

Delta Smelt Culture and Research Program

Project Information

1. **Proposal Title:**

Delta Smelt Culture and Research Program

2. **Proposal applicants:**

Serge Doroshov, University of California, Davis

3. **Corresponding Contact Person:**

Ahmad Hakim-Elahi
University of California at Davis
Office of the Vice Chancellor for Research, Sponsored Programs 118 Everson Hall University of
California One Shields Ave. Davis, CA 95616
530 752-2075
vcresearch@ucdavis.edu

4. **Project Keywords:**

At-risk species, fish
Fish, Nongame
Hatchery Management

5. **Type of project:**

Research

6. **Does the project involve land acquisition, either in fee or through a conservation easement?**

No

7. **Topic Area:**

At-Risk Species Assessments

8. **Type of applicant:**

University

9. **Location - GIS coordinates:**

Latitude: 37.8271

Longitude: -121.5954

Datum:

Describe project location using information such as water bodies, river miles, road intersections, landmarks, and size in acres.

The project is on the State Water Project site, Fish grow-out facility adjacent to Clifton Court Forebay in the south Delta. Nearest town is Byron.

10. Location - Ecozone:

1.1 North Delta, 1.2 East Delta, 1.3 South Delta, 1.4 Central and West Delta

11. Location - County:

Contra Costa

12. Location - City:

Does your project fall within a city jurisdiction?

No

13. Location - Tribal Lands:

Does your project fall on or adjacent to tribal lands?

No

14. Location - Congressional District:

10th

15. Location:

California State Senate District Number: 7

California Assembly District Number: 15

16. How many years of funding are you requesting?

2

17. Requested Funds:

a) Are your overhead rates different depending on whether funds are state or federal?

Yes

If yes, list the different overhead rates and total requested funds:

State Overhead Rate: 10

Total State Funds: \$601,918

Federal Overhead Rate: 26

Total Federal Funds: \$685,832

b) Do you have cost share partners already identified?

Yes

If yes, list partners and amount contributed by each:

California Department of Water Resources 5,000

c) Do you have potential cost share partners?

No

d) Are you specifically seeking non-federal cost share funds through this solicitation?

Yes

If yes, list total non-federal funds requested:

5,000

If the total non-federal cost share funds requested above does not match the total state funds requested in 17a, please explain the difference:

18. **Is this proposal for next-phase funding of an ongoing project funded by CALFED?**

No

Have you previously received funding from CALFED for other projects not listed above?

Yes

If yes, identify project number(s), title(s) and CALFED program.

00FC200113 Delta Smelt Culture, Phase II and III ERP

B-81581 Delta Smelt Culture, Phase I ERP

B-103 Assessing Impacts of Selenium on Restoration of the San Francisco Bay-Delta Ecosystem ERP

98-C15 Biological Assessment of Green Sturgeon in the Sacramento-San Joaquin Watershed (Phase I) ERP

00FC200142 Biological Assessment of Green Sturgeon in the Sacramento-San Joaquin Watershed (Phase II) ERP

19. **Is this proposal for next-phase funding of an ongoing project funded by CVPIA?**

No

Have you previously received funding from CVPIA for other projects not listed above?

No

20. **Is this proposal for next-phase funding of an ongoing project funded by an entity other than CALFED or CVPIA?**

No

Please list suggested reviewers for your proposal. (optional)

Randy Baxter Cal Fish and Game 209-942-6081

Mike Chotkowski US. Bur. Reclamation 916-9789524

21. **Comments:**

Environmental Compliance Checklist

Delta Smelt Culture and Research Program

1. CEQA or NEPA Compliance

a) Will this project require compliance with CEQA?

No

b) Will this project require compliance with NEPA?

No

c) If neither CEQA or NEPA compliance is required, please explain why compliance is not required for the actions in this proposal.

Not required at this time. If project is approved and federally funded then CEQA or NEPA compliance will be completed.

2. **If the project will require CEQA and/or NEPA compliance, identify the lead agency(ies). If not applicable, put "None".**

CEQA Lead Agency: None

NEPA Lead Agency (or co-lead:) None

NEPA Co-Lead Agency (if applicable): None

3. **Please check which type of CEQA/NEPA documentation is anticipated.**

CEQA

Categorical Exemption

-Negative Declaration or Mitigated Negative Declaration

-EIR

-none

NEPA

Categorical Exclusion

-Environmental Assessment/FONSI

-EIS

-none

If you anticipate relying on either the Categorical Exemption or Categorical Exclusion for this project, please specifically identify the exemption and/or exclusion that you believe covers this project.

4. **CEQA/NEPA Process**

a) Is the CEQA/NEPA process complete?

Not Applicable

b) If the CEQA/NEPA document has been completed, please list document name(s):

5. **Environmental Permitting and Approvals** (*If a permit is not required, leave both Required? and Obtained? check boxes blank.*)

LOCAL PERMITS AND APPROVALS

Conditional use permit

Variance

Subdivision Map Act

Grading Permit

General Plan Amendment

Specific Plan Approval

Rezone

Williamson Act Contract Cancellation

Other

STATE PERMITS AND APPROVALS

Scientific Collecting Permit Obtained

CESA Compliance: 2081

CESA Compliance: NCCP

1601/03

CWA 401 certification

Coastal Development Permit

Reclamation Board Approval

Notification of DPC or BCDC

Other

FEDERAL PERMITS AND APPROVALS

ESA Compliance Section 7 Consultation

ESA Compliance Section 10 Permit

Rivers and Harbors Act

CWA 404

Other

PERMISSION TO ACCESS PROPERTY

Permission to access city, county or other local agency land.

Agency Name:

Permission to access state land.

Agency Name:

Permission to access federal land.

Agency Name:

Permission to access private land.

Landowner Name:

6. Comments.

Land Use Checklist

Delta Smelt Culture and Research Program

1. **Does the project involve land acquisition, either in fee or through a conservation easement?**

No

2. **Will the applicant require access across public or private property that the applicant does not own to accomplish the activities in the proposal?**

No

3. **Do the actions in the proposal involve physical changes in the land use?**

No

If you answered no to #3, explain what type of actions are involved in the proposal (i.e., research only, planning only).

Research only.

4. **Comments.**

Conflict of Interest Checklist

Delta Smelt Culture and Research Program

Please list below the full names and organizations of all individuals in the following categories:

- Applicants listed in the proposal who wrote the proposal, will be performing the tasks listed in the proposal or who will benefit financially if the proposal is funded.
- Subcontractors listed in the proposal who will perform some tasks listed in the proposal and will benefit financially if the proposal is funded.
- Individuals not listed in the proposal who helped with proposal development, for example by reviewing drafts, or by providing critical suggestions or ideas contained within the proposal.

The information provided on this form will be used to select appropriate and unbiased reviewers for your proposal.

Applicant(s):

Serge Doroshov, University of California, Davis

Subcontractor(s):

Are specific subcontractors identified in this proposal? No

Helped with proposal development:

Are there persons who helped with proposal development?

Yes

If yes, please list the name(s) and organization(s):

Joel Van Eenennaam UC Davis

Comments:

None

Budget Summary

Delta Smelt Culture and Research Program

Please provide a detailed budget for each year of requested funds, indicating on the form whether the indirect costs are based on the Federal overhead rate, State overhead rate, or are independent of fund source.

State Funds

Year 1												
Task No.	Task Description	Direct Labor Hours	Salary (per year)	Benefits (per year)	Travel	Supplies & Expendables	Services or Consultants	Equipment	Other Direct Costs	Total Direct Costs	Indirect Costs	Total Cost
1	Delta Smelt Culture	6832	135018	26855	2400	12000	0	16000	0	192273.0	17627	209900.00
2	Larval Nutrition	936	16032	3204	600	4630	0	0	0	24466.0	2447	26913.00
3	Spawning Behavior	984	16032	3204	600	3570	0	0	0	23406.0	2341	25747.00
4	Project Management	1080	24516	4908	2400	1800	0	0	0	33624.0	3362	36986.00
		9832	191598.00	38171.00	6000.00	22000.00	0.00	16000.00	0.00	273769.00	25777.00	299546.00

Year 2												
Task No.	Task Description	Direct Labor Hours	Salary (per year)	Benefits (per year)	Travel	Supplies & Expendables	Services or Consultants	Equipment	Other Direct Costs	Total Direct Costs	Indirect Costs	Total Cost
1	Delta Smelt Culture	6832	139835	27862	2400	12000	0	9000	0	191097.0	18210	209307.00
2	Larval Nutrition	936	16032	3204	600	3600	0	0	0	23436.0	2344	25780.00
3	Spawning Behavior	984	17244	3444	600	4600	0	0	0	25888.0	2589	28477.00
4	Project Management	1080	25896	5184	2400	1800	0	0	0	35280.0	3528	38808.00
		9832	199007.00	39694.00	6000.00	22000.00	0.00	9000.00	0.00	275701.00	26671.00	302372.00

Year 3												
Task No.	Task Description	Direct Labor Hours	Salary (per year)	Benefits (per year)	Travel	Supplies & Expendables	Services or Consultants	Equipment	Other Direct Costs	Total Direct Costs	Indirect Costs	Total Cost
		0	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00

Grand Total=601918.00

Comments.

Budget Justification

Delta Smelt Culture and Research Program

Direct Labor Hours. Provide estimated hours proposed for each individual.

Two Assistant Researchers (one @ 164 hr/mo and one @ 82 hr/mo) Two full time Lab Assistants IV, s@ 164 hr/mo One post graduate researcher @ 82 hr/mo Three or four hourly Lab Assistant I's total of 150 hr/mo Year Two will have the same commitments.

Salary. Provide estimated rate of compensation proposed for each individual.

Assistant Researchers (full time @ \$ 4,292/mo) Lab Assistant IV's @ \$ 3,030/mo. Post Graduate Researcher @ \$ 1,576/mo. Lab Assistant I's @ \$ 11.45/hr Year Two will include a 4% salary increase for all positions.

Benefits. Provide the overall benefit rate applicable to each category of employee proposed in the project.

Academic positions (Assistant Researchers, Post Graduate Researcher @ 20%) Staff positions (Lab Assistant IV's full time @ 25% and hourly Lab Assistants @ 2%)

Travel. Provide purpose and estimate costs for all non-local travel.

During both years, two Assitant Researchers to attend two national conferences to present results @ \$ 1,200 each person, each conference = \$ 4,800. Remaining \$ 1,200 for field sampling/broodstock collection, local meetings: UCD, Stockton, Sacramento, Pacific Grove

Supplies & Expendables. Indicate separately the amounts proposed for office, laboratory, computing, and field supplies.

Laboratory = \$20,000 [fish feed \$1,000; water quality analyses \$2,500; lab chemicals \$ glassware \$1,500; misc lab supplies \$1,500; tank culture supplies \$3,000; hardware supplies \$2,000; rotifer and algae supplies \$2,000; site maintenance \$2,500; PVC pipe and fittings \$1,000; fatty acid analyses \$2,000. Office supplies \$500. Computing \$500. Field supplies \$1,000.

Services or Consultants. Identify the specific tasks for which these services would be used. Estimate amount of time required and the hourly or daily rate.

No services or consultants are anticipated at this time.

Equipment. Identify non-expendable personal property having a useful life of more than one (1) year and an acquisition cost of more than \$5,000 per unit. If fabrication of equipment is proposed, list parts and materials required for each, and show costs separately from the other items.

Year One: Heat pump replacement \$10,000; Current meter \$4,000; Custom net \$2,000. Year Two: Heat pump replacement \$4,000; Storage container \$2,500; Fish tanks \$2,500

Project Management. Describe the specific costs associated with insuring accomplishment of a specific project, such as inspection of work in progress, validation of costs, report preparation, giving presentatons, reponse to project specific questions and necessary costs directly associated with specific project oversight.

Because of the off-campus location this project requires a larger amount of labor-hours to manage, compared to on-campus projects (an estimated 90 hours per month). The two Assistant Researchers, living near the site, and the post graduate researcher(PGR, UCD)will be responsible for project management. Travel costs include round-trips to UCD, Sacramento, Stockton, and other in-state cities for report preparation, presentations, human resource issues, inspections of labs and work in progress. Supplies include costs for preparing and dissemination of reports and presentations.

Other Direct Costs. Provide any other direct costs not already covered.

No other direct costs are anticipated.

Indirect Costs. Explain what is encompassed in the overhead rate (indirect costs). Overhead should include costs associated with general office requirements such as rent, phones, furniture, general office staff, etc., generally distributed by a predetermined percentage (or surcharge) of specific costs.

Indirect costs are determined by the UCD Office of Research and projected costs for state funds during the time of this project is 10%. Projected off-campus indirect costs for federal funds is 26%.

Executive Summary

Delta Smelt Culture and Research Program

Project Description, Location, and Project type The on-going Delta Smelt Culture Project is a successful research and development program currently funded by the CALFED Bay-Delta Program. The culture facility is located in the south Delta on State Water Project land near Byron, CA. Advances in culture methodologies allowed production of nearly 10,000 larval-to-juvenile-age delta smelt this year. Numerous research programs requested the smelt as they became available. The cultured animals also serve as a refuge against extinction of this threatened fish native to the Delta. Through careful observation and experimentation production capabilities increase and information on the smelt basic biology and life history is obtained. This information improves predictability of smelt performance in culture and may aid in predicting smelt performance in the field and in restoration projects. In the proposed work we will investigate larval fish nutrition and spawning behavior, while continuing to expand culture production capabilities. **Goals and objectives:** This project aims to deliver a reliable and enhanced supply of cultured delta smelt to numerous research programs, while improving culture protocols through our own investigative work. **Research efforts are directed in two areas which currently restrict production: larval nutrition and spawning. Approach to implement the program:** The culture of delta smelt requires the annual capture of sub-adults and daily maintenance of the progressive life stages (broodfish, eggs, larvae, juveniles and subadults) for 12 months of the year (Task1). Effect of varying the (n-3) fatty acid levels (eicosapentaenoic and docosahexaenoic) in larval food will be evaluated in terms of growth, survival, and stress tolerance of larval smelt (Task 2). Effect of spawning-substrate type on spawning behavior will be measured in terms of total number of eggs deposited on each substrate type in large outdoor tanks (Task 3). The remote field location of the site requires increased efforts for project management and communications, as well as for the planning and distribution of live animals (Task 4). **Working hypotheses and expected outcomes for current proposal:** (1) Delta smelt culture production will increase with continued research and development, doubling by 2004. (2) Delta smelt larvae require eicosapentaenoic and docosahexaenoic fatty acids in diets, and the enrichment will increase larval survival, growth and stress tolerance. (3) Delta smelt spawns are heterogeneous with respect to substrate anticipate better understanding of factors influencing spawn deposition. **Relationship of Proposed Program to CALFED ERP Goals and Priorities** The CALFED Ecosystem Restoration Program (ERP) goals are met in the creation of a supply of all life stages of delta smelt for research programs. The proposed work addresses 3 of the 8 priorities of the current CALFED ERP and PSP for the Delta and Eastside Tributaries Region: (1) Develops a better understanding of the life history of the smelt. For example the proposed spawning study may offer life history information applicable to habitat restoration concerns of at-risk species (Delta Region Priority-4; Goal 1, at-risk species; Goal 4, habitats); (2) Provides a unique stock of this native fish with known rearing history - required for delta contaminant studies (Delta Region Priority-6; Goal 6, water and sediment quality); (3) Provides a supply of smelt to test methods for reducing the impact of water diversions in the Delta, ie. to test new fish screen designs, (Delta Region Priority-7; Goal at-risk species).

Proposal

University of California, Davis

Delta Smelt Culture and Research Program

Serge Doroshov, University of California, Davis

DELTA SMELT, *Hypomesus transpacificus*, CULTURE AND RESEARCH PROGRAM

Serge Doroshov, Ph.D. (Principal Investigator)

Dept. of Animal Science, UC–Davis, Davis CA 95616, 530-752-7603 (voice), 530-752-0175 (fax),
sidoroshov@ucdavis.edu

Joan Lindberg, Ph.D.

Dept. of Animal Science, UC–Davis, Davis CA 95616, 925-443-2448, lindberg@jps.net

Bradd Baskerville-Bridges, Ph.D.

Dept. of Animal Science, UC–Davis, Davis CA 95616, 209-839-0752,
Braddbridges@mindspring.com

A PROPOSAL TO THE CALFED BAY–DELTA PROGRAM

September 2001 Proposal Solicitation Package

Financial Contact

Ann Day, Dept. of Animal Science, UC–Davis, Davis CA 95616, 530-752-4512(voice), 916-752-0175 (fax), jaday@ucdavis.edu.

Participants

California Department of Water Resources

September 2001

EXECUTIVE SUMMARY

Project Description, Location, and Project type

The on-going Delta Smelt Culture Project is a successful research and development program currently funded by the CALFED Bay-Delta Program. The culture facility is located in the south Delta on State Water Project land near Byron, CA. Advances in culture methodologies allowed production of nearly 10,000 larval-to-juvenile-age delta smelt this year. Numerous research programs requested the smelt as they became available. The cultured animals also serve as a refuge against extinction of this *threatened* fish native to the Delta. Through careful observation and experimentation, production capabilities increase and information on the smelt's basic biology and life history is obtained. This information improves predictability of smelt performance in culture and may aid in predicting smelt performance in the field and in restoration projects. In the proposed work we will investigate larval fish nutrition and spawning behavior, while continuing to expand culture production capabilities.

Goals and objectives:

This project aims to deliver a reliable and enhanced supply of cultured delta smelt to numerous research programs, while improving culture protocols through our own investigative work. Research efforts are directed in two areas that currently restrict production: larval nutrition and spawning.

Approach to implement the program:

The culture of delta smelt requires the annual capture of sub-adults and daily maintenance of the progressive life stages (broodfish, eggs, larvae, juveniles and subadults) for 12 months of the year (Task 1). Effect of varying the (n-3) fatty acid levels (eicosapentaenoic and docosahexaenoic) in larval food will be evaluated in terms of growth, survival, and stress tolerance of larval smelt (Task 2). Effect of spawning-substrate type on spawning behavior will be measured in terms of total number of eggs deposited on each substrate type in large outdoor tanks (Task 3). The remote field location of the site requires increased efforts for project management and communications, as well as for the planning and distribution of live animals (Task 4).

Working hypotheses and expected outcomes for current proposal: (1) Delta smelt culture production will increase with continued research and development, doubling by 2004. (2) Delta smelt larvae require eicosapentaenoic and docosahexaenoic fatty acids in diets, and the enrichment will increase larval survival, growth and stress tolerance. (3) Delta smelt spawns are heterogeneous with respect to substrate – anticipate better understanding of factors influencing spawn deposition.

Relationship of Proposed Program to CALFED ERP Goals and Priorities

The CALFED Ecosystem Restoration Program (ERP) goals are met in the creation of a supply of all life stages of delta smelt for research programs. The proposed work addresses 3 of the 8 priorities of the current CALFED ERP and PSP for the Delta and Eastside Tributaries Region:

(1) Develops a better understanding of the life history of the smelt. For example the proposed spawning study may offer life history information applicable to habitat restoration concerns of at-risk species (Delta Region Priority-4; Goal 1, at-risk species; Goal 4, habitats); (2) Provides a unique stock of this native fish with known rearing history - required for delta contaminant studies (Delta Region Priority-6; Goal 6, water and sediment quality); (3) Provides a supply of smelt to test methods for reducing the impact of water diversions in the Delta, i.e. to test new fish screen designs, (Delta Region Priority-7; Goal at-risk species).

A. PROJECT DESCRIPTION: PROJECT GOALS AND SCOPE OF WORK

A. 1. Description of the Problem to be Addressed

Decline in delta smelt abundance in the 1980's and lack of recovery in the '90s resulted in listing of the smelt as *threatened*. Low abundance and threatened status limits the number of delta smelt available for restoration-oriented research. The CALFED Bay-Delta Program and other agencies consider the recovery of this and other at-risk species a priority to ecosystem restoration. Restoration goals are often hampered by lack of information on the life history of at-risk species. The Delta Smelt Culture and Research Program creates a refuge population of delta smelt and a supply of smelt for research. In the proposed work we will refine and improve culture techniques to increase production of smelt for research, and initiate studies on larval nutrition and spawning behavior which will improve the culture system and provide new information on biology and life history of delta smelt.

Objectives of the proposed Delta Smelt Culture and Research Program, for a period November 2002 – October 2004, are:

- Improve efficiency of delta smelt culture system for reliable supply of all life stages for research and creating refuge population;
- Determine effects of larval food enrichment with (n-3) fatty acids (eicosapentaenoic and docosahexaenoic) on growth, survival, and stress tolerance of smelt larvae;
- Determine effects of different spawning substrates on smelt spawn deposition in large outdoor tanks.

Review of Past Work. The delta smelt culture program is an ongoing project supported by state and federal agencies (DWR, CALFED and IEP) over the last several years. This support enabled the program to develop successful methods for culturing all life stages of delta smelt. Delta smelt bloodstocks' fragility and, especially, its prolonged larval phase have limited the production of juveniles for a number of years. However, culture system and rearing to juvenile stage have markedly improved in the recent years due to improvements in broodstock management, disease prevention, and larval rearing techniques (Swanson et al., 1996; Antonio et al., 2000; Baskerville-Bridges et al., 2001). Continued support will enable the culture program to meet the increasing demands for delta smelt eggs, larvae, juveniles, and adults needed for restoration-oriented research. The culture method derives from the experience with other species of smelt (Moring, 1985; Akielaszek et al., 1985; Kashiwagi et al. 1988) and marine fishes with small pelagic larvae (Blaxter 1981; Rosenlund et al. 1993; Reitan et al. 1994; Baskerville-Bridges 1999; Baskerville-Bridges and Kling 2000). Culture methodology for delta smelt is based on studies conducted at two locations, UC-Davis and the State Water Project, SWP, Byron, CA (the south Delta). Since 1998, the culture effort was consolidated at the SWP site, offering advantages of closer location to broodstock capture and availability of natural delta water that stimulates smelt spawning. The culture program has advanced methods for the capture, spawning, incubation, and rearing of smelt (Mager 1996; Mager et al. 1996; Lindberg 1992; Lindberg et al. 1996, 1998, 1998a, 1999, 2000; Baskerville-Bridges et al. 2001). The spring season of 2001 was the most successful to date, with 10,000 larvae and juveniles produced. We anticipate to further increase production in 2002 by optimizing larval rearing temperatures and tank design for rearing juveniles (Baskerville-Bridges et al. 2001).

Goals of project. The main goal of the program is to create a reliable supply of live delta smelt at all stages to meet the needs of the research community. This supply also represents a refuge population needed for conservation should the population decline further. With improved efficiency of the culture system, we also propose to investigate the role of polyunsaturated (n-3) fatty acids in the larval diet and the spawning substrate preferences that may influence egg deposition in culture and in the wild.

A 2. Justification (concepts, hypotheses and project type)

Delta Smelt Culture and Research Program is a research and development hatchery operation located on State Water Project land in the south Delta. Supported by state and federal funds, this project supplies delta smelt eggs, larvae, and juveniles to a wide variety of research projects. The research element of the Delta Smelt Culture Program provides insight into the reproductive biology, development, growth, and behavior of delta smelt. The availability of research material and the project findings stimulate further hypotheses and research questions from managers and members of the scientific community, leading to more in-depth investigations into the smelts' biology and natural habitat. This process represents the basis of the scientific method that is supported by the CALFED as contributing to restoration of at-risk native species. The supply of live delta smelt allows many investigations to move forward. This year 10,000 larvae and juveniles were produced to supply research animals for testing fish screens, effects of contaminants, predator-prey interactions, and to create a laboratory standard for growth in known age animals from which comparisons can be made to larvae collected in the field. In addition, we determined growth and survival rates of larvae and juveniles at three different temperatures, which provides valuable data for field comparisons and for population and habitat modeling. This proposal responds to at least three of the current PSP's priorities: (1) need for basic life history information of at-risk species, (2) need for knowledge of contaminant effects on at-risk species, and (3) need to understand (and minimize) effects of water diversions on at-risk species. Products of the proposed work that will address these three points include: (1) information on factors influencing nutrition and growth of the larval smelt and the spawning behavior of smelt; (2) a supply of native fish with known rearing history for studies of toxicology and fish health, and (3) live smelt for testing new screens at water diversion sites.

In the next section we present a conceptual model for each of the three objectives of our proposal. The major thrust of the project will be to improve delta smelt culture system (Objective 1), in order to provide for a continuous supply of research animals. The Objectives 2 and 3 are the experimental studies on larval nutrition and spawning behavior of smelt, which may enhance delta smelt culture.

Conceptual model for smelt culture (Task 1). Culture system for delta smelt evolved from the field and laboratory observations on life history (Moyle et al., 1992), development (Mager 1996), and larval culture (Baskerville-Bridges et al. 2001) of this estuarine species. Delta smelt are small and fragile pelagic fish, living in the brackish water of upper estuary, having predominantly annual life span, and spawning during spring season in the river delta. Delta smelt have low fecundity, adhesive mesolecithal eggs, relatively short duration of embryo (pre-hatch) development, and a prolonged pelagic larval phase with late acquisition of functional swim bladder and hydrostatic regulation. The larval delta smelt is a most sensitive stage of the life cycle in culture, vulnerable to the physical (water flow and tank design), chemical (water quality), and biological (feeding requirements in live micro-zooplankton) factors. Because of the need to maintain optimal environment and to avoid excessive domestication, the culture of delta smelt evolved as an intensive, low-density, one-cycle system, with minimum handling and close to natural seasonality. The culture procedures involve the annual procurement of wild broodstock (as juveniles, in October - November), rearing them in tanks on prepared feed to full sexual maturity (November - March), natural spawning in tanks (March - May), and rearing larvae and juveniles on live (cultured rotifers and brine shrimp nauplii) and prepared (manufactured) diets.

Our simple hypothesis is that smelt production is a function of the survival and optimal performance of all life stages in the above described system. We assume that all life stages of delta smelt can be reared effectively in culture given the correct conditions for each life stage, including culture density, water quality, tank size and color, feed types, nutritional qualities, and feeding practices, seasonal temperature and photoperiod. Our current production (as of this writing, September 2001) of delta smelt juveniles and advanced (viable) larval stage was ca.10,000 resulting from 700 broodfish (males and females, 1:1 ratio). By optimizing culture methods, husbandry, and environment (based on our current and proposed experimental studies), we should be able to double production of juveniles in 2004 from the same broodfish number and, potentially, to increase the

production to 30,000 juveniles that can be used for research and testing new fish screens at water diversion sites. *Delta smelt culture project* is a small-scale research and development program with the goal of creating a sustained production of this cultured, *threatened*, species for research. Studies with cultured fish enhance knowledge of smelt' basic biology, habitat requirements, and environmental impact on the organism and population. Thus the delta smelt culture program contributes to restoration goals of CALFED by conducting and supporting research programs important to the restoration of this at-risk species.

Conceptual model for larval nutrition (Task 2). Production of delta smelt can be improved by investigating larval requirements in polyunsaturated long-chain omega 3 and omega 6 fatty acids, particularly eicosapentaenoic (EPA, 20:5 (ω -3)) and docosahexaenoic (DHA, 22:6 (ω -3)) acids. These fatty acids play a significant role in the development of brain, function of eye retina, integrity of cell membrane, synthesis of prostaglandins, and tolerance to stress (Henderson et al. 1985; Kanazawa 1985; Bell et al. 1995; Sargent et al. 1997). The ability to improve stress resistance is linked in larval fish to the level of DHA in their diet (Kanazawa 1997). Stress-induced mortality currently results in smelt from excessive handling (tank cleaning, grading, and transport of fish), negatively impacting the larval production system. The EPA and DHA requirements vary among fish species and differ in the freshwater and marine fish. Freshwater fish are able to elongate and desaturate linolenic acid (LLA, precursor of EPA and DHA) and do not require DHA and EPA in their diet (Watanabe 1982; Sargent et al. 1997), although some exceptions were found (e.g. striped bass, Tuncer and Harrell, 1992). Marine fish have limited capabilities to elongate and desaturate LLA and do require EPA and DHA in their diet (Watanabe 1982; Takeuchi 1997; Sargent et al. 1999). In addition, marine fish are not able to convert sufficient quantities of EPA to DHA during larval growth (Sargent 1997). Marine fish also lack the ability to convert linoleic acid to arachidonic acid (omega-6 fatty acid), which is important for eicosanoid synthesis (Sargent et al. 1997). Little is known about the fatty acid requirements of delta smelt living in an area of low salinity and feeding on estuarine zooplankton (Moyle et al. 1992), which provide a source of EPA and DHA during the larval period. The primary source of these fatty acids is marine or estuarine microplankton, such as dinoflagellate and certain species of phytoplankton.

Rotifers and brine shrimp, used as the main live food for fish larvae, are rich in LLA, but EPA and DHA are generally absent or present in low amounts (Sargent et al. 1999). The enrichments of prey with emulsions of marine oil or ethyl esters of fatty acids have been extensively used in aquaculture to improve the nutritional quality of these organisms by increasing the levels of EPA and DHA (Robin et al. 1991; Southgate and Lou, 1995). This is accomplished by feeding the secondary nutrients to a live prey prior to its introduction into the fish tank (Watanabe et al. 1983; Leger et al. 1986; Sargent et al. 1997). The current enrichment procedure for smelt larvae utilizes DC-Super Selco (Inve Aquaculture, Inc., Grantsville, UT) but its efficacy has not been evaluated for smelt. We propose experiments to determine the effects of enrichments with EPA and DHA on the performance of larval smelt in culture.

Our *null hypothesis* is that delta smelt larvae are able to utilize LLA to satisfy their fatty acid requirements and do not require EPA and DHA in their diet. To test this hypothesis, delta smelt larvae will be fed rotifers enriched with EPA and DHA, and their performance (survival, growth, and tolerance to stress) as well as body lipid composition will be compared to non-enriched controls. If the smelt larvae do not require supplemental EPA and DHA then we could eliminate the enrichment process and simplify our current larval rearing protocol. If the supplementation of EPA and DHA significantly improves larval performance, we will further investigate the optimization of enrichment protocol. The proposed study is a research project, which will test how varying levels of EPA and DHA influence performance of smelt larvae and their fatty acid profiles. This research will provide initial information on fatty acid requirements of delta smelt larvae and may lead to further improvement of the smelt culture system. It may also be of interest for the evaluation of health of larval smelt in the natural habitat where the changes in microplankton (primary source of polyunsaturated long-chain omega-3 fatty acids) may affect nutritional quality of larval prey.

Conceptual model for spawning behavior (Task 3). A host of factors, both biotic and abiotic, affect spawning behavior of delta smelt. Delta smelt appear to be broadcast demersal spawners: distributing their adhesive eggs low to the river bottom and providing no parental protection. The

eggs attach to the substrate by a highly adhesive, elastic outer chorion that everts to form the attachment “foot” (Mager 1996). Out of all potential spawning habitats we assume smelt seek out a set of conditions, a microhabitat, which optimizes embryo survival. The proposed study will test the effect of the spawning substrate on spawn deposition rate. Several substrates such as gravel, sand, plants, and smooth surfaces will be tested. We expect that a variety of dense substrates will prove suitable for spawning. Water flow perturbations associated with certain substrates such as vertical plants could also affect spawn deposition. Factors found important to spawn deposition in the large outdoor test tanks may also be important to smelt in their natural habitat. Smelts spawn on the river or tidal currents, but some species may spawn in lakes and ponds. Observations in the field have shown that delta smelt disperse to fresh water from the estuarine mixing zone to spawn in dead end sloughs of the Delta (Radke 1966), or in areas with higher water flows (Moyle 1976; Wang 1991; Lindberg and Marzuola 1993). Unfortunately, the visibility is often less than 30 cm, precluding direct observations of spawning behavior in these areas. Studies using artificial spawning substrates have been unsuccessful in gathering delta smelt eggs in the field, even in areas known to contain spawning adults (pers. com. Aasen, DFG; pers. com., Fleming, DFG). Wang (1986) found a few eggs of smelt on submerged tree roots and firm leaves, but not on soft substrates. Moyle (1976) suggests that one may find smelt eggs on hard substrates such as sand on the river bottom, or on submerged vegetation near the river bottom.

Observations in the laboratory provide some insight into delta smelts’ spawning behavior. In our early culture experiments, fish were observed to swim flank to flank, broadcasting their eggs and sperm within 2-3 cm of the tank floor (Lindberg 1992). The eggs drifted to the floor and adhered. Eggs that we currently collect from broodfish tanks are routinely found evenly distributed on the tank floor and not up the tank walls or on the standpipe higher than 3-4 cm from the bottom (Lindberg et al. 1998). These observations suggest that delta smelt may broadcast their eggs on the bottom with some current so that spawned eggs are uniformly distributed on horizontal substrates, and not on the vertical substrate (Lindberg 1992). The proposed experiment stems from the interest to understand the natural spawning behavior of the smelt, and to test whether we can alter the tank environment to obtain more complete spawning of captive broodfish. Environmental factors found important in stimulating spawning of captive fish may also prove important for the wild fish. In our culture system, most broodfish spawn in tanks with a bare bottom, but some females spawn incompletely or not at all. These broodfish later succumb to disease and die, as expected of a near-annual fish (Radke 1966). We surmise, from the number of females retaining eggs, that the homogeneous tank environment may lack physical or other factors that stimulate egg deposition. Our spawning tanks receive natural delta water with presumed springtime spawning cues for smelt (e.g. turbidity, plankton bloom, seasonal temperature and day length). However, some other important cues may be absent, for example the variation in substrate or water current velocity. In the current study we propose to quantify spawning response to several naturalistic substrates, while maintaining current velocity at a level comfortable for delta smelt swimming (Swanson et al. 1998).

Our *null hypothesis* is that spawns will be evenly distributed on all substrates. The hypothesis will be tested by offering several different substrates and quantifying incidence and density of spawns on all substrates, including the tank floor. Experimental results may allow rejection of the null hypothesis to suggest that smelt prefer some of the substrates, which will be important to know for smelt culture and field studies. The proposed study is an experimental research project to test for factors that may affect the location and densities of delta smelt spawning. Knowledge of these factors will be used to improve production of smelt in culture and in restored spawning habitat of delta smelt.

A. 3. Approach to Study Design

Location of the delta smelt culture and experimental facilities is at the mouth of the canal to Banks Pumping Plant, SWP land, in the south Delta near Byron. The site culture facilities include: indoor and outdoor broodstock rearing/spawning tanks, indoor water recirculation and temperature controlled systems for egg incubation and larval and juvenile rearing, culture systems for the production of live food (rotifers and brine shrimp nauplii), and large outdoor tanks (20’ diameter) for the proposed spawning studies. The laboratory is equipped for basic water quality analysis (dissolved oxygen, electric conductivity, pH, and ammonia), microscopy, weighing, measuring, and fixation of

fish and samples. The analyses of dry weight and fatty acids will be conducted in the nutrition and biochemistry laboratories of the Department of Animal Science, UC-Davis.

Task 1. Delta smelt culture. Basic techniques and methodologies for delta smelt culture have been previously described (Lindberg 1992; Mager 1996; Lindberg et al. 1998; Baskerville-Bridges et al. 2000). In brief, the juveniles or sub-adults will be collected in the fall of each year, netted by purse seine deployed from a boat, usually from the lower Sacramento River south of Rio Vista. Fish are transported to the SWP, Byron, acclimated to tank conditions and weaned to prepared diets. Fish mature and spawn naturally on tank bottoms during March-June. The disease management includes disinfection (broodfish tanks) and water sterilization (larval tanks) and rearing animals in multiple isolated tanks, at proper temperatures, feeding regimes, and reduced handling stress (Antonio et al. 2000). Spawning eggs are collected from tanks, incubated in flow-through containers, and resulting larvae are reared in tanks on live (rotifers and brine shrimp nauplii) and prepared diets (Biokyowa Inc.) to metamorphosed juveniles (August-October). Care and maintenance are provided for broodfish, eggs, larvae, juveniles and sub-adults for 12 months of the year, seven days a week. Larval rearing protocols will be updated with results from studies conducted in 2001-02. The new protocol will include a staged increase in rearing temperature and tank size with age and phase of larval development. The 0-60 d old larvae (length from 5 to 18 mm) will be reared at temperature 17°C, and the temperature will then be increased to 20°C (at 1°/d) to accelerate the growth and metamorphosis. Larvae and juveniles will be transferred to larger tanks at age 60 d (200 L tanks) and 120 d (550 L tanks) to improve survival and growth. Rotifers (*Brachionus plicatilis*) and brine shrimp nauplii (*Artemia* sp.) are cultured to provide the appropriate prey size to the larvae as they develop. Live prey are initially (from age 5 d) fed 7 times a day to maintain desired concentrations (5-7 /ml). Older larvae (age 18-22 d) are weaned onto brine shrimp nauplii, and shortly thereafter onto a prepared commercial diet. Prey organisms are enriched with Super Selco (Inve Aquaculture, Inc., Grantsville, UT) to increase their nutritional quality. Co-feeding of live and prepared diet is employed to improve survival during transfer on prepared diet. The smelt culture is evaluated each year by determining: total number of eggs spawned (count or volumetric estimate), growth rates of larvae and juveniles (length and dry weight), and survival to specified age and size. Our production target for Year 2 (2004) is 20,000 juveniles > 20 mm length, which doubles current production. The production by stages (i.e. eggs, larvae, juveniles) will be determined by requirements of different laboratories and approval of allocations based on consensus of the Resident Fish Project Work Team (for the Delta area) of the IEP. Distribution of early life stages (eggs and larvae) to various research programs supplies life stages heretofore unavailable for research, however these distributions reduce the total number of juveniles and older fish the culture program will produce.

Task 2. Larval nutrition. A feeding trial will be conducted in Year 1 to test the effects of three enrichments on growth, survival and tolerance to stress of smelt larvae. Twelve tanks (70-L) will be stocked with 2,800 larvae each (40 larvae/L) and randomly assigned one of the three enrichment diets (n=4). Rotifers will be enriched with emulsions containing triacylglycerol or ethyl esters to provide three fatty acid profiles of larval prey (EPA and DHA as a percent of the dry weight): 1) control (yeast-fed rotifers containing little or no EPA and DHA), 2) 1% EPA, 3) 1% EPA and 1% DHA. These treatments will provide information relative to the ability of smelt larvae to produce sufficient quantities of DHA from shorter chained, more saturated precursors (such as LLA or EPA).

One hundred larvae will be sampled on day 5 for initial length and weight measurements, and fatty acid analysis, prior to their transfer into the experimental tanks and start of feeding. Twenty larvae will be sampled from each tank on days 20 and 40, for measuring length, weight and fatty acid analysis. The remaining larvae at the end of the experiment (day 40) will be counted to calculate survival. A vitality test will also be conducted at this time to examine the effect of each treatment on stress tolerance: ten fish per tank will be exposed to the air for 20 seconds and survival will be determined after one hour upon returning larvae to water (Kanazawa 1997).

Total lipids will be extracted from rotifers and larvae sampled on day 5 and day 40 (Bligh and Dyer 1959). Polar and neutral lipids will be separated by thin layer chromatography and total lipid in each fraction will be expressed as a percentage of the dry weight. Fatty acid analysis will be conducted as described by Sukhija and Palmquist (1988), except that hexane will be used instead of benzene during the methylation procedure. Separation of fatty acid methyl esters will be performed

on a 5890A gas liquid chromatograph. Fatty acid methyl esters will be identified by comparison of their retention times to that of known standards and quantified using nonadecanoic acid as an internal standard.

Fatty acid composition of the live prey and larvae at the end of the experiment will be compared to examine differences in uptake of the enrichments. In particular, linoleic, linolenic, arachidonic, eicosapentaenoic, and docosahexaenoic acid contents will be examined to compare the effects of each enrichment procedure. Fatty acids will be expressed as a percent of the dry weight and as a percent of the total fatty acids. This study will provide preliminary information regarding the fatty acid requirements and essentiality of long-chain HUFA's for larval delta smelt. If the results demonstrate significant effects (either on body fatty acid composition or larval performance, or both), a similar experiment will be conducted during Year 2, with an objective to optimize enrichment. Choice of the enrichment media and fatty acid enrichment levels will be decided based upon the results of first experiment. For statistical analysis, treatments will be compared at each sampling time. Normality of the data (Shapiro and Wilk, 1965) and homogeneity of variance (Snedecor and Cochran, 1993) will be tested to ensure the assumptions for analysis of variance are satisfied. Proportion data (survival) will be arcsine-transformed before conducting analysis of variance (Snedecor and Cochran, 1993). Differences due to level of EPA and DHA will be considered significant at $P < 0.05$ and Duncan's multiple range test will be used for mean separation (Snedecor and Cochran, 1993).

Task 3. Spawning behavior. Three large outdoor circular tanks (20' diameter x 20" water depth) will be used as replications for the spawning experiments. Natural delta water will provide a flow-through system, with inflow distributed evenly around the perimeter of the tank to create a low velocity (< 8 cm/sec) appropriate for sustained 'stroke-and-glide' swimming (Swanson et al., 1998). Captive sub-adult smelt populations (from the culture program) will be randomly assigned to each of three tanks (100/tank), and each group will be acclimated to tank conditions for at least one month before the onset of spawning season (end of February-March). Fish will be fed and tanks cleaned daily. Five different substrate types will be tested in each tank, including tule plants (*Scirpus acutus*) artificial plants, sand, gravel, and empty tray (4 trays per substrate). Two complete sets of each substrate will be prepared. Trays will be randomly distributed in tanks in early February and, after onset of spawning, will be examined for the presence of eggs every 3 times per week depending on water temperature (recorded as min/max daily). When a spawn is detected all trays will be removed and replaced with identical second set. The adhesive eggs are de-adhered by bathing tray (3-4 min) in diluted (1:300) commercial bleach and washing eggs from substrate by water. Total egg number on each tray will be estimated volumetrically. Eggs will be also collected from the floor of the tank by scraping. The expected spawning duration in experimental tanks is approximately 4 weeks during March-April. The response variable is number of eggs per tray. The effects of substrate on egg density will be examined by analysis of variance and appropriate mean comparison tests, to determine if substrate type plays significant role in smelt spawning. The analysis may suggest retesting of a few substrates or the testing of new substrates. Therefore, similar a study will be conducted during the second year using modified design.

Task 4. Project Management. Doroshov, Lindberg, and Baskerville-Bridges will manage the project jointly. Principal Investigator will guide the project and hold regular meetings to review the work in progress for each task. Baskerville-Bridges will supervise the studies on Tasks 1 and 2. Lindberg will supervise Task 3 and communications of project with researchers, managers, and agencies. Van Eenennaam will supervise budget planning and assist in reports. Project leaders will participate in scientific conferences and prepare jointly authored publications.

A. 4. Feasibility of Approach to Conducting the Delta Smelt Culture and Research Program

The Delta Smelt Culture Program has had several years of good performance establishing successful methodologies and training personnel that will ensure the achievement of project goals, therefore we seek continued funding. This past year we had excellent results, producing 10,000 larval to juvenile aged fish. Continuity of funding, particularly to retain experienced personnel, is

critical to the success of the project. Currently we hold both a state and federal permit for the capture of delta smelt broodstock and for subsequent culture; these permits will be renewed. The project has a UC-Davis Animal Care Permit required for research with live animals. To the best of our knowledge we do not require other permits to conduct the proposed research. We have appropriate smelt culture facilities at the SWP site in Byron and supporting laboratory facilities at UC Davis. State Department of Water Resources provides logistic support to the project and maintenance of electricity and water supply systems. We do not foresee any specific constraints that will negatively impact this project.

A. 5. Performance Measures

Delta smelt culture (Task 1):

- Culture system: successful management of broodstock, spawning, and larval rearing; implementation of new techniques; survival and growth of cultured fish; achieving production targets.
- Project outputs: supply of delta smelt to researchers and managers (two years); updated manual of delta smelt culture on computer disc (at termination of project); research publication on delta smelt culture.

Larval Nutrition (Task 2):

- Experiment: quality of experimental design, procedures, and results.
- Project outputs: research publication; potential implementation of improved enrichment procedure in culture practice.

Spawning Behavior (Task 3):

- Experiment: quality of experimental design, procedures, and results
- Project outputs: research publication; potential use of preferred spawning substrates in culture and for the field studies

Project Management (Task 4): timely and successful coordination of tasks, budget, and reports.

A. 6. Data Handling and Storage

Data summaries of project tasks will be made available for uploading to the CALFED web site.

A. 7. Expected Products and Outcomes

Quarterly reports will include financial status, brief description of project activities, tasks accomplished, and deliverables produced. A final report describing the results of the project will be submitted by the end of the project (Oct 30, 2004). Study results will be presented at scientific and technical meetings, and in IEP Newsletter articles. Manuscript "Manual of delta smelt culture" (to be compiled during 2002 under current CALFED Agreement # 00FC200113) will be updated by incorporating description of new techniques and will be made available on computer disc to all interested parties. Manuscripts will be submitted for publication in peer-reviewed research journal (Aquaculture, J. Fish Nutrition, Trans. Amer. Fish. Soc.). Delta smelt eggs (live embryos), larvae, juveniles, and subadults will be supplied to research and testing laboratories over two year duration of the project; the requests and supplies will be monitored to determine needed production output and the quality of material supplied.

A. 8. Work Schedule

We are requesting funding for two years beginning November 1, 2002 (at the completion date of our current project “Culture of delta smelt *Hypomesus transpacificus* in support of environmental studies, Phase II and III”, CALFED Agreement # 00FC200113). Culture and experiments with early life stages of delta smelt require trained and skilled personnel, making timely project funding critical for success. The proposed tasks and schedule (Table) are based on the annual cycle of smelt reproduction and contingent on personnel support by project funds. Tasks 1 and 4 represent major effort and cost. The Task 1 (smelt culture) supplies animals for Tasks 2 and 3.

Table. Tasks and schedule for proposed delta smelt culture and research program. Budget represents State funding level at 10% overhead (Federal overhead is 26%).

Task	Schedule and (budget) by task
Task 1. Delta Smelt Culture	Nov 2002 – Oct 2004 (419,207)
Task 2. Larval Nutrition	Nov 2002 – Oct 2004 (52,693)
Task 3. Spawning Behavior	Nov 2002 – Oct 2004 (54,224)
Task 4. Project Management	Nov 2002 – Oct 2004 (75,794)

Annual Time Line with Milestones

Project’s time line (below) is determined by seasonality of reproductive migration, spawning, and larval-to-juvenile development and growth of delta smelt. Some activities overlap in time because of extended spawning season of smelt.

Year 1: Nov 1, 2002-Oct 31, 2003

Nov-Jan	Capture, acclimation, and rearing of smelt broodstock (Task 1)
Feb-Jun	Spawning and egg incubation (Task1)
Feb-Apr	Experiment on spawning behavior (Task3)
May-Jul	Experiment on larval nutrition (Task 2)
Apr-Aug	Larval rearing (Task 1) and data analysis on spawning behavior (Task 3)
Jul-Sep	Juvenile rearing (Task 1) and data analysis on larval nutrition (Task 2)
Oct	Summary report (Task 4)

Year 2: Nov 1, 2003-Oct 31, 2004

Nov-Jan	Capture, acclimation, and rearing of smelt broodstock (Task 1)
Feb-Jun	Spawning and egg incubation (Task 1)
Feb-Apr	Experiment on spawning behavior (Task 3)
May-Jul	Experiments on larval nutrition (Task 2)
Apr-Aug	Larval rearing (Task 1) and data analysis on spawning behavior (Task 3)
Jul-Sep	Juvenile rearing (Task 1) and data analysis on larval nutrition (Task 2)
Oct	Data analysis and final reporting (Tasks 1-4)

B. APPLICABILITY OF PROPOSED RESEARCH TO CALFED GOALS AND IMPLEMENTATION PLAN

B. 1. Applicability to ERP, Science Program, and/or CVPIA Priorities

Delta smelt culture and research program is applicable to the CALFED ERP goals and implementation plan. Initiation of the smelt culture project was suggested by the Department of Water Resources (DWR) in 1992, and the project was supported by DWR and US Bureau of Reclamation through the Interagency Ecological Program for the Sacramento – San Joaquin Estuary (IEP) for several years, as a way of creating a refuge population in the event of extinction, and as a supply of animals for research. The CALFED Program has supported the smelt culture program in the past (Phase I, 7/89-6/99) and currently supports the Phase II and III (7/00-10/02). The CALFED ‘Study Needs for At-Risk Species’ states as an objective: “determine appropriate methods for rearing delta smelt in captivity, and evaluate need to acquire rearing facilities if delta smelt populations continue to decline after restoration actions begin” (Attachment 3 of CALFED’s Stage 1 Implementation Plan). The smelt culture program is now demonstrating success in rearing this delicate native species. This past season 10,000 larval-to-juvenile-stage smelt were provided to several agencies (UCD, DWR, USBR) for delta smelt research. The culture project made progress towards CALFED goals by creating a supply of this at-risk fish for research. Continued support will permit improvements to rearing techniques that will ensure a reliable supply of delta smelt in the future and, if needed, maintenance of a refuge population.

The current CALFED ERP states: “Highest priority is given under the Draft Stage 1 Implementation Plan to native at-risk species ... that most strongly affect the State Water Project’s (SWP) or the Central Valley Project’s (CVP) diversions in the south Delta, such as *delta smelt*, all runs of Chinook salmon, steelhead trout, and splittail.” (Strategic Goal 1), and “The recovery of at-risk species is at the heart of the ERP. There is a need to better understand the life history, abundance, distribution, and habitat requirements of species for which CALFED has the responsibility for their recovery or needs to contribute to their recovery.” (CALFED Draft Stage 1 Implementation Plan). The main contribution the delta smelt culture program provides to CALFED ERP’s goals is the supply of all life stages of smelt for research programs and a refuge population of smelt against the possibility of extinction. The supply of cultured smelt directly reduces the take by some researchers (also saving time and money). Other research programs simply have not been able to obtain animals in the early life stages they need to conduct their research until the establishment of this cultured supply. By 2004 we aim to double our current production rate creating a more efficient supply of smelt.

Within the Delta and Eastside Tributaries Region (DR) the current PSP lists 8 restoration priorities. Our proposal addresses one or more aspects of 3 of the 8 priorities by: (1) developing a better understanding of smelt’ life history (e.g. growth curves for larvae and juveniles at 3 temperatures, creating standard for comparisons to field samples; and spawning behavior study proposed here may offer life history information applicable to habitat restoration (DR Priority #4; Strategic Goal 1, at-risk species, Strategic Goal 4, habitats, p.39); (2) creating a unique stock of smelt with known rearing history for delta contaminant studies (DR Priority #6;

Strategic Goal 6, water and sediment quality, p. 42. A request for 2400 eggs and 2400 larval delta smelt was submitted to the culture program for 2002 to test for toxicity effects of herbicide used in the delta (Pesticide Investigative Unit, DFG); (3) providing smelt for testing fish screen designs for reducing the impact of water diversions in the Delta (DR Priority #7; Strategic Goal 1, at-risk species, p. 43). The CVP (Tracy Pumping Station) in the south Delta has put forward a notification to the Smelt Culture Program that thousands of fish, including delta smelt, are needed for the purpose of developing and testing new fish screen designs that minimizes stress to the fish in just a few years. They requested 1500 larval-to-juvenile age fish for 2002.

B. 2. Relationship to Other Ecosystem Restoration Projects

The Delta Smelt Culture Project currently receives CALFED funding (Jul 00 – Sep 02) for Phase II and III of a three-phase project. The Smelt Culture Program has also received support from the State Department of Water Resources (DWR) and the U. S. Department of Reclamation (US Bureau), and the Interagency Ecological Program (IEP). These State and Federal agencies, and the University of California, continue to show interest in the Smelt Culture Program's ability to provide basic information on the biology of the smelt, and in having access to a reliable supply of live smelt at all ages. The US Bureau is currently donating labor hours to our efforts. They stand to benefit from information obtained regarding smelt rearing and holding techniques as they begin determining the effectiveness of new fish screen designs for native at-risk fishes. DWR personnel also contribute labor and some additional funding for operation and maintenance of smelt facility at the SWP site.

During the last several years we have supplied smelt at various life stages to research programs. We supplied healthy post-spawn fish to Hanson and Associates (Walnut Creek) for testing sensitivity to an acoustical barrier. We created a preserved developmental series of embryos and juveniles for two projects: the comparative morphology of delta and wakasagi smelts (J. Wang) and for the larval otolith-aging study (L. Grimaldo, DWR, and D. Sweetnam, DFG). We supplied live embryos for toxicity testing of a locally used herbicide (C. Huang, DFG 1998). In 1999 - 2001 we supplied delta smelt for ecological (B. Bennett, BML, UCD) and physiological studies (J. Cech, UCD). We also supplied preserved larvae and juveniles to the US Bureau at the Tracy CVP site for developing larval identification techniques (J. Wang and B. Baskerville-Bridges). In future year indications are that demand for cultured smelt will increase. The Federal Bureau of Reclamation plans to build a new water diversion channel at Tracy and anticipates using large numbers of delta smelt as a sensitive native fish species for testing new screens.

B. 3. Requests for Next Phase Funding

Our current CALFED project (Phase 2 and 3) will be completed 10/31/02. This new proposal represents the next phase of Delta Smelt Culture Program – that of increasing production capabilities through continued research and development. The program refines culture methodologies leading to improved culture system and production. The program successfully rears and distributes delta smelt, has retained experienced personnel, and conducts experiments at a facility built with state and federal funding.

B. 4. Previous Recipients of CALFED Program or CVPIA funding

Our current and previous delta smelt culture projects funded by CALFED for a total of three years are identified below. Over the last several years the smelt culture program has made significant progress in creating a supply of delta smelt for research. With the continued support, we aim to further improve the production system to rear 20,000 juveniles by October of 2004. This improvement relies on our continued studies of factors important to reproduction, development, growth, and health of delta smelt. We are on target with all deliverables and have several journal papers in preparation.

Current /previous support from CALFED:

Jul '00 – Oct '02: Culture of delta smelt *Hypomesus transpacificus* in support of environmental studies, Phase II and III. S.I. Doroshov, J.C. Lindberg and J Van Eenennaam, Agreement # 00FC200113 with US Bureau of Reclamation; \$559,446 funded by CALFED Bay Delta Program.

July '98 – June '99: Culture of delta smelt *Hypomesus transpacificus* in support of environmental studies, Phase I. S.I. Doroshov, J.C. Lindberg and J Van Eenennaam, Contract B-81581 with Department of Water Resources; \$194,870 funded by CALFED Bay Delta Program.

B. 5. System-Wide Ecosystem Benefits

Delta Smelt Culture Program can benefit the delta-smelt recovery by providing information on the biology of the animal and by creating a supply of smelt for research. The experiments with cultured smelt raise new hypotheses and yield reference information for comparison to natural populations; findings of these experiments may be useful for habitat restoration projects. The supply of live animals at all life stages benefits restoration-oriented studies, such as: (1) monitoring the health of delta smelt in the wild, (2) contaminant levels and exposure studies, (3) development of taxonomic keys for larvae, (4) studies on delta smelt swimming and improving fish screen design for water diversions.

B. 6. Additional Information for Proposals Containing Land Acquisitions

No land acquisitions are proposed.

C. Qualifications

SERGE I. DOROSHOV, Ph.D., Professor of Animal Science, UC Davis, 1983 to present.

Four Selected Publications: (1) Doroshov, S.I., G.P. Moberg and J.P. Van Eenennaam. 1997. Observations on reproductive cycle of cultured white sturgeon, *Acipenser transmontanus*. *Env Biol Fishes* 48: 265-278; (2) Doroshov, S.I., J.P. Van Eenennaam, and G.P. Moberg. 1999. Development of white sturgeon broodstock. *J Appl Ichthyol* 15: 326-327; (3) Antonio, D.B., C. Swanson, J.J. Cech, Jr., R.C. Mager, S. Doroshov, and R.P. Hedrick. 2000. Prevalence of *Mycobacterium* in wild and captive delta smelt. *Calif Fish Game* 86: 233-243; (4) Webb, M.A.H., J.P. Van Eenennaam, G.W. Feist, J. Linares-Casenave, M.S. Fitzpatrick, C.B. Schreck, and S.I. Doroshov. 2001. Effects of thermal regime on ovarian maturation and plasma sex steroids in farmed sturgeon, *Acipenser transmontanus*. *Aquaculture* 201: 137-151.

JOAN C. LINDBERG, Ph.D., Postdoctoral Researcher, Animal Science, UC Davis, 1996 to present.

Four Selected Publications: (1) Lindberg, J.C. and S. I. Doroshov, 1986. Effect of diet switch between natural and prepared foods on growth and survival of white sturgeon juveniles. *Trans Amer Fish Soc* 115:166-171; (2) Lindberg, J., R. Mager, B. Baskerville-Bridges and S. Doroshov. 1997. Status of delta smelt culture project. Interagency Ecological Program for the Sacramento-San Joaquin Estuary Newsletter, 10, (3): 31-32; (3) Lindberg J.C., B. Baskerville-Bridges, J.P. Van Eenennaam, S.I. Doroshov. 2000. Update on delta smelt culture with an emphasis on larval feeding behavior. Interagency Ecological Program for the Sacramento-San Joaquin Estuary Newsletter 13 (3): 45-49; (4) Lindberg, J.C., B. Baskerville-Bridges, J. P. Van Eenennaam, S.I. Doroshov. 1999. Development of Delta Smelt Culture Techniques; Year-end report 1999. Report to California State Department of Water Resources, Sacramento (DWR B-81581).

BRADD BASKERVILLE-BRIDGES, Ph.D., Postdoctoral Researcher, Animal Science, UC Davis, 1998 to present.

Four Selected Publications: (1) Baskerville-Bridges, B. and L.J. Kling, 2000. Development and evaluation of microparticulate diets for early weaning of Atlantic cod (*Gadus morhua*) larvae. *Aquaculture Nutrition* 6:171-182; (2) Baskerville-Bridges, B. and L.J. Kling, 2000. Larval culture of Atlantic Cod (*Gadus morhua*) at high stocking densities. *Aquaculture* 181:61-69; (3) Baskerville-Bridges, B. and L.J. Kling, 2000. Early weaning of Atlantic cod (*Gadus morhua*) larvae onto a microparticulate diet. *Aquaculture* 189:109-117; (4) Baskerville-Bridges, B., J. Lindberg, J. Van Eenennaam, and S. Doroshov, 2001. Progress and Development of Delta Smelt Culture: Year-end Report 2000. *IEP Newsletter* 14 (1): 24-30.

JOEL P. VAN EENENNAAM, M.S., Research Associate, Animal Science, UC Davis, 1985 to present.

Four Selected Publications: (1) Van Eenennaam, J.P., S.I. Doroshov, G.P. Moberg, J.G. Watson, D.S. Moore and J. Linares. 1996. Reproductive conditions of the Atlantic sturgeon (*Acipenser oxyrinchus*) in the Hudson River. *Estuaries* 19: 769-777; (2) Doroshov, S.I., G.P. Moberg and J.P. Van Eenennaam. 1997. Observations on reproductive cycle of cultured white sturgeon, *Acipenser transmontanus*. *Env Biol Fishes* 48: 265-278; (3) Van Eenennaam, J.P. and S.I. Doroshov. 1998. Effects of age and body size on gonadal development of Atlantic sturgeon. *J Fish Biol* 53: 624-637; (4) Van Eenennaam, J.P. and seven co-authors. 2001. Artificial spawning and larval rearing of Klamath River green sturgeon (*Acipenser medirostris*). *Trans Amer Fish Soc* 130: 159-165.

BOBBY RENSCHLER, B.S., Laboratory Assistant, Animal Science, UC Davis, 2000-present. Education in Animal Science/Aquaculture and excellent experience with breeding and rearing larval smelt

LUKE ELLISON, Laboratory Assistant, Animal Science, UC Davis, 1999-present. Excellent two-year experience with smelt culture.

D. Proposed Cost

Budget is provided with State overhead (10%); Federal overhead for off-campus (University) research is 26%. Project total cost for the two years (11/02-10/04) with State funds is \$601,918. Broken down, cost to State is \$299,546 for year 1, and \$302,372 for year 2. Cost share for on-site repairs and maintenance is approved by DWR (\$2500 per year; attachment A). The major part of the budget supports four personnel at the delta smelt culture facility (Baskerville-Bridges, Lindberg, Renschler, and Ellison). Their previous experience and technical skill are important to the continued success of the Delta Smelt Culture and Research Program. All proposed work, except for the analysis of fatty acids, will be conducted outside the University Campus, at the SWP-site facilities near Byron.

E. Local Involvement

The Delta Smelt Culture Project is a small-contained operation, located on state property (SWP land, DWR) in the south Delta. This proposal does not involve land acquisitions or restoration of public or private lands. Therefore the project is not impinging on other landowners, and it is unlikely to have any adverse effects on the public or private sector.

Local support has been shown for the project by DWR personnel on site, and by the Environmental Services Office. Local support also comes from the Tracy Fish Facility (CVP, and from parent agency - US Bureau of Reclamation) personnel whose land borders the state's land to the south. Letters describing our project in Contra Costa County were sent to the County Board of Supervisors and to the County Board of Planning (3/99).

F. Compliance with Standard Terms and Conditions

Applicant complies with State and Federal contract terms to the best of our knowledge, as described in Attachments D and E.

G. Literature Cited

- Akielaszek, J. J., J. R. Moring, S. R. Chapman, and J. H. Dearborn. 1985. Experimental culture of young rainbow smelt, *Osmerus mordax*. *Trans Amer Fish Soc* 114: 596-603.
- Antonio, D.B., C. Swanson, J.J. Cech, Jr., R.C. Mager, S. Doroshov, and R.P. Hedrick. 2000. Prevalence of *Mycobacterium* in wild and captive delta smelt. *Calif. Fish Game* 86: 233-243.
- Baskerville-Bridges, B. 1999. Studies on rearing and early weaning of Atlantic cod (*Gadus morhua*) larvae onto commercial and experimental microparticulate diets. Ph.D. Dissertation. University of Maine, Orono, Maine, 162 pp.
- Baskerville-Bridges, B. and L.J. Kling, 2000. Larval culture of Atlantic Cod (*Gadus morhua*) at high stocking densities. *Aquaculture*. 181:61-69.
- Baskerville-Bridges, B., J. Lindberg, J. Van Eenennaam, and S. Doroshov. 2001. Progress and development of delta smelt culture: Year-end report 2000. Interagency Ecological Studies Program for the Sacramento-San Joaquin Estuary Newsletter, Winter 2001.
- Bell, M.V., R.S. Batty, J.R. Dick, K. Fretwell, J.C. Navarro and J.R. Sargent. 1995. Dietary deficiency of docosahexaenoic acid impairs vision at low light intensities in juvenile herring (*Clupea harengus* L.). *Lipids* 30: 373-476.
- Blaxter, J.H.S. 1981. The rearing of larval fish. Aquarium Systems. Hawkins, A.D. London, Academic Press: 304-323.
- Bligh, E. G. and W.J. Dyer. 1959. A rapid method of total lipid extraction and purification. *Can J Biochem Physiol* 37: 911-917.
- Boehloert, G.W. and J.B.Morgan. 1985. Turbidity enhances feeding abilities of larval pacific herring, *Clupea harengus pallasii*. *Hydrobiologia* 123:161-170.
- Henderson, R.J., M.V. Bell, and J.R. Sargent. 1985. The conversion of polyunsaturated fatty acids to prostaglandins by tissue homogenates of the turbot, *Scophthalmus maximus* (L.). *J Exp Mar Biol Ecol* 85: 93-99.
- Kanazawa, A. 1985. Essential fatty acids and lipid requirement of fish. In: Cowey, C.B., A.M. Mackie, and J.G. Bell (Eds.). Nutrition and Feeding in Fish. Academic Press., London, pp. 281-298.
- Kanazawa, A. 1997. Effects of docosahexaenoic acid and phospholipids on stress tolerance of fish. *Aquaculture* 155:129-134.
- Kashiwagi, M., T. Iwai, and A. N. G. Lopes. 1988. Effects of temperature and salinity on egg hatch of the pond smelt, *Hypomesus transpacificus*. *Bull Fac Bioresources*, Mie University 1: 7-13.
- Leger, P., D. A. Bengtson, K. L. Simpson, and P. Sorgeloos. 1986. The use and nutritional value of *Artemia* as a food source. *Oceanogr Mar Biol Ann Rev* 24:521-623.
- Lindberg, J. C. 1992. Development of delta smelt culture techniques. Report to California State Department of Water Resources, Sacramento. (August 1992).
- Lindberg, J. C. 1996. Delta smelt culture, State Water Site, 1995. Final report, California Department of Water Resources, Contract B-59776, 24 pp
- Lindberg J. C., Baskerville-Bridges B, Van Eenennaam J. P., Doroshov S. I. 1999. Development of Delta Smelt Culture Techniques; Year-end report 1999. Report to California State Department of Water Resources, Sacramento (DWR B-81581).

- Lindberg J. C., Baskerville-Bridges B, Doroshov S. I. 2000. Update on delta smelt culture with an emphasis on larval feeding behavior. Interagency Ecological Program for the Sacramento-San Joaquin Estuary Newsletter 13 (3): 45-49.
- Lindberg, J. C., and C. Marzola. 1993. Delta smelt in a newly-created flooded island in the Sacramento - San Joaquin Estuary, Spring 1993. Report to California State Department of Water Resources., Sacramento. (September 1993)
- Lindberg, J., R. Mager, B. Baskerville-Bridges, J. Kulczyk, J. Van Eenennaam, S. Doroshov. 1998. Delta smelt culture; year-end report 1998. Progress report, California Department of Water Resources, Sacramento; October 1998; Contract B-81355, 20 pp.
- Lindberg, J., R. Mager, B. Baskerville-Bridges, J. Kulczyk, J. Van Eenennaam, S. Doroshov. 1998a. Delta smelt culture, 1997. Progress report, California Department of Water Resources, Sacramento; March 1998; Contract B-80999, 24 pp.
- Mager, R. C. 1996. Gametogenesis, reproduction, and artificial propagation of delta smelt, *Hypomesus transpacificus*. Ph.D. Dissertation, University of California–Davis, 125 pp.
- Mager, R. C., S. I. Doroshov, and J. P. Van Eenennaam. 1996. Development of laboratory culture of delta smelt. Final report, Department of Water Resources, Fund title DWR B-59306, 65 pp.
- Moring, J. C. 1985. Smelt culture. Maine Fish and Wildlife Summer, pp 13-15.
- Moyle, P. B. 1976. "Inland Fishes of California". University of California Press, Berkeley.
- Moyle, P. B., D. E. Stevens, and L. W. Miller. 1992. Life history and status of delta smelt in the Sacramento–San Joaquin Estuary, California. *Trans Amer Fish Soc* 12: 67-77.
- Radtke, L.D. 1966. Distribution of smelt, juvenile sturgeon and starry flounder in the Sacramento-San Joaquin Delta. Pages 115-119, In S.L. Turner and D.W. Kelley, eds. Ecological Studies of the Sacramento-San Joaquin Estuary, Pt. 2. California Department of Fish and Game, *Fish Bull* 136.
- Reitan, K. I., J. R. Rainuzzo, G. Oie and Y. Olsen. 1994. Nutritional effects of algal addition in first feeding of turbot (*Scophthalmus maximus* L.) larvae. *Aquaculture* 118: 257-275.
- Robin, J.H., M.M. Le Gall, and H. Le Delliou. 1991. Comparison of three kinds of rotifer enrichments for turbot larval culture. *Fish Nutrition in Practice*, June 24-27, pp. 619-622.
- Rosenlund, G., Meslo, I., Rodsjo, R. and H. Torp. 1993. Large scale production of cod. Proceedings of the first International Conference on Fish Farming Technology, Trondheim, Norway, A.A. Balkema.
- Sargent J.R., L.A. McEvoy, J.G. Bell. 1997. Requirements, Presentation and sources of polyunsaturated fatty acids in marine fish. *Aquaculture* 155: 117-127.
- Sargent, J., L. Mc Evoy, A. Estevez, G. Bell, M. Bell, J. Henderson, and D. Tocher. 1999. Lipid nutrition of marine fish during early development: current status and future directions. *Aquaculture* 179:217-229.
- Shapiro, S.S., and M.B. Wilk. 1965. An Analysis of Variance Test for Normality (complete samples). *Biometrika* 52: 591-611.
- Snedecor, G.W. and W.G. Cochran. 1993. Levene's test for homogeneity of variance. In: Statistical methods, 8th edition. Iowa State University Press, Ames.
- Southgate P.C., D.C. Lou. 1995. Improving the n-3 HUFA composition of *Artemia* using microcapsules containing marine oils. *Aquaculture* 134: 91-99.
- Sukhija, P.S. and D.L. Palmquist. 1988. Rapid method for determination of total fatty acid content and composition of feedstuffs and feces. *J. Agric. Food Chem*, 36: 1202.
- Swanson, C., R.C. Mager, S.I. Doroshov, and J.J. Cech, Jr. 1996. Use of salts, anesthetics, and polymers to minimize handling and transport mortality in delta smelt. *Trans. Amer. Fish. Soc.* 125: 326-332.

- Swanson, C., P.S. Young and J. J. Cech, Jr. 1998. Swimming performance of delta smelt: maximum performance, and behavioral and kinematic limitations on swimming at submaximal velocities. *J. Exp. Biol.* 201: 333-345.
- Takeuchi, T. 1997. Essential fatty acid requirements of aquatic animals with emphasis on fish larvae and fingerlings. *Rev Fish Sci* 5:1-25.
- Tamaru, C.S., R. Murashige, C. Lee, H. Ako, and V. Sato. 1993. Rotifers fed various diets of bakers yeast and / or *Nannochloropsis ocolata* and their effect on the growth and survival of striped mullet (*Mugil cephalus*) and milkfish (*Chanos chanos*) larvae. *Aquaculture* 110: 361-372.
- Tuncer, H. and R. M. Harrell. 1992. Essential fatty acid nutrition of larval striped bass (*Morone saxatilis*) and palmetto bass (*M. saxatilis* x *M. chrysops*). *Aquaculture* 101:105-121.
- Wang, J.C.S. 1986. Fishes of the Sacramento-San Joaquin Estuary and adjacent waters, California: a guide to the early life histories. Interagency Ecological Study Program, Sacramento-San Joaquin Estuary Technical Report 9, Sacramento, California.
- Wang, J.C.S. 1991. Early life stages and early life history of the delta smelt, *Hypomesus transpacificus*, in the Sacramento-San Joaquin Estuary, with comparison of early life stages of the longfin smelt, *Spirinchus thaleichthys*. Interagency Ecological Study Program for the Sacramento-San Joaquin Estuary. Technical Report 28. 52 pp.
- Watanabe, T. 1982. Lipid nutrition in fish. *Comp Biochem Physiol* 73B: 3-15.
- Watanabe, T. C. Kitajima and S. Fujita. 1983. Nutritional value of live organisms used in Japan for mass propagation of fish: a review. *Aquaculture* 34:115-143.

Attachments:

Attachment A – Letter of support. Pledge of matching funds (\$2500/year) for site-maintenance costs and repairs from Department of Water Resources.