In Vitro Antioxidant Properties of Coffee Pulp and Cocoa Shell Infusions for Sustainable Functional Beverages

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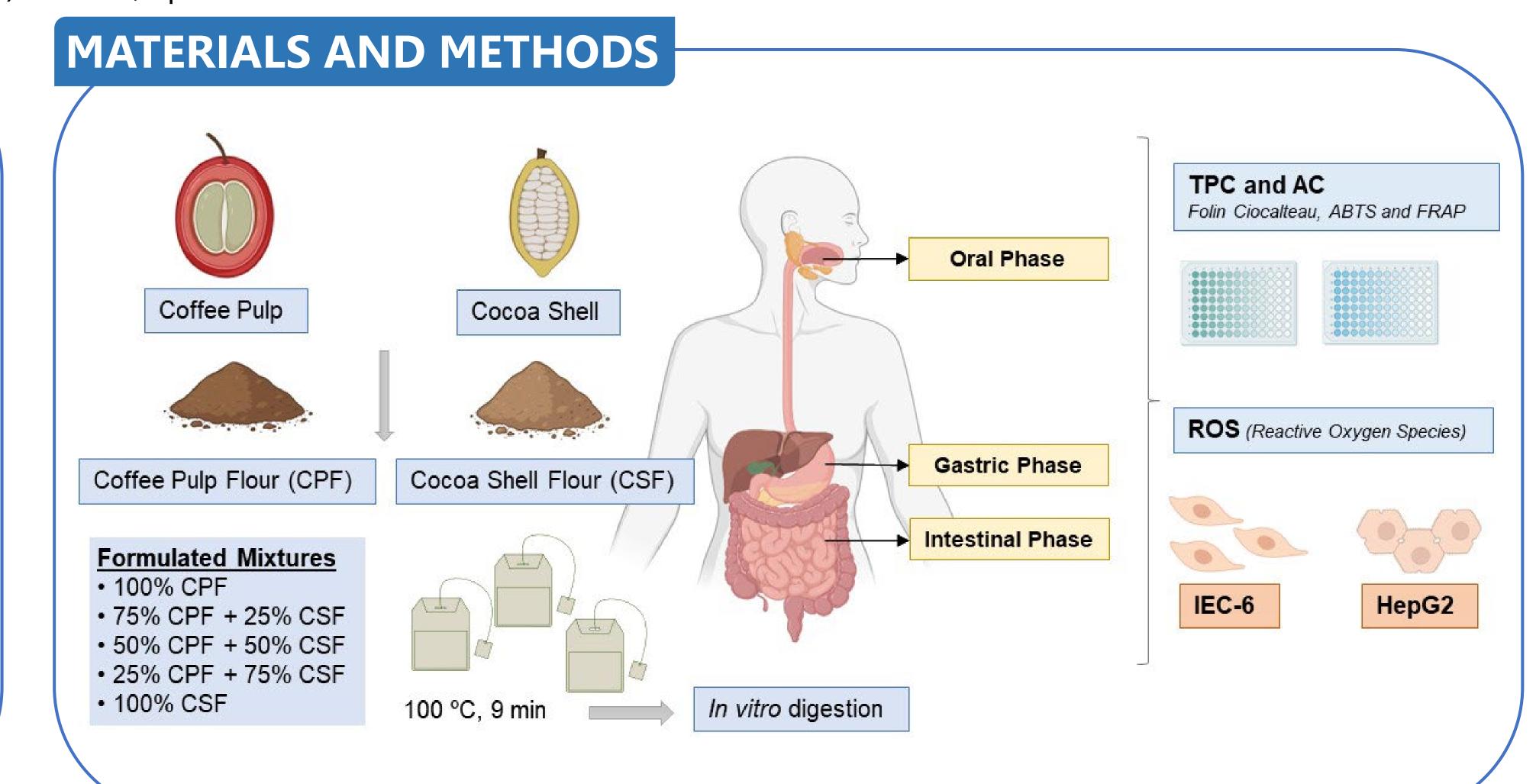
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INTRODUCTION

Coffee pulp and cocoa shell are by-products from coffee and cocoa processing, rich in bioactive compounds with health-promoting properties. Combining them in a single infusion represents a sustainable approach to develop functional beverages while reducing food waste. This work explores the antioxidant potential of these infusions before and after digestion, aiming to identify possible synergistic effects [1].

The objective was to evaluate the antioxidant potential of coffee pulp and cocoa shell infusions before and after simulated digestion, exploring possible synergistic effects and their suitability as sustainable ingredients for functional beverages.

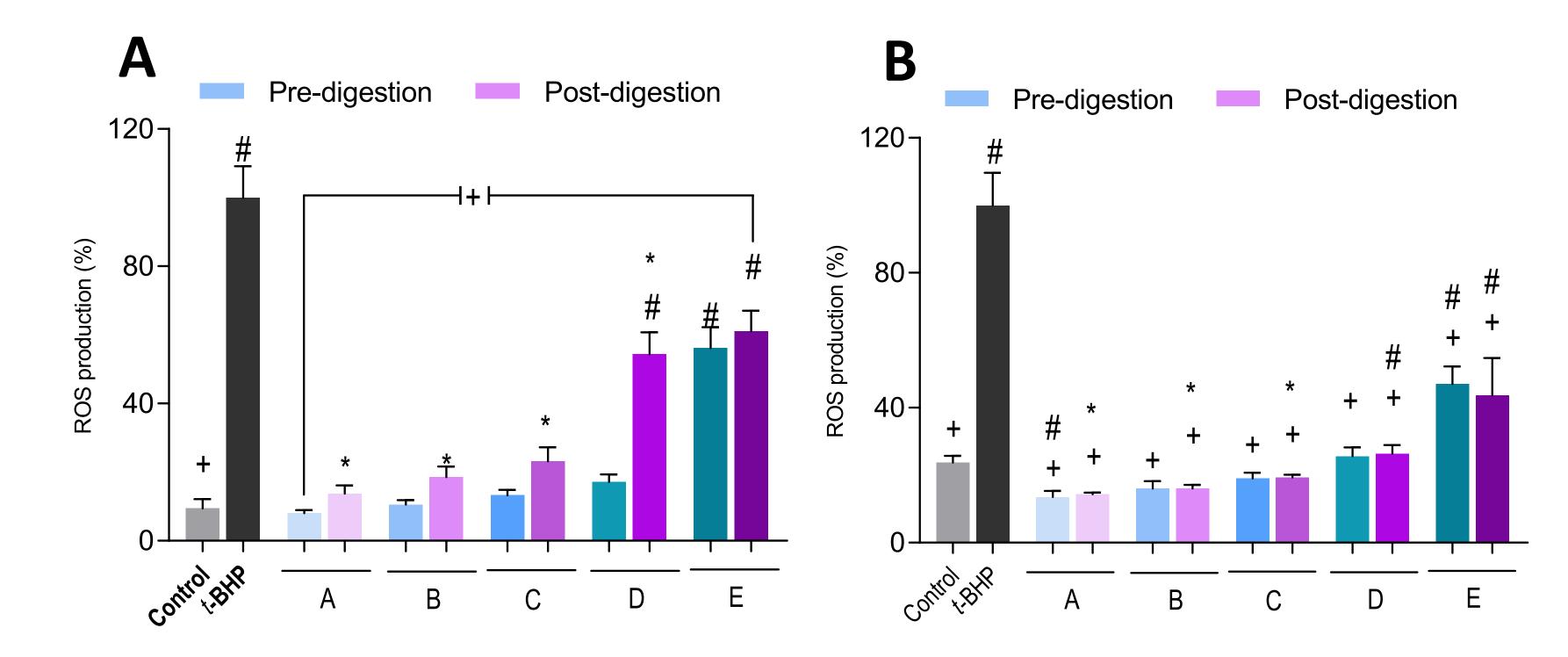


RESULTS

Table 1. Total phenolic content (TPC) and antioxidant capacity (ABTS and FRAP assays) of freshly prepared infusion blends of coffee pulp (CP) and cocoa shell (CS) at different ratios (100:0, 75:25, 50:50, 25:75, 0:100, CP:CS).

CP:CS ratio	TPC	ABTS	FRAP
	(mg GAE L ⁻¹)	(mg TE mL ⁻¹)	(mmol TE/mL^{-1})
100:0	1341.3 ± 445.9^{a}	$3.2\pm0.4^{\rm a}$	$14.9\pm1.1^{\rm a}$
75:25	1025.2 ± 454.3^{b}	$0.7 \pm 0.1^{\text{b}}$	$3.5\pm0.3^{\text{b}}$
50:50	536.4 ± 359.2^{c}	$0.4 \pm 0.0^{\rm c}$	$2.0\pm0.2^{\rm c}$
25:75	500.2 ± 171.1^{c}	$0.2 \pm 0.0^{\text{d}}$	$1.2 \pm 0.1^{\text{d}}$
0:100	308.4 ± 105.6^d	$0.1 \pm 0.0^{\rm d}$	$0.6 \pm 0.1^{\rm e}$

Results are expressed as mean \pm SD (n = 3). Mean values within rows followed by different superscript letters (a–e) are significantly different according to ANOVA and Tukey's multiple range test (p < 0.05). GAE: gallic acid equivalent; TE: Trolox equivalent.



- The infusion made exclusively from coffee pulp showed the highest total phenolic content (1341.3 mg GAE/L), which gradually decreased with the addition of cocoa shell, reaching 308.4 mg GAE/L in the 100% cocoa shell infusion. Antioxidant capacity followed the same trend, with ABTS decreasing from 3.2 to 0.1 mg TE/mL and FRAP from 14.9 to 0.6 mmol TE/mL.
- After simulated digestion, all infusions retained antioxidant activity, although values were slightly lower than before digestion. The decrease was more pronounced in samples with higher cocoa shell content, suggesting a lower stability of its phenolic compounds during the digestive process.
- In both cell lines (IEC-6 and HepG2), oxidative stress induced by *t*-BOOH was significantly reduced by the infusions, with ROS inhibition ranging from 48–114% before digestion and 43–112% after digestion.

Figure 1. Production of reactive oxygen species (ROS) in rat intestinal epithelial cells (IEC-6) (**A**) and human hepatocytes (HepG2) (**B**) in infusions made from coffee pulp (CP) and cocoa shell (CS) flours: 100% CP + 0% CS (A), 75% CP + 25% CS (B), 50% CP + 50% CS (C), 25% CP + 75% CS (D), and 0% CP + 100% CS (E). Blue bars represent infusions before digestion, and purple bars represent infusions after in vitro digestion. Results are expressed as mean \pm standard deviation (n = 3). Asterisks (*) indicate differences between cells treated with the same infusion before and after in vitro digestion according to the Student's t-test (p < 0.05). The (+) symbol above the bars indicates statistically significant differences between cells treated with the infusions (p < 0.05) and t-BHP, according to Dunnett's test (p < 0.05). The (#) symbol above the bars indicates statistically significant differences between cells treated with the infusions (p < 0.05) and the control, according to Dunnett's test (p < 0.05).

CONCLUSIONS

These findings reveal that coffee pulp and cocoa shell infusions retain antioxidant activity after digestion and reduce oxidative stress in cells. They represent promising and sustainable sources of bioactive compounds for the development of functional beverages.