Project Purpose

Steelhead (*Oncorhynchus mykiss*) exhibit a remarkable diversity of life histories. At the end of their first year of life, steelhead follow three possible trajectories: smoltification and emigration to the ocean, remaining in freshwater as an immature parr, or precocious maturation. Following the first year, multiple pathways are again possible, such as emigration or continued freshwater residence. Some individuals never emigrate, thus becoming de facto rainbow trout, the non-anadromous form of *O. mykiss*. In contrast to other Pacific salmonids, steelhead are iteroparous and may spawn over several years, returning to the ocean between each spawning.

This complexity of life histories makes understanding population dynamics and environmental effects on steelhead very challenging. Fisheries science typically focuses on analysis of annual cohorts (year classes) and how they fare as they progress through their life span. However, steelhead progeny produced in a single year will very likely split into many trajectories, inhibiting the tracking of birth cohorts. The returning run of adults will be comprised of fish of different ages that have spent varying periods in freshwater and seawater, making linkages of abundance to environmental conditions (i.e., flow rates in the birth year) problematic. An improved understanding of how individuals arrive at a particular life history pathway will greatly improve our ability to monitor and predict effects of changing environments on steelhead populations. The diversity of life histories in *O. mykiss* and the apparent high plasticity in phenotypic expression of these life histories theoretically has developed as a bet-hedging strategy promoting persistence of populations in a highly variable environment (Mangel and Clark 1988, Thorpe et al 1998). However, despite this flexibility steelhead populations continue to decline in abundance. In California, all Evolutionarily Significant Units (ESUs) of steelhead are currently listed as either threatened or endangered under the Endangered Species Act, with the exception of the Klamath Mountain ESU. In the Central Valley, declines are clearly linked to water management (McEwan 2001). Dam construction throughout the Sacramento and San Joaquin watersheds has eliminated approximately 82% of historical spawning and rearing habitat (Yoshiyama et al. 1996 in McEwan 2001) and constrained steelhead to spawning in lower elevations. However, the continuing decline of steelhead populations 40+ years after the major period of dam construction suggests that other factors are contributing to population losses. McEwan (2001) suggests three possible stressors that continue to impact steelhead populations. First, increasing water exports by State Water Project and Central Valley Project pumping facilities contribute to further degradation of habitat quality in the remaining spawning and rearing locations accessible to steelhead. Second, density-dependent growth responses associated with reduced populations may increase the proportion of steelhead that residualize, thereby reducing the number of individuals that adopt an anadromous life history. Third, metapopulation dynamics (Cooper and Mangel 1999) interacting with habitat loss may be contributing to continuing abundance declines. In a dynamic system such as the Central Valley, extirpation of small populations that occasionally replenish large populations or reduction of large source populations that sustain smaller sink populations may lead to overall declines in total abundance of the ESU.

This project will focus on determining the environmental conditions that underlie the three pathways available to steelhead in their first year. Prior studies on juvenile salmonids have demonstrated the influence of growth and lipid accumulation, interacting with genetic factors, on timing of life history transitions. Fast growth is typically associated with smoltification and emigration at age 1. Fish with poor growth typically remain in freshwater for at least another year. Precocious maturation may occur in fish that are able to both grow quickly and accumulate high levels of lipids. Once initiated, these trajectories may be fixed, with limited opportunity to switch if conditions change. These varying strategies presumably enhance the persistence of steelhead populations by allowing flexibility in individual responses. However, major shifts in the environment can result in a high proportion of fish that have entered an inappropriate pathway. The overall hypothesis is that water flow levels and the temporal pattern of water delivery have a major impact on growth opportunity and life history expression in age-0 steelhead. Alteration of water flow patterns potentially disrupts the natural adaptive responses of juvenile steelhead, resulting in reduced survival as fish make crucial mistakes in selected life history trajectories.

Grantee proposes to investigate a series of questions concerning the factors determining the timing of emigration in juvenile steelhead and the role of environment, particularly water flows, in shaping this behavior. These include:

1) How does one modify the conceptual framework of Thorpe et al. (1998), which is described below, to account for the relatively unique biogeography of California?
2) What predictions emerge from this modified framework?
3) What information do size distributions and growth rate provide about the probability of migration?
4) How do coastal steelhead differ from Central Valley steelhead in these regards? Is this evidence for local adaptation or developmental plasticity?
5) What are the implications of these results for the effects of water flows on steelhead survival and migration?

Grantee will address these questions from approaches of field studies, lab experiments, and modeling.

Background

Life-history strategies are the means by which organisms achieve successful reproduction in varying environments. It is common to approach the study of life histories from two perspectives. With the ultimate or functional perspective the goal is measuring fitness in terms of the number of descendants or the number of genes in future generations. Ultimate considerations are post-hoc because they attribute fitness to individuals at the completion of the particular phase of the life cycle and do not attempt to characterize the mechanisms that animals use to achieve the optimum life history pattern. The proximate or physiological perspective focuses on the developmental pathways that are the consequences of individual responses to the opportunities offered by the environment.

Salmonid life histories are characterized by two developmental conversions: smolt metamorphosis and maturation. Fish generally spawn in autumn or winter, and first feeding of the offspring is in the following spring (April/May). In Atlantic salmon *Salmo salar*, determination of whether an individual will undergo smolt metamorphosis the following spring (at age 1) occurs soon after midsummer (Thorpe et al 1998), but this timing is not known for any other salmonids. Regarding maturation, the key observation (Policansky 1983) is that fish with access to abundant resources and stable conditions for development mature as soon as they are able to do so. Germinal tissue differentiates very early and investment in gonadal growth begins during the embryo stage. Both males and females have the potential to mature at age 0+ (typically described as 'precocious' maturation), but it is more commonly expressed in males. (However, in cases of landlocked populations, both males and females mature without migration and at small size (13-15 cm); see Behnke (2002) pg 243 for an example.)

Thorpe et al. (1998, also see Mangel 1994) developed an approach to understanding the life history of Atlantic salmon that combines ultimate (evolutionary) and proximate (physiological) considerations. In this approach, which represents the culmination of a 20-year investigation begun by John Thorpe in the late 1970s, fish life histories are regulated by inhibition (of smolt metamorphosis and of maturation) or the release of inhibition at certain points in the calendar year (decision windows). Whether or not inhibition is released depends on projections of physiological state into the future, based on current information (Figure 1) This framework has been successfully applied to Atlantic salmon in both North America and Europe, and to chinook salmon *Oncorhynchus tschawytscha* in British Columbia and Washington respectively (W. Dickhoff, presentation to the Recovery Science Review Panel, 30 Aug 04, using data from Beckman et al (2003) and Larsen et al (2004).

Experimental studies have elucidated the importance of early growth rates and the position of an individual along an expected growth trajectory in shaping the probability of early maturation and/or residualization, as well as the timing of emigration for anadromous individuals, with the following general sequence for Atlantic salmon (outlined in Thorpe et al. 1998). In the spring, a maturation switch occurs in which current energetic state (lipid reserves) and the rate of change in state are compared with a genetically determined maturation threshold. Individuals exceeding the threshold adopt a pathway of early maturation. Near the end of their first summer, juveniles enter a second decision window of assessment of internal state (body size) and its rate of change (i.e. growth rate). If the threshold is exceeded, the individual adopts a migratory pathway and continues to feed and have high activity levels during the winter in preparation for smolting in the spring. If the threshold is not attained, the individual adopts a non-migratory pathway, reducing its activity and thus its vulnerability during the winter. Following this decision, the population in general will divide into two size modes as fish on the emigrating pathway continue to increase in size relative to the non-emigrating mode. Fish that have adopted the maturation pathway do not initiate the emigration process. As a consequence of these life history decisions, fast growing age-0 individuals with high lipid accumulation may mature as parr, individuals growing at moderately fast rates are likely to undergo smolt transformation in the spring and emigrate at age 1, and the slowest growers remain in the stream for another year, again entering the maturation and emigration decision windows at their respective times.

The relevance of this framework for steelhead populations has not been previously assessed. There is evidence of at least moderate heritability of early maturation, smolting timing, and growth in steelhead (Thrower et al. in press), providing support for the concept of varying genetic thresholds for life history transitions. Grantee will focus on the environmental factors underlying the expression of different pathways. Although steelhead and Atlantic salmon share a similar repertoire of life history strategies and variability in those strategies, we believe the situation of steelhead in California is different. Atlantic salmon on a life history pathway of delaying emigration until age 2 will reduce activity and feeding in winter, presumably as a means of reducing predation risk during a period of poor growth opportunity due to cold temperatures and low food availability. A similar response is evident in steelhead populations of Vancouver Island, where fish reduce growth rates in winter even when provided with elevated temperatures and unlimited food (Johnsson et al. 1993). Johnsson et al. (1993) suggested that high feeding activity in winter was maladaptive due to the associated risk and costs. However, for some California populations it is winter, rather than summer, that is the good growing season and summer may be the harshest season, from the perspective of growth (Figure 2). Thus, the timing of life history decisions in southern populations of steelhead is not necessarily comparable to Atlantic salmon, despite their similarity of plasticity in life histories. Models developed for Atlantic salmon may be more applicable in current form to northern populations of steelhead, which experience much harsher winter conditions. Furthermore, steelhead populations within California may experience very different biogeographic conditions. For example, preliminary studies of the four systems to be examined in this study have found extreme differences in early growth rates and likely proportions of age-1 emigrants between the central coast and Central Valley. Development of appropriate life history models will require detailed comparisons of contrasts between the two regions.

The central role of early growth in determining life history trajectories provides a tractable means of generating the empirical data necessary to develop life cycle models for California steelhead. Grantee's overall focus in the proposed research is to understand the mechanisms underlying variability in potential growth rates and how different stressors impact growth and consequent life history pathways. As outlined below, Grantee believes management decisions affecting the growth environment, including habitat availability, food delivery via drift, and physical conditions such as temperature, can dramatically alter the natural distribution of life history patterns exhibited in steelhead populations. Development of appropriate, well-supported life history models for steelhead will be useful for both improved management of water resources for threatened populations and improved predictive capabilities for future environmental impacts such as global warming and drought regimes.

Most of the knowledge of steelhead ecology has been derived from northern populations. However, local adaptation of steelhead appears to be extensive; high levels of genetic differentiation among stream systems has been observed for both the Central Valley (Nielsen et al. 2003) and along the entire coast of California (Garza 2004). Thus, one expects to see many contrasts between California steelhead and northern residents, as well as contrasts among streams within California.

A notable exception in which California steelhead have been intensively studied is the analysis of Waddell Creek fish in the 1930s and 1940s, reported by Shapovalov and Taft (1954). Their detailed account provides invaluable data on variability in the proportions of different life history pathways over a ten year time period in a relatively pristine system on the central coast. For example, based on scale analyses of returning adults, only approximately 10% of individuals surviving to adulthood migrated to sea in their first year, with most survivors emigrating at age 2 (69%) or age 3 (19%) (Figure 3). Although their results from a downstream migrant trap did not directly measure emigration (many fish caught moving downstream may actually have remained in the creek another year), they are suggestive of a proportion higher than 10% for fish entering the ocean at age 1. Mortality rates of salmonids in general appear to increase immediately after ocean entry and are size-selective, with smaller fish less likely to survive (Ward and Slaney 1988, Holtby et al. 1990). Because size at emigration increases with freshwater age, individuals emigrating at older ages are more likely to survive the initial period of ocean residence (Ward and Slaney 1988). In Central Valley populations such as the American and Mokelumne rivers, growth during the first year is markedly higher than in central coast populations, likely resulting in a high proportion of fish emigrating at age 1 (Titus, unpublished data). The proportion of age-1 emigrants prior to dam construction and extensive habitat alteration in these systems is unknown. Clearly a significant trade-off is evident between survival to age 2+ in freshwater and size-selective survival upon ocean entry. Individuals remaining in the stream for a second year of growth must survive through the highly variable flow conditions present in winter. Likewise, emigrating fish must survive the initial gauntlet of predators awaiting them as they enter marine habitats.

Preliminary results:

In field collections of age-0 steelhead in Soquel Creek, Grantee has not observed the bimodal size distributions in fall/winter that seem common in Atlantic salmon. Thus, this population does not appear to split into two life history trajectories prior to winter. Based on Grantee's direct measurements of growth rates (Figure 2) and indirect estimates based on size-frequency distributions, growth in Soquel Creek is inhibited in summer but increases during winter/spring, opposite to the pattern typical of more northern populations of steelhead (Johnsson et al. 2003) and the Atlantic salmon modeled by Thorpe et al. (1998). Similar reversed seasonal patterns of growth have been observed in Scott Creek (Sean Hayes, NMFS, unpublished data).
In academic year 2002-03, (Mangel and Sogard) sponsored a senior thesis student who conducted preliminary studies in the lab examining some of the predictions of the life history models developed for Atlantic salmon by Thorpe et al (1998). On the theoretical side, the framework of Thorpe et al (1998) was modified, based on a literature review (Figure 4). The results (Atcheson 2003) demonstrate the expected size relationship with spring Na^+K^+ATPase levels but unexpected patterns of behavior of non-emigrating fish (Figure 5). Fish that were initially large in size (>130 mm in January) had significantly higher ATPase levels in April than smaller fish (< 100 mm in January), suggesting a higher probability of smoltification and emigration. One interesting result was that low ration levels appeared to reverse the life history decision for some large fish. For large fish held on high rations, 76% had ATPase levels > 3.0, our index of seawater readiness, in contrast to 43% for large fish held on low rations. None of the small fish had elevated levels of the gill enzyme. General behavioral activity during the winter was expected to be lower in small fish than in large fish based on the assumption that fish selecting a non-emigration pathway would reduce activity and aggression in winter as a risk avoidance strategy. However, Atcheson (2003) found that small fish had higher activity levels than large fish, suggesting continued searching for prey and no evidence of the near torpor observed in non-emigrating Atlantic salmon.

Overall, these results emphasize the likelihood that Grantee will need to modify the Atlantic salmon models for southern steelhead populations.

Goals, Objectives and Hypotheses

The overall goal of our proposed research is to extend the framework developed by Thorpe et al. (1998) for Atlantic salmon to California steelhead, using field and lab studies to derive appropriate empirical data and modeling to modify and refine the theory for Central Valley and central coast populations. Grantee assumes that smoltification and subsequent emigration depend on the physiological state of an individual at some time in advance of the actual initiation of the required physiological transformations; once this decision is made an individual fish is committed to a particular developmental pathway. In Atlantic salmon and potentially in northern populations of steelhead, this decision window is in the late summer or early fall, and greatly influences winter growth and behavior for individuals depending on the selected life history trajectory (emigrating in the spring or staying in the stream for another summer). For southern populations, Grantee hypothesizes that local adaptation results in a shift in timing of the emigration decision window. Shapovalov and Taft (1954) suggested that on Waddell Creek growth rates of juvenile steelhead declined in late summer due to high water temperatures and low flows, remained low through the winter flooding period, then rapidly increased in spring as temperatures increased and food delivery remained high. Recent tagging studies on the central coast (Scott and Soquel creeks) similarly demonstrate very slow growth rates in summer and fall, and accelerated growth in the winter/spring, when temperatures are mild and food availability presumably increases due to higher flows. Thus, the decision to emigrate likely occurs much closer to the actual start of transformation processes compared to populations that undergo harsh winters with minimal growth potential.

Growth rates of age-0 fishes are presumably driven by the interactions of several factors, including density of conspecifics and potential competitors, water temperature, genetic differences in growth capacity, and food availability. Grantee hypothesizes that variability in water flow is a major determinant of variability in food availability through the impact on delivery of insect prey, resulting in a direct relationship of flow with growth potential. Drifting terrestrial invertebrates are a rich and preferred prey type for juvenile steelhead, and increase in abundance as water velocity increases (Smith and Li 1983). Poff and Ward (1989) emphasize the importance of stability in flow patterns for the productivity of stream fish. Seelbach (1993), for example, found that stable flows were associated with high survival of steelhead through their first winter. Management decisions that alter the natural patterns of flow rates thereby have the potential to alter growth rates and, consequently, life history pathways of steelhead. For example, if the emigration decision window occurs prior to a period of normally high flow, disruption of food delivery may have a negative impact on fish that have adopted an early emigration trajectory. Grantees do not know of any data linking these growth components on a seasonal basis and thereby addressing the issue of growth potential for steelhead in different seasons. Central Valley and central coast steelhead populations are likely to differ in life history patterns as a consequence of different environmental conditions in the two regions.

Hypotheses to be Tested.

Work in the laboratory will be guided by the following hypotheses:

H1: Life history pathways of early maturation, emigration and non-emigration are established during specific time periods (decision windows) in the first year of life.

H2: Growth rates and body condition during the decision windows establish the life history pathway selected by an individual fish.

H3: Timing of the decision windows differs among populations.

H4: Altered temperature regimes during the decision window influence the proportion of fish adopting different life history pathways.

H5: Behavior during winter months varies as a function of life history pathway and temperature.

The field work will be guided by the following hypotheses

H6: Age-0 steelhead exhibit bimodal size distributions in the winter, reflecting a split into emigrating and non-emigrating trajectories.

H7: Availability of insect prey differs among stream systems and seasonally.

H8: Prey delivery via drift is a function of flow rate.

H9: Growth of age-0 steelhead is an interactive function of temperature, prey density, and competitor density.

Models will be developed concomitantly with the laboratory and field work, so that the empirical studies provide insight for formulating the models and data for parameterizing the models and so that the models provide detailed predictions that can be tested. Examples of this interplay between empirical and modeling work can be found in Clark and Mangel (2000), Chapters 4 and 5.

Project Description:

Plan of work

Lab component

Laboratory experiments will address the following questions:
1. When does the decision window for emigration occur?
2. Does the timing of the decision window vary among populations?
3. Does variability in winter temperatures modify the emigration decision?
4. Does fish behavior in winter vary as a function of life history trajectory?
5. Is early maturation of parr controlled by a similar decision window?

Grantee will examine these processes in two populations of steelhead representing two contrasting environmental regimes. Scott Creek, on the central California coast, typifies an unmodified coastal system with low summer flows leading to poor summer growth conditions and mild winters when growth potential improves. A conservation hatchery operated by the Monterey Bay Salmon and Trout Project on a tributary of Scott Creek propagates steelhead for release in Scott Creek and a limited number of nearby creeks with depleted steelhead runs. Only wild fish returning to Scott Creek are used as spawners, ensuring minimal genetic alteration and domestication (sensu Price 2002) in the progeny. In the Central Valley, Coleman National Fish Hatchery propagates steelhead on Battle Creek. Genetic studies have concluded that these fish are closely related to fish from the upper Sacramento River and its tributaries (Nielsen et al. 2003), and thus are representative of populations in the northernmost Central Valley. These populations presumably experience the coldest winters within the Central Valley ESU, although still comparably mild compared to populations in the northern part of the species range. We predict that these two populations will differ in the timing of life history decisions as a function of their local adaptation to contrasting environmental regimes. As a consequence, behavioral responses to different winter temperatures are also likely to vary.

Standard egg and sperm removal and incubation procedures will be conducted by hatchery staff at each location, using aggregated males and females to ensure a varied mix of lineages within each population. Under a cooperative agreement with each hatchery, we will collect progeny just prior to the first feeding stage and transport them to our aquarium facilities at the NMFS Santa Cruz lab for subsequent growth experiments. The fry will likely be ready for transfer some time in May. This timing will allow us to have full control over the food availability experienced by different treatments of fish. In the laboratory, fish will be randomly assigned in groups of 20 fish to one of four treatments, with two replicate tanks for each. The two source populations will be maintained separately throughout the experiment. Treatments will vary the timing of a period of rapid growth, simulating fluctuating exposure to high food availability in a stream. All fish will be constrained to a moderate but continual growth rate via limited rations. Then, for a two-month period, the fish will receive unlimited rations, allowing rapid increases in growth. Prior studies in our lab have demonstrated a marked capacity in the Scott Creek population to capitalize on these feeding opportunities with accelerated growth rates (exceeding 1 mm d^{-1}), and Grantee expects the Coleman population to respond similarly. Following the two-month fast growth opportunity the fish will be returned to restricted rations for the remainder of the experiment. The four treatments will be comprised of enhanced growth in August/September, October/November, December/March, or February/March. Grantee expects the decision window to be limited to one of these periods, and fish experiencing rapid growth during this window will adopt an emigration strategy, with full seawater readiness in April/May.

Fish will be maintained under a natural photoperiod using the lab's automated lighting system, which adjusts light exposure to simulate dawn and dusk transitions and the daily progression of seasonality in photoperiod. Grantee will use a photoperiod intermediate to Scott Creek and Battle Creek latitudes. In year one temperatures will be varied on a daily basis to match typical seasonal patterns of Scott Creek (mild winter temperatures). In year two, the experiment will be repeated using a temperature regime comparable to the average pattern on Battle Creek (colder temperatures). Results of the two experiments will provide additional information on the effects of temperature on the decision window and behavior patterns of the two populations. Our objective is to match the natural physical conditions of the two systems but manipulate the timing of growth opportunities. Food provided will be a standard hatchery pellet, BioDiet, with pellet sizes continually increased as fish grow into appropriate sizes.

Growth will be monitored by measuring and weighing all fish on a monthly basis during restricted feeding and bi-weekly during the period of high rations. Rations will be adjusted accordingly to maintain the desired growth trajectories. Behavior of fish in each treatment will be examined in late winter and early spring to determine any differences associated with life history trajectory and temperature regime. Pairs of fish from each tank will be place in circular tanks with overhead video cameras and filmed for 4-hour periods during the day and at night. A PVC shelter will be placed in the middle of the tank. General motor activity, shelter use, and aggressive interactions will be quantified and compared among treatments and between day and night. Our preliminary results using these methods with the Scott Creek population have suggested that fish selecting a non-emigration pathway (at age 1) do not reduce activity in the winter (Fig. 5), in contrast to results for Atlantic salmon (Metcalfe 1998). However, the northern California population from Coleman Hatchery, which typically has much colder winters, may be more likely to behave similarly to Atlantic salmon.

Grantee expects fish that have selected a life history trajectory leading to emigration in the spring to exhibit changes in physiological state as they prepare for smolt transformation. Smolting fish typically have lower condition indices (Fulton's K), increased silvering, increased metabolic rates, and increased activity levels compared to non-smolting fish (Folmar and Dickhoff 1980). We will use a combined index of these factors to identify individuals likely to smolt in the spring. Fulton's K will be calculated at each measurement interval. Fish will be photographed periodically throughout the experiments to assess changes in coloration. Grantee will monitor physiological condition of the fish in our experiments by measuring swimming capability and metabolic rate in a swim tunnel respirometer once a month, using a representative sample of fish from each population and treatment. Grantee predicts that metabolic rates will increase for individuals that have entered the emigration trajectory (Forseth et al. 1999). The Santa Cruz Lab has recently acquired a Loligo model 10 tunnel respirometer, which will be available for these tests. Fish will be tested individually in the tunnel, with respiration measured after 30 min of acclimation at a moderate swimming speed of 3 body lengths s^{-1}. A series of four randomly selected fish from each of the 8 population/ration treatments will be tested each month.

In late April, the expected time of emigration for all fish in the emigration life history pathway, gill filament samples will be collected for Na+K+ATPase enzyme analysis, using the methods of McCormick (1993). After a one week recovery period from gill sampling, fish will receive a seawater challenge to assay their ability to osmoregulate in full strength seawater. Fish will be transferred directly from freshwater to seawater at a salinity of 32 ppt and left for 48 hours. Individuals will be scored as smolts or non-smolts depending on their ability to survive and maintain equilibrium in seawater. Seawater readiness will be compared with Na+K+ATPase levels to verify the ability of the enzyme assay to accurately predict smoltification. Seawater readiness will be equated with a decision to emigrate at age-0 and compared across factors of population source and ration treatment. Grantee predicts that fish exposed to high food availability during the appropriate decision window will exhibit a greater likelihood of seawater readiness than fish experiencing poor growth conditions during this decision window. Fish in the latter groups will presumably select a non-emigration pathway and will fare poorly in seawater challenges. At the conclusion of the experiment all fish will be sacrificed and dissected to determine early maturity. Mature parr are not expected to exhibit characters associated with smolting and will be excluded from treatment comparisons for determination of the decision window.

In year 3 Grantee will focus on factors leading to early maturation of parr. Pre-feeding fry will be obtained from the two respective hatcheries and held in laboratory tanks as in years 1 and 2. We hypothesize that early maturation is induced by rapid growth rates during a decision window that precedes the emigration decision window (Thorpe et al. 1998). Manipulation of food availability has been demonstrated to influence early maturation in brown trout *Salmo trutta* (Pirhonen and Forsman 1999). Accordingly, fish will be maintained on ad libitum diets up to a selected time period, then fed a restricted ration for the remainder of the experiment. Treatments will shift from ad libitum to restricted diets at the end of July, September, or November. A control group will receive continuous ad libitum rations. If early maturation is turned on by a developmental switch during a specific time window, we predict that those individuals will mature in the early spring, regardless of their feeding success subsequent to the decision window. At the end of February, in the middle of the normal steelhead spawning period, all fish will be sacrificed and dissected to determine gender and stage of maturity.

Field component

Field studies will focus on evaluating seasonal patterns in growth potential for steelhead populations in four different watersheds, two in the Central Valley and two on the central California coast. Central Valley systems will consist of the American River, a tributary of the Sacramento, and the Mokelumne, which flows into the Sacramento-San Joaquin delta system. On the central coast, we will conduct studies in Soquel and Scott creeks, both of which flow directly into the Pacific Ocean, passing through small estuaries/lagoons that are typically blocked from ocean access during summer months due to sandbars. Prior research has indicated vastly different growth rates of age-0 steelhead at the regional level, but similarities between the two streams within each region. Within each stream system we will establish four 100 m reaches for sampling. Our focus here is not to establish index reaches that will be representative of a particular stream or watershed. Instead, our objective will be to select sampling sites that cover a range of conditions and habitats currently being used by age-0 *O. mykiss* in each system. Our goal is to examine the variability in growth potential as a function of different environmental conditions and to document the breadth of responses exhibited in natural systems by young steelhead.

Physical parameters of temperature and flow will be monitored on a continuous basis. Temperatures will be recorded using Onset TidBits placed in housings at each site. Water flow data will be obtained from USGS records available for 3 locations on the American River and 1 location on Soquel Creek. Flows at the 16 individual sampling sites will be periodically calibrated against USGS values with a hand held stream flow meter.

The diet of age-0 and age-1 steelhead is comprised primarily of insect larval and adult stages (Merz & Vanicek 1996). Grantee expects availability of these prey to be highly variable depending on seasonal cycles of production and fluctuating delivery via drift. Documenting seasonal patterns of food abundance will be a key component of our field research and one of the most time intensive efforts. It is also a vital information gap, as relatively little effort has been expended to address the annual variability of prey in natural steelhead waters (or any salmonid, for that matter). Insect prey for young steelhead is derived from in situ (benthic) sources and delivered via drift from upstream sources. To sample these two components Grantee will use benthic samplers (Surber or modified Hess) and stream drift nets, deployed according to standard protocols. Collected organisms will be identified to broad taxa and categorized as potential prey for either age-0 or age-1 steelhead. Dr. Joe Merz (EBMUD) has graciously agreed to assist us with training in identification of stream invertebrates, and processing of benthic and stream samples will take place in his laboratory. Sampling will be conducted at the 16 sites on a seasonal basis (four time periods per year). In addition, Grantee will examine variability in food availability on a diel basis by sampling across a 48 hour time period at one site in each stream and each season. To provide more detailed comparisons of prey abundance with flow rates, we will conduct sampling on an opportunistic basis as water flows vary within a season. Grantee expects this variability to be expressed primarily during fall and winter in association with rainstorms. Standard parametric statistics will be used to evaluate differences in food abundance across regions, streams within regions, and as a function of season and flow rate.

Fish density estimates, steelhead size distributions, and the species composition of potential predators and competitors will be determined using snorkeling observations at two sites within each stream. Standard snorkeling transects will be conducted along the 100 m reach during four time periods (spring, summer, fall, winter) each year. Abundance and size distribution of *O. mykiss* and other fish species present in each reach will be recorded. Roni and Fayram (2000) found that nighttime observations provided estimates comparable to electrofishing for small salmonids, whereas daytime snorkeling underestimated abundance, particularly at low water temperatures (small fish are more likely to feed at night and hide during the daytime). Grantee will conduct preliminary tests to determine if night snorkeling counts exceed day counts and plan further sampling accordingly. When low flow rates restrict snorkeling capabilities, as is likely in summer for the coastal streams, density estimates will be supplemented with electrofishing. A series of depletion sampling with 3 passes of the shocker will be completed when necessary to complement snorkeling efforts. For Central Valley systems, low water clarity may inhibit snorkeling surveys. In this case, the fish community will be sampled with seines and trawls to provide a coarse estimate of fish density and potential competitors and predators.

Growth rates will be monitored indirectly using the time series of length-frequency estimates derived from density sampling and directly using mark and recapture methods at the same eight sites. In late spring/early summer of each year, steelhead will be caught via seining or electroshock fishing. Fish < 65 mm FL (all presumed to be age-0) will be tagged with different colors of fluorescent elastomer (Northwest Marine Technology) in a series of 10 mm size classes. Fish > 65 mm will be tagged with PIT (Passive Induced Transponder, Allflex corporation) tags, which provide a unique identifier for each individual. Subsequent recaptures at each sampling site during the four seasonal efforts will provide data on growth rates within each season. As fish attain the 65 mm target, they will be PIT tagged, providing increasing sample sizes for tracking of individual growth rates. These methods have proven to be highly effective in evaluating growth in one of our systems, Soquel Creek (Fig. 2).

All of the field studies will be conducted across the full 3 years of the project. Grantee expects to encounter a broad range of environmental conditions across this time, providing the opportunity to evaluate the maximum variability in growth potential experienced by natural steelhead populations in the four watersheds. This intensive effort will provide a wealth of data on the physical environment, food availability, and fish density across seasons in four different stream systems. Growth rate measurements will elucidate the interactive effects of these factors and any population differences potentially attributable to local adaptation. Grantee expects to see markedly different results between the two broad regions of Central Valley and central coast. All results will feed directly into the modeling component of this project as they become available.

Modeling component:

The modeling component of our work has three main goals:

1) To modify the theory of Thorpe et al. (1998) to account for the relatively unique biogeography (at least for salmonids) of California, including frequency dependence.

2) To apply this theory to steelhead trout by appropriate parameterization of the timing of the life history windows and the estimation of the thresholds from a combination of literature review and connection to laboratory and field experiments conducted in the course of this project.

3) To explore the fitness consequences of steelhead developmental plasticity (sensu Bateson et al. 2004), so the consequences of individual life history patterns can be scaled to the population.

The theory of Thorpe et al. (1998) recognizes that salmonid life histories are characterized by two primary developmental conversions (Smith-Gill 1983), smolting and sexual maturation. In Atlantic salmon, although smolting occurs in the spring, the critical decision at which physiological decisions are taken occurs in mid-summer. The framework is one in which a fish is assumed to project a measure of size and condition at the time of smolting with a genetically determined threshold (Figure 1). If the projection is greater than the threshold, then the fish commits to a developmental pathway leading to smolting; otherwise it is resident in freshwater another year. The timing of this process in Atlantic salmon was confirmed in independent studies using appetite (Metcalfe et al 1986, Huntingford et al. 1988), body growth (Thorpe et al. 1989), and otoliths (Wright et al. 1990.). This process leads to diversity in life histories, and generates the great diversity in life histories of Atlantic salmon. Similar diversity is seen in the life history of steelhead trout (Shapovalov and Taft 1954). Furthermore, this theoretical framework leads us to understand that each salmonid migration

Egg ---> Gravel redd
Redd --> Stream bed
Stream bed --> Feeding territory
Feeding territory --> Downstream
Freshwater ---> Sea

is an abandonment of a habitat that can no longer satisfy the needs of the fish. That is, steelhead trout migrate because the freshwater environment is insufficient for them, not because the ocean is good for them. This is confirmed, for example, in studies of growth rates of steelhead and rainbow trout (Johnsson et al 1993).

To develop a computationally practicable description of the developmental switches, proximate mechanism, and fitness consequences requires the following components:

a) <u>Development of a growth model that will characterize the anticipated and actual</u>

trajectories. Grantee believes that the initial framework can be developed from existing models (Weatherley and Gill 1981, Weatherley and Gill 1995, West et al. 2001, Forseth et al. 2001, Elliott et al. 1996, Elliott 1994, Hill and Grossman 1993), parameterized initially with the data in Shapovalov and Taft (1954) and refined through our own empirical work. These models are typically of the form:

$$\frac{dW}{dt} = q\Phi(T(t))W^a - \alpha e^{0.0717T(t)}W^b$$

where W(t) is mass at time t, q is maximum food assimilation efficiency and related to the abundance of food, $\Phi(T(t))$ characterizes the temperature dependence of food assimilation, a and b are allometric parameters (a=2/3, b=1 corresponds to von Bertalanffy growth), and α is a measure of standard metabolic rate. We will make such general models specific to steelhead, separating coastal and Central Valley fish.

b) A measure of relative performance. This measure compares the actual and anticipated performance over the decision window used by the fish. For steelhead, we do not know the timing or the length of the window, and the empirical work will determine those. Given those, a natural measure of relative performance is the relative specific growth rate (e.g. actual divided by anticipated). A measure of growth potential is $\frac{q}{\alpha}$, and in the salmonids, they have coefficient of variation about 30% (Jobling 1994). Grantee will assume that length is given by an allometric relationship with weight, determined by our empirical studies.

c) Projection to the time of smolting. Grantee will iterate the growth model forwards in time, from the end of the assessment period to the time of smolting. Once the growth model is set, this is straightforward.

d) Comparison of the projected size at the time of smolting and the genetically determined threshold. Grantee will use the empirical work, plus literature reviews, to determine information about the distribution of the thresholds for smolt metamorphosis. This is a relatively complicated inverse problem, in which we observe the distribution of growth rates and life history pattern and infer the thresholds.

e) Computation of fitness. By fitness, Grantee means expected reproductive success, which has components of survival and reproduction. Salmonids, and fish in general, have mortality rates with size dependent and size independent components and the size dependent component scales inversely with mass to approximately the 1/3 (Lorenzen 1996); Grantee will use mortality $m(W) = m_0 + m_1W^{-0.37}$, where m(W) is the mortality rate of a fish of mass W and m_0 and m_1 are the size independent and size dependent components of mortality. By combining the growth and survival models, and comparing them with survival data, Grantee will be able to estimate these parameters. We will follow the procedure of (Mangel 1996) for computation of female fitness from smolt size; this procedure relies on a combination of literature based parameters and our empirical work. The situation with males is a little bit more complicated, because it is well known that in the salmonids males have two main reproductive strategies

(Gross 1985, Gross 1991, Gross and Repka 1995, Aubin-Horth and Dodson 2002). Larger males fight for access for females, while smaller males 'sneak' matings at the time that a larger male is mating. In the extreme, male parr that have grown very well will mature in freshwater and attempt such matings. This behavior is currently understood in the context of conditional strategies related to a threshold (Hazel et al. 1990, Gross and Repka 1995, Aubin-Horth and Dodson 2002, Hazel et al. 2004) and thus generally consistent with the framework of Thorpe et al (1998). Although the theory of Thorpe et al (1998) did not account for early maturation in males, we will do so for steelhead.

f) Forward projection of survival, size and fitness in relation to flow. The maximum food assimilation efficiency q and water temperature at time t, T(t), behave in predictable and understood relations with respect to water flow (e.g., reduced flow, all else being equal, will reduce q and increase T). Consequently, the Grantee will be able to use the model to predict the consequences of changes in flow at different times of the year on the growth, survival and smolt metamorphosis of juvenile steelhead. (An example of how this might be done, for a preliminary version of the theory for Atlantic salmon, is found in Mangel (1994)).

Task Table: Task summary, schedule, and list of deliverables

Task ID	Task Name	Start Month	End Month	Personnel Involved	Description	Task Products
1	Extend Theory of Thorpe et al	1	24	Mangel, Marc, PhD *Postdoctoral researcher 1 *Postgraduate Research Assistant	Mangel and Post-doc, in consultation with Sogard and Titus will extend the theory of Thorpe et al to account for steelhead in California and will parameterize the models with data collected during the empirical work	Computer models, peer-reviewed journal articles, final report to CalFed presentations at CalFed and other scientific meetings

Task ID	Task Name	Start Month	End Month	Personnel Involved	Description	Task Products
2	Test the Theory	25	36	Mangel, Marc, Ph.D. *Postdoctoral researcher 1 *Postgraduate Research Assistant	The theory developed in Task 1 will be tested through experimental and observational studies (Tasks 3-8)	Data analyses development of a database with field and experimental data, peer-reviewed journal articles, final report to CalFed, presentations at CalFed and other scientific meetings
3	Laboratory Growth Experiment 1	5	17	Sogard, Susan, Ph.D. *Postdoctoral researcher 2 *Laboratory Assistant	Conduct lab experiments designed to determine the influence of growth patterns on emigration at age 1 in juvenile steelhead. Factors include two populations (Scott Creek and Battle Creek) and four ration treatments, with fish held at restricted levels except for a varying two month period of unlimited food availability. Photoperiod will match the natural cycle of a	Final report to CalFed, presentations at CalFed and other scientific meetings, publication in peer-reviewed scientific journal

Task ID	Task Name	Start Month	End Month	Personnel Involved	Description	Task Products
3 cont.					latitude mid-way between the two population locations, and temperatures will be moderate, matching those experienced on average by the Scott Creek population. Measured variables will include monthly growth rates in length and weight, behavioral assays of activity and aggression in mid-winter, Na+K+ATPase levels in April, and responses to seawater challenges in April	
4	Laboratory Growth Experiment 2	17	29	Sogard, Susan, Ph.D. *Postdoctoral researcher 2 *Laboratory Assistant	Conduct lab experiments designed to determine the influence of growth patterns on emigration at age 1 in juvenile steelhead. Factors include two populations (Scott Creek and Battle Creek) and four ration treatments, with fish held at	Final report to CalFed, presentations at CalFed and other scientific meetings, publication in peer-reviewed scientific journal

Task ID	Task Name	Start Month	End Month	Personnel Involved	Description	Task Products
4 cont.					restricted levels except for a varying two month period of unlimited food availability. Photoperiod will match the natural cycle of a latitude mid-way between the two population locations, and temperatures will be moderate, matching those experienced on average by the Battle Creek population. Measured variables will include monthly growth rates in length and weights, behavioral assays of activity and aggression in mid-winter, Na+K+ATPase levels in April, and responses to seawater challenges in April.	
5	Laboratory Maturation Experiment	29	36	Sogard, Susan, Ph.D. *Postdoctoral researcher 2 *Laboratory Assistant	Conduct laboratory experiments to define the timing of the maturation decision window. Factors include	Final report to CalFed, presentations at CalFed and other scientific meetings,

Task ID	Task Name	Start Month	End Month	Personnel involved	Description	Task Products
5 cont.					two populations and four ration treatments with variable timing of a switch to restricted rations. Measured variables include monthly length and weight and gender and maturation status in April.	publication in peer-reviewed scientific journal
6	Field estimates of juvenile steelhead densities	1	36	Sogard, Susan, Ph.D. Titus, Robert G., Ph.D. *Postdoctoral researcher 2 *Postgraduate Research Assistant *Technician	Estimate seasonal densities of juvenile steelhead in two central California coast creeks and two Central Valley rivers using snorkeling techniques supplemented by electro-shock fishing and seining	Final report to CalFed, presentations at CalFed and other scientific meetings, publication in peer-reviewed scientific journal
7	Field estimates of invertebrate prey densities	1	36	Sogard, Susan, Ph.D. Tutus, Robert G., Ph.D. *Postdoctoral researcher 2 *Laboratory Assistant Technician	Estimate seasonal patterns in food availability at four sites in each of four streams using standard benthic and drift sampling techniques. Sampling will occur over four time periods per year in each of three years, with	Final report to CalFed, presentations at CalFed and other scientific meetings, publication in peer-

Task ID	Task Name	Start Month	End Month	Personnel Involved	Description	Task Products
7 cont.					one 48 hour sampling series for each of the 16 sites during the 3 year period.	
8	Natural growth variability in juvenile steelhead	1	36	Sogard, Susan, Ph.D. *Postdoctoral researcher 2 *Laboratory Assistant *Technician	Estimate natural growth rates of juvenile steelhead in four stream systems, using mark and recapture techniques in each of 3 years. Fish< 65 mm will be tagged with elastomer and fish > 65 mm will be tagged with PIT tags. Recaptures will be conducted in association with seining and electroshock fishing efforts.	Final report to Cal-Fed, presentations at CalFed and other scientific meetings, publication in peer-reviewed scientific journal

Deliverables:

- Semi-annual reports will be submitted every 6 months following the project start date;
- Final report will be submitted 36 months from the project start date;
- Draft manuscript or a project closure summary report will be submitted 36 months from the project start date;
- Final manuscript(s) will be submitted after publication.