Normal Embryonic Stages of Polyodon spathula (Walbaum)

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The paddlefish, Polyodon spathula (Walbaum), is a degenerate survivor of the Cretaceous chondrostean family Polyodontidae. Except for its anatomy, the biology of this sharklike “living fossil” is almost unknown. Some aspects of its gametogenesis are in record (Larimore, '50), but only a dozen or two postlarval young have been collected (Thompson, '33), and the observations published on its life history have been extremely scanty.

This is unusual for such a conspicuous species, whose individuals may grow to a length of seven feet and a weight of 160 pounds. Polyodon has been fished commercially at a number of places along the Mississippi River system, but in each case the resource was soon exhausted. Local renewal of the population has been noticed in recent decades where new breeding grounds were established above certain great power and storage dams. At one of these, on the upper Osage River near Osceola, Mo., Polyodon was seen spawning for the first time in '60, by Charles Purkett. That year he collected the first eggs and newly hatched larvae ever seen (Purkett, '61), and the next year he stripped and fertilized eggs from mature fish caught near the same place by sportsmen. Some of the young fish which developed from these eggs reached a length of nearly three feet at the age of 17 months (Purkett, '63a).

The Fisheries Section of the Missouri State Conservation Commission is making life history and population studies on Polyodon, with a view to stabilizing and increasing its numbers as a permanent resource (Purkett, '63b). Since it is not yet clear that Polyodon succeeds in spawning normally every year, the junior author of this paper is investigating the possibility of insuring a supply of spawn through treatment of adults with pituitary injections.

For this first systematic account of Polyodon embryology, eggs were stripped from one female and fertilized on April 25, '63, and eggs from a second on June 3. The percentage of fertilization was very low, but more than 500 normally developing embryos were collected for observation. They were reared to the hatching stage in running water at approximately 14°C at the Little Dixie Wildlife Area near Columbia, Mo. Some of them were then shipped by airplane to Hanover, N. H., where they were reared in flat dishes of spring water at room temperatures that slowly rose to 20°C. A few individuals survived long enough to exhaust their yolk and to feed actively. These swam incessantly day and night in erratic courses close to the surface, pursuing daphnids, but consistently refusing copepods. The last specimen available reached a length of 60 mm at an age of 70 days from fertilization. Its external form was then quite like that of a tiny adult.

Definition of stages

Our observations on the Polyodon development from fertilization to first cleavage are by no means as complete as those for sturgeon published by Salensky (1881), by Dean (1895), and by Detlaf and Ginsburg ('54). No figures are submitted for the cleavage stages of Polyodon since these resemble those of the sturgeon in all major features, and the figures published by these authors will do as well.

The ovarian eggs are oval and uniformly gray-brown in color. At Stage 1, after fertilization but before the effects of activation are visible, each egg lies with its
animal pole — vegetal pole axis in a horizontal position. At Stage 2 the egg rotates through 90° bringing the animal pole uppermost, and a spectacular redistribution of pigment takes place. White cytoplasm gathers at the animal pole, surrounded by an irregular but usually complete narrow ring of very dark pigment. Perivitelline fluid accumulates above the somewhat depressed animal pole. At Stage 3, a light area, in all respects reminiscent of the amphibian gray crescent, appears at one side of the animal pole.

As in the sturgeons, cleavage in Polyodon is holoblastic, but the cleavage furrows spread slowly down over the surface and they cut even more slowly inward through the yolk. In our material, the appearance of the first three cleavage furrows (Stages 4, 5, 6) took place between two and six hours after fertilization. The third cleavage was not horizontal as in Amphibia, but variably meridional, and the first separation of macromeres from micromeres occurred during the fourth cleavage, Stage 7, at seven hours. The fifth and seventh cleavages, defined here as Stages 8 and 9 to parallel the stages described for the sturgeon by Detlaf and Ginsburg (‘54), followed in our material at eight and ten hours. At Stage 10, late cleavage, a marginal zone of cells intermediate in size between macromeres and micromeres became noticeable.

The later stages are defined by successive appearances of new surface features, as shown in the figures. The times given are from fertilization.

Stage 11, 24 hours. Early blastula. The micromeres still give a pebbled appearance to the upper surface.

Stage 12, 28 hours. Late blastula. The upper surface is now a smooth epithelium.

Stage 13, 32 hours. The blastocoel roof has thinned out and protrudes like a blister. Concentration of pigment in the marginal zone indicates that the gastrulation movements have started, but there is not yet any invagination furrow (fig. 1, A).

Stage 14, 35–38 hours. A short straight furrow at the equator on the future dorsal side shows that invagination has begun at the junction between the gray-white epibolic cells and the dark brown vegetal cells (fig. 1, B).

Stage 15, 38 hours. The lighter cells have carried their margin a little below the equator by epibolic movement. The darker brown yolk plug of macromeres has a diameter more than half that of the whole embryo (fig. 1, C).

Stage 16, 42 hours. The yolk plug has a diameter half that of the whole. The pigment of the invaginated endodermal cells is dimly visible through the gastrocoel roof on the dorsal side (fig. 1, D).

Stage 17, 45–55 hours. The diameter of the yolk plug has shrunk to about one-fourth that of the whole. At this stage the embryo rotates in its membranes, the future dorsal side coming to the top and the yolk plug to the equator (fig. 1, E).

Stage 18, 55–58 hours. The slit blastopore usually shows a dorsal-lip notch, and the yolk plug has been nearly buried. The medullary plate has thickened, and it extends 120° forward around the embryo from the blastopore (fig. 1, F).

Stage 19, 65 hours. This is the earliest neurula, with a slit blastopore but not normally any yolk plug showing on the surface. (Some of our specimens showed mild exogastrulation.) A neural groove extends a short distance craniad from the slit (fig. 2, A).

Stage 20–21, 68 hours. (The combining of stages is explained in the Discussion section.) Neural folds have become prominent but are not closed together (fig. 2, B).

Stage 22, 71 hours. The neural folds have met at trunk level, but not at brain level. The embryonic axis, extending from the blastopore to the anterior end of the neural folds, wraps around 150° of the yolk mass (fig. 2, C).

Stage 23, 78 hours. Neural folds are completing their closure at brain levels. One to three pairs of somites are visible, and there are short whitish kidney ridges diverging caudal, lateral to them (fig. 2, D).

Stage 24, 90 hours. The embryonic axis extends 200° around the yolk, but the head parts are not yet raised up. The trunk-tail bud is a very broad slight mound, not undercut. The kidney duct is beginning to bow outward opposite somites 4–7 (fig. 2, E).

Stage 25, four days. The pronephric bends of the kidney ducts now extend slightly forward (fig. 2, F).
Stage 26, four and three-quarter days. The trunk-tail bud has become a flat blunt paddle, slightly undercut. The mandibular arches have formed winglike expansions forward to the level of the optic vesicles.

Stage 27–28, five and one-half days. The embryo is not yet motile. Its trunk-tail bud projects, flat or rodlike, though not as far as the brain is long. The heart is a straight or slightly bent tube without chambers, and is beating feebly (figs. 3, A and B).

Stage 29, six and one-quarter days. There are now slow squirming trunk movements. The blunt, curved and fingerlike trunk-tail bud is as long as the brain, and the head is raised up off the yolk surface, but not undercut (fig. 3, C and D).

Stage 30–31, six and three-quarter days. The free trunk-tail is one-third the length of the yolk sac and beginning to straighten. The head is undercut as far as the eye level. Both the median finfold and the segmentation process have developed well.
Fig. 2. *Polyodon* neurulation Stages 19–25. The scattered stipple represents the dark pigment of the archenteron floor showing through the tissues of the roof. The close stipple in A, C and F is asymmetrically distributed ectodermal pigment which has converged on the neural fold of one side. Plain arrows and arrows with X as in figure 1.

Beyond the cloaca, but not to the tail tip (fig. 3, E).

**Stage 32–33,** seven and three-quarter days, length 7.5 mm. The finfold is narrow but reaches nearly to the tip of the tail. The trunk-tail axis is straightening but not straight. The opercular border of the hyoid segment now bulges prominently, anterior to the branchial segments (fig. 4, A and B).

**Stage 34–35,** nine days, length 8 mm. First hatching. The tail is nearly straight, and the segmentation is almost complete. The head is undercut in the ventral midline past the lower jaw. The outlines of the ear vesicles have changed from round to oval. The heart begins to show chamber bulges (fig. 4, C and D).

**Stage 36,** ten days. Mass hatching. The tail is now completely straight and the head is undercut in the midline as far as the pericardial cavity. There is no indication of a stomodeum, nor of pigment in the eyes (fig. 5, A).

**Stage 37,** 11 days, length 9 mm. There is now a stomodeal groove or central pit, but the mouth is not open. A black spot is dimly visible at the inner pole of each optic cup. Pectoral fin anlagen are visible as mesenchyme condensations. There is
no indication where the dorsal fin will emerge in the continuous finfold, and there are no gill filaments or barbels. The yolky stomach is still indistinguishable from the yolky midgut (fig. 5, B).

Stage 38, 12 days. The continuous dorsal finfold now shows a local widening, the anlage of the dorsal fin, but there is no corresponding expansion in the ventral finfold to represent the anal fin. Pectoral fin anlagen project slightly as surface ridges. A pair of low swellings represent the future barbels, and the olfactory pits have changed from round to oval. Rows of tubercles, the rudiments of gill filaments, appear on the first branchial arches. Right and left grooves in the yolk mass extend toward the liver, forming between them an angle of 180°; they set apart the future stomach and midgut (figs. 4, E and 5, C).

Stage 39, twelve and one-half days. A line of projecting epidermis marks the location of each pelvic fin anlage, but it has not projected enough to constitute a flange. The anal fin lobe is detectable. The mouth has opened. As the right and left lines of separation between the future stomach and midgut approach the liver, they form an angle of 150° (fig. 4, F).

Stage 40, 13 days. The angle formed by the lines of separation between the yolky midgut and stomach has been reduced to 90°. The pelvic fin rudiments are long but
very narrow flanges, barely projecting free. The barbels have not increased in length (fig. 6, A).

Stage 41, 14 days. The barbels are short fingerlike projections, as long as wide. Rows of gill-filament tubercles have appeared on the second branchial arches. The yolky midgut, which continues to be wider than the growing liver, is beginning to accumulate fluid internally. The gall-bladder remains empty (fig. 6, B).

Stage 42, 15 days. The barbels are longer than wide. The flanges on the pelvic fins remain narrower than the median finfold that separates them. The yolky midgut has shrunk to a width less than that of the liver, and it contains yellow bile. The pericardial cavity still occupies more space than the liver (fig. 6, C).

Stage 43, 16 days. The liver takes more space than the pericardial cavity. The width of the pelvic fin flanges equals that of the median finfold that separates them. Only a small amount of yolk remains in the wall of the midgut. The jaw makes occasional movements (figs. 4, G and 5, D).

Stage 44, 18 days. The midgut has become a narrow tube swung to the right side, and is yolkless, in contrast to the stomach. The pelvic fins have increased in width so that they project beyond the median finfold that separates them. A slight finfold still connects the dorsal fin with the caudal fin. There is no sign of a rostrum. Opposed flaps of skin approach each other across the dumbbell-shaped nasal grooves but have not fused (fig. 6, D).

Stage 45, 21 days. The median finfold separating the two pelvic fins has dis-
DEVELOPMENT OF POLYODON

Fig. 5 Later developments on the flank of Polyodon. A and D are dorsolateral views, B is a lateral view, and C is ventrolateral. Plain arrows and arrows with X as in figure 1.

appeared, and so has the section between caudal and dorsal fins. A rounded anterior bulge suggests the beginning of outgrowth of the rostrum. Yolk still crowds the walls of the stomach. Separation of anterior and posterior nasal apertures has been achieved by the fusion of opposing flaps of skin across the middle part of the nasal groove (fig. 6, E).

Stage 46, 26 days. Feeding stage. Length 15 mm. The bluntly pointed rostrum is by now 1 mm long. The stomach, no more yolky, lies on the left side (fig. 6, F and G).

Notes on Polyodon organogenesis

The following observations could be made on the intact embryos. Materials from each of the stages were preserved for further study in serial sections.

1. Central nervous and sensory structures. Brain vesicles appear promptly after closure of the neural folds at Stage 23. Half a day later, at Stage 24 (fig. 2, E), optic vesicles have formed as forebrain bulges, and four pairs of neuromeres show in the walls of the hindbrain ventricle. The eyes complete their differentiation from Stage 25 to Stage 45 with almost no increase in size. A spot of black pigment has formed at the inner pole of each optic cup at Stage 37, and this spreads to encompass the inner hemisphere at Stage 40, enveloping the whole eyeball internal to the cornea by Stage 42.

Small olfactory placodes, detectable at Stage 24, are separated from each other by a small hatching gland (fig. 3, D) which regresses quickly at Stage 36. The growth of the olfactory organs surpasses that of the eyes by Stage 29. The external nasal openings, which are at first round pits, become slanted oval grooves at Stage 38 (fig. 4, E), and then dumbbell-shaped at
Fig. 6 Later developments of Polyodon intestine, fins and rostrum, Stages 40–46. Plain arrows and arrows with X as in figure 1.

Stage 41. By fusion of the opposed walls of the middle of the nasal groove at Stage 45, the original single external nasal aperture is converted into anterior and posterior openings (figs. 5, D; 6, E; 6, G). Shortly before the feeding stage, the nasal sense organs enlarge and protrude so much that they cut off most of the anterior field of vision from the tiny eyes.

Otic placodes appear at Stage 24, become pits at Stage 25, and round closed vesicles by Stage 27–28. Their outlines are oval by Stage 34–35 (fig. 4, C), and the commencements of saccular enlargements may then be seen from above. Otoliths are present by Stage 37.

The caudal migration of the post-otic lateral line placode takes place after the hatching stage in Polyodon, and most of its extension toward the tip of the tail has been completed before it is obscured by pigment. Only certain favorable urodele embryos show the process with greater clarity. The slightly swollen tip of the outgrowth reaches the level of the first somite at Stage 38 (fig. 5, C), and passes above the pronephros at Stage 39. It is found posterior to the pectoral fin at Stage 42, at pelvic fin level in Stage 43 (fig. 4, G). By Stage 46 it has migrated beyond the anal fin and halfway down the tail. Transparency of the epidermis in earlier stages allows one to see the neuromasts that have been dropped off in the trail of the migrating tip, and the simultaneously elongating vagus nerve branch.

Sensory patches multiply in profusion on the head skin after their first appearance on the operculum at Stage 42. The pattern of special glands, sensory patches and lat-
eral-line canals of the head, is nearly complete, and conspicuous, at Stage 46 (figs. 5, D and 6, G).

2. Mouth and pharyngeal segments. The mandibular segments of mesoderm first appear lateral to the hindbrain at Stage 24 (fig. 2, E), and spread forward like wings as far as the optic vesicles at Stage 26. The hyomandibular pouch, when it approaches the surface, allows deep endodermal pigment to show through as a dark line, defining the anterior border of the hyoid segment. At Stage 26 the hyoid wings are only half as long as the mandibular wings. At Stage 27–28 (fig. 3, A), all these still lie flat on the yolk mass, and the dark hyomandibular lines make an angle of 90° as they converge toward the ear vesicles. The pre-otic pharyngeal segments begin to lift themselves off the yolk and swing together ventrad at Stage 30–31 (fig. 3, E), and a gular region is completed ventrally at Stage 36, together with elongation and projection of the whole head.

The stomodeum appears at Stage 37 as a shallow transverse groove with a central pit, the deepening of which opens up the mouth at late Stage 38. The two barbels appear as raised knobs on the upper jaw at about the same time, but their lengthening is postponed until Stage 41 (fig. 6 A–E). The gaping mouth shows tooth germs on both jaws beginning at Stage 42.

The posterior border of the hyoid segment suddenly begins to bulge at Stage 32–33 (fig. 4, B) and is almost at once undercut. The extension of the operculum then proceeds rapidly, as shown in figures 5, A–C, and 4, E–G. It flares more and more conspicuously laterad over the gill arches, and the dorso-posterior projection, which remains as a prominent feature in the adult, first shows dorsal to the gills at Stage 40. The bulging gill masses are gradually covered over (fig. 6, A–F), but for several weeks the red color of the blood coursing through them can be plainly seen through the weakly pigmented opercular skin.

The branchial segments cannot be distinguished in the general visceral mesoderm until about Stage 30–31, when a superficial groove sets the first of them off from the hyoid segment (fig. 3, E). Each of them is visibly segregated by Stage 34–35, though the gill slits break through later.

Tiny tubercles show in two rows on the first branchial arch at Stage 37 (fig. 4, E), and have developed into long transparent blood-bearing gill filaments by Stage 40. Similar developments do not appear in the second branchial arch until Stage 41.

3. Somites and the kidney system. Formation of somites from the epimERIC mesodERM starts at Stage 23 and continues rapidly toward the tip of the tail, ending in Stage 34–35 (figs. 2, D–E; 3, E; 4, C). By Stage 30–31, chevron-shaped myotomes are developing from the block-like somites in the trunk region, and from these a series of hypaxial muscle processes spread out over the yolk mass (fig. 5, A and B). The hypaxial divisions of the somites beyond the yolk in the projecting trunk-tail form later and in a different way. As late as Stage 37 the muscle processes that extend over the yolk are string-like cords, and they do not widen to meet their neighbors in a hypaxial sheet until they have spread outward beyond the pectoral fins (fig. 5, B and C). The spreading outer tips of the muscle processes are slightly thickened so that they collectively form a visible scalloped border to the thin almost transparent hypaxial sheet. This border is seen at the level of the pectoral fin at Stage 38, and halfway between the fin and the ventral midline at Stage 42 or 43 (fig. 5, C and D). The hypaxial musculature later becomes thick and opaque, but the ventral borders of the two sides have not met, even at the feeding stage. As in amphibian larvae, there remains for some weeks a broad transparent strip of belly wall between them, through which abdominal viscera may be seen.

The intermediate mesoderm makes its appearance as two short divergent streaks lateral to the first somites (fig. 2, D). As the materials of the embryonic axis gather closer to the midline posteriorly, the wide angle of divergence of these streaks is quickly closed (fig. 2, D and E), and by the time the trunk-tail bud begins to project free they have become parallel. The future pronephros bends outward opposite somites 4–7 at Stage 24, and this bend
rapidly elongates (figs. 2, F and 3, C) into a hairpin-shaped tubular excursion, anterolaterad and return. Such a formation has not been found in any other fishes except sturgeons. The departing and returning limbs of this extraordinary hairpin tubule are at first straight, but from Stage 29 on they become increasingly beaded and twisted until the definitive pronephros is a tangled mass of convolutions at Stage 39, when melanophores begin to obscure it. The anatomy of such a kidney, though not its development, was described for the sturgeon by Jungersen (1893).

The primitive kidney duct meanwhile reaches the cloaca at Stage 27–28, becomes tubular at Stage 29, and is drawn inward parallel with the notochord. Expanding myotomes then conceal it.

4. Fins. At Stage 29, a seam appears on the outgrowing trunk-tail bud. This grows out as the dorsal and ventral parts of the finfold, eventually connecting around the tip of the tail (fig. 3, C–D, and fig. 4). The embryo has enough of a tail-fin to be an accomplished swimmer by Stage 34–35. At Stage 38, muscle slips grow from the adjacent myotomes into the parts of the finfold which will later be the dorsal and anal fins, in the former area while the fin lobe is forming, in the latter area before there is any hint of a lobe (fig. 4, E). The separation of dorsal, caudal and anal fins is accomplished by regression of intervening parts of the median finfold during Stages 44 and 45, and the caudal fin has become fully heterocercal by the time the larva begins to hunt for food (figs. 3, G and 6, E).

The paired pectoral fins appear first as thin mats of mesenchyme at Stage 37. Mollier (1897) showed that in Acipenser sturio the pectoral fin musculature arose from ingrowing sprouts of myotomes 6–10, quite as in elasmobranchs. Harrison (1895) just as clearly showed that in the teleost Salmo the myotome sprouts do not enter the pectoral fin, the musculature forming from local mesenchyme, as in Amphibia. In Polyodon, the wavering threadlike muscle processes from myotomes 9–12 show a tendency to converge toward the pectoral mats at this stage (fig. 5, B), but one cannot be convinced from surface examination that they contribute any cells to them. The question requires further study.

Whatever the situation in the pectoral fins, there is no doubt about the pelvics. Their musculature is provided in Polyodon, as in all other fishes so far examined, by ingrowth of sprouts from all the adjacent myotomes. The pelvic fins are detectable as rudiments as early as Stage 39, and the muscle slips are visible through the transparent skin, extending into them at Stage 42.

5. Intestine. The early Polyodon archenteron is a very wide shallow cavity lined by heavily pigmented endoderm, extending on each side from the embryonic axis halfway to the equator, and in a wide strip across the midline posterior to the closed blastopore (figs. 2, D and 3, B). The chocolate-brown pigment shows quite plainly through the overlying whitish layers, as suggested by scattered stippling in figures 1–3. The roof of this cavity is thin, its floor consists of large yolky cells. Not until Stage 27–28 does the projecting trunk-tail bud grow out beyond the posterior limit of the pigmented zone. Part of the definitive intestine develops in the trunk section of this projecting bud. It becomes visible when the body has started to straighten at Stage 32–33. The endodermal lining of this hindgut is not pigmented, and it develops a spiral valve (fig. 4, E).

Polyodon shares with the sturgeons the peculiarity that its liver is formed, not dorsal to the yolk as in holosteans and teleosteans, but on its ventral surface, clearly visible in external view (fig. 4, D). Its anterior border is marked off at Stage 30–31, its posterior border at Stage 32–33, and the gallbladder appears at Stage 37.

A vitelline network of veins is established at Stage 30, and almost at once one can distinguish two yolky areas in ventral view. The more posterior half, the future midgut (as the term will be used here), is yellowish and contains a pattern of veins that branch out into capillaries from the subintestinal vein at the cloaca, and gather together more anteriorly in a vitelline vein that skirts the liver on its right side (fig. 4, D). Much of the blood from this latter vein is subsequently diverted through the liver before being sent to the heart. The
more anterior half of the yolk mass, the future stomach, is a mottled gray color, and it is only sparsely infiltrated by veins and capillaries, whose blood comes from the dorsal aorta and the pronephros. This blood is sent, not to the liver, but directly to the sinus venosus.

At first, there is nothing more to distinguish these two parts of the yolk mass than their blood pattern and their color. Later, at Stage 38, a transverse superficial furrow begins to separate them, and as this furrow deepens it forms a progressively narrowing angle whose arms approach the liver at Stages 39 and 40 respectively at 150° and 90°. It may be noted in figure 6 that this furrowing, visible through the transparent somatopleure, involves only the intestinal wall.

A strong circulation having been established by Stage 34–35, the originally spherical yolk mass becomes oval and then progressively pear-shaped. The shrinkage shows first at its posterior pole, where the greatest influx of blood occurs. The midgut, i.e. the part of the intestine lying between the bile duct and the spiral valve, rapidly loses its yolk after Stage 38. It still exceeds the liver in width at Stage 41 but begins to contain fluid, which takes on the color of bile in Stage 42. Epithelial ridges may be seen within it by Stage 43, and at Stage 44, all its yolk gone, it has collapsed to a straight tube on the right side of the coelom (fig. 6, A–D). Yolk lasts a week longer in the stomach wall, but with its disappearance at the feeding stage, the empty stomach swings to the left side and the pattern of the intestine is complete.

Jet black pigment accumulates in the turns of the spiral intestine, starting at Stage 40. This suggests that the highly pigmented cells that line the floor of the original archenteron are probably broken up and digested, as is known to happen in amphibian embryos.

6. Heart. As in many other fishes, the rudiment of the heart is first found flat on the surface of the yolk, directly anterior to the forebrain. At Stage 36, it appears as a Y-shaped accumulation of cells, the branches of the Y spreading out on the yolk laterally, and the stem projecting toward the unraised forebrain vesicle (fig. 3, D). During Stage 27–28 it changes from a short straight tube to a slightly bent one and starts beating in a slow irregular rhythm. The S-shaped coil is established at Stage 29 and strengthened with increasing flow of blood during Stage 30–31. Heart chambers are not clearly defined until Stage 34–35 (fig. 4, D). By this time there is a heavy circulation of red blood, the principal pattern of the aortic arches and the dorsal aorta has been established, and abundant capillaries are separating the primordial continuum of arteries and veins.

7. Pigment. The early Polyodon embryo is strikingly colored. There is not only the heavy chocolate-brown pigment that moves into the archenteron walls with the yolk plug, there is a variably distributed dusting of pigment in the predominantly white cells of the upper surface. During gastrulation, some individuals show streamers of this surface pigment carried by cells converging toward the blastopore and invading into the mesoderm (fig. 1, C–E). When convergence movements draw the definitive ectodermal cells toward the embryonic axis, an asymmetric distribution of the surface pigment often produces a sharp line down the middle, so that one side of the neural plate or the trunk-tail bud is dusty gray and the other side snow white. Particular examples of this are shown in figures 2 A, C and F; 3 B and D. The early embryonic surface pigment is still prominent through Stage 32–33, but then quickly disappears.

The newly hatched embryo at Stage 36 shows practically no melanophores. The first wispy black processes of such cells appear on the anterior flanks and over the midbrain at Stage 37. A triangular concentration of melanophores gradually accumulates with its anterior base over the pronephros and its long point trailing caudad along the primitive kidney duct. By Stage 41, this has quite obscured the pronephros. The boundaries of the brain lobes are not hidden under the concentrating melanophores until about Stage 43. Over most of the body, it is no longer possible to see internal structures through the skin after the feeding stage. The rostrum becomes dense black after it starts to grow out at Stage 46.
8. **Behavior.** When removed from their membranes at Stage 29, *Polyodon* embryos can be stimulated to squirm slowly. At Stage 30-31, continuous and vigorous wiggling occurs. Active swimming efforts by the released embryos at Stage 30-31 are thwarted by the heavy yolk load and inadequate fin development, but by the normal hatching time they can make long direct trips through the water. Older embryos swim incessantly unless they are ill or fouled in algae. They travel in wide random circles close to the surface, changing direction abruptly in response to stimuli from possible prey.

**DISCUSSION**

Most of the peculiarities of the embryonic development of *Polyodon* are shared by sturgeons. The latter, comprising the family Acipenseridae, are grouped in recent classifications with the Polyodontidae as Acipenseroformes, the only surviving order of the Superorder Chondrostei. Their development is sharply contrasted with that of the Teleostei, but somewhat less so in comparison with the Holostei, in such matters as cleavage pattern, disposition of yolk, and the maneuvers of gastrulation and neurulation. The internal events of *Polyodon* gastrulation will be described in a subsequent paper.

Detlaf and Ginsburg ('54) have recently described the development of *Acipenser stellatus*, *A. guldenstadtii* and *Huso huso* at a range of temperatures from 10° to 25°C. To facilitate the comparison of sturgeon and paddlefish development, our *Polyodon* stages have been defined as closely as possible in accordance with theirs for *Acipenser*. Their account ends with Stage 36, the period of mass hatching, and to cover the period of several weeks until yolk is exhausted and the larva is feeding we have added ten more stages. Furthermore, some of the Detlaf-Ginsburg stages have had to be combined to fit the *Polyodon* material.

Their stages 20 and 21 are defined by characters of the neural folds which appear with insufficient clarity in *Polyodon*, so that we define only a Stage 20-21. In their rather modest development of neural folds, our *Polyodon* resemble the neurula stages of *Acipenser sturio* figured by Dean (1895) more than they do the sturgeon species available to the Russian authors.

Similarly Stages 30-31, 32-33 and 34-35 have been combined for *Polyodon* because of a lack of morphological characters sufficiently precise to define stages like those of Detlaf and Ginsburg. The sole criterion for their Stages 30-35 is the position of the tip of the tail against the body of the unhatched embryo, as it is seen curled up within the chorion. At Stage 30 it has not reached as far forward as the heart, but it does so at Stage 31, meets the anterior end of the head at 32, passes beyond at 33, overtakes the midbrain at 34, and touches the ear at Stage 35.

Sturgeon and paddlefish have reached precisely comparable levels of morphological development at the hatching Stage 36, but at markedly different rates. *Polyodon* hatches in nine days at 14°C. According to Detlaf and Ginsburg, *Acipenser guldenstadtii* hatches in seven days at this temperature, and *A. stellatus* in less than five days. Dean (1893) saw *A. sturio* hatching in from 92 to 100 hours at 16° to 23°C.

**SUMMARY AND CONCLUSIONS**

1. The embryonic development of *Polyodon spathula*, heretofore undescribed, is like the development of sturgeon embryos in nearly all details. Detlaf and Ginsburg ('54) defined 36 stages of sturgeon embryology from fertilization to hatching. Five pairs of their stages were defined by trivial characters, and for *Polyodon* each of these pairs is reduced to one. Otherwise a nearly exact correspondence exists between their stages and ours. We have added ten more stages beyond hatching, to carry the account of *Polyodon* development to the onset of feeding. The age of each stage is given in hours or days from fertilization.

2. As in sturgeon, *Polyodon* cleavage is total and unequal, and the gastrulation involves an amphibian-like cytoplasmic crescent, a blastopore, yolk plug, and gastrocoel. In contrast to conditions in holosteans and teleosteans, the yolk mass is contained within the blastomeres, and the liver grows out ventral to the yolk mass, not dorsal to it.

3. A conspicuous feature of the late neurula stage is a long hairpin-shaped
excursion of the primitive kidney duct, from which the pronephros is derived. Only in sturgeons has a similar development been found.

4. Relatively huge nasal organs contrast with extremely small eyes. The latter complete their histogenesis with very little increase in size beyond that of the optic vesicles. The caudad migration of the lateral line placode is exceptionally clear.

5. Development of the gut, and of the hypaxial muscles, takes a different course in the part of the trunk filled at first by the yolk mass, compared with the part formed by the projecting trunk-tail bud.

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LITERATURE CITED