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Influence of solvents on the extraction efficiency and biological activity of marbled eel mucus

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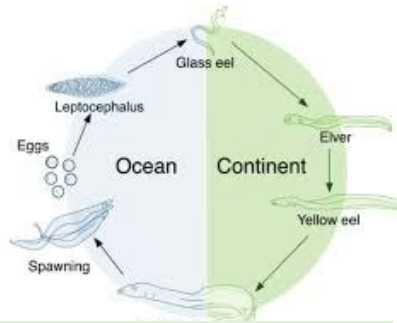
1. INTRODUCTION

- *Anguilla marmorata* is a catadromous life species.
- Eel mucus has a high biological compounds related to the body's natural immune activity in protecting against environmental pathogens.
- Vibrio bacteria are a common pathogen in aquatic animals.
- The composition and biological activity of mucus depend on endogenous and exogenous factors.
- The extraction process also affects the antibacterial ability of mucus.



This study aimed to investigate the effect of solvents on extracting bioactive compounds from marbled eel mucus for both aquaculture and human healthcare applications.

2. MATERIALS AND METHODS

2.1. Materials

MARBLED EEL AND CRUDE MUCUS EXTRACT (CM)

MUCUS EXTRACT

ME-1(Acetone)
ME-2 (Chloroform)
ME-3 (ethanol)
ME-4 (methanol)

BACTERIA

The extracts have 19-23 other peaks of compounds, with retention times ranging from 0.8 to 60.0 minutes on high-performance liquid chromatography (HPLC

2.2. Collect crude of mucus and extract mucus

Marbled eel

Crude skin mucus

Freeze dryer

Freeze-dried sample

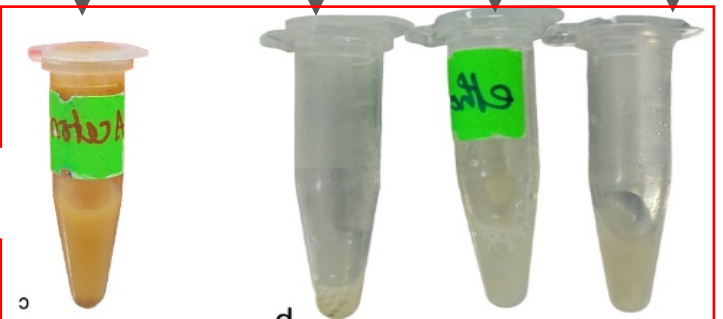
NaCl 0.9%

CM

AOAC, 2005

HPLC

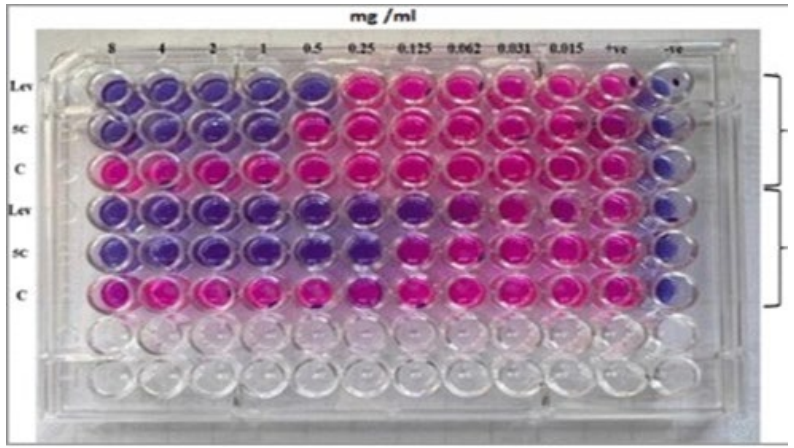
Acetone Chloroform ethanol methanol



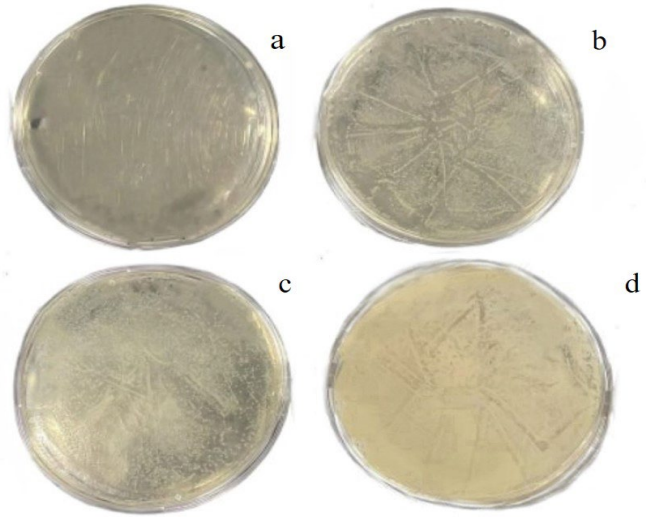
ME-1 ME-2 ME-3 ME-4

2.3. Activity methods

Antibacterial activity test
(Turker et al., 2009)



MIC determination
CLSI (2012)



Determine MBC
CLSI (1998)

2.3. Analysis data methods

Activity test

The selective extraction rate of components (% area) has significant differences between extracts with different solvents.

Faikoh et al., 2014

No.	Diameter of antimicrobial ring (mm)	Level of antimicrobial resistance
1	≥ 15	Strong (***)
2	10 – 14	Medium (**)
3	< 9	Weaky (*)
4	0	None

MBC/MIC

Canillac and Mourey (2001)

No.	BMC/ MIC	Antimicrobial ability
1	≥ 4	Antibacterial
2	< 4	Bacteriostatic
3	-	None

3. RESULTS

3.1. Components of crude mucus

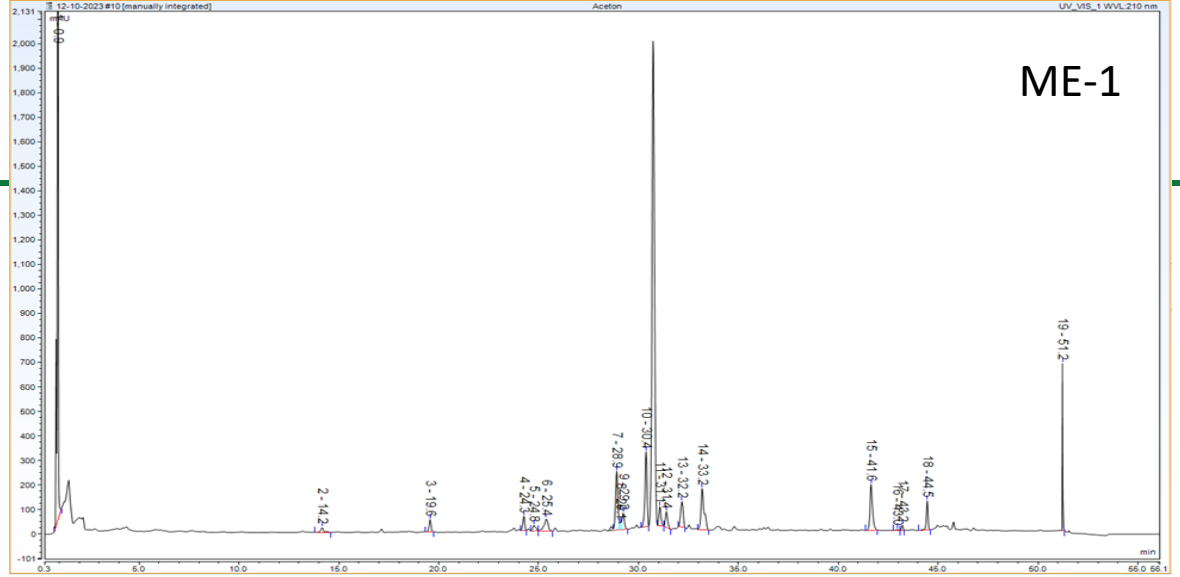
Table 1. Components of *A. marmorata* mucus

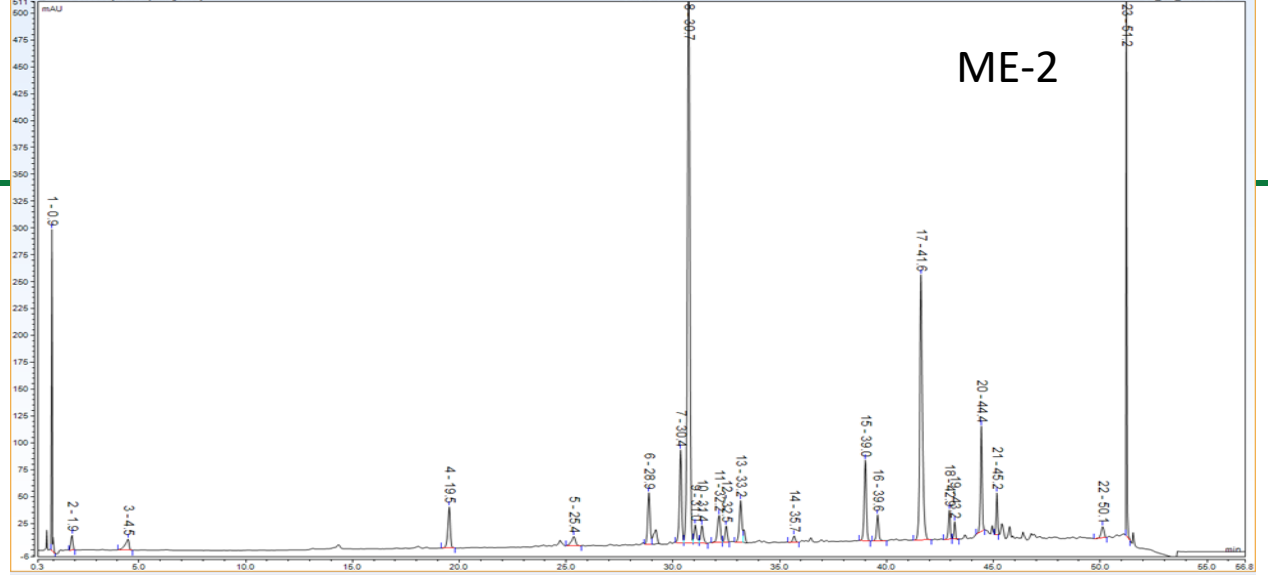
No.	Components	%
1	Total protein	13.74 ± 0.098
2	Total lipid	9.14 ± 0.038
3	Crude ash	6.83 ± 0.094
4	Moisture	76.29 ± 0.021

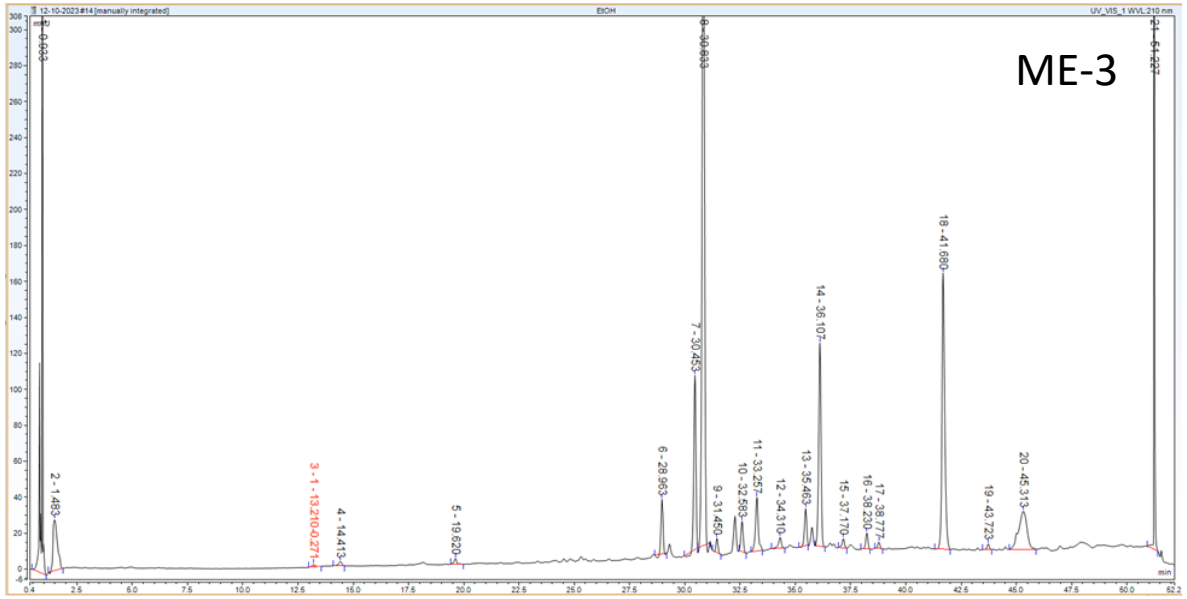
3.2. Recovery efficiency and biological compounds in mucus extracts

Table 2. Recovery efficiency of mucus extracts

Index	ME-1	ME-2	ME-3	ME-4
RE (%)	18.475	18.45	56.025	23.375







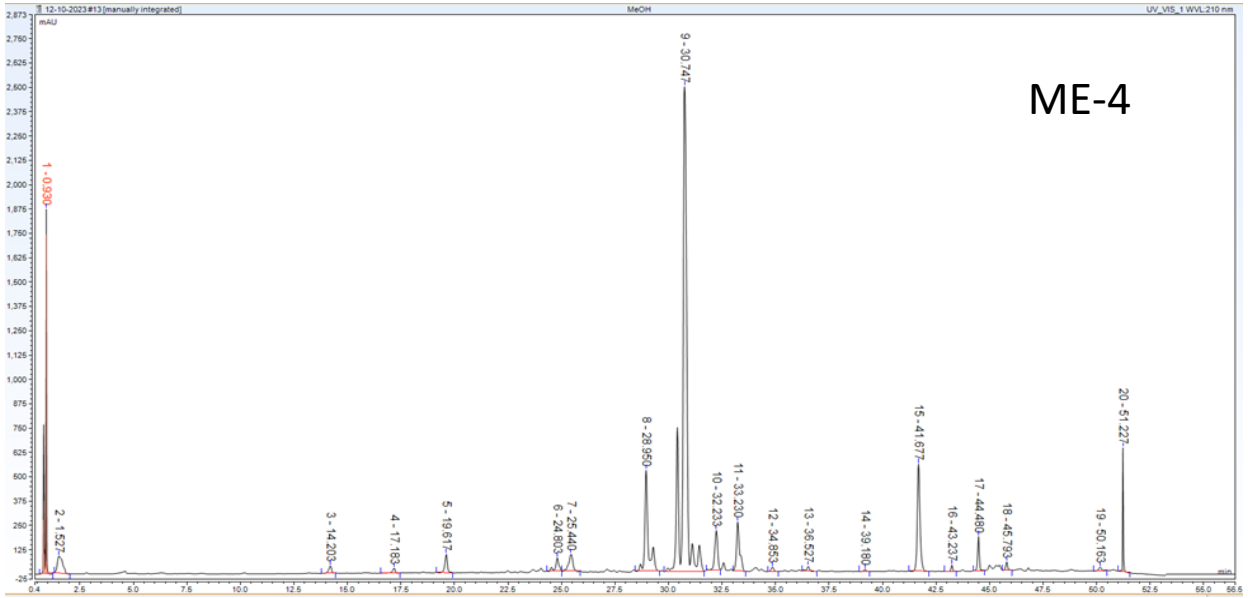


Figure 2. The biological compounds in marbled eel mucus extracts

3.3. Antibacterial ability of eel mucus

Table 3. Diameter of antibacterial zone of eel mucus (mm)

The eel mucus extracted with acetone solvent exhibited the highest biological activity in all tests: IC50 = 36.603 mg/mL and SC% = 98.424% at 100 mg/mL for antioxidant activity; Antibacterial activity was demonstrated against *S. aureus* (IC50 = 65.00 ± 3.01 µg/mL) and *B. subtilis* (IC50 = 112.00 ± 7.89 µg/mL);

NT	*V. parahaemolyticus*	*V. alginolyticus*	*V. harveyi*	*P. damselae*
CM	11.7 – 12.3 (12 ± 0.3)	9.7 – 10.3 (10 ± 0.3)	17.6 – 18.3 (17.96 ± 0.2)	15.6 – 16.3 (16.0 ± 0.35)
ME-1	-	-	12.0 – 13.0 (12.5 ± 0.5)	11.7 – 12.2 (12 ± 0.25)
ME-2	-	11.6 – 12.2 (11.9 ± 0.3)	9.5 – 10.2 (9.9 ± 0.36)	11.4 – 12.3 (11.9 ± 0.45)
ME-3	10.8 – 11.3 (11.03 ± 0.25)	16.9 – 17.4 (17.1 ± 0.26)	13.7 – 14.5 (14.06 ± 0.4)	13,7 – 14.2 (14.0 ± 0.25)
ME-4	8.5 – 9.2 (8.9 ± 0.36)	-	-	-
Amociciline	17.6 – 18.2 (17.93 ± 0.3)	31.6 – 32.7 (32.1 ± 0.5)	19.7 – 12.3 (20.0 ± 0.3)	16.8 – 17.2 (17.0 ± 0.2)
DMSO5%	-	-	-	-

Table 4. The MIC values of mucus extracts (µg/mL)

The ability to inhibit human skin cancer cells (IC50 = 465.96 ± 8.54 µg/mL) and normal cells (IC50 = 453.36 ± 17.88 µg/mL).

NT	*V. parahaemolyticus*	*V. alginolyticus*	*V. harveyi*	*P. damselae*
ME-1	-	-	100	50.0
ME-2	-	100	-	100.0
ME-3	12.5	12.5	25	12.5
ME-4	100	-	-	-
CM	100%	100%	100%	100%

Table 5. The MBC value (µg/mL) of mucus extracts

Extracts extracted with n-Hexane solvent showed higher safety for cells than extracts from other solvents (IC50 = 632.43 ± 18.80 µg/ml, higher than the value IC50 = 545.25 ± 26.36 µg/ml for skin cancer cells).

NT	*V. parahaemolyticus*	*V. alginolyticus*	*V. harveyi*	*P. damselae*
ME-1	-	-	100	100
ME-2		100	-	100
ME-3	100	100	100	100
ME-4	100	100	-	-

Table 6. The BMC/MIC ratio of mucus extracts on *Vibrio* bacteria

These results provide essential data for selecting and applying products derived from extracts of marbled eel mucus in the future, sparking excitement and optimism about this research's potential impact on aquaculture and human health care.

NT	*V. parahaemolyticus*	*V. alginolyticus*	*V. harveyi*	*V. damselae*
ME-1	-	1	-	1
ME-2	-	-	1	2
ME-3	8	8	4	8
ME-4	1	-	1	2

4. CONCLUSION

- Eel mucus has antibacterial properties against some *Vibrio* bacteria in aquaculture
- Extraction solvent affects antibacterial properties and composition of mucus extracts
- The mucus extract from ethanol solvent (ME-1) has the highest antibacterial properties against the tested *Vibrio* bacteria
- Research on fish mucus extraction methods and antibacterial mechanisms is necessary.
- The results confirmed that eel mucus is a valuable source for protecting aquatic animal health and can apply to human skin care.
- It has opened up new potentials in nutritional research, application, and exploitation of the pharmaceutical value of the marbled eel.

Products from study will be commerce soon

THANKS FOR YOUR ATTENTION!