Coral nutrition, defense and adaptation to changing environment: the role of it's associated microbial community

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June 2012

The Environmental Biotechnology Lab at BGU

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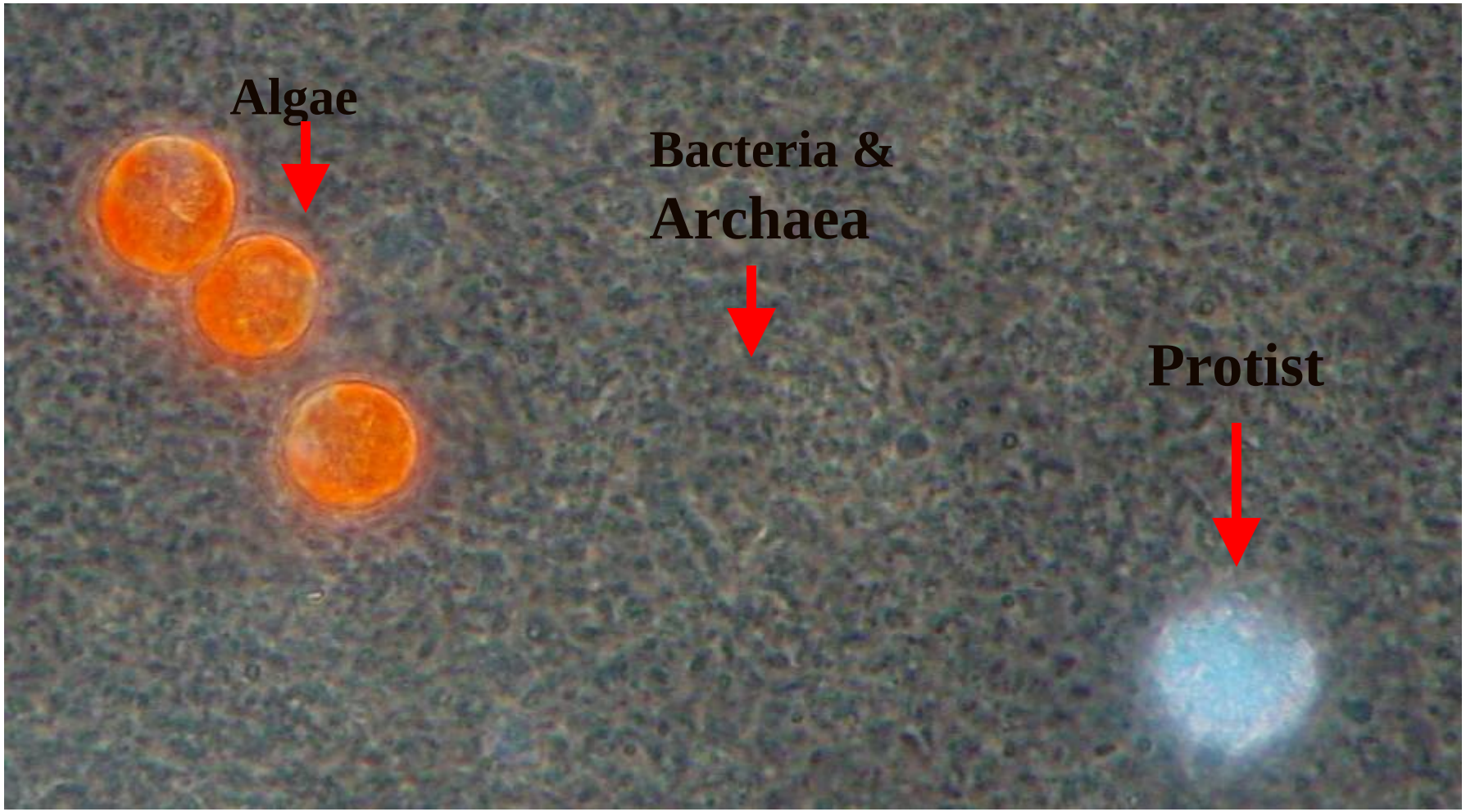
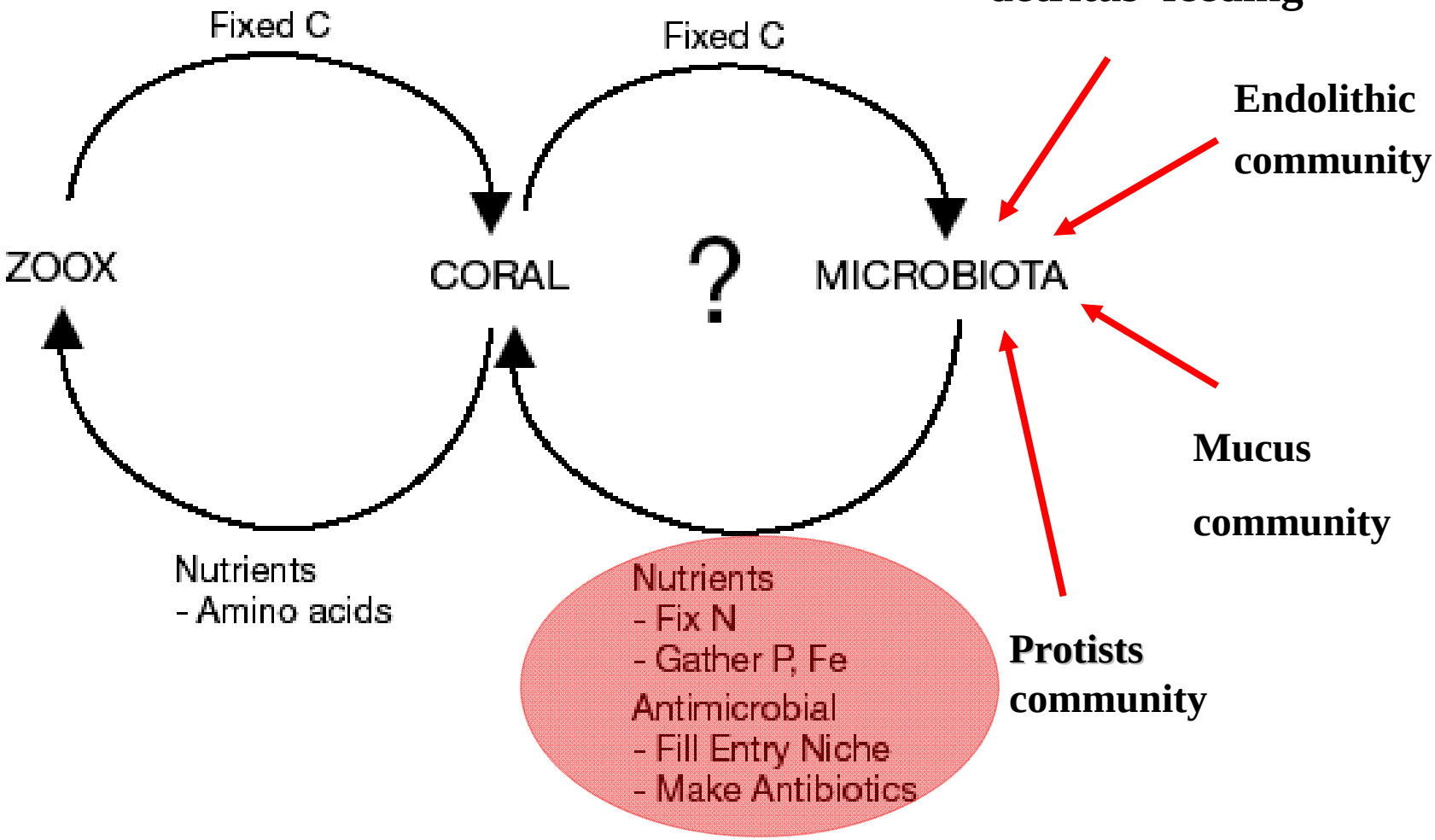
Outline

- Holobiont concept
- Microbial shift
- Bacterial contribution to coral health
- Coral diseases
- Coral Probiotic hypothesis
- Conclusions

Coral mucus (X1000)



Coral holobiont



Rohwer et al.: Coral-associated bacteria

Mar Ecol Prog Ser 243: 1–10, 2002

Shifting of microbial populations

Change in

Environmental conditions

Coral's physiological state

Mucus composition changes

Microbial communities change

Bacterial contribution to coral nutrition

- Nitrogen & Phosphorous
- Corals -passive suspension feeders – ingest:
  - Bacteria attached to particulate food
  - Suspension bacteria trapped in mucus
  - Bacterial aggregates
- Nitrogen fixation (acetylene reduction)

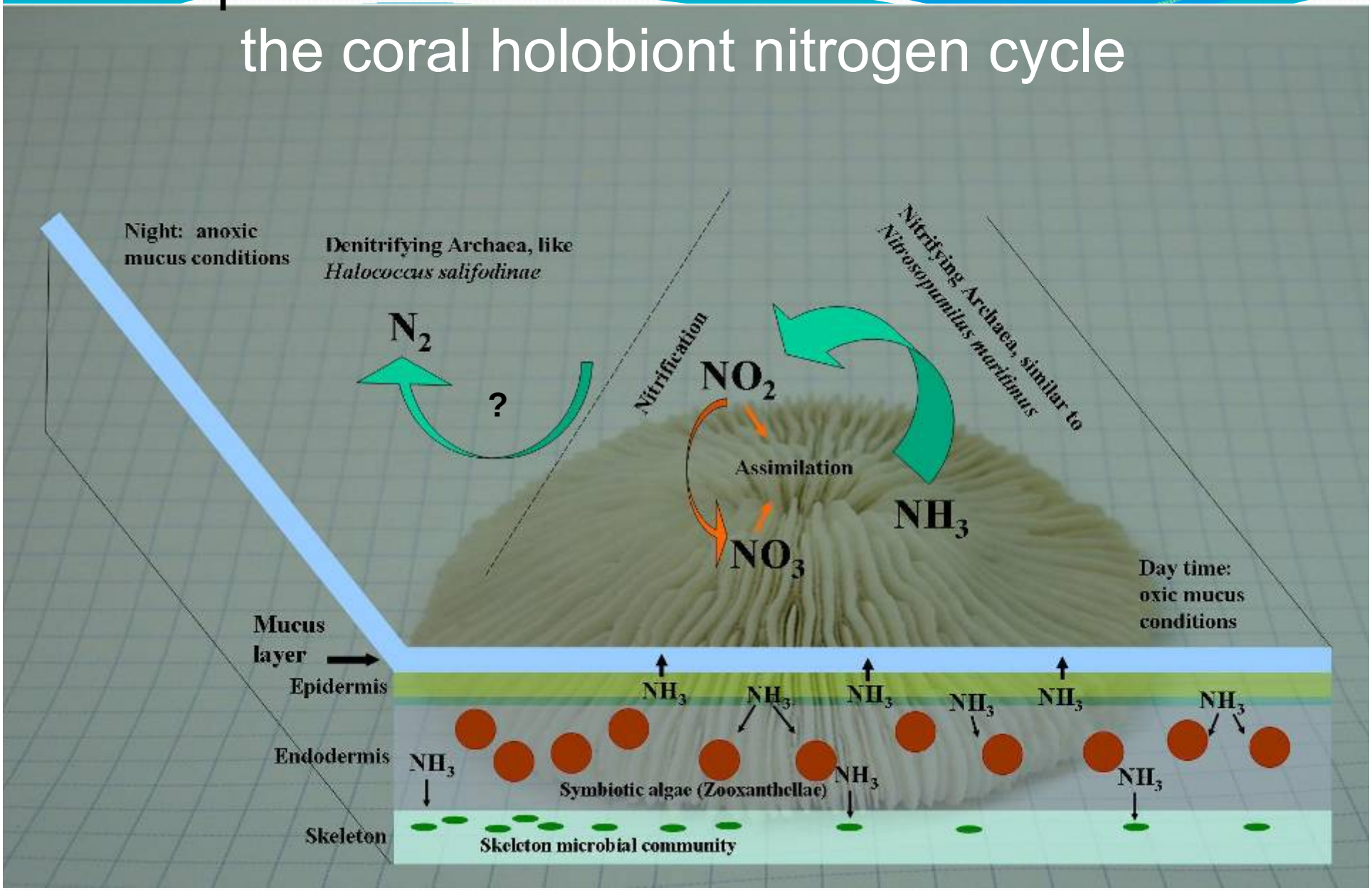
Bacterial contribution to coral health

- Nutritional contribution
- Antioxidant activity (catalase, superoxide dismutase)
- Sunscreen protection
- Antimicrobial activity

Coral-associated Archaea: Major players in the coral holobiont nitrogen cycle

Corals samples: *Acanthastrea* sp., *Favia* sp. and *Fungia* sp.

Proposed model for the role of Archaea in the coral holobiont nitrogen cycle

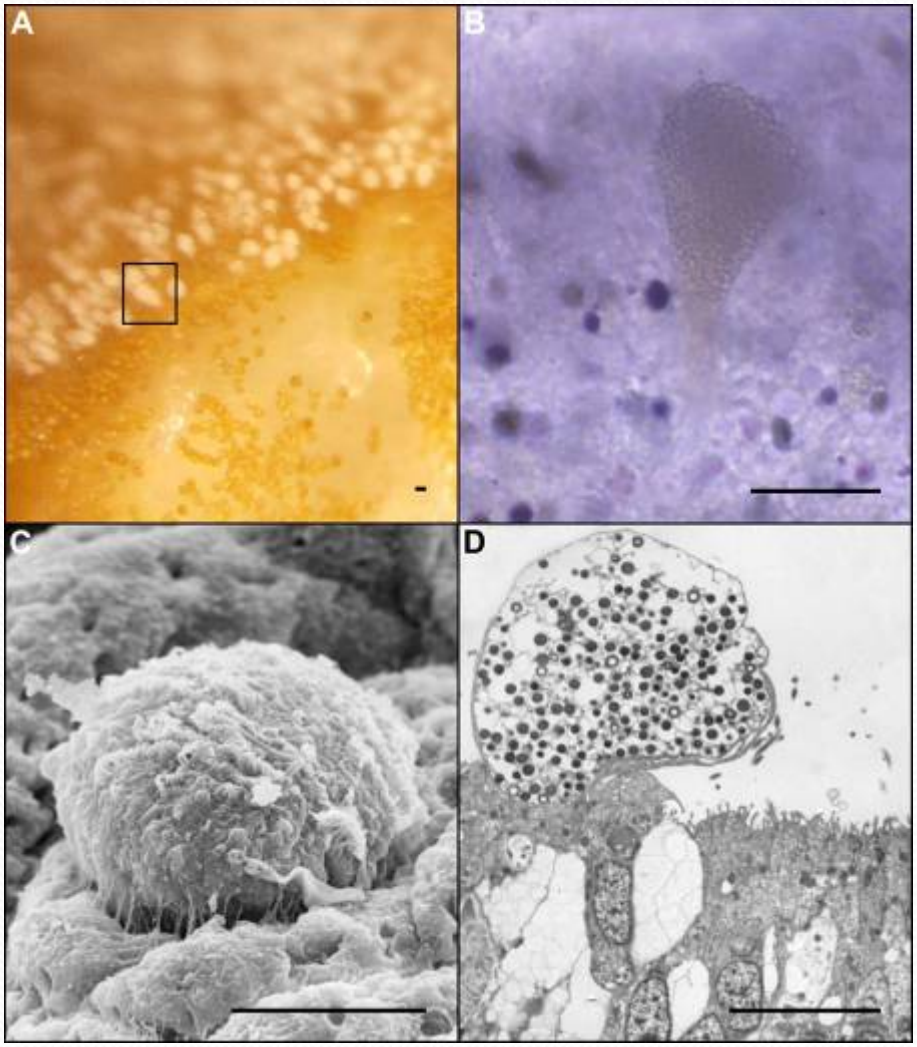


Siboni et al. (2008), *Environmental Microbiology*

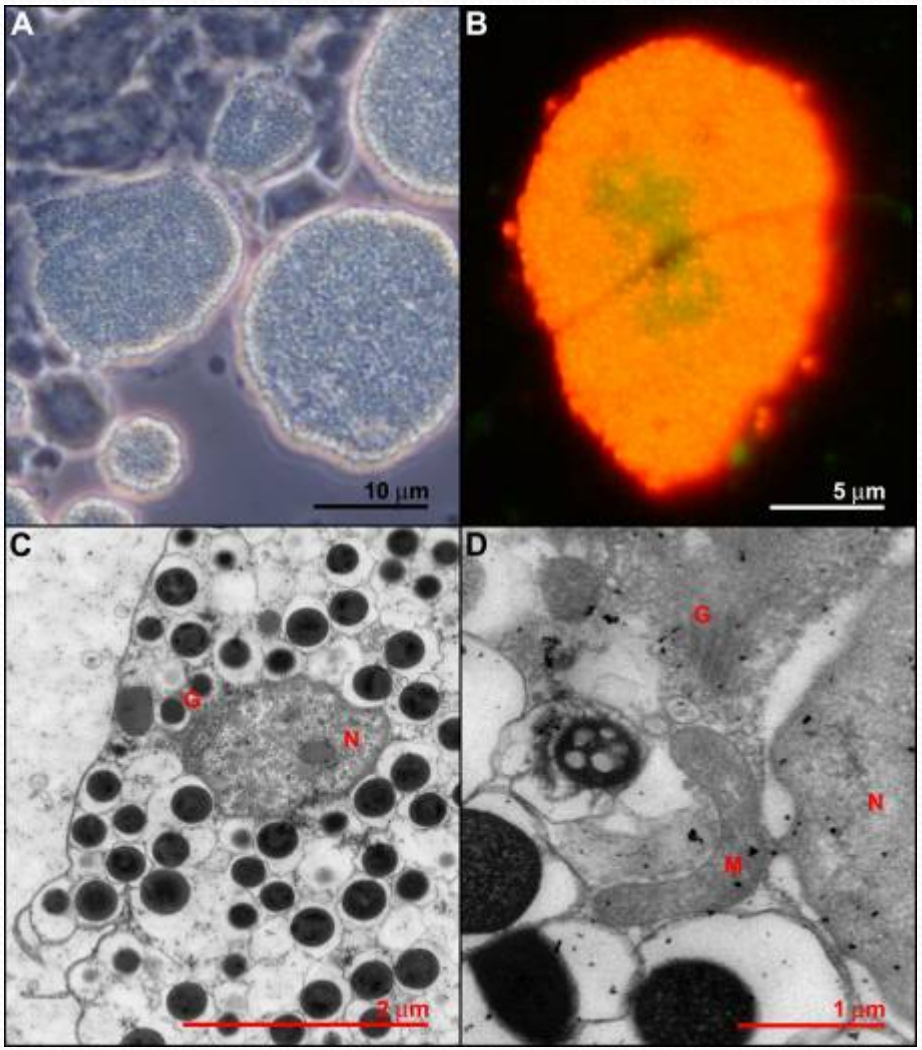
N. Siboni

Protists in corals

(A) Protist in the epidermis of *Fungia granulosa*. Black box denotes a single aggregate. (B) Photomicrograph of a single protist; (C) SEM of a protist in epidermis; (D) TEM of a protist showing its nucleus.



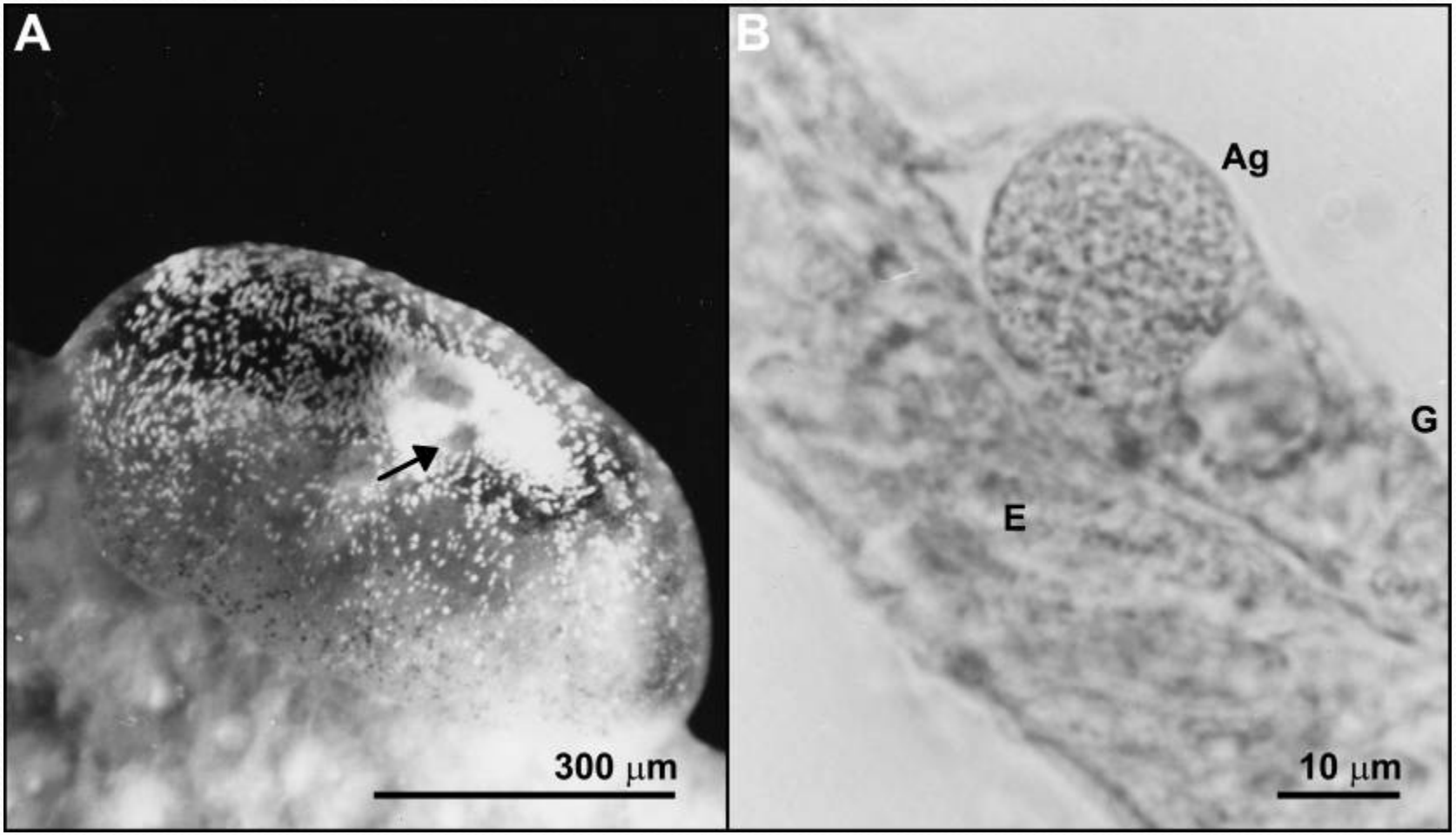
Protist from *Fungia granulosa* coral mucus. (A) Phase contrast micrograph; (B) Fluorescent micrograph; (C) TEM of a single protist; (D) Higher magnification TEM; nucleus (N), golgi (G), mitochondrion (M)



Kramarsky-Winter et al. 2006, Mar Ecol Prog Ser

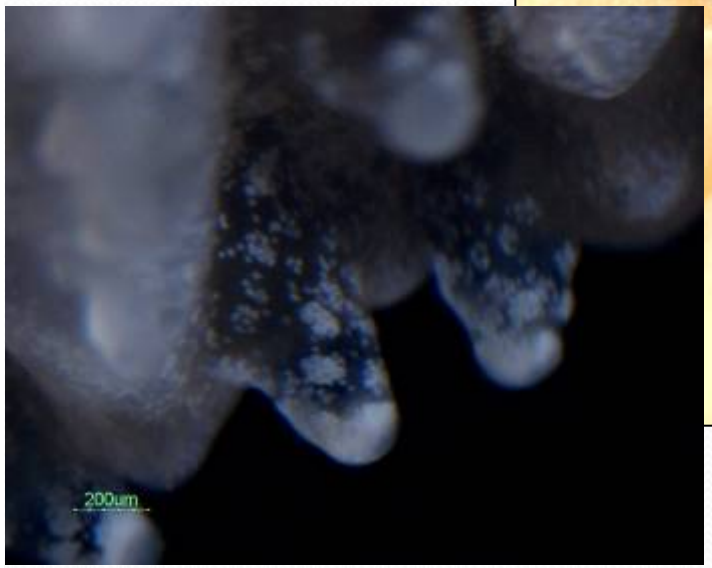
Protists in corals

Primary polyp developing from tissue fragments of *Fungia granulosa*. (A) Aggregates are dispersed on the body surface, as fluorescing bodies. Note the high density surrounding the developing mouth (arrow). (B) *F. granulosa* histological section demonstrating an aggregate (Ag) in the coral gastroderm (G). E: coral ectoderm

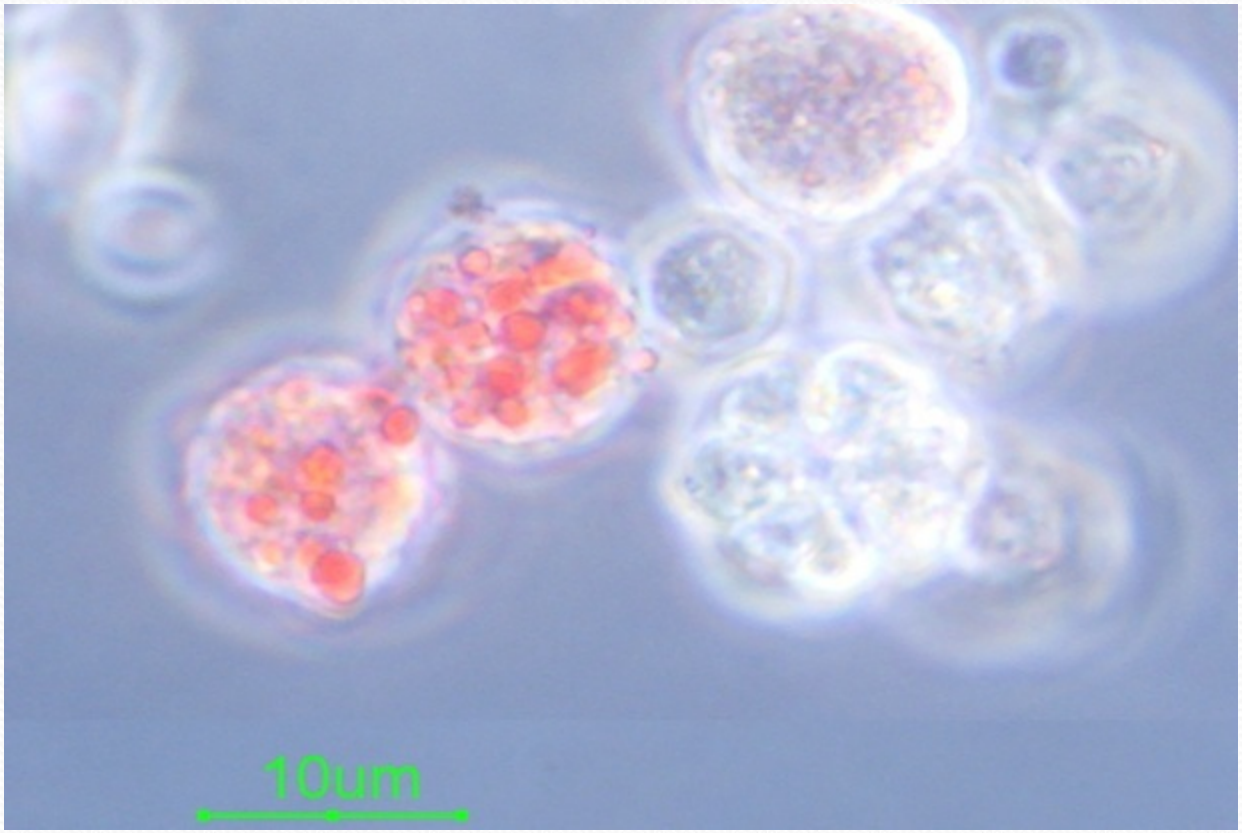


Kramarsky-Winter et al. 2006, Mar Ecol Prog Ser

Fungia granulosa after six months of complete darkness



Coral-associated Thraustochytrid fatty acids



Natural Quorum Sensing Inducers and Inhibitors from Coral Associated Bacteria

K. Golberg, E. Eltzov. M. Shnit- Orland, R. S. Marks and A. Kushmaro. 2011. Characterization of Quorum Sensing Signals in Coral Associated Bacteria Microbial Ecology 61(4): 783-92.

Coral diseases

Pathogenic microorganism Environmental stress

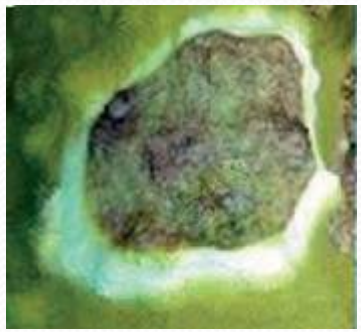
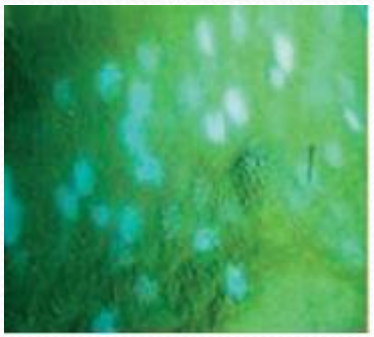
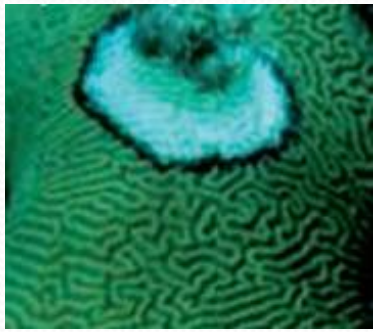
↓ ↓

Weakening host's innate immune system

↓

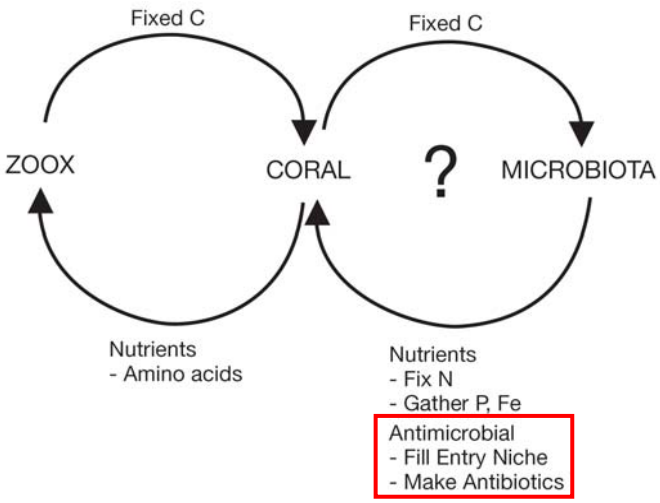
Coral diseases

Coral bleaching | White plague | Yellow blotch | White pox | Black band disease (BBD)

Rosenberg et al., 2007

The Coral *Holobiont*



Rohwer et al., 2002

The Coral Probiotic Hypothesis

Reshef et al., 2006

http://www.clipartguide.com/cgi-bin/images_clipartguide.pl

Coral Probiotic hypothesis (Reshef et al., 2006)

- Symbiotic relationship between the coral and a diverse metabolically active microorganisms

- Environmental conditions change

↓

Relative abundance of microbial species changes

- Coral holobiont adapt to the new conditions more efficiently

Hypothesis

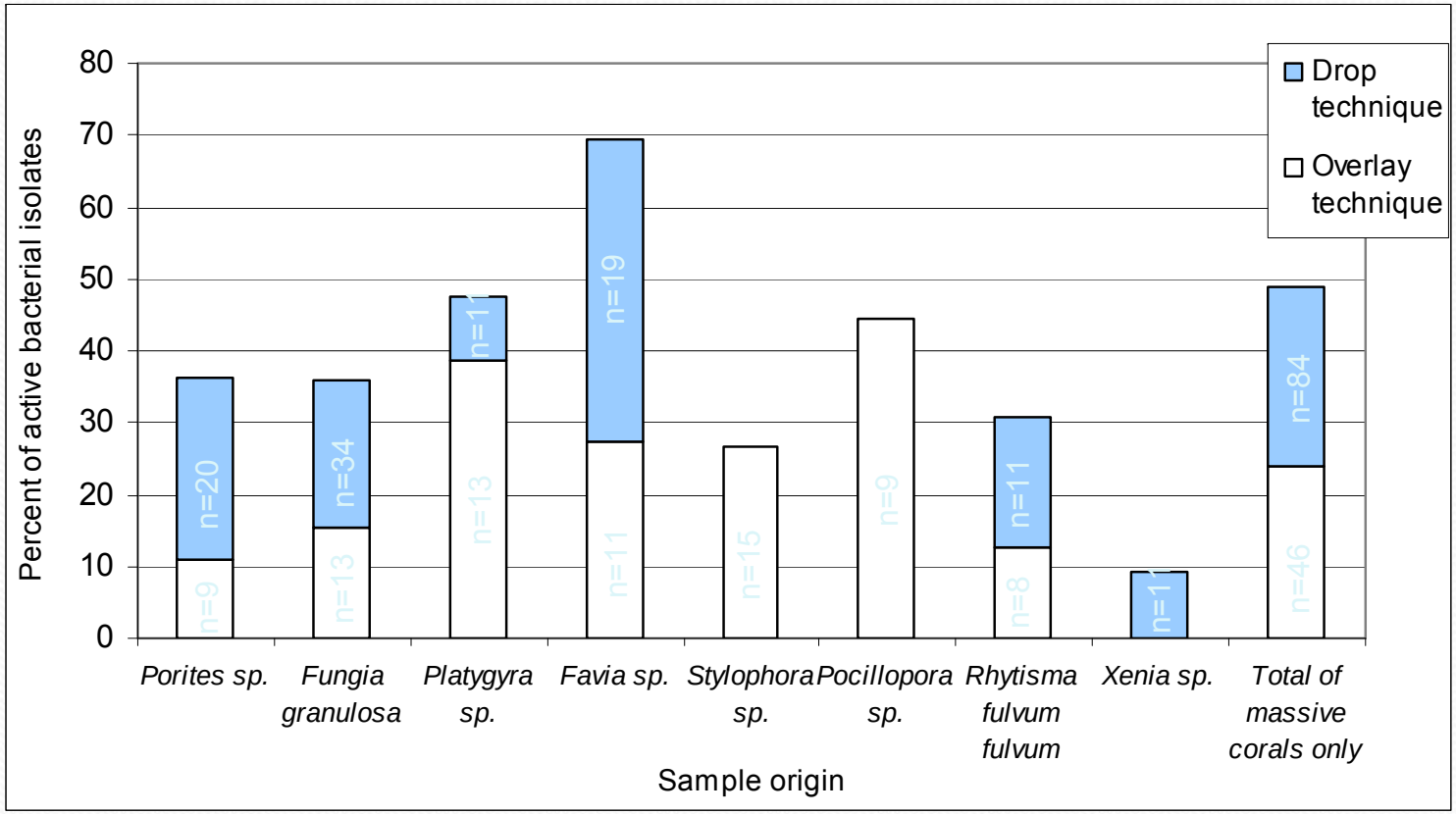
Coral mucus-associated bacteria take part in coral defense strategy

Methods

- Sampling mucus-associated bacteria, sea water & sediment
- Dilution & agar plating
- Screening techniques: overlay, drop
- Molecular techniques: DNA extraction, PCR, sequencing, phylogenetic analysis
- Characterization of antibacterial activity
- Chemical extraction

Antibacterial activity method comparison

- Percent of active bacterial isolates from screened coral colonies, obtained using overlay or drop techniques.
- n = number of screened bacterial isolates



Shnit-Orland & Kushmaro, 2009 *FEMS Microbiol Ecol*

Antibacterial activity method comparison

Comparison between stony coral isolate activities

	No. of screened isolates	No. of active isolates	Percent of activity
Drop	84	21	25%
Overlay	78	19	24.4%

- No activity overlap (except one isolate)
- Only few of bacterial supernatants exhibited antibacterial activity using drop technique

Different mechanisms:

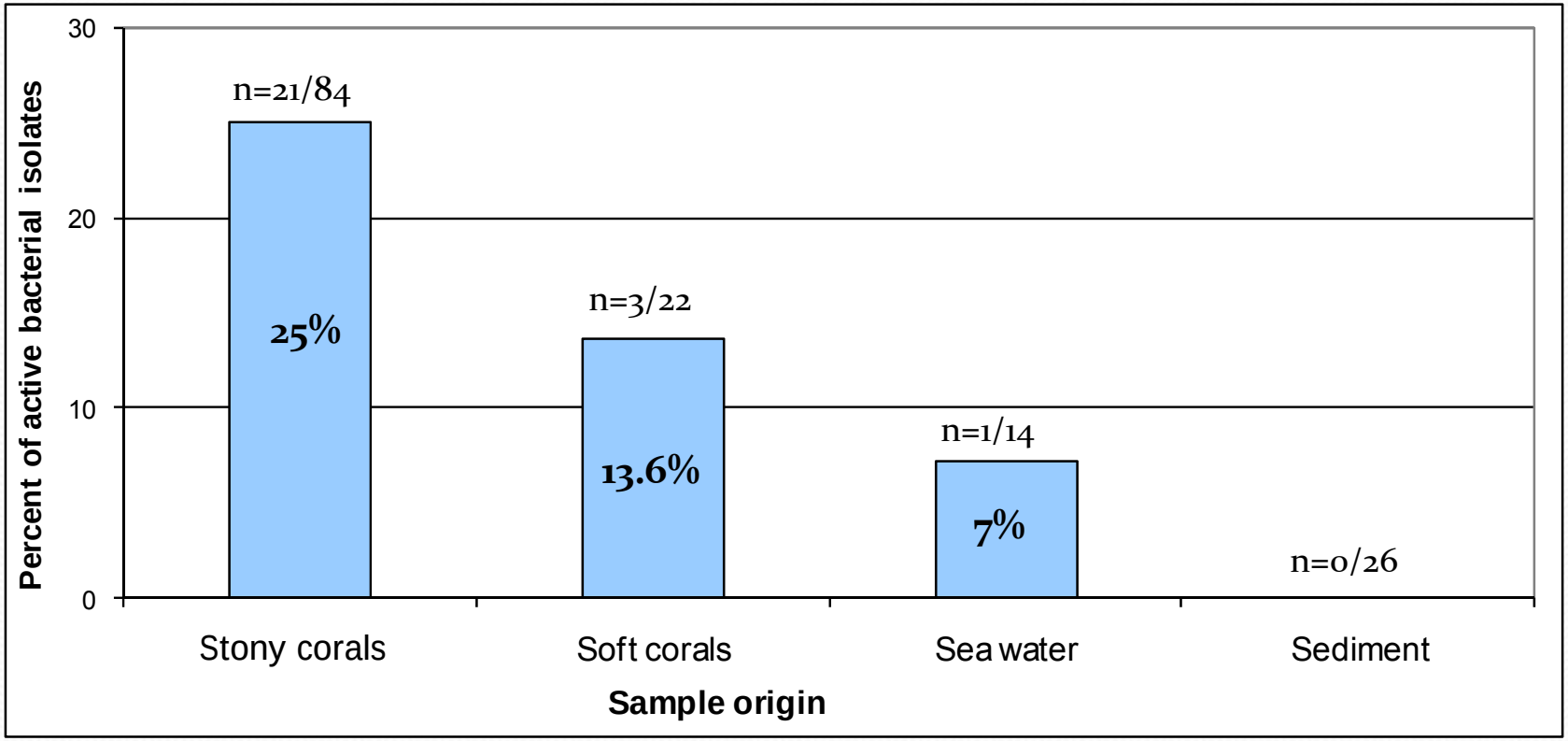
drop + overlay = **50% of isolates active**

(in stony corals' isolates)

Antibacterial activity sample-origin comparison

- 146 isolates were screened using the drop technique

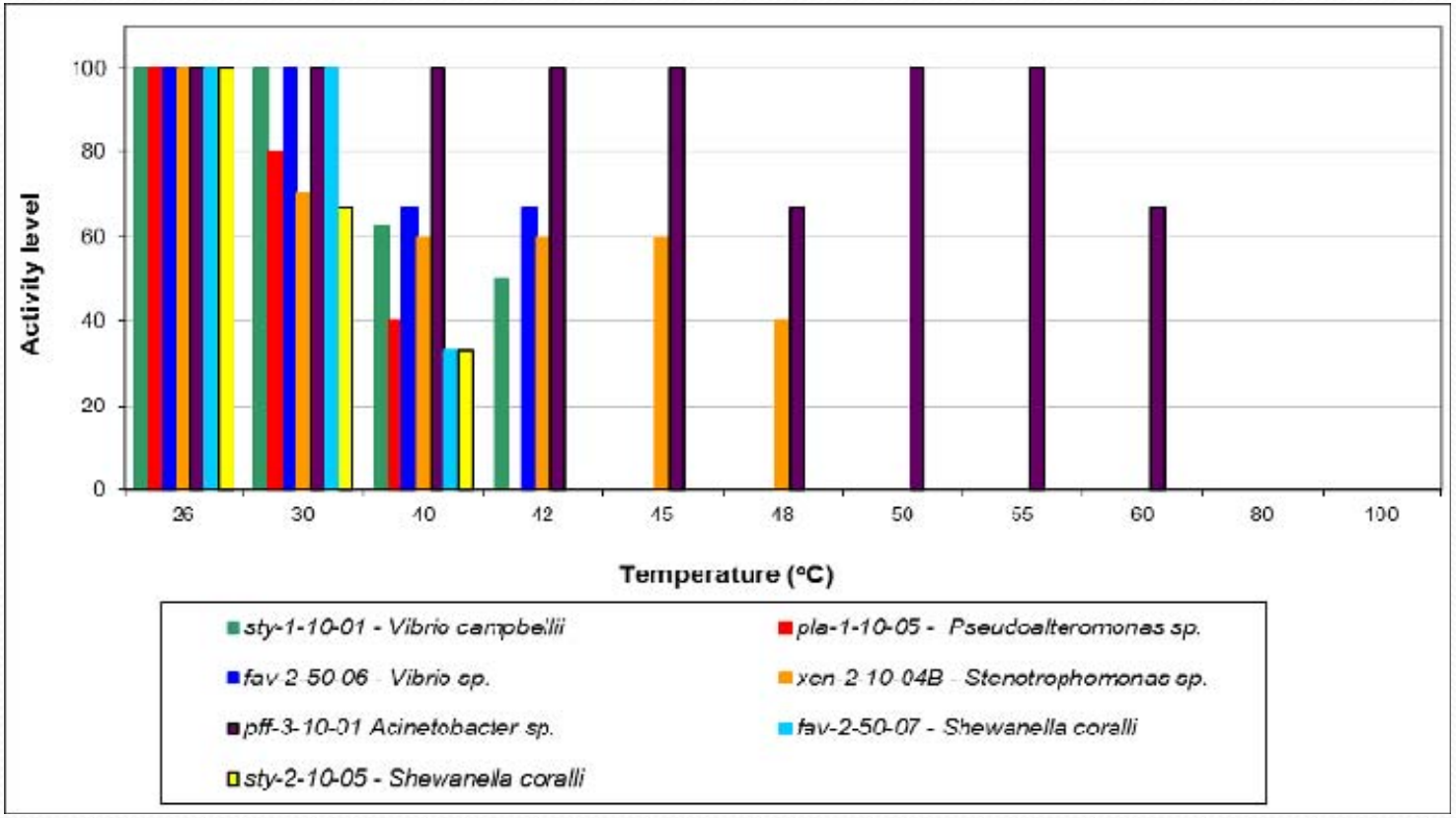
- n = fraction of active bacterial isolates



Shnit-Orland & Kushmaro, 2009 *FEMS Microbiol Ecol*

Inactivation temperature

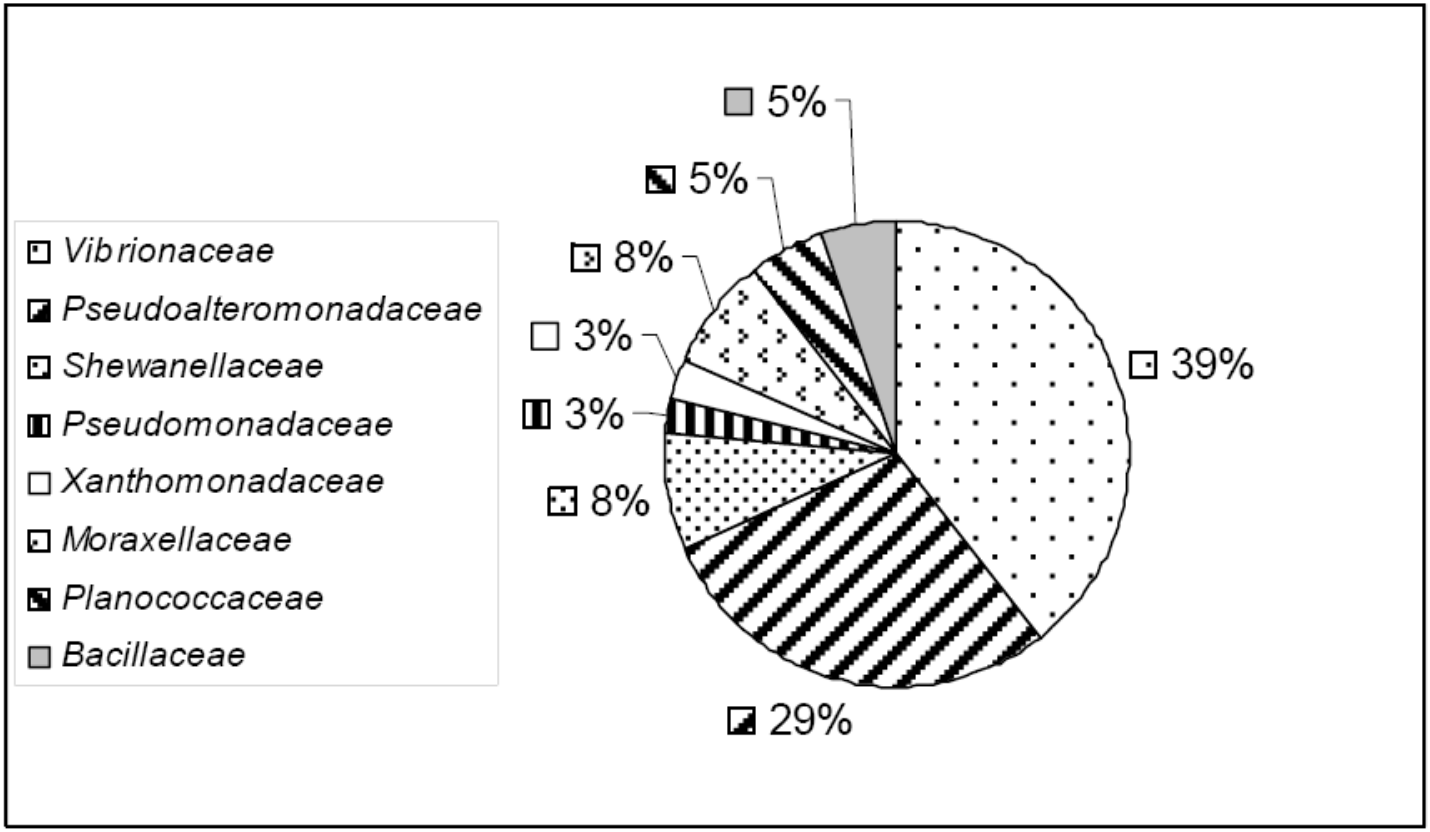
- 10 min X 26-100°C (screened using drop technique)
- Inactivation occurred before 45°C (in most cases)



- Protein component may be involved ?

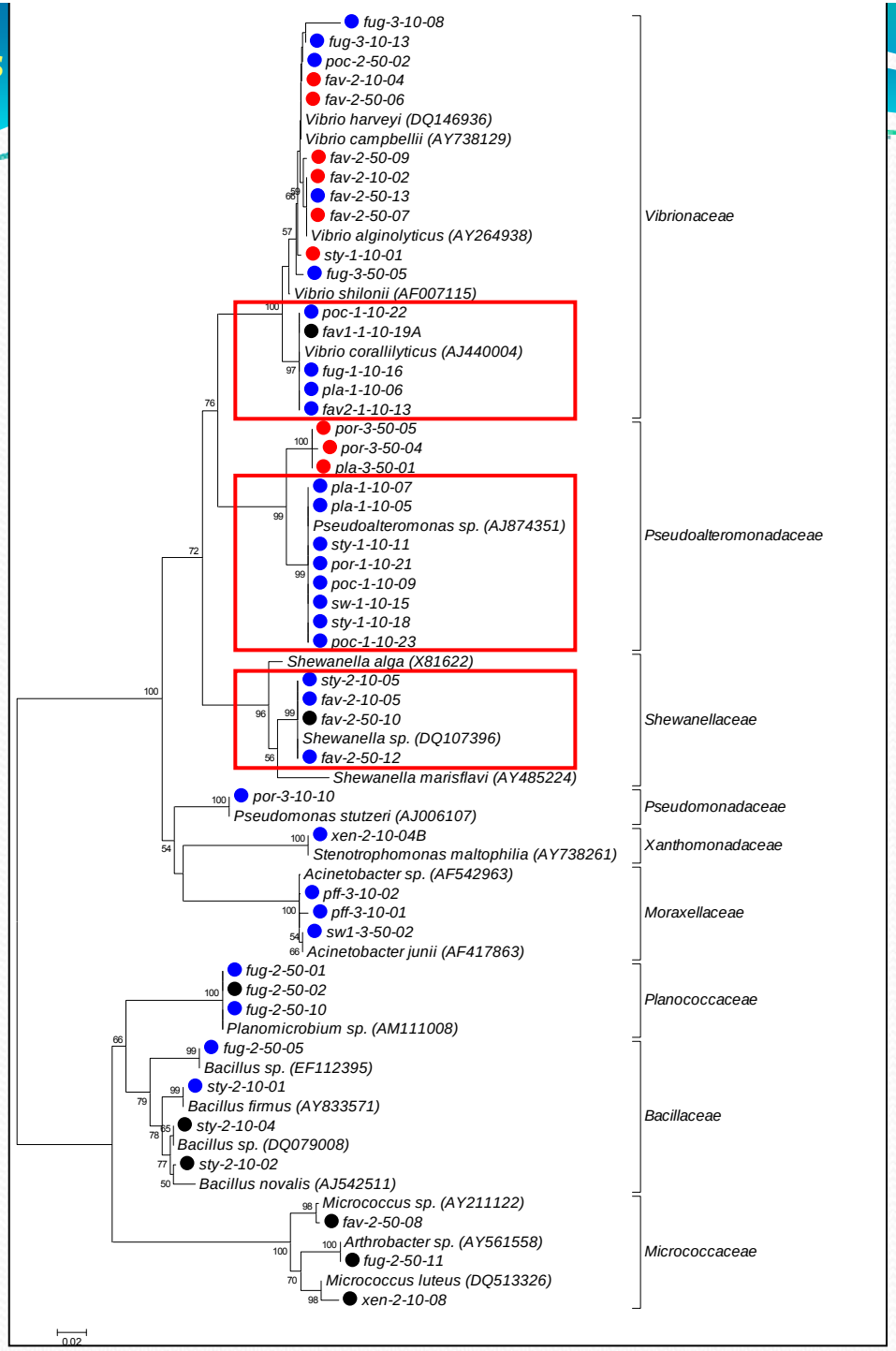
Antibacterial activity phylogenetic comparison

Percent of active isolates from total active isolates sequenced (16S rRNA gene), distributed according to family taxa (<93%)



Shnit-Orland & Kushmaro, 2008 *Proc 11th Int Coral Reef Symp*

Results



Phylogenetic analysis

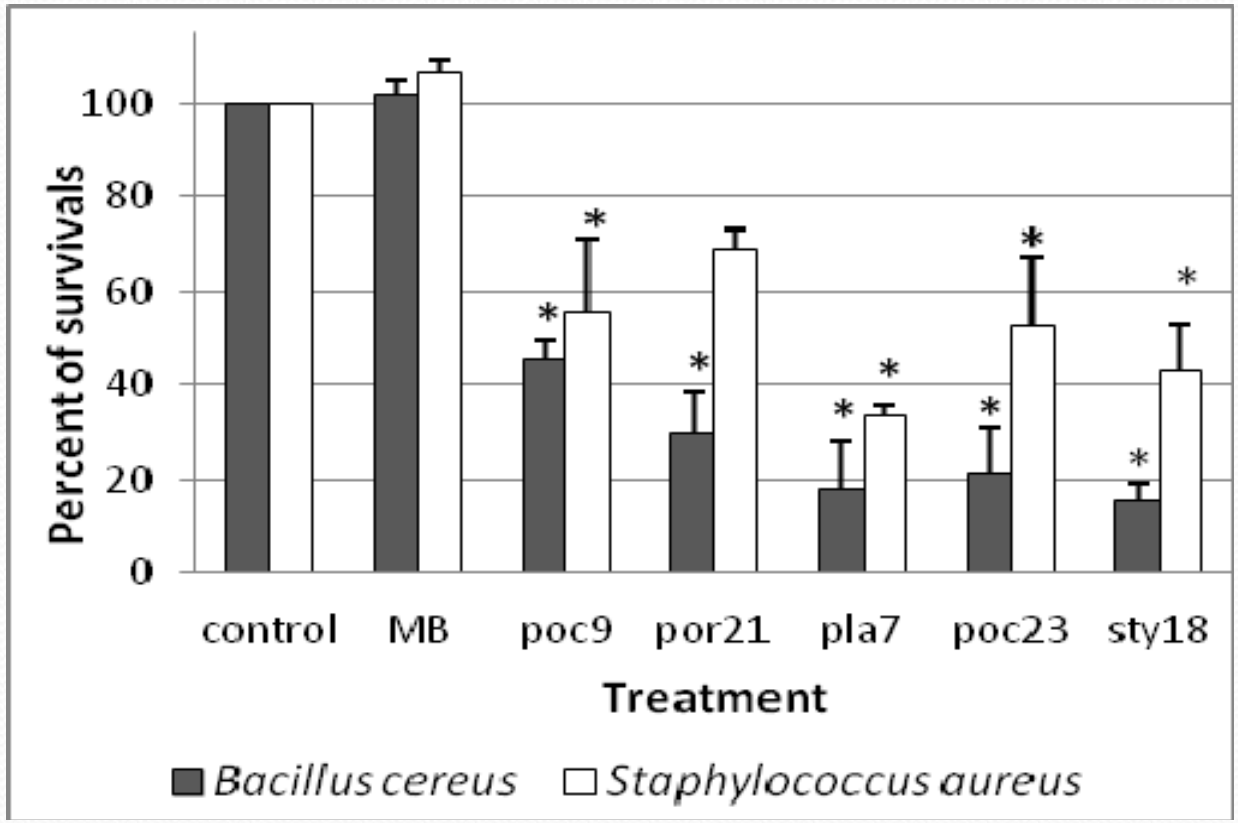
16S rRNA gene

- 🔴 - Activity against Gram (-) & (+) strains
- 🔵 - Activity against Gram (+) strains
- ⚫ - Non active isolates

Not a true representation
- only cultivable bacteria

Antibacterial activity of *Pseudoalteromonas* sp.

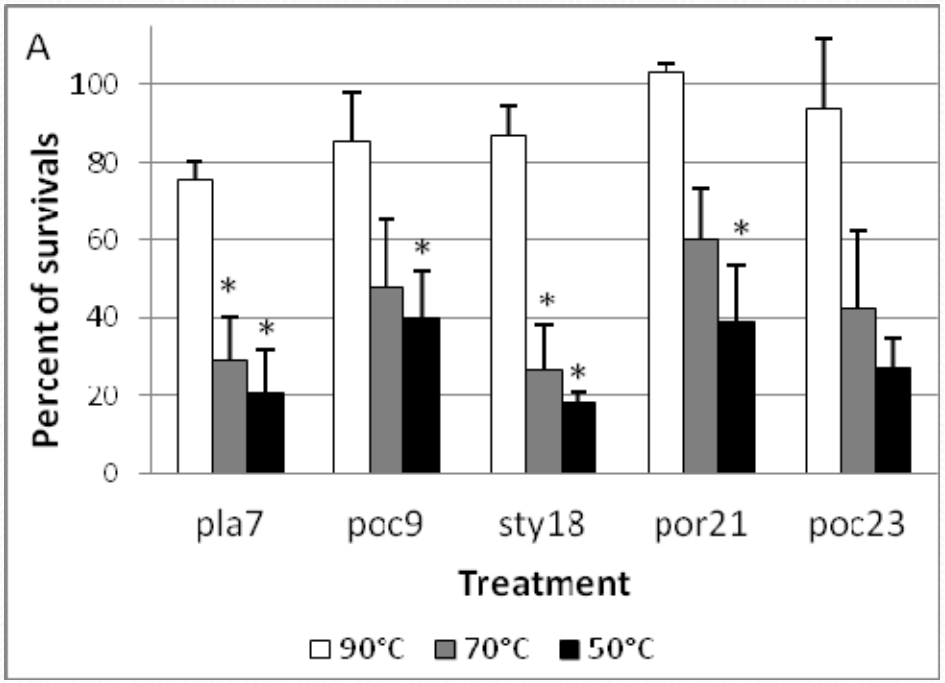
Effect of *Pseudoalteromonas* spp. cell-free supernatant (CFS) on growth of *Bacillus cereus* and *Staphylococcus aureus* ($P \leq 0.05$)

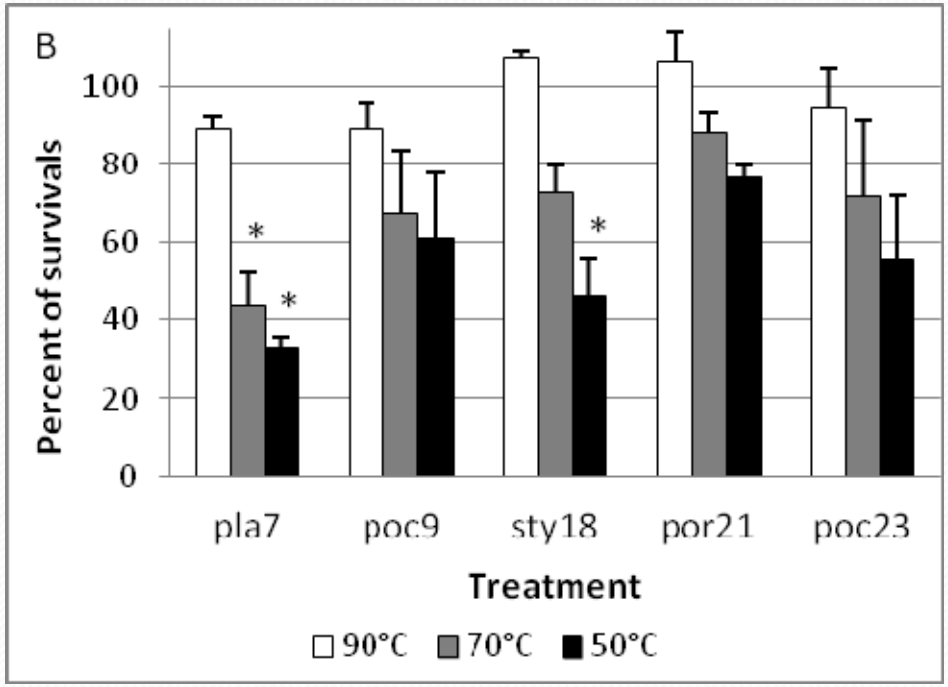


Shnit-Orland et al., 2012 *Microbial Ecology, in press*

Antibacterial activity of *Pseudoalteromonas* sp.

Effect of heat-treated *Pseudoalteromonas* spp. cell-free supernatant (CFS) on growth of a) *Bacillus cereus* and b) *Staphylococcus aureus* ($P \leq 0.05$)

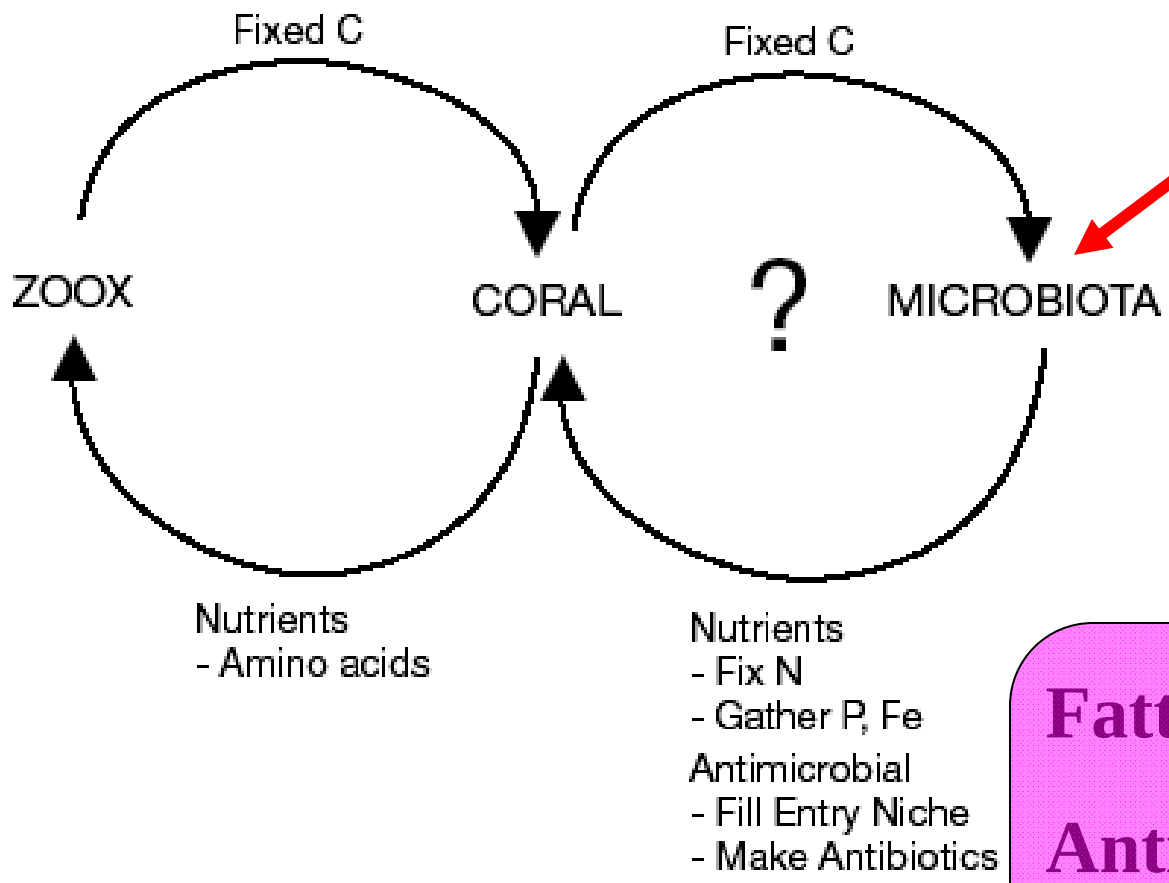




Shnit-Orland et al., 2012 *Microbial Ecology, in press*

Summary

- Cultivable coral mucus bacteria demonstrated antibacterial activity
- Stony coral isolates found to be more active than soft coral isolates
- Antibacterial activity had dual mechanism - diffusible and cell-associated
- Activity was sensitive to temperature
- Some clusters associated with different coral sp
- Marine *Pseudoalteromonas* sp. produced extracellular substances inhibiting growth of indicator strains
- **Coral mucus-associated bacteria may play a role in coral holobiont's defense strategy**



Mucus community

Fatty acids (PUFA) ★

Antimicrobial ★

N cycling ★

Rohwer et al.: Coral-associated bacteria

Mar Ecol Prog Ser 243: 1–10, 2002

The hunt continues

QuickTime™ and a
TIFF (Uncompressed) decompressor
are needed to see this picture.

Thanks….

- **M. Shnit-Orland**
- M. Harel
- N. Kooperman
- D. Zeevi Ben-Yosef
- D. Rasoulouniriana
- **N. Siboni**
- L. Arotsker
- E. Ben-Dov
- E. Eltzov
- O. Barnea
- Valentina Pavlova
- Yulia Goldovsky
- Alex Sivan
- K. Goldberg

- E. Kramarsky-Winter (TAU)
- A. Sivan (BGU)
- I. Brickner (TAU)
- Y. Loya (TAU)
- E. Rosenberg (TAU)
- Z. Barak (BGU)
- Z. Wiesman (BGU)
- R. Marks (BGU)
- O. Hoegh-Guldberg (AIMS)
- Hanny Faibish
- Jacob Gopas

- ISF and NIBN for financial support