The Melting Point Phase Diagram of a Co-Crystal Between L-Proline and Ezetimibe

Wajeeha Raqeeb
Florida State University Panama City

Abstract

This study examined the melting point phase diagram of an L-Proline and Ezetimibe co-crystal as an alternative to differential scanning calorimetry (DSC). Mixtures in a 1:1 molar ratio were crystallized in methanol and analyzed using a Vernier melt station. Melting points from 11 mole fractions indicated co-crystallization at a 0.5 ratio, aligning with the expected 1:1 stoichiometry. While broader melting ranges were anticipated, the data did not clearly show this. The results support using melting point analysis for co-crystal characterization, emphasizing the need for precise sample preparation and measurement in future studies.

Introduction

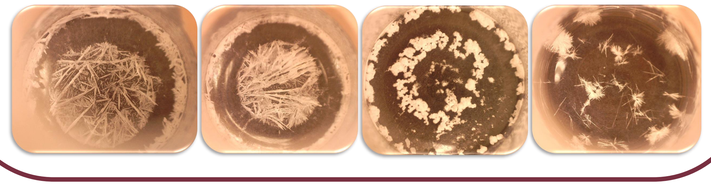
Co-crystals are of significant interest to the pharmaceutical industry and researchers because they can improve the properties of active pharmaceutical ingredients (API), such as solubility, stability, and bioavailability (Bag et al., 2023). This study focuses on the co-crystallization of L-Proline and Ezetimibe to determine whether a melting point phase diagram can indicate the formation of a cocrystal. L-Proline is a naturally occurring amino acid that plays a role in protein synthesis and structural stability, while Ezetimibe is a cholesterol-lowering drug that inhibits intestinal cholesterol absorption (Shimpi et al., 2014). The melting point phase diagram was determined from solids resulting from solutions with varied mole fractions of L-Proline and Ezetimibe to assess whether the cocrystal formation would be evident from the data. This method provides an alternative to Differential Scanning Calorimetry (DSC) and allows for a better interpretation of the results.

I hypothesize that by using the melting point phase diagram, we will be able to determine whether co-crystallization has occurred based on distinct melting point patterns, which will indicate the formation of a co-crystal.

Methods

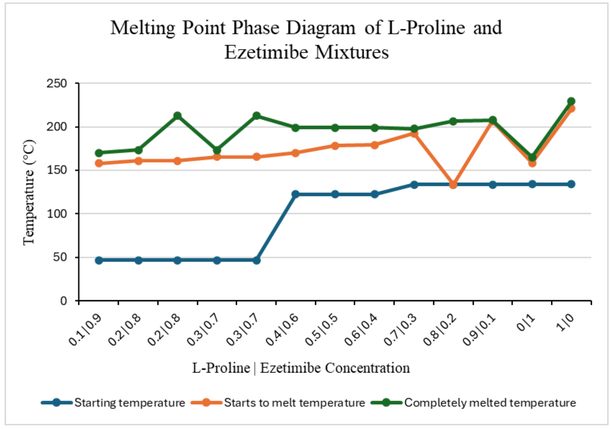
The experiment involved dissolving 0.0500 grams of L-Proline and 0.178 grams of Ezetimibe in 10 mL of methanol to create a 0.0434 M solution. Eleven (11) vials were prepared with varying mole fractions by mixing different volumes, such as 0.1 mL of L-Proline with 0.9 mL of Ezetimibe. The solutions were left to evaporate in a fume hood for a week, forming crystals that were collected and analyzed for melting points using a Vernier melt station.

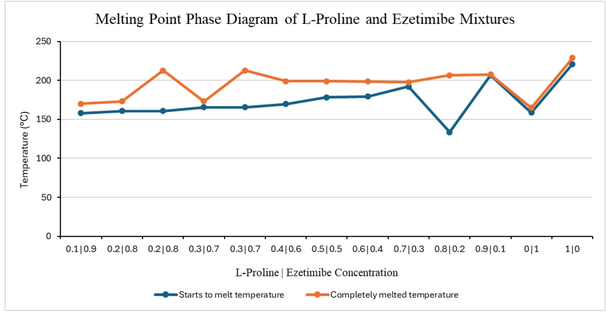
Figure # 1: Crystals of L-Proline and Ezetimibe at Varying Concentrations

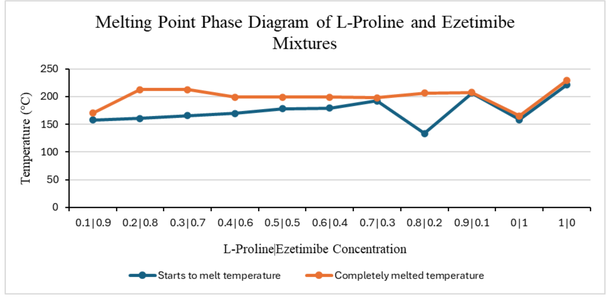


Results

Figure # 2a-c: Melting Behavior of L-Proline and Ezetimibe Mixtures at Different Concentration







Discussion

Ezetimibe melts at 165°C, while L-Proline melts at 229°C, however, when mixed, the melting point ranged from 174.22°C to 175.98°C (Shimpi et al., 2014). At 0.2 and 0.3 mole fractions, two melting points were observed: an initial melt at 173.3°C, forming a light brown precipitate, followed by a second melt at 212.8°C, corresponding to residual L-Proline.

Vials 4–7 exhibited melting points between 197°C and 199°C, gradually darkening upon heating, reflecting the increasing L-Proline content. Similarly, vials 8 and 9 showed melting points of 206.6°C–207.7°C, confirming the trend of higher temperatures with increased L-Proline. Vials 10 and 11, containing pure Ezetimibe and L-Proline, melted at their expected 165°C and 229°C, respectively.

The co-crystal is most likely at a concentration of 0.5, as this corresponds to the expected 1:1 molar ratio of L-Proline and Ezetimibe, which aligns with the findings from the reference study. Co-crystals typically form a distinct and stable solid phase with a sharp melting range, and in this experiment, the 0.5 mole fraction sample exhibited characteristics consistent with co-crystallization. While broader melting point ranges were anticipated between the pure components and the co-crystal, the data did not show this as clearly.

Conclusion

The experiment indicates that a 1:1 molar ratio of L-Proline and Ezetimibe forms a co-crystal, aligning with previous findings. However, factors such as sample size limitations, impurities, and preparation differences may have influenced the results. Small-scale sampling could have caused missed early melting transitions, making the co-crystal melt appear more distinct. While the study used a melting point phase diagram instead of DSC, the data still support co-crystallization at this ratio.

For future experiments, improved precision in sample preparation, accurate concentration measurements, and careful observation of melting transitions are important for more reliable results.

References

Bag, P. P., Singh, G. P., Subba, A., & Chettri, A. (2023). Pharmaceutical co-crystals: A green way to enhance drug stability and solubility for improved therapeutic efficacy | Journal of Pharmacy and Pharmacology | Oxford academic. https://academic.oup.com/jpp/article/76/1/1/7339341.

Shimpi, M. R., Childs, S. L., Boström, D., & Velaga, S. P. (2014). New cocrystals of ezetimibe with l-proline and imidazole. CrystEngComm, 16(38), 8984–8993. https://doi.org/10.1039/c4ce01127a.