# *Whey–pectin microcapsules improve the stability of grape marc phenolics during digestion*

Article

Supplemental Material

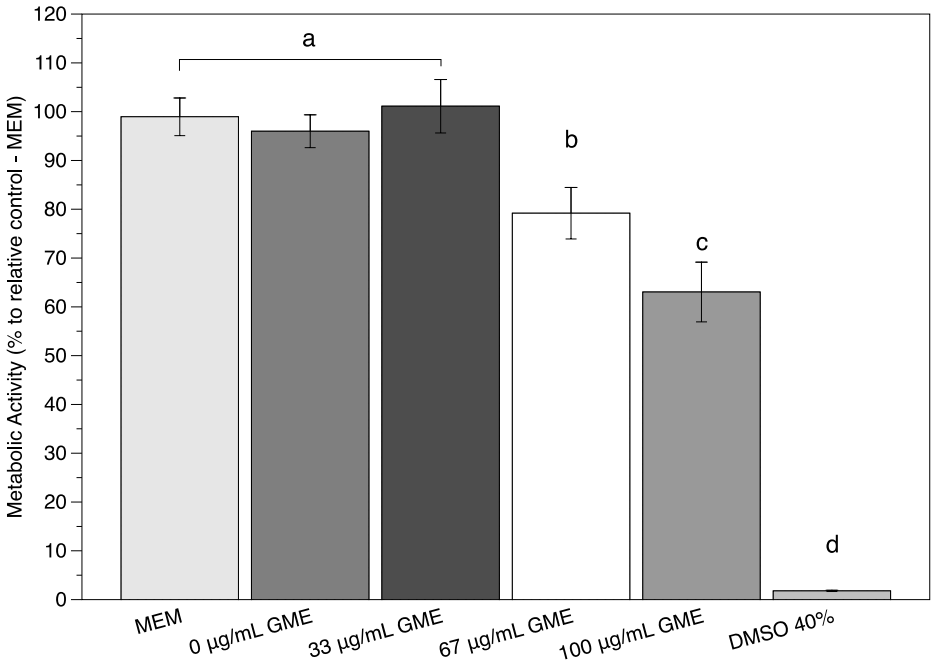


Figure 1. Viability of Caco-2 cells after 24 h-incubation with different concentrations of GME (33, 67 or 100 GAE µg/mL), measured through the resazurin assay. Culture medium (MEM) was used as a positive control (100% cell viability), and 40% DMSO (v/v) as a negative control. Values are the mean ± sd from 2 independent assays analyzed in quadruplicate. Different letters denote statistical significance ($p<0.05$) determined using the Dunnett method.

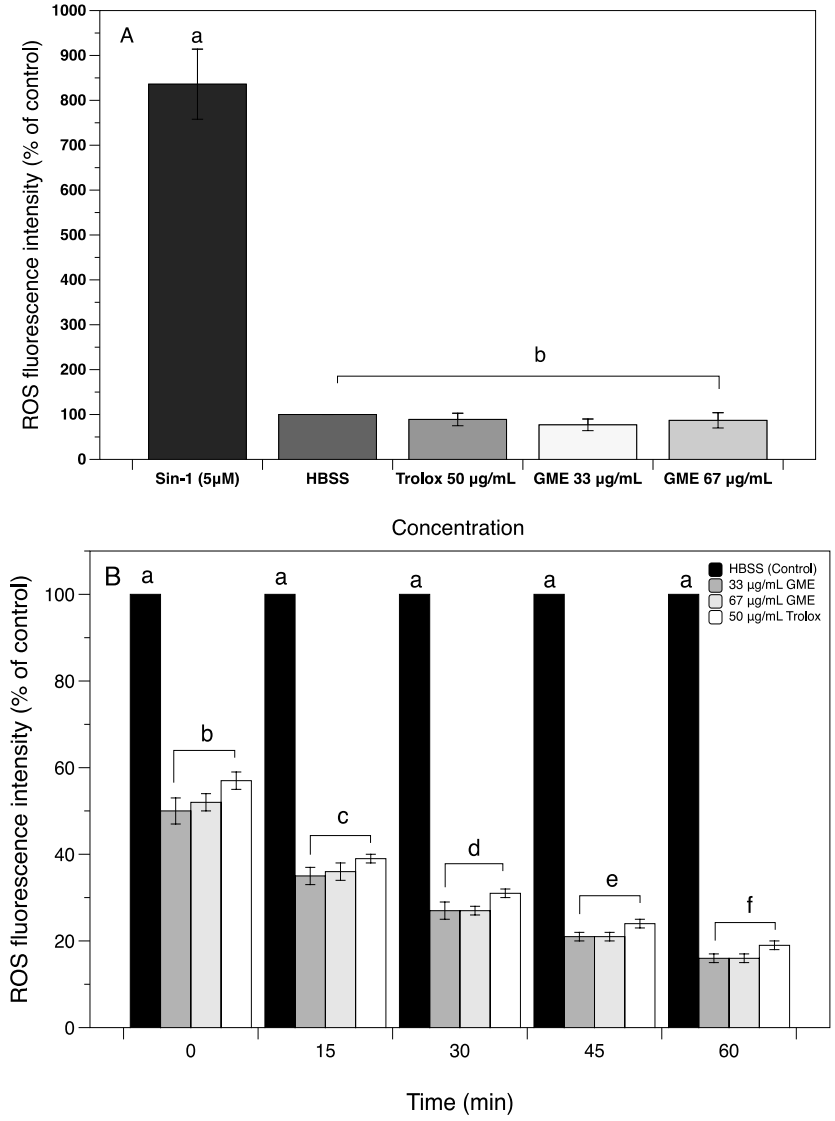


Figure 2. (A) Effect of different concentrations of GME (33 or 67 GAE µg/mL) on the ROS basal levels of Caco-2 cells after incubation for 4h, measured through the DCFH-DA assay. HBSS was used as a negative control, and Sin-1 (5 uM), as a positive control. (B) Protective effect of different concentrations of GME (33 or 67 GAE µg/mL) on the ROS levels of Caco-2 cells after incubation for 4h, followed by stimulation with 5 uM Sin-1 (oxidant) for 1 h. ROS was measured through the DCFH-DA assay. Cells treated with HBSS and stressed with Sin-1 were used as a positive control. Values are the mean ± sd of two independent assays analysed in quadruplicate. Different letters show statistical significance ($p<0.05$) determined using the Dunnett method.

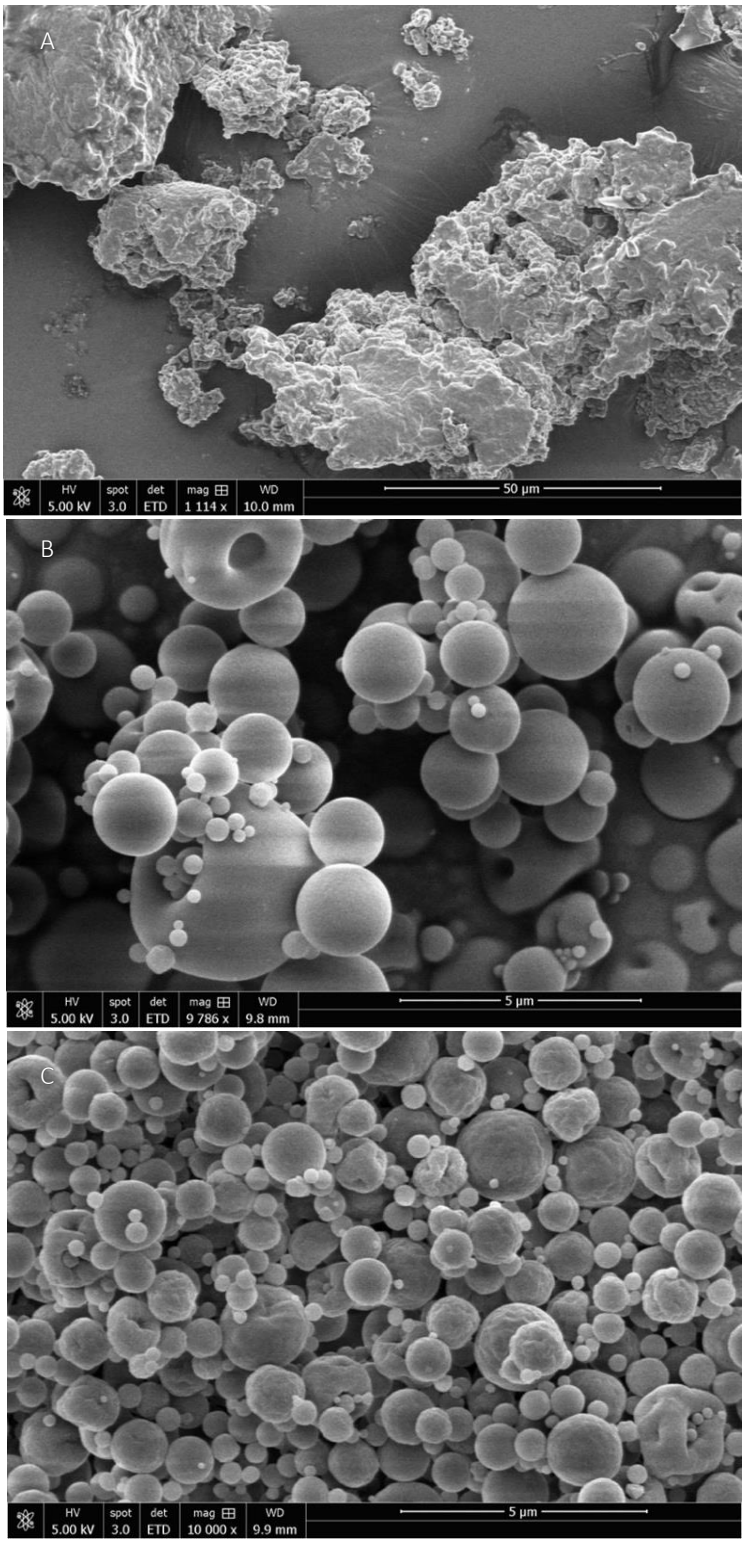


Figure 3. SEM images of GME (A), whey-pectin blank microparticles (B), and GME encapsulated in whey-pectin microparticles (C).

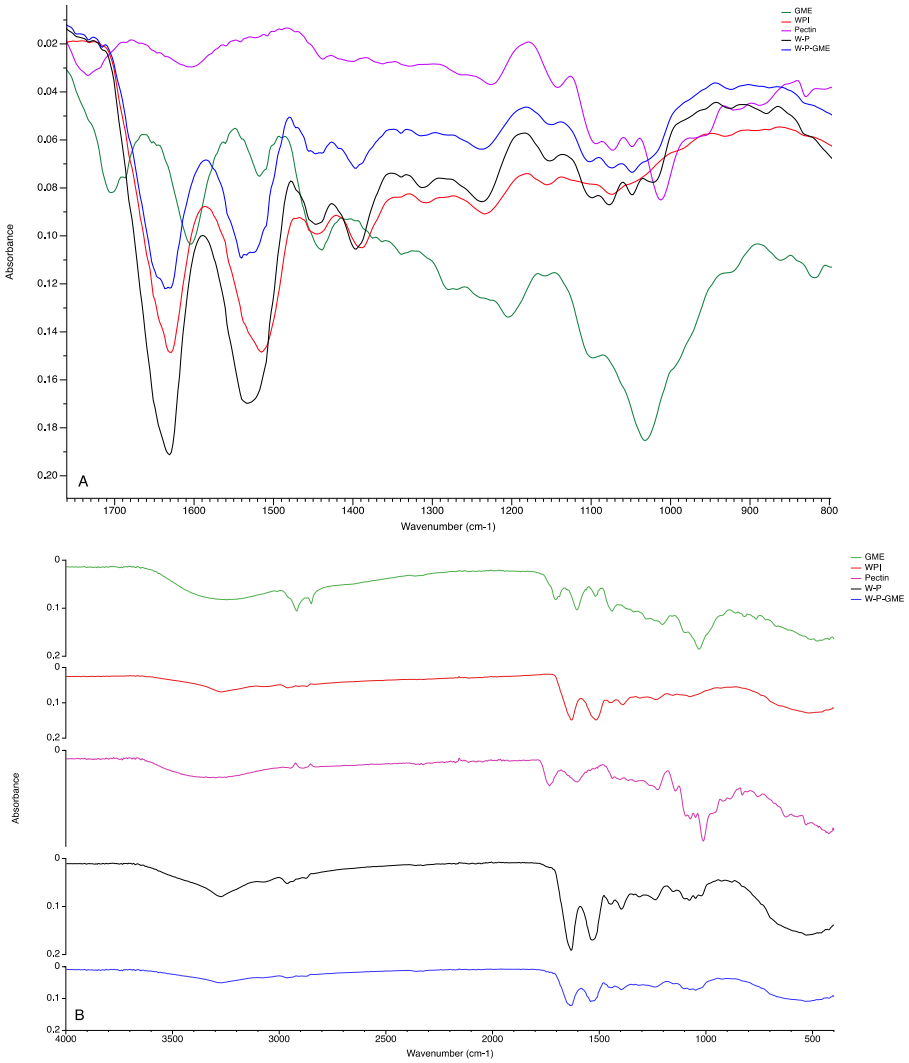


Figure 4. FT-IR of GME, whey-pectin blank microparticles, GME encapsulated in whey-pectin microparticles, pectin and whey. Amplified FTIR spectra of wavelengths 1750 to 800 cm-1 (A) and full spectra (B). The results of each material are the average of three independent spectra.

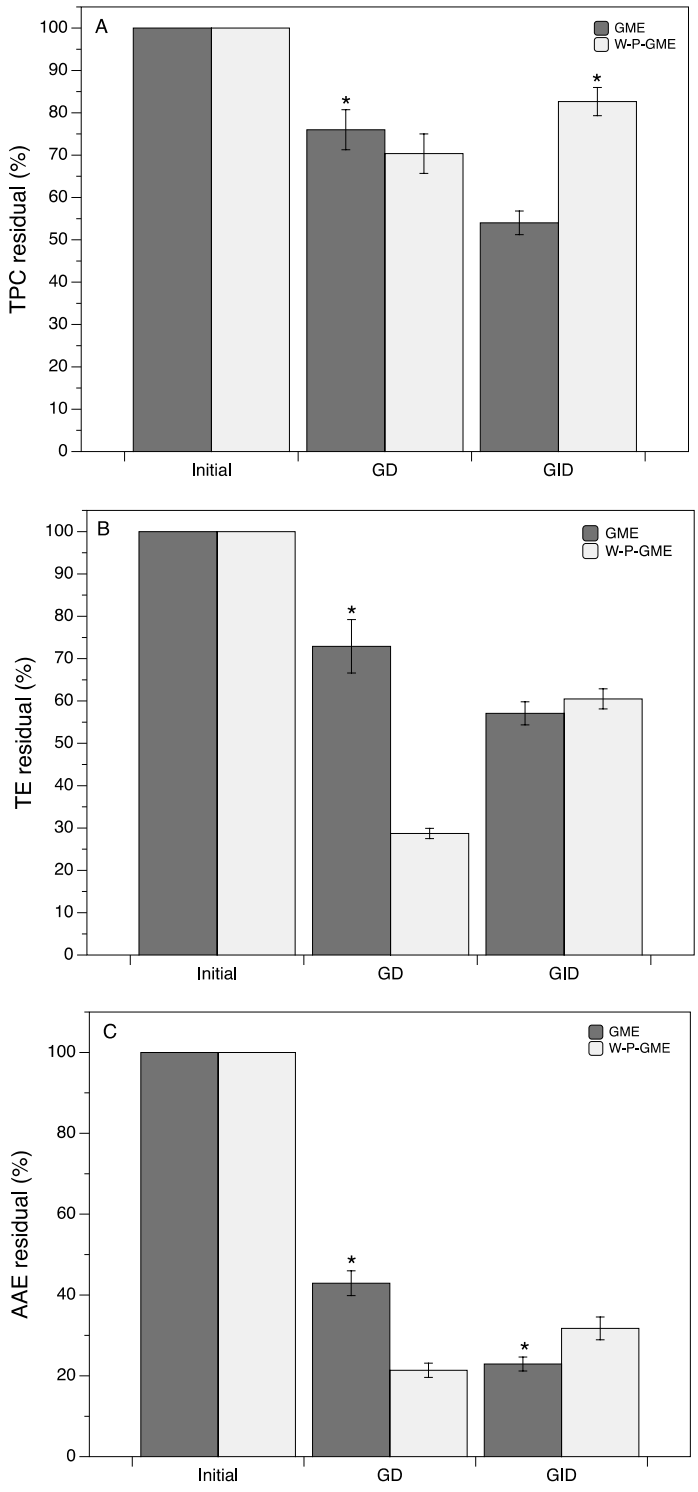


Figure 5. Results of the residual TPC (A), TE (B), and AAE (C) of GME and GME encapsulated in whey-pectin microparticles after *in vitro* gastric digestion (GD) and gastrointestinal digestion (GID). Values are the mean ± sd from three independent assays, each analysed in triplicate.* Denotes statistical significance between GME and W-P-GME ($p<0.05$) determined using the Tukey test.

# Tables

**Table 1.** Content of polyphenols and antioxidant activity in GME.

| | |
|---|---|
| **Total phenol content** | 219.62 ± 11.50 |
| **Total flavonoid content** | 151.69 ± 5.29 |
| **Total monomeric anthocyanins** | 12.80 ± 0.63 |
| **Antioxidant capacity by ABTS** | 1389.71 ± 97.33 |
| **Antioxidant capacity by FRAP** | 848.95 ± 43.99 |

TPC: mg GAE/g dry extract; TAC: mg MV3GE/ L; TFC: mg CE/ g dry extract

ABTS: µmol TE/ g dry extract; FRAP: µmol AAE/ g dry extract

Values are represented as mean ± sd (n=6 from 3 replicates and 2 determinations)

Table 2. Z-potential, size and PDI of particles

| Particles | Z-potential | Size (µm) | PDI | Yield (%) |
|---|---|---|---|---|
| W-P-GME | -28.3 ± 6.1 | 1.0 ± 0.5 [a] | 0.7 | 73 |
| W-P | ND | 1.3 ± 0.7 [b] | 0.7 | ND |

Z-potential: based on 1mg/mL particle suspension in water; average ± sd from 5 measurements

ND: non determined

Different letters denote significate difference ($p<0.05$) using the independent samples t-test.