

ALASKA CRAB STOCK

Enhancement and Rehabilitation

Workshop Proceedings

March 14-16, 2006

Kodiak, Alaska

Bradley G. Stevens, Editor

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Book design is by Jen Gunderson, cover design is by Brooks Pleninger, and copy-editing is by Sue Keller. **Front cover** photo by Art Sutch, adult red king crab. **Back cover** photos by Bradley G. Stevens. **Left:** Glaucothoe larva, transitional between zoea and first crab stage. Its job is to select habitat that will protect it from predation. **Middle:** 388-day-old blue king crab embryo, showing the large black pigmented eye. This embryo would hatch in about one month. **Right:** First stage zoea larva of blue king crab.

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Acknowledgments

Alaska's crab stocks were once the envy of the world. And while Alaska fishermen today still enjoy bountiful harvests of several crab species, stocks of some species—notably Kodiak red king crab and Pribilof blue king crab—have markedly declined. These declines have spurred rebuilding efforts through improved state and federal fisheries management, and led to grassroots calls to employ mass culture of crab stocks in hatcheries as a rebuilding tool. The concept of using hatcheries was first suggested in the early 1990s. The January 1992 meeting, International King Crab Rehabilitation and Enhancement Symposium, was convened by Jeff Stephan and Lonnie White. More recently, fishermen again raised the prospect of crab enhancement, noting that red king crab stocks around Kodiak Island and blue king crab stocks near the Pribilof Islands had not recovered. Additionally, enhancement techniques have advanced considerably in the intervening years—now several countries have successful crab culture programs.

Which brings us to March 14-16, 2006, when fishermen, scientists, fishery managers, coastal community leaders, and others gathered in Kodiak, Alaska, to listen to a panel of experts from around the world detail the state of the science regarding crab culture for wild stock enhancement. This proceedings is the result of that meeting.

Convening this workshop was largely the undertaking of key individuals and organizations, who are gratefully acknowledged here.

Alaska King Crab Stock Enhancement and Rehabilitation Workshop 2006

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- United Fishermen's Marketing Association

Agenda

Tuesday, March 14, 2006

8:30 **Welcome by Kodiak Representatives**

8:45 **Introduction**

*Brad Stevens, NOAA Fisheries, Kodiak Fishery Research Center,
Kodiak, Alaska*

9:00 **Technical Workshop—Invited Speakers**

Culture of king crab at the Kodiak Fishery Research Center
*Sara Persselin, NOAA Fisheries, Kodiak Fishery Research Center,
Kodiak, Alaska*

Research on king crab cultivation in Russia
Nikolina Kovatcheva, VNIRO, Moscow, Russia

Research on cultivation of European lobsters,
Homarus gammarus, in Norway
Gro van der Meeren, Institute of Marine Research, Bergen, Norway

Cultivation of golden king crabs
*Tom Shirley, University of Alaska Fairbanks, Juneau Center SFOS,
Juneau, Alaska*

Basis for stock enhancement of *Lithodes santolla* in Argentina
Gustavo Lovrich, CADIC, Ushuaia, Argentina

Cultivation of *Lithodes santolla* in Chile
Kurt Paschke, Universidad Austral de Chile, Puerto Montt, Chile

Cultivation of Chesapeake Bay blue crab, *Callinectes sapidus*
*Odi Zmora, Center of Marine Biotechnology, University of Maryland
(Presented by Tuck Hines, Smithsonian Environmental Research Center,
Edgewater, Maryland)*

Cultivation of lobsters
*Michel Comeau, Department of Fisheries and Oceans,
New Brunswick, Canada*

2:45 Roundtable Discussion

Discussion among researchers. Some questions are:
 What worked? What didn't work? What are the research gaps? Speculate on how to proceed in Alaska.

Wednesday, March 15**8:30 Welcome and announcements**

Brian Allee, Alaska Sea Grant, University of Alaska Fairbanks, Fairbanks, Alaska

8:40 Case Studies

Case study 1: Stock enhancement of Chesapeake Bay blue crab
Tuck Hines, Smithsonian Environmental Research Center

Case Study 2: Enhancement of Maine lobsters
Ted Ames, MacArthur Fellow and fisherman, Stonington, Maine

Enhancement and Rehabilitation Concepts and Potential (Panels)**10:00 Technical Considerations**

Discussion and questions from the audience

Moderator

Ray RaLonde, Alaska Sea Grant Marine Advisory Program, Anchorage, Alaska

Panel members

Brad Stevens, NOAA Fisheries
Tuck Hines, Smithsonian Environmental Research Center
Other researchers

Panel questions

- Is crab enhancement feasible?
- Should it be attempted?
- What are the limitations?
- What species should be considered? King, tanner, others?
- What techniques should be considered? For example, brood stock transfer; larval settlement traps; habitat enhancement; cultivation and release (stock enhancement).
- How could enhancement be implemented in Alaska (from a technical viewpoint)?
- Who would regulate and who would harvest?

1:15 Potential Models for Crab Enhancement and Rehabilitation

Discussion and questions from the audience

Moderator

Brian Allee, Alaska Sea Grant

Panel members

Ted Ames, MacArthur Fellow and fisherman

Ray RaLonde, Alaska Sea Grant Marine Advisory Program

*Pete Esquiro, Northern Southeast Regional Aquaculture Association,
Sitka, Alaska*

Panel questions

- What are the benefits and costs associated with lobster enhancement activities in Maine?
- What general economic opportunities, costs, and benefits are anticipated or desired for crab enhancement and rehabilitation activities that may take place in Alaska?
- What funding mechanisms may be reasonable, possible or available for the purpose of funding crab enhancement and rehabilitation activities (e.g., to fund preliminary research, feasibility testing, pilot project, etc.)?
- What structural, operational, and administrative mechanisms may be reasonable for the purpose of crab enhancement and rehabilitation activities in Alaska? Are there elements of the current salmon hatchery structural model that are applicable to crabs (i.e., Alaska legislation that provides for regional aquaculture associations, private nonprofit entities, etc.)?
- What are the infrastructure needs for the early phases of evaluating and testing the feasibility of crab enhancement and rehabilitation activities (facility location, size, and space; equipment and materials needs)?

3:00 Industry and Community Perspectives

Discussion and questions from the audience

Moderator

Heather McCarty, McCarty and Associates, Juneau, Alaska

Panel members

Arni Thomson, Alaska Crab Coalition, Seattle, Washington

Jeff Stephan, United Fishermen's Marketing Association, Kodiak, Alaska

Dave Woodruff, Alaska Fresh Seafoods, Kodiak, Alaska

*Gale Vick, Gulf of Alaska Coastal Communities Coalition,
Anchorage, Alaska*

Mel Morris, Alaska Board of Fisheries, Kodiak, Alaska

Thursday, March 16**10:30 Kodiak ComFish Seminar: Alaska Crab Enhancement and Rehabilitation—This Isn't Crab Farming!**

Keynote Presentation: Maine Lobster Enhancement
Ted Ames, MacArthur Fellow and fisherman

Summary Presentations

Crab enhancement and rehabilitation research
Brad Stevens, NOAA Fisheries

Technical considerations
Ray RaLonde, Alaska Sea Grant Marine Advisory Program

Potential models for crab enhancement and rehabilitation
Brian Allee, Alaska Sea Grant

Industry and community perspectives
Heather McCarty, McCarty and Associates

Public Forum: Ask the Experts

Moderator
Jeff Stephan, United Fishermen's Marketing Association

Alaska Crab Stock Enhancement and Rehabilitation: An Introduction

Bradley G. Stevens

NOAA Fisheries, Alaska Fisheries Science Center,
Kodiak Fisheries Research Center, Kodiak, Alaska

King crab enhancement is not “crab farming.” I have to say that up front because the issues are commonly confused. The goal of this symposium is not to develop “farms” for crabs, but rather to discuss the possibility of enhancing or restoring natural populations of crabs in the ocean. There are numerous potential methods for achieving that result. Perhaps the most widely discussed technique involves cultivation and release of small crabs. But none of these methods have been critically evaluated, much less undergone intense scientific or public scrutiny. With this volume, we hope to enlighten the reader with discussions of the feasibility and desirability (or not) of crab stock enhancement in Alaska.

Background

Crab populations in Alaska have declined steadily in recent decades. Populations of the red king crab *Paralithodes camtschaticus* (Tilesius 1815) in Alaska have fluctuated greatly over the last three decades (Stevens et al. 2001). Four other commercial fisheries in the Bering Sea, including snow crab (*Chionoecetes opilio*), Tanner crab (*C. bairdi*), and blue king crab (*P. platypus*) at St. Matthew Island and the Pribilof Islands, are considered to be overfished. The fishery for Bering Sea Tanner crab has been closed since 1996. The Pribilof Islands blue king crab fishery began to decline in the 1980s, was closed from 1988 to 1994, and reopened in 1995; both fisheries were closed in 1998 and have remained closed since then (NPFMC 2002). Simultaneously, the population of red king crab in the Pribilof Islands has increased since 1991, but no directed fishing for red king crab has occurred there since 1998 in order to prevent bycatch of blue king crab.

These fluctuations are probably the result of variable recruitment of juveniles, but there is great uncertainty about the ultimate cause of recruitment variability (Blau 1986). Many hypotheses have been proposed, including egg predation (Kuris et al. 1991), disease, overfishing (Orensanz et al. 1998), bycatch (Dew and McConnaughey 2005), and climatic changes (Zheng and Kruse 2000). Changes in spatial distribution associated with climate variability may be a primary cause of population fluctuations (Loher and Armstrong 2005), but the link between environmental change and population abundance is not yet understood.

Whatever the cause of the population declines, these trends are expected to continue in the near future. Crab stocks around the world have suffered similar fates and many are overfished and non-viable. Traditional management techniques have not helped stocks to recover—after 25 years without fishing, Kodiak king crab populations are still depressed. Declining catches and income have caused fishermen and communities to begin seeking alternative solutions to traditional management schemes, such as the possibility of enhancing natural stocks. To counteract declining abundance, scientists in many parts of the world have begun research into cultivation and artificial enhancement of crab populations. Because of its high value, cultivation of red king crab was begun by Japanese scientists in the 1960s. In the last decade, advancements in the cultivation of several species of king crab have been made in Japan, Norway, Argentina, Chile, and Russia, as well as in Alaska. A commercial stock enhancement facility currently exists at Akkeshi, Hokkaido, and more are being developed in Russia and Norway. Similar techniques and approaches are being used worldwide, yet scientists conducting this work rarely have the opportunity to communicate with each other because of the distance between research centers.

Research on king crab cultivation has been conducted at the Kodiak Fisheries Research Center since 2000, primarily as a tool for ecological research. Initial success was poor due to the steep learning curve, but in 2004 we concluded a highly successful project with blue king crab (Persselin 2006, Stevens et al. submitted for publication). Our efforts have been conducted on a small scale, raising a few thousand crabs for use in experimental research, as opposed to the millions that will be needed for enhancement. However, the same techniques we use in the laboratory are applicable to commercial stock enhancement, if scaled up to a larger level.

In January 1992, Kodiak College of the University of Alaska hosted a small workshop titled International Crab Rehabilitation and Enhancement Symposium. The symposium was held in response to a proposal from the Alaska Department of Fish and Game (ADFG) to cultivate larval king crabs and release them into the ocean. At that time, the idea of enhancing crab stocks was new, and nobody in the United States had ever cultivated crabs of

any kind, especially king crabs. At the conclusion of the meeting most of the participants agreed that stock enhancement of king crabs was not technically feasible at that time.

But things have changed. King crabs are now being routinely cultivated in many countries. By 2004, we recognized that the time had come to bring together those scientists who are doing this work, to reevaluate the state of crab cultivation technology and learn from each other. Rather than re-inventing the wheel in isolated laboratories, we needed to focus on technologies that work, and find ways to improve our efforts. At the same time, we needed to examine larger questions. Cultivation and release of small crabs is one technology that has merit. But is it the best method for enhancing crab stocks? Would it create problems with disease, genetics, or waste pollution? Are there cheaper, quicker, or more efficient methods? What about enhancing natural habitat? Who would benefit, and who should pay for these efforts?

In January 2005, an ad hoc group of people representing industry, coastal communities, and research agencies came together to promote an international conference on Alaska crab enhancement. The goal of the workshop was to bring together scientists who are doing cutting-edge research on cultivation and stock enhancement of king crabs and lobsters. Why lobsters? They share many similarities with king crabs, including life history, habitat requirements, size, age at maturity, and cannibalistic tendencies, and they are one of the few large cold-water crustaceans for which enhancement studies are starting to bear fruit. The workshop focused on three major themes: technical issues, alternative approaches, and management and social issues. An intensive one-day workshop provided scientists with opportunities to exchange information on technology, methodology, and goals of various crab cultivation and enhancement programs. Additional workshops addressed issues of concern to the greater public, including appropriate technology and species, environmental effects of enhancement, and management issues. We did not expect to answer all of these questions at one time. However, we did hope that a discussion focused around king crab cultivation will help guide research and development for the near future.

This volume presents summaries of the presentations made at the workshop. We hope that it will serve as a guiding light for future discussion, research, and policy development regarding enhancement of Alaska crab stocks, and other cold-water crustaceans in general.

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Cultivation of King Crab Larvae at the Kodiak Fisheries Research Center, Kodiak, Alaska

Sara Persselin

NOAA Fisheries, Alaska Fisheries Science Center,
Kodiak Fisheries Research Center, Kodiak, Alaska

Introduction

Red king crabs (*Paralithodes camtschaticus*) and blue king crabs (*P. platypus*) have historically supported an extensive commercial fishery in Alaska waters. However, red king crab stocks declined precipitously in the 1980s followed by blue king crab stocks in the 1990s, and both stocks have since remained depressed. To investigate this decline in the crab population, Brad Stevens of the National Marine Fisheries Service (NMFS) began studies of king crab life history at the Kodiak Fisheries Research Center (KFRC) in 1998. Projects at the KFRC have addressed the needs of larvae in cultivation (food, density, temperature), settling behavior of glaucothoe, and habitat preferences of juveniles. In 2001 I began work to refine larval cultivation techniques to increase the number of juvenile crabs produced for study at the KFRC. The development of successful laboratory cultivation techniques has the additional benefit of potential application to large scale cultivation for stock enhancement.

Red and blue king crab larval development consists of a brief prezoal stage, four zoeal stages, and one glaucothoe stage (equivalent to megalops) before metamorphosis to the first juvenile crab stage (C₁) and settlement on the ocean bottom. The four zoeal stages last 8-10 days depending on water temperature; the colder the temperature the longer the development. The glaucothoe stage is a nonfeeding stage and lasts 3-8 weeks.

Red king crab culture

Red king crab (RKC) larvae for all research projects at the KFRC are obtained from adult ovigerous female crabs collected from Womens Bay on Kodiak Island by NMFS divers. The crabs are brought into the lab and held communally in 2,500 L tanks on a flow-through seawater system. When the crabs

begin releasing larvae they are isolated into 120 L tubs. Newly hatched larvae are collected from these tubs and transferred to treatment containers.

Initially, the newly hatched larvae were collected and placed in 22 L square plastic tanks at a density of 40 zoeae per L. Larvae were fed *Artemia salina* nauplii and 75-90% of the culture water was changed at 3-5 day intervals (Stevens 2003). Water circulation was provided by an airstone at the bottom of each tank. This culture method produced glaucothoe, but at sub-optimal levels. Water changes were difficult and water circulation allowed uneaten food, molts, and live and dead larvae to collect on the bottom of the tanks.

I felt that the two main issues to be addressed in improving larval culture were (1) improvement of culture containers and (2) improvement of food quality. To improve the culture system, I designed a set-up that would allow for easy, daily water changes. This consisted of a 10 L plastic aquarium filled with 9 L of filtered seawater and containing three PVC cylinders 100 mm in diameter, and 150 mm long with a disc of 670 μm nylon mesh attached to the bottom with silicon adhesive (Fig. 1). Each cylinder was set mesh-side down



Figure 1. Red king crab larval culture system. Each treatment consisted of a 10 L plastic aquarium filled with 9 L of filtered seawater, containing three PVC cylinders 100 mm in diameter and 150 mm long with a disc of 670 μm nylon mesh attached to the bottom with silicon adhesive.

inside the tank and zoeae and *Artemia* nauplii were placed in each cylinder. Each day the cylinders and their larvae were lifted from the tank and placed in an identical tank of acclimated seawater. The mesh size allows for uneaten *Artemia* to be flushed out the bottom of the cylinder yet retain the larger zoeae. It is important that the *Artemia* are removed on a daily basis as they lose their nutritional value after 24 hours.

Artemia nauplii are a commonly used food in larval culture, although they have little, if any, of the highly unsaturated fatty acid (HUFA) docosahexaenoic acid (DHA, 22:6n-3) that is considered crucial to the survival of crustacean larvae (Navarro et al. 1991, 1993). Larvae do not synthesize most HUFA de novo and must ingest it from a food source. Enrichments can be fed to *Artemia* nauplii to enhance their nutritional quality prior to feeding them to crab larvae. Enrichments are generally selected based on the nutritional needs of the larvae being cultured and include microalgae, microalga pastes, yeast, emulsified products, and micro-encapsulated diets. I chose to compare a diet of newly hatched *Artemia* nauplii to nauplii enriched with one of the following: the microalgae *Rhodomonas salina*, *Isochrysis* sp., *Thalassiosira nordenskiöldii*, or *Thalassiosira aestivalis*, or the commercial enrichment products *Isochrysis* Instant Algae® paste or Algamac 3050. These enrichments were chosen because they are known to be high in DHA (*Isochrysis* sp., *Isochrysis* paste and Algamac 3050), to have been used successfully in aquaculture (*R. salina*, *Isochrysis* sp. *Isochrysis* paste, and Algamac 3050), or known to be ingested by crab larvae (*T. aestivalis* and *T. nordenskiöldii*). Seven tanks were set up with three cylinders each and 20 zoeae per cylinder for a total of 60 zoeae per treatment. All treatments were placed in a temperature-controlled room at 8°C.

The treatment that resulted in the highest survival to the nonfeeding glaucothoe stage was *T. nordenskiöldii* (73%); however, there was no significant difference between the *T. nordenskiöldii*, *R. salina*, *Isochrysis* sp., and *Isochrysis* paste treatments. Lowest survival occurred in the newly hatched unenriched nauplii treatment (42%). Based on these results, a diet of enriched *Artemia* nauplii is superior to unenriched newly hatched nauplii for increasing survival of red king crab larvae cultured in the laboratory.

Blue king crab culture

In 2004, Brad Stevens, Julie Matweyou, and I undertook a study to develop cultivation techniques for the larvae of blue king crab, *P. platypus* (Stevens et al., submitted for publication). Based on the diet results from the RKC larval study, we chose to compare unfed larvae (to determine lecithotrophy) (UNFED) to larvae fed on a diet of *Artemia* nauplii enriched on *Isochrysis* paste (ISO) or the diatom *T. nordenskiöldii* (THAL), or larvae fed newly hatched *Artemia* nauplii in combination with *T. nordenskiöldii* in the treatment water (A+THAL). I included the latter diet based on the suspicion that the zoeae might benefit

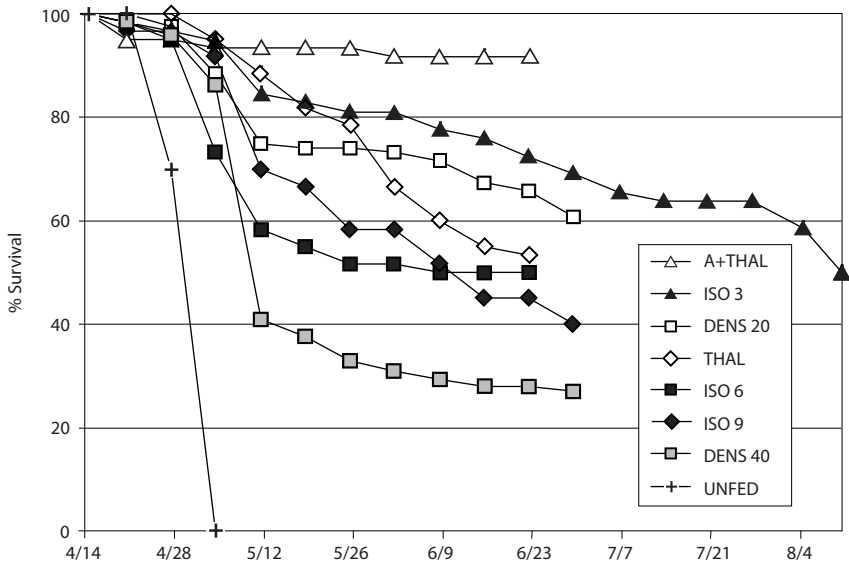


Figure 2. Percent survival of blue king crab (*P. platypus*) larvae at weekly intervals, from hatching to stage C1, under different culture conditions (from Stevens et al., submitted for publication). All points are means of 6 replicates. See text for description of treatments.

from consuming the diatoms directly. The zoeae might also benefit from the diatoms improving the culture water by taking up nitrogenous wastes, producing oxygen, or enhancing the water in some other aspect. All diets were tested at 6°C, and a density of 10 zoeae per liter, with six replicates per treatment.

In addition to diet, we tested different water temperatures and zoeal densities. We chose the ISO diet as our “base” diet and had treatments at 3°C (ISO 3) and 9°C (ISO 9) and at densities of 20 (DENS 20) and 40 zoeae per L (DENS 40). For each treatment, zoeae were placed in PVC cylinders in individual 1 L glass beakers (rather than a plastic aquarium). Survival on the A+THAL diet (91.7%) was significantly higher than all others, whereas UNFED larvae died within two weeks (Fig. 2). Survival decreased slightly with increasing temperature, but not significantly. Density had no significant effect on survival, but final mean density (16 per L) was similar in the DENS 20 and DENS 40 treatments suggesting that a maximum carrying capacity for these conditions had been reached. We concluded from this research that blue king crab larvae can be cultivated with high survival using the proper diet and that larvae are not lecithotrophic.

The results of king crab larval culture studies at the KFRC indicate that proper diet is crucial to larval development and survival. The development of successful larval cultivation techniques at the KFRC has allowed us to raise zoeae and produce juveniles for studies on early life stages. The techniques developed at the KFRC could be modified for use in king crab stock enhancement and rehabilitation projects.

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Red King Crab (*Paralithodes camtschaticus*) Reproduction and Cultivation in Artificial Conditions in Russia

Nikolina P. Kovatcheva

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and Oceanography (VNIRO), Moscow, Russia*

Introduction

In recent years there has been a sharp decline in the stocks of red king crab in virtually all the conventional fishing areas in the Russian Far Eastern seas. As a consequence, the recommended volumes of the allowable catch for the recent five years have been reduced to nearly one tenth: from 30,000 tons in 1999 down to 3,583 tons in 2003. That is why we should employ every possible method to promote the introduction of more optimum techniques for the use of red king crab resources. These objectives can be reached using techniques of commercial culturing of crab. Since 2000, a closed recycling seawater system has been operating at VNIRO aquaria to experiment in obtaining and rearing red king crab larvae up to the viable juvenile stages. Optimum conditions of development absent in natural habitats, or tampered, are being maintained in the process, especially at the plankton larval stage.

Despite the increasing abundance of red king crabs in the Barents Sea, some problems have arisen connected with fullness of the appendages. Additional rearing of prerecruits and fishable males caught at sea up to marketable quality recently became important in Russia. Work in this direction is of interest for trade companies as well.

The main goals of our studies were as follows:

1. Investigation of the biology and behavior of crustaceans (red king crab, several crayfish species, giant freshwater prawn).
2. Elaboration of technology for artificial reproduction and rearing of crustaceans (red king crab, several crayfish species, giant freshwater prawn).

3. Designing recirculating and flow-through water systems for the facilities for artificial reproduction and rearing of red king crab.

The results of those studies were basic in developing tentative biological standards for culturing red king crab, and were used in designing experimental crab integrated facilities in Kamchatka, in the Far East, and the Barents Sea (Russian Federation Patent No. 2200386 and No. 2261594, 2005; Kovatcheva et al. 2005).

Culture systems and equipment

Recirculating culture system

In inland culture facilities or in the areas where environmental conditions are not optimal for the red king crab, dynamic recirculation culture systems may be used. This system is based upon continuous water circulation through mechanical and biological filters, thus providing continuous removal of solid and nitrogen wastes. The closed recirculation water system used in *Paralithodes camtschaticus* culture includes the following main components: holding and rearing tanks, mechanical and biological filters, water pumps, cooling equipment (chillers), protein skimmers, and ultraviolet disinfection unit (UV-sterilizer). Additionally, wastewater treatment units may be included.

A major bottleneck in red king crab culture is that all the processes mentioned above require very low temperatures of seawater (4–11°C). To date, the experience of exploiting recirculating systems in this temperature range was very limited. Thus, it was necessary to conduct special investigations in order not only to develop the biotechniques for red king crab rearing but also to elaborate the technology for recirculating systems in a low temperature regime. System efficiency is dependent upon filter and tank shape and size, type of bio-filter media, and rate and pattern of water circulation. Both natural seawater and artificial water prepared from the dry mixtures of marine salts may be used. All recirculating systems require periodical water replacement in order to compensate water losses. Thus the system should incorporate a tank for water storage or preparation (if artificial water is used).

The investigations included

- The use of biological filters, protein skimmers (flotators), and UV disinfection units in recirculating systems.
- Start-up (initial phase) and functioning of biological filters in a low temperature regime (Kovatcheva and Kalinin 2005, Kovatcheva et al. 2005, Kalinin et al. 2005).

Culture techniques

Capture and handling of ovigerous females

It is expedient to start artificial reproduction with the capture of ovigerous females when the eggs are at late zoeal stage of embryogenesis, i.e., several weeks before hatching. In the North Pacific Ocean, the females should be caught in late March or early April, whereas in the Barents Sea it is better to catch the females no later than the end of February, when the embryos are about 300 days old (Kovatcheva 2002b, Kovatcheva et al. 2005). Female crabs may be collected by scuba divers or captured by standard king crab pots (rectangular or conical) set at a depth from 30 to 70 meters.

Transportation time should preferably not exceed 20 hours. Ovigerous females and their eggs are very sensitive to water temperature fluctuations. Therefore, water temperature during transportation should not differ significantly from the water temperature in the area where the females were caught, and should preferably be held relatively constant in a range from 1.5 to 3.0°C. The water temperature in the tanks should be adjusted so that it is the same as in transportation container, in order to minimize stress. Afterwards it is recommended to keep water temperature at 3-4°C. Monitoring of the females' behavior thus provides an opportunity to predict the beginning of hatching, which is very important in terms of larval survival. When the females are held under optimal conditions, the hatching rate comprises 95-100%.

Once hatching has occurred, the females should be released back to the area where they were caught. Prior to transportation the females should be acclimated to the water temperature in their natural habitat at the time of release, as it may differ from handling conditions.

Larval culture

Red king crab larvae are especially vulnerable to stress, i.e., fluctuations of environmental factors. Therefore, culture conditions and parameters, such as

- water quality parameters,
- rearing densities,
- feed quality and availability, etc.,

should be carefully monitored and maintained at optimal levels during the whole larval phase.

Red king crab larvae are highly cannibalistic even at the first zoeal stage (Borisov et al. 2005). Therefore, in mass-culture the rearing density should preferably not exceed 50 larvae per liter (Russian Federation Patent No. 2200386).

Feeding

Fresh *Artemia* nauplii hatched at 28°C for 24 hours were used as food. *Artemia* cyst sources were lakes of the Altai Region (Russia). Our previous experiments have shown that while kept in the seawater at 8°C and 32 g per L all *Artemia* nauplii remain alive and active for 12-14 hours; the decrease of nutritive value of nauplii under these conditions is less than 5% (Lavens and Sorgeloos 1996). Therefore, we have chosen a 12 hour interval between feedings. Optimal initial *Artemia* nauplii concentrations for feeding zoeae I-IV constitute 400-600, 600-800, 800-1,000, and 1,000-1,200 nauplii per L, respectively (Kovatcheva et al. 2005, Epelbaum and Kovatcheva 2005). Daily food intake for each zoeal instar fed *Artemia* nauplii at 7-8°C are summarized in Table 1.

Development and growth

Average values and standard deviations for growth and development parameters for each zoeal instar are shown in Table 2. Average duration of larval period constituted 39 days (284 cumulative degree days). Our data on growth and development of larvae differ from those of Nakanishi (1978), who worked with larvae obtained from the females caught in the North Pacific. In our experiment, carapace length of zoea I was 1.390 mm, whereas in the experiment of Nakanishi it was only 1.099 mm (Kovatcheva 2002a,b; Kovatcheva and Epelbaum 2003).

Table 1. Daily food intake of zoeae I-IV at 8°C.

Zoeal stage	Daily food intake (M)		
	nauplii/ind	mg (wwt)/ind	µg (dwt)/ind
I	11.3	0.294	47.46
II	22.4	0.582	94.08
III	33.2	0.863	139.44
IV	41.8	1.087	175.56

Abbreviations: dwt = dry weight; wwt = wet weight.

Table 2. Development and growth of red king crab zoeae reared at 7-8°C

Stage	Duration, days/ degree-days	Carapace length (±SD), mm	Rostrum length (±SD), mm	Individual wet/ dry weight, mg
Zoea I	10 / 66.0	1.39 ± 0.029	1.29 ± 0.038	0.86 / 0.110
Zoea II	10 / 68.7	1.63 ± 0.027	1.52 ± 0.089	1.41 / 0.165
Zoea III	9 / 69.3	1.83 ± 0.044	1.53 ± 0.121	2.00 / 0.250
Zoea IV	10 / 79.7	2.07 ± 0.043	1.63 ± 0.084	2.67 / 0.300

Survival

The results of our study have shown that rearing red king crab larvae under controlled laboratory conditions at 7-8°C and rations shown above provides an opportunity to increase survival of larvae up to 35%. In our experiments, relatively high mortality rate was observed throughout larval development, especially during the molting process. The highest mortality rate was observed after the first molt and during second zoeal stage.

Glaucothoe phase

Feeding

Studies on the anatomy of digestive tracts of larvae, glaucothoe, and juveniles (Abrunhosa and Kittaka 1997; Epelbaum 2002) and laboratory observations (Kovatcheva 2002b) confirm the conclusion that red king crab glaucothoe do not feed and represent a secondarily lecithotrophic stage. The term “secondary lecithotrophy” was proposed by Anger (1989) for nonfeeding stages that develop with energy reserves accumulated during the preceding larval phase.

Substrate preferences

The following types of substrata were tested:

- Vertically oriented nylon net (0.5 mm) stretched over a plastic framework (“net 1”).
- Horizontally oriented nylon net (0.5 mm) stretched over a plastic framework (“net 2”).
- Vertically oriented flat mats of plastic filament (mechanical filter media) (“mat 1”).
- Horizontally oriented flat mats of plastic filament (“mat 2”).

From 79.2 to 83.8% of glaucothoe preferred horizontally oriented flat mats of plastic filaments. This substrate can be easily grasped by glaucothoe and provides a high degree of interstitial space. Therefore, safe and easy maintenance mats of plastic filaments can be recommended as suitable substratum for rearing red king glaucothoe (Kovatcheva et al. 2005).

Development and growth

In our experiments, carapace length of glaucothoe averaged 1.85 ± 0.035 mm, and carapace width 1.63 ± 0.05 mm. Individual wet weight averaged 3.77 mg, dry weight 0.679 mg. The duration of glaucothoe phase at 10-11°C was 18-20 days (177.7-200.0 degree-days).

Survival

When glaucothoe were reared under our conditions (closed recycling water system) survival was approximately 90%.

Early juvenile phase

First-stage juveniles are considerably more viable than zoeae and glaucothoe, but they exhibit strong cannibalistic behavior and thus are highly prone to mortality due to cannibalism, especially when reared in mass culture at high densities (Borisov et al. 2005). Therefore, when the red king crab is cultured with the aim of repopulating depleted natural stocks, it is expedient to release first-stage juveniles into the ocean as soon as possible (Kovatcheva 2002b, Kovatcheva et al. 2005).

Rearing reservoirs, substrata, and transfer methods

An important factor for successful rearing of juveniles is the availability of suitable substrata that the juveniles can easily grasp and walk on. Juveniles may be simply reared in the cells with a rugged bottom. Another option is to use the cells with a smooth bottom, but place additional substrata into them (Kovatcheva et al. 2005). When juveniles are reared in mass-culture, we recommend using the substrata that maximize the rearing reservoir volume usage, e.g., loosely arranged thick mats of plastic thread. This will ensure more even distribution of the juveniles and reduce the level of cannibalism.

Feeding

Raw meat of marine invertebrates (squid, shrimp, and mussels) was found to be the most appropriate food type for red king crab juveniles reared under laboratory conditions. Feeding should be started on the first day after metamorphosis. Juveniles should preferably be fed twice a day, with 12-hour interval between feedings. Prior to feeding, food should be cut in small pieces.

Transportation and release to the ocean

Prior to transportation, the juveniles should be acclimated to the water temperature in their natural habitat at the time of release, as it may differ from the rearing conditions. Juveniles may be released at previously selected sites, which should have enough natural shelters and food or prepared artificial substrates (collectors, reefs, etc.).

The knowledge of the biology of red king crab early life history stages has formed a basis for establishing basic culture methods and techniques: technical specifications and documentation for planning, designing, and building experimental crab rearing facilities with the aim to further work on the technology of red king crab artificial reproduction and cultivation.

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King Crab Cultivation and Stock Enhancement in Japan and the United States: A Brief History

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Introduction

Much of what we know about the biology and cultivation of king crabs is the direct result of research conducted by Japanese scientists. Likewise, initial efforts to cultivate and release crabs have been conducted by the Japan Sea Farming Association. This report discusses the history of those efforts.

Early work

The development of king crab larvae was first described by Marukawa (1933) who raised individual larvae captured in plankton samples. Sato (1944, 1945), and Sato and Tanaka (1949a,b) raised larvae from eggs in the laboratory and made detailed descriptions of larval development. Kurata (1960a) studied the consumption of *Artemia salina* nauplii by king crab zoea larvae at 8 to 12°C, and found that each successive stage consumed more nauplii. An average total of 90, 163, 230, and 296 nauplii were consumed during zoeal stages I-IV, respectively. Daily consumption during stages III and IV was 26 and 37 nauplii per day at 10°C, and 48 and 42 nauplii per day at 12°C. Zoeal stage I consumed more nauplii at night, but later stages showed no preference. Kurata (1961) reared larvae from eggs to stage C₁ in the laboratory of Hokkaido University at Akkeshi, then shipped them by train to another laboratory at Yoichi, 15 hours away, but because of the temperature changes they experienced, few crabs survived past stage C₃. Nonetheless, he was able to demonstrate the relationship between growth rate and degree-days. He also showed that survival was poor at salinities below 12 ppt (Kurata 1960b).

The most comprehensive studies on cultivation of king crab larvae were conducted by Takashi Nakanishi, who studied the effect of temperature and salinity on growth, survival, and oxygen consumption of king crab larvae

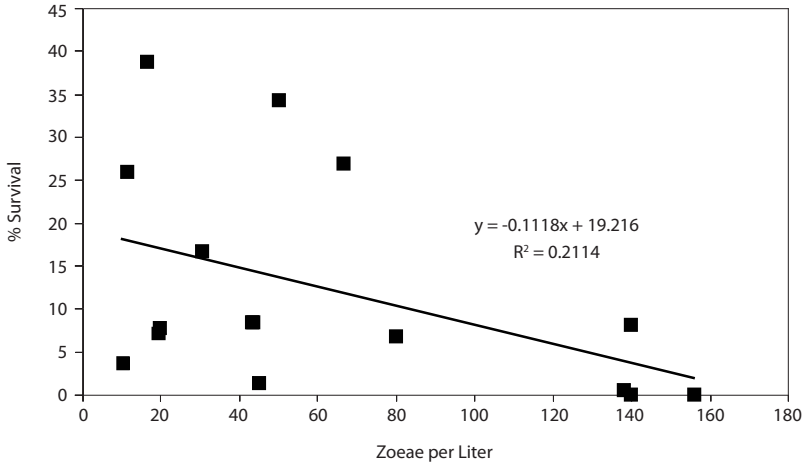


Figure 1. Relationship between cultivation density and survival of red king crab zoeae (Nakanishi 1988).

(Nakanishi 1981,1988). He concluded that the optimum temperature for cultivation was 8°C. Early efforts to cultivate larger numbers of larvae (500 to 70,000 per tank) were conducted in 30-50 L tanks at 3°C, and zoeae were fed with *Artemia salina* nauplius larvae, rotifers (*Brachionus plicatilis*), and minced clams (Nakanishi 1988). Densities of larvae ranged from 11 to 156 zoeae per L, and mean survival from the first zoea (Z₁) to the first crab stage (C₁) was 15.5% (range 4.4-34.3%) in 30 L tanks, but only 2.3% (range 0-16.6%) in 500 L tanks. Although overall survival declined with density, there was no relationship at densities below 80 zoeae per L (Fig. 1). Nakanishi noted that the majority of larval mortality occurred as a result of cannibalism, and a smaller but still significant percentage died of “failure to molt.” Like other researchers before him, Nakanishi fed *Artemia* to glaucothoe-stage larvae, and concluded they were consumed, based on their disappearance and the survival of the glaucothoe. However, he did not raise unfed controls to verify if consumption was actually occurring. Nakanishi summarized his research on crab cultivation in a magnum opus titled *Rearing Condition of Eggs, Larvae, and Post-Larvae of King Crab* (Nakanishi 1987). This document was published in Japanese, but has been translated into English for the National Marine Fisheries Service. In it, he describes the embryonic development, oxygen consumption, temperature and salinity tolerance, lighting conditions, and food requirements for optimal cultivation of king crab larvae and juveniles. To date, it is the primary source for information on the biology of larval and juvenile king crabs, but the information contained is too comprehensive to summarize herein.



Figure 2. Conical-bottom plastic kreisels used for cultivation of red king crab zoeae by Dr. Jiro Kittaka.

Later work

For a decade, from the mid 1980s to the mid 1990s, little further research was conducted on king crab cultivation. In 1993, Dr. Jiro Kittaka was hired to direct the Nemuro City Fisheries Research Laboratory in Nemuro, Hokkaido, Japan. Dr. Kittaka had previously been instrumental in the development of cultivation techniques for penaeid shrimp (Kittaka 1981), as well as for spiny lobsters (Kittaka 1988, Kittaka and Kimura 1989, Kittaka 1994). He developed a recirculating kreisel system in which lobster phyllosoma larvae could be cultivated with high densities of microalgae (Kittaka 1997). In Nemuro, he applied similar technology to the cultivation of king crab larvae, and concluded that the glaucothoe stage did not feed on *Artemia* (Kittaka 1995). The non-feeding nature of glaucothoe was verified by subsequent work showing that the morphology of the digestive system degenerated during the glaucothoe stage (Abrunhosa and Kittaka 1997a,b).

In 1996, I spent a year working with Dr. Kittaka in his laboratory in Nemuro. During that time, we conducted research on the optimal temperature and density for larval cultivation of both red king crab (*P. camtschaticus*) and the spiny or Hanasaki king crab (Hanasaki-gani, *P. brevipes*), which is most

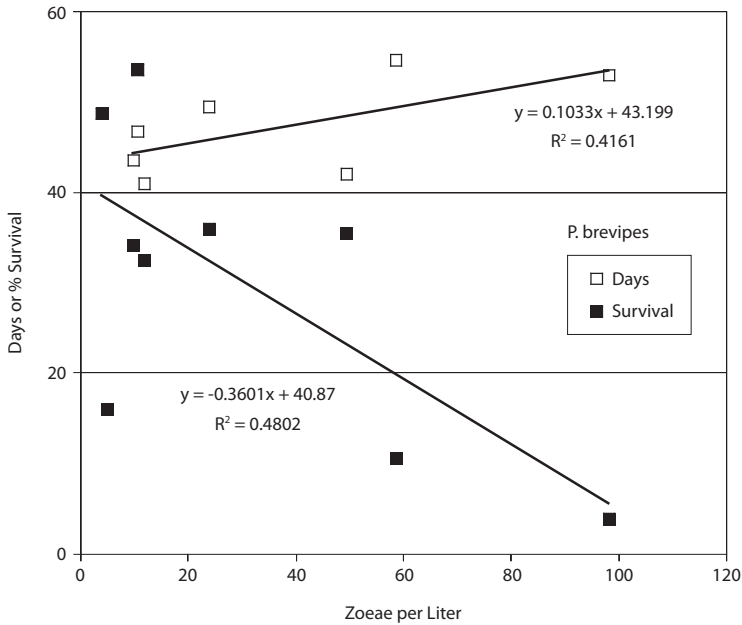


Figure 3. Relationship between survival and development period for zoea larvae (to stage C1) and density (from Kittaka et al. 2002).

common in Hokkaido. Dr. Kittaka's techniques were similar to those of his predecessors, in that many different combinations were tried, with little replication or statistical analysis. Nonetheless, he had surprisingly good results. Larvae were raised in 100 L conical bottom kreisels (Fig. 2), and fed on various combinations of *Artemia* and the chain-forming diatom *Thalassiosira nordenskiöldii*. Survival was negatively correlated with density (Fig. 3), but the correlation between density and length of larval development (to stage C1) was not significant (Kittaka et al. 2002). The best survival rates to the first crab stage were in the range of 30-50%. This technique worked well enough that Dr. Kittaka developed a larger version capable of holding 2 t of water.

Kittaka et al. (2002) also experimented with different ways to enhance the nutritional value of *Artemia* by feeding it with *Thalassiosira* that had been coated in tuna oil, *Spirulina* enriched with highly unsaturated fatty acids (HUFA), or beer yeast enriched with HUFA. Survival of *P. brevipes* fed with diatom-enhanced *Artemia* was low (>12%) but still significantly higher than other treatments, all of which had survival <6%. Fatty acid analysis showed that *Artemia* has some eicosapentaenoic acid (EPA, 20:5(n-3), but very low amounts of docosahexaenoic acid (DHA, 22:6(n-3); *Thalassiosira* had twice as

much EPA as *Artemia*, and enhanced feeds had 1-2 orders of magnitude more EPA and DHA than un-enhanced feeds (Kittaka et al. 2002). After feeding with enhanced diets, levels of EPA and DHA in *Artemia* were five times higher than in non-enhanced *Artemia*. Ultimately, improving the quantity of HUFAs such as EPA and DHA should result in better survival of king crab zoea larvae.

Settlement by glaucothoe larvae is also an important consideration. Besides understanding the types of substrata required for natural settlement, such knowledge can help provide insight into the best ways to release cultivated juveniles. Laboratory experiments with red king crab (*P. camtschaticus*) have shown that glaucothoe prefer to settle in habitats that are structurally complex, can be easily grasped, and provide a high degree of interstitial space, i.e., those with a high fractal dimension. In experiments using sand, gravel, and synthetic mesh aquarium filters, glaucothoe began settling on the first day, and <10% remained swimming after day 6 (Stevens and Kittaka 1998). Glaucothoe showed a significant preference for the structurally complex mesh substrate, and occupancy increased from 49% on day 2 to 75% by metamorphosis to the first crab (C1) instar. Glaucothoe rejected sand, and only 1% were observed on it. Glaucothoe in gravel- or mesh-only aquaria settled rapidly, whereas 40% of glaucothoe in sand-only aquaria continued swimming until metamorphosis to stage C1. In experiments with natural substrata, glaucothoe preferred hydroids > algae > airstones and tank walls > sand > calcareous worm tubes (Stevens 2003). Complex habitats may provide settling crabs with shelter from predation during critical early stages. This hypothesis was tested by placing glaucothoe and juvenile crab in aquaria with or without artificial habitats, and with or without predators (1-3 year old red king crab) (Stevens and Swiney 2005). Predators caused increased mortality of glaucothoe regardless the presence of shelter habitats, possibly due to the active, substrate-testing behavior of glaucothoe. In contrast, juvenile crabs occupied habitats in higher densities than did glaucothoe, which tended to reduce predation. These results indicate the importance to settling larvae of biogenic oases, and underscore the importance of conserving such habitats. Furthermore, knowledge of settlement, habitat selection, and substratum preference are essential prior to considering the potential of king crabs for stock enhancement or aquaculture.

Commercial scale production

Marine organisms produce many eggs, but few survive due to predation and starvation. Recognizing this, the Japan Sea Farming Association was formed to support fish and shellfish propagation. The organization is a cooperative venture between national and prefectural fisheries research agencies, and local fisheries cooperative associations. The objective of sea farming is to produce large quantities of "seeds" (juvenile fish and shellfish) under human control, and release them into the ocean after they become large enough to improve



Figure 4. Cultivation tank at the Akkeshi Sea Farming hatchery in Hokkaido, Japan.

their chances of survival. Sea farming also requires measures to ensure adequate management, conservation of brood stock, and rational conservation. Sea farming centers were established at many locations around Japan, and are responsible for cultivating over 50 species of fish and shellfish. One such center in Akkeshi, Hokkaido, includes five buildings containing over 52,000 ft² of floor space. The Akkeshi hatchery has produced seeds of the hanasakigani (*P. brevipes*) since the mid 1980s. Juvenile crabs are produced in seasonal rotation with barfin flounder, herring, and hair crab (*Erimacrus isenbeckii*). In September 1996, I visited the hatchery and interviewed the director, Mr. Imamura.

Cultivation consisted of four major steps: brood stock production and egg collection; seed production; intermediate rearing, and release. In March, wild-caught females were brought to the laboratory and held at 4°C until their eggs hatched. After hatching, approximately one million larvae were raised at 8°C in large vats of 100 m³ volume (Fig. 4), at densities of 10,000 per m³ (or 10 zoeae per L). Larvae were fed with a diet of *Artemia* and diatoms (*Thalassiosira* sp., at 3,000 cells per ml). In addition, microalgae (*Nanochloropsis* sp., at 500,000 cells per ml) were added to stabilize the diatom population. Upon reaching the glaucothoe stage about 42 days later, they were allowed to settle into net bags. The bags were then placed in the ocean to acclimatize (a process called “hardening the seed”). After 3 months, scuba divers released the small crabs from their net bags onto a specially prepared substratum of gravel. The little crabs presumably crawled into the spaces between the gravel where they found plenty of natural food.

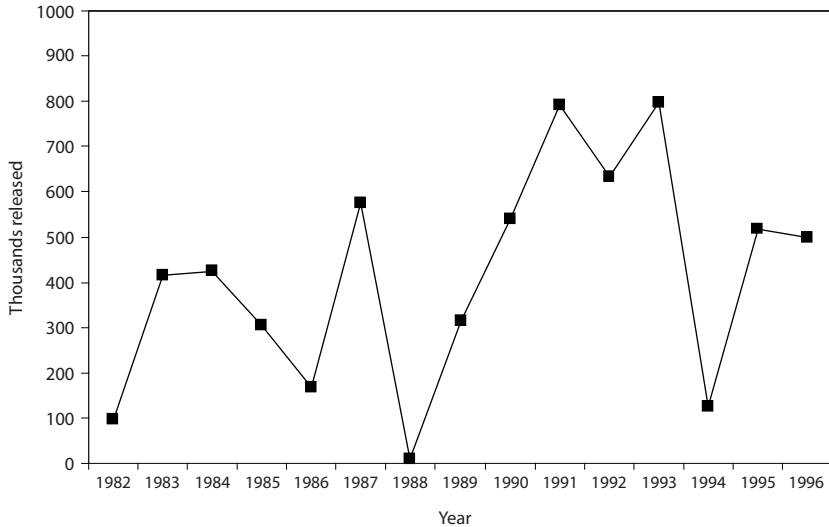


Figure 5. Production of stage C1 Hanasaki-gani at the Akkeshi hatchery (data through 1996).

From 1982 to 1996, production of Hanasaki-gani seed at Akkeshi was highly variable, ranging from 0 to 800,000 per year (Fig. 5); average production was 415,000 crabs per year, representing survival of about 42%. In 1996, Akkeshi released 500,000 seeds. However, they did not conduct follow-up research to determine the survival rate of released crabs, so nothing is known about survival, impact on the fishery, cost of production, or the overall effectiveness of the program. One can only conclude that culture of marine fisheries species in sea farming hatcheries is socially and economically important to Japan whatever the cost, so it is conducted routinely. However, the questions of effectiveness and cost/benefit have not been adequately addressed.

Recent progress

After the opening of the Kodiak Fisheries Research Center in 1998, we began small scale efforts to cultivate king crab larvae. Our primary goals were to produce a few thousand juvenile crabs for use in ecological research (detailed above). Kittaka's success in growing both *Thalassiosira* and crab larvae gave us perhaps unrealistic expectations, which turned out to be rather optimistic. Our early efforts to cultivate *Thalassiosira* were not very successful, due to differences in strains, temperatures, and water conditions, and attempts to raise king crab zoeae using other diatoms were also disappointing. Several different strains of *Thalassiosira* were tested, including wild strains from local waters, but they were quickly overwhelmed by other diatoms. Several years of

diligent work and highly variable results were required to develop repeatable techniques. The culmination of this work, using blue king crab (*P. platypus*), was an experiment in which we achieved an unprecedented 92% survival from hatching to first crab stage (Persselin et al. 2006).

The information presented herein can now be used as the basis for the next phase of crab enhancement research. Questions specific to cultivation technology will need to be addressed. Is it possible to cultivate king crab larvae in Alaska on a large scale, as it has been done in Japan? What equipment, techniques, and costs will be necessary? How should juvenile crabs be released: what stages, time of day, length of “hardening,” and type of substratum should be used? How will cultivated crabs be identified, and the effectiveness of such a program evaluated? And finally, what is the cost of such a program? Other articles in this volume may provide some answers to those questions.

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Improving the Survival of Cultivated and Released Juveniles

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Abstract

Release of hatchery-reared lobsters is often suggested for enhancing recruitment-limited populations. Due to lack of ecological considerations ahead of the releases and the unsuccessful efforts to find juvenile lobsters in the sea, little has been known about survival rates, ecological impacts of the releases, and how to improve the performance of releases. Recent studies of morphology and behavior in hatchery-reared homarid lobsters demonstrate that qualitative, small-scale laboratory experiments, in combination with larger scale field studies, can yield such information. Based on significant results from the laboratory, we should be able to design field studies with focus on expected ecological “bottlenecks” instead of the trial-and-error field studies known from the past, thereby reducing time and investment in the development of viable stock enhancement/sea ranching activities. This is a review of studies of rearing conditions, transportation, and handling during release that aimed to diminish the occurrence of conspicuous morphology and behavior in the reared juveniles, causing reduced competitive ability, slow sheltering speed, and thereby high mortality rates in the sea. We need to combine small-scale studies with field studies to be able to confirm the significance of the laboratory results. This presentation is based on van der Meeren (2003) and the Ph.D. thesis van der Meeren (2005).

“For every complex problem there is a simple, easy to understand, incorrect answer.” —Albert von Szent-Györgyi (1893-1986)

Introduction

Cultivation for release purpose

Rearing useful organisms has been a part of human culture for thousands of years. Manipulation of plants and animals has transformed most terrestrial parts of our globe. It seems to be part of human nature to interfere with nature on a large scale. Until the last century, the consequences of such actions were

not considered. As long as the cultivation was on land or in drainable dams, it was possible to see and measure how well the cultivation techniques functioned by the size of the production. Cultivation of the ocean has turned out to be more challenging. Lack of basic knowledge about life history and behavior of the chosen organism, the environmental requirements of that organism, and the ecosystem it lives in, has led to many failed marine cultivation projects, where only a few studies have been done to explain the lack of success. Three major types of population conservation or cultivation have been described: mitigation (altered or alternated habitat with recruit limitation); augmentation (habitat expected to be below carrying capacity due to recruit limitation); and community change (species transplantation) (Bartley 1999).

The purpose of mitigation is to compensate for lost habitat, and it requires preparation of a high quality release environment (Bartley 1995). Construction of artificial reefs has been tried in marine environments (Jensen et al. 2000).

Augmentation is suggested as a management tool to reintroduce or increase the natural stock above the present level. This is often the case when the natural population for some reason has decreased to such a low level that it is reasonable to expect failure in natural recruitment. In the cases of the gray wolf (*Canis lupus*) in Yellowstone National Park, in the northern Rocky Mountains of the United States (Fritts 2000), and white-tailed eagles (*Haliaeetus albicilla*) in Scotland (Gregory et al. 2002), re-establishment of the breeding populations was successful. An attempt to save the African rhinoceros from extinction has been made, by using captive breeding, release, and relocation (Emslie and Brooks 1999). It is not common to find successful cases from marine releases. No return has yet been seen in reared Kemp's ridley sea turtles (*Lepidochelys kempi*) after releases on Padre Island in Texas (Fontaine et al. 1989). However, boosting of a local stock of lobsters (*Homarus gammaurus*) has been successful in Norway (Agnalt et al. 1999, 2004).

The third type, community change, is where exotic species are introduced into a new biotope. It has been done in Europe, for instance, by releasing red king crabs (*Paralithodes camtschaticus*) in the Barents Sea (Kuzmin and Olsen 1994, Jørstad et al. 2002) and muskoxen (*Ovibos moschatus*) in Russia, Norway, and Alaska (Wilson and Reeder 1993). The best known case is the introduction of European mammals, such as rabbits (*Oryctolagus cuniculus*), to Australia (Wilson et al. 1993). Introduction to new locations raises a lot of special concerns, since it can lead to unexpected ecological impacts, from unexpected performance of the species, unlike in the natural home-range, via out-competing the native species to the introduction of new pathogens (Bartley 1996). Also unintentional introductions have happened and will occur in the future, both in terrestrial and aquatic environments. The potential biological and economical damage of such introductions in the marine environment can be illustrated in cases of the Pont-Caspian zebra mussels (*Dreissena polymor-*

pha), driving native mussel species to extinction and destroying water pipes and pumps in the Great Lakes between the United States and Canada worth several billions of dollars a year (Khalanski 1997). Another account was the change in the whole pelagic food chain in the Black Sea after the introduction of the North American ctenophore *Mnemiopsis leidyi*, predated on pelagic plankton and leading to more than 90% reduction in fish landings in the area (Gollasch and Leppäkoski 1999).

No matter if the release is intentional and based on native or exotic species, or unintentional by exotic species, it is the same ecological forces that lead to either successful establishment of the introduced organisms or failure followed by disappearance of the organisms.

Species of interest for cultivative releases

Species thought fit for ocean ranching are usually high-priced sessile or very slow-moving species, which will stay at the place they are released. This is the case for mollusks and echinoderms. In addition, motile, but stationary species such as decapod crustaceans, e.g., prawns, shrimps, and clawed lobsters are suggested as potential species for sea ranching. It is important that they stay in one place, to allow the owner to have control over the area and the stock. Sea ranching of mollusks such as scallops is already established in a number of countries (Fleury et al. 1997, Bell 1999, Dao et al. 1999, Nadeau and Cliché 1999). Yet the impact such “monocultures” might have on the surrounding biotope is not studied. Naturally occurring predators have been a major problem (Fleury et al. 1997).

Stock enhancement has been tried out or is suggested for a larger range of invertebrates and vertebrates, both sessile and motile species, as a method for conservation of biodiversity. While ocean ranching is based on low-cost production of organisms of commercial value, motivation for stock enhancement arises from a range of situations, from variable annual landings to recruitment failure, destruction, addition of suitable nursery habitat, climate changes, food supply, and pollution (Addison and Bannister 1994, Grossman et al. 1997, Lindberg 1997, Gendron 1998, Doherty 1999, Smedstad et al. 1994, Castro et al. 2001) Cost-efficiency is not the only motivation, and species mobility is no longer of importance in the same way as for sea ranching. The aim is to reconstruct a natural stock that is managed by fishery and natural management rules.

Even if the motivations behind sea ranching and stock enhancement in many ways are different, they have the same foundation of breeding, rearing, and release of young organisms. The strategy is safe rearing of young larvae and/or juveniles through the most vulnerable life stages, and release to their natural biotope when they are thought to be more robust or safe from predators, in order to maintain a stable recruitment of the selected species in the

chosen area. The term “settlement” is therefore used in a wider sense in this thesis. It is usually a definition for the termination of a pelagic larval phase and assumption of a benthic life (Scheltema 1974), but in this thesis it is also used to define the first time an already bottom-settled organism is transferred from land-based holding facilities to natural or semi-natural habitats.

If recruitment analyses show why a stock is depleted, and are followed by ecology studies that document a recruitment bottleneck, it should be possible to suggest how this can be avoided by rearing the vulnerable life stage. This must be the foundation when initiating release of hatchery-reared animals for stock enhancement purposes. However, it is quite common that knowledge of both recruitment biology and the species ecology is lacking and difficult to study (Laurec 1999, Tsukamoto et al. 1999).

Another objection to stock enhancement has been the lack of controlled research in connection to the releases, as well as missing assessments of the result of the releases (Laurec 1999). In addition, investment has been put into the construction of rearing facilities and cultivation of animals in the most cost effective way, as it is quite accomplishable to calculate the cost and economic value of the production of these technical and fully controlled operations (Wickins and Lee 2002). Least investment has been put into biological and ecological studies of the chosen organism, the most suitable release sites, and long-term monitoring to evaluate the impact of the releases. Such studies are usually expensive and it is difficult to evaluate the economic value of the results. Even studies of the basic needs of the release organism itself, except for food, growth, and survival, are rarely accomplished. The result is that most of the release attempts worldwide have failed to give reliable conclusions on how they succeeded in enhancing the manipulated stocks. No documentation on the impact on the biotope is available.

The North Atlantic clawed lobster (*Homarus* sp.) has been the subject for cultivation and release-programs since the 1880s in Norway (Dannevig 1885, Nicosia and Lavalli 1999), and particularly in Europe, as the European lobster (*Homarus gammarus*) seems to have lower fecundity and reproduction rates than the American lobster (*Homarus americanus*) (Aiken and Waddy 1980, Free 1998). Enhancement efforts based on reared and released juveniles, as is now suggested for the red king crab *Paralithodes camtschaticus*, was also the motivation behind the Norwegian lobster stock cultivation, which is representative for enhancement enterprises, based on severe stock depletion and well-functioning rearing technology, but with lack of biological and ecological data (Svåsand et al. 2005).

Behavioral and ecological studies

It is important to develop release strategies in ways that ensure survival of the released organism and at the same time consider how the biotope might be affected by the release. The selection forces—settling conditions and behav-

iors, shelter need, food availability, growth, mobility, inter- and intraspecific interactions, including anti-predatory response, and eventually reproductive ability and mortality—are working at the individual level, and the properties of populations and communities emerge from the behavior of individuals (Butler 1997) (Fig. 1). This talk is based on studies of the European lobster but in many aspects is of general relevance also for other species. The research is based on ecological and behavioral studies connected to research programs for lobster stock enhancement and management in Norway and Europe since 1988 (see Appendix).

Standardized rearing techniques might affect the morphology as well as the behavior and thereby the ability of the animal to survive and respond in a functional way to complex and changing environmental challenges. Also, transportation and release techniques might have short-time influence on behavior. Thorough field studies and understanding of the ecology of wild juveniles will allow for designing the best possible release strategy. Since wild European lobsters have been impossible to find between larval and emerging phase two to five years after settling, there was no reliable information available on natural demands, behavior, and morphological development during their early life stages; the research was done on hatched and reared juveniles only. In addition, the benthic ecosystem of lobster biotopes has previously not been thoroughly studied.

The ability to survive and adapt are related to:

1. Morphology, physiology, and neurology.
2. Stressors prior to and at release (handling, temperature, light).
3. Substrate and shelter-related behavior.
4. Competition, both intra- and interspecific.
5. Predators.
6. Foraging and food.

The talk at this workshop discusses survival, functionality, and adaptation to several of these topics, and are based on the publication by van der Meeren (2005).

Conclusions

Quality of the release organism

Hatchery-reared lobster juveniles grown in traditional hatcheries cannot exercise and have no experience of the seasonal cycles, the use of shelters, predators and techniques for predator avoidance, foraging, social interaction, or competition. It is also a possibility that brain development in decapod crustaceans might be underdeveloped because of lack of stimuli (Sandeman and Sandeman 2000). As the development of the crusher claw is dependent on physical exercise between molt stages V and VIII, this development usually

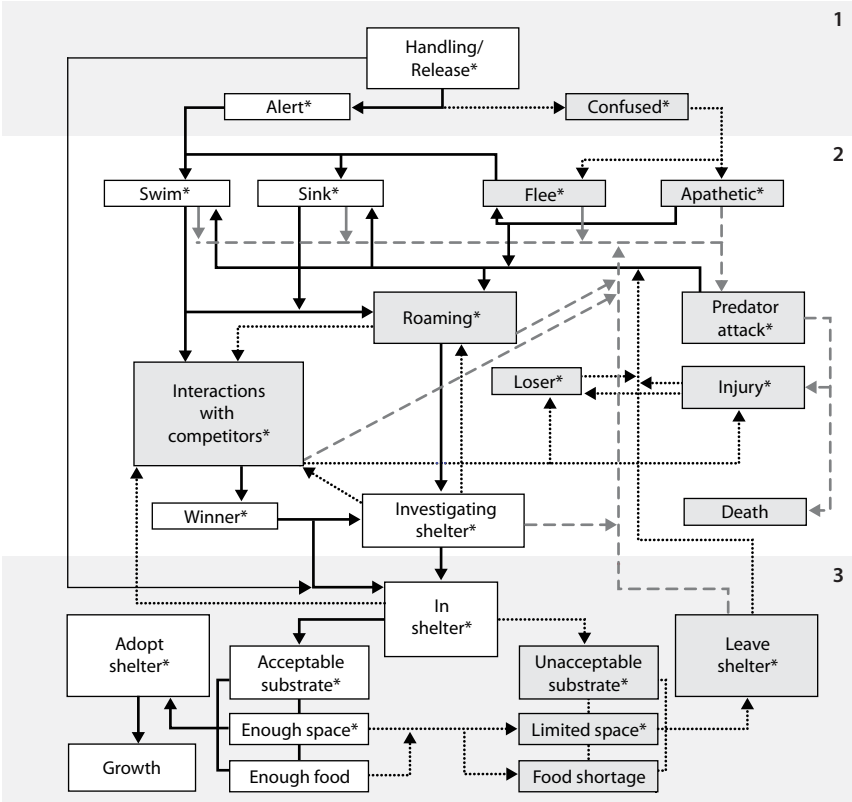


Figure 1 covers the three parts:
 1. "Handling and Release," which is connected to the rearing- and release procedures, and
 2. "Roaming and Investigating" as well as
 3. "In shelter," which is common for all organisms, both reared and released and wild ones.

The different actions and incidents are
 Wanted actions = Risk-decrease (open boxes; solid line);
 Other actions = Risk-increase (gray boxes; dashed line); and
 Unwanted incidents = Life-threatening (gray boxes; broken line).

The asterisks (*) indicate the actions presented and discussed in the present paper.
 The thick line is expected to be the optimal development of a successful establishment. Impacts imposed on the organism by morphological or neurological abnormalities due to the rearing conditions, and physical impact by temperature, currents, and/or salinity is left out of this model.

Figure 1. Flow diagram describing the settling process of released benthic organisms. Source: van der Meeren (2005).

fails in hatcheries that rear juveniles in boxes with no supplement of coarse shell sand or shell spat (Wickins 1986). In addition, growth rate after release, nutritional status, and mobility might be different between reared and wild lobsters (Addison and Bannister 1994).

The behavior of hatchery-reared species has been linked to increased predation rates (Olla et al. 1998, Svåsand et al. 1998) and reared lobsters may become more prone to predation (Spanier 1994). In addition, stress induced by packing, transportation, handling, and the sudden change of environment when transferred from the hatchery to the sea must have an impact on the lobster juveniles (van der Meeren 1991, 1993).

It is of utmost importance that the organism is reared free of disease and that genetic components are matching the wild population to avoid the changing of the gene pool (Allendorf and Ryman 1987, Ferguson et al. 2002, Strohmeier et al. 2002). Next, the life quality of the released organism must be fit for survival in the sea. Functional morphology and behavior at the time of release will influence how well the released animals manage to establish themselves after release (van der Meeren 1991, van der Meeren and Uksnøy 2000). Prior experience and training that increases the ability to avoid predators and competitors are probably important factors that will influence the survival chances of each individual (van der Meeren 1991, 2000, 2001). If the olfactory neural system is affected by rearing conditions, then this is another topic that should be investigated, as well as the fitness of the offspring of released organisms.

Biological considerations

Marine technical development has improved our ability to study the sea, but there is still a long way to go before we can understand benthic shallow-water ecology. Classical ecosystem theory is in general too abstract and oversimplified to address real-world issues (Suter 1981). Individual-based ecological models have now been developed, for a range of taxonomic groups, and also for aquatic fish and smaller planktonic crustaceans. Most of them are based on more realistic assumptions than state variable models, and are designed to understand how the system's properties emerge from the behavior of individuals that make up the system (Grimm 1999).

Lack of biological knowledge that should be considered and used in preparations ahead of the releases is perhaps the most common cause when release attempts have been deemed as failures, and leaves us without proper models to explain how the organisms as well as the ecosystem respond to the attempted management regimes. It is plausible that the stocks decrease, that initiated release actions, was actually caused by weak management due to lack of biological understanding in the first instance. Understanding the ecosystem of the sea is challenging as it is very complex and we are not a part of it. Temperature loss,

low visibility, currents, and limited air supply prevent us from observing the system in the same way that we can study terrestrial biology. Laboratory-scale experiments give us some insights and hypotheses, but need to be followed up by field studies for hypothesis confirmation or rejection.

Environmental considerations

Environmental considerations are usually focused on optimal conditions for the released organism, which in many cases is only known from hatchery-related research. Most releases take place in areas where the species are already established, or in similar areas with the same environmental impact, i.e., salinity, temperature, depth, bottom substrate, and currents. When information about environmental demands are missing or insufficient, the releases will be based on speculation, belief, and guesswork, and the risk of misplacement for the released animals is high. It is both time and money consuming to invest in costly rearing procedures if the animals are to be released in unsuited areas, chosen by speculation due to lack of knowledge.

It is also important to understand how the different environmental factors affect the organism, both physically and behaviorally. Increased growth potential due to high temperatures can lead to increased energetic demands, but if the organism is forced to face increased predation risk and fierce intraspecific, aggressive competition in order to obtain enough food it may be lost altogether. Environmental conditions that will allow the animals to avoid competitors and predators while they adjust to their new environments should be the best choice. Light intensity, currents, and temperature are all factors that influence animal behavior. Therefore, conditions that induce natural alertness and protection-seeking behavior are preferred.

Biodiversity and annual fluctuations in density of both the target species and the other species that are known to have the largest impact on the released animals should be known prior to the release. If low recruitment is the result of the population being close to carrying capacity, or limited nursery habitat, then the release of additional specimens will hardly be successful. With enough basic information about natural recruitment, including identification of the most important bottlenecks, a model could be run to evaluate how release of hatchery-reared animals might contribute to the wild stock, and perhaps to the biodiversity. It would then be possible to calculate if the release will lead to an increase in the output of the fishery. Such knowledge will also allow the releaser to decide size and density of the release batch and when and how releases should be done in order to avoid places and times when predators or competitors are present in high densities. Last, but not least, a documentation of the biodiversity ahead of the first release is needed, to be able to monitor the effect the released organism has on the environment. A thorough knowledge of the local biodiversity will also provide information that is needed to evaluate the consequence from release of alien species, as there is no ecolog-

ical difference in the constraints met by hatchery-reared or unintentionally introduced animals.

Considering the reasons for stock reduction

Finally, but not least important, the cause for the reduced, or collapsed stock must be known. Only if recruitment analyses show that a stock is depleted, due to a recruitment bottleneck in juvenile life stages, will it be useful to rear the vulnerable life stage for release purposes, if the release can be done by a ecologically sound strategy. The stock must then be managed in a way that ensures the stock to increase to and maintain within a reproductively sound level.

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Appendix

Recent research programs for ecological studies related to cultivation of the European lobster

Programme for Development and Stimulation of Ocean Ranching (PUSH)

The PUSH programme ran from 1990 to 1997, and was financed as a Norwegian governmental research programme to develop and evaluate the potential for commercial sea ranching of salmon, arctic charr, cod, and lobster. Prior to this, a pilot project was carried out on lobsters, and financed by the Fisheries Research Council from 1988 to 1990. The large-scale lobster releases in the PUSH programme were designed on the experiences from the pilot project and a British lobster release programme on the Yorkshire coast (Addison and Bannister 1994). A report on the study with preliminary results is presented in Agnalt et al. (1999, 2004).

*The Influence of Competitive Interactions on the Abundance of Early Benthic Stage European Lobsters (*Homarus gammarus* L.) and Hence on the Carrying Capacity of Lobster Habitat (LEAR)*

The LEAR study ran from 1997 to 1999, supported by the European Commission under the contract FAIR CT-96-1775. The participating institutions were the National University of Ireland in Galway, Ireland; Institute of Marine Research at the Centre of Aquaculture; Austevoll Marine Research Station, University of East Anglia in Norwich, UK; Centre of Environment, Fisheries and Aquaculture Science in Lowestoft, UK; and Università Degli Studi de Bologna, Italy. Although it failed to find wild early benthic phase lobsters, it was the first study in Europe to perform extensive faunistic sampling in lobster biotopes (Linnane et al. 2001).

*Coexistence between European lobsters (*Homarus gammarus* L.) and American lobsters (*Homarus americanus* Milne Edwards); Shelter use, activity, dominance and social behavior*

The Norwegian Directorate of Fisheries, the Norwegian Research Council and the Aquarium in Bergen supported this study, which took place in 2001. It was initiated by the fact that American lobsters were introduced into Norwegian waters (van der Meeren et al. 2000).

Cultivation Potential of Golden King Crab, *Lithodes aequispinus*

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Introduction

The golden king crab *Lithodes aequispinus* is commercially harvested on rocky substrates on both coasts of the North Pacific, from the fjords of northern British Columbia to Suruga Bay in central Japan. Although the golden king crab is similar in size to both red and blue king crabs, its deeper depth range, 150 to 900 m, slowed commercial interests until declines occurred in the red and blue king crab fisheries in the early 1980s.

Egg size and fecundity

Studies on golden king crab roughly paralleled commercial interests and little was known about the biology of the species until recent decades. The eggs are unusually large and yolky, with a length of 2.2 mm; for comparison, eggs of red king crab are 1.0 mm and blue king crab are 1.2 mm in length (Somerton and Otto 1986, Stevens 2006). This difference in egg length translates into a volume that is approximately 12 times greater than the egg volume of red king crab and six times greater than the volume of blue king crab eggs. The larger egg volume results in a correspondingly lower fecundity, approximately 10,000 eggs per female in comparison to 300,000 and 100,000 eggs for red and blue king crabs, respectively, although the fecundity of each species varies as a function of female size.

Incubation and hatching periods

One aspect of golden king crab biology that is attractive to aquaculturists is that females may hatch eggs any month of the year (Fig. 1). Red king crabs and blue king crabs are relatively synchronous and seasonal in egg extrusion, with the exception that primiparous females extrude eggs several months earlier (Shirley and Shirley 1989). Also, eclosion, hatching or release of larvae, is seasonal and pulsed for both red and blue king crabs, generally synoptic with the

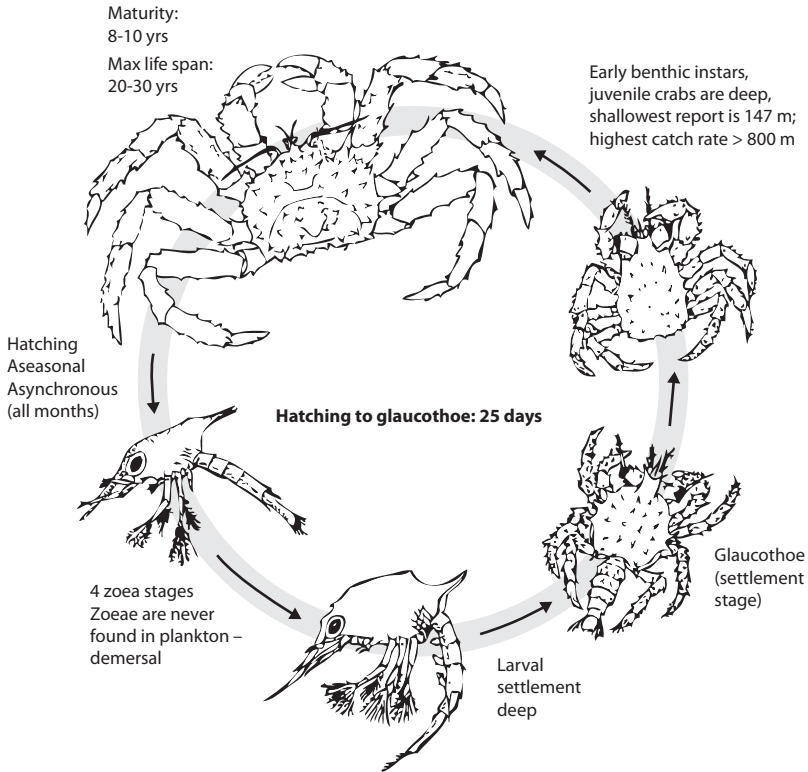


Figure 1. Life history of golden king crab.

spring phytoplankton bloom, again with the exception of primiparous females, which hatch eggs several weeks to a month earlier for red king crabs (Shirley and Shirley 1988, 1989; Shirley et al. 1990; Stevens and Swiney, submitted for publication; Stone et al. 1992). Hatching of blue king crabs is also thought to coincide with the spring phytoplankton bloom (Jensen and Armstrong 1989). In contrast, golden king crabs are asynchronous in the timing of egg extrusion, larval development (Somerton and Otto 1985), and time of hatching (Paul and Paul 2001a, Shirley and Zhou 1997). The mean span of hatching, e.g., the time between first and last hatching of eggs, for individual golden king crab females in the laboratory was $34 \text{ d} \pm \text{SD of } 16 \text{ d}$ (Paul and Paul 2001a), but hatching occurs in all months for captive females (Paul and Paul 2000, Shirley and Zhou 1997). This hatching span is similar to the span reported for red king crabs (Stevens and Swiney, submitted for publication).

A striking difference between golden king crabs and both red and blue king crabs is that the average time span prior to molting after eggs are hatched is exceeding long, $192 \text{ d} \pm \text{SD of } 72 \text{ d}$ ($n = 111$) (Paul and Paul 2001a), whereas the time prior to molting after hatching for red king crabs is approximately two weeks (T. Shirley, unpubl. observations; A.J. Paul, unpubl. observations). The long incubation period ($362 \text{ d} \pm \text{SD of } 78 \text{ d}$, $n = 59$) and long period between hatching and molting results in an unusually long period (590 d) between production of successive clutches (Paul and Paul 2001). These values should be considered conservative, as the golden king crab females were incubated in laboratory water temperatures that were $1\text{-}4^\circ\text{C}$ warmer than water temperatures at the depth where the females were collected; at colder temperatures surely ontogeny would have required longer. It is entirely possible that golden king crab might require 18-22 months for larval development if females are incubated at their in situ water temperatures of $2\text{-}4^\circ\text{C}$. The southern king crab *Paralomis granulosa* requires 18-22 months for embryonic development and has biennial reproduction (Lovrich and Vinuesa 1995).

Larval morphology, lecithotrophy, and development

Larval morphology and ontogeny of golden king crab are similar to most lithodid crabs, with four zoeal stages and a glaucothoe; one oddity is the lack of a prezoal stage (Haynes 1982). Another oddity is that in laboratory culture, all golden king crab larvae skipped one zoeal stage, either zoeal stage 3 or stage 4 (Shirley and Zhou 1997). Skipping larval stages is not unusual in crabs, but relatively few researchers notice the event because larvae are not cultured individually. Haynes (1982) mass cultured larvae in his experiments and did not report any missing stages. The larvae of *Lithodes antarcticus* (Vinuesa et al. 1985) and *Lithodes maja* (Anger 1996) were reported to have only three zoeal stages, but the larvae were not individually raised and larval stages might have been missed. However, the larvae of *Paralomis granulosa* have been cultured individually and have only two zoeal stages and a megalopa (Anger et al. 2003).

One aspect of the early life history of golden king crab that perhaps makes the species attractive to mass culture is that all larval stages are fully lecithotrophic (Shirley and Zhou 1997). The dry weight of unfed golden king crab larvae decreases slightly during development, from an average of 1.55 mg for the first stage zoeae (Z1) to 1.42 mg for glaucothoe and 1.39 mg for the first crab stage (C1). This total decrease in dry weight represents a 10% loss. No significant difference was found in dry weights of those larvae in fed treatments (food was provided) and the unfed treatments. Lecithotrophy often goes unnoticed because researchers fail to include an experimental treatment in which the larvae are unfed (Haynes 1985, Vinuesa et al. 1985). Several Japanese researchers have even provided optimum diets for rearing golden king crab larvae, with-

out substantiating that food was required for growth and development. Other species of lithodids (e.g., *Paralomis granulosa*, *Lithodes maja*) with large yolky eggs are now recognized as being fully lecithotrophic (Anger 1996, Anger et al. 2003) and others are strongly suspected. Introducing food to larval cultures increases the need for cleaning and water exchange, while also increasing the probability of introducing bacteria and other pathogens.

Another aspect that makes culture of golden king crab attractive is the relatively short larval period. The intermolt period for each zoeal stage averages less than 7 d. The larval period from hatching to the glaucothoe stage was approximately 25 d, regardless of which zoeal stage was skipped (Shirley and Zhou 1997); this is very similar to the larval period of *Paralomis granulosa*, another lithodid crab whose lecithotrophic larvae have a larval period of 24 d (Vinueza et al. 1989). The glaucothoe of golden king crab persisted an average of 41.3 d before settlement (Shirley and Zhou 1997). Again, these development and settlement times were in laboratory cultures of 7.0 to 9.5°C (although no treatment varied by more than 1.5°C), several degrees warmer than would have occurred in natural conditions. No special settlement substrates were tried, which might have shortened settlement times of glaucothoe. However, survival rate of larvae to the glaucothoe stage, particularly in the unfed treatments, remained high. Additional experiments with different water temperatures, different culture containers, and other culture conditions might yield even higher survival, without the necessity of rearing prey items or the possible introduction of pathogens or contamination. In sharp comparison to this relatively short 25 d larval period, the larval period for red king crabs is approximately 64 d, but varies with temperature, salinity, and food (Nakanishi 1987).

The larvae of golden king crab are almost unknown from plankton collections and are assumed to be demersal, remaining near the bottom or within specialized substrata. We have examined hundreds to thousands of zooplankton samples from a variety of depths in Alaska in our studies of red king crab larvae, without observing any golden king crab larvae (Shirley and Shirley 1989, 1990). In laboratory cultures the zoeae and glaucothoe of golden king crab remain near the bottom of culture containers and have reduced movements (Shirley and Zhou 1997), even though they are positively phototrophic (Adams and Paul 1999), as are most crab zoeae (Shirley and Shirley 1988).

Juvenile biology

After settlement and metamorphosis, the first crab stage (C₁) begins feeding, although sufficient energy reserves exist for some individuals to continue to molt to the C₃ stage. The duration of survival of unfed C₁ specimens averaged 89 d (\pm 28 d SD) (Shirley and Zhou 1997). The average carapace length (CL) of 32 C₁ juveniles was 2.5 mm (\pm 0.06 SD) and the average percent increase in CL was

28% (\pm 8% SD) for 76 specimens (Paul and Paul 2001b). These molt increments are similar for those reported for the lithodids *Paralomis granulosa* (Lovrich and Vinuesa 1994) and *Lithodes santolla* (Vinuesa et al. 1990).

Juvenile golden king crabs, like the larvae, are rarely collected in nature. The primary reason is that glaucothoe apparently settle in deep water, as juveniles become more abundant with increasing depth. Pot sampling is often biased, as juveniles of many crab species hesitate entering pots where adults are present. In a survey conducted in the Aleutian Islands, small (between 40 and 100 mm CL, carapace length) golden king crabs made up a large proportion of the catch in pots set at depths \geq 730 m, while the CPUE for legal sized males (152.4 mm CL) was highest at depths $<$ 183 m (Blau et al. 1996). Most (97%) of the commercially fished pots were in depths $<$ 548 m, while the highest CPUE for juveniles was at the deepest depth (913 m) sampled (Blau et al. 1996). An aggregation of juvenile golden king crabs was observed from a manned submersible between 623 and 583 m depth on Patton Seamount in July 2002; adult golden king crab were more common in depths as shallow as 275 m (B. Stevens, NOAA, unpubl. observations; T. Shirley, unpubl. observations).

Growth of juveniles and adults

Assuming an average benthic temperature of 6°C (which is much higher than ambient temperatures), a newly settled golden king crab C1 would molt 6 times during its first year and require 866 days or 2.4 years to grow to 35 mm CL (Paul and Paul 2001b). The intermolt duration for adult female golden king crabs held in laboratory culture is about 1.5 to 2 years (Paul and Paul 2001a) while adult males tagged in the field had an intermolt duration of 10-33 months. The age of golden king crab at maturity and legal size, and maximum age, is unknown. Field tagging of 899 male golden king crabs from 1970 to 1972 and the recapture of 112 crabs from 1972 to 1976 in southeastern Alaska provided some insights into size at age (Koeneman and Buchanan 1985). The average time at large for tagged crabs was 17.5 months, with a range of 10 to 33 months for a single molt. The largest growth recorded was 60 mm in 51 months by a crab that was initially 133 mm CL. In the Aleutian Islands, one female tagged as an adult was recaptured 11 years after her release (Alaska Department of Fish and Game (ADFG), unpubl. observations). These few bits of evidence suggest that golden king crab are slow growing and require more time than red king crab to attain legal size. A lower growth rate would be expected in the colder, deeper waters they normally inhabit.

Mating

Pre-mating embraces of adult golden king crab have been observed in situ from manned submersibles. Hand-holding and coupled pairs were observed from

the DSV *Alvin* on Patton Seamount in July, 1999 (B. Stevens, NOAA, unpubl. observations). Similarly, 17 courting pairs were observed from the DSV *Delta* in May 2000, between depths of 140 and 340 m in rocky substrate in southeastern Alaska (Z. Hoyt, ADFG, unpubl. observations; T. Shirley, unpubl. observations; Hoyt et al. 2002). In several of these pairs, the female was in a subordinate, ventral to ventral position and difficult to observe as the male walked or ran along the bottom. The 17 courting pairs were from a total of 92 crabs observed, suggesting a large percentage of the adult population was involved in mating. The time span of mating activities, from May to July, suggests that courting and mating is a lengthy process, or perhaps more likely, that mating is aseasonal and occurs continuously.

Summary

Golden king crabs possess a number of attributes that make them amenable for aquaculture:

1. Adult females are aseasonal and asynchronous in reproduction, with ovigerous females being available year-round, and females hatching eggs in every month.
2. Larvae are large and easy to handle. The relatively inert larvae are demersal, remaining near the bottom of culture dishes.
3. Larvae are fully lecithotrophic, eliminating the need for feeding, decreasing cleaning efforts, and decreasing the potential for culture contamination.
4. The larval period is short, only 25 d from hatching to glaucothoe. This larval period might be decreased with more suitable culture conditions.
5. Survival of larvae is high, and larvae can easily be cultured individually.

Golden king crabs also possess a number of life history attributes that detract from their use in aquaculture:

1. The habitat is much deeper than that of red king crabs. Release of glaucothoe or early crab stages into deep waters might be problematic and monitoring their survival would be challenging. Early life history stages might not survive well in shallow waters, as they do not occur there naturally.
2. Many aspects of the early life history remain unknown in comparison to red king crabs, which have been more extensively studied. Whether or not the non-feeding larvae are agonistic and will survive well in mass culture is unknown.
3. Growth rates of juveniles and adults are slower than red king crabs
4. An unusually long period between successive clutches of ovigerous females would necessitate acquiring new brood stock.

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Basis for Stock Enhancement of *Lithodes santolla* in Argentina

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Introduction

In southern South America two lithodid crabs, *Lithodes santolla* and *Paralomis granulosa*, constitute a mixed trap fishery. In Chile and Argentina landings have been ca. 3,000 t per year, with Chile reporting 90% of total king crab landings. In Argentina the fishery for crabs started in the 1960s; landings of *L. santolla* peaked during the 1980s and decreased dramatically thereafter, from ca. 300 to 10 t per year. In 1994 the fishery for *L. santolla* was closed, and *P. granulosa*—with lower price, quality, and meat yield—began to be regularly fished. Since 2004 the population parameters of *L. santolla* have shown a recovering trend. We believe that the present population of *L. santolla* in the Beagle Channel can only sustain modest landings for the local market. We propose that a population subsidy can be one of the ways for increasing the population size, in order to obtain increased abundance and a mixed size distribution with many size/age components for harvest.

Our objective is to provide the biological basis for promoting *L. santolla* stock enhancement. The major goal is to skip the supposedly high mortality during the larval period in the natural environment by rearing huge numbers of larvae in the laboratory. This process will have as its final product the first crab stage, which could be “inoculated” in an appropriate environment, either natural or artificial. Our research has been focused on three topics: basic larval and early postlarval physiology, sampling early stages with passive collectors, and massive culture of larvae.

Methods

Lithodes santolla seems to be a good candidate for crab enhancement in the way we propose. Larvae hatch once a year during September, and this represents an important constraint for continuous cultivation. Hatching is extended in time and requires about one month for a single female to hatch the whole

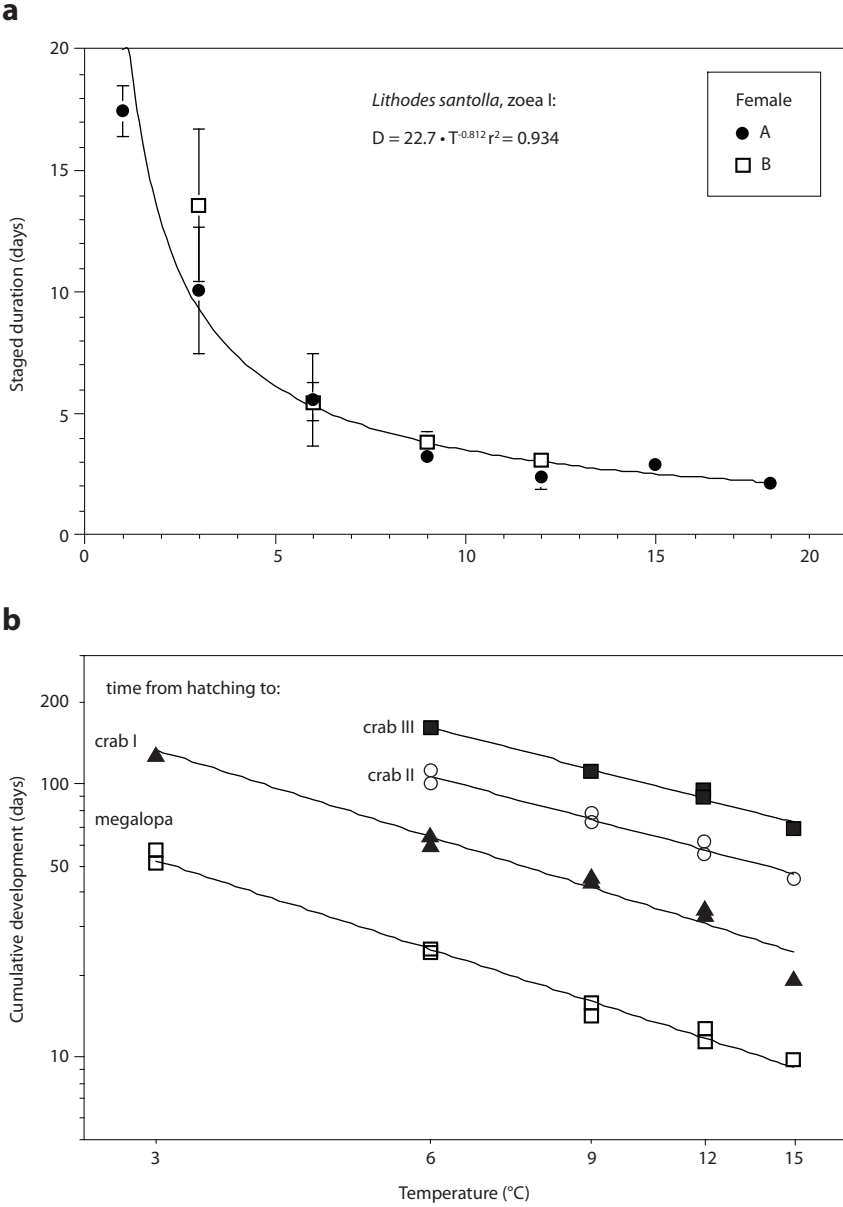


Figure 1. *Lithodes santolla*. Duration of larval and early juvenile development in relation to temperature (from Anger et al. 2004). (a) Illustration of the nonlinear relationship (power function): zoea I stage duration at 1–15°C; (b) cumulative time of development from hatching to later stages (megalopa–crab III).

egg mass (Thatje et al. 2003). Thus, obtaining huge numbers of larvae on a given date requires collecting from numerous different females.

Larval development of *Lithodes santolla* consists of 3 zoeae and 1 megalopa (glaucothoe) stages, and the whole larval span lasts 62 days at 6°C. So far, the only place where the very first juvenile stages were found is the holdfast of the kelp *Macrocystis pyrifera*. This is a three-dimensional structure that provides refuge for the recently settled animals. Densities at the holdfast are between 1 to 5 crabs per holdfast.

Larvae are lecithotrophic, i.e., during the entire larval phase they use their yolk reserves as an energy source and do not need external food for survival. Larvae have no enzymes for digesting exogenous food. The first juvenile crab stage (C1) is exotrophic, needing external food as a source of energy, and begins to have the appropriate enzymes to digest exogenous food. Larval developmental time strongly depends on temperature, and stage duration decreases exponentially with temperature (Fig. 1) (Anger et al. 2004). For example, the duration of zoea I is 17 days at 1°C but only 3 days at 15°C. The same holds true for complete larval development, which can be completed in 20 days at 15°C. Larval mortality depends on temperature and female (Fig. 2). The highest larval mortality was recorded at 3°C whereas the highest survival was at 6°C.

During the juvenile phase growth is also dependent on temperature (Fig. 3). In individual cultures for 180 days, the maximum size attained at 6°C was

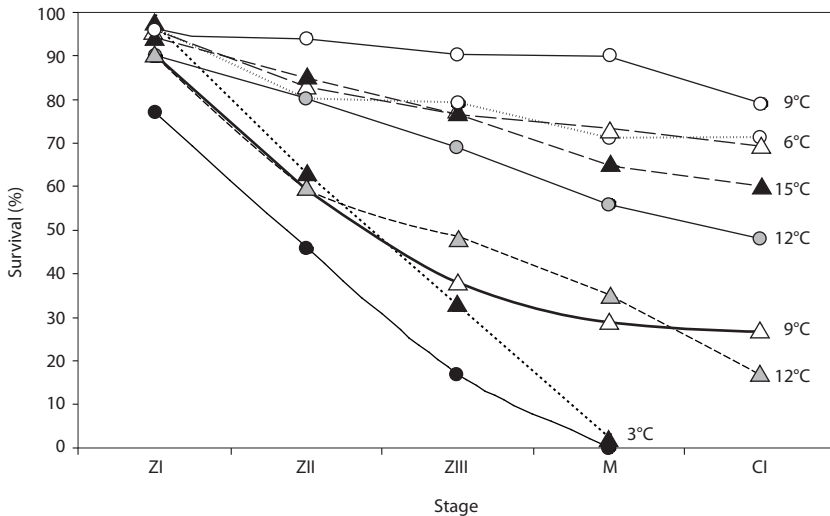


Figure 2. Cumulative survival of different larval stages from two different females (different symbols) at different temperatures (different line patterns).

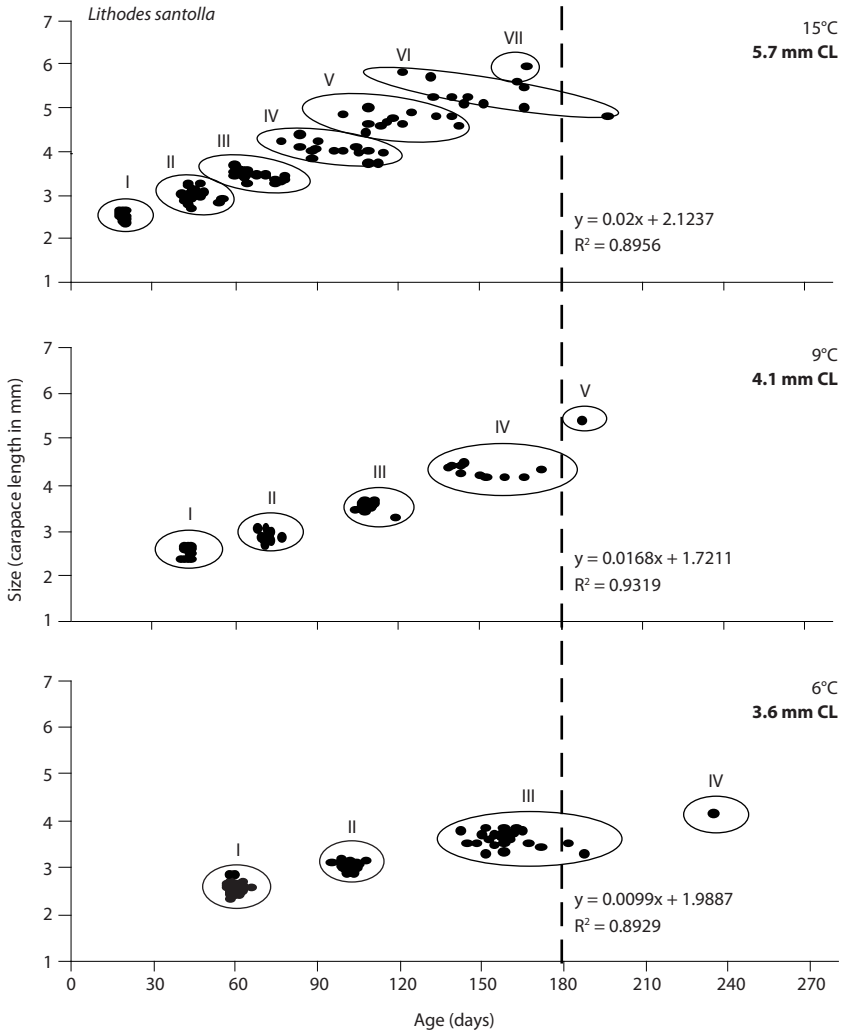


Figure 3. Size at age of different juvenile stages cultured at three different temperatures (from Calcagno et al. 2005). The vertical dotted line represents an age of 180 days and numbers on the right are the sizes (in mm CL) attained at that time.

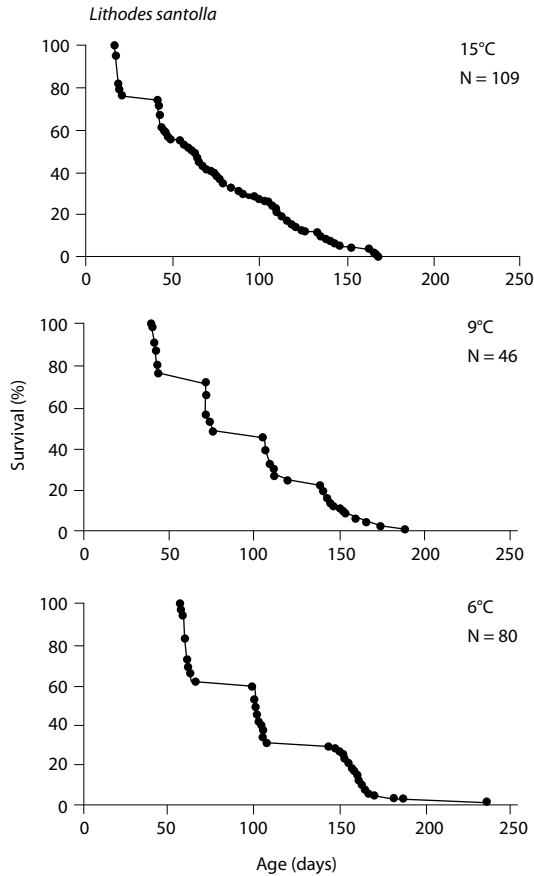


Figure 4. Survival of juvenile stages cultured at different temperatures (from Calcagno et al. 2005).

3.6 mm carapace length (CL) at stage C₃, whereas the maximum size attained at 15°C was 5.7 mm CL and stage C₇. Mortality of juvenile stages is strongly associated with the molting process (Fig. 4). As the intermolt period shortens with increasing temperature, molting frequency and mortality increases. At the natural average temperature of the Beagle Channel, i.e., 6°C, *L. santolla* attains a size of 70 mm CL (puberty molt) at about 6 years old, whereas increasing temperature would shorten this time by about half. However, more studies on growth and mortality at 15°C are needed.

With the aim of collecting recently settled crabs, we designed artificial collectors to emulate the three dimensional structure of the kelp holdfast. As

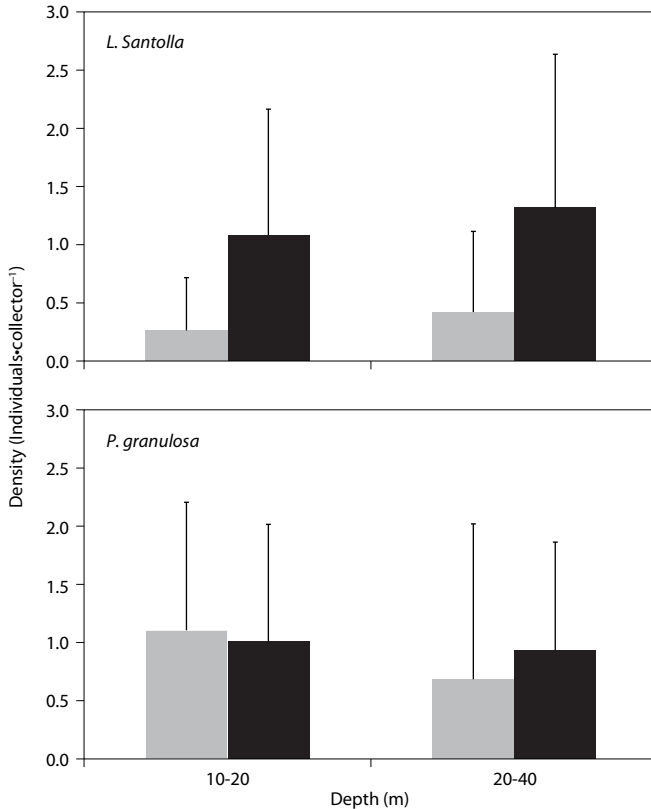


Figure 5. Average abundance of young-of-the-year *Lithodes santolla* and *Paralomis granulosa* settled in SAC and BOX (light and dark columns, respectively) deployed at two depths in the Beagle Channel. Sample sizes were 19 and 24 for SAC and BOX collectors, respectively.

a first trial we adapted the “sausaged artificial collector” (SAC) previously used in Alaska (Donaldson et al. 1991, 1992), and we built a collector that was a polypropylene bag filled with the same material. Our first objective was to test the settlement depth of crabs. We deployed lines with collectors arranged in a vertical or horizontal position. Invertebrate biomass decreased with depth, with a maximum at 10-20 m and minimum at 80-140 m. Horizontal collectors at the shallowest depth collected less invertebrate biomass than the vertical ones; we attributed this to waves that affected the former ones and washed the invertebrate fauna by scratching against the bottom. *Lithodes santolla* and *Paralomis granulosa* settled in the SACs at <40 m depth at densities between 0.5 and 1 crab per collector. We also tested another model of collector—BOX—that

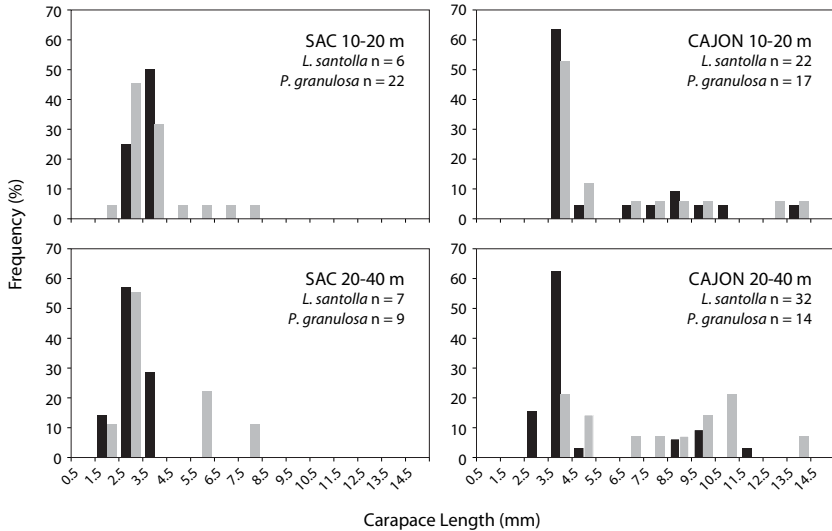


Figure 6. Size frequency distributions of young-of-the-year *Lithodes santolla* and *Paralomis granulosa* (dark and light columns, respectively) caught with SAC and BOX (=CAJON) artificial collectors deployed at two depths in the Beagle Channel. Sample sizes for each species are shown in each graphic.

consisted of an opened plastic box filled with polypropylene and rocks as ballast. At both tested depths (10-20 and 20-40 m) BOX and SAC collectors were similarly effective for capturing both *P. granulosa* and *L. santolla*, although box collectors were more efficient collecting *L. santolla* (Fig. 5). Either in BOX or SAC >60% of collected *L. santolla* were at an age less than a year, i.e., <3.0 mm CL (Fig. 6).

For massive larval rearing, we tested larval survival in two different tank designs and at three different densities. We used the traditional “plankton-kreisel” and a rectangular tank that maximizes the bottom surface. We chose the latter type of design because in individual cultures larvae remain near the bottom, rather than swimming in the water column. Controls were done by placing larvae individually in 100 ml beakers, either with massive culture water (Cult) or 20 μ m filtered seawater (F20). Larval abundance decreased in both types of tanks regardless of density, and mortality was greater in the individual cultures with the massive culture water than in individual cultures with filtered water (Fig. 7). We attribute this problem to an inefficient filtering system for our massive culture water. Regardless of the water quality, larvae in plankton-kreisel tanks survived better than in the rectangular tanks. We conclude that lack of turbulence in the rectangular tanks causes oxygen to be

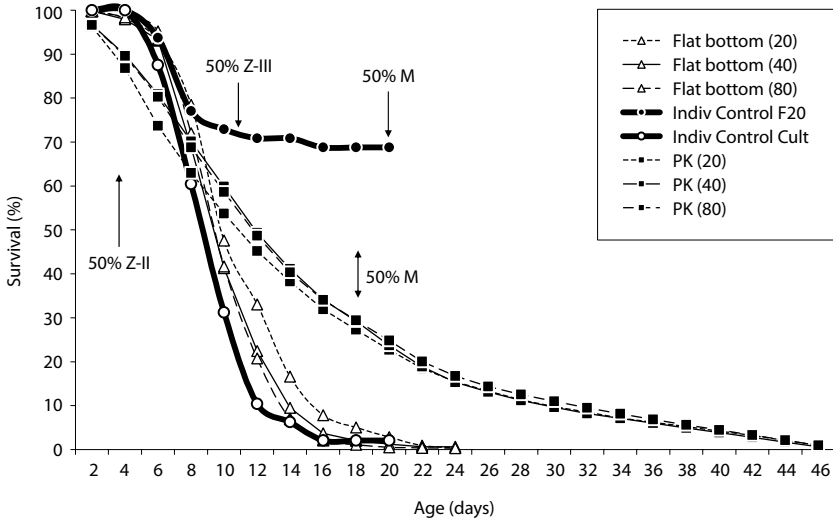


Figure 7. Survival of larvae in massive cultures in two different types of tanks: plankton-kreisel and rectangular with flat bottom, with different densities. Controls are larvae raised individually with water filtered at 20 μm (F20) or with massive culture water (Cult). Arrows indicate the 50% of molting from one to other larval stage. Z-II is zoea II, Z-III is zoea III, and M is megalopa.

unevenly distributed, causing higher mortality rates. In contrast, the mortality observed in the plankton-kreisels was mostly due to the accumulation of larvae in the tank drain.

We are optimistic that *L. santolla* populations can be subsidized with individuals produced in the laboratory. Improvements in the massive culture conditions along with finding optimum artificial collectors will allow important advances in our ability to accomplish this task.

These studies have been funded by the Argentine Agencia para la Promoción Científica y Técnica (PICT 01-10042) and by an international cooperation project funded by the International Bureau of the German Ministry of Research (BMBF, project No. ARG 99/002), and the Argentine Secretaría Nacional para la Ciencia Tecnología e Innovación Productiva (SECyT).

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Cultivation of *Lithodes santolla* in Chile: Advances in the Last Six Years Using Multiple Approaches in Puerto Montt, Chile

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Introduction

This work summarizes the main advances made in the area of developing technologies for the culture of the Southern king crab (centolla), *Lithodes santolla*, by the Laboratory of Crustacean Physiological Ecology belonging to the Instituto de Acuicultura of the Universidad Austral de Chile, located in Puerto Montt, Chile. Data presented here correspond to data collected since 1999, during the development of two consecutive grants: Fondef D99I1087 and D02I1163. The work is based on multiple approaches mainly to develop the technology needed for larval culture and the subsequent nursery phase. Finally, in the last couple of years some experiments regarding growth in the field have also been carried out.

The information available in 1999 about larval development in crabs belonging to this family (Lithodidae) reported two different nutritional strategies: planktotrophic (*Paralithodes camtschaticus* and lecithotrophic (*Lithodes maja*, *Lithodes aequispinus*). At that time, the nutritional strategy of *Lithodes santolla* larvae was still in debate as the experiments carried out were not conclusive regarding feeding type. On the other hand, *L. santolla* has a wide thermal range (3-15°C) observed from its wide geographical distribution. In addition, it was also necessary to complete a series of experiments, aiming for an optimal culture temperature.

Methods

Three different approaches were used for answering the above questions: a traditional (or basic) approach, evaluating developmental time and survival in

different feeding trials and temperature conditions; a physiological approach, specifically bioenergetics, quantifying physiological parameters such as energy balance and proximal composition at different culture temperatures; and finally a behavioral approach evaluating swimming responses to gravity and light in different larval stages.

Larvae were collected in laboratory conditions from ovigerous females collected in the field near Puerto Montt. *L. santolla* larvae ($n = 540$) were individually cultured at 9, 12, and 15°C in three different food regimes (starved, *Artemia*, *Brachionus*). Mortality and molting were checked daily during water exchange.

Results

Differences were observed in developmental time for each larval stage, and were related to temperature but not to feeding conditions. The highest survival during larval development was observed for the treatment at 12°C plus starvation (38%; $p < 0.05$). The highest mortality occurred during the zoea I and megalopa stages. The relation between percent survival and development time shows that the best results for temperature occurred at 12°C regardless of feeding conditions, and best results for diet trials occurred in starvation regardless of temperature (Fig. 1). From these results it is possible to conclude that whole larval development in *Lithodes santolla* is lecithotrophic, rejecting facultative lecithotrophy for any larval stages. Moreover, the presence of food increases mortality as a result of water quality degradation. Finally, temperature (in the range we studied) did not affect larval dependency on food.

Larval bioenergetics results showed that both ammonium-N excretion and oxygen consumption increase with larval development, from zoea I to zoea II in all temperature regimes studied. The decrease observed for both metabolic rates in the megalopa stage is believed to be the result of behavioral effects, as this stage becomes benthic late in its development. Reduced swimming activity is coupled with a decrease in metabolic rates.

There is an effect of temperature on larval physiology. When the values of these physiological rates (corresponding to energetic loss) are multiplied by its respective time of development, it is possible to obtain the accumulated losses for larval development at the studied temperatures. Accumulated energy loss at 15°C is double the loss recorded at 12 and 9°C. This approach explains the high mortality observed at 15°C which is believed to occur as a result of early exhaustion of energetic reserves caused by increased metabolic rates due to the use of high temperatures in larval culture. In addition, this can explain the northern limit for the geographical distribution of *L. santolla*. Waters located north of Puerto Montt can reach temperatures as high as 15°C which, as we know now, are inadequate for larva.

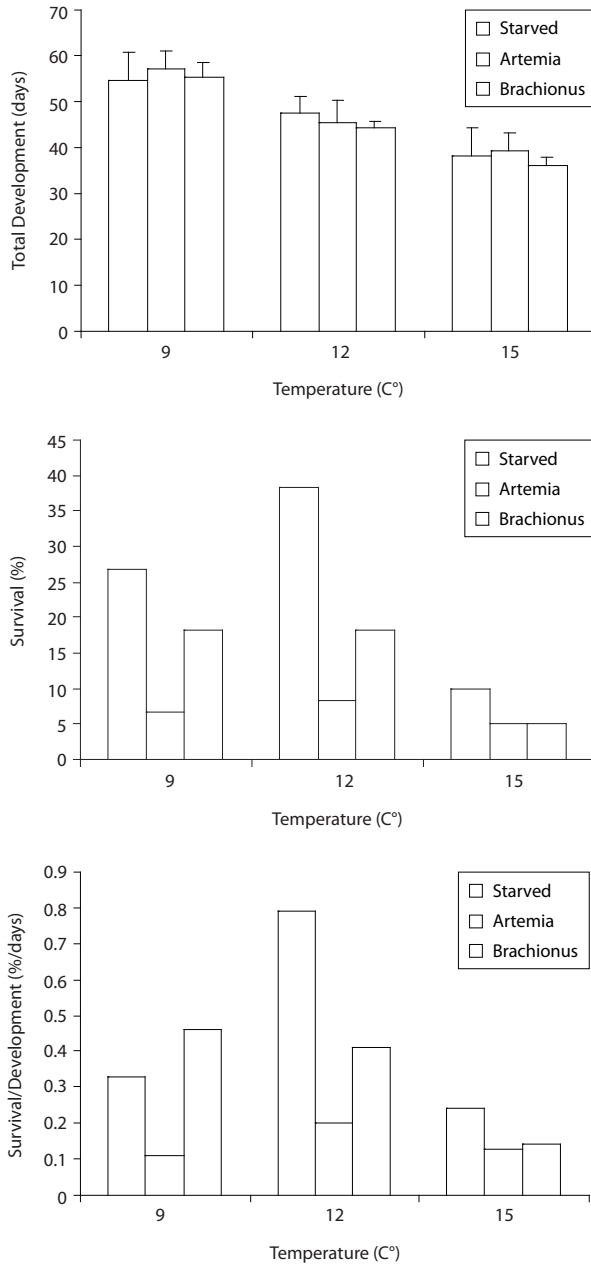


Figure 1. *Lithodes santolla*—effect of temperature and feeding regime on (a) development time, (b) mortality, and (c) development/mortality ratio, for the whole larval phase.

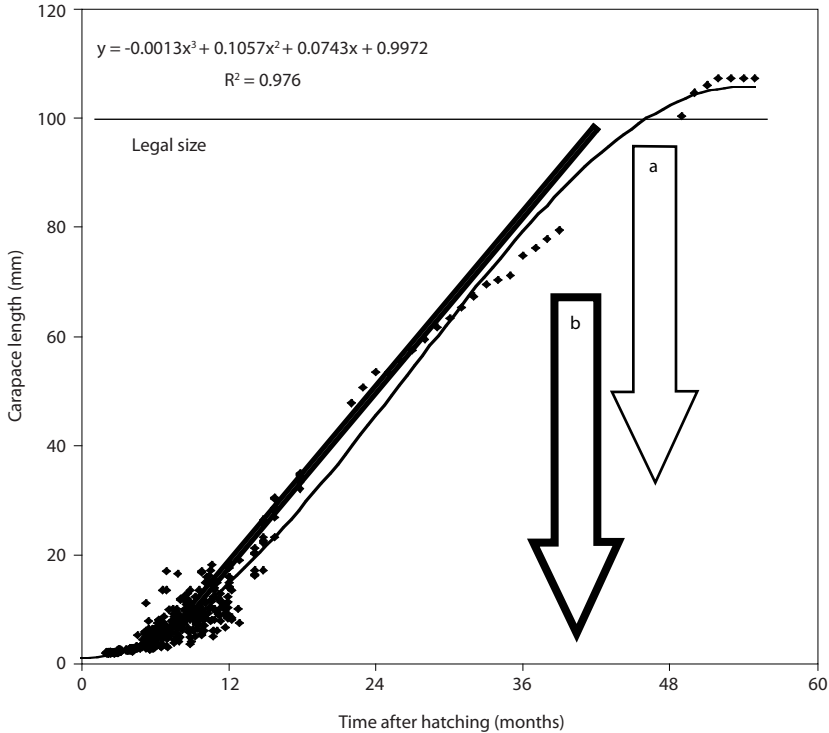


Figure 2. Composite growth curve with laboratory and sea data. Arrows indicate time required to reach legal size using (a) all data and (b) “best growers” in the laboratory.

The behavioral approach used is based on swimming experiments in a 2 m high experimental water column. Three groups of larvae ($n = 200$) from each centolla larval stage were injected in the central part of the column. Different light regimes were used on larvae: darkness, top-down illumination, and bottom-up illumination. Video recording was used to quantify larvae located in any of four different strata along the column: 10, 50, 90, and 130 cm (from top to bottom). In addition, maximum swimming speed was estimated for each larval stage. Results showed that all larval stages present an unequivocal positive geotaxis. Light (positive phototaxis) becomes a major stimulus for swimming only in the megalopa stage. Finding larvae located in the lower stratum of the column is not the result of reduced swimming ability as all larval stages showed high swimming velocity; it is rather a positive geotaxis which allows us to speculate a benthic or hyperbenthic habit for southern king crab larvae.

The finding of positive phototaxis in megalopa larva (which is also the longest stage) implies in *L. santolla* culture that larvae will make a special effort to swim toward a light source, resulting in the use of energy reserves (yolk). An experiment comparing survival rate among larvae cultured in darkness and larvae cultured with a 12:12 photoperiod showed that survival was almost twice higher for those cultured in the absence of light.

Discussion

All these findings make us think that we are not in the presence of a common decapod larva but somewhat in the presence of an “unusual egg.” Massive culture of centolla larvae with traditional methodologies did not work properly. The methodology in use by us is based on the idea of not manipulating the larvae, but culturing them in high quality waters, and discarding the dead larvae, similar to the methods used for salmon culture. Experiments carried out with massive culture of larvae at different densities and water exchange rates allow us to guarantee a survival of 30% for larval development up to the first juvenile, predicting a production of 19,000 juveniles per cultivation chamber per year considering two production seasons.

Research carried out on juveniles allowed identifying the best conditions for culturing under laboratory conditions (nursery) regarding water exchange and evaluation of artificial diets. Preliminary results showed centolla can grow well in suspended culture in the sea, having a high survival in individual culture. From growth data collected, both in laboratory and field experiments, it is possible to develop a model and predict a total production time, from hatching to harvest (100 mm CL), of 3-4 years (Fig. 2).

However, the southern king crab culture still has some unsolved problems such as identifying the factors generating the great variation in growth observed in juveniles under laboratory conditions; the variability in larval quality not only among females but also within a female; the development of technologies to start up massive culture in the sea; and the current problem of having to depend on ovigerous females collected from the field, among others. There is still some room for more research in these crustaceans.

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Lobster (*Homarus americanus*) Enhancement Project in the Southwestern Gulf of St. Lawrence, Canada

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Introduction

Lobster (*Homarus americanus*) landings in some areas of the southwestern Gulf of St. Lawrence (sGSL) have been declining after reaching historically high landings in the late 1980s to the early 1990s. In some areas, landings have declined steadily for the past fifteen years and are currently at their all-time low (Comeau et al. 2004). Fishermen in those areas have been interested in enhancement projects to complement sound conservation measures to manage the lobster population. However, efforts to enhance lobster populations for increasing harvests through the addition of hatchery-reared larvae/juvenile lobsters have been widespread worldwide and controversial (Bannister and Addison 1998). The main reason for this controversy is the lack of measurable data to show an increase in the adult lobster population that can be attributed to larvae/juvenile release programs (van der Meeren 2005). In a habitat restoration project on the East Coast of the United States, Castro et al. (2001) used microwire tags to mark 4,000 hatchery-reared stage V and VI lobsters to determine whether the addition of hatchery-raised young-of-year lobsters to the habitat would increase population densities. Using intensive sampling techniques, including a before-after-control-impact (BACI) design with scuba diver-operated airlift samplers, they only recovered one hatchery-reared lobster. Based on their field observations, they concluded that seeding with hatchery-reared lobsters produced no apparent increase in density. Nevertheless, it seems that some questions about the possibility of enhancing lobster populations remain unanswered.

In 2001, the Department of Fisheries and Oceans Canada (DFO) was approached by the Maritime Fishermen's Union (MFU) to collaborate in a

lobster enhancement program in the sGSL. Although DFO does not financially support hatchery programs, its mandate includes gathering information on coastal habitats, lobster juvenile ecology, and lobster population dynamics. Hence a collaborative project was initiated, and the *Homarus* group was formed to carry out the lobster enhancement project. The *Homarus* group is composed of members from fishermen's associations, research institutes, the private sector, and both provincial and federal governments. Their goals are to:

1. Increase lobster stocks and achieve a sustainable lobster fishery.
2. Introduce innovative and practical approaches to enhance the lobster habitat and increase landings.
3. Increase scientific knowledge of lobster biology, coastal habitat structure, and ecosystem processes.
4. Introduce educational tools to better explain ecological processes to the fishing industry.

In practical terms, the *Homarus* group wants to develop an efficient method for producing stage IV lobster larvae (high production, low cost) and then transfer that technology to interested fishermen's groups. However, it would be pointless to efficiently produce stage IV lobsters if they don't survive in their natural habitat. Therefore, the development of a sound protocol to determine if a hatchery-reared stage IV release program could actually increase or enhance recruitment, and eventually landings (the ultimate expectation from the fishing industry), is paramount. Lobster density was monitored by scuba between 2003 and 2005 according to a BACI design in the area where hatchery-reared stage IV larvae were released. The BACI sampling design consists of a series of samples taken before and after the treatment in the control and treatment experimental units. The treatment in this project was the release of hatchery-reared stage IV lobsters. In order to properly conduct a BACI experiment and statistically detect changes, replicates within several treatment and control experimental units were surveyed.

Methods

Hatchery

The hatchery used to produce juvenile haddock in Shippagan, New Brunswick, Canada, during winter and early spring has been used to rear lobster larvae between June and early September from 2002 to 2005. Larvae used in the hatchery come from berried female lobsters collected in the wild. The advantages of using wild berried females are their large abundance in nature (easy access), low cost (no extra cost related to holding a brood stock in tanks over long periods), and the development of the eggs being well synchronized with the natural conditions (females held in artificial conditions tend to desyn-

chronize their natural spawning cycle). Females are captured in late May and brought to the hatchery where they are held in large tanks with water temperature below 5°C. The cold temperature will delay the release of larvae until they are needed. A thermal shock, by placing the berried females in tanks of temperature above 10°C, will promote the release of larvae that are then transferred to rearing tanks. After the hatching, females are returned to the wild.

The American lobster has four larval stages. Stages I to III are pelagic and they are found swimming in the water column. At stage IV the animal goes through a metamorphosis with the appearance for the first time of chelipeds. It is also the first benthic stage as the pelagic larvae will settle to the bottom. Besides being the first benthic stage, stage IV was selected as the released stage because it can be produced in 9 to 14 days in a hatchery, thus lowering the production cost. It is felt that less time spent in the hatchery will not only be important to lower the cost but could increase survival in the wild, i.e., allowing stage IV to find a shelter and begin its life under natural conditions.

Major changes have occurred in the hatchery between 2002 and 2005. During that time, several techniques were tried with various successes and under that experimental setting (trial-and-error-type of experiments) over 150,000 stage IV lobsters were produced and released. The first production in 2002 was done using 250 L K-wall tanks equipped with a filtering system (closed system). The density of larvae per L was 5-9, and they were fed with live *Artemia*. The survival rate ranged between 2% and 18% for a production of 1,500 larvae. This system demanded a great deal of manipulation and was labor intensive. The rearing of *Artemia* and its cost was also a problem for the type of hatchery the Homarus group wanted to develop (low-tech, low-cost, high-production hatchery). In 2003, a comparison between 250 L K-wall tanks and 1,200 L cylindrical tanks equipped with a flow-through system (eliminating a great deal of labor-intensive daily cleaning) was done. With experimental density ranging from 3 to 20 larvae per L, the survival rate between the two types of tanks was similar (ranging between 2% and 9%). Hence, the K-wall tanks were replaced by easy to maintain larger cylindrical tanks. A total of 3,500 larvae were produced and released in 2003. It was noticed during the release, however, that stage IV lobsters swam to the surface seeking light instead of the typical cryptic behavior (seeking a shelter). It was hypothesized that stage IV lobsters were conditioned by their feeding during the day, and associated light with food. It was proposed to change their feeding from day to night to alleviate this undesirable behavior that was also observed by Castro et al. (2001). The main focus in 2004 was to test, on a small scale, alternative and less expensive food (to replace the expensive live *Artemia*), and improve the tank system. A total of eight types of aeration prototypes were tested, and one significantly reduced cannibalism and promoted a higher survival rate. Of the eleven alternative foods that were tested, dry pellets (various pellets of

Table 1. Production cost of stage IV larvae. The production cost includes labor, food, and electricity to run the hatchery system developed by the Homarus group. The 2003-2005 productions were done in an experimental setting; larval mortality was caused by studies (trial-and-error-type) carried out to compare tanks, feeds, and aeration prototypes.

Year	Total cost (\$)	Number of stage IV	\$ per larva
2003	100,000	3,500	\$28.57
2004	100,000	60,000	\$1.67
2005	100,000	90,000	\$1.11
2006*	200,000	500,000	\$0.40
2007*	200,000	1,000,000	\$0.20

*Objectives in a commercial setting.

shrimp and fish) and flaked *Artemia* were not fully consumed and accumulated in tanks causing fouling problem and larvae mortality. The best alternative to live *Artemia* (\$500 per kg) was frozen *Artemia* (\$20 per kg). With a density of 10 larvae per L, the survival rate observed in 2004 was over 20% and a total of over 60,000 larvae was produced. Finally in 2005, with three years of improvement, it was possible to produce (in an experimental setting with a density of 20 larvae per L) over 90,000 stage IV lobsters with a survival rate of over 35%. More importantly for the Homarus group, the improvement achieved through years of experiments diminished the cost per larva produced yearly from \$29 to \$1 (Table 1). Our goal for 2006 and 2007 is to change the setting from experimental to commercial with the production of at least 1,000,000 larvae by 2007. The focus of the research will now shift to study the “quality” of stage IV larvae produced and enhance (or enrich) the diet of frozen *Artemia*. Ultimately, the Homarus group wants to transfer this new technology to fishermen’s groups so they could launch their own larval production system locally.

Stage IV transfer and release

Hatchery-reared stage IV lobsters are released in the wild between July and mid-September, as soon as temperature reaches 10°C, via the pipe method. Stage IV lobsters are transferred by truck from the hatchery to a boat equipped with a tank fitted with a hose. The hose is lowered to the seafloor with the help of weights, and stage IV lobsters are then transferred to the seafloor using gravity as the boat drifts over the release area. Rocky habitats at depths of less than 10 m are preferred for the release of stage IV lobsters. The hose could be guided by scuba divers to insure that the larvae are released over the best rocky habitat. The advantage of this technique, instead of a surface release, is that stage

IV is the first benthic stage for lobsters and the pipe method allows them to be released directly on the seafloor where they can immediately seek refuge and hide instead of swimming from the surface in the water column and be vulnerable to predation before settling (Johns and Mann 1987, Wahle and Steneck 1991). An important aspect of the transfer is the continuous aeration within the transfer tanks to lower cannibalism of stage IV lobsters.

Hatchery-reared stage IV survival in nature

In both 2003 and 2004, hatchery-reared lobsters were released in July (early release) and late August (late release) in Caraquet New Brunswick, Canada. The Caraquet area was divided in three experimental units: one impact unit where hatchery-reared stage IV lobsters were released and two control units located 5 km east (with two replicates) and west (with four replicates). The release unit was divided in eight sites: two sites for each of the early and late release with their replicates (four replicates). To detect a difference in the mean abundance of the lobster population in the impact and control sites before and after the release of hatchery-reared stage IV and assess the survival in the wild of hatchery-reared stage IV animals, lobster densities were monitored according to a BACI design (Underwood 1991, 1992, 1994). The BACI was carried out in July each year using scuba transect surveys. One-hundred-meter transect lines marked every 5 m were used. A minimum of three transects were randomly placed in each site for the first “before” survey in 2003. Transects for the “after” surveys in each given year were identical to those done during the 2003 “before” survey. Two divers, one on each side, surveyed an area 2 m wide perpendicular to the transect line for the entire transect length. Therefore, 40 sections covering 10 m² each (400 m² total) were surveyed for each transect. Lobsters observed during these transect surveys were counted, measured, and sexed.

A total of 3,500 hatchery-reared stage IV lobsters were released in summer 2003 following the first “before” survey. In 2004, the “after” survey of the 2003 release took place in July. Since lobsters grow to a maximum of 18 mm of carapace length during their first year (Hudon 1987), animals smaller than that size observed in the summer of 2004 were considered as 1-year-old lobsters (assumed to be lobsters that settled the previous summer). Results showed that the increase in recruitment of 2.5-fold observed in the impact sites (release sites and replicates) was smaller than the 4- and 5-fold recruitment increase observed in the control east and control west sites respectively (Fig. 1). Furthermore, it was noted by scuba divers during the 2003 release that hatchery-reared stage IV lobsters actually swam to the surface instead of seeking a shelter on the bottom. Hence, it seems that the release of a small number of animals (3,500) coupled with a behavior that is not typical for stage IV animals did not enhance lobster recruitment. Immediately following the 2003 release, it

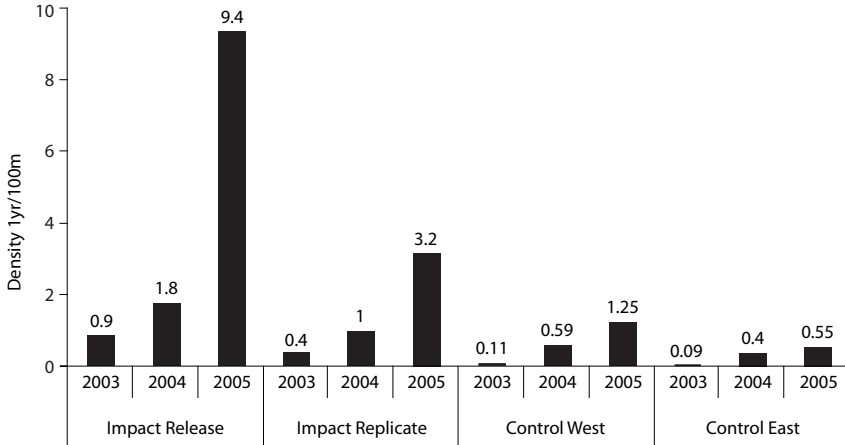


Figure 1. Density of 1-year-old lobsters (per 100 m²) captured in the impact (release sites and replicates) and control sites. The scuba surveys were carried out during the month of July 2003 (“before”), 2004 (“after” for the 2003 release, “before” for the 2004 release), and 2005 (“after” for the 2004 release).

was decided to do another release in 2004 with stage IV lobsters that were fed during the night to, hopefully, correct for the light-seeking behavior. Hence, following the “after” survey in 2004, a total of 53,000 stage IV lobsters were released in the impact area. The 2004 scuba survey was considered the “before” of the 2004 release. Results from the 2005 “after” survey revealed that recruitment (the 2004 recruitment) was higher in the impact sites compared to the control sites (Fig. 1). The density of 1-year-old lobster was higher in the release sites (5 times higher than the previous year) and the replicates (3.2 times higher) within the impact unit. Scuba divers noticed that the behavior of the hatchery-reared stage IV release in 2004 was to settle to the bottom and seek shelter, a more typical behavior for stage IV lobsters. Densities observed in the control sites increased by about twofold. The 2005 results showed that the survival in the wild of hatchery-reared stage IV lobster over the first year (through winter) seems to be good and could increase recruitment.

Summary

In summary, improvement to the rearing system between 2002 and 2005 has allowed the survival of stage IV lobsters to increase without increasing the production cost. The major improvements included (1) rearing larvae from circular 1,200 L tanks with a flow-through system instead of 250 L K-wall tanks equipped with a filtering system, (2) changing the feed from live to frozen

Artemia, and (3) improving the aeration system to reduce cannibalism. With these improvements, the survival rate increased from 10% to over 35% and the cost of production for stage IV decreased from \$28 to \$1. Another improvement to the rearing system included adjusting the feeding pattern from day to night, as it was noted during the first release that stage IV fed during the day were actively swimming to the surface and seeking light instead of settling to the bottom and seeking a shelter. Ultimately, we hope to transfer this proven low technology hatchery system that has a high production and a low operation cost to fishermen's groups.

The survival in the wild of hatchery-reared stage IV lobsters over the first year seems to be good. Using the BACI approach, the release in 2003 of 3,500 stage IV lobsters could not be statistically detected the following year suggesting that releasing a small number of stage IV lobsters does not enhance recruitment. However, the release of over 53,000 stage IV lobsters in 2004 enhanced recruitment as the 2005 survey showed that the density of 1-year-old lobsters in the release areas was significantly higher compared to the control sites. Hence, the release of a large number of hatchery-reared stage IV lobsters seems to enhance juvenile recruitment, i.e., good survival over the first winter of stage IV released the previous summer. However, before assessing the long-term success of releasing hatchery-reared stage IV lobsters more monitoring is needed to demonstrate measurable increased recruitment to the fishable population (6-7 years).

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Is It Possible to Enhance King Crab Populations in Alaska?

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Introduction

The question posed as the title of this article actually contains three critical sub-questions: Can populations of crabs be enhanced? Will it work for king crabs? And, can it be done in Alaska? Each of these must be addressed individually.

Enhancement of crab populations is being conducted in many parts of the world. Crab cultivation is thriving in the tropics, using methods ranging from low-tech family operations in ponds, to high tech industrial hatcheries. Many of these operations focus on the mud crab *Scylla serrata*, because it is a large, high value crustacean that grows rapidly in warm water (Christensen et al. 2004, Genodepa et al. 2004). In Japan, major enhancement efforts have been undertaken with the gazami or swimming crab *Portunus trituberculatus* (Secor et al. 2002). In the United States, similar work is being conducted with the Chesapeake Bay blue crab *Callinectes sapidus*. This species is an excellent candidate for stock enhancement because they live in relatively warm water, produce many larvae, have a short development period, reach 20 mm in 2 months (as opposed to 2 years for red king crab), mature at 18 months (vs. 5 years for red king crab), and may only live 4 years (A.H. Hines, Smithsonian Environmental Research Center, pers. comm.). They require a complex feeding regimen, but can be stocked at high densities, up to 150 per L in 1,000 L tanks, and survival in culture can be almost 30% to the first crab stage. Crabs released earlier in the summer grew faster and matured a year earlier than later releases, survival ranged from 6% to 25%, enhancement (increase over natural densities) ranged from 25% to 150%, and production ranged from 100 to 600 crabs per ha, over twice normal densities. Hatchery-raised crabs tended to have shorter spines and naïve behaviors, but adapted quickly if given the opportunity.

Unlike mud crabs or blue crabs, king crabs live in cold water, grow slowly, and have high cannibalism rates (Stevens and Swiney 2005). Nevertheless, Japanese scientists have been conducting research on king crab cultivation for over 70 years. The Akkeshi hatchery of the National Sea Farming Association

has been producing juvenile Hanasaki-gani since 1982, and released 500,000 crabs in 1996 (Stevens 2006). At the Kodiak Fisheries Research Center, several thousand larvae of the blue king crab (*Paralithodes platypus*) were raised in plastic containers or PVC tubes, with survival up to 92% to the first crab stage on a diet of *Artemia* and *Thalassiosira*, at a temperature of 6°C and densities up to 16 zoeae per L (Persselin 2006). In Moscow, red king crabs have been raised with up to 35% survival to the first crab stage (Kovatcheva 2006). Golden king crabs (*Lithodes aequispinus*) should be easier to cultivate because their larvae are lecithotrophic, so it is not necessary to feed them; they hatch with a large quantity of lipid, and can live over 6 months without feeding (Shirley 2006). The southern king crab, *Lithodes santolla* is also lecithotrophic (Lovrich and Tapella 2006), and can be successfully cultivated by raising larvae in the dark with upwelling, at densities up to 800 per tray (Paschke et al. 2006).

Many lessons relevant to king crab enhancement can be learned from people working with lobsters. Lobsters are very similar to king crabs, and have many of the same life history traits, including similar habitats, temperature ranges, and longevity. In New Brunswick, Canada, the *Homarus* group has produced over 90,000 stage IV (S4) lobsters with up to 35% survival. In 2006, they will produce 500,000 S4s, and by 2007 they plan to produce 1,000,000 larvae, at a cost of about 20-40 cents per lobster, and turn over the technology to industry. The effectiveness of the program ranges up to five times natural densities (Comeau 2006). However, few enhancement programs have been successful due to lack of ecological understanding. The goals of most programs are simple: to release small crabs or lobsters, allow them to settle and grow to maturity, and recapture them. But there are many pitfalls. The animals must be physiologically adapted to their new conditions. They need to be released under correct light and temperature conditions, or they will be confused or shocked, and not adapt. They need to find shelter immediately; if they go roaming they will be subject to conflict, injury, poor growth, predation, and death. They need to be released into appropriate habitat that has enough space, food, and shelter. They need opportunities to turn naïve behavior into adaptive behavior. And finally, they need to be released when and where there are few predators so they don't become eaten immediately (van der Meeren 2006).

Common to all of the presentations at this workshop were a few recurrent themes. First of all, cultivation of king crabs to the settling stage is not particularly difficult. The most difficult work is raising diatoms and other food sources for the larvae. A good source of artificial feed would be a great improvement. Survival rates to the first crab stage from 30% to 90% can be achieved, but 50% is probably a reasonable goal for large scale cultivation. After settlement, cannibalism is the biggest problem. Isolation of individual crabs is too space- and labor-intensive, but providing adequate habitat and diets can help. However, the best solution is to outplant the juveniles as soon as possible. Before doing

that, we need to know where the best habitats are, and what normal densities are. Hatchery-produced crabs should be given the chance to acquire adaptive behaviors by challenging them in the lab with natural habitats, foods, and predators. And finally, in order to determine the effectiveness of such a program, we need methods to mark the hatchery crabs in order to distinguish them from wild crabs. On a small scale this can be done with magnetic coded wires or elastomers, but it is very labor intensive. Better tools would be to identify genetic markers that can distinguish crabs by their source.

Options for king crab enhancement

Aquaculture of king crabs, in the sense of “farming or ranching” is not economically feasible at the present time, because of high cannibalism and slow growth rates. Those obstacles could perhaps be overcome with time, by selecting for individuals with more agreeable traits. However, there are varying levels of population enhancement that could be functional.

1. Transplantation. One option includes transferring females from the Bering Sea to the Gulf of Alaska. This may not be the best option for various reasons, including the potential for introduction of diseases and changes to a gene pool that may already be well adapted to Gulf of Alaska conditions. In addition, a survival bottleneck may be occurring in the first year of life, perhaps due to ocean conditions, food availability, or predator abundance that is preventing survival of small crabs. Furthermore, this option is not feasible for blue king crabs, whose populations are all depressed, and no population can supply a large number of females.
2. Augmentation of habitat. Larvae are naturally dispersed into many areas where juvenile habitat simply does not exist, and as a result, will not settle in those locations (Loher and Armstrong 2000). Normally, most of those would die and be lost from the system. A simple mechanism to overcome this would be to distribute larval settlement devices such as SACS (Donaldson et al. 1991, 1992), collect the settling crabs, and move them to better habitat. This could be done at relatively low cost by local communities. Another technique would be to enhance benthic habitat that is important for settlement. However, the occurrence of high population levels in the past for many king crab stocks suggests that habitat is not limited.
3. Stock enhancement. A third strategy is to cultivate and release early stage crabs, as is being done in Japan; that option will require research on techniques for cultivation, feeding and diet, and release timing, methods, and locations. Prior to conducting enhancement, it is important to determine whether populations have declined due to recruitment lim-

itations or some other factors (van der Meeren 2006). This may not be possible for red king crab due to the lack of historical data on recruitment of small crabs. However, historically high abundance indicates that current recruitment levels are below the environmental carrying capacity.

Where to start?

In the United States, federal (NMFS) and state (ADFG) government agencies will not take responsibility for crab enhancement because it is too expensive and benefits only a minority of users. They should be expected to conduct much of the original research, but will require support from industry to obtain the necessary budget to do it. Construction and operation of an enhancement operation will only occur with industry investment, as has been done for salmon fisheries. However, before any of this can become a reality, certain questions need to be answered.

For example, how do we define a king crab population for enhancement purposes? Are crabs from Alitak Bay genetically similar to those in Uyak or Ugak bays? If so, there should not be any problems caused by releasing juveniles from one bay into another. How will cultivated crabs perform in the wild? Will they have the same ability as wild crabs to find shelter and food, avoid predators, and survive? Finally, how can the effectiveness of stock enhancement be determined? Hatchery-produced crabs will need to be distinguished from wild crabs in order to determine their survival and eventual entry into the fishery. Tracking of hatchery crabs will require development of sophisticated techniques for tagging or identifying them based on genetic signatures.

Given our current level of technology and understanding, it will take several years of dedicated research and enhanced funding before a test program of king crab enhancement would be possible. Another three years would probably be required to bring it to the level at which it could support a commercial hatchery-enhancement operation. At least seven years of consistent operation would be required before released crabs would appear in a fishery, and success could be determined. Therefore, any economic benefit is at least 10-15 years off. However, once a project of this nature is undertaken, surprises and technological advancements may shorten that agenda.

How many crabs are needed?

To support a viable fishery, enhancement would have to produce about one million adult male crabs. The following discussion is based on certain justifiable assumptions about king crab enhancement. Although female red king crabs can produce upwards of 250,000 eggs, typical numbers of larvae released are about 150,000 per female (Stevens and Swiney, submitted for publication).

Natural mortality of crabs is poorly known, but is probably associated with molting events. Rather than estimating mortality on an annual basis, it was estimated per molt. Survival was calculated as

$$N_{n+1} = N_n \times e^{-mm}$$

where N_n is number at molt n , N_{n+1} is number at molt $n + 1$, m is mortality and n is number of molts. A two-tiered mortality schedule was used including one level for all larval molts (to the first crab stage, C1), and a second for all further juvenile to adult molts. The range of survival possibilities was calculated by varying survival of both tiers from 0.05 to 0.3 in increments of 0.05. Survival to sexual maturity (age 6) ranges from a low of 50,000 (using larval and juvenile mortality rates of 30% and 30%, respectively) to 5.8 million crabs (using rates of 15%/15%) (Fig. 1). The ratio of age-6 survivors to starting numbers of larvae (and thus, efficiency) was also calculated for varying mortality rates (Fig. 2).

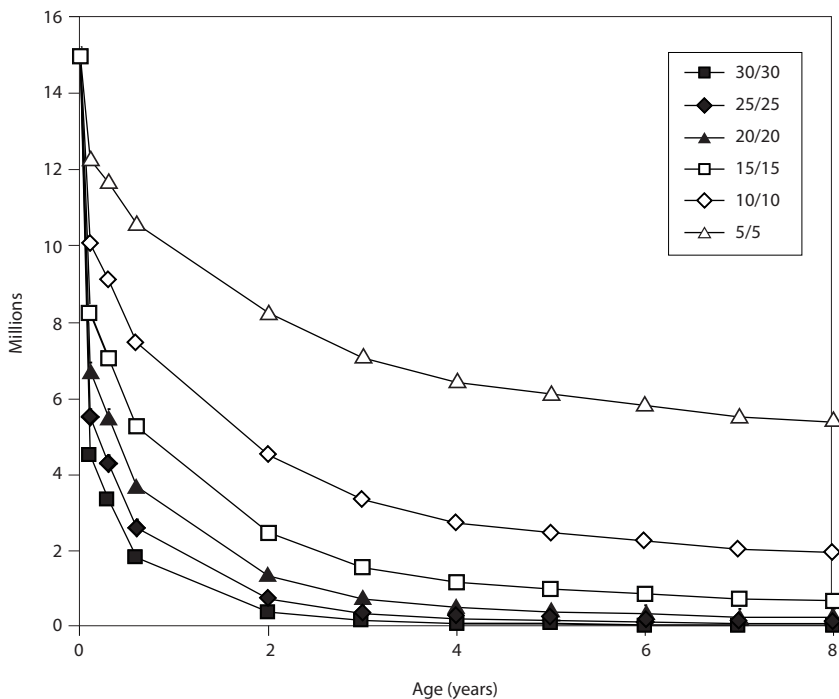


Figure 1. Millions of crab at various ages resulting from a two-tiered survival schedule. A range of combinations from 5% to 30% were used for each tier. For convenience, only those combinations are shown where larval and juvenile survival are equivalent.

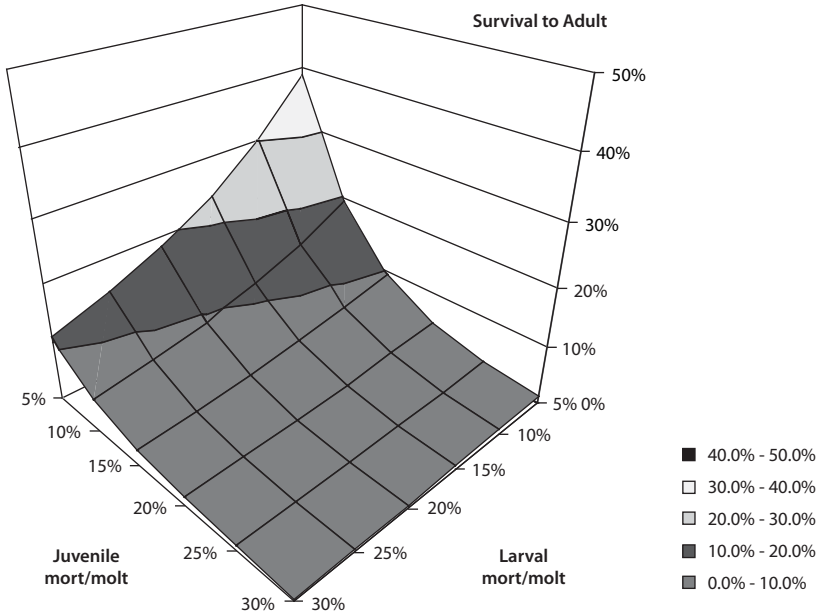


Figure 2. Survival ratio for cultivated and released king crabs using mortality rates from 5% to 30% for larval and juvenile molts.

Survival rates >0.1 (10%) can be considered “good,” but are only obtained when mortality is <0.15 for one or both tiers. Therefore, to successfully raise crabs to the point of release, an enhancement program needs to produce stage C1 crabs with a mortality rate <0.15 per molt (approximately 47% total, close to the target of 50%). While mortality during the larval phase is controllable in an enhancement operation, mortality after release is not. However, careful selection of a release technique, timing, and location can influence survival for the following one or two molts.

For this exercise, assume a starting brood stock of 100 females that release 150,000 larvae each, or a total of 15 million larvae. Using mortality rates of 0.15 for both tiers (15/15) as a benchmark, these larvae would produce 7 million stage C1 crabs, which become 693,000 age-8 crabs, of which half (346,422) are male, averaging 6.5 lbs each (Table 1). Using values of \$6 per pound and an exploitation rate of 15%, respectively, they could be worth \$2.025 million.

But what will be the cost of producing those crabs? A hatchery large enough for this undertaking will require an initial investment of at least \$5 million. A professional staff of five people (manager, culturist, engineer, and two technicians) will require about \$450,000 per year, and building maintenance,

Table 1. Life table for red king crab cultivation. Results assume that 100 females each produce 150,000 larvae; larval and juvenile mortality equals 0.15 per molt; 50% of surviving age-8 adults are males with an average weight of 6.5 lbs, and exploitation ratio is 15%. Other scenarios with varying mortality, value, and exploitation rate are discussed in text.

No. female brood stock			100
Larval mortality/molt			15%
Juvenile mortality/molt			15%
Larvae per female			150,000
	Age	Molts	Million crabs
Hatch	0	0	15,000
Larval stage	0.1	4	8.232
Glaucothoe stage	0.3	1	7.085
Outplant mortality	0.6	2	5.249
Mortality Y1	2	5	2.479
Mortality Y2	3	3	1.581
Mortality Y3	4	2	1.171
Mortality Y4	5	1	1.008
Mortality Y5	6	1	0.868
Mortality Y6	7	1	0.747
Mortality Y7	8	0.5	0.693
Survival to C1			47.3%
Survival to adult			5.78%
Results of cultivation from above			
Results		Rates	Totals
Total males		50%	346,422
Total lbs		6.5 each	2,251,744
GHL crabs		0.15	51,963
GHL lbs		0.15	337,762
Total value		\$6	\$2,026,570
Cost/benefit			3.35

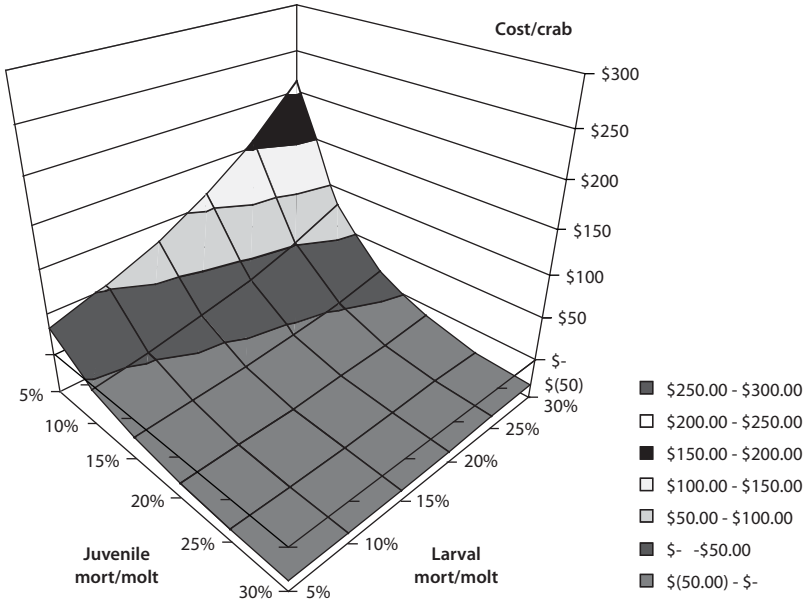


Figure 3. Cost per crab captured (after subtracting ex-vessel value of crabs) for cultivated and released king crabs using mortality rates from 5% to 30% for larval and juvenile molts. Negative values imply a positive benefit.

supplies, and debt retirement will cost another \$150,000 per year, making the annual cost (after construction) about \$605,000 (Table 2). This is the lowest cost probable. At this level of production, the cost for each crab released ranges from \$0.05 to \$0.18 each. The cost for each crab captured varies from a low of \$1.50 to a high of \$252, but after subtracting the ex-vessel value of \$39 per crab these costs become \$-37.50 to \$213.07 (Fig. 3); negative values indicate that crab enhancement is generating more value than it costs. The value of a fishery on these crabs would be correlated directly with both the ex-vessel value and the guideline harvest level (GHL) or exploitation rate. Using the 15/15 benchmark, values of \$6 to \$10 per lb, and exploitation rates from 0.1 to 0.25, the potential benefit/cost ratio for adult crabs captured by the fishery ranges from 2.2 to 9.3 (Table 3). However, if 200 female crabs were used (and 30 million larvae hatched), the benefit/cost ratios would double.

Conclusion

That brings us back to the original question: Is it possible to enhance king crab stocks in Alaska? The answer is Yes, maybe. With an initial investment of \$5-

Table 2. Projected start-up and annual costs for a king crab hatchery.

Facility cost	\$5,000,000
Annual cost	
Director	\$150,000
Engineer	\$112,500
Culturist	\$112,500
Technicians	\$75,000
Total salaries	\$450,000
Building	\$50,000
Mortgage	\$50,000
Taxes	\$5,000
Supplies, equipment	\$50,000
Total annual cost	\$605,000

Table 3. Projected cost/benefit ratios based on expected costs of hatchery production (Table 1), mortality rates of 0.15 for both larval and juvenile molts (Table 2), and a range of ex-vessel values and fishery exploitation rates.

\$/lb	Exploitation rates			
	0.1	0.15	0.2	0.25
\$6.00	2.23	3.35	4.47	5.58
\$7.00	2.61	3.91	5.21	6.51
\$8.00	2.98	4.47	5.96	7.44
\$9.00	3.35	5.02	6.70	8.37
\$10.00	3.72	5.58	7.44	9.30

10 million and good luck, it might turn a positive benefit/cost ratio after 10-12 years. If you were a banker and your goals were to make a reasonable profit, that might be a questionable investment. But if your goals were

- To maintain fishing opportunities;
- To provide employment for fishermen and their children;
- To replenish a depleted stock;
- To maintain socioeconomic structures of coastal communities;
- To produce a high value, high quality seafood product;

Then it might be a beneficial thing to do.

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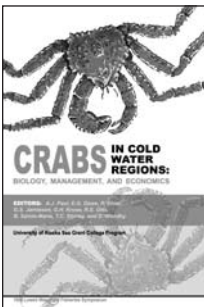


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