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Stability of Tetracycline-Methanol Solution under Different Storage Conditions

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Abstract

As one of the commonest antibiotics, investigating the impact of different storage conditions on tetracycline-methanol solution can provide theoretic foundation for its better preservation. This paper mainly discussed the effects of different ambient temperatures (-20°C, 25°C) and various initial concentrations (10 mg/L, 50 mg/L, 100 mg/L) on the stability of tetracycline-methanol solution. High-performance liquid chromatography-UV detection (HPLC-UV) was used to detect changes in the concentration of tetracycline-methanol solution at different time. The experimental results show that the stability of the tetracycline-methanol solution was closely related to the initial concentration and storage temperature. Low temperature and high initial concentration were beneficial to preserve the tetracycline-methanol solution.

Keywords

Tetracycline; Initial concentration; Temperature.

1. Introduction

High performance liquid chromatography (HPLC) is mainly used to efficiently convert organic compounds into organic compounds by means of separation and analysis by chemical ways [1]. Instrumental analysis methods for the detection of tetracycline residues are HPLC, GC and LC/MS, but HPLC is the most used method, which is a more reliable method [2].

Tetracycline antibiotics are broad-spectrum antibiotics produced by actinobacteria or semi-synthetic, with phenanthrene parent core. Due to the low price, tetracycline antibiotics are widely used not only as a drug for the treatment of human and animal diseases, but also as an additive to improve feed utilization efficiency and promote animal growth. However, its stability and persistence results in the residues of tetracycline antibiotics in the environment, the presence of tetracycline antibiotics is currently detected in soil, surface water and groundwater. Studies have shown that long-term consumption of foods containing antibiotics can do harm to human health, mainly damage organs such as stomach, intestines, liver, etc., detriment normal physiological functions, and lead to the emergence of drug-resistant pathogens [3-5]. The tetracycline detected in the environment poses a potential threat to people's health and even lives. As another typical tetracycline antibiotics, oxytetracycline antibiotics has also been proved its stability is related to the concentration and temperature [6]. The study of the stability of tetracycline will give us a way to preserve the tetracycline solution properly and also provide a method to degrade tetracycline in the environment.

As for methanol is one of the universally-used solvents for tetracycline as well as for dissolution, extraction and elution in many solute analysis work, this study aims to research the stability of tetracycline solution preservation. Therefore, it is significant to study the stability of tetracycline-methanol solution under different conditions.

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2. Materials and methods

2.1 Chemicals and materials

Tetracycline (powder, purity >99%) was purchased from Solarbio, methanol and acetonitrile are HPLC grade, oxalic acid was analytical reagent grade, and experimental water was ultrapure water, supplied by Molgenepure ultrapure water system. The tetracycline powders was accurately weighed by an electronic balance and then dissolved in methanol to prepare three groups of different concentrations (10 mg/L, 50 mg/L and 100 mg/L), each of which was 50 mL and preserved in brown glass bottles. One group was stored at -20 ° C (I) and the other group was stored at 25 ° C (II).

2.2 High performance liquid chromatography detection

Tetracycline-methanol solution was analysed by Flexar high performance liquid chromatography coupled with UV detector (PerkinElmer, USA). Separation was completed by Brownlee analysis C18 column (5 μm × 4.6 mm × 250 mm, PerkinElmer, USA). Isocratic elution was performed using methanol-acetonitrile-0.01 M oxalic acid (8:16:76, v/v/v) as the mobile phase. The injection volume was 20 μL and flow rate was 1 mL / min. Detection wavelength was 268 nm

2.3 Standard curve

A total of 1.00 mg tetracycline powder was dissolved in 10 mL of methanol and 100 mg/L mother liquor was obtained. Dilute the solution with methanol to 2 mg/L, 2.5 mg/L, 5 mg/L, 10 mg/L, 16 mg/L, 20 mg/L, 50 mg/L, and 100 mg/L which were used to draw a standard curve.

3. Results and discussions

3.1 Chromatogram of tetracycline-methanol solution

As shown in the Figure 1, methanol-acetonitrile-oxalic acid (8:16:76) was used as the mobile phase for the determination of HPLC-UV. The peak of tetracycline was good and appeared at about 8.30 min.

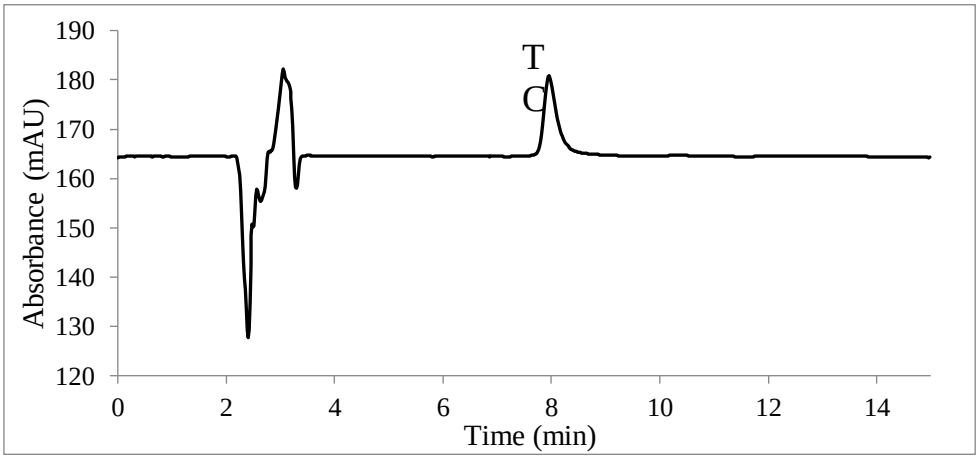

Figure 1. Chromatogram of 10 mg/L tetracycline-methanol solution at the first determination

3.2 Effect of concentrations on stability of tetracycline-methanol solution

As shown in the Figure 2, the concentration of tetracycline-methanol solution in 10 mg/L group finally reduced to 7.93 mg/L, and the changes are not obvious; the concentration in 50 mg/L group finally decreased to 44.33 mg/L; the fluctuation of 100 mg/L group was within 2 mg/L. Overall, under the same temperature (-20 °C), the tetracycline-methanol solution with a lower concentration (10 mg/L) was slightly less stable. In contrast, higher concentration (100 mg/L) of tetracycline-methanol solution has better stability.

At 25 °C, the 10 mg/L tetracycline-methanol solution reduced to 6.78 mg/L, and the 50 mg/L declined to 24.18 mg/L. While the concentration of tetracycline-methanol solution decreased to 47.73 mg/L

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from 100 mg/L, a 52% reduction. In conclusion, under the same temperature (25 °C), lower concentration (10 mg/L) of tetracycline-methanol solution has better stability.

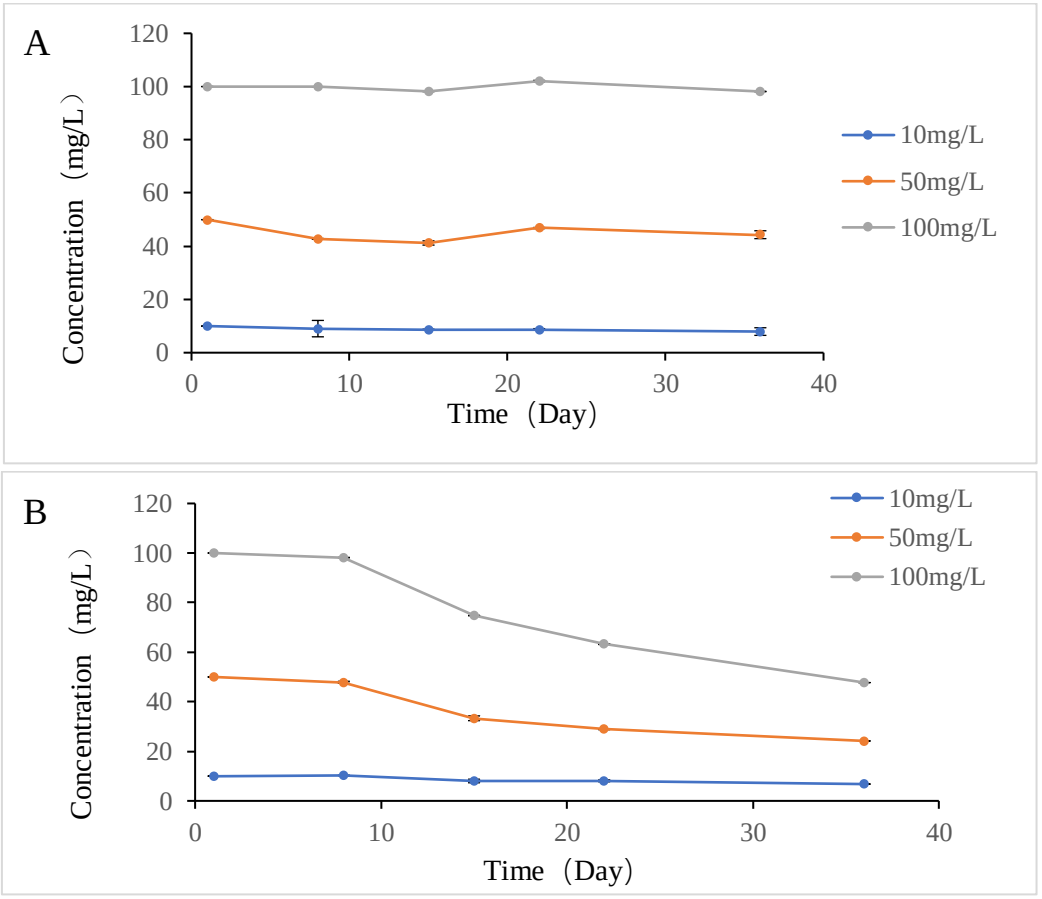
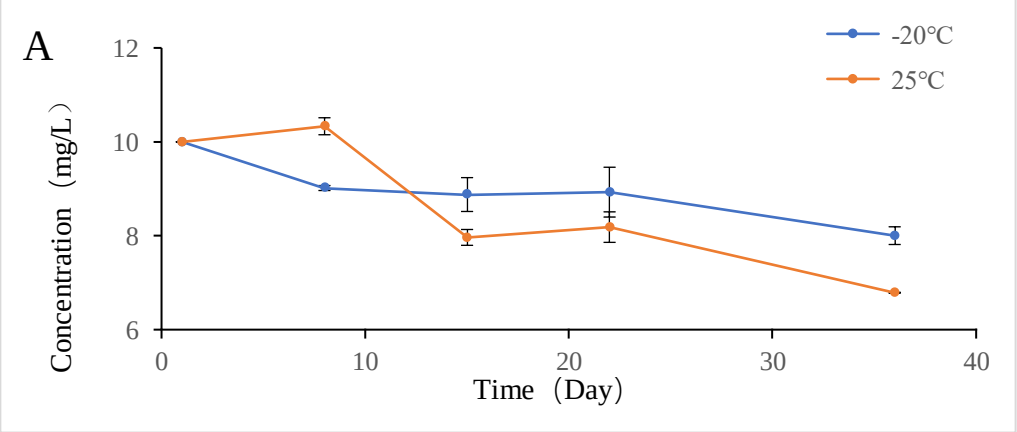


Figure 2. Changes in concentrations of tetracycline-methanol solution at different initial concentrations (A. Temperature at -20 °C, B. Temperature at 25 °C)

3.3 Effect of temperature on stability of tetracycline-methanol solution

In this study, the results illustrate that the stability of tetracycline-methanol solution is better when the temperature is lower (-20 °C) compared with higher temperature (25 °C) under the same initial concentration. The concentration of 10 mg/L tetracycline-methanol solution declined to 8.00 mg/L at -20 °C, 6.78 mg/L at 25 °C. In the 50 mg/L group, the concentration of tetracycline-methanol solution reduced to 45.40 mg/L at -20 °C while 24.25 mg/L at 25 °C. In the ending of 100mg/L group, the concentration in the cooler environment was higher (97.13 mg/L) than that at 25 °C which was 47.80 mg/L (Figure 3).



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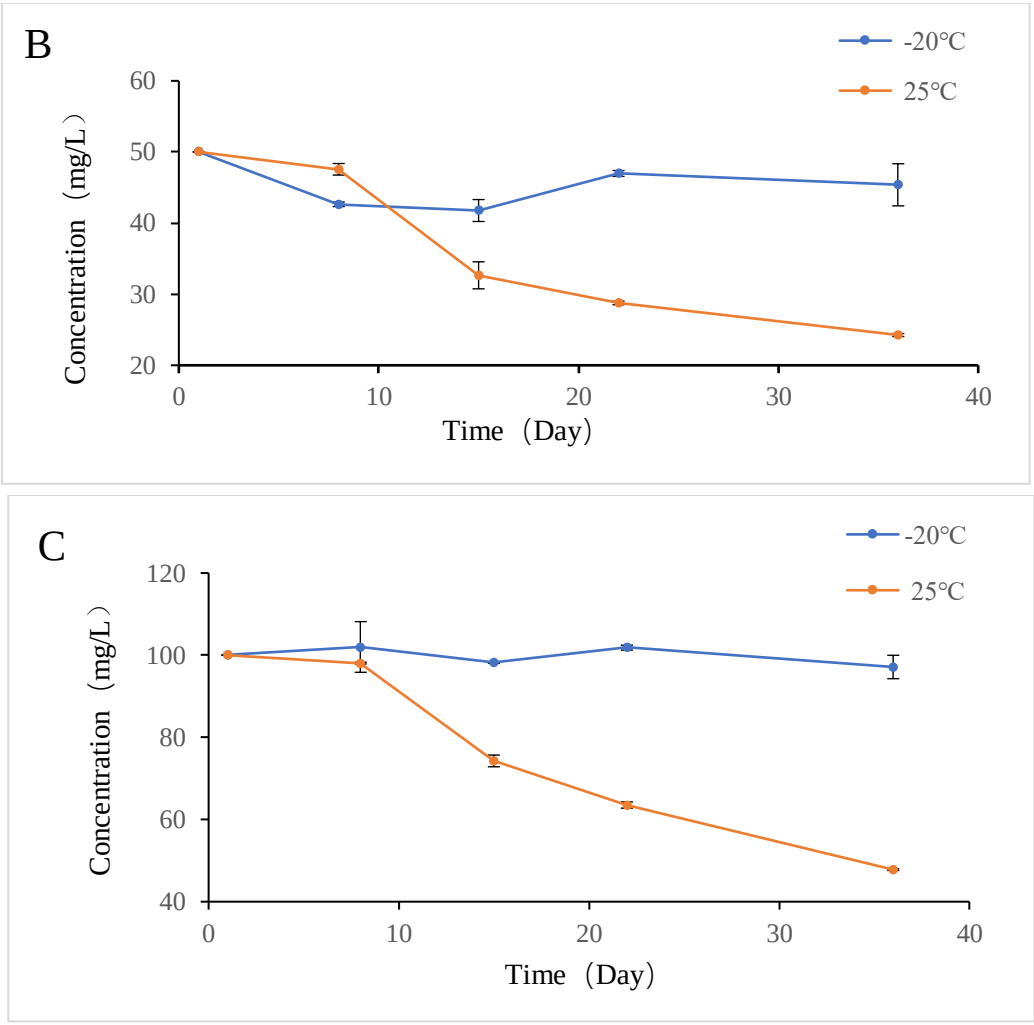

Figure 3. Changes in concentrations of tetracycline-methanol solution at different temperatures with the same initial concentration (A. Concentration of 10 mg/L, B. Concentration of 50 mg/L, C. Concentration of 100 mg/L)

4. Conclusion

On the basis of the experiment, the stability of the tetracycline-methanol solution is closely related to the storage temperature and initial concentration. Low temperature and high initial concentration benefit to maintain the stability of tetracycline-methanol solution. However, further studies are necessary to conduct to find out better conditions to preserve tetracycline-methanol solution and improve degradation rate of tetracycline in the environment.

Acknowledgments

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