

# Unveiling Host-Parasite Dynamics: Proteomic Insights into *Sacculina carcini* Parasitization of Green Crabs (*Carcinus maenas*)

## Implications for Reproduction, Immunity, and Host Manipulation

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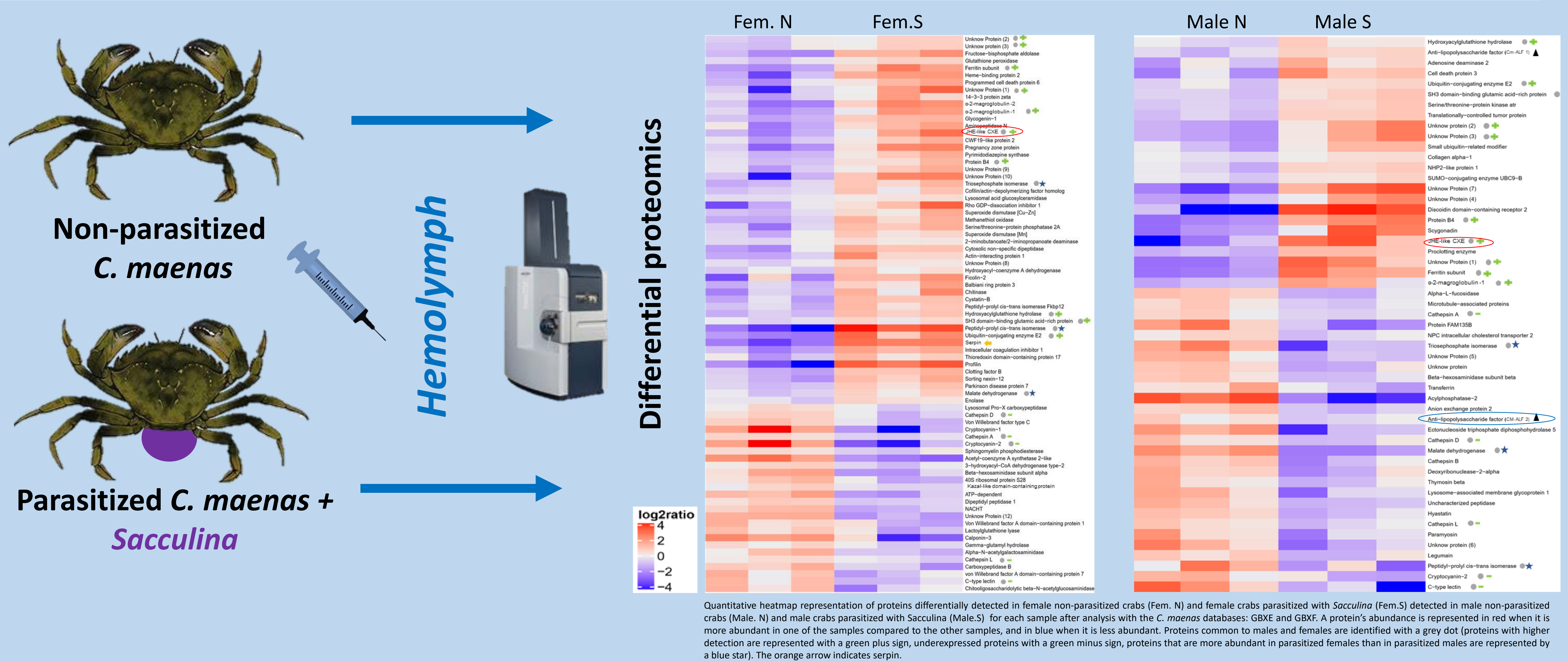
### Introduction

In its natural habitat, the green crab faces parasitization by *Sacculina carcini*, a Cirriped crustacean. This parasite alters the crab's morphology, behavior, and physiology, ultimately affecting its survival. Despite extensive observations, molecular data on this interaction are lacking. Our study aims to bridge this gap by identifying differentially expressed proteins in parasitized versus healthy crabs, focusing on the hemolymph proteome. Our results shed light on the molecular mechanisms driving the complex relationship between *Sacculina* and its green crab host.

### Materials and Methods

Crabs were collected from the Rance estuary (Normandy, France). The sample preparation involved pooling hemolymph samples from three small-sized crabs (< 3 cm wide) in each group: non-parasitized females, non-parasitized males, parasitized females, and parasitized males, resulting in nine individuals per group.

Hemolymph samples were analyzed using mass spectrometry. The PeaksXPro software conducted a database search using MS/MS fragmentations from the GBXE and GFXF transcriptomes of *C. maenas*, encompassing various tissues, followed by independent analysis and subsequent data grouping for relative quantification using label-free XIC. Protein annotation involved blasting proteins with differential expression in parasitized crabs against NCBI databases, and conducting a targeted search for proteins associated with molting, reproduction, and immune responses to confirm their presence in all samples.



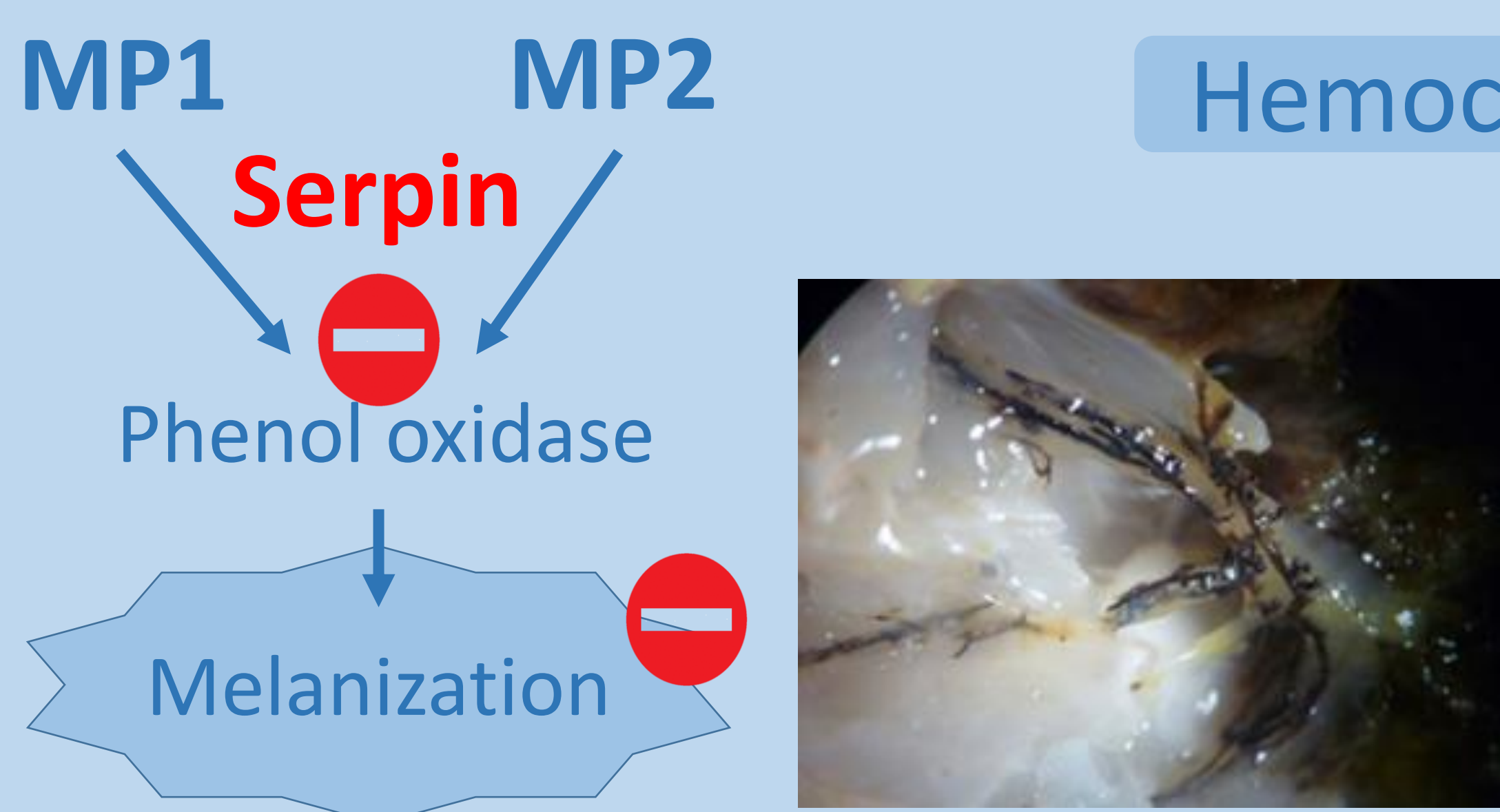
### Results

The mass spectrometry analysis of *C. maenas* hemolymph identified 1652 proteins in the GBXE database and 1934 proteins in the GFXF database, revealing differences in protein abundance between parasitized and non-parasitized crabs. Proteins involved in the immune response, such as α2 macroglobulin 1, superoxide dismutase, and ferritin, showed differential abundance. The presence of serpin in female parasitized crabs, coupled with the absence of melanization-related proteins, suggests a potential manipulation by the parasite to inhibit key defense pathways (e.g. melanization), rendering the host more susceptible to parasitic infestation. Also, proteins involved in regulation of molting and reproduction, such as juvenile hormone esterase-like carboxylesterase (JHE-CXE, red circle), which was 17-fold higher, were notably more abundant in parasitized crabs, suggesting potential manipulation by the parasite to inhibit molting and reproduction through increased metabolism of methyl farnesoate. This indicates a potential mechanism for the parasite to influence host physiology, including molting and reproductive cycles, highlighting the complex interplay between host and parasite in this relationship.

#### Modulation of the immune response

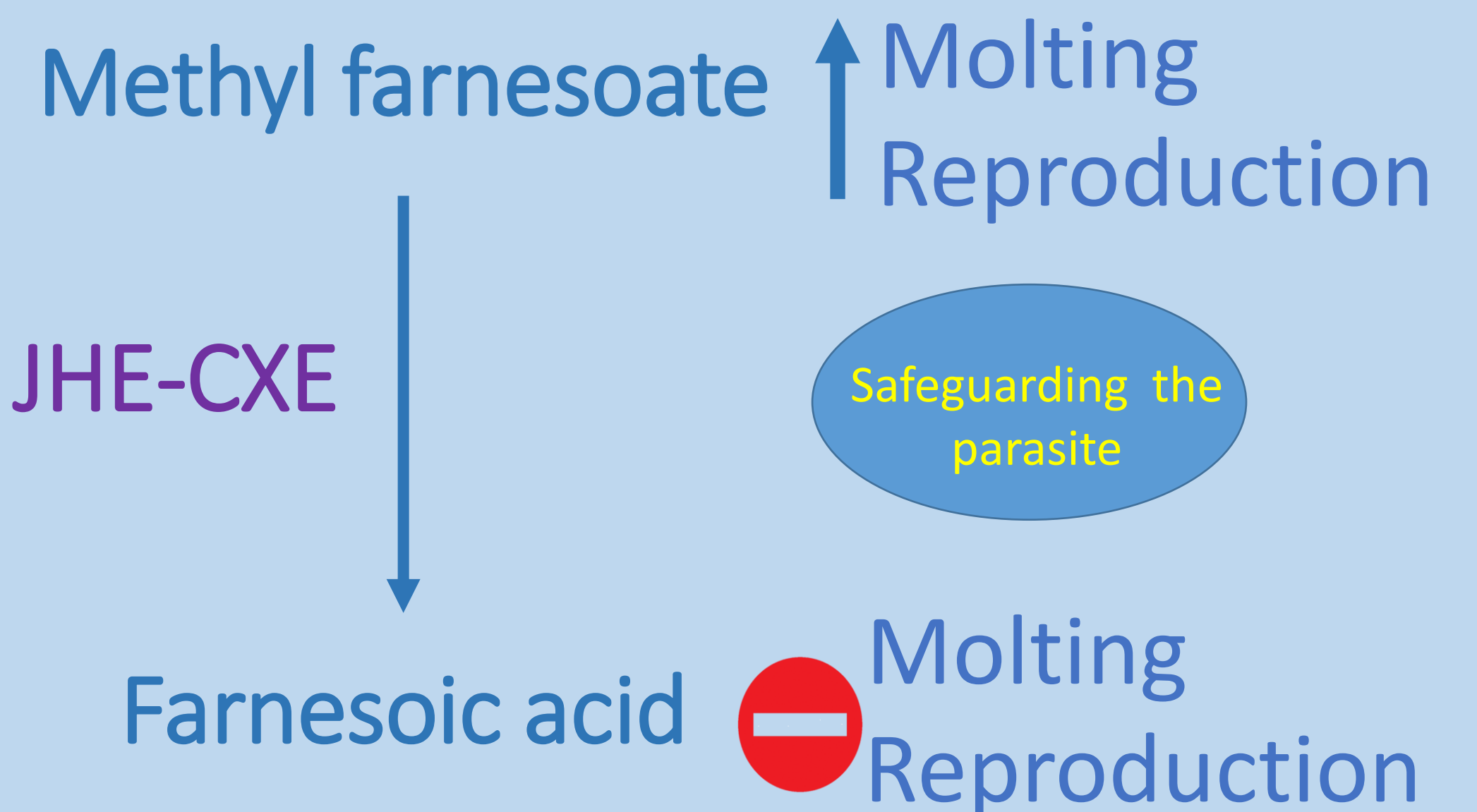
##### Inhibition of melanization

##### Secretion of immune factors



#### Inhibition of Molting and Reproduction

##### Secretion of Juvenile Hormone Esterase



### Conclusion

The identification of proteins in the proteome of *C. maenas*, whose expression is influenced by the presence of *S. carcini*, sheds light on the significant impact of parasitic secretions on host immune and reproductive processes. This study unveils how *Sacculina* subtly controls its host's immune response by modulating the secretion of target proteins, such as antimicrobial peptides and proteins involved in melanization. Furthermore, the high levels of crustacean JHE-CXE in parasitized crabs suggest a mechanism for inhibiting host molting and reproduction. Thus, by altering its host's proteome to its advantage, the parasite ensures its own protection and reproduction, offering a fascinating glimpse into the molecular intricacies of host-parasite interactions.