

Influence of natural preservatives on the bioactivity and stability of a nutraceutical formulation

Filipa A. Fernandes¹, Márcio Carochó¹, Sandrina A. Heleno¹, Paula Rodrigues¹, José Pinela¹, Miguel A. Prieto², Lillian Barros^{1,*} and Isabel C. F. R. Ferreira¹

¹Centro de Investigação de Montanha (CIMO), Instituto Politécnico de Bragança, Campus de Santa Apolónia, 5300-253 Bragança, Portugal;

²Nutrition and Bromatology Group, Department of Analytical and Food Chemistry, Faculty of Food Science and Technology, University of Vigo-Ourense Campus, E-32004 Ourense, Spain.
[*lillian@ipb.pt](mailto:lillian@ipb.pt)

Introduction

The medicinal properties of plants have been well known from an early age, but nowadays there are many scientific studies supporting these claims. *Aloe Arborescens* Mill. is one of the most abundant species of the *Aloe* genus, native to South Africa which has been imported into many countries as an ornamental and medicinal plant due to its biological effects, namely antimicrobial, anti-inflammatory and scab healing properties, which have been proven by scientific studies [1]. Beyond this, *A. arborescens* is also used for the extraction of active ingredients with cosmetic and nutraceutical interest [2].

The growing concern about the amount of chemicals added to foods as well as pharmaceutical and cosmetic products lead consumer preference to tilt towards products with low additives, and, when possible, natural additives, which are perceived by consumers as less harmful to human health [3].

Objective

The main objective of this work was to study the influence of natural ingredients (quercetin, citric acid, chestnut flower and chestnut flower extract) on the antioxidant activity as well as on the stability of a commercial nutraceutical formulation of *Aloe arborescens* Mill., with the aim at replacing the artificial preservatives commonly applied (sodium benzoate).

Results and discussion

Antioxidant activity

The antioxidant capacity of the different formulations is presented in **Table 1**.

- Regarding the **TBARS** assay, this analysis was carried out to verify the capacity that the preservatives showed in inhibiting the formation of thiobarbituric and malodialdehyde complexes. The best preserving capacity was found for both quercetin and chestnut flowers preserved formulations, which showed the lowest EC₅₀, followed by the chestnut flower extract preserved formulation.

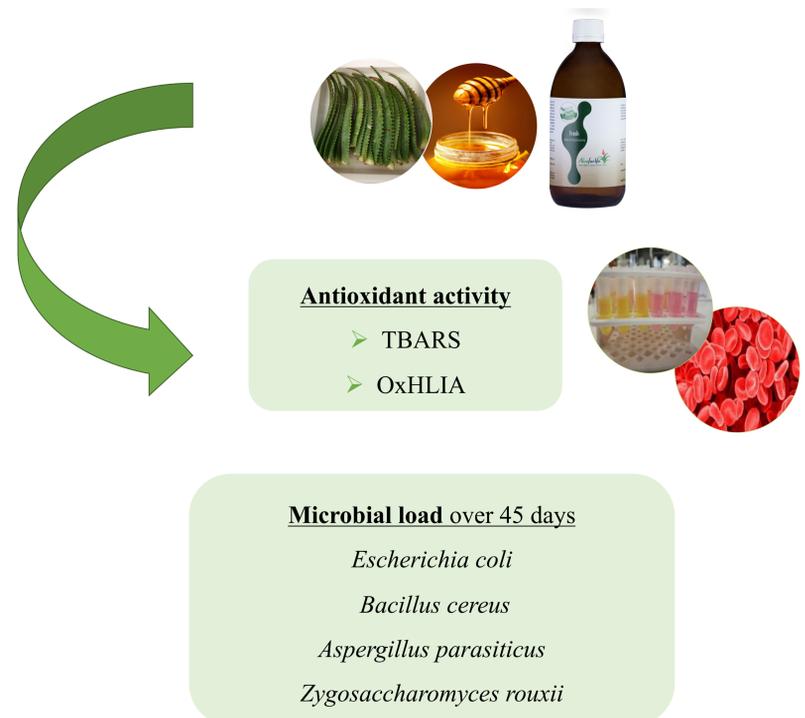
Table 1. Antioxidant profile of the different nutraceutical formulations, expressed in mg and µg/mL of extract.

| Type of Preservative | TBARS | OxHLIA 60 min | OxHLIA 120 min |
|----------------------|-------------|------------------|-------------------|
| | Control | 4.3±0.5 | 332±192 |
| Sodium Benzoate | 6.1±0.9 | 273±40 | 576±161 |
| Citric Acid | 5±2 | 381±101 | 792±286 |
| Quercetin | 0.037±0.008 | 139±62 | 208±99 |
| Chestnut Flower | 0.109±0.005 | 228±136 | 393±244 |
| Chestnut Extract | 0.6±0.2 | 370±144 | 962±377 |

- **OxHLIA** results, that are presented as IC₅₀, representing the amount needed to quench 50% of the free radicals, and thus, a lower amount is correlated to a higher antioxidant activity. Comparing the 60 to the 120-minutes time of assay, there seems to be lower IC₅₀ values in the 60 minutes, showing that all the studied ingredients have high antioxidant potential, highlighting quercetin and chestnut flower as the most antioxidant compounds.

Methodology

Preparation of the different nutraceutical formulations with the preservative ingredients



Microbial loads

The Microbial load of the different formulations is presented in **Figure 2**.

- Regarding the growth of **coliforms** (Figure (a)), after 45 days of storage, the formulations do not show bacterial growth, and the formulation with chestnut flower proved to be the most effective compound.
- Figure 2 (b) show the counts of *Bacillus cereus* and interestingly, after 30 days, the bacterial load was reduced to zero in all the formulations.
- Regarding the **yeasts**, (Figure 2 (c)), these microorganisms showed an increase from T0 to T30 followed by a decrease from T30 to T45. Although all formulations followed this tendency, the formulation with chestnut flower display a better preserving action than the positive control.
- Regarding the **molds**, in Figure 2 (d), at T30 for the benzoate samples (positive control), no molds were detected; and, at T45 the samples preserved with citric acid, also the molds were not detected, with all other samples not showing any change over time.

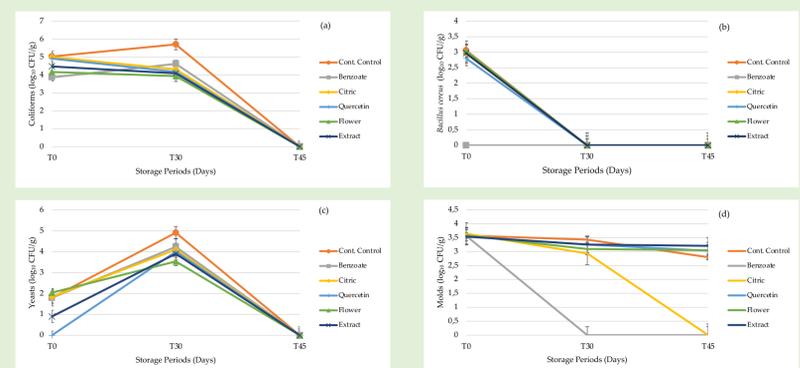


Figure 2. Microbial development along the 45 days of storage time for (a) coliforms, (b) *Bacillus cereus*, (c) yeasts, and (d) molds.

Conclusion

The natural studied natural ingredients proved the ability to promote the antioxidant capacity of the nutraceutical formulation. Moreover, they also revealed the capacity to control microbial growth; two essential activities to develop a strong and viable preservative.

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