CARRAGEENAN-MEDIATED POLYELECTROLYTIC PRECIPITATION FOR BROMELAIN EXTRACTION FROM ANANAS COMOSUS: EVALUATING THE EFFECTS OF BROMELAIN SOLUTION VARIABLES AND CARRAGEENAN LEVELS ON YIELD AND SPECIFIC ACTIVITY

ANA M. VILAS-BOAS, DÉBORA CAMPOS, MANUELA PINTADO*

UNIVERSIDADE CATÓLICA PORTUGUESA, CBQF – CENTRO DE BIOTECNOLOGIA E QUÍMICA FINA – LABORATÓRIO ASSOCIADO, ESCOLA SUPERIOR DE BIOTECNOLOGIA, RUA ARQUITETO LOBÃO VITAL, 172, 4200-374 PORTO, PORTUGAL
*CORRESPONDING AUTHOR

INTRODUCTION

Pineapple (*Ananas comosus* Merr.) is a widely popular fruit, commonly used in various fruit-based products (Ataide et al., 2019; Campos et al., 2019). The byproducts from pineapple processing, which are rich in bioactive compounds like bromelain, offer an environmentally friendly approach to waste management. Bromelain, a protease primarily found in pineapple stems, is typically extracted using techniques such as solvent precipitation and ultrafiltration (Ketnawa & Rawdkuen, 2011; Pratiwi et al., 2020; Schieber et al., 2001; Simões et al., 2022). Recently, polyelectrolyte precipitation has gained attention as a green alternative for protein concentration and purification, ensuring the integrity of proteins while facilitating their separation (Chakraborty et al., 2021; Mala et al., 2021; Rajan & Dunna, 2022).

OBJECTIVE

This study aimed to employ the polyelectrolytic precipitation with carrageenan (Carr) to assess a process for efficient bromelain extraction from pineapple stem and peel fractions.

Several sequential steps for bromelain purification were applied to obtain the best end-product for potential commercialization, like centrifugations, pellet washing and different stock Carr concentration.

METHODOLOGIES

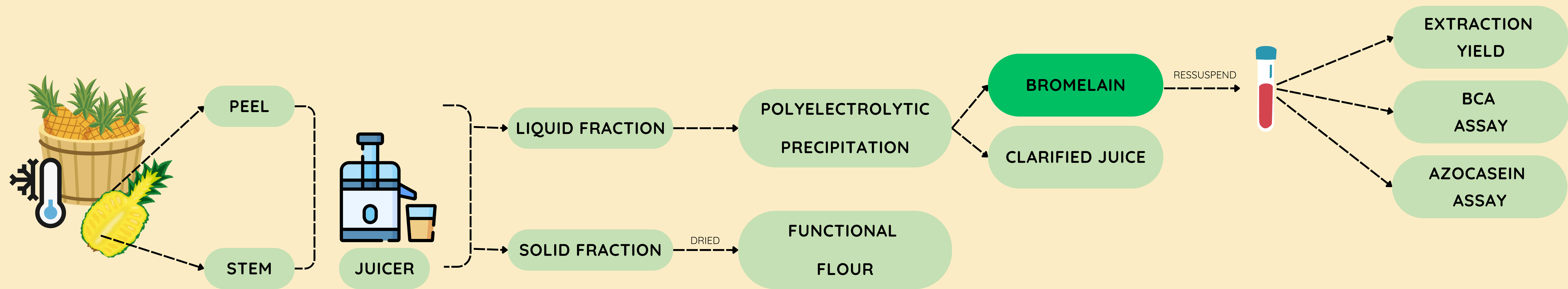

Figure 1 - General schematic representation of the methodologies used for the obtention and analysis of bromelain from pineapple parts.

RESULTS

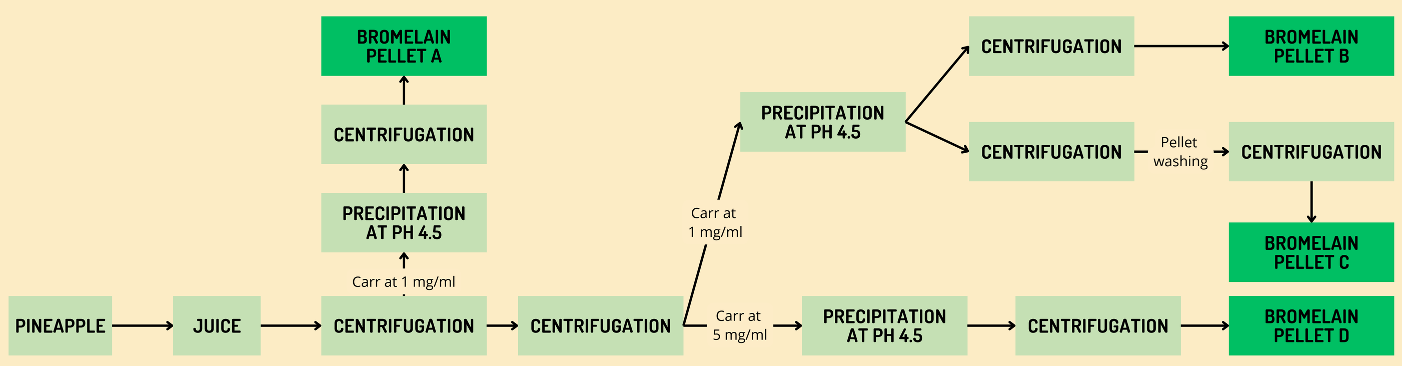

Figure 2 - Polyelectrolytic precipitation flowchart description for different bromelain pellets production (Pellet A, B C and D). "Method for Extraction and/or isolation of bromelain from pineapple": Patent no. EP 3 252 156 A1.

Overall higher extraction yield at higher Carr concentrations

Specific Proteolytic Activity was significantly affected by the processes

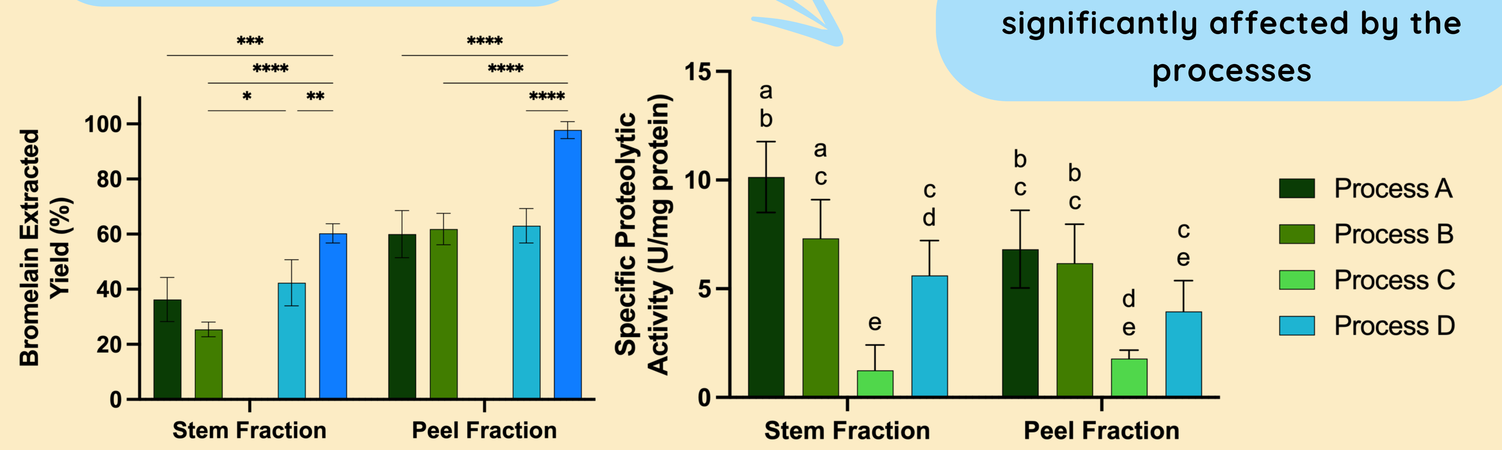

Figure 3 - (left) Extrated bromelain percentage yield obtained for all tested samples; (right) Specific proteolytic activity obtained for bromelain samples from stem and peel pineapple fractions.

Outermost pHs significantly affected one another, but no significant differences were found on the tested samples. pH 8 exhibits overall highest absorbance results.

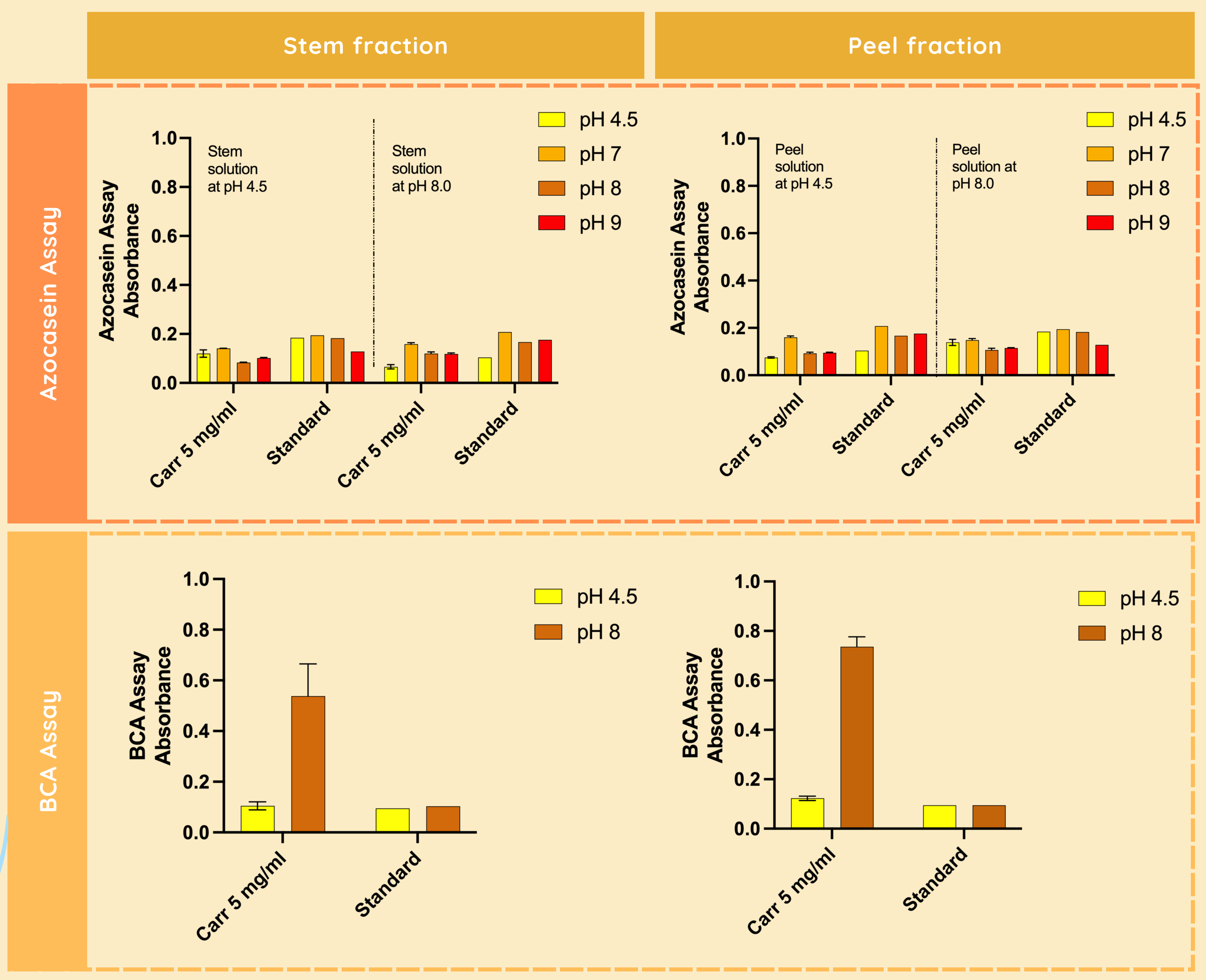

Figure 4 - (top) Azocasein assay results for stem and peel pineapple samples at 5 mg/ml Carr concentration tested for pH 4.5, 7, 8 and 9; (bottom) BCA assay results for stem and peel pineapple samples at 5 mg/ml Carr concentration tested for pH 4.5 and 8.

CONCLUSION

Polyelectrolytic precipitation with Carr concentrations of 5 mg/ml, or higher, significantly impacts precipitate yield (ca. >60%) and specific proteolytic activity for both fractions. The enzyme separation pH affects the activity, exhibiting higher values for alkaline pH. Extracting bromelain from pineapple waste offers a sustainable solution for waste reduction and promotes health-focused products, supporting a zero-waste approach in pineapple production.

ACKNOWLEDGMENTS

We would like to thank the scientific collaboration of Faculty of Biotechnology of Universidade Católica Portuguesa through CBQF under FCT-Fundação para a Ciência e a Tecnologia, to accept Ana M. Vilas-Boas through a Ph.D. grant reference 2022.14462.BD. As well as, the PRR Proj. nº 16 - Pacto de Inovação da Bio Economia Azul (C644915664-00000026) - Vertical Rações – FEED.