

STABILITY STUDIES OF EXTEMPORANEOUS ZINC SULFATE INJECTIONS FOR HOSPITAL USE

Piyawat Chaivichacharn¹, Papitchaya Bunditritidej¹,
Pittha Phannasorn¹, Watcharaphong Chaemsawang¹, Sompol Prakongpan¹,
Putthiporn Khongkaew^{1,*}

Faculty of Pharmaceutical Science, Burapha University, 169 Long-Hard Bangsaen Road, Saen Sook Sub-district,
Mueang District, Chonburi 20131

*Corresponding author: E-mail: putthiporn@go.buu.ac.th

Abstract: Zinc is an important trace mineral that people need to stay healthy. Zinc sulfate injection is used as a supplement to intravenous solutions for patients who cannot take zinc orally. In Thailand, zinc sulfate injection is not commercially available, instead being aseptically prepared in extemporaneous preparation in individual hospitals. However, the stability of zinc sulfate injection was not studied. The objective of this study was to evaluate the stability of zinc sulfate injections prepared from different vehicles (sterile water and pH 2-3 sterile water adjusted with sulfuric acid) and sterilization methods (moisture sterilization and sterile by filtration). It was found that zinc sulfate injection had the greatest solubility in acidic condition and zinc sulfate injection 1 mg/mL was not precipitate at pH 2-9. The stability of zinc sulfate 6 formulas that have been stored at $40\pm 2^\circ\text{C}$, 75% \pm 5%RH for 3 months by the standard analytical method described in USP 38, the results show that content of zinc did not change and still be in the standard level of 90.0-110.0% labeled amount. The stability study of zinc sulfate injection in various IV admixtures indicated that the percentage of zinc content did not decrease after mixing for 24 hours. In conclusion, the different vehicles and sterilization methods have no effect on content of zinc when storage in 3 months. Sterile water was recommended as vehicle and moisture sterilization as the sterilization method, because it is convenient and cost saving.

Keywords: zinc sulfate injection, stability test, complexometric titration

บทคัดย่อ: สังกะสี (Zinc) เป็นแร่ธาตุที่มีความสำคัญต่อร่างกายของมนุษย์ โดยยาฉีดสังกะสีเฟดนำมาใช้เสริมสภาวะการขาดสังกะสีในผู้ป่วยที่ไม่สามารถรับประทานได้ โดยให้ผ่านหลอดเลือดดำ ซึ่งในประเทศไทยยังไม่มียาฉีดสังกะสีเฟดจำหน่ายในรูปแบบผลิตภัณฑ์ ทำให้แต่ละโรงพยาบาลต้องมีการเตรียมยาฉีดสังกะสีเฟดขึ้นมาใช้เอง (extemporaneous) ด้วยเทคนิคไร้เชื้อ อย่างไรก็ตามยังไม่มีการศึกษาความคงตัวของยาฉีดสังกะสีเฟดที่เตรียมขึ้นมา ทำให้ไม่ทราบวันหมดอายุที่แน่นอน ในการศึกษาครั้งนี้จึงมีวัตถุประสงค์ เพื่อศึกษาความคงตัวของยาฉีดสังกะสีเฟดที่เตรียมขึ้นจากน้ำกระสายยาต่างชนิด (sterile water และ sterile water ที่ปรับ pH 2-3 ด้วย sulfuric acid) และศึกษาผลของการทำไร้เชื้อ (moisture sterilization และ sterile by filtration), ผลการทดลองพบว่า ซิงค์ซัลเฟตมีค่าการละลายที่ดีในสภาวะกรด โดยพบว่าที่ pH 2-9 ยาฉีดสังกะสีเฟด ความแรง เมก./มล. ไม่มีการตกตะกอน และเมื่อทำการศึกษาความคงตัวของตำรับเมื่อเก็บที่สภาวะ $40\pm 2^\circ\text{C}$, 75% \pm 5%RH เป็นเวลา 3 เดือน ตามวิธีวิเคราะห์มาตรฐาน USP 38 พบว่าสูตรตำรับทั้ง 6 สูตร มีความคงตัว โดยปริมาณซิงค์ซัลเฟต มีปริมาณอยู่ในช่วง 90-110% หลังจากเก็บไว้เป็นระยะเวลาอย่างน้อย 3 เดือน ในการศึกษาความคงตัวของยาฉีดสังกะสีเฟดเมื่อผสมกับสารอาหารทางหลอดเลือดชนิดต่างๆ พบว่าหลังผสม 24 ชั่วโมง ปริมาณซิงค์ไม่เกิดการเปลี่ยนแปลงและไม่เกิดการตกตะกอนให้เห็น โดยสรุป พบว่ากระบวนการทำไร้เชื้อและชนิดน้ำกระสายยาไม่มีผลต่อความคงตัวของยาฉีดสังกะสีเฟดเมื่อทำการเก็บรักษาเป็นเวลา 3 เดือน ซึ่งจากผลการศึกษาสามารถแนะนำโรงพยาบาลให้ใช้กระบวนการทำไร้เชื้อด้วยวิธีความร้อนชื้นเนื่องจากเป็นวิธีที่สะดวกและมีประสิทธิภาพรวมถึงประหยัดค่าใช้จ่ายอีกด้วย

คำสำคัญ: ยาฉีดสังกะสีเฟด, การศึกษาความคงตัว, การวิเคราะห์ปริมาณโดยปฏิกิริยาการเกิดสารประกอบเชิงซ้อน

INTRODUCTION

Zinc is an important trace mineral that people need to stay healthy. The World Health Organisation's (WHO) Model List of Essential Medicines recommends the use of zinc sulfate in paediatric diarrhoea treatment. In Thailand, zinc sulfate injections are not commercially available. Therefore, Zinc sulfate injection forms have to be prepared aseptically in extemporaneous conditions by Thai technicians in individual hospitals. Few research studies exist that focus on the stability of zinc sulfate injections. The existence of such studies would make it a waste of time to study zinc sulfate and the cost of preparation. Therefore, the purpose of this study is to evaluate the stability of zinc sulfate injections using different vehicles. The sterile water is a typical approach used by hospitals in Thailand for the preparation of zinc sulfate injections. For comparison with sterile water, of which pH 2-3 is adjusted by sulfuric acid, the formula is recommended by the WHO and found in zinc sulfate injections distributed abroad.

MATERIALS AND METHODS

Material

Zinc sulfate heptahydrate (Loba chemie), Ethylenediaminetetraacetic acid (EDTA) (Loba chemie), Eriochrome black TS (Loba chemie), Calcium carbonate, Diluted hydrochloric acid, Sodium hydroxide, Ammonium chloride, Ammonium hydroxide, Sterile water for injection, Hydroxy naphthol blue indicator, Normal saline solution (NSS) (Thai-Otsuka), 5% Dextrose in 0.45% Sodium chloride (D51/2S) (Thai-Otsuka), 5% Dextrose in water (D5W) (Thai-Otsuka) and Total Parenteral Nutrition (TPN) solution (Thai-Otsuka)

Limits of solubility for Zinc sulfate in different pH

Different pH solutions were prepared by adjusting the pH 2-12 of sterile water with hydrochloric acid (HCl) and sodium hydroxide (NaOH). One ml of pipetted solution was placed in a test tube, then 10 mg of zinc sulfate was added and swirled until zinc sulfate dissolved. Zinc sulfate was added until it could no longer dissolve. Total amount of zinc sulfate in mg in each pH of solutions was then calculated.

Stability test for zinc sulfate concentration of 1 mg/mL in pH 2-9

Sterile water was adjusted to pH 2-9 with hydrochloric acid (HCl) and sodium hydroxide (NaOH). Zinc sulfate was then added to the solution to make a final concentration of 1 mg/ml. The samples were stored in $40\pm 2^{\circ}\text{C}$ 75%RH \pm 5%RH for 1 month, after which they were analysed by a Nano-sizer for detection of particles.

Study of the type of vehicles and sterilization methods associated with the stability of zinc sulfate injections

The zinc sulfate solution was prepared in a concentration of 1 mg/ml in 2 types of vehicles, including 1) sterile water for injection (following hospital formulation) and 2) sterile water for injection adjusted to pH 2-4 with sulfuric acid (following original formulation). The solutions were divided by sterilisation with 2 sterilisation methods, autoclave and membrane filtration. The finished products were stored in $40\pm 2^{\circ}\text{C}$ 75%RH \pm 5%RH for 3 months. Percentage content of zinc sulfate was analysed with complexometric titration following USP 38. The zinc sulfate injection was diluted in 100 ml of purified water and added to an ammonia-ammonium chloride buffer of TS 5 ml and eriochrome black TS 0.1 ml. The solution was titrated with 0.05 M edetate disodium (EDTA) VS until the colour changed to deep blue.

Stability test of zinc sulfate injection in 4 different IV admixtures

The zinc sulfate injection was mixed in IV admixtures (D5W, D51/2S, NSS and TPN) to a final concentration of 2 mg/100ml. The solutions were kept at room temperature for 24 hours, after which the solutions were analysed by complexometric titration to determine the physical properties and the percentage of zinc sulfate content.

RESULTS AND DISCUSSION**Limits of solubility for Zinc sulfate in different pH**

The maximum solubility for zinc sulfate was pH 2-3. The dissolution values for zinc sulfate solutions reduced continuously until the pH values increased up to pH11. Consequently, the solutions could not be dissolved.

Stability test of zinc sulfate concentration 1 mg/mL in pH 2-9

Zinc sulfate injection was prepared in a concentration of 1 mg/mL in pH 2-12. Basically, injection solutions must be filtrated through membranes with porous holes of size 0.22 microns in order to remove particles. Thus, this study measured the sizes of particles after storage. Particles larger than 0.22 microns were expected to be found after 1 month of storage at $40\pm 2^{\circ}\text{C}$, $75\%\pm 5\%\text{RH}$. The results revealed that no particles larger than 0.22 micron were found when measured by a nanosizer.

Study of the type of vehicles and sterilisation methods associated with the stability of zinc sulfate injections

The content of zinc did not change from the standard level between 90.0-110.0% of the labelled amount followed by USP 38. The results show that type of vehicle and sterilization method did not affect the content of zinc.

Stability testing of zinc sulfate injections in 4 different solutions

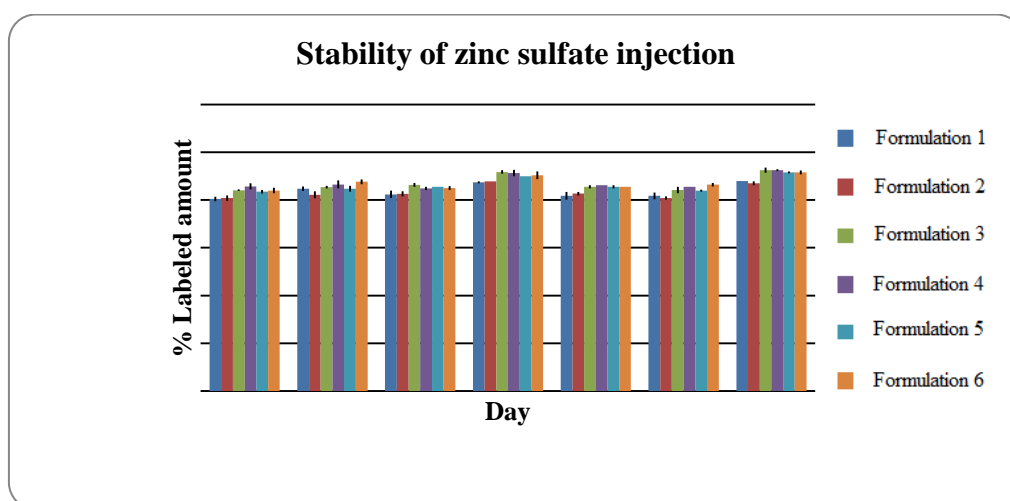
The mixing of zinc sulfate injections with D5W, D51/2S and NSS found that the solutions were physically stable and the percentage content of zinc sulfate did not change after being mixed for 24 hours. The amount of zinc sulfate in the TPN solutions could not be analyzed with the titrate method because TPN components were mixed solutions of various electrolytes, distributing the complexation between zinc and EDTA, which causes analysis inaccuracy. Only physical stability could be evaluated. The results showed that no physical change occurred when TPN solutions were mixed with prepared zinc sulfate.

Table 1. The amount of zinc sulfate dissolved in sterile water with different pH.

pH	Average (g/mL)
2	2.17±0.25
3	2.04±0.16
4	1.95±0.14
5	1.90±0.14
6	1.92±0.10
7	1.98±0.01
8	1.83±0.31
9	1.87±0.25
10	1.03±1.44
11	N/A
12	N/A

Table 2. Percentage content of zinc after being mixed in an IV solution for 24 hours.

	Zinc (%Content) n=3
NSS	101.37 ± 0.92
D5 $\frac{1}{2}$ S	101.10 ± 0.80
D5W	101.64 ± 0.46

**Figure 1.** Labelled amount of zinc sulfate injection in 6 formulations for Day 0, Day 30, Day 45, Day 60, Day 75 and Day 90, with results showing that the content of zinc did not change and remained in the standard level between 90.0-110.0 % of the labelled amount.

- Formulation 1: Zinc sulfate in sterile water sterilised by moisture sterilization
- Formulation 2: Zinc sulfate in sterile water sterilised by filtration
- Formulation 3: Zinc sulfate in sterile water pH 2-3 sterilised by moisture sterilization
- Formulation 4: Zinc sulfate in sterile water pH 2-3 sterilised by filtration
- Formulation 5: Zinc sulfate in sterile water non-sterilised
- Formulation 6: Zinc sulfate in sterile water pH 2-3 non-sterilised

CONCLUSION

Different vehicles and sterilization methods have no significant effect on the zinc content over 3 months. The suitable formula recommended is sterile water for injection as the vehicle and moisture sterilization as the sterilization method. This formula appears to be the most convenient and cost-effective. However, this experiment was conducted over a span of only three months. Therefore, date of expiry should be determined by not over 3 months. For periods longer than 3 months, further studies should be conducted in the future.

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