

## Antioxidant and anti-inflammatory actions of red seaweed (*Grateloupia turuturu* and *Porphyra umbilicalis*) extracts linked to phytochemical characterization and cytotoxicity evaluation

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### INTRODUCTION & AIMS

The number of

functional foods

and

nutraceuticals

from marine origin is increasing in the worldwide market [1].

Specifically, the red seaweeds



*Porphyra umbilicalis*

*Grateloupia turuturu*

have shown their potential considering relevant bioactive compounds and nutritional value [1,2].

Nevertheless, it is pertinent to explore knowledge gaps regarding their phytochemical composition and bioactivities.

The main objectives were to prepare *G. turuturu* and *P. umbilicalis* extracts to determine their phytochemical composition, evaluate their cytotoxicity and assess antioxidant and anti-inflammatory potentials, validating their use as functional food and nutraceuticals.

### METHODS

Harvested from the Western Portuguese coast



*Grateloupia turuturu*

*Porphyra umbilicalis*

Hydroethanolic, infusion and decoction extracts

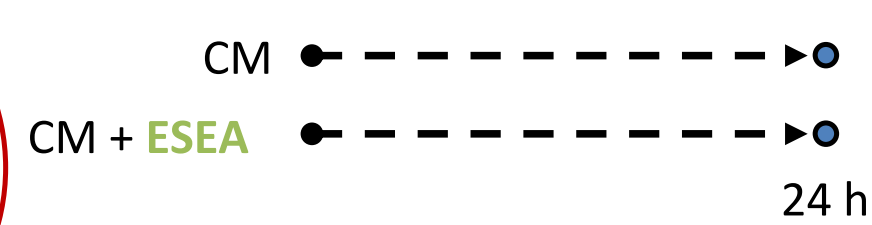
#### Phytochemical characterization

- Total phenols content was determined using Folin-Ciocalteu method [3].
- RP-HPLC-DAD analyses was used to determine the phenolic profile.
- Mycoporine-like amino acids (MAAs) content was determined using RP-HPLC-DAD and LC-DAD-ESI-MS.
- The phenol-sulfuric acid method was used for determining total carbohydrates content [4].

#### Antioxidant potential

- ABTS<sup>•+</sup>, •OH (salicylic acid) and •NO (Griess method) scavenging activities were determined [5,6].

#### Citotoxicity and anti-inflammatory activity



Cell viability assay  
alamarBlue<sup>®</sup> assay [5,6]

Anti-inflammatory activity  
Lipopolysaccharide stimulated cells  
Griess method for •NO quantification [5,6]

RAW 264.7 cells

CM – culture medium | ESEA – seaweed extract | ● – sample collection

### RESULTS & DISCUSSION

#### Folin-Ciocalteu and HPLC data for phenolics

- Results showed a higher content in reducing compounds for aqueous extracts (~22-28 mg gallic acid equivalents (GAE) g<sup>-1</sup> extract) compared to hydroethanolic extracts (~17-18 mg GAE g<sup>-1</sup> extract).
- Chromatograms did not show the presence of typical terrestrial phenols.

#### MAAs and total carbohydrates contents

- P. umbilicalis* extracts showed greater MAAs content, and porphyra-334 was the main MAA (Table 1).
- The aqueous extracts showed higher carbohydrate contents, with *P. umbilicalis* decoction exhibiting the highest level (~40% extract).

Table 1. Content of MAAs in *G. turuturu* and *P. umbilicalis* extracts (in mg g<sup>-1</sup> extract).

Seaweed extract	MAAs concentration (mg g <sup>-1</sup> extract)				TOTAL
	Shinorine	Palythine	Porphyra-334	Aplysiapalythine A or Palythanol	
GH	3.96 ± 0.30 <sup>a</sup>	0.65 ± 0.11 <sup>a</sup>	-	-	4.61 ± 0.32 <sup>a</sup>
GI	6.23 ± 0.04 <sup>b</sup>	0.57 ± 0.03 <sup>a</sup>	t	-	6.80 ± 0.05 <sup>b</sup>
GD	5.43 ± 0.08 <sup>c</sup>	0.55 ± 0.04 <sup>a</sup>	-	-	5.98 ± 0.09 <sup>c</sup>
PH	1.71 ± 0.01 <sup>d</sup>	0.39 ± 0.03 <sup>b</sup>	18.35 ± 0.01 <sup>a</sup>	0.41 ± 0.01 <sup>a</sup>	20.86 ± 0.03 <sup>d</sup>
PI	1.72 ± 0.02 <sup>d</sup>	0.21 ± 0.06 <sup>c</sup>	21.64 ± 0.01 <sup>b</sup>	0.39 ± 0.01 <sup>a</sup>	23.96 ± 0.06 <sup>e</sup>
PD	1.34 ± 0.02 <sup>e</sup>	0.12 ± 0.02 <sup>c</sup>	17.85 ± 0.08 <sup>c</sup>	0.31 ± 0.01 <sup>b</sup>	19.62 ± 0.09 <sup>f</sup>

GH, *G. turuturu* hydroethanolic extract; GI, *G. turuturu* infusion extract; GD, *G. turuturu* decoction extract; PH, *P. umbilicalis* hydroethanolic extract; PI, *P. umbilicalis* infusion extract; PD, *P. umbilicalis* decoction extract; t, trace. Different superscript letters within each column indicate statistically significant ( $p < 0.05$ ) differences. Data in bold show the predominant MAA in each seaweed extract ( $p < 0.0001$ ).

#### Antioxidant activity

- Aqueous extracts demonstrated the greatest ABTS<sup>•+</sup> scavenging activities, being greater for *P. umbilicalis* decoction (97 mmol trolox equivalents g<sup>-1</sup> extract).
- Antioxidant actions against •OH were more accentuated for hydroethanolic extracts (55-57% inhibition).
- G. turuturu* aqueous extracts developed the highest protection against •NO (54-56% inhibition).

#### Citotoxicity and anti-inflammatory activity

- P. umbilicalis* infusion extract had an IC<sub>50</sub> of 0.43 mg mL<sup>-1</sup>, while other extracts showed IC<sub>50</sub> values above 0.75 mg mL<sup>-1</sup>.
- The highest anti-inflammatory activity was shown in *P. umbilicalis* hydroethanolic extract (Fig. 1).

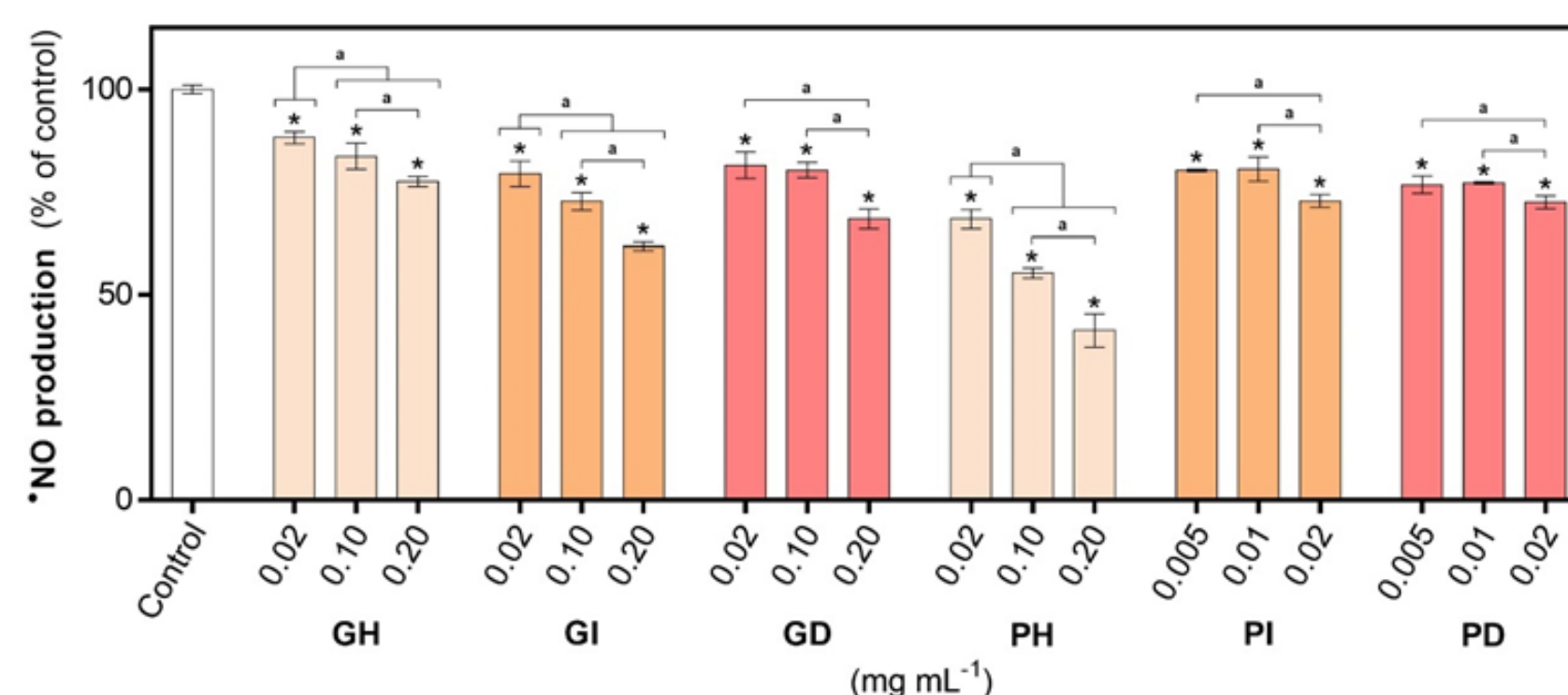


Fig. 1. Anti-inflammatory activity of the *G. turuturu* and *P. umbilicalis* extracts on RAW 264.7 cells upon stimulation with lipopolysaccharide. *G. turuturu* extracts: hydroethanolic (GH), water infusion (GI) and water decoction (GD) and *P. umbilicalis* extracts: hydroethanolic (PH), water infusion (PI) and water decoction (PD). \* $p < 0.05$ , significant differences relative to control; \*\* $p < 0.05$ , significant differences between concentrations of the same extract.

- MAAs and carbohydrates are believed to be the main compounds accountable for the reported antioxidant and anti-inflammatory activities.
- G. turuturu* and *P. umbilicalis* demonstrated their potential as functional food and source of bioactive compounds.

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