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Investigation of the Stability of Acetylsalicylic Acid Solution at Different Temperatures

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Abstract

This study investigated the stability of acetylsalicylic acid (ASA) in aqueous citric acid solutions at various temperatures (21°C, 37°C, 45°C, and 60°C). The degradation of ASA was monitored using a titrimetric method to quantify its hydrolysis product, salicylic acid. Kinetic analysis based on the Arrhenius equation was performed to evaluate the stability profile of ASA under different storage conditions. The findings indicate that hydrolysis occurs even at room temperature, leading to a significant decrease in ASA concentration over time. The activation energy was calculated as 7.48 kcal, suggesting a rapid degradation process. The results highlight the instability of ASA in aqueous formulations, making it unsuitable for liquid dosage forms. To improve stability and prevent hydrolysis, alternative solvents such as propylene glycol and polyethylene glycol may be used instead of water. Additionally, microencapsulation techniques can offer a protective barrier against degradation, ensuring extended shelf life and improved pharmaceutical efficacy. These findings provide crucial insights for the formulation of stable liquid aspirin preparations and emphasize the necessity of selecting appropriate solvents and excipients in pharmaceutical development.

Keywords

Acetylsalicylic acid, arrhenius equation, citric acid, hydrolysis, stability, pharmaceutical formulation.

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INTRODUCTION

The formulation of pharmaceutical products must ensure that all characteristics comply with predefined specifications and remain within acceptable limits throughout their entire lifecycle, from manufacturing to patient use, without undergoing significant changes. Therefore, when developing a pharmaceutical product, careful consideration is given not only to the active ingredient(s) but also to all excipients and even the packaging materials to maximize stability over time. The designed dosage form should maintain the integrity of the formulation, enhance the stability of the active ingredient(s), and minimize potential interactions with other active or excipient components. Ensuring long-term stability is a critical aspect of pharmaceutical development, as it directly affects the efficacy, safety, and overall quality of the final product (González et al., 2022; Chinchole et al., 2014).

Acetylsalicylic acid, widely recognized as aspirin, is a crucial substance in pharmaceutical science due to its multifaceted therapeutic effects, including analgesic, antipyretic, anti-inflammatory, and antithrombotic properties. Its mechanism of action involves irreversible inhibition of cyclooxygenase (COX-1 and COX-2) enzymes, leading to the suppression of prostaglandin synthesis, which plays a key role in pain, fever, and inflammation. Aspirin is extensively prescribed for the treatment of various inflammatory conditions such as rheumatoid arthritis and osteoarthritis, as well as for symptomatic relief of mild to moderate pain and fever. Beyond its conventional uses, aspirin's role in cardiovascular health is well established, particularly in the prevention of thrombosis, myocardial infarction, and ischemic stroke. Recent researches also suggest its potential application in cancer chemoprevention and neurodegenerative diseases. Despite its broad clinical utility, aspirin's stability in solution remains a critical challenge, as hydrolysis leads to the formation of salicylic acid, affecting its efficacy and safety in pharmaceutical formulations (Dominiak et al., 2022; Merimi et al., 2023).

The stability of pharmaceutical compounds in solution is a critical factor affecting their efficacy, safety, and shelf life. Chemical degradation in aqueous environments can lead to loss of therapeutic activity and the formation of potentially harmful by-products. Factors such as temperature, pH, light exposure, and oxidative conditions significantly affect the stability of active pharmaceutical ingredients (APIs). Hydrolysis, oxidation, and

photodegradation are among the most common degradation pathways in solution. Hydrolysis, especially in ester- and amide-containing drugs such as acetylsalicylic acid, results from interaction with water and leads to the degradation of the parent compound to hydrolyzed derivatives. Oxidative degradation, usually catalyzed by dissolved oxygen or transition metals, can generate reactive oxygen species that accelerate the degradation of sensitive functional groups. Understanding these mechanisms and designing solutions of active substances, especially acetylsalicylic acid, as stable pharmaceutical formulations is important to optimize storage conditions and ensure consistent therapeutic performance (Kowalska et al., 2022).

Citric acid, a widely utilized weak organic acid in pharmaceutical formulations, serves as an effective buffering agent in aqueous solutions. Its primary function is to regulate and stabilize pH levels, which is crucial for preserving the integrity of pH-sensitive APIs. In solution, citric acid establishes an equilibrium with its conjugate bases (citrate ions), forming a buffer system that mitigates drastic fluctuations in pH. This buffering capability helps to reduce hydrolytic degradation, oxidative breakdown, and other pH-dependent chemical reactions that may compromise drug stability. Additionally, citric acid acts as a chelating agent by binding metal ions that can otherwise catalyze oxidative degradation. Through these combined effects, sodium citrate, a sodium salt of citric acid, can contribute to the enhanced stability of pharmaceuticals such as acetylsalicylic acid, potentially slowing its hydrolysis into salicylic acid and acetic acid, thereby extending its shelf life and therapeutic efficacy (Ciaramitaro et al., 2023).

This study focuses on evaluating the stability of acetylsalicylic acid in an aqueous sodium citrate solution under various storage temperatures. By analyzing the degradation kinetics at 21°C, 37°C, 45°C, and 60°C, the study aims to quantify the hydrolysis rate and identify the temperature at which acetylsalicylic acid demonstrates the highest instability. The results will offer valuable insights into the impact of temperature on the degradation process of acetylsalicylic acid and assess the potential role of citric acid as a stabilizing agent in liquid pharmaceutical formulations.

MATERIALS AND METHODS

Materials

Pharmaceutical-grade acetylsalicylic acid, sodium citrate, sodium hydroxide, and phenolphthalein were obtained from Sigma-Aldrich®. Purified water was used for the preparation of all solutions to ensure consistency and quality in the experimental procedures.

Methods

Preparation of acetylsalicylic acid solution

A solution containing 3.6 g of acetylsalicylic acid and 10.8 g of sodium citrate was prepared in 200 mL of purified water under continuous stirring. The mixture was stirred at room temperature using a magnetic stirrer until complete dissolution was achieved, ensuring homogeneity and stability for further analysis.

Determination of acetylsalicylic acid by titrimetric method

For the quantification of acetylsalicylic acid, 10 mL of the prepared solution was taken, and 2–3 drops of phenolphthalein indicator were added. The solution was then titrated with 0.1N NaOH solution until a persistent color change was observed, remaining stable for at least 1 minute, indicating the endpoint of the titration (Khouri et al.,2024).

The concentration of acetylsalicylic acid in the solution was calculated using the following formula.

$$Remining\ acetylsalicylic\ acid\ concentration = (2A - B)x100/A$$

A: The amount (ml) of 0.1 N NaOH consumed when acetylsalicylic acid is 100% present in the medium before hydrolysis starts.

B: It is the amount (ml) of 0.1N NaOH that neutralizes the hydrolyzed and unhydrolyzed acetylsalicylic acid in the sample after a certain time.

Stability studies

The prepared acetylsalicylic acid solution was divided into four separate beakers, each containing 50 mL of solution, and stored at 21°C (room temperature), 37°C, 45°C, and 60°C. To determine the initial concentration of acetylsalicylic acid before degradation, a sample was analyzed with titrimetric method immediately after preparation (at zero time). Subsequently, at 30, 60, 90, and 120 minutes, aliquots were taken from the solutions stored at each temperature, and the acetylsalicylic acid concentration was determined. The method allowed the assessment of the remaining stable acetylsalicylic acid concentration over time at different storage temperatures (Yenda et al., 2023).

RESULTS AND DISCUSSION

Determination of acetylsalicylic acid by titrimetric method

The concentrations (%) of acetylsalicylic acid remaining without degradation in solutions held at four different temperatures for up to 120 minutes, along with the natural logarithm (ln) values of these concentrations, are shown in Table 1.

Table 1: The concentrations of acetylsalicylic acid remaining without degradation in solutions at different temperatures.

	21 °C			37 °C			45 °C			60 °C		
t (min)	B (mL)*	C (%)*	In C	B (mL)*	C (%)*	In C	B (mL)*	C (%)*	In C	B (mL)*	C (%)*	In C
0	9.5	100	4.6	9.5	100	4.6	9.5	100	4.6	9.5	100	4.6
30	9.6	98.95	4.59	9.6	98.95	4.59	10	94.74	4.55	10.1	93.68	4.54
60	9.9	95.79	4.56	9.8	96.84	4.57	10.8	86.32	4.46	11.5	78.95	4.37
90	10.1	93.68	4.54	10.2	92.63	4.53	11	84.21	4.43	11.7	76.84	4.34
120	10.3	91.58	4.52	10.5	89.47	4.49	11.5	78.95	4.37	12.5	68.42	4.23

*B: The amount (ml) of 0.1N NaOH that

Determination of degradation equations for each temperature condition

The graphs of time (t) values as independent variables (x) and ln concentration values as dependent variables (y), were plotted as shown in Table 1. The slope (m), intercept (n), and determination coefficient (r^2) of the resulting lines were calculated. Since four different neutralizes the hydrolyzed and unhydrolyzed acetylsalicylic acid in the sample; C: The concentrations (%) of acetylsalicylic acid remaining without degradation in solutions. Temperatures were used, four separate lines were obtained. These equations are shown in Table 2 and Table 3 according to temperatures.

Table 2: First-order degradation kinetics equations at different temperatures.

Temperature (°C)	Degredation Equation
21	$y= 4.604-7.10^{-4} x$, r= -0.9922
37	$y= 4.612-9,33.10^{-4} x$, r= -0.971
45	$y=4.598-1,93.10^{-3} x$, r= -0.9906
60	$y=4.604- 3,13. 10^{-3} x$, r= -0.9815

Degradation kinetics and Arrhenius Equation

The relationship between reaction rate and temperature is defined by the Arrhenius equation below. According to Table 3, the activation energy (Ea) was calculated from the slope of the line that was calculated, and the frequency factor, which gives the frequency of molecules colliding with each other, was calculated from the intersection value. According to Table 3, the equation y = -5.3911 + 3,765.71x was obtained. Here, the Ea/R value, where R is a gas constant, can be read as 3,765.71.

Table 3: Details of the degradation equations with respect to temperatures.

Temperature °C	Temperature °K	1/T (x)	Slope (k)	In k (y)
21	294	$3{,}4.10^{-3}$	7.10^{-4}	7.26
37	310	$3{,}23.10^{-3}$	$9.3.10^{-4}$	6.98
45	318	$3{,}14.10^{-3}$	$1.9.10^{-3}$	6.25
60	333	3.10^{-3}	$3.1.10^{-3}$	5.77

CONCLUSION

The Arrhenius equation plays a crucial role in stability studies, as it allows for the prediction of the degradation rate of pharmaceutical compounds at different temperatures. By establishing a relationship between the reaction rate constant (k) and temperature (T), the equation helps to estimate a drug's shelf life and determine optimal storage conditions. In stability studies, accelerated stability testing is conducted at elevated temperatures, and the degradation rates are extrapolated to predict long-term stability at room temperature. This approach significantly reduces the time required for stability assessments while ensuring the reliability and safety of pharmaceutical products over their intended shelf life.

In the study, based on the values in Table 3, the degradation reaction was determined using the Arrhenius equation as: $y = -5.3911+3{,}765.71x$.

In this equation, the Ea/R value was found to be 3,765.71, and the activation energy (Ea) was calculated as 7.48 Kcal. It is well-known that if the activation energy of a degradation reaction is below 10 Kcal, the degradation process occurs easily. The prepared acetylsalicylic acid solution was found to be unstable at all studied temperatures (21°C, 37°C, 45°C, and 60°C), including room temperature, indicating a stability issue.

To enhance the stability of acetylsalicylic acid solutions and prevent hydrolysis, alternative solvents such as propylene glycol and polyethylene glycol can be used instead of water. Additionally, the preparation of microcapsules containing the active ingredient can create a protective barrier against degradation, making this approach a viable option for formulation development.

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