gao Yang, et al. Application of Microwave Digestion in the Analysis of Rare Earth Elements and Heavy Metals in Tea and Soil [J]. China Testing, 2010, 36(2): 37-40.
[15] Miao Lixin. Determination of Zinc in Fortified Foods by Microwave Digestion-Flame Atomic Absorption Spectrometry [J]. Henan Preventive Medicine, 2015, 26(3): 187-188.

https://doi.org/10.62852/csa/2024/62