Appendix 1. Bighead Carp

These pictures are images of live and preserved specimens of bighead carp (*Hypophthalmichthys nobilis*). Text descriptions and line drawings from Chapman and Wang (2006), and Yi and others (1988) are also presented, when available, for comparison purposes. Although Yi and others (1988) describe 45 stages of development, data are presented on only the first 40 stages.

Preliminary data on timing and required cumulative thermal units (CTUs) are also presented, though this data is based on a single experiment, and is likely to be revised with further experiments. The magnification listed is the total magnification (ocular x objective) for each picture.

Table 1–1. Timing and required CTUs for achieving developmental stages of bighead carp.

[This data is based on a single experiment, and is likely to be revised with further experiments. CTU, cumulative thermal unit; °C, degrees Celsius; T_{min}, thermal minimum; --, not available]

Stage		Time			CTU (Calculated using 14.0°C as T_{min})		
		Yi	High	Low	Yi (21°C)	High (22.3°C)	Low (20.1°C)
1	1-cell	--	0	0	--	0	0
2	2-cell	0:55	0:47	1:12	6.05	6.5	7.32
3	4-cell	1:03	1:05	1:58	6.93	8.99	12.00
4	8-cell	1:12	1:32	--	7.92	12.73	--
5	16-cell	1:24	1:48	2:37	9.24	14.94	18.30
6	32-cell	2:00	2:31	--	13.20	20.89	--
7	64-cell	2:20	--	3:29	15.40	--	21.25
8	128-cell	3:00	3:11	4:34	19.80	26.42	27.86
9	Morula	3:38	4:04	4:59	23.98	33.75	34.36
10	Early blastula	4:10	4:38	6:02	27.50	38.46	36.80
11	Mid-blastula	5:30	5:06	8:49	36.30	42.33	53.78
12	Late blastula	7:20	6:30	9:42	48.40	53.95	59.17
13	Early gastrula	8:10	8:00	10:21	53.90	66.40	63.14
14	Mid-gastrula	9:55	10:07	13:18	65.45	83.97	81.13
15	Late gastrula	11:20	12:10	15:16	74.80	100.98	93.13
16	Neurula	12:45	12:53	16:17	84.15	106.93	99.33
17	Blastopore closure	14:00	13:26	18:34	92.40	111.50	113.26
18	Somite appearance	14:50	14:22	19:59	97.90	119.24	121.90
19	Optic primordium	15:40	15:20	21:38	103.40	127.27	131.96
20	Optic vesicle	16:35	16:20	22:06	109.45	135.57	134.81
21	Olfactory placode	17:45	17:58	23:16	117.15	149.12	141.92
22	Tail bud	19:05	18:57	23:54	125.95	157.29	145.79
23	Otic capsule	19:35	19:23	24:48	129.25	160.88	151.28
24	Tail vesicle	22:00	19:47	27:17	145.20	164.20	166.53
25	Caudal fin	24:28	21:20	29:16	161.48	177.07	178.53
26	Lens formation	25:20	21:57	30:23	167.20	182.19	185.34
27	Muscular effect	26:25	24:19	32:33	174.35	201.83	198.56
28	Heart rudiment	28:40	25:22	33:50	189.20	210.54	206.38
29	Otolith appearance	32:30	26:53	35:45	214.50	223.13	218.08
30	Heart pulsation	33:00	28:48	38:48	217.80	239.04	236.68
31	Hatching	39:00	31:11	45:05	257.40	255.70	275.01
32	Pectoral fin bud	49:00	36:34	--	323.40	299.85	--
33	Gill arch	59:00	--	68:35	389.40	--	418.36
34	Xanthic eye	65:00	68:40	80:23	429.00	563.07	490.34
35	Gill filament	76:00	76:07	99:45	501.60	624.16	608.48
36	Melanoid eye	98:00	92:29	106:49	646.80	758.36	651.58
37	Gas bladder emergence	111:00	117:11	131:52	732.60	960.90	804.39
38	One chamber gas bladder	135:00	131:53	167:07	891.00	1,081.44	1,019.42
39	Dorsal fin differentiation	166:00	--	213:00	1,095.60	--	1,299.30
40	Yolk absorption	202:00	189:48	261:00	1,333.20	1,556.36	1,592.10

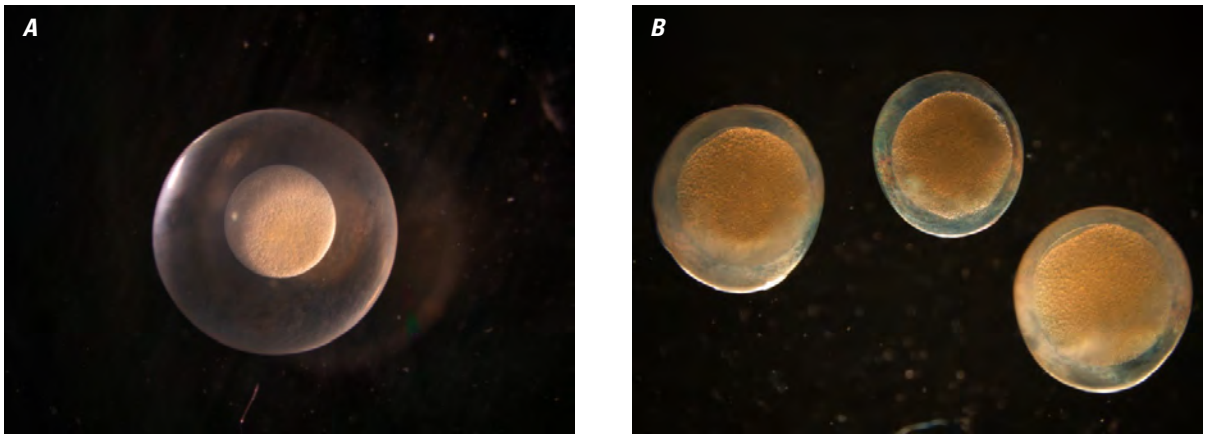


Figure 1–1. Stage 1: 1-cell stage of bighead carp development. *A*, Live specimen, high-temperature treatment, 0:47 post fertilization, picture taken with 20x magnification. *B*, Preserved specimen, high temperature treatment, 0:16 post fertilization, picture taken with 40x magnification.

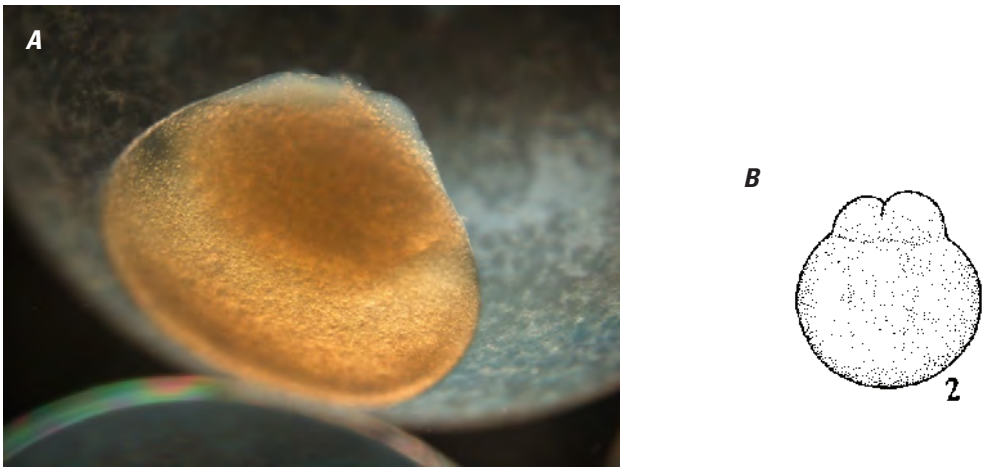


Figure 1–2. Stage 2: 2-cell stage of bighead carp development. The cytoplasm is widely distributed in the yolk (Yi and others, 1988). *A*, Preserved specimen, low temperature treatment, 1:38 post fertilization, picture taken with 50x magnification. *B*, Line drawing of 2-cell stage from Yi and others (1988).

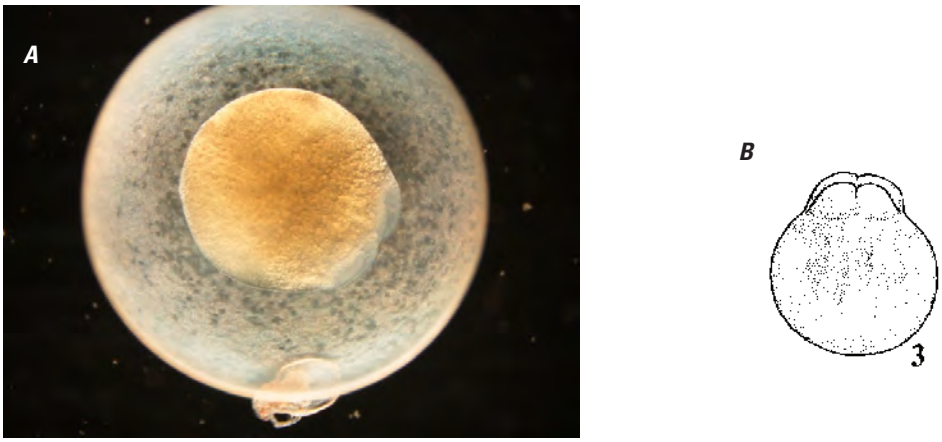


Figure 1–3. Stage 3: 4-cell stage of bighead carp development. Cells divide vertically and the cytoplasm diminishes gradually (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 1:05 post fertilization, picture taken with 40x magnification. *B*, Line drawing of 4-cell stage from Yi and others (1988).

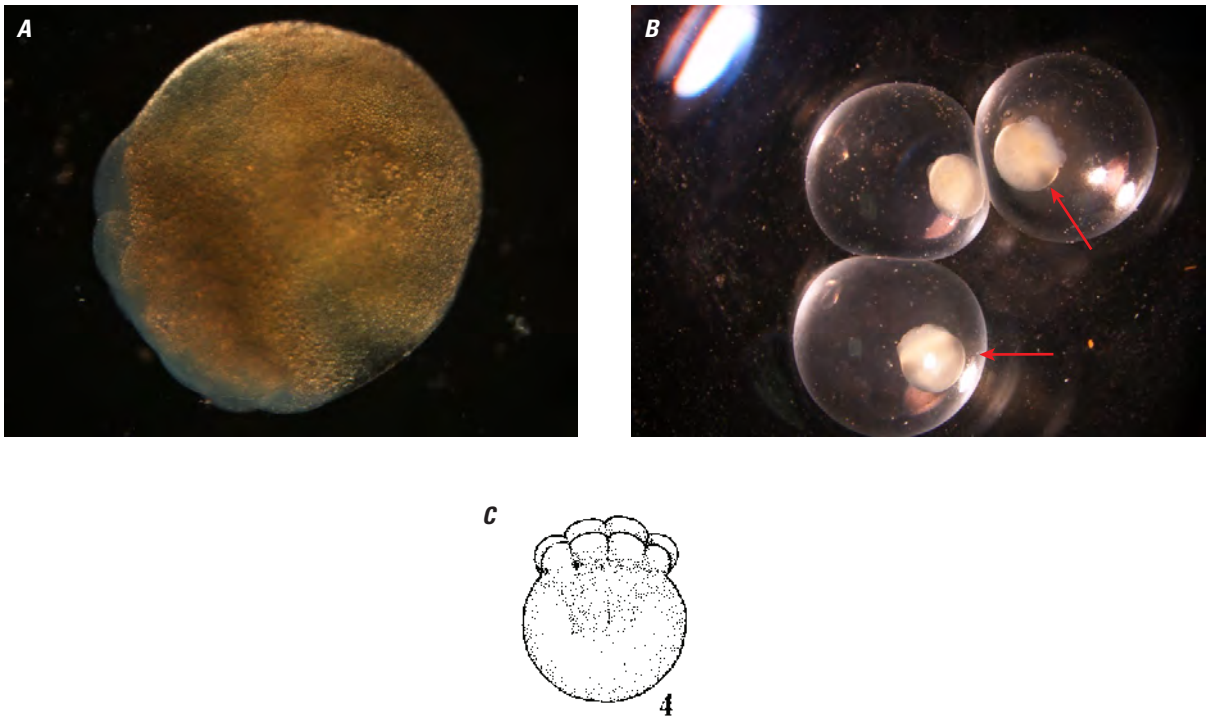


Figure 1–4. Stage 4: 8-cell stage of bighead carp development. Cells divide vertically and the cytoplasm diminishes gradually (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 1:32 post fertilization, picture taken with 50x magnification. *B*, Live specimen, high temperature treatment, 1:32 post fertilization, picture taken with 10x magnification. *C*, Line drawing of 8-cell stage from Yi and others (1988).

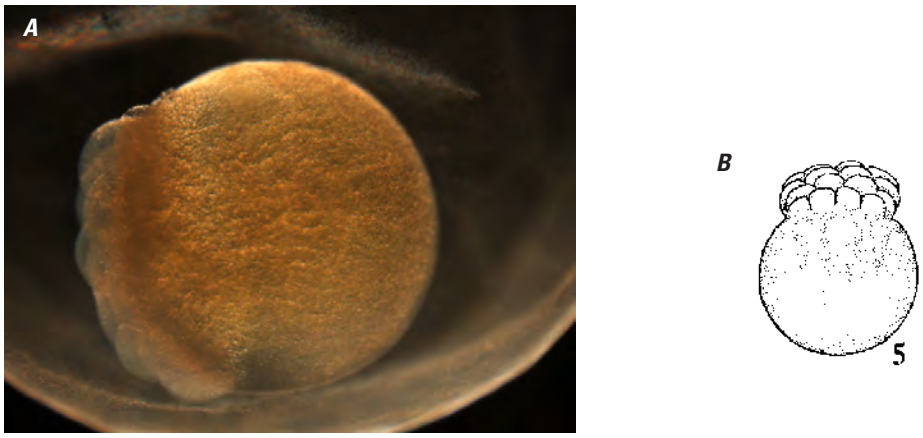


Figure 1–5. Stage 5: 16-cell stage of bighead carp development. Cells divide vertically and the cytoplasm diminishes gradually (Yi and others, 1988). *A*, Preserved specimen, low temperature treatment, 2:37 post fertilization, picture taken with 50x magnification. *B*, Line drawing of 16-cell stage from Yi and others (1988).

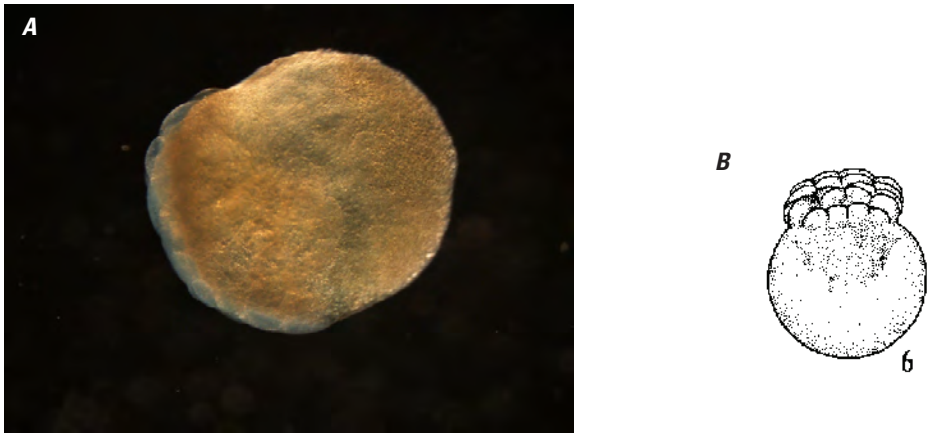


Figure 1–6. Stage 6: 32-cell stage of bighead carp development. Cells divide vertically and the cytoplasm diminishes gradually (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 2:31 post fertilization, picture taken with 40x magnification. *B*, Line drawing of 32-cell stage from Yi and others (1988).

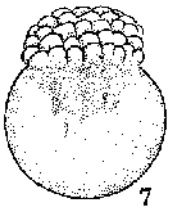


Figure 1–7. Stage 7: 64-cell stage of bighead carp development. Cells divide vertically and the cytoplasm diminishes gradually (Yi and others, 1988). Line drawing of 64-cell stage from Yi and others (1988).

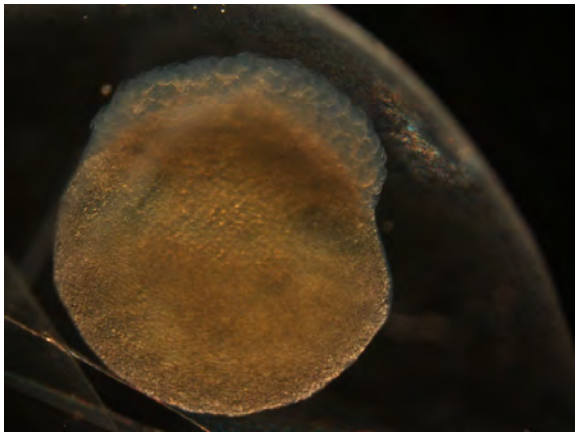


Figure 1–8. Stage 8: 128-cell stage of bighead carp development. Cells divide horizontally (Yi and others, 1988). Preserved specimen, high temperature treatment, 3:11 post fertilization, picture taken with 50x magnification.

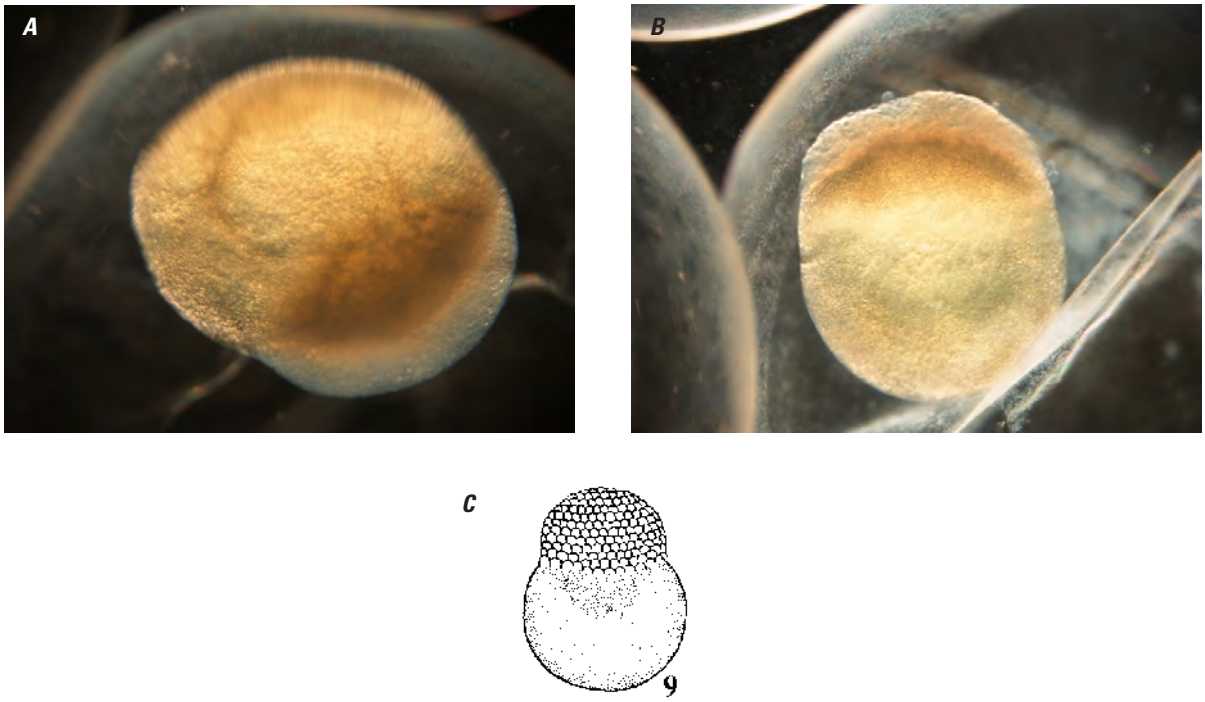


Figure 1–9. Stage 9: Morula stage of bighead carp development. The blastodisc is highly raised above the yolk (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 4:04 post fertilization, picture taken with 50x magnification. *B*, Preserved specimen, low temperature treatment, 4:59 post fertilization, picture taken with 40x magnification. *C*, Line drawing of morula stage from Yi and others (1988).

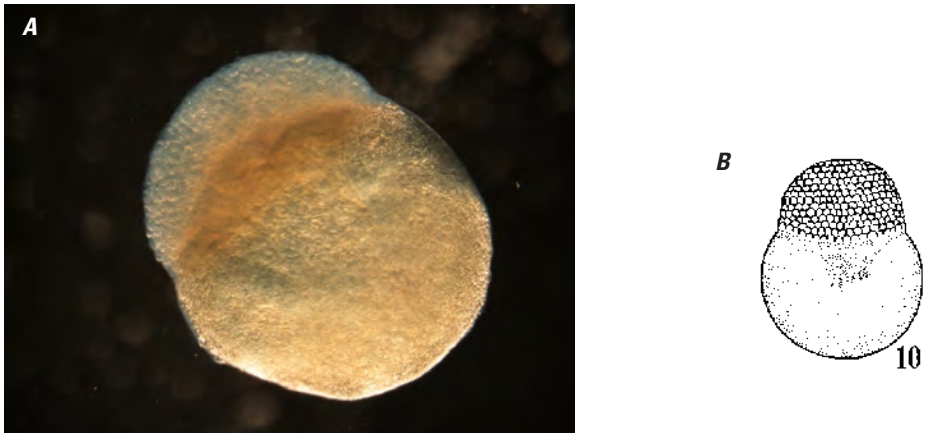


Figure 1–10. Stage10: Early blastula stage of bighead carp development. Blastodisc remains high, cells small (Yi and others, 1988). *A*, Preserved specimen, low temperature treatment, 6:02 post fertilization, picture taken with 50x magnification *B*, Line drawing of early blastula stage from Yi and others (1988).

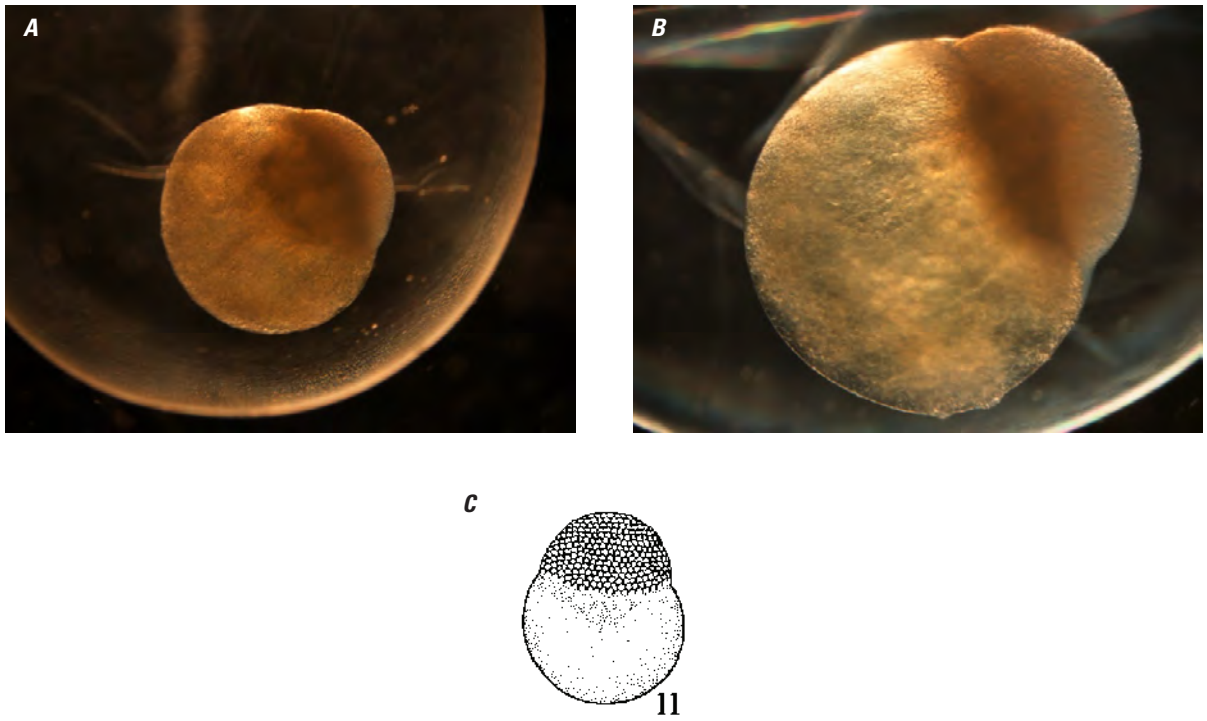


Figure 1–11. Stage 11: Mid-blastula stage of bighead carp development. Blastodisc begins flattening (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 5:55 post fertilization, picture taken with 30x magnification. *B*, Preserved specimen, low temperature treatment, 9:11 post fertilization, picture taken with 60x magnification. *C*, Line drawing of mid-blastula stage from Yi and others (1988).

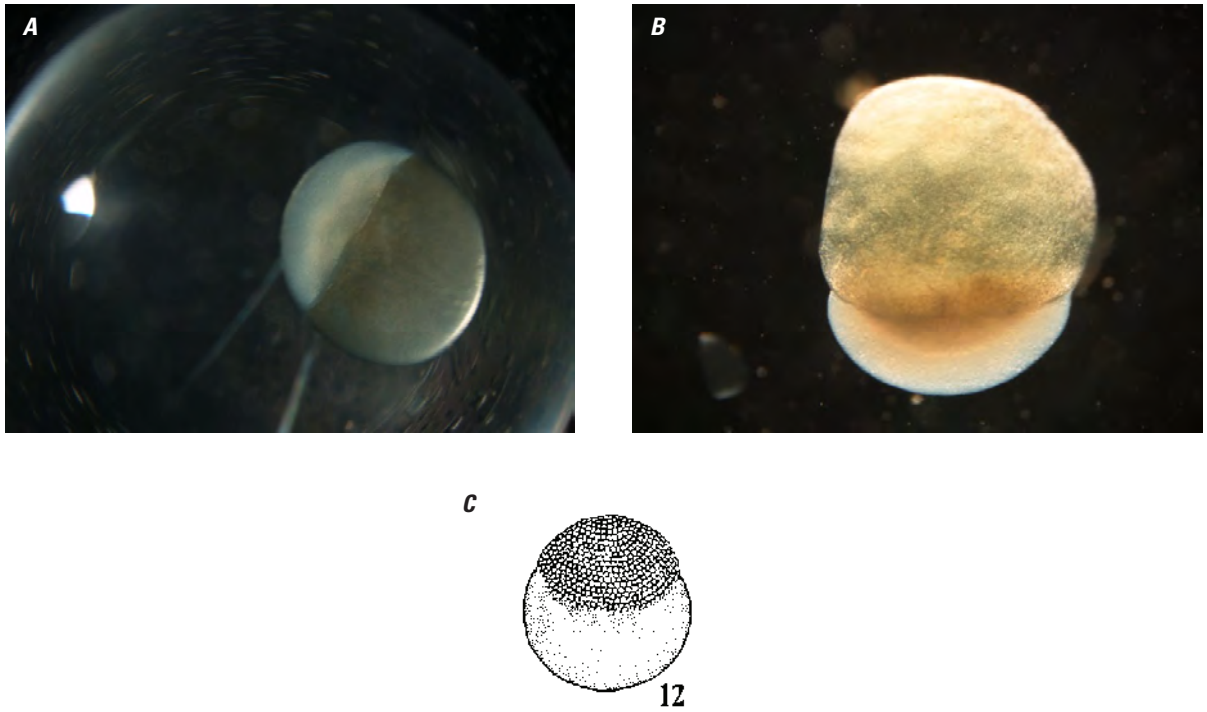


Figure 1–12. Stage 12: Late blastula stage of bighead carp development. Blastodisc gradually expands over yolk, cytoplasm almost disappears (Yi and others, 1988). *A*, Live specimen, low temperature treatment, 9:47 post fertilization, picture taken with 30x magnification. *B*, Preserved specimen, low temperature treatment, 9:42 post fertilization, picture taken with 40x magnification. *C*, Line drawing of late blastula stage from Yi and others (1988).

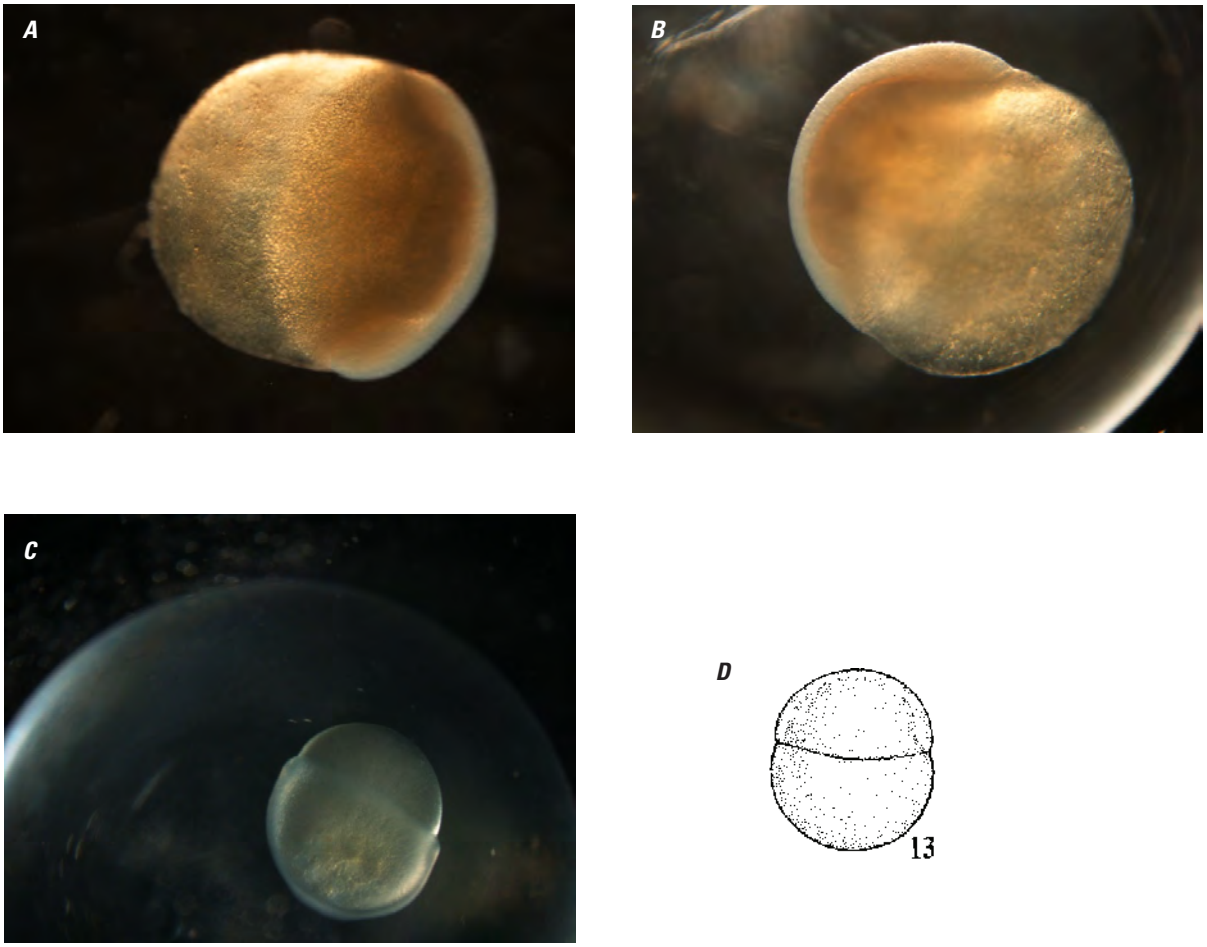


Figure 1–13. Stage 13: Early gastrula stage of bighead carp development. Egg looks round, dorsal lip appears, germ ring forms, blastoderm covers 1/3 of yolk (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 8:51 post fertilization, picture taken with 50x magnification. *B*, Preserved specimen, high temperature treatment, 8:00 post fertilization, picture taken with 40x magnification. *C*, Live specimen, high temperature treatment, 10:09 post fertilization, picture taken with 30x magnification. *D*, Line drawing of early gastrula stage from Yi and others (1988).

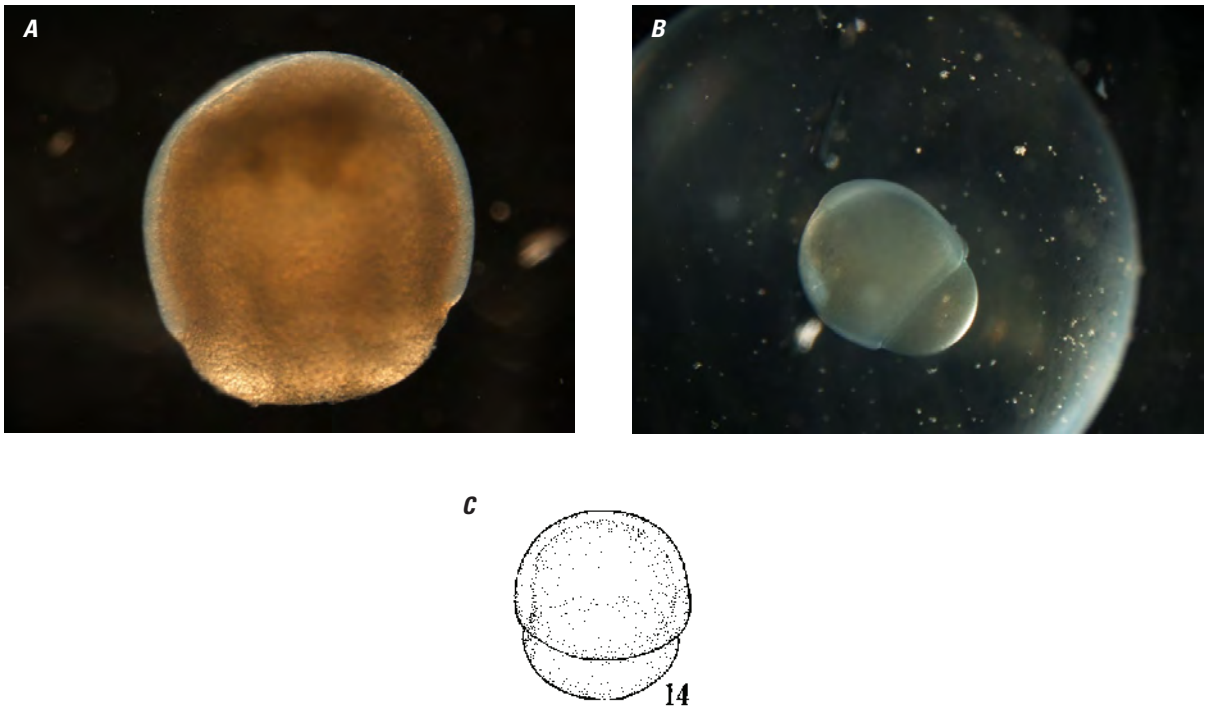


Figure 1–14. Stage 14: Mid-gastrula stage of bighead carp development. Blastoderm covers 2/3 of yolk, embryonic shield appears (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 10:59 post fertilization, picture taken with 60x magnification. *B*, Live specimen, high temperature treatment, 11:03 post fertilization, picture taken with 30x magnification. *C*, Line drawing of mid-gastrula stage from Yi and others (1988).

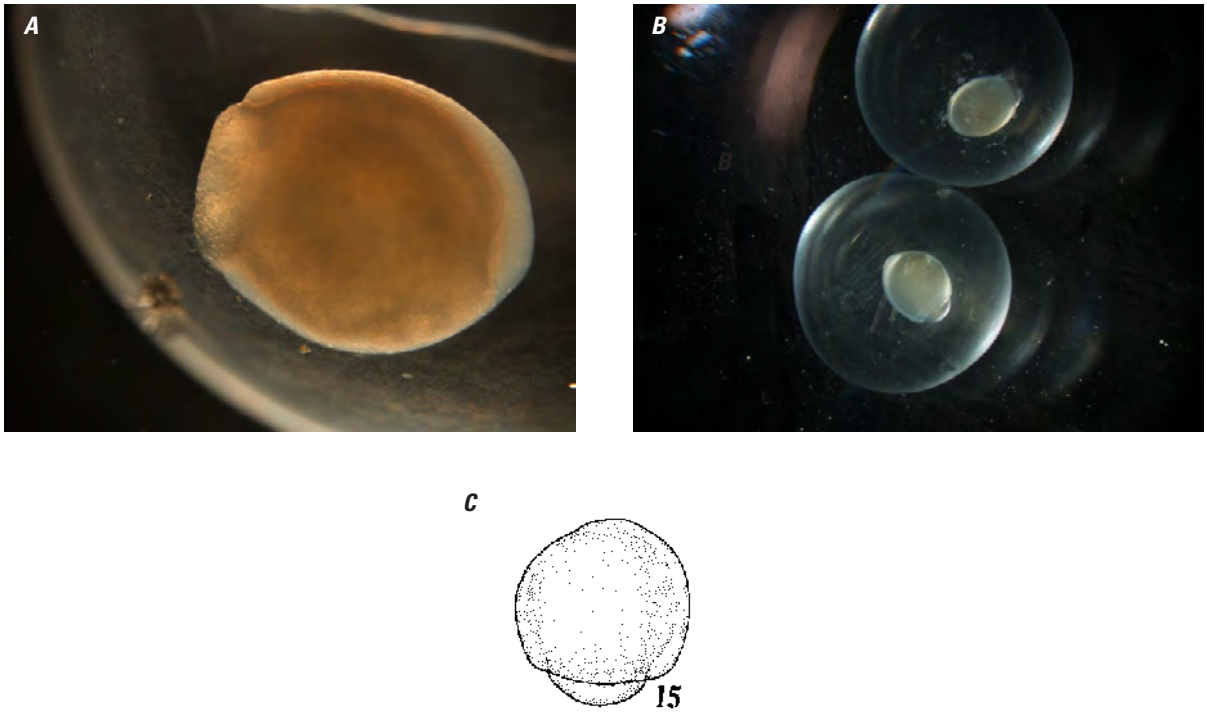


Figure 1–15. Stage 15: Late gastrula stage of bighead carp development. Blastoderm covers 5/6 of yolk, rudimentary embryo visible (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 12:10 post fertilization, picture taken with 50x magnification. *B*, Live specimen, high temperature treatment, 11:35 post fertilization, picture taken with 10x magnification. *C*, Line drawing of late gastrula stage from Yi and others (1988).

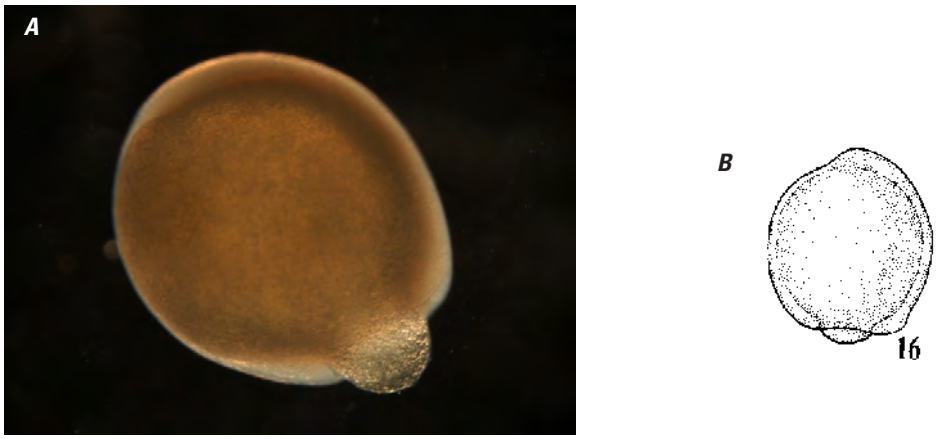
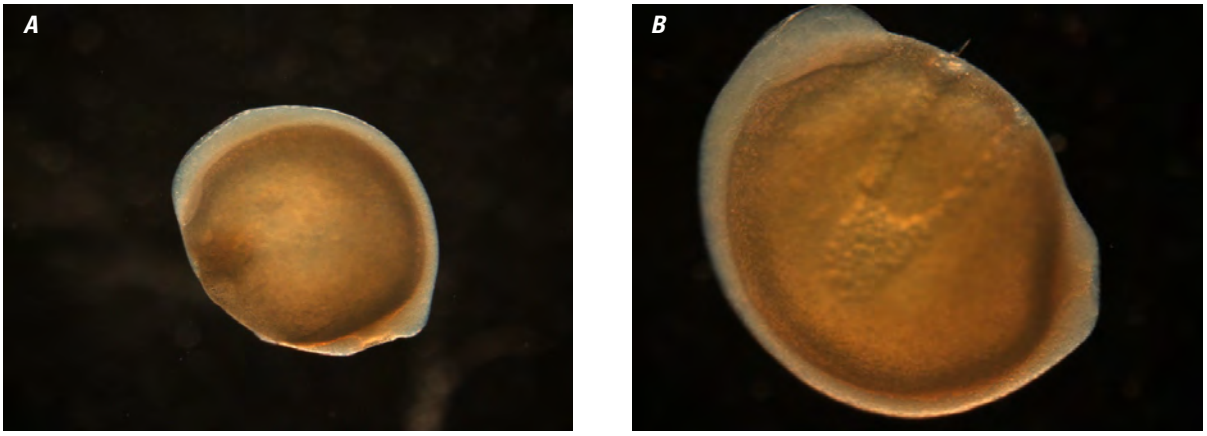


Figure 1–16. Stage 16: Neurula stage of bighead carp development. Head enlarges, blastoderm covers almost entire yolk, except for yolk plug (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 12:53 post fertilization, picture taken with 50x magnification. *B*, Line drawing of neurula stage from Yi and others (1988).



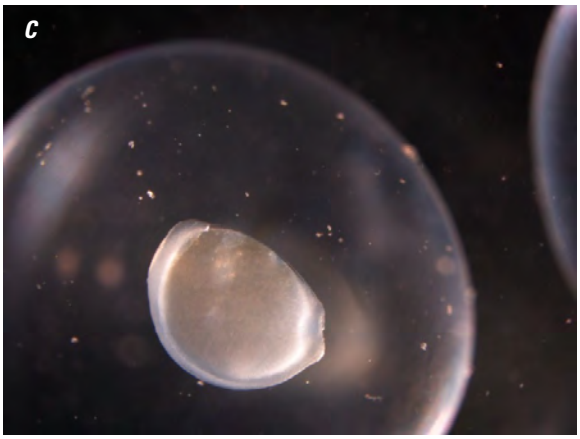


Figure 1–17. Stage 17: Blastopore closure stage of bighead carp development. Front part of embryo head is slightly curved (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 13:26 post fertilization, picture taken with 40x magnification. *B*, Preserved specimen, low temperature treatment, 19:59 post fertilization, picture taken with 60x magnification. *C*, Live specimen, high temperature treatment, 13:26 post fertilization, picture taken with 30x magnification.

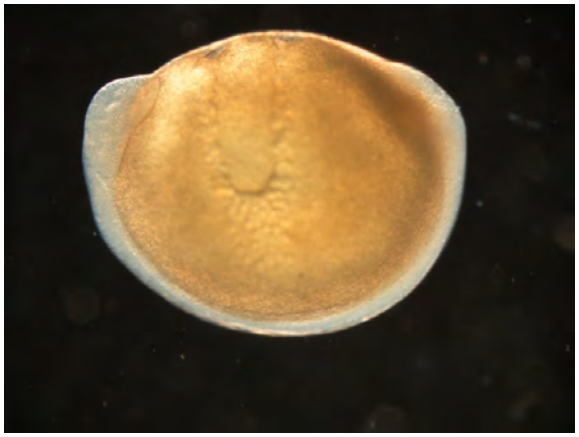
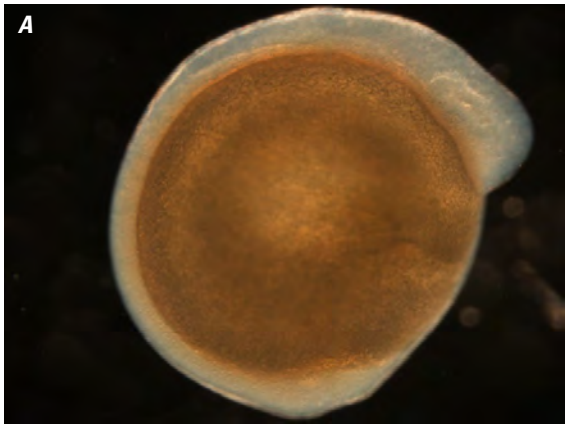


Figure 1–18. Stage 18: Somite appearance stage of bighead carp development, 1–3 pairs of somites present, head is round with a flat front margin (Yi and others, 1988). Preserved specimen, low temperature treatment, 19:59 post fertilization, picture taken with 50x magnification.



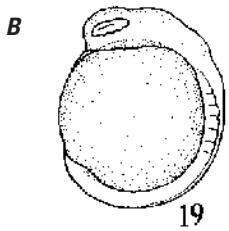


Figure 1–19. Stage 19: Optic primordium stage of bighead carp development. Optic primordium dimly visible as a long oval shape with a slightly crenulated lower margin. Embryo embraces 3/4 of yolk, no oil droplets in yolk, 4–6 pairs of somites present (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 15:20 post fertilization, picture taken with 50x magnification. *B*, Line drawing of optic primordium stage from Yi and others (1988).

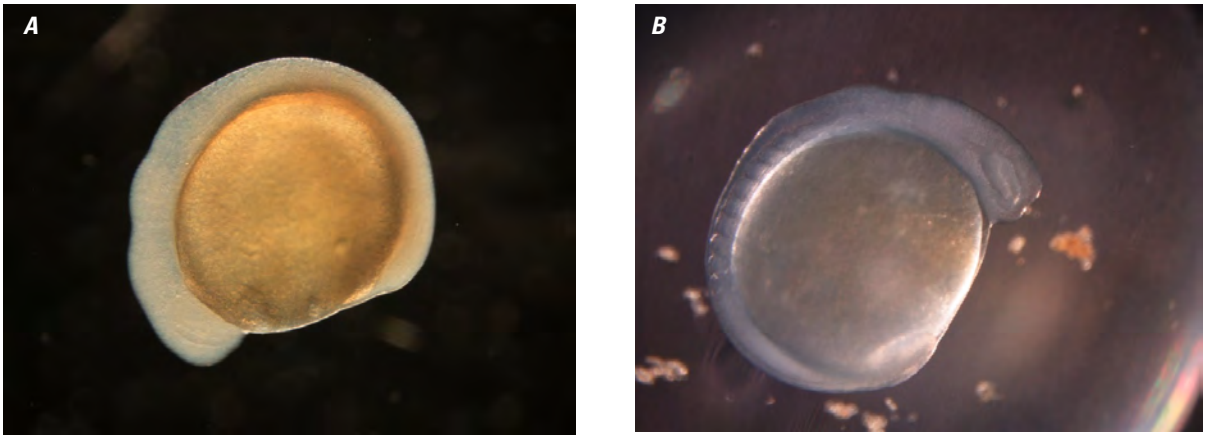


Figure 1–20. Stage 20: Optic vesicle stage of bighead carp development. Eyes enlarge, 7–10 pairs of somites present (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 16:20 post fertilization, picture taken with 40x magnification. *B*, Live specimen, high temperature treatment, 16:52 post fertilization, picture taken with 50x magnification.

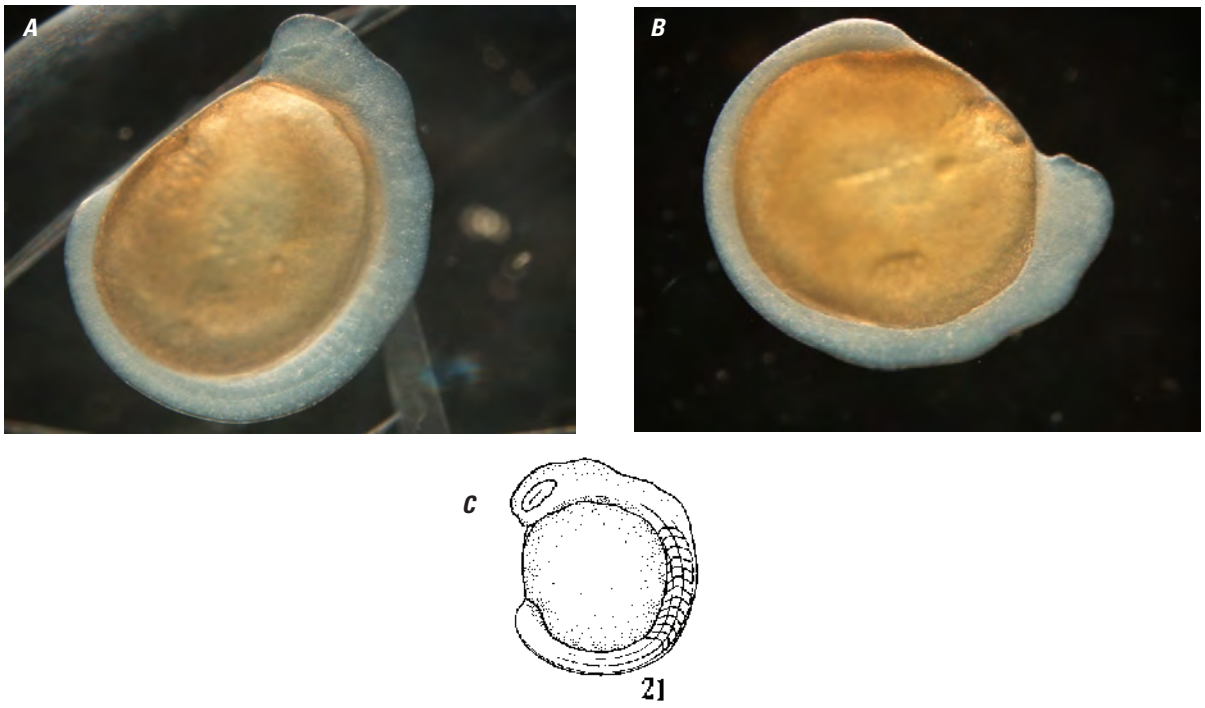


Figure 1–21. Stage 21: Olfactory placode stage of bighead carp development. Eyes enlarging, lower edge of eye still slightly crenulated, olfactory placode appears, notochord clearly visible, 11–14 pairs of somites present (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 17:58 post fertilization, picture taken with 50x magnification. *B*, Preserved specimen, low temperature treatment, 23:16 post fertilization, picture taken with 50x magnification. *C*, Line drawing of olfactory placode stage from Yi and others (1988).

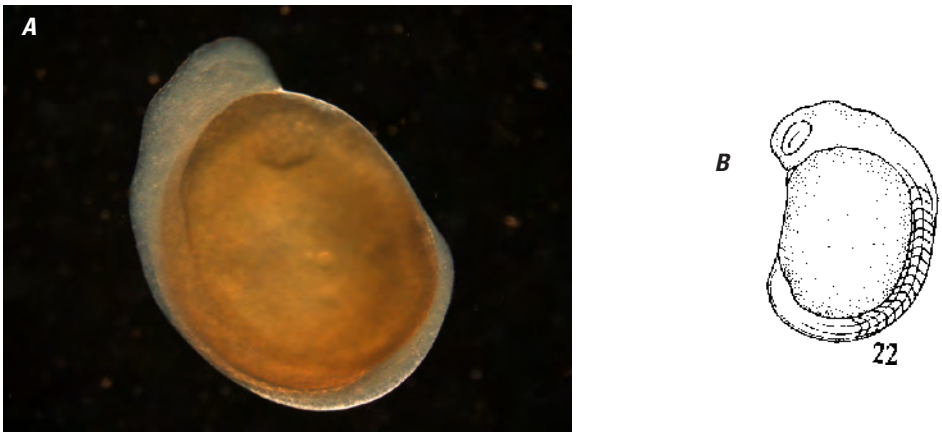


Figure 1–22. Stage 22: Tail bud stage of bighead carp development. Optic vesicle large and clearly visible, tail bud appears, yolk elongates, 15–17 pairs of somites present (Yi and others, 1988). *A*, Preserved specimen, low temperature treatment, 23:54 post fertilization, picture taken with 50x magnification. *B*, Line drawing of tail bud stage from Yi and others (1988).

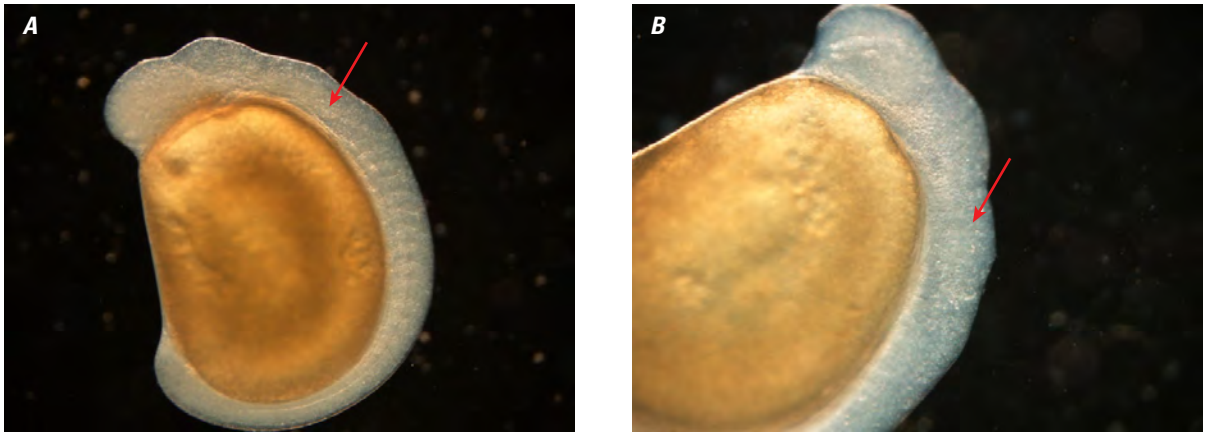


Figure 1–23. Stage 23: Otic capsule stage of bighead carp development. Yolk between head and tail begins invagination, 18–19 pairs of somites present (Yi and others, 1988). *A*, Preserved specimen, low temperature treatment, 26:18 post fertilization, picture taken with 50x magnification. Arrow points to the otic capsule. *B*, Preserved specimen, low temperature treatment, 25:16 post fertilization, picture taken with 60x magnification. Arrow points to the otic capsule.

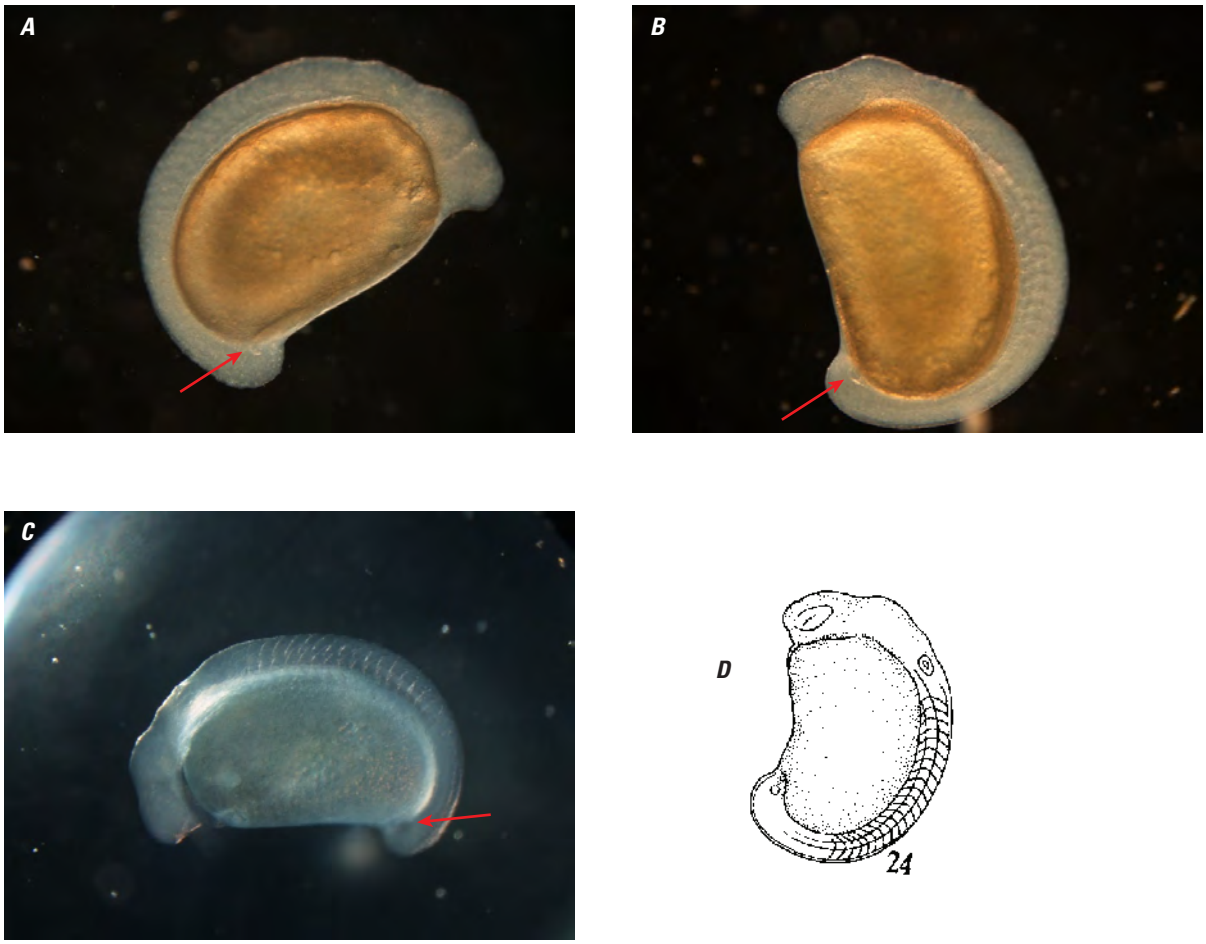


Figure 1–24. Stage 24: Tail vesicle stage of bighead carp development. Otic capsule clearly visible, tail bud expands outward, tail vesicle appears, embryo becomes thicker, yolk elongates and becomes kidney shaped, 20–21 pairs of somites present (Yi and others, 1988). *A*, Preserved specimen, low temperature treatment, 28:52 post fertilization, picture taken with 40x magnification. Arrow points to the tail vesicle. *B*, Preserved specimen, high temperature treatment, 19:47 post fertilization, picture taken with 40x magnification. Arrow points to the tail vesicle. *C*, Live specimen, high temperature treatment, 20:06 post fertilization, picture taken with 40x magnification. Arrow points to the tail vesicle. *D*, Line drawing of tail vesicle stage from Yi and others (1988).

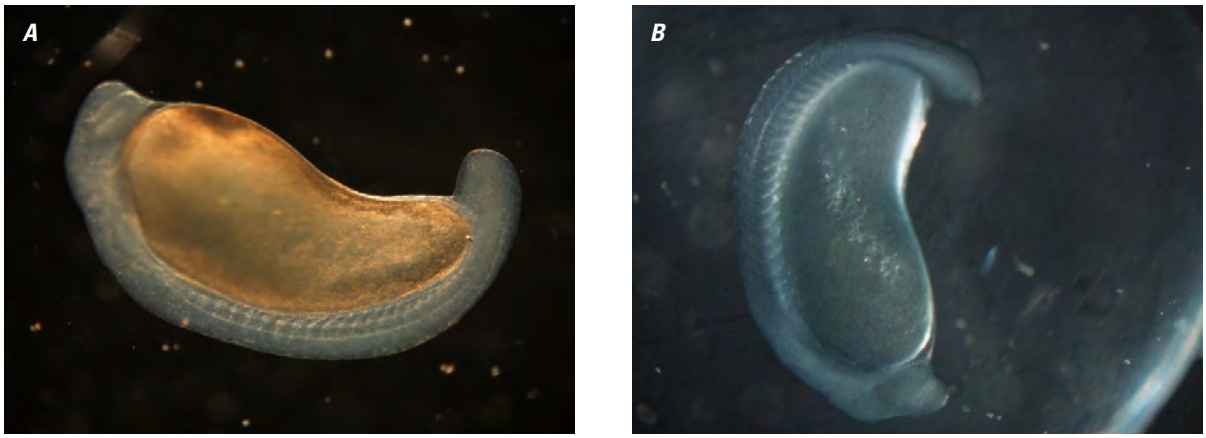


Figure 1–25. Stage 25: Caudal fin stage of bighead carp development. Caudal fin expands outward, 22–23 pairs of somites present (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 21:20 post fertilization, picture taken with 40x magnification. *B*, Live specimen, high temperature treatment, 20:55 post fertilization, picture taken with 40x magnification.

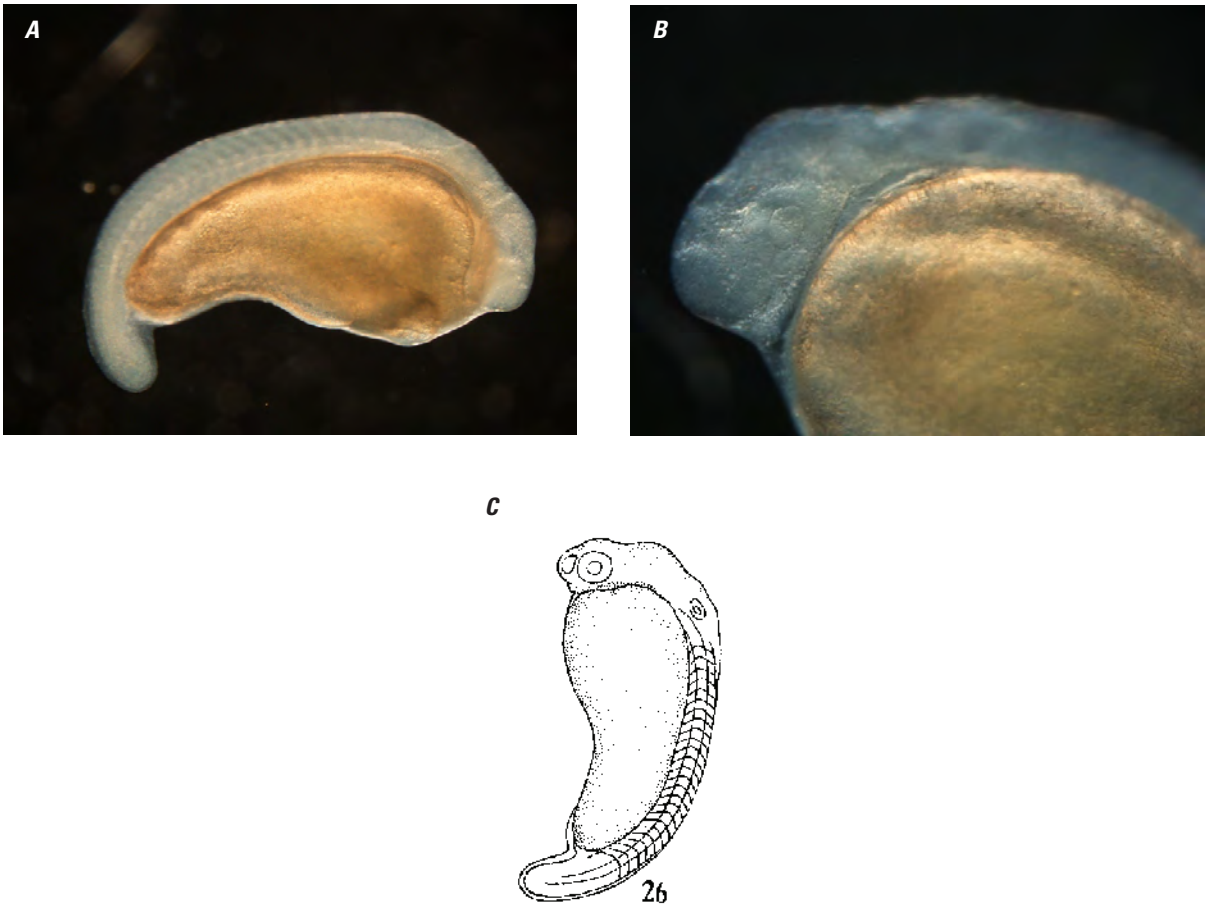


Figure 1–26. Stage 26: Lens formation stage of bighead carp development. Olfactory vesicle clearly visible; lens forms, embryo and yolk elongate. Bump on brain area slightly raised, tail vesicle disappears, 24 pairs of somites present (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 21:57 post fertilization, picture taken with 40x magnification. *B*, Preserved specimen, high temperature treatment, 24:19 post fertilization, picture taken with 100x magnification. *C*, Line drawing of lens formation stage from Yi and others (1988).

Figure 1–27. Stage 27: Muscular effect stage of bighead carp development. Embryo lashes slightly, end of yolk–light gray/blue, 25–27 pairs of somites present (Yi and others 1988). Preserved specimen, low temperature treatment, 32:33 post fertilization, picture taken with 40x magnification.

Figure 1–28. Stage 28: Heart rudiment stage of bighead carp development. Embryo lashes occasionally, 28–29 pairs of somites present (Yi and others, 1988). Preserved specimen, high temperature treatment, 27:24 post fertilization, picture taken with 30x magnification (Yi and others, 1988).

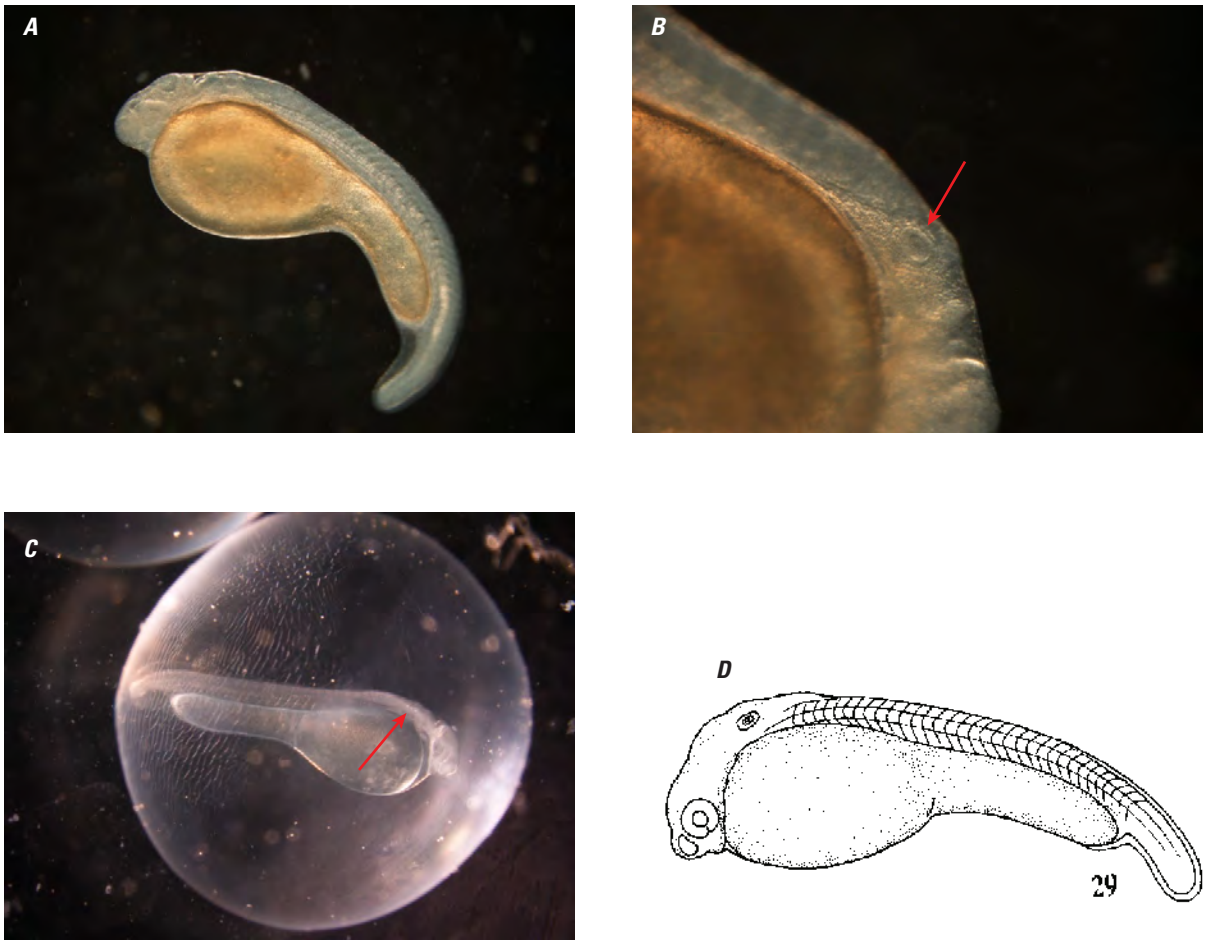


Figure 1–29. Stage 29: Otolith appearance stage of bighead carp development. Otolith appears, embryo further elongates, tail relatively long and often moves to one side, embryo rotates occasionally, anterior half of yolk sac oval and wide, whereas posterior half is narrow, 30–32 pairs of somites present (Yi and others, 1988). *A*, Preserved specimen, low temperature treatment, 37:45 post fertilization, picture taken with 30x magnification. *B*, Preserved specimen, low temperature treatment, 35:45 post fertilization, picture taken with 80x magnification. Arrow points to otoliths. *C*, Live specimen,high temperature treatment, 27:24 post fertilization, picture taken with 20x magnification. Arrow points to otoliths. *D*, Line drawing of otolith appearance stage from Yi and others (1988).

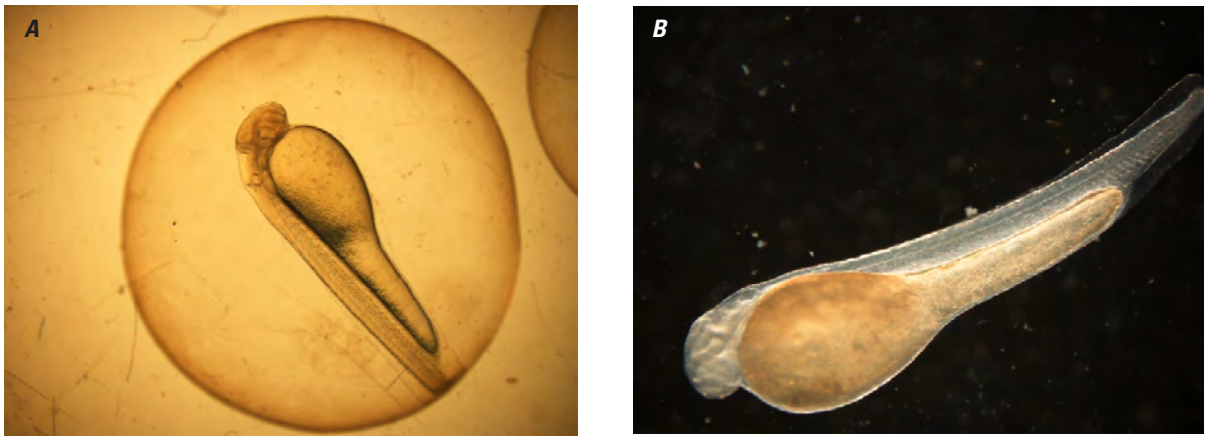


Figure 1–30. Stage 30: Heart pulsation stage of bighead carp development. Heart begins to pulsate; embryo rotates continuously. 33–35 pairs somites present (Yi and others, 1988). *A*, Live specimen, low temperature treatment, 45:03 post fertilization, picture taken with 20x magnification. *B*, Preserved specimen, low temperature treatment, 41:41 post fertilization, picture taken with 30x magnification.

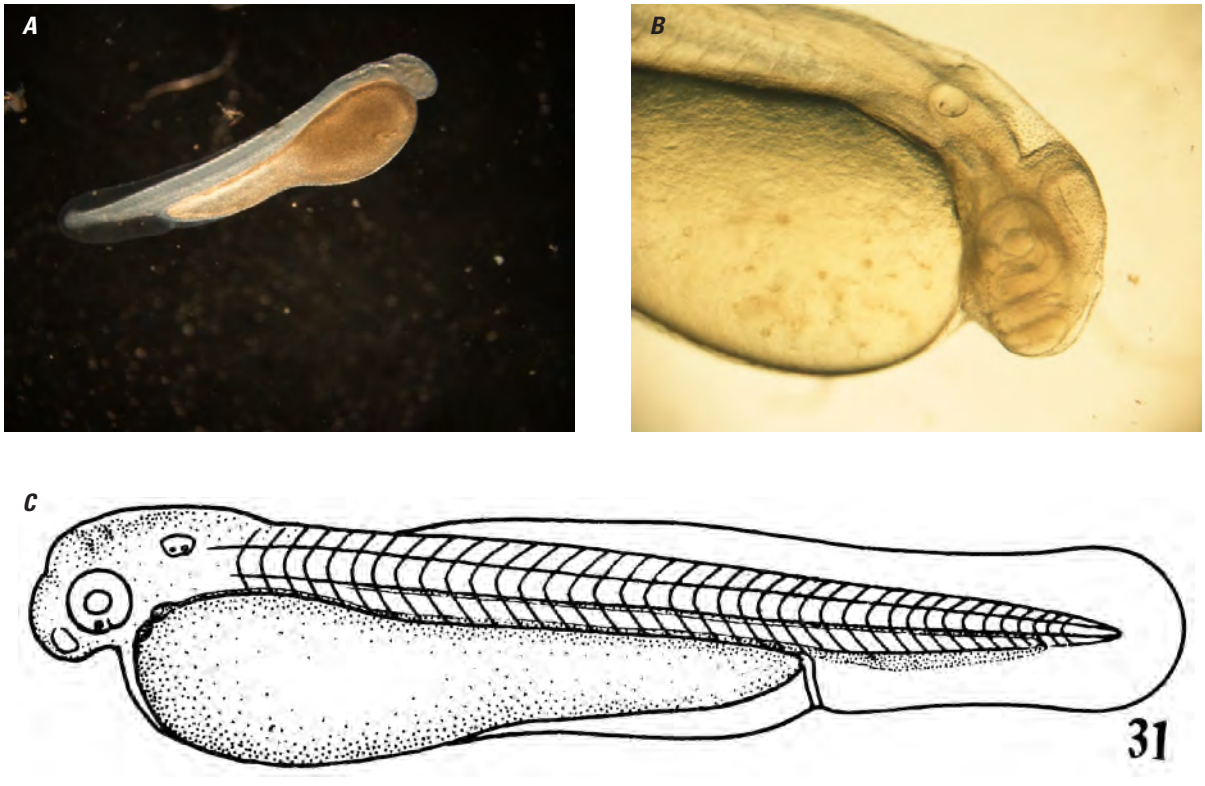


Figure 1–31. Stage 31: Hatching stage of bighead carp development. The larvae is approximately 7.0 millimeters; tail length long; head extends straight forward; anterior part of yolk sac large and oval, posterior part of yolk sac narrow and lengthened tear drop shape. Heart at the top of anterior edge of yolk sac. Black spot appears below eye, caudal vein large and flat (Yi and others, 1988). The black spot below eye was not apparent until stage 32. *A*, Preserved specimen, high temperature treatment, 31:11 post fertilization, picture taken with 30x magnification. *B*, Live specimen, low temperature treatment, 45:05 post fertilization, picture taken with 70x magnification. *C*, Line drawing of hatching stage from Yi and others (1988).

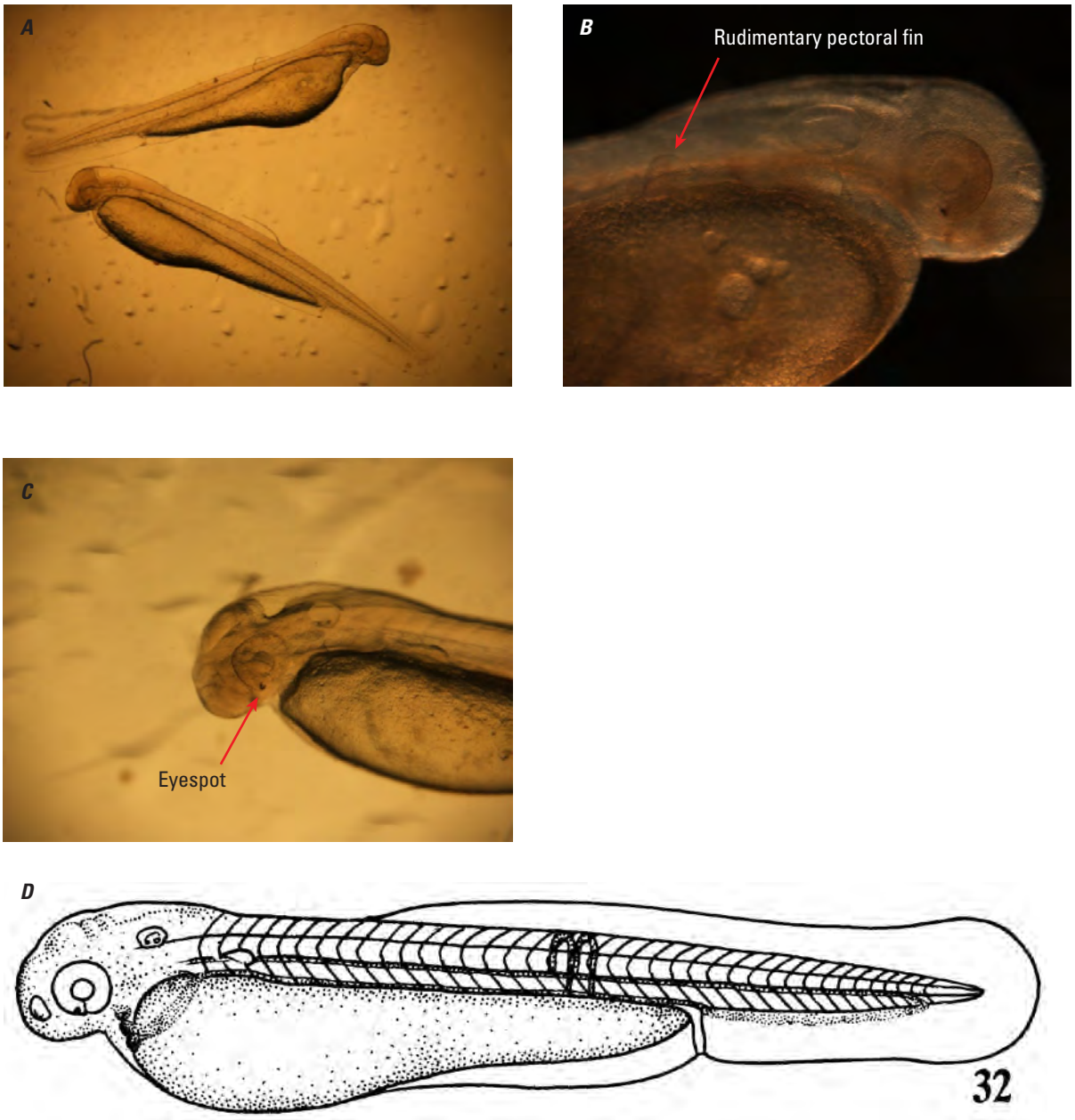


Figure 1–32. Stage 32: Rudimentary pectoral fin stage of bighead carp development. The larvae are approximately 7.1 millimeters; rudimentary pectoral fin appears between myomeres 2 and 3. Caudal vein wide and long. Cuvierian ducts appear on anterior tip of yolk sac (Yi and others, 1988). *A*, Live specimen, high temperature treatment, 42:35 post fertilization, picture taken with 20x magnification. *B*, Preserved specimen, high temperature treatment, 36:34 post fertilization, picture taken with 80x magnification. Arrow points to the rudimentary pectoral fin. *C*, Live specimen, high temperature treatment, 42:35 post fertilization, picture taken with 50x magnification. Arrow points to the eyespot. *D*, Line drawing of rudimentary pectoral fin stage from Yi and others (1988).

Figure 1–33. Stage 33: Gill arch stage of bighead carp development. The larvae are 7.5 millimeters; four gill arches appear. Indentation of mouth appears; caudal vein and cuvierian ducts obvious. Yolk sac shrinks and anterior potion narrows (Yi and others, 1988). *A*, Live specimen, low temperature treatment, 68:35 post fertilization, picture taken with 30x magnification. *B*, Line drawing of gill arch stage from Yi and others (1988).

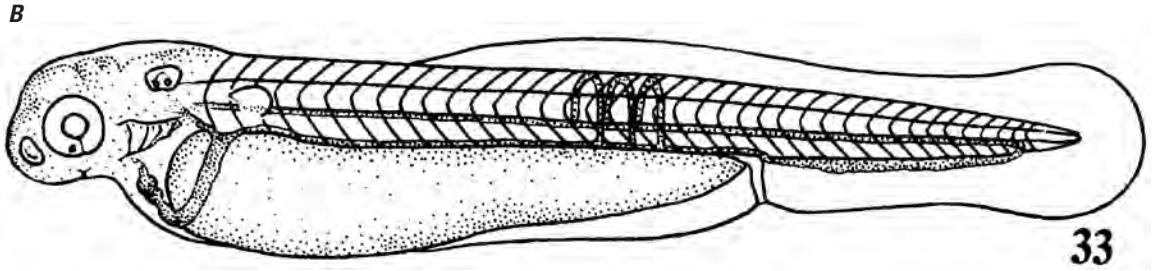


Figure 1–34. Stage 34: Xanthic eye stage of bighead carp development. The larvae are approximately 7.8 millimeters; yellow pigmentation of eye appears, mouth open and can move. Lower edge of caudal vein crenulated (Yi and others, 1988). *A*, Preserved specimen, low temperature treatment, 80:23 post fertilization, picture taken with 60x magnification. *B*, Line drawing of xanthic eye stage from Yi and others (1988).

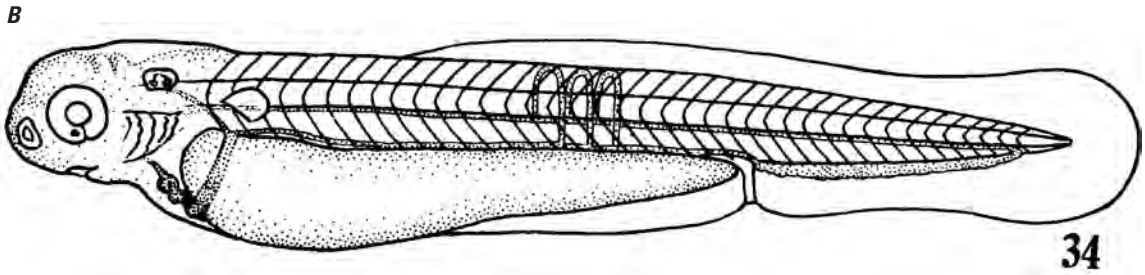
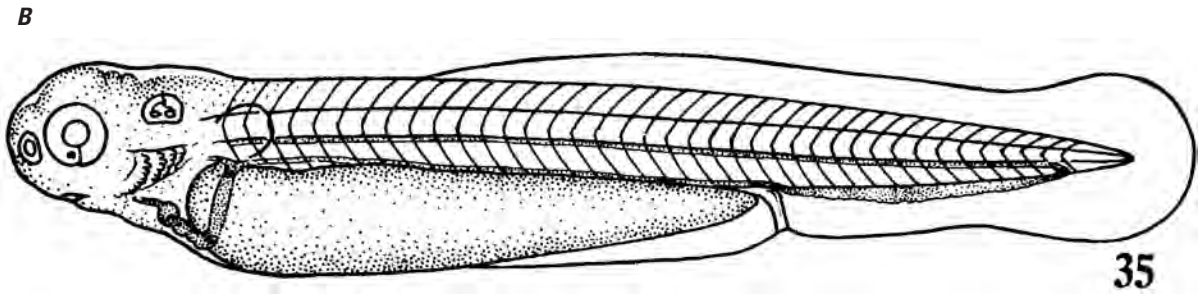


Figure 1–35. Stage 35: Gill filament stage of bighead carp development. The larvae is approximately 8.1 millimeters; gill filaments appear, otic capsule and pectoral fins enlarge. Body relatively thick (Yi and others, 1988). *A*, Live specimen, low temperature treatment, 99:45 post fertilization, picture taken with 50x magnification. *B*, Line drawing of gill filament stage from Yi and others (1988).



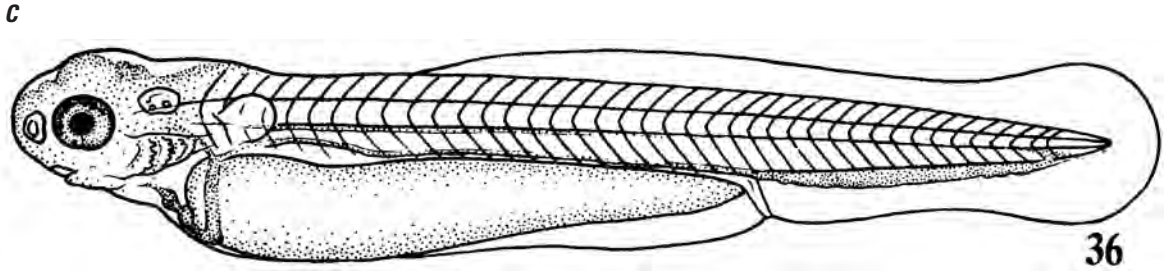


Figure 1–36. Stage 36: Melanoid eye stage of bighead carp development. The larvae is approximately 8.3 millimeters; melanophores appear around eye. Caudal fin wide, long, dark yellow. Normal swimming begins (Yi and others, 1988). *A*, Live specimen, high temperature treatment, 79:50 post fertilization, picture taken with 40x magnification. *B*, Preserved specimen, high temperature treatment, 99:43 post fertilization, picture taken with 20x magnification. *C*, Line drawing of melanoid eye stage from Yi and others (1988).

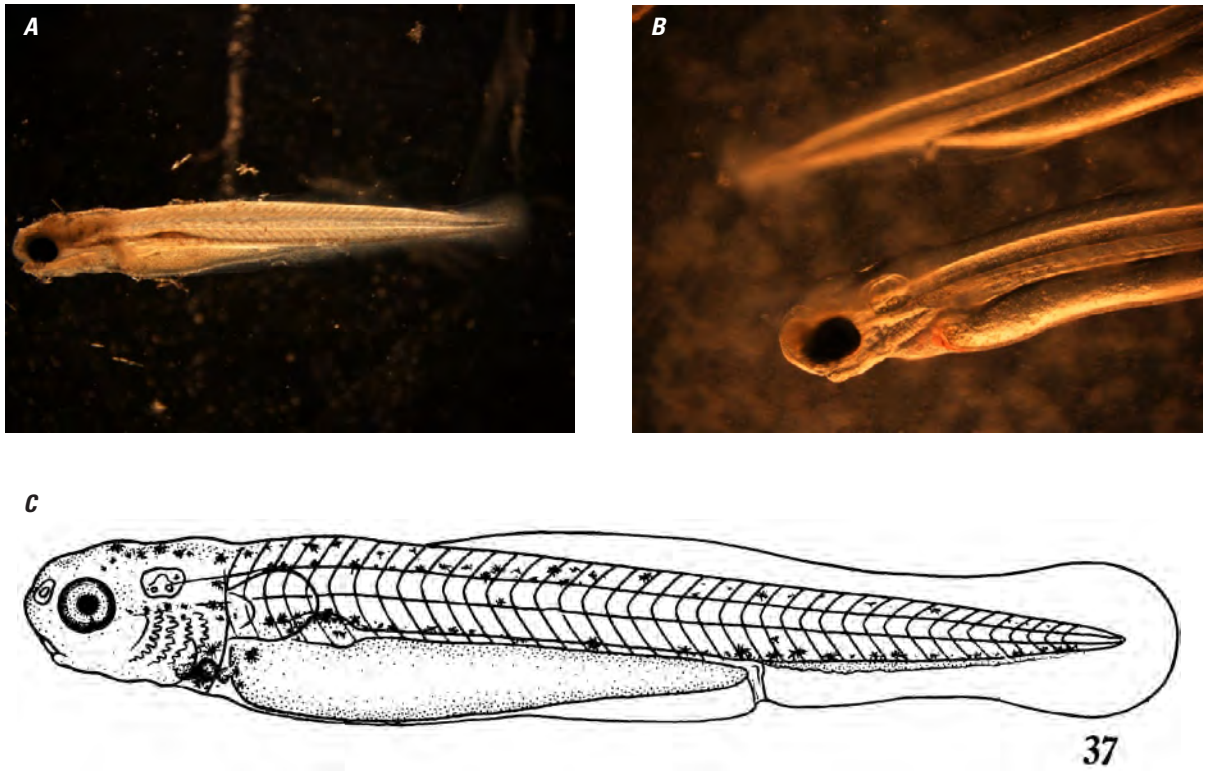


Figure 1–37. Stage 37: Gas bladder emergence stage of bighead carp development. The larvae is approximately 9.2 millimeters; blunt snout, initial gas bladder appears. Gill filaments extend, gut continuous, increasing pigmentation. Swims normally (Yi and others, 1988). *A*, Preserved specimen, high temperature treatment, 117:11 post fertilization, picture taken with 20x magnification. *B*, Live specimen, high temperature treatment, 117:11 post fertilization, picture taken with 30x magnification. *C*, Line drawing of gas bladder emergence stage from Yi and others (1988).

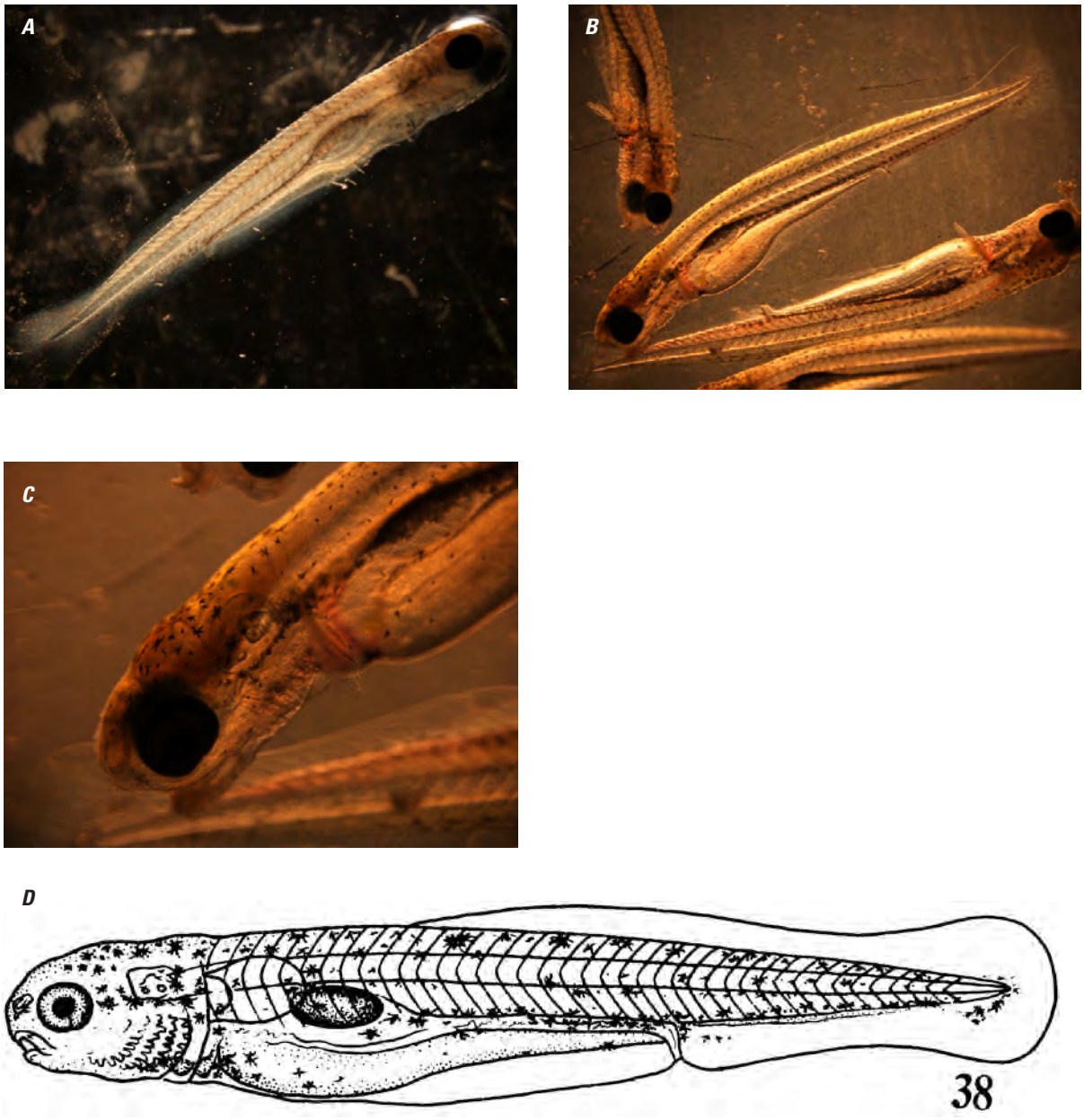


Figure 1–38. Stage 38: 1-chamber gas bladder stage. The larvae is approximately 9.4 millimeters; the gas bladder appears and is oval. Inside of gut appears wavy. Yolk sac shrinks to become a curved strip. Upper jaw forms. Pigment on head increases. Pre-anal finfold extends forward to posterior margin of gas bladder (Yi and others, 1988). *A*, Preserved specimen, low temperature treatment, 189:46 post fertilization, picture taken with 20x magnification. *B*, Live specimen, high temperature treatment, 141:21 post fertilization, picture taken with 20x magnification. *C*, Live specimen, high temperature treatment, 141:21 post fertilization, picture taken with 50x magnification. *D*, Line drawing of one chamber gas bladder stage from Yi and others (1988).

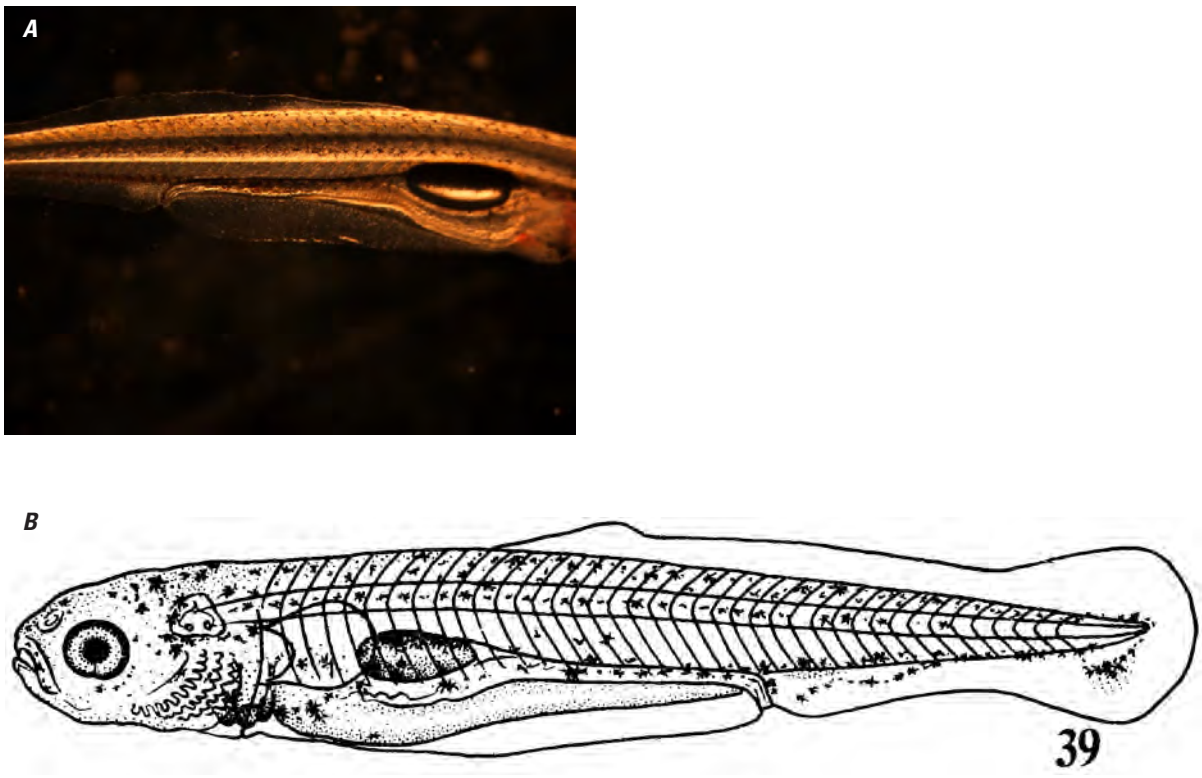


Figure 1–39. Stage 39: Dorsal fin differentiation stage of bighead carp development. The larvae is approximately 9.7 millimeters; dorsal margin of anterior part of dorsal finfold has a sigmoid shape. Narrow strip of yolk sac remains. *A*, Preserved specimen, low temperature treatment, 213:00 post fertilization, picture taken with 30x magnification. *B*, Line drawing of dorsal fin differentiation stage from Yi and others (1988).

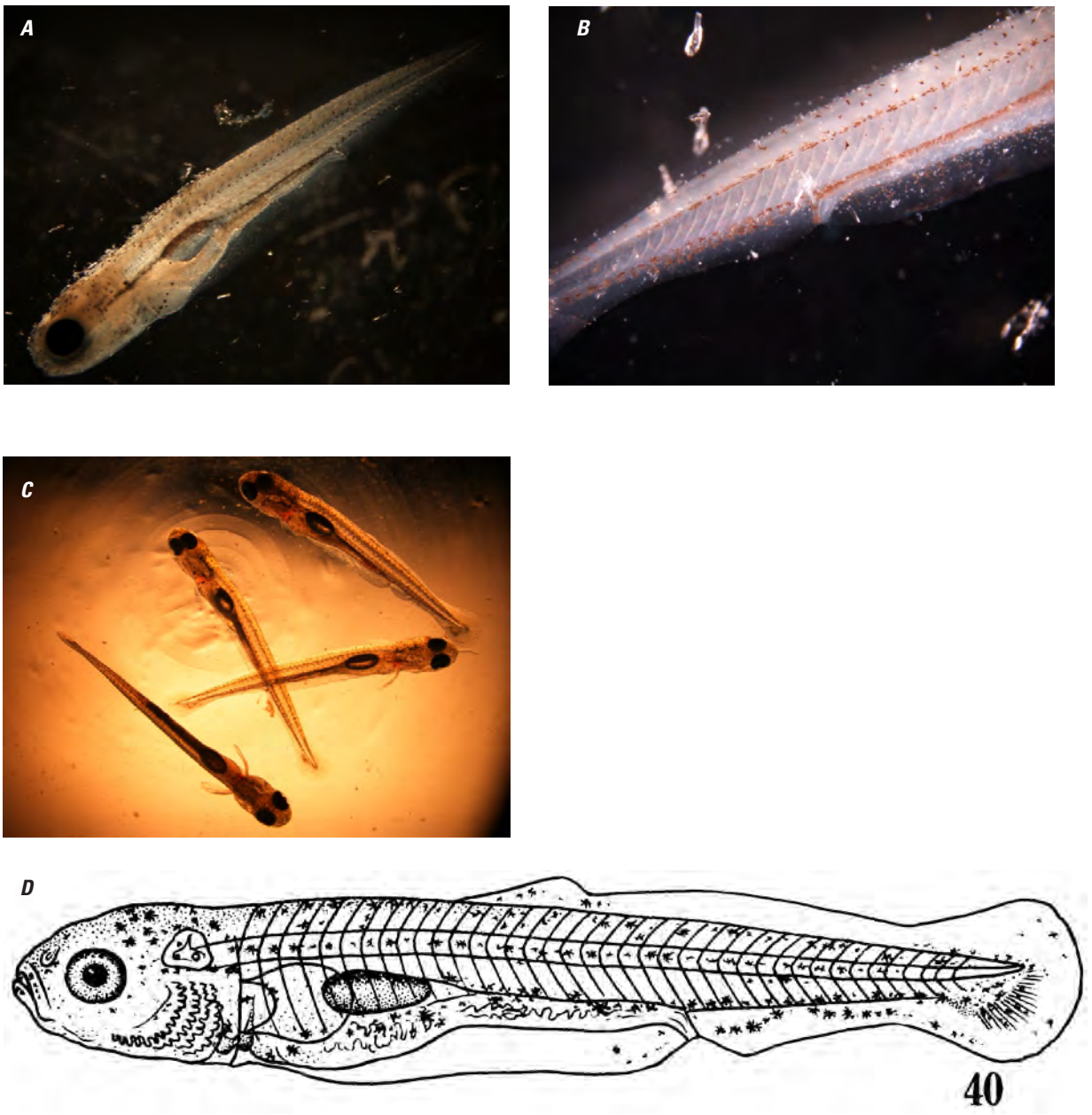


Figure 1–40. Stage 40: Yolk absorption stage of bighead carp development. The larvae is approximately 10.0 millimeters; yolk sac exhausted; dorsal fin further differentiates; posterior margin of caudal finfold becomes crenulated. Pre-anal finfold enlarges (Yi and others, 1988). *A*, Preserved specimen, low temperature treatment, 261:00 post fertilization, picture taken with 20x magnification. *B*, Preserved specimen, high temperature treatment, 213:00 post fertilization, picture taken with 40x magnification (melanophore detail). *C*, Live specimen, low temperature treatment, 261:00 post fertilization, picture taken with 10x magnification. *D*, Line drawing of yolk absorption stage from Yi and others (1988).

Abnormalities

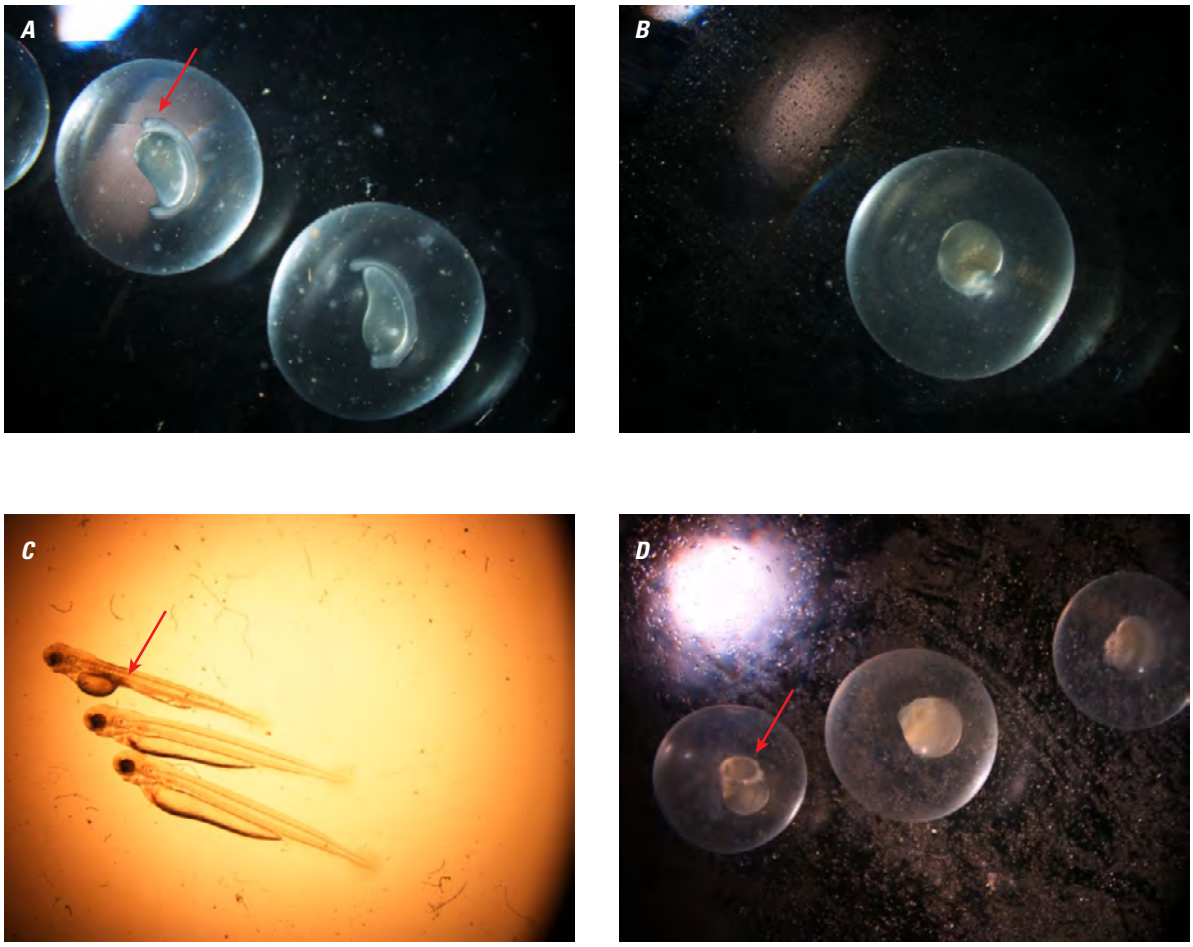


Figure 1–41. Developmental abnormalities of bighead carp. *A*, Missing heads. *B*, Cell development abnormality. *C*, Yolk sac abonormality. *D*, Cell development abnormality.