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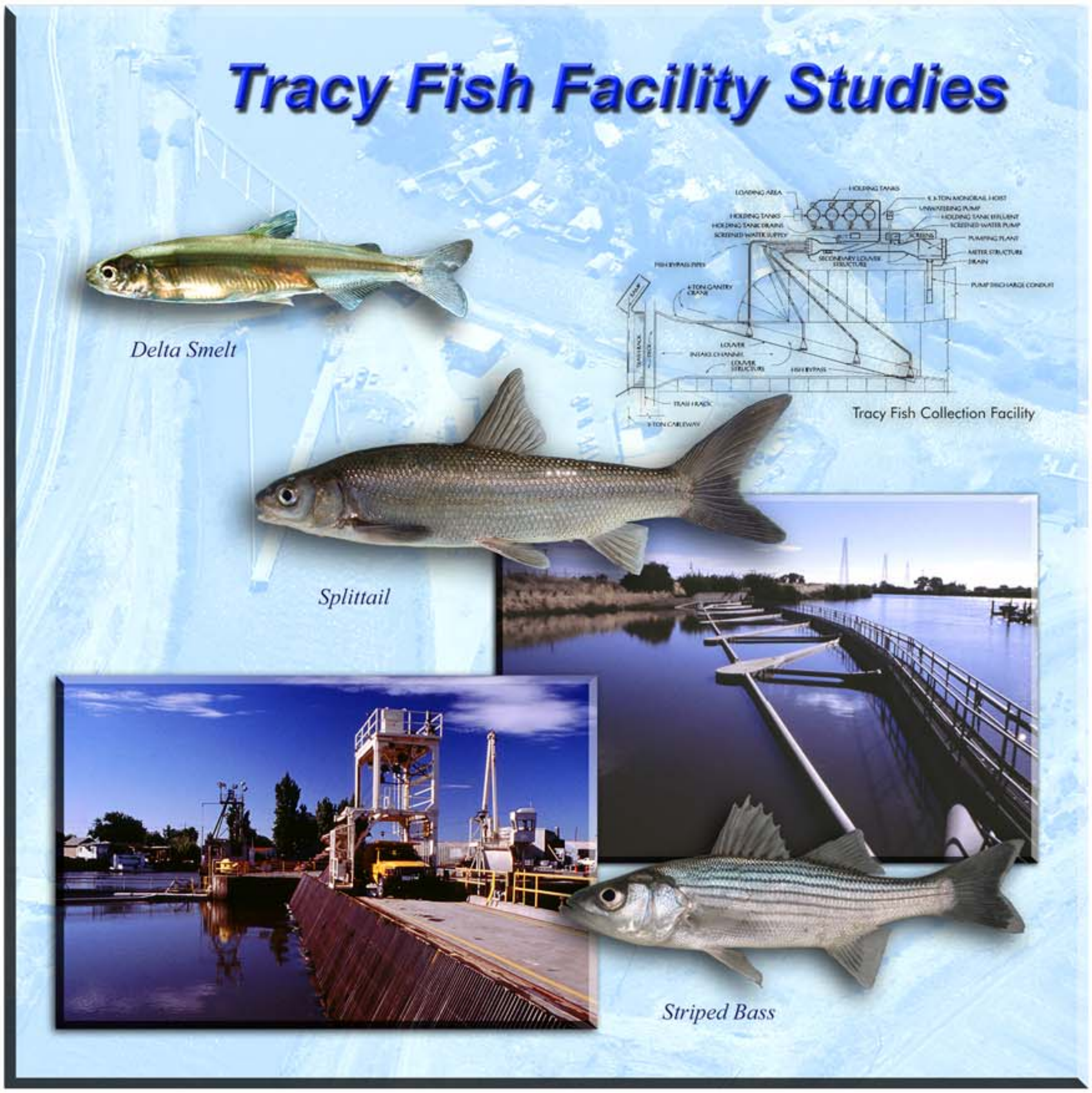

Using Morphometric Characteristics to Identify the Early Life Stages of two Sympatric Osmerids (Delta Smelt and Wakasagi - *Hypomesus transpacificus* and *H. nipponensis*) in the Sacramento-San Joaquin Delta, California

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Delta smelt, *Hypomesus transpacificus*, an indigenous fish to the Sacramento-San Joaquin Delta, California, USA, has been listed as a threatened species by the U.S. Fish and Wildlife Service. The introduced wakasagi, *Hypomesus nipponensis*, is a very similar smelt, and it is difficult to distinguish between the two species in the Delta. This report describes how to identify and distinguish the early life stages of the two species. Four early life stages are reported: (1) prolarvae or yolk sac larvae; (2) postlarvae; (3) prejuvenile; and (4) juvenile. Field specimens were compared with laboratory reared specimens to develop the key. Morphological characteristics and morphometric measurements were used to compare the two species. Illustrations and photographs help identify the features that distinguish these two species. Due to the similar development of early life stages in these two species, it is necessary that the trained biologist use multiple characteristics to correctly identify these similar species.

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Using Morphometric Characteristics to Identify the Early Life Stages of Two Sympatric Osmerids (Delta Smelt and Wakasagi, *Hypomesus transpacificus* and *Hypomesus nipponensis*) in the Sacramento-San Joaquin Delta, California

Volume 30

by

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Illustrations and Photographs – Rene C. Reyes[3]

April 2005

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EXECUTIVE SUMMARY

Delta smelt, *Hypomesus transpacificus*, an indigenous fish to the Sacramento-San Joaquin Delta, California, USA, has been listed as a threatened species by the U.S. Fish and Wildlife Service. The introduced wakasagi, *Hypomesus nipponensis*, is a very similar smelt, and it is difficult to distinguish between the two species in the Delta. This report describes how to identify and distinguish the early life stages of the two species. Four early life stages are reported: (1) prolarvae or yolk-sac larvae; (2) postlarvae; (3) prejuvenile; and (4) juvenile. Field specimens were compared with laboratory reared specimens to develop the key. Morphological characteristics and morphometric measurements were used to compare the two species. Illustrations and photographs help identify the features that distinguish these two species. Due to the similar development of early life stages in these two species, it is necessary that the trained biologist use multiple characteristics to correctly identify these similar species.

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INTRODUCTION

The delta smelt, *Hypomesus transpacificus,* is indigenous to the upper reaches of the San Francisco Bay Estuary (McAllister, 1963; Radtke, 1966; Moyle, 1976, 1995, 2002). Due to population declines beginning in the early 1980s, in 1993 delta smelt was listed as threatened under the State and Federal endangered species acts. The delta smelt spends the majority of its life cycle in the freshwater/brackish portion of the estuary—mostly near the confluence of the Sacramento and San Joaquin Rivers—henceforth called the Delta (Plate 1). This region provides a significant portion of the water supplies for the San Francisco Bay area, the San Joaquin Valley, the Central Coast and Southern California. Much of this water supply comes from State and Federal water project diversions in the southern Delta. To reduce water supply-related threats to delta smelt, Federal and State resource agencies have implemented restrictions on water supply operations and placed limits on the numbers of delta smelt that can be taken at the water project intakes.

In recent years, understanding delta smelt life history and the numbers taken at the intakes to the pumping plants has been complicated by the presence of another osmerid species, the introduced wakasagi, *Hypomesus nipponensis*. The two organisms are morphologically similar, and until 1963 taxonomists considered them one species called pond smelt (McAllister, 1963). Delta smelt, *Hypomesus transpacificus,* was officially recognized as a separate species by the California Department of Fish and Game (CDFG) in 1972 (Miller and Lea, 1972). Ecologically, the two species may compete for similar habitat and food resources (Aasen et al., 1998). In a comparative environmental tolerance study using delta smelt and wakasagi, some differences in response were noted with the wakasagi having physiological advantage over delta smelt with respect to wider salinity and water temperature range tolerances (Swanson et al., 2000).

Despite the need for correctly identifying all life stages of these two species, until recently this has not been possible. In particular, the early life stages have been difficult to distinguish. A complete identification guide for the two species starting from the early life stages is important to understand their speciation and ecology. We hope that this report helps researchers, managers, and operators distinguish between the two species, and that this ability will lead to increased understanding and protection for delta smelt.

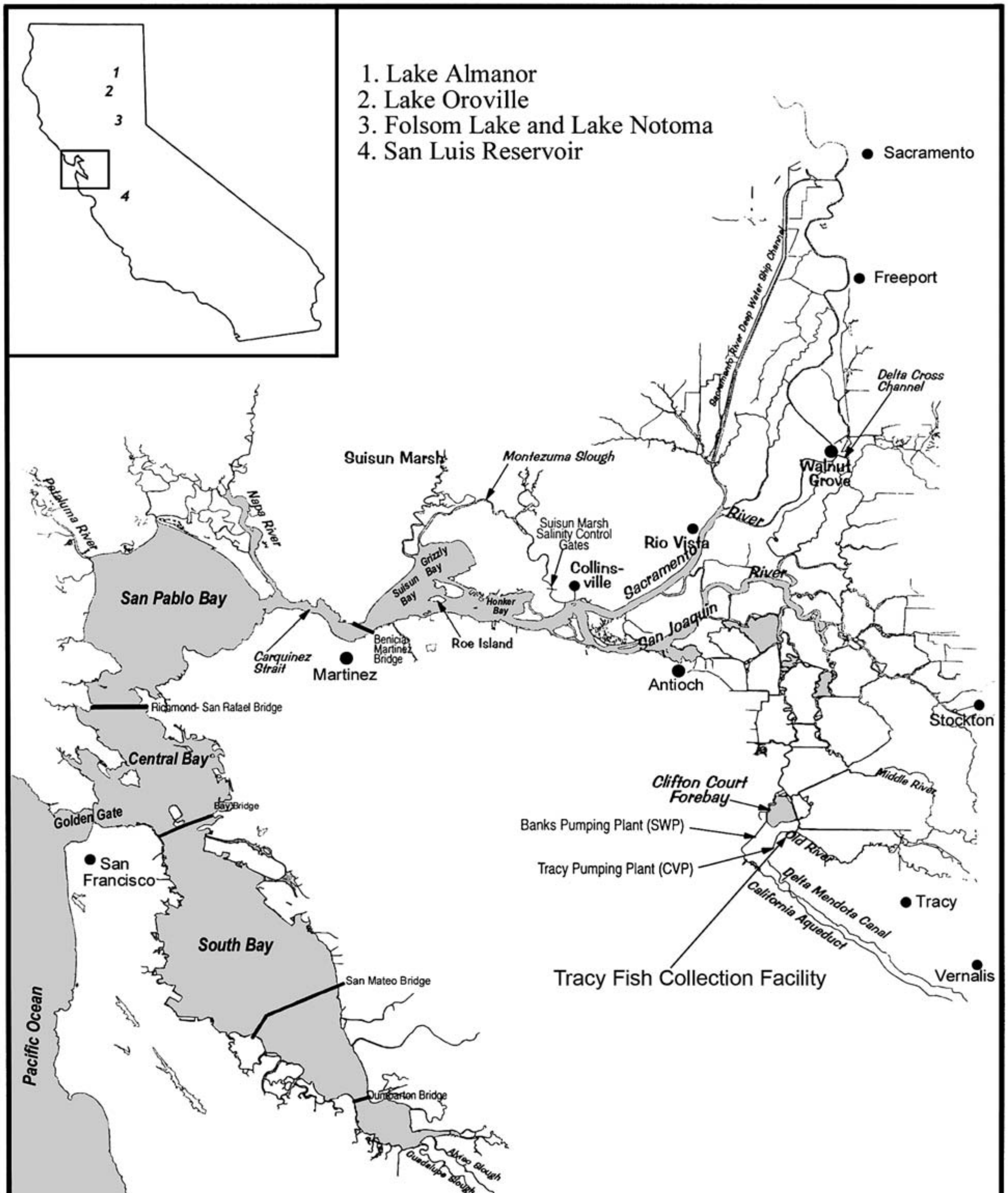


PLATE 1.—Map showing the Sacramento-San Joaquin Estuary and Tributaries.

In 1959 (before delta smelt were recognized as a distinct species), CDFG introduced the wakasagi, a native Japanese osmerid, throughout California. Initial stockings were made in Dodge Reservoir, Shastina Reservoir, Freshwater Lagoon, Spaulding Reservoir, Jenkinson Lake, and Big Bear Lake. The stocked fish were to provide a forage base for salmonids stocked in lakes and reservoirs (Wales, 1962). Some of the reservoirs in which wakasagi were initially or subsequently stocked were above dams in the Sacramento River watershed. The wakasagi have since moved out of the reservoirs and now are relatively common in streams above the Delta, including the Lower American River. Wakasagi have been observed on occasion in the Delta and adjacent waters over the past two decades. The earliest recorded occurrence of the wakasagi was a single specimen collected in Montezuma Slough by a CDFG biologist in July 1974. A few adult wakasagi were captured in 1978–82 between the confluence of the Sacramento and the San Joaquin Rivers. It is unclear why the wakasagi were rarely seen in the Sacramento-San Joaquin Delta between the 1960s and 1980s, followed by a rapid increase in mid-1990s. One possible explanation is that when one of the gates on the American River's Folsom Dam broke in 1995, many wakasagi were washed out of Folsom Lake into the Delta via the American and Sacramento Rivers. Currently, wakasagi are commonly observed during various Interagency Ecological Program (IEP) sampling events in the Delta and adjacent waters (Hess et al., 1995; Aasen et al., 1998).

There are four osmerid species commonly found in the Delta. The native surf smelt, *Hypomesus pretiosus*, delta smelt, and longfin smelt, *Spirinchus thaleichthys* (Miller and Lea, 1976; Moyle, 1976), and the introduced wakasagi. With respect to larval smelt identification, the surf smelt, a coastal marine species, has a myomere count of at least 60 while the other three species all have less than 60 (Wang, 1986). In addition, surf smelt are generally intolerant of the Delta's low salinity water and only a few surf smelt larvae enter the Delta during their winter/spring spawning period. Distinguishing larval longfin smelt and the delta smelt relies on the air bladder's developmental morphology as a primary characteristic (Wang, 1991). Developing a key to separate larval wakasagi from larval delta smelt has been challenging because of morphological similarities in their early life stages (Wang and Brown, 1993; Wang, 1995), although the two species are thought to be in divergent trend in evolution (Moyle, 2004).

The following is a brief history of the recorded occurrences of wakasagi and the related information about this species in the Delta:

1. In 1990, wakasagi larvae were first found in fish egg and larvae (E&L) samples collected by the CDFG at the confluence of the Sacramento and Feather Rivers in 1990 (CDFG, 2004; Wang, 1995). Specimens were initially identified as the delta smelt, and subsequently as wakasagi after additional early life stage information was available. Wakasagi is thought to be the only osmerid collected in the mid and upper reaches of the Sacramento River and in the Feather River (Seesholtz, 2004).

2. In 1991, additional wakasagi larvae were collected near Fair Oaks on the American River (a few miles below the Folsom/Nimbus Dam complex) by Hanson Environmental (Wang, 1995).

3. Also in 1991, wakasagi larvae were found in the CDFG's E&L samples from a station on the Sacramento River below the mouth of the American River, (CDFG, 2004; Wang, 1995).

4. In 1992, one wakasagi larvae was observed from CDFG's E&L sample collected at the station on the Sacramento River just upstream of Walnut Grove (CDFG, 2004; Wang, 1995).

5. In 1991 and 1994, wakasagi larvae, prejuveniles, and juveniles were captured by Bureau of Reclamation (Reclamation) biologists at a site on the Sacramento River just above its confluence with the American River (Wang, 1995).

6. In 1993 and 1994, approximately 110 wakasagi larvae were collected at stations on the American River, Sacramento River, Cache Slough, Miner Slough, Sacramento River Deep Water Ship Channel, Barker Slough, and the lower Mokelumne River. The Mokelumne River specimen was the first recorded appearance of wakasagi larvae on the San Joaquin River side of the Delta and probably came from the Sacramento River via the Delta Cross Channel in the springs of 1993 and 1994 (CDFG, 2004; Wang, 1995).

   Because more wakasagi ecophenotypes were observed in the Sacramento-San Joaquin Delta than in previous years, and these ecophenotypes were different from the wakasagi that resided in the lakes and reservoirs above the Delta, some of these fish were initially misidentified (Wang and Hess, 2000). Environmental factors may help produce those ecophenotypes.

7. In the wet year of 1993, many wakasagi were collected at the Tracy Fish Collection Facility (TFCF) and State Water Project (SWP) fish salvage facilities along with delta smelt (Siegfried et al., 2000). The TFCF is the Reclamation fish salvage facility for the Central Valley Project's (CVP) Tracy Pumping Plant.

   Fewer wakasagi were collected in 1994, a dry-water year. We estimated that wakasagi probably were introduced to San Luis Reservoir, a joint CVP/SWP storage reservoir located south of the Delta during 1993–94. This conclusion is based on observations by Spaar (1990a, 1990b, and 1991) and Hiebert (1995) who did not observe wakasagi in the vicinity of CVP and SWP intakes in the late 1980s and early 1990s. Hess et al. (1995) reported capturing wakasagi in San Luis Reservoir in 1994–95.

8. In the wet-water years of 1995–2000, wakasagi moved from the Sacramento-San Joaquin Delta further westward to Suisun Bay, Montezuma Slough (a tributary of the Suisun Marsh along the eastern edge of the San Pablo Bay), and the Napa River (CDFG, 2004; Matern et al., 1996; Wang, 1996).

9. From February through April 2000, over 200 spawning wakasagi and over 150 larvae were collected at San Luis Reservoir. In early 2001, another 150 adult wakasagi were collected at the same location. Wakasagi (1 to 2 years old) spawned in the University of California at Davis Delta Smelt Culture Project at the Skinner Fish Collection Facility. These researchers reared the complete wakasagi life cycle—from fertilized eggs to adults—and archived a complete series of specimens including yolk sac larvae, larvae, prejuvenile, juvenile, and adults. These samples have also been used for genetic verification of the species.

10. More spawning-sized wild wakasagi were also observed in 2000 and 2001 samples from the SWP and CVP fish salvage facilities suggesting that wakasagi had established a successful reproducing population in the Delta and adjacent waters.

METHODOLOGY

Specimens

We examined approximately 10,800 delta smelt, and 4,200 wakasagi specimens from various locations and waters (including hatchery stocks) that were collected from 1978 to 2003. Voucher specimens collected in the last 5 years are currently preserved at the CDFG Stockton office, and the TFCF. Further descriptions of samples used in developing this key are found in appendix 1.

Morphological terminology and description:

1. To describe the larval fish, we refer to the following morphological features: the snout, mouth (upper jaw, lower jaw, maxillary, mandible, isthmus, lip, superior, terminal), lower chin, eye, head, nape, thoracic, pectoral girdle, yolk sac, oil globule, gut, or digestive tract (stomach, intestine, anus), air bladder, finfolds (dorsal finfold, ventral finfold, postanal finfold), finbud, finray, adipose fin, pelvic girdle, cleithrum, myomere (preanal myomere, postanal myomere) notochord, flexion, caudal peduncle, vertebra, urostyle, tail (the body section behind anus), and body profile.

2. Pigmentation patterns are important larval fish descriptors. The morphology of the melanophores (chromatophores) can be classified into three types: stellate (with pseudopodia), dash, and block (with less pseudopodia). Pigmentation may be found externally and internally at various locations on head and body (such as mid-dorsum, mid-ventrum, postanal, and various locations of caudal region). Note that the melanophores generally fade shortly after the specimens are immersed in formalin.

3. Changes in the morphological characteristics of the second section of dorsal finfold (formation process of the adipose fin) between the two species are compared.

4. Anatomical formation of the air bladder between the two species is compared.

5. There are five early life stages of larval osmerids described in the key. They are:

    a. Yolk sac larvae (prolarvae) are the newly hatched larvae with a yolk sac, oil globule, and full finfold.

    b. Post-yolk sac larvae (postlarvae) have the entire yolk sac absorbed, but the small oil globule may or may not exist.

c. Larvae (large postlarvae) have two sections of dorsal finfold, and the fin bud, flexion and urostyle have become visible.

d. Prejuveniles have most finrays developed, the finfold has largely retreated, but scales have not developed.

e. Juveniles have completely developed finrays, scales present, and resemble adults except in size.

Morphometric measurement:

1. Length ratio (see plate 2a, No. 1): Total length (TL) is measured from the tip of the snout to the tip of caudal finfold. Preanal length (PAL) is measured from the tip of snout to the opening of the anus. The length ratio is represented as a percentage by PAL/TL. This formula is applied to larval specimens prior to development of the tail fork.

2. Eye size ratio (see plate 2a, No. 2): The diameter of the eye is measured vertically from the rim of the eye to the opposite rim of the same eye. The depth of the head is measured through the mid-eye position, in a vertical line. The eye size ratio is calculated by dividing the diameter of the eye by the depth of head at mid-eye position. This ratio is applied to the prejuvenile and juvenile life stages.

3. Dorsal finfold ratio (see plate 2a, No. 3): This ratio is based on measurements at the highest point of the dorsal finfold. A vertical line drawn at that point is used to obtain the depth of the dorsal finfold (a) and the depth of body (b). Ventral finfold is not included in this ratio. The ratio is represented by the measurement categories, such as finfold > body, finfold = body, and finfold < body. This ratio is applied to the yolk sac larvae and the early postlarvae stages with residual oil globule present after commencement of feeding.

4. Nape ratio (see plate 2a, No. 4): The measurement of depth at the narrowest point of the nape (behind the pectoral fin), and in front of the yolk sac. The other measurement is the total length of the larval fish. The nape ratio is calculated as nape depth divided by TL. This ratio is applied to the late yolk sac larvae, postlarvae, larvae, and prejuvenile life stages.

5. Myomere count (see plate 2a, No. 5):: Total myomeres: count starts at the first complete myomere (between two myosepta) behind the head to the end of caudal (or flexion in the advanced stages). Preanal myomeres and postanal myomeres: a vertical line is drawn at the opening of the anus through the body; two counts are obtained from this partition.

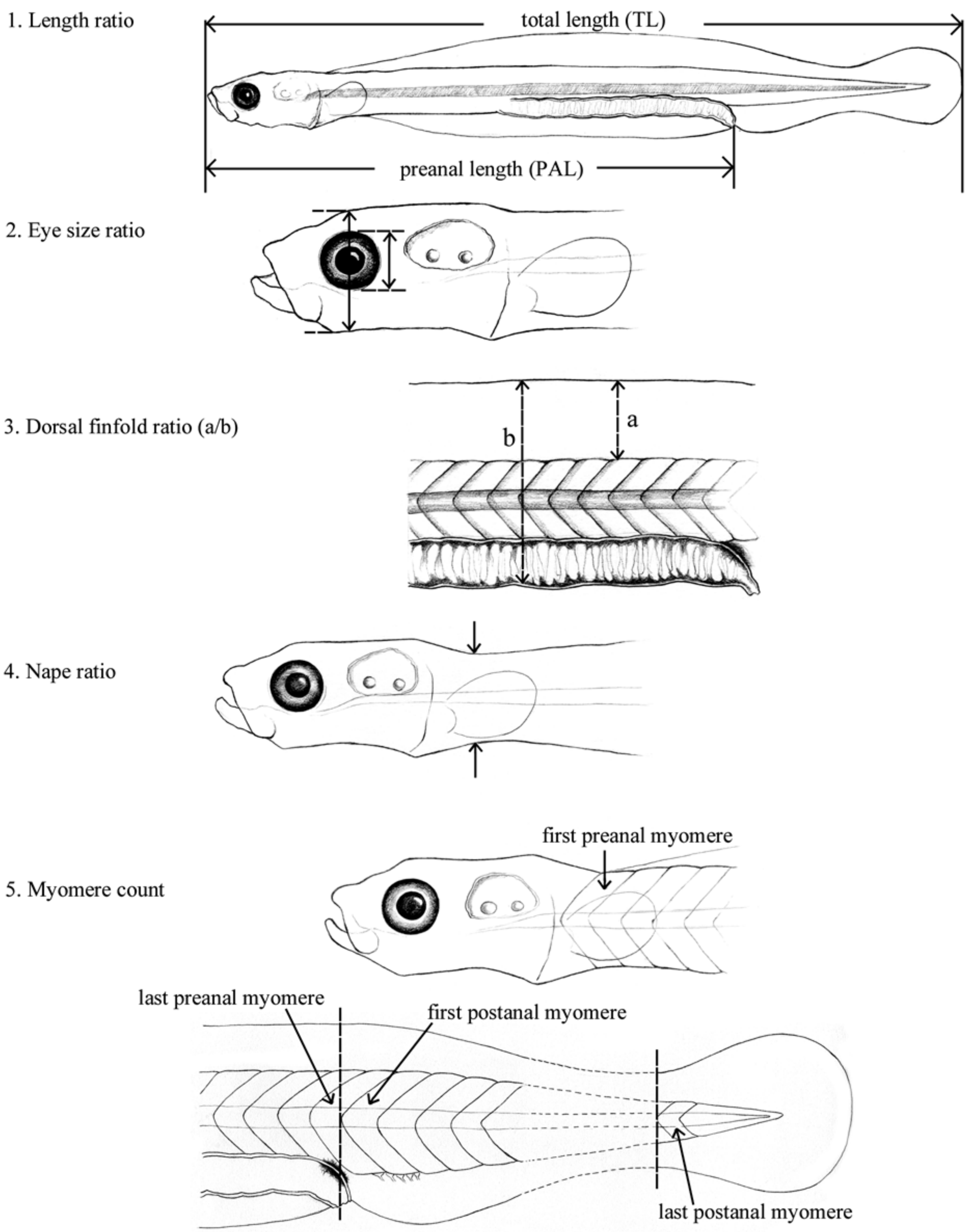
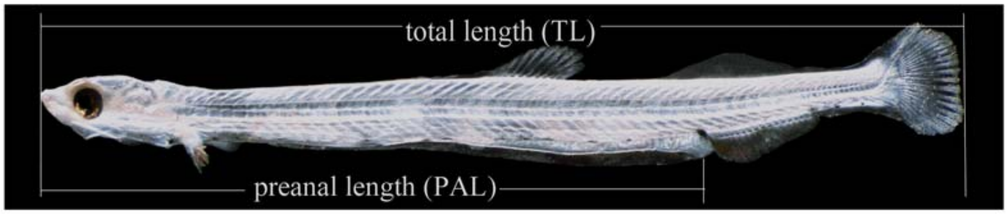
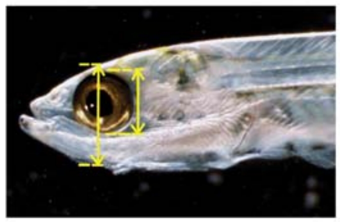


PLATE 2a.—Morphometric measurement (illustrations).

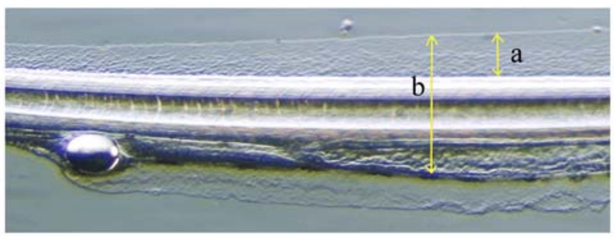
1. Length Ratio



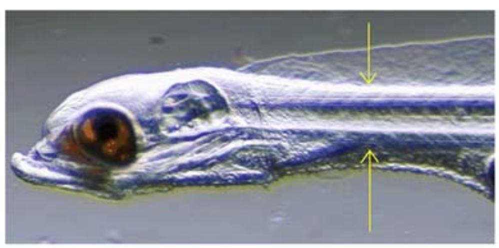
2. Eye size ratio



3. Dorsal finfold ratio (a/b)



4. Nape Ratio



5. Myomere count

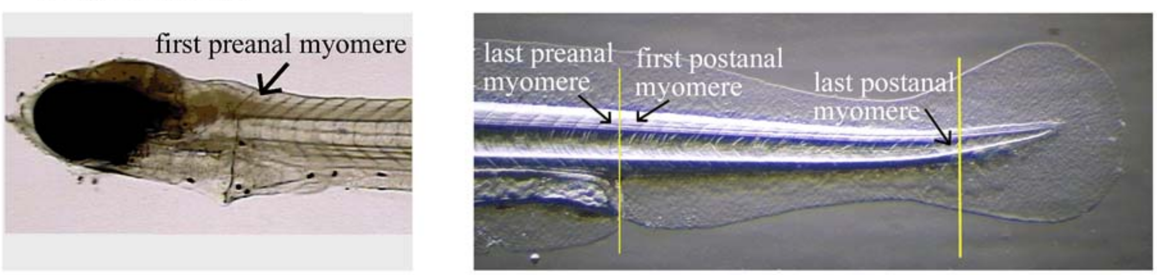


PLATE 2b.—Morphometric measurement (photographs).

KEY

The following are the comparative diagnostic keys with emphasis on the morphometric and anatomical differences in distinguishing yolk sac or prolarvae, post-yolk sac or postlarvae, larvae and juveniles, prejuveniles and juveniles, and juvenile life stages of delta smelt and wakasagi. We recommend using the descriptive characteristics collectively to achieve the greatest accuracy.

Yolk sac larvae or Prolarvae, ca. 4.0–6.0 mm TL (see plates 3a and 3b)

Wakasagi:

1. Oil globule, small, ca. 0.10–0.20 mm in diameter for newly hatched larvae
2. Yolk sac: small, round, oval, and teardrop
3. Yolk sac absorption: faster, absorbed at less than 5.5 mm TL
4. Yolk sac pigmentation: on mid-ventral and anterior region
5. Mid-pectoral girdle pigmentation: 3 pairs below the pectoral girdle
6. Postanal (on ventrum) pigmentation: from 0 to 6, various patterns
7. Profile of dorsal finfold: high; a “curve” or an “arch”
8. Postanal myomere: mainly 17–18 (ranged: 16–19)
9. Length ratio: mainly 71–73 percent (ranged: 70–75 percent)
10. Overall profile body thin and elongate, tail short; larval fish with a small oil globule

Delta smelt:

1. Oil globule: large, ca. 0.20–0.50 mm in diameter for newly hatched larvae
2. Yolk sac: large, mainly round, some oval or teardrop
3. Yolk sac absorption: various, some may last to 6.0 mm TL

4. Yolk sac pigmentation: all over the yolk sac

5. Mid-pectoral pigmentation: 3–5 pairs, both above and below girdles

6. Postanal (on ventrum) pigmentation: mostly 3 to 4, clustered

7. Profile of dorsal finfold: low; dorsal finfold ratio: the depth of dorsal finfold is equal or less than the body depth

8. Postanal myomere: mainly 15–16 (ranged: 14–18)

9. Length ratio: mainly 70–71 percent (ranged: 68–72.5 percent)

10. Overall profile: body stubby and short, tail long, larval fish with a large oil globule.

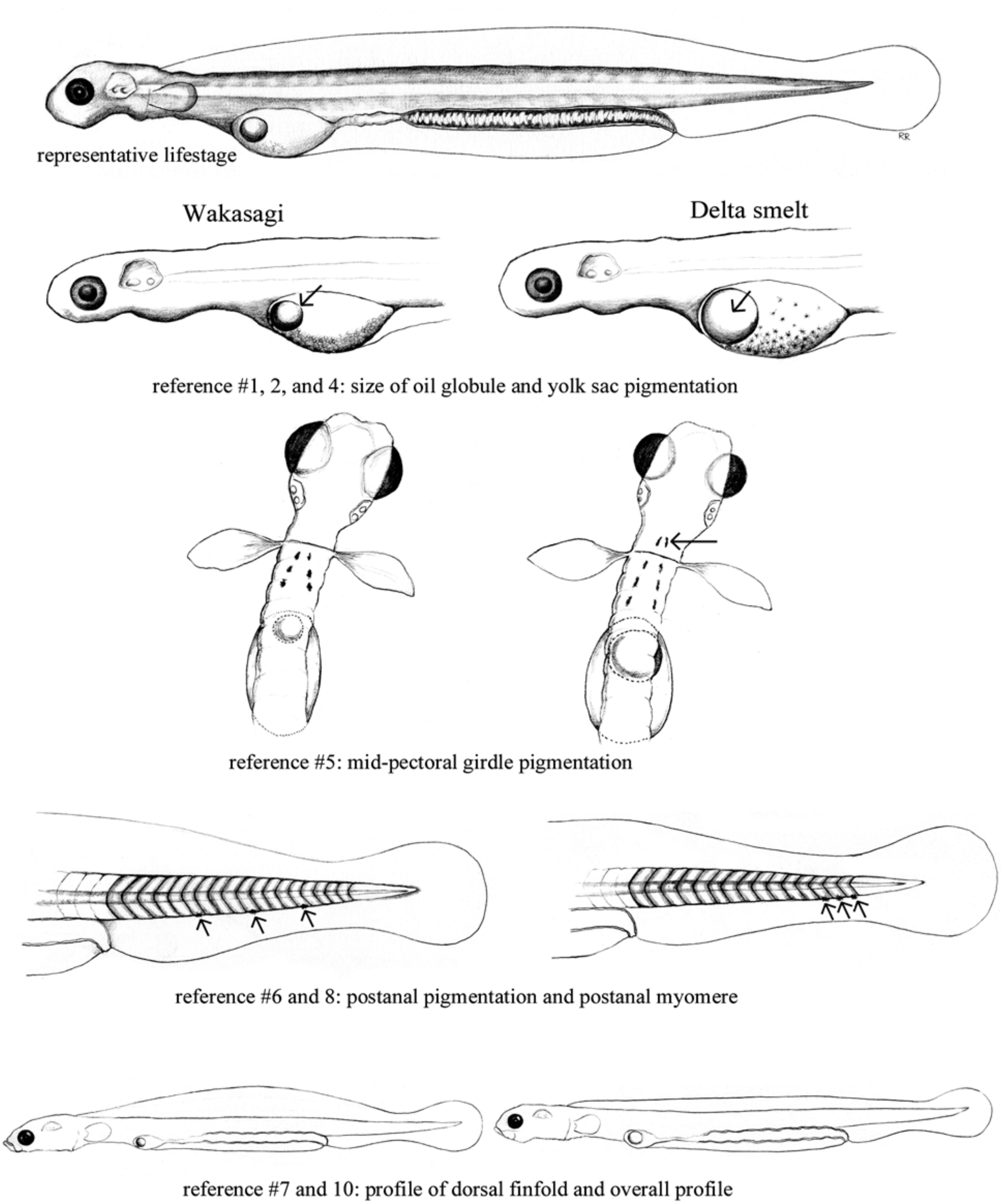


PLATE 3a.—Yolk sac or prolarvae, ca. 4.0–6.0 mm TL (illustrations).

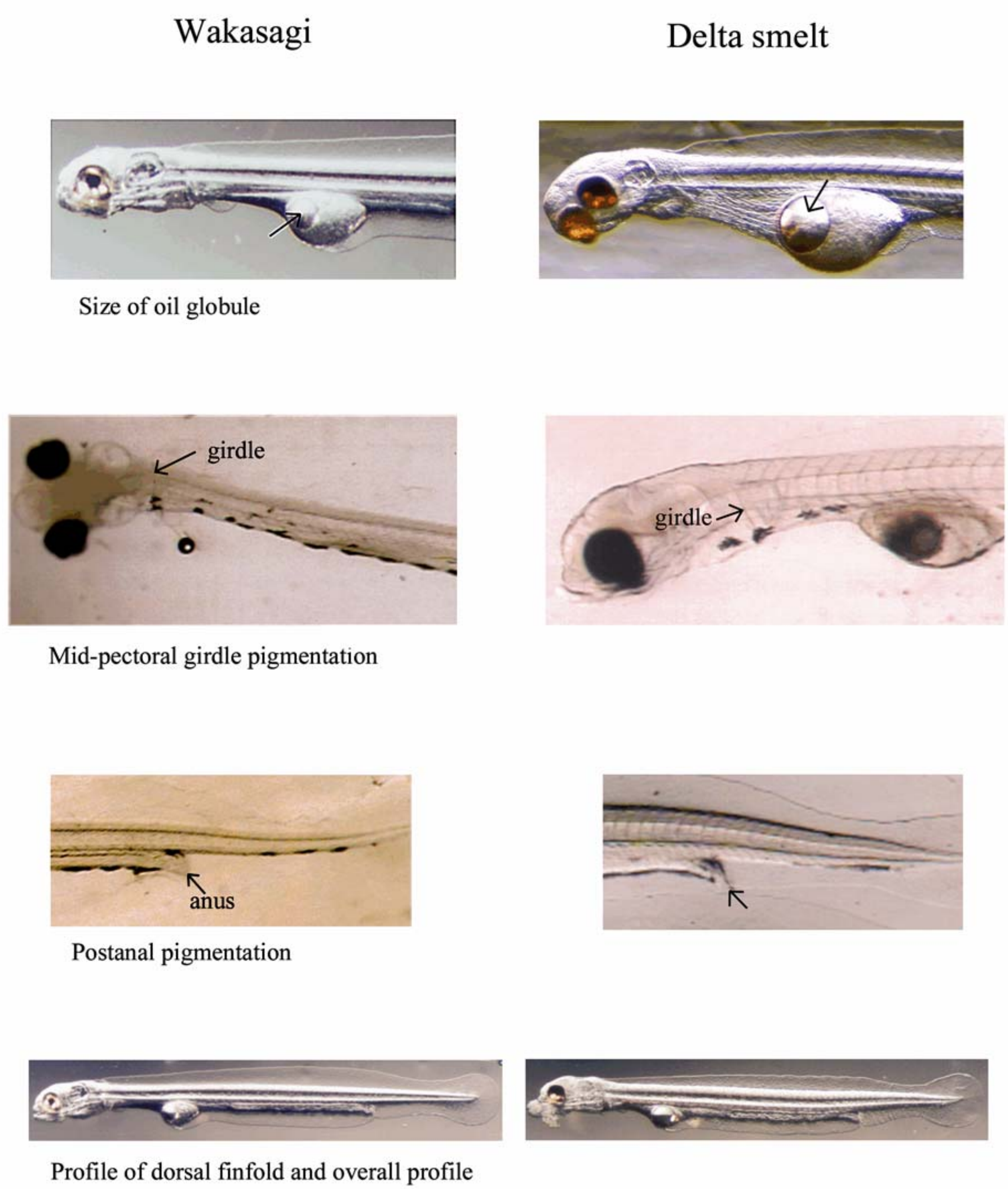


PLATE 3b.—Yolk sac or prolarvae, ca. 4.0–6.0 mm TL (photographs).

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Post-yolk sac larvae or postlarvae, ca. 6.0–10.0 mm TL (see plates 4a and 4b)

Wakasagi:

1. High dorsal finfold, the highest point of dorsal finfold is equal or greater than the body depth (this is true mainly before feeding); dorsal finfold separated into 2 sections at ca. 9.0–10.0 mm TL, also, flexion begins at caudal peduncle region
2. The highest point of second section of dorsal finfold is located above to slightly in front of anus
3. Development of the pigmentation at the ceiling of air bladder as early as 9.0 mm TL
4. Mid-ventral pigmentation: mainly greater than 16 (ranged: 8–21)
5. Lateral ventral pigmentation: mainly all the way to anus
6. Postanal pigmentation (on dorsum): common, 1–2 melanophores
7. Length ratio: mainly 73–75 percent (ranged: 71–77 percent)
8. Nape ratio: mainly 4–6 percent, a ventral notch is noticeable
9. Body profile: fish larvae with a hammer (large) head, narrow nape, slender body, and high dorsal finfold

Delta smelt:

1. Low dorsal finfold, the highest point of dorsal finfold is less than the body depth; dorsal finfold separated into 2 sections at ca. 8.0–9.0 mm TL, also, flexion begins at caudal peduncle region
2. The highest point of second dorsal finfold is located at above to slightly behind the anus
3. Development of the pigmentation at the ceiling of air bladder: not occurring at this stage
4. Mid-ventral pigmentation: mainly ca. 14 or less (ranged: 6–20)
5. Lateral ventral pigmentation: mainly half way to the intestine
6. Postanal (on dorsum) pigmentation: very rare, 1 melanophore
7. Length ratio: mainly 70–72 percent (ranged: 68–73 percent)
8. Nape ratio: mainly 5–8 percent, a ventral notch is less noticeable
9. Body profile: fish larvae with smaller head, chunky (muscular) nape and body, and lower dorsal finfold

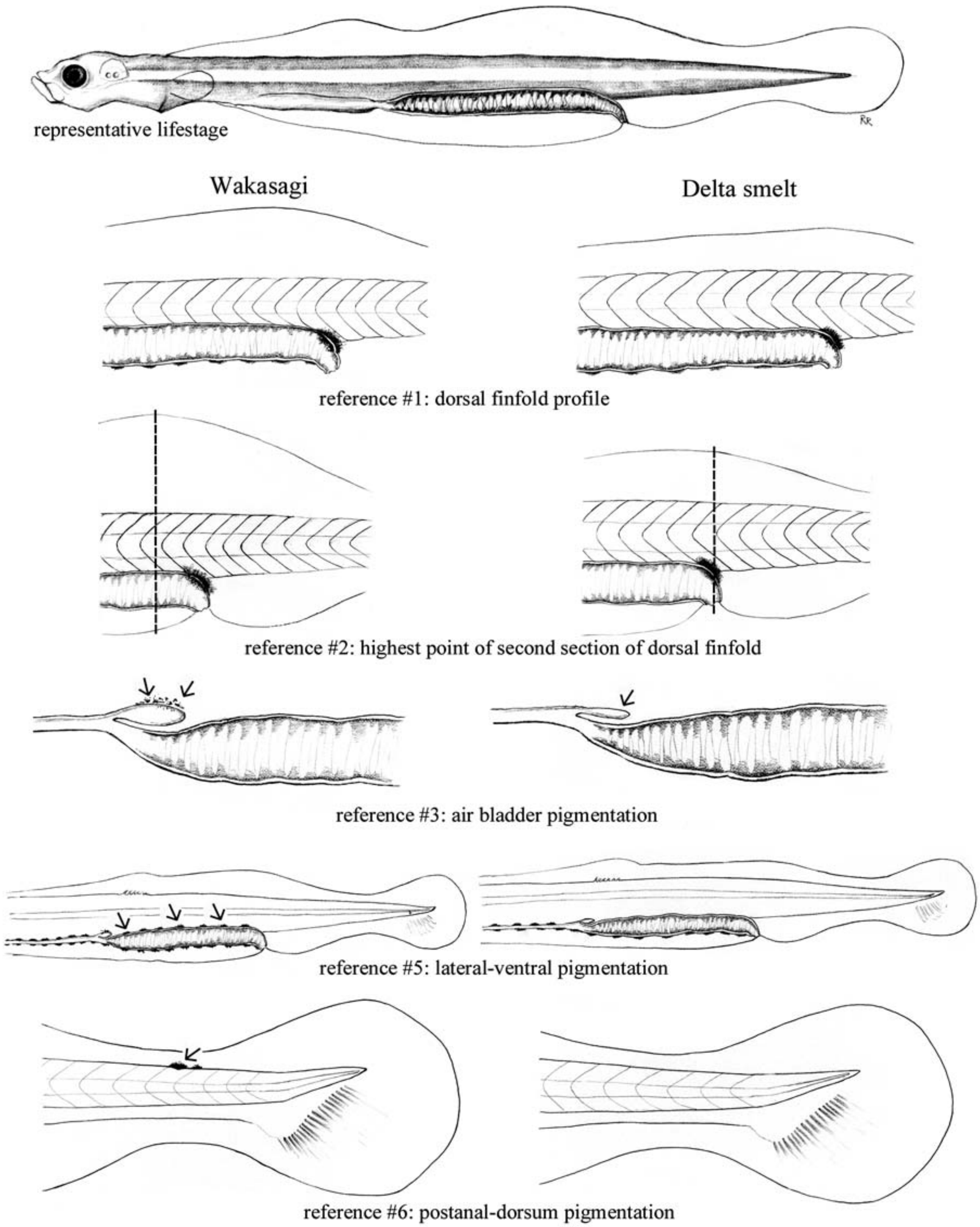
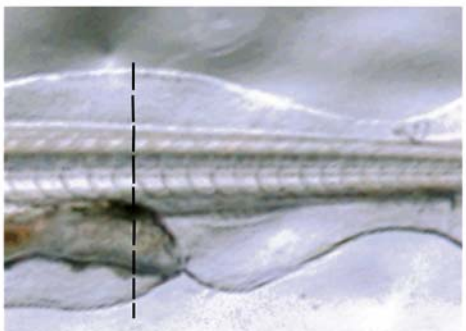
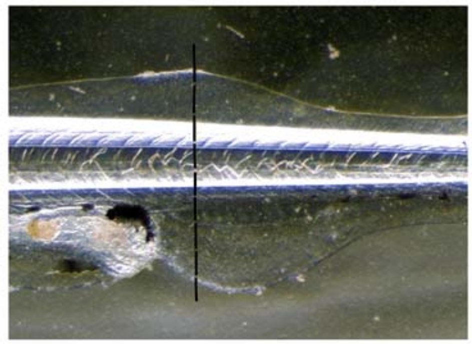


PLATE 4a.—Post-yolk sac larvae or postlarvae, ca. 6.0–10.0 mm TL (illustrations).

Wakasagi | Delta smelt

Dorsal finfold profile

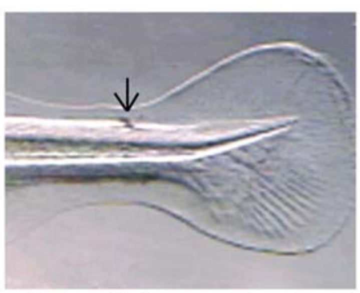




Highest point of second section of dorsal finfold

Air bladder pigmentation (see Figure 5 for air bladder key)

Lateral-ventral pigmentation



Postanal-dorsum pigmentation

PLATE 4b.—Post-yolk sac larvae or postlarvae, ca.6.0–10.0 mm TL (photographs).

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Larvae and Prejuvenile, ca. 10.0–16.0 mm TL (see plates 5a and 5b)

Wakasagi:

1. Eye ratio: ca. 50 percent and greater, larger

2. Air bladder: inflated at ca. 12.0 mm TL, pigmentation covers the ceiling of air bladder

3. Nape: An obvious notch on ventral side (behind thoracic) of nape

4. Pigmentation often observed at base of pectoral and cleithrum

5. Profile of second section of dorsal finfold: base covers both front and back of anus (on dorsum), profile fuller and symmetrical on both slopes

6. Body profile: large head, large eye, narrow nape, obvious air bladder, and elongate body

Delta smelt:

1. Eye ratio: ca. 50 percent and lesser, small

2. Air bladder: may be inflated at ca. 13.0 mm TL, but pigmentation at the ceiling of air bladder starts at ca. 15.0–16.0 mm TL

3. Nape: no obvious ventral notch

4. Pigmentation not often observed at base of pectoral and cleithrum

5. Profile of second section of dorsal finfold: base covers in both front and back of anus, but base migrates behind anus at ca. 14.0–15.0 mm TL, profile symmetrical and less full on both slopes

6. Body profile: head not so large, small eye, thicker nape, obscure air bladder, and chunky body

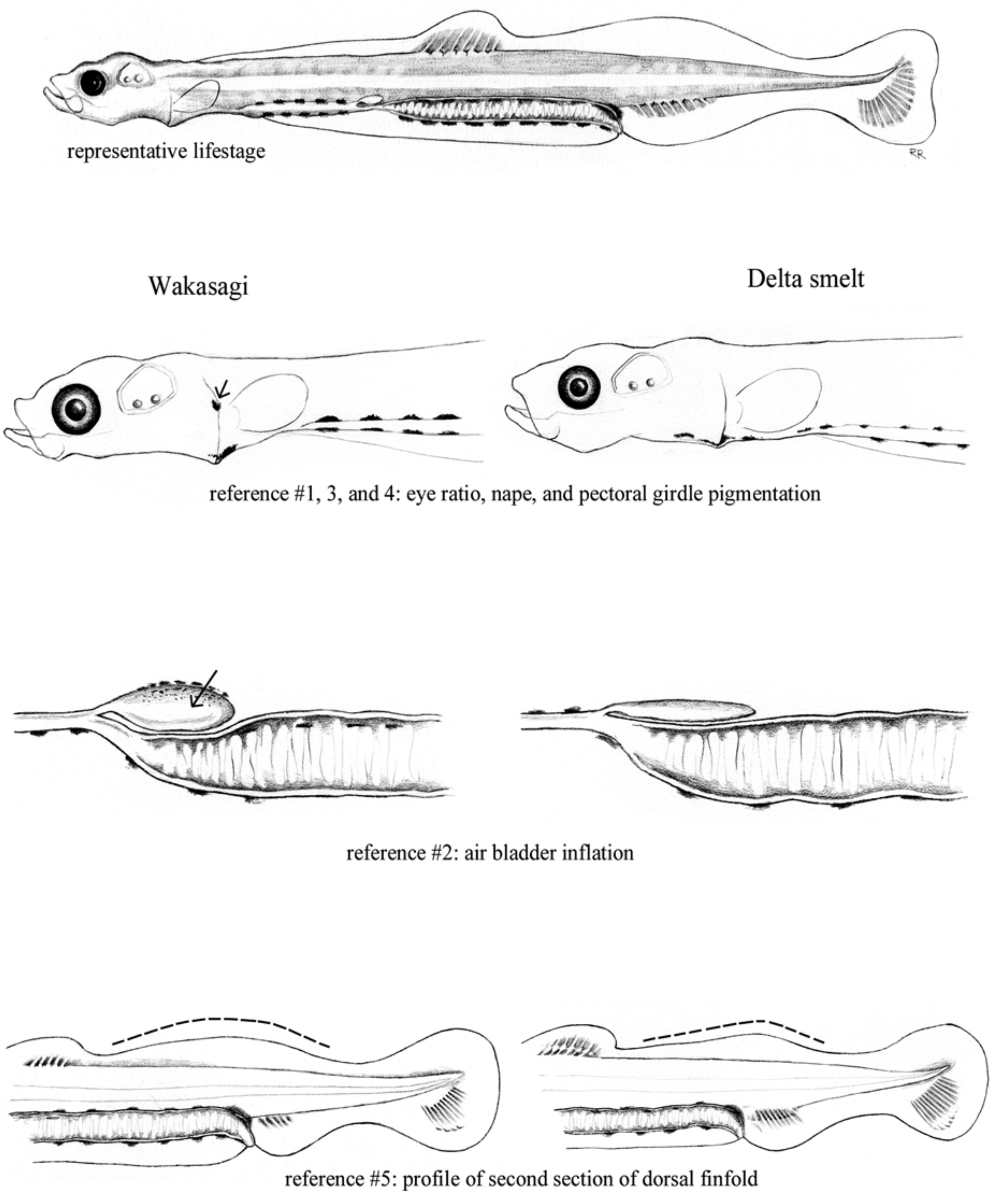

PLATE 5a.—Larvae and prejuvenile, ca. 10.0–16.0 mm TL (illustrations).

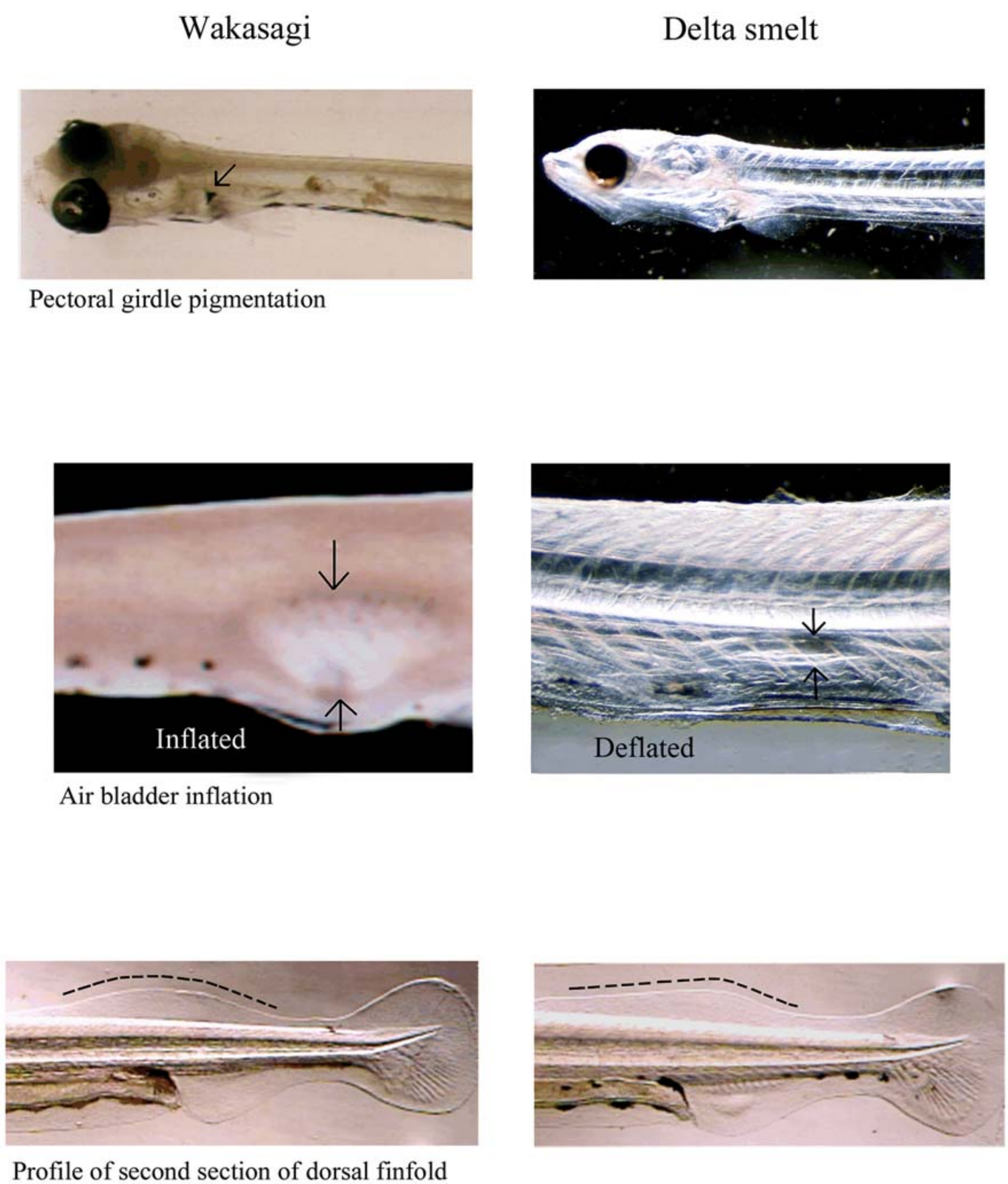

PLATE 5b.—Larvae and prejuvenile, ca. 10.0–16.0 mm TL (photographs).

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Prejuvenile and Juvenile, ca. 16.0–25.0 mm TL (see plates 6a and 6b)

Wakasagi:

1. Eye: large
2. Air bladder chamber: clear and well defined
3. Migration of adipose fin: base of adipose fin migrates behind anus at ca. 20.0–22.0 mm TL, base broader
4. Isthmus pigmentation: some field specimens may develop at ca. 20.0–25.0 mm TL
5. Flexion pigmentation: a black spot observed at upper base of flexion, at ca. 20.0–25.0 mm TL
6. Base of anal fin: mainly short
7. Profile of dorsal fin: narrow and tall, sail-like
8. Body profile: fish tapering look, head > body (trunk) > tail (caudal)

Delta smelt:

1. Eye: small
2. Air bladder chamber: not so clear, and not well defined
3. Migration of adipose fin: base of adipose migrates behind anus as early as ca. 14.0–15.0 mm TL, and completes at ca. 17.0–18.0 mm TL, base shorter
4. Isthmus pigmentation: none (exception: may have one melanophore at just below the mandible)
5. Flexion pigmentation: mainly none above base may have below
6. Base of anal fin: mainly long
7. Profile of dorsal fin: wide and short, fan-like
8. Body profile: fish chunky look, head > body (trunk), or head = body (trunk), and body (trunk) > tail (caudal)

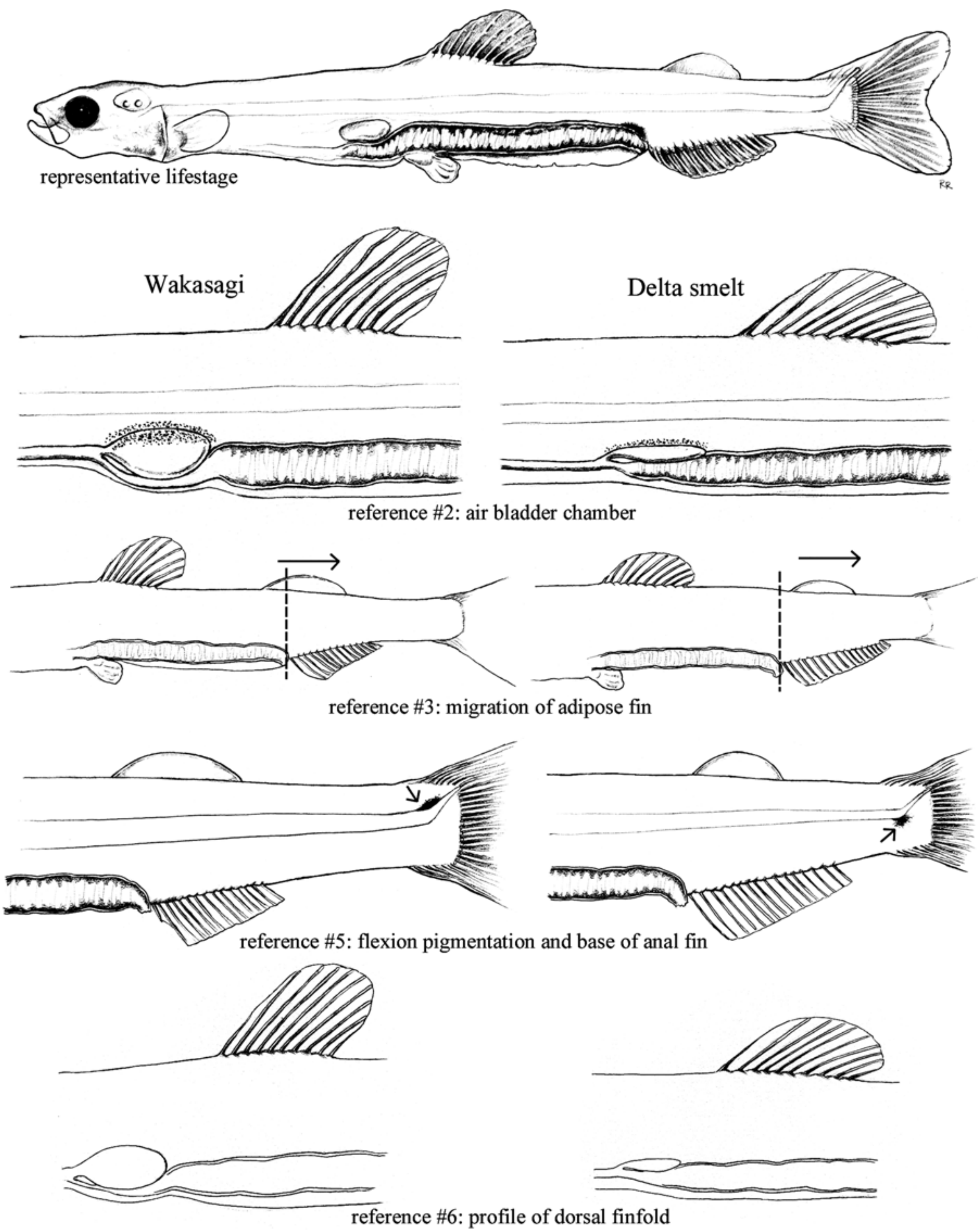

PLATE 6a.—Prejuvenile and juvenile ca. 16.0–25.0 mm TL (illustrations).

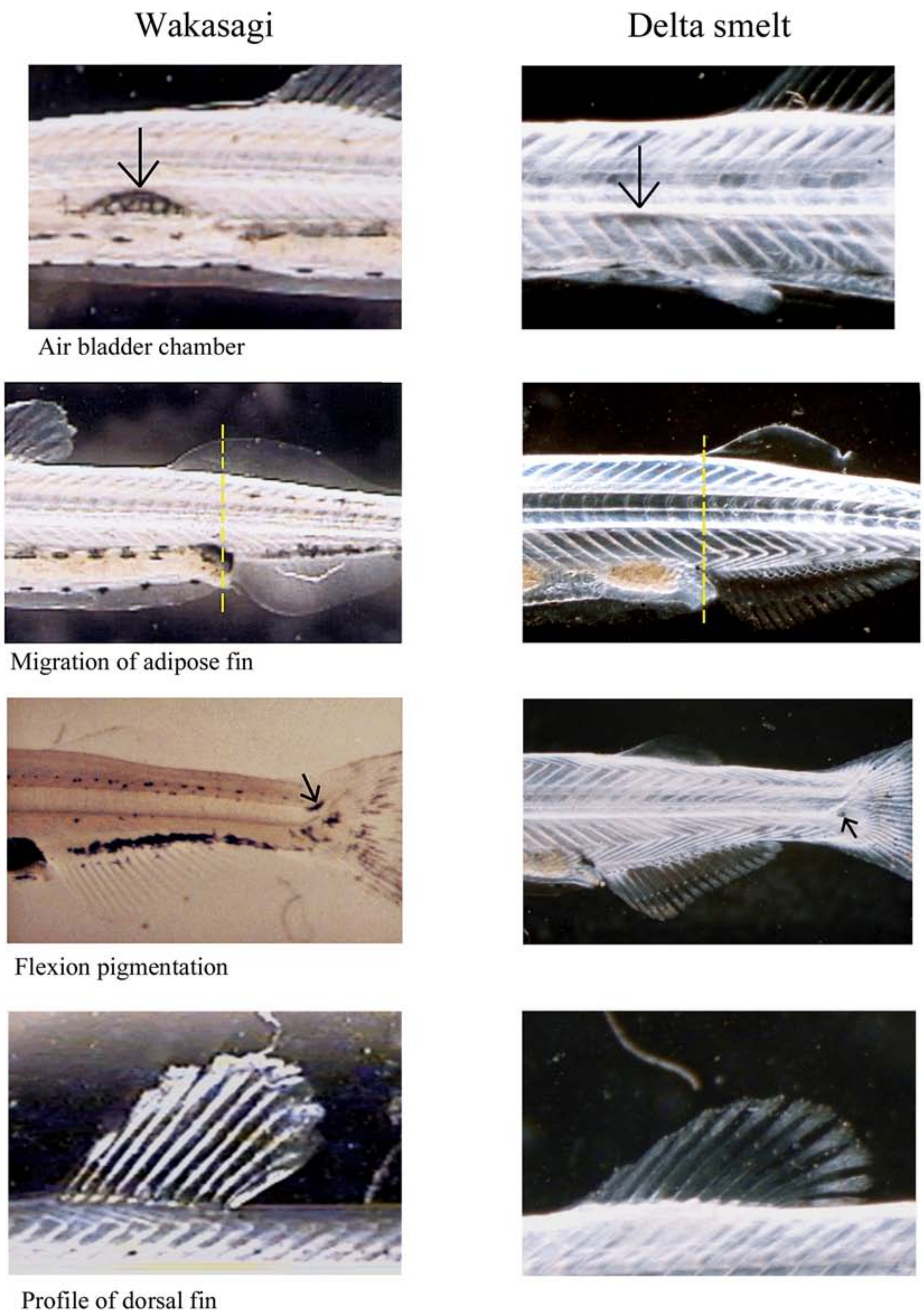

PLATE 6b.—Prejuvenile and juvenile ca. 16.0–25.0 mm TL (photographs).

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Juvenile, ca. 25.0–50.0 mm TL (see plates 7a and 7b)

Wakasagi:

1. Eye: large
2. Pigmentation at upper base of caudal: random scattered
3. Mouth: superior
4. Isthmus pigmentation: yes, for most of the fish
5. Body profile: big eye, superior mouth, and mainly with isthmus goatee (pigmentation)

Delta smelt:

1. Eye: small
2. Pigmentation at upper base of caudal: pigmentation fused, and formed a “<” mark
3. Mouth: terminal
4. Isthmus pigmentation: none (may have one melanophore at just below mandible)
5. Body profile: small eye, terminal mouth, and no goatee for field specimen

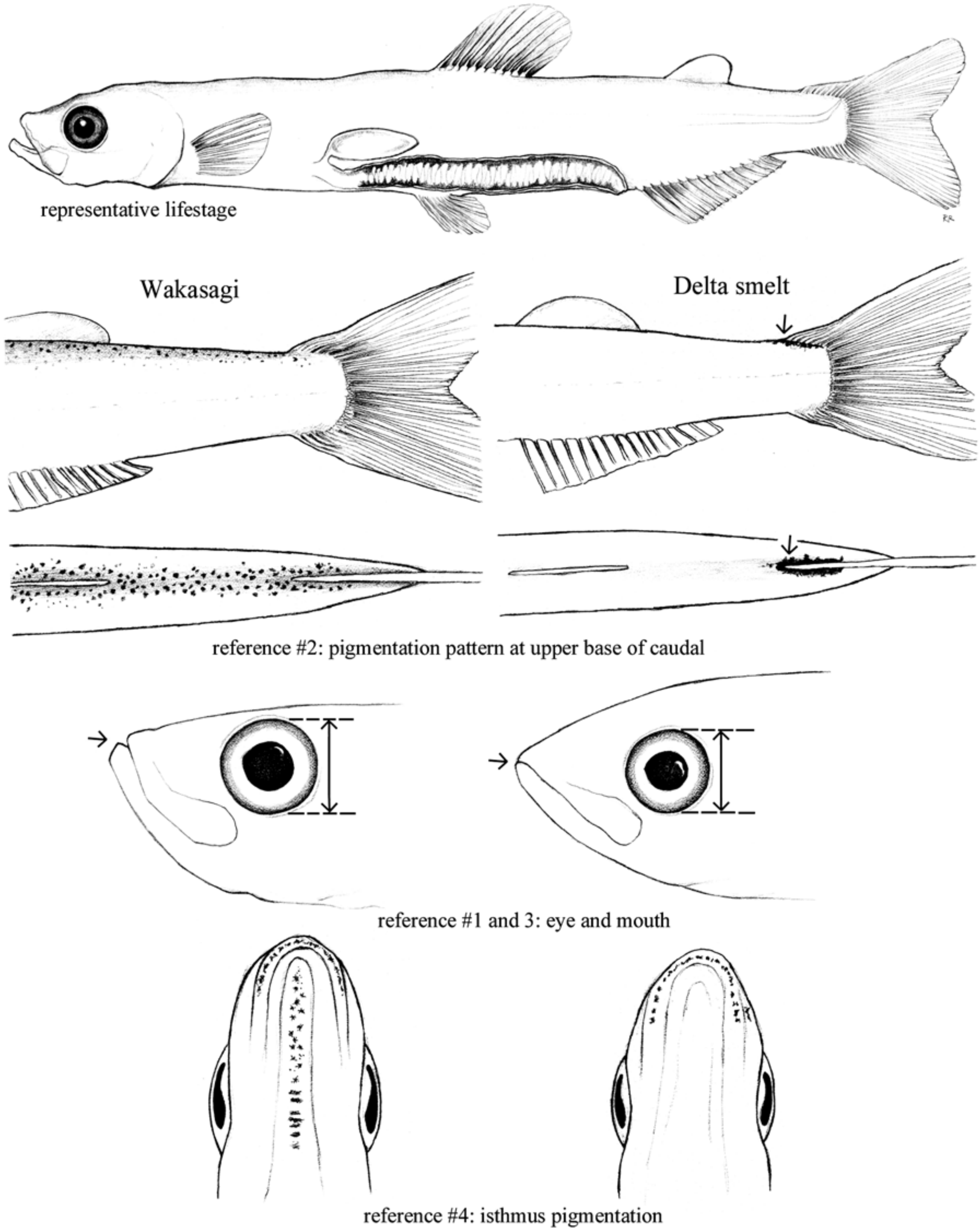

PLATE 7a.—Juvenile, ca. 25.0–50.0 mm TL (illustrations).

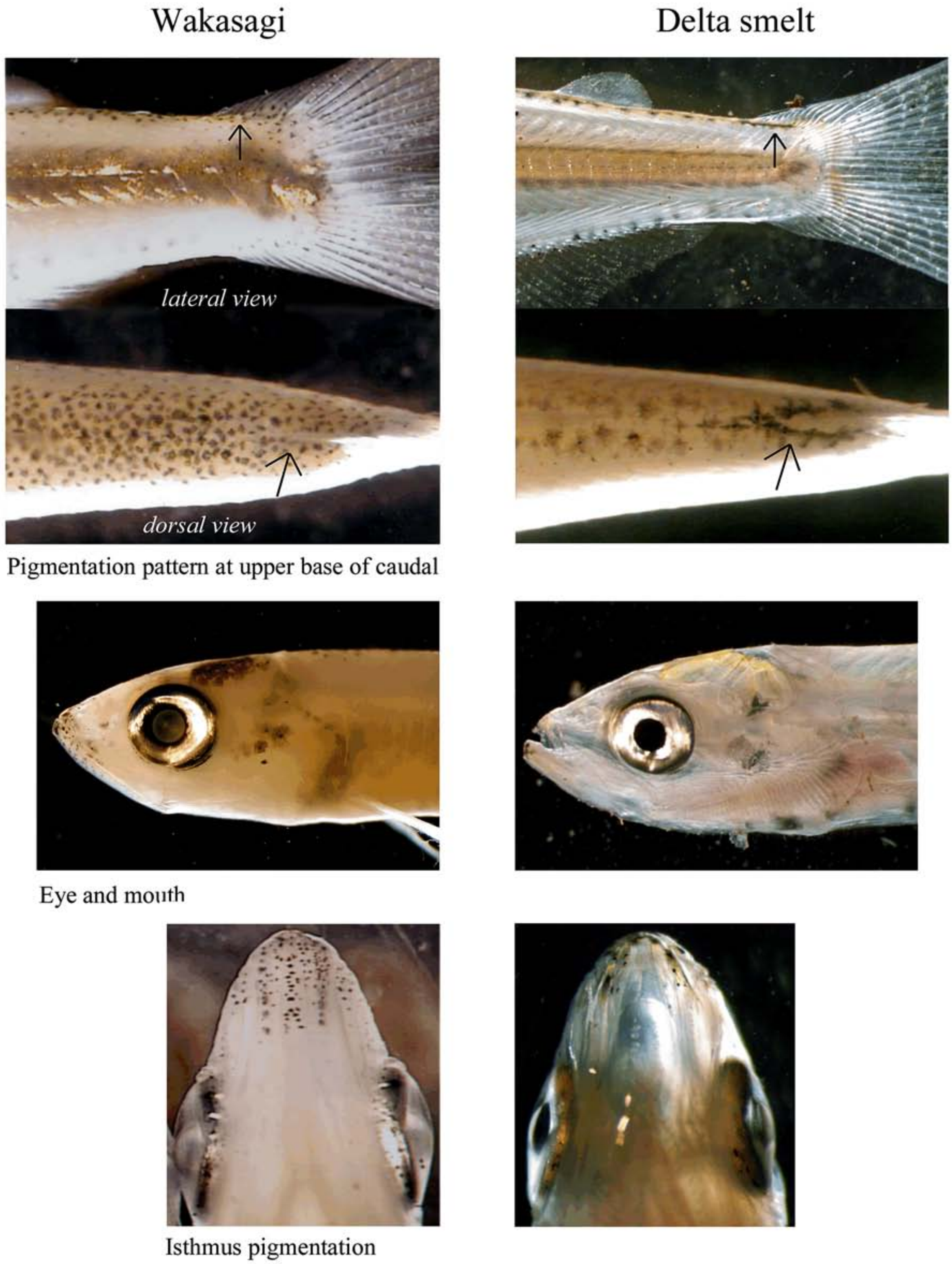

PLATE 7b.—Juvenile ca. 25.0–50.0 mm TL (photographs).

DISCUSSION

The early development of the wakasagi was described by Okiyama (1988), and Shadrin (1989). The early development of the delta smelt was investigated by Simonson (1977) and Wang (1991). The life history and ecological status of these fish has been studied by Moyle et al. (1992), and various Federal and State agencies from the late 1980s. A comparative study between the two species was not performed until the early 1990s (Wang 1995). This study was complicated by the resemblance of the two species during early developmental stages, and was further complicated by the following environmental factors:

a. The spawning periods overlap. Wakasagi spawn in the winter months (December, January and February, and early March); while delta smelt spawn in spring months (March through May, and may extend to June in some years). The delta smelt can also spawn earlier in a mild winter, thus overlap occurs with wakasagi spawning.

b. The areal distribution of the wakasagi was formerly restricted by dams to the lakes and reservoirs above the Delta. Now, the wakasagi has successfully established a reproductive population in the Sacramento-San Joaquin Delta.

c. The spawning grounds overlap. During dry water-years in the early 1990s, wakasagi were found mostly in the upper river, such as the Feather River (below Oroville Dam), and the American River (below Nimbus Dam). Arthur et al. (1990) did not collect delta smelt larvae in their egg and larvae study at Bryte (on the Sacramento River near Sacramento), but they collected delta smelt larvae near the entrance of the Delta Cross Channel (Plate 1). The partition of the spawning ground of two species seems to be located in the vicinity of Garcia Bend on the Sacramento River (CDFG, 2004). During the wet years of 1993, and 1995 to 2000, the wakasagi moved downstream from the Sacramento River to the Cache Slough, Suisun Marsh and the Mid-Napa River. After 2000, delta smelt and wakasagi occupy very similar spawning ground (CDFG, 2004; Maytern et al., 1996).

d. Ecophenotype: As described earlier, we found a range of ecophenotypes of the wakasagi, and recently observed a similar phenomenon in the delta smelt. Lakes and reservoir wakasagi have overall darker body pigmentation, and all have the isthmus pigmentation. The wakasagi from the Sacramento-San Joaquin Delta, San Luis Reservoir and laboratory cultures, however, have a lighter pigmentation. Some delta smelt and cultured wakasagi do not have any isthmus pigmentation. Laboratory-reared delta smelt take on isthmus pigmentation (not found in wild fish), at early life stages (Wang and Hess, 2000), although all other morphometric measurements match the delta smelt. Hence, we suggest isthmus characteristics be considered as reference characteristics, but not the "single key" characteristics to be used to separate the two species during their juvenile and adult life stages (McAllister, 1963; Sweetnam, 1995).

e. A hybrid between the wakasagi and the delta smelt was detected by use of electrophoretic analysis (Trenham et al., 1994; May, 1996; Stanley, et al., 1996). In the laboratory, we crossed the two species by artificial fertilization (male wakasagi x female delta smelt) to examine hybridization potential and produced many abnormal embryos and very low hatching success. Apparent hybrids have been observed in field collections on rare occasions during wet years; however, it is unlikely that extensive hybridization is occurring in the wild.

Because of the similarities between the two species, interested individuals should examine several hundred specimens from wild and voucher collections to gain experience distinguishing these species. The voucher specimens are available from the author.

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REFERENCES

Aasen, G.A., D.A. Sweetnam, and L.M. Lynch. 1998. Establishment of the wakasagi, *Hypomesus nipponensis,* in the Sacramento San Joaquin Estuary. California Department of Fish and Game. 84 (1): pp. 31–35.

Arthur, J.F, M.D. Ball, G. Collins, L. Hess, and C. Liston. 1990. Continuous monitoring of striped bass eggs, and larvae in the San Francisco Bay-Delta Estuary – A potential tool. Bureau of Reclamation, Mid-Pacific Region, Sacramento, California. 55 pp.

CDFG, 2004. Unpublished data. California Department of Fish and Game.

Hess, L., C. Karp, and J. Wang. 1995. Sacramento perch, wakasagi, splittail, Sacramento blackfish, and shimofuri goby in the San Luis Reservoir, and O’Neil Forebay. IEP Newsletter. Autumn 1995: pp. 18–20.

Hiebert, S. 1995. Continuous monitoring of fish eggs and larvae during 1991 and 1992. Tracy Fish Collection Facility Studies, Volume 2. Bureau of Reclamation, Mid-Pacific Region and Denver Technical Services Center. 46 pp.

Matern, S.A., L. Meng, and P.B. Moyle. 1996. Trend in fish population of Suisun Marsh January 1995–December 1995. Report to Department of Water Resources, from University of California at Davis, Davis, California. 41 pp.

May, B. 1996. Identification of smelt species and their interspecific hybrids in the Sacramento-San Joaquin Estuary by allozyme analysis. IEP Newsletter. Summer 1996: pp. 9–10.

McAllister, D.E. 1963. A revision of the smelt family, Osmeridae. National Museum of Canada Bulletin 191. 53 pp.

Miller, D.J., R.N. Lea. 1972, 1976. Guide to the coastal marine fishes of California. California Department of Fish and Game, Fish Bulletin 157. 249 pp.

Moyle, P.B. 1976. Inland Fishes of California. University of California Press, Berkeley. 405 pp.

__________ 1995. Searching the origin of the delta smelt. IEP Newsletter. Spring: pp. 9–10.

__________ 2002. Inland Fishes of California, Revised and Expanded. University of California Press. 502 pp.

__________ 2004. Personal communication.

Moyle, P.B., B. Herbold, D.E. Stevens, and L.W. Miller. 1992. Life history and status of delta smelt in the Sacramento-San Joaquin Estuary, California. Transaction of American Fisheries Society 121: pp. 67–77.

Okiyama, M., *Ed.*, 1988. An atlas of the early stages fishes in Japan. Tokai University Press. 1145 pp.

Radtke, L.D. 1966. Distribution of smelt, juvenile sturgeon, and starry flounder in the Sacramento-San Joaquin Delta with observations on food of sturgeon, in Ecological studies of the Sacramento-San Joaquin Delta, Part II. (J.L. Turner and D.W. Kelley, editors). California Department of Fish and Game Fish, Bulletin 136. pp. 115–129.

Seesholtz, A. 2004. Personal communication.

Shardin, A.M. 1989. Embryonic and larval development of smelt (Osmeridae) of the Far East IV. *Hypomesus nipponensis.* Journal of Ichthyology. 29(6): pp. 960–972.

Siegfried, S., D. Craft, S. Hiebert, and M. Bowen. 2000. Continuous monitoring of fish eggs and larvae at the Tracy Fish Collection Facility, Tracy, California, February–June 1994. Tracy Fish Collection facility Studies, California. Volume 6. Bureau of Reclamation, Mid-Pacific Region, South Central California Area Office and Denver Technical Services Center. 30 pp. Plus appendix.

Simonson, M. 1977. The use of discrimination function analysis in the identification of two species of larval smelt, *Spirinchus thaleichthys* and *Hypomesus transpacificus*, in the Sacramento-San Joaquin Estuary, California. MS Thesis, University of Pacific, Stockton, California. 53 pp.

Spaar, S.A. 1990a. Results of the 1988 striped bass egg and larva study near the State Water Project and Central Valley Project facilities in the Sacramento-San Joaquin Estuary. IEP Technical Report 25 (FF/BIO-IATR/90-25).

__________ 1990b. Losses of striped bass eggs and larvae at SWP and CVP intakes. pp. 36–49 in 1989 annual report: Interagency Studies Program for the Sacramento-San Joaquin Delta. Department of Water Resources.

__________ 1991. 1990 entrainment of striped bass eggs and larvae to the State Water Project and Central Valley Project facilities in the Sacramento-San Joaquin Delta. Department of Water Resources.

Stanley, S.E., P.B. Moyle, and H.B. Shaffer. 1995. Allozyme analysis of delta smelt, *Hypomesus transpacificus* and longfin smelt, *Spirinchus thaleichthys* in the Sacramento-San Joaquin Estuary, California. Copeia 1995: pp. 390–396.

Swanson, C., T. Reid, P.S. Young, and J. J. Cech Jr. 2000. Comparative environmental tolerances of threatened delta smelt (*Hypomesus transpacificus*) and introduced wakasagi (*Hypomesus nipponensis*) in an altered California estuary. Oecologia (2000) 123: pp. 384–390.

Sweetnam, D.A. 1995. Field identification of delta smelt and wakasagi. IEP Newsletter. Spring 1995: pp. 1–3.

Trenham, P., P.B. Moyle, and H.B. Shaffer. 1994. Electrophoretic identification of smelt Genus *Hypomesus* from Sacramento-San Joaquin Delta. Report to Department of Water Resources, University of California at Davis, Davis, California. 7 pp. Plus tables.

Wales, J.H. 1962. Introduction of pond smelt from Japan to California. California Department of Fish and Game. 48 (2): pp. 141–142.

Wang, J.C.S. 1986. Fishes of the Sacramento-San Joaquin Estuary and adjacent waters, California: A guide to the early life histories. IEP Technical Report 9. 612 pp.

__________ 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.* IEP Technical Report 28. 52 pp.

__________ 1995 Observation of possible larval and prejuvenile and delta/wakasagi hybrids in the Delta and tributary streams. IEP Newsletter. Summer 1995: pp. 18–19.

__________ 1996. Notes on spawning of smelt and splittail in the Sacramento-San Joaquin Estuary in a dry year 1994 and wet year 1995. IEP Newsletter. Summer 1996: pp. 31–35.

Wang, J.C.S., and R.L. Brown. 1993. Observation of early life stages of delta smelt, *Hypomesus transpacificus*, in the Sacramento-San Joaquin Estuary in 1991, with a review of the ecological status in 1998 to 1990. IEP Technical Report 35. 18 pp. Plus appendix.

Wang, J.C.S., and L. Hess. 2000. Similarities between hatcheries reared delta smelt and wild wakasagi from the Sacramento-San Joaquin Delta. IEP Newsletter. Winter 2000: pp. 49–51.

APPENDIX 1

The following describes the voucher collections used to develop this diagnostic key:

Wakasagi:

1. Field or wild specimens:

    a. Various early life stages of the wakasagi were collected mainly by the CDFG at Folsom Lake, Lake Natoma, Lake Almanor, Lake Oroville, and San Luis Reservoir from 1990 to 2001.

    b. Wakasagi larvae were collected from the CDFG's E&L samples at various stations of the Sacramento River from 1990–1995.

    c. Wakasagi larvae were collected from CDFG's E&L samples at Barker Slough, Lindsay Slough, Sacramento Deep Water Shipping Channel, Miner Slough, and Cache Slough by the North Bay Aqueduct (NBA) project from 1993 to the present.

    d. Wakasagi larvae were collected by University of California at Davis' E&L samples at Suisun Marsh, Montezuma Slough, and its tributaries from 1995 to the present.

    e. Most of the prejuvenile and early juvenile life stages of wakasagi were collected (10-minute holding tank fish sampling program) at the CVP and SWP in the Sacramento-San Joaquin Delta from 1990 to the present.

    f. Postlarvae and early juvenile life stages of the wakasagi were collected by the CDFG's 20 mm smelt survey project in the Sacramento-San Joaquin Delta (including the Napa River and eastern part of the San Pablo Bay) in 1995 to the present.

    g. Early life stages of wakasagi were collected at the mid-Napa River (south of downtown Napa) by Stillwater Science, Inc. for the Army Corps of Engineers in 2001.

2. Laboratory specimens:

    Artificial propagation was successfully performed at SWP (Skinner) smelt hatchery and CVP Tracy Aquaculture Facility (TAF) in 2000 and 2001. Series of wakasagi eggs at different developmental stages and at various temperatures were closely monitored. Approximately 10 wakasagi eggs were preserved in 10 percent buffered formalin solution every day from first fertilization until eggs hatched. A similar number of prolarvae was preserved every other day from the day of hatching to the day of yolk sac and oil globule absorption (about 1 week after hatching). Then the postlarvae, prejuvenile, and juvenile were preserved at 5-day intervals, until the fish reached a size of approximately 50.0 mm in total length (TL).

Delta smelt:

1. Field or wild specimens:

   a. Larval and juvenile life stages of the delta smelt were collected by Ecological Analysts, Inc, and then Tenera Energy, Inc. for Pacific Gas and Electric Company, Inc., during 316a and 316b studies in the west Delta between 1978 and 1990.

   b. Larval life stages of the delta smelt were collected by the CDFG's E&L program between 1988 and 1995. These specimens formed the initial foundation of the delta smelt larval fish systematic voucher collections. DWR had a similar E&L program conducted in the central and south Delta between 1988 and 1995.

   c. Larval life stages of the delta smelt were collected in the Barker Slough, Lindsey Slough, Cache Slough, Miner Slough and its vicinities for the NBA project by the CDFG from 1990 to the present.

   d. Larvae of the delta smelt were collected at Suisun Marsh, Montezuma Slough, and its tributaries by University of California at Davis from 1995 to 2002.

   e. Delta smelt larvae were also observed in the E&L samples collected by the CDFG at the fish barrier in the south Delta from 1997 to 2001.

   f. Most of the postlarvae to juvenile life stages of the delta smelt were collected by CDFG's 20-mm smelt survey at various locations in the Delta, the eastern part of the San Pablo Bay, and the lower Napa River from 1995 to the present.

   g. Larvae to juvenile life stages of the delta smelt were collected at the mid-Napa River by Stillwater Science, Inc. for the U.S. Army Corps of Engineers in 2001.

   h. Late postlarvae to juvenile life stages of the delta smelt were collected (10-minute holding tank fish sampling program) by CDFG at SWP (Skinner) and Reclamation at CVP (Tracy Fish Collection Facilities) from 1990 to the present.

2. Laboratory specimens:

a. Various early life stages of the laboratory reared delta smelt specimens were obtained from Joan Lindberg and Bradd Baskerville-Bridges at the Skinner Fish Collection Facility from 1998 to 2001.

b. Fertilized eggs from natural and artificial reproduction at CVP (Tracy Fish Collection Facility) in 2000 and 2001. Several complete series of the delta smelt eggs and larvae were prepared and preserved (10 percent formalin) for genetic verification as for the wakasagi specimens.