

Aquaculture of the giant opihi *Cellana talcosa*
(ko`ele). Development of an artificial diet.

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Introduction and Background

“Opihi overharvesting means slim pickings”

Honolulu Advertiser, Wed, June 1, 2005

- Scientists fear that the largest and most prized species of the hardy 'opihi a uniquely Hawaiian delicacy may be essentially extinct on O'ahu, and the population of other limpets statewide is also on the decline.
- “Pupu” in Hawaiian means “snail” and in modern times it is used to mean hors d'oeuvres. Opihi were the most favored pupu traditionally.



Opihi

- High value potential aquacultured product in Hawaii, \$150/gallon with shell on. A century ago, 'opihi pickers were selling 140,000 pounds of the limpets annually. In recent years the number has been less than 10 percent of that, around 13,000 pounds.
- They dubbed 'opihi “the fish of death” because so many people were swept away while prying it off the rocks.

www.nature.org



Three main species of opihi in Hawaii

- *Cellana sandwicensis* - opihi alinalina – yellow foot – most common – preferred
- *Cellana exarata* - opihi makaiauli – black foot – *not preferred*
- *Cellana talcosa* - opihi ko`ele – giant opihi – grows fast – lives in calm, deep water – we targeted this



Other views

Cellana exarata
(opihi makaiauli)



Cellana sandwicensis
(opihi alinalina)

Cellana talcosa
(opihi ko`ele)

Outline of this talk

- To talk story about optimization of capture and holding strategies. Problem: 75% mortality in early days of holding and transferring. Starvation after a few days.
- To talk story about natural feeds and holding on biofilm. Benthic diatoms
- To describe early feed preference studies. Fish/soy and biofilm
- To talk story about a feed that may be capable of supporting long term growth and survival. We now have an artificial feed for the opihi. Diets containing commercial porphyra



Survival after capture, a new problem

(we attribute mortality to damage during capture)

| Trial | individuals (started) | Mortalities after 4 days | % survival |
|-------------------|-----------------------|--------------------------|------------|
| First collection | 38 | 21 | 48 |
| Second collection | 45 | 12 | 73 |
| Third collection | 29 | 5 | 83 |
| Fourth collection | 12 | 2 | 83 |



- We are getting better at collecting by being careful

Survival during research

(Problematic when removing from one tank wall to another tank)

| | Individuals start | individuals after moving | % survival |
|-----------------------|-------------------|--------------------------|------------|
| 1 st group | 17 | 9 | 53 |
| 2 nd group | 33 | 0 | 100 |
| 3 rd group | 24 | 0 | 100 |
| 4 th group | 10 | 0 | 100 |

- Problem scraping off tanks walls
- Put plastic liner and easy to remove



Biofilm. What is it?

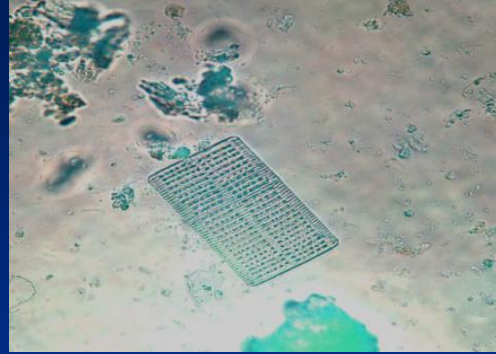
- Stomach content of opihi. Unidentifiable particles as well as *Bacillaria*, *Fragilaria*, *Melosira*, *Navicula*. *Rhabdonema*. What they normally eat.
- Biofilm on tanks. From sand filtered seawater or salt spray. In the sun. Looks like a mat of benthic diatoms (brown film on tank plastic). In the microscope these included *Nitzschia*, *Rhizosolenia*, *Melosira*, *Coscinodiscus*, and *Navicula*. Also bacteria, macroalgae, and unidentifiable particles.



Some stomach contents



Melosira



Rhabdonema



Navicula



Fragilaria



Nitzschia

Lab grown biofilm
seems to be similar
to natural food.

Feeding on biofilm

- Opihi eat biofilm
- They do not eat every day
- The eating rate of 0.47% may be a natural feeding rate (slow growth)
- When in doubt we can hold opihi on biofilm.



| | % drymatter/bodyweight/day | | | | | | | |
|-----|----------------------------|------|------|------|------|------|------|------|
| 1 | 0.7 | 0.59 | 1.3 | 0 | 0 | 1.2 | 0.25 | 0 |
| 2 | 0.93 | 0.60 | 0 | 0 | 0.84 | 0 | 0.30 | 0 |
| 3 | 0.12 | 0.42 | 0.42 | 0.74 | 0.78 | 0 | 0.87 | 0.75 |
| 4 | 0 | 1.12 | 0.55 | 0 | 0 | 0.56 | 0.50 | 0.37 |
| 5 | 0.12 | 0.56 | 0.84 | 1.06 | 0.52 | 1.11 | 0.33 | .037 |
| avg | 0.37 | 0.66 | 0.62 | 0.36 | 0.43 | 0.57 | 0.45 | 0.30 |
| | 0.47±0.13 | | | | | | | |

For aquaculture

- Need an artificial feed.
- Need to hold animals for long periods of time, to breed them, and rear the larvae.
- Phase 1. What do they like to eat?
- Phase 2. Design a nutritious feed, spawning, larval rearing etc.



Feeding preferences

dry feeds

| diet | %dry matter/bodyweight/day |
|-------------------------|----------------------------|
| 6% fishmeal+6%squidmeal | 0.17 |
| 12% fishmeal | 0.08 |
| 12% squidmeal | 0.09 |
| mussel homogenate | 0.02 |
| squid homogenate | 0.02 |

| Diet | %dry matter/bodyweight/day |
|-------------------------|----------------------------|
| 6% fishmeal+6%squidmeal | 0.02 |
| 12% fishmeal | 0.03 |
| 12% squidmeal | 0.03 |

1. Have to feed animals before they die.
2. First test shows preference for marine meals
3. Second test cannot be compared to first. Shows no preference among marine meals. N=3

Feeding preferences

(gelatin feed: easier to make for us)

| % dry matter/bodyweight/day | | | |
|-----------------------------|-----------|------------------|------------------|
| Fish/soy | Soy/corn | Fish/soy/betaine | Soy/corn/betaine |
| 0.16±0.06 | 0.10±0.03 | 0.03±0.05 | 0.01±0.01 |

N=3 (three animals/diet)

T=5 (5 days/diet)

Conclusions. Within limits of experimental uncertainty

1. The fish soy diet seems to be eaten in largest amount.
2. The fish soy diet eaten often, 53% of the nights.
3. Betaine not an attractant

Feeding preferences

(agar feeds, also easy to make)

| % dry matter/bodyweight/day | | | |
|-----------------------------|------------------|---------------|---------------|
| Fish/soy | Fish/soy/biofilm | Fish/soy/GABA | Fish/soy/DMPT |
| 0.07±0.12 | 0.17±0.19 | 0.08±0.11 | 0.04±0.07 |

N=3 in most cases

Time = 5 days

Conclusions

1. The diet with biofilm was eaten in largest amount and often (43% of nights)
1. Gamma amino butyric acid and dimethyl propiothetin are not attractants. Spirulina too (data not shown).

Focus in

- Fish/soy and biofilm seem preferred.
- Focus in on this. Are we right or wrong?



Focusing in

Is high fishmeal diet without biofilm also liked? Not especially.

| | % dry matter/bodyweight/day |
|--------------------------|-----------------------------|
| High fishmeal no biofilm | 0.012 \pm 0.01 |

N=5; Time=4 days

Is high fishmeal diet and biofilm liked? Yes. Or
biofilm with more fishmeal liked more? No.

| | % dry matter/bodyweight/day |
|---------------------------|-----------------------------|
| Fishmeal and biofilm | 0.08 \pm 0.03 |
| High fishmeal and biofilm | 0.07 \pm 0.01 |

N=6; Time = 6 days

Conclusions

1. Removal of biofilm decreases feeding
2. Adding additional fishmeal does not help.
3. Hence, biofilm is the key.



Biofilm is the key to feed acceptance

- But biofilm is a randomly recruited assemblage of wild benthic diatoms. It might be unreliable.
- Biofilm would have to be grown.
- Vernon Sato suggested a trip to Don Quixote to find a commercial substitute.



Sustained feeding on feed with *Porphyra* %DM/BW/day

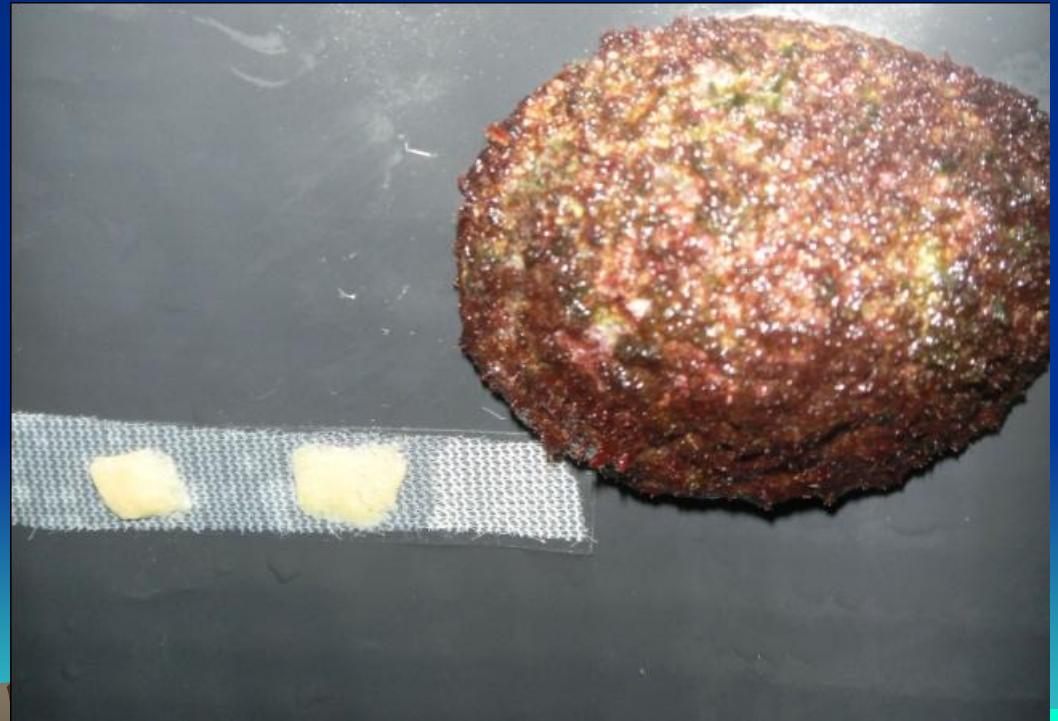
1. One giant opihi lived 15 days and ate 0.074%/day.
2. Another giant opihi lived 45 days and ate 0.11%/day.
3. A blackfoot lived 32 days, ate 0.076%
4. We believe we are on the cusp of keeping opihi forever.
5. Diet is fish meal, soy meal, Nori, algenate, vitamins, cholesterol, agar



| | | |
|-------|-------|-------|
| 0.16 | 0 | |
| 0 | 0.12 | |
| 0.1 | 0 | 0 |
| 0 | 0.22 | 0 |
| 0.18 | 0 | 0.2 |
| 0 | 0.2 | 0.18 |
| 0 | 0 | 0 |
| 0.12 | 0.1 | 0 |
| 0.1 | 0.15 | 0 |
| 0.21 | 0 | 0.17 |
| 0 | 0.16 | 0 |
| 0 | 0.32 | 0.12 |
| 0.16 | 0 | 0 |
| 0 | 0.12 | 0.11 |
| 0.074 | 0 | 0.15 |
| | 0.21 | 0 |
| | 0.13 | 0 |
| | 0.12 | 0.18 |
| | 0 | 0 |
| | 0.11 | 0.13 |
| | 0 | 0.12 |
| | 0.18 | 0 |
| | 0.08 | 0.23 |
| | 0 | 0 |
| | 0.23 | 0.18 |
| | 0.08 | 0 |
| | 0.15 | 0.2 |
| | 0.17 | 0.08 |
| | 0.11 | 0 |
| | 0 | 0.2 |
| | 0.3 | 0.12 |
| | 0.12 | 0 |
| | 0.22 | 0.12 |
| | 0.23 | 0.1 |
| | 0 | 0 |
| | 0.1 | died |
| | 0.22 | |
| 0.074 | 0.113 | 0.076 |

Choice experiments

- Preference to feed on food attached to the vertical side of the aquarium
- Feed pressed to net attached by double sided tape to plastic in aquarium.



Summary

- We believe that we have figured out how to capture and hold opihi. Plastic lined tanks are the key.
- We believe that we have determined that a key to feed palatability is biofilm, an aggregate of benthic diatoms and we can replace grown biofilm with a commercial *Porphyra* preparation.
- Instead of struggling to keep opihi alive with our artificial diets we can hold opihi and study them for an extended time. This is a big deal for us.



Future work

- We need more animals to do nutritional experiments.
- We must get ripe opihi to spawn. Either wild caught or preferably aquacultured. Several spawning methods will be applied to trigger spawning of specimens such as **vigorous aeration, thermal shock, hydrogen peroxide...**
- **Larval rearing, settlement and metamorphosis.** Substratum preference, diatoms species as biofilm, chemical cues GABA...
- Growout and closing the life cycle.

- **The future of opihi is hopeful.**



Thank you very much for your attention!!!

MAHALO!

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