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# Anti-inflammatory activity of exopolysacharides (EPS) from New Caledonian marine bacteria

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LPS + EPS (250 ng/mL



### Background

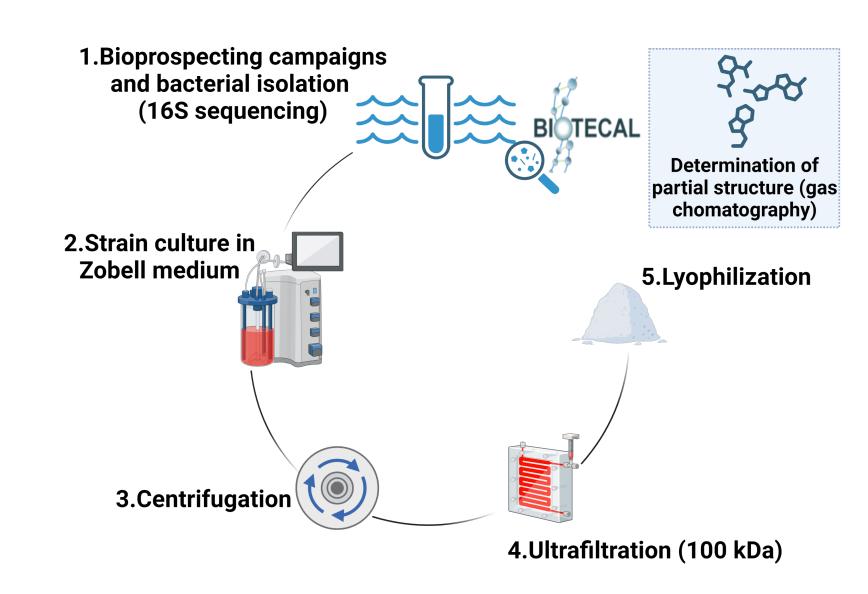
- Chronic inflammatory diseases affect
   5-7% of Western population<sup>1</sup>
- Treatments based on corticoids and NSAIDs lead to severe side effects, whereas monoclonal therapy caused the development of opportunistic diseases
- Research is still ongoing to find other molecules: chelators<sup>2</sup> (deferoxamine...)
- Marine natural products (MNPs) are a potential source of bioactive molecules
- Bioprospecting campaigns in New Caledonia led to the characterisation of bacterial strains expressing bioactive molecules (EPS)<sup>3</sup>

# OBECTIVE: Evaluate the anti-inflammatory activity of EPS from New Caledonian marine bacteria

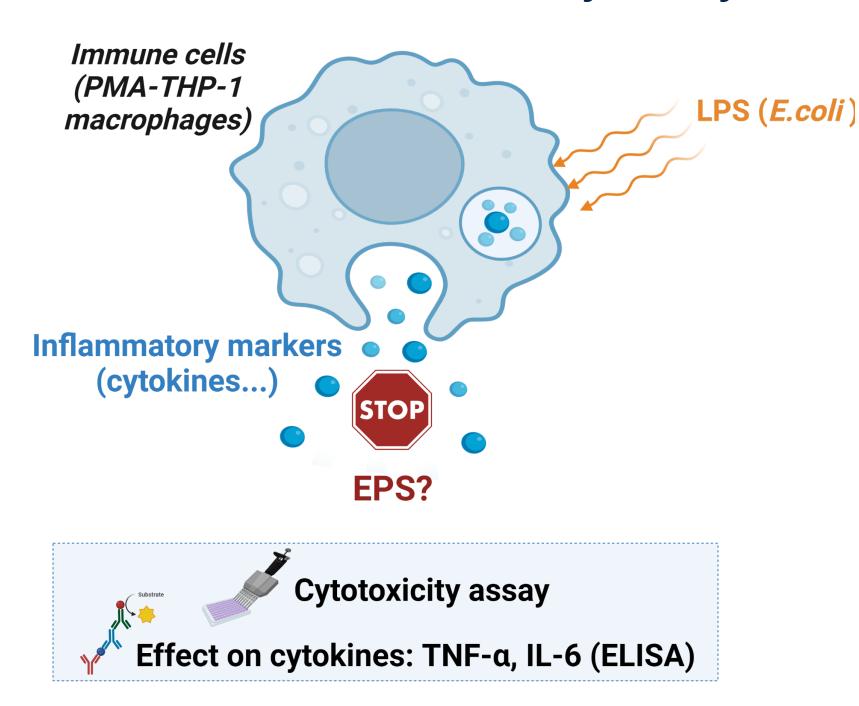


## Methodology

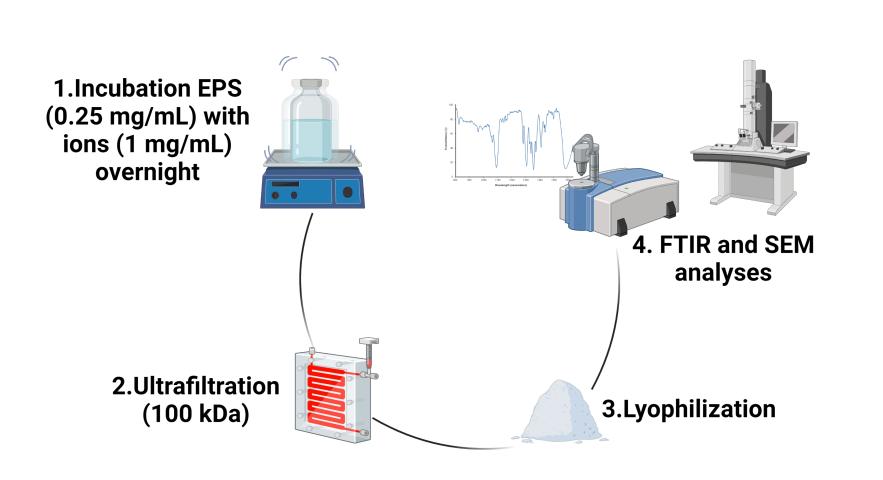
#### **EPS Production**



#### *In vitro* anti-inflammatory assays



#### **Tests for metal chelation**



#### Results and discussion

#### 1. Anti-inflammatory activity of EPS from marine bacteria

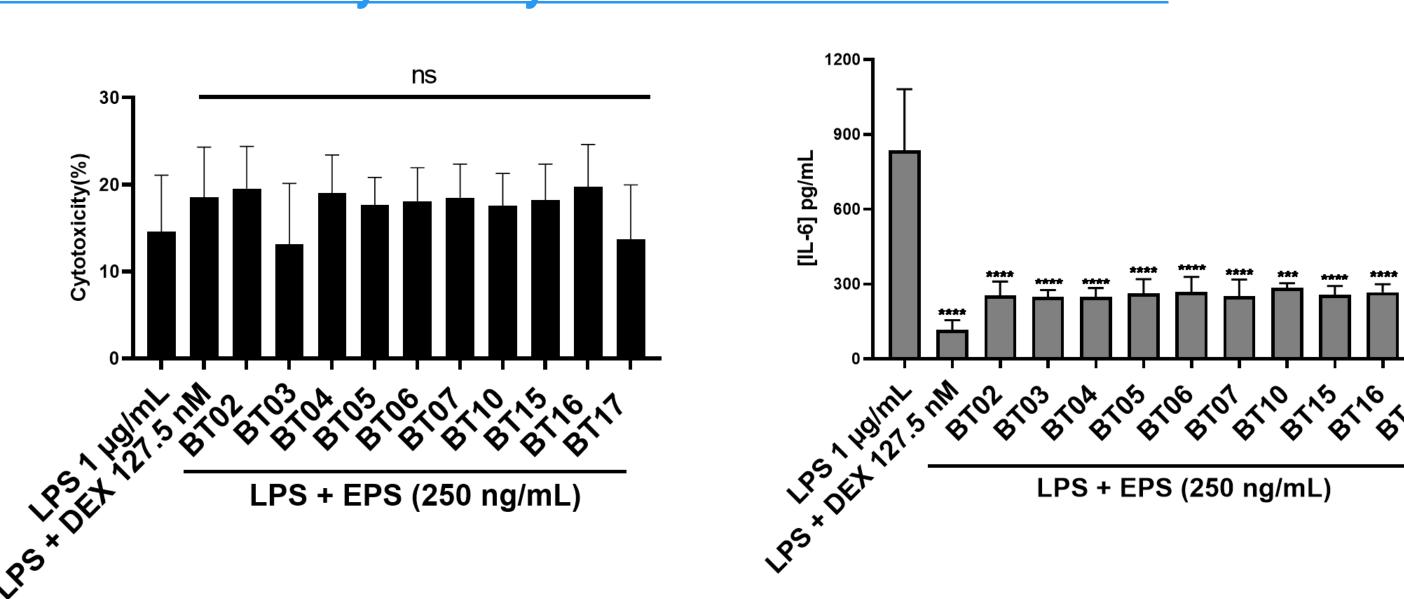


Figure 1. Inhibitory effect of 10 EPS on cytokine production (Mann Whitney Test, n=9 wells, \*p<0.05,\*\*p<0.01,\*\*\*p<0.005,\*\*\*p<0.0001,ns: non significant, DEX: dexamethasone)

- No significant cytotoxicity was observed compared to the LPS condition
- EPS have an anti-inflammatory activity on the production of IL-6 and TNF-α
- BT03, BT15 and BT17 have the most significant inhibitory activity on the production of TNF-α
- Further experiments were carried out for BT17

#### 2. <u>Bacterial identification, chemical composition and anti-inflammatory activity of BT17</u>

- 16S sequencing datas showed that BT17 is produced by a Vibrio bacteria
- N-acetyl glucosamine is predominant
- with a concentration up to 16%
   Interestingly, Vibrio alginolyticus is known to produce an anti-inflammatory EPS that also contain N-acetyl glucosamine<sup>4</sup>

	Mass percent
Glucose	
Galactose	
Glucuronic acid	
N-acetyl glucosamine	

Table 1. Chemical composition of BT17 (gas chromatography)

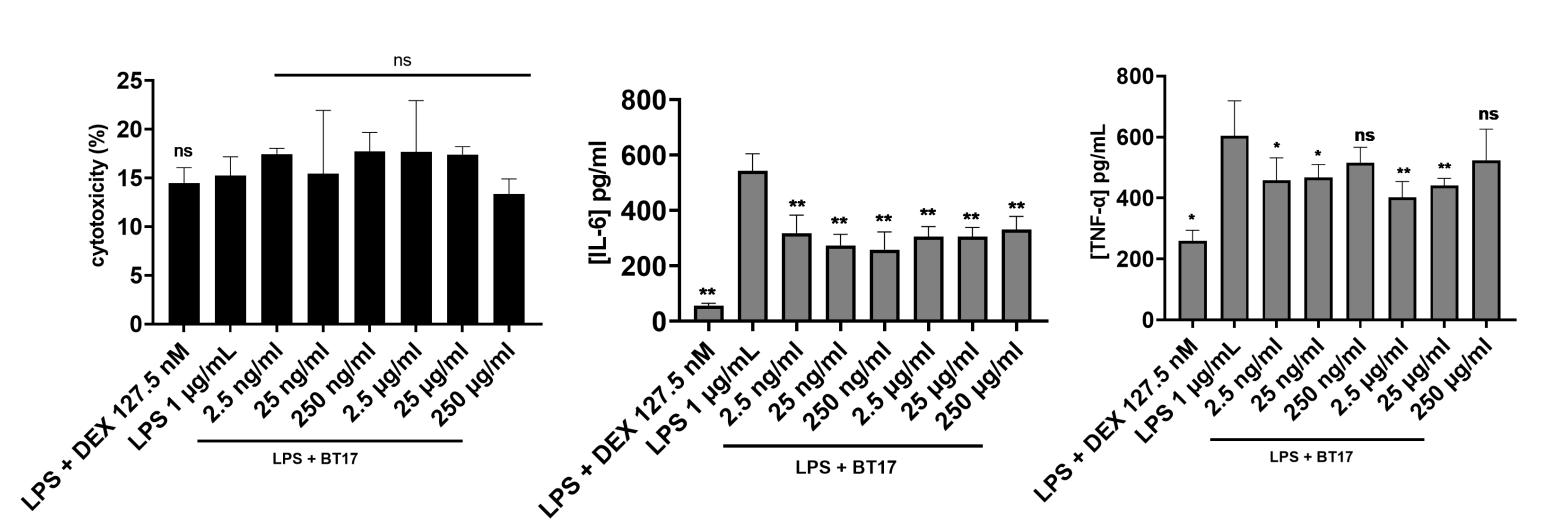


Figure 2. Inhibitory effect of BT17 on cytokine production (Mann Whitney Test, n=6 wells, \*p<0.05, \*\*p<0.01, \*\*\*p<0.005, \*\*\*\*p<0.0001,ns: non significant, DEX: dexamethasone)

- No significant cytotoxicity was observed compared to the LPS condition
  BT17 have an anti-inflammatory effect by reducing the production of
- BT17 have an anti-inflammatory effect by reducing the production of cytokines but no dose effect was observed
- Chelating metal ions is a strategy in the metalloimmunology process: interestingly, EPS are known to be potential chelators<sup>5</sup>

#### 3. BT17 is a potential chelator of metal ions

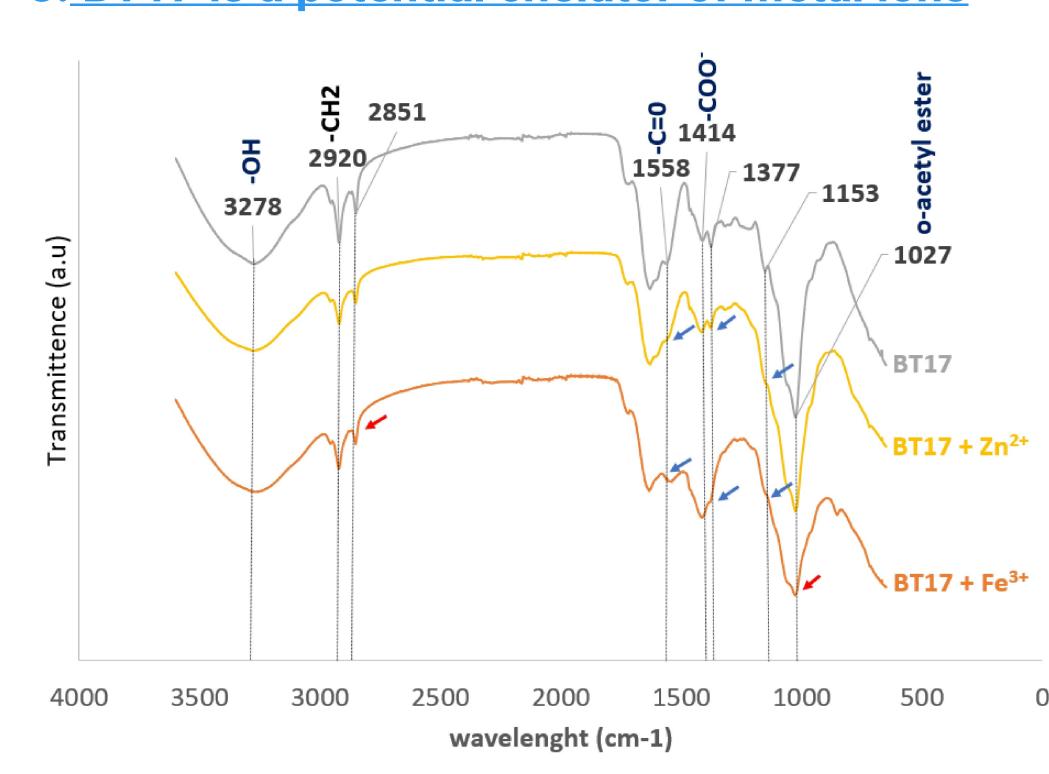


Figure 3. Fourier transform Infrared spectroscopy spectra of BT17

- Groups (in blue) have been reported to interact with metal ions (cations)<sup>5</sup>
- Shifting of peaks (red arrows) as well as changes in the intensity of the peak (blue arrows) areas suggest increase or decrease in the functional groups contents directly engaging with the metal

- Metal ions have minor changes on the conformation of the polymer
- The presence of metal in the EPS matrix was also confirmed by the EDX
- Homogeneous distribution of metal ions suggests that they are chelated by the EPS

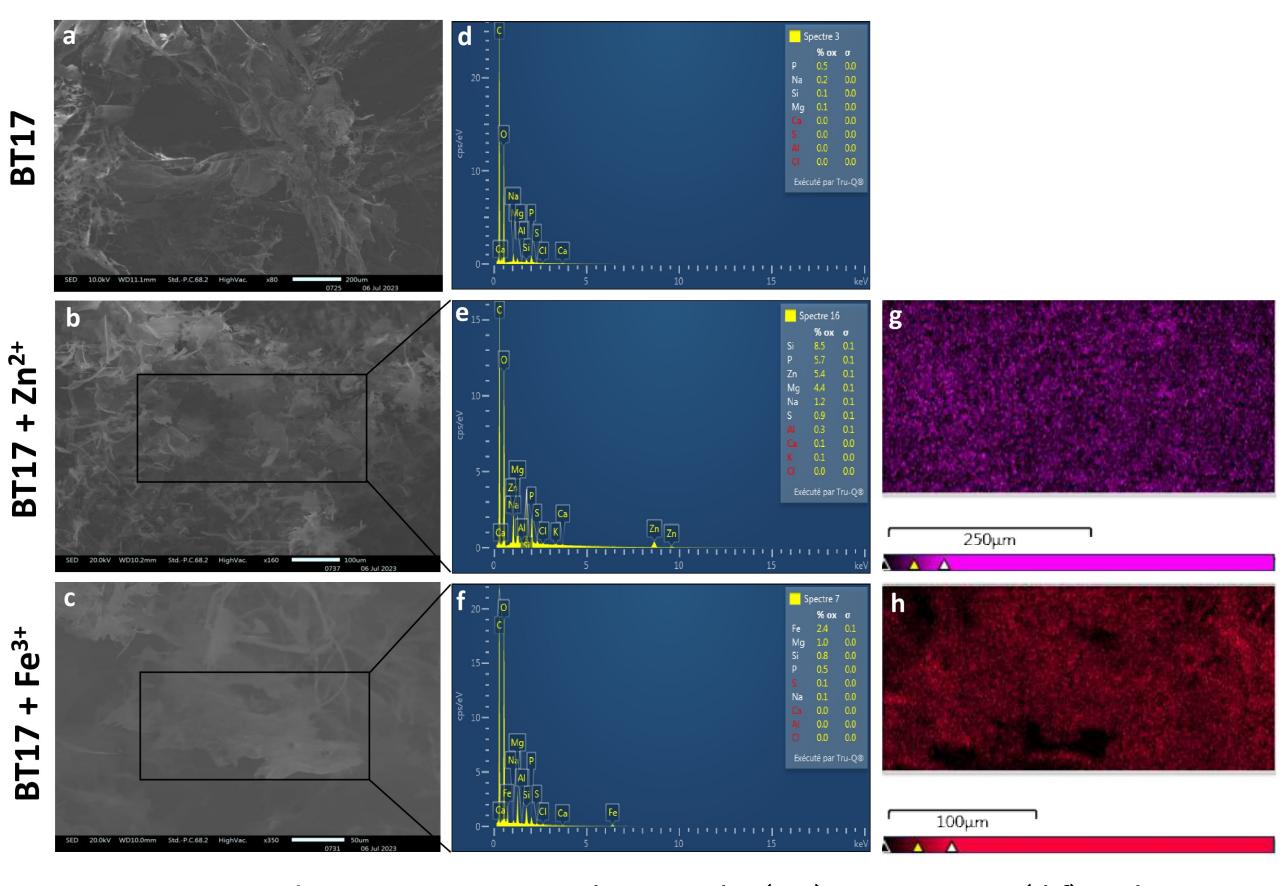


Figure 4. Scanning electron microscopy photography (a-c), EDX spectra (d-f) and cartography (g-h) of BT17 with metal ions

#### Conclusion and perspectives

- 10 bacterial EPS have an anti-inflammatory activity on the production of IL-6 and TNF-α in the PMA-THP-1 model
- Among them, BT17 is a potential chelator of metal ions
- Further experiments will be carried out to complete the results (ICP-OES...)
- Considering the importance of metal ions in immunity, BT17 could play a role in the research of novel therapeutics

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<sup>5</sup> Parmar et al., 2020, Microbiological research