CERTAIN PELVIC AND SACRAL ANOMALIES IN ANURA

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The structure of the sacrum, pelvic arch and hind limbs in ectromelia and polymelia in froglings of ground toad (*Bufo bufo*) is described. The variants presented along with the formerly described anomalies of these structures in ground frogs cast doubt on the main role of pelvis and/or hind limbs in sacrum formation.

Key words: Anura, Bufo, sacrum, pelvic arch, hind limbs, ectromelia, supernumerary legs.

The morphology research group of the Department of Vertebrate Zoology (St. Petersburg State University) has been investigating the morphogenetic relations in the development of sacrum, pelvic arch, and hind limbs for a number of years (Kovalenko and Anisimova, 1987; Borkhvardt, 1995). The data on the structure anomalies are widely used to analyze the causative factors of their development (Kovalenko, 1992; Kovalenko and Danilov, 1994; Kovalenko and Danilevskaya, 1994; Kovalenko, 1995). These investigations are carried out on the basis of the collection of osteological anomalies at the Department of Vertebrate Zoology (CACVZ), collected by researches and students of the morphology group. Skeletal anomalies of Anura are presented on a large scale in this collection (several thousands of specimens).

The structure anomalies in ground toad (*Bufo bufo*), which demonstrate the new variants of the relationship between sacrum, pelvis and limbs, are described, namely: 1) there is sacrum in the presence of the pelvic process and the absence of the hind limb (3 specimens); 2) there is sacrum in the presence of the supernumerary leg and supernumerary ilium. The variants of ectromelia and polymelia in Anura are not uncommon and more than once were described in the literature (see Voitkevich, 1958; Dubois, 1979; etc.). But there are no any data on the skeletal morphology, and particularly on the backbone structure, in these articles which prevents using this information for morphogenetic analysis.

MATERIAL AND METHODS

The described anomalies were found in 4 froglings of ground toad (*Bufo bufo*). The series were captured during coupling in the natural ponds (spring 1995 in the environs of St. Petersburg) and spawn in the laboratory. The offsprings from 5 couples were obtained (approximately 2500 hatchlings and 1300 – 1500 froglings after metamorphosis). The abnormal froglings were taken from the posterity of one couple (4 froglings from 500 hatchlings and 300 specimens were brought to the end of metamorphosis). The breeders of this series had normal limbs in outward appearance, except a damaged left foot in the female more likely of a traumatic character.

The collection number of these specimens: AG-B.b.-1-4. Study was conducted on alizarin-alcian total preparations (Wassersug, 1976). The drawings were made by Yu. I. Kovalenko.

RESULTS

Case 1 (AG-B.b.-3, Fig. 1). The larva is at the end of metamorphosis. There is only one, hind, limb (right), and the left leg is fully absent. The right leg is formed normally, but the femur is slightly distorted (Fig. 1*a*); the femur normally articulates with the pelvic arch and is directed at an angle of 60 to the body axis. The pelvic arch (from below) is located on the midline (Fig. 1*b*), but is turned to the left relatively to the body axis. The right part of the pelvis is normally formed; the left half is presented by a small rounded gristly plate, which has no acetabulum but the ilium

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Fig. 1. Ectromelia in larvae of *Bufo bufo: a*, View from above; *b*, *c*, structure of the pelvic arch and femur from below (*b*) and from the left (*c*); VII - X, numbers of vertebrae; *F*, femur; *il*, ilium; *plp*, pelvic plate; *sc*, scapula; dotted line, body axis (*b*) and ventral line of chorda dorsalis (*c*).

grows from its center (Fig. 1*c*). The left ilium is shorter than the right one (approximately by one fifth of its length). The anterior tip of the right ilium is winded. The left pelvic process is gristly and the right one is covered by a bony cuff by two thirds of its length.

The axial skeleton demonstrates one of the variants of the "DS syndrome" (disturbance of segmentation); the segments of the left side are longer than those of the right side so that the effect of the asymmetric shift (accumulation of asymmetry) arises (Kovalenko, 1983, 1992). There are eight trunk anlages on the left side of the body, and nine ones on the right side. The opposite neural arches in the seventh and eighth segments are located in staggered rows (in the chess order). That is why the last trunk vertebra is formed by a pair of different anlages (left eight and



Fig. 2. Ectromelia in larvae of *Bufo bufo: a*, View from above; b, c, structure of the pelvic arch and femur from the left (b) and right (c); dotted line, the ventral line of chorda dorsalis. Designations are the same as in Fig. 1.

right ninth). So there are one sacral vertebra and two sacral diapophyses of a common size.

Case 2 (AG-B.b.-4, Fig. 2). The larva is at the end of metamorphosis. The right hind limb is absent. The left leg is nearly normally formed (the femur is slightly distorted). A single hind limb is located practically in the middle. The head of the femur adjoins the acetabulum by its lateral surface (dysplasia of the thurl) and as a result the thigh is directed not aside as in the norm, but practically backwards (Fig. 2*a*).

The pelvis consists of an oval gristly plate and two iliac processes. On the left of the plate there is the acetabulum (more hollow than in the norm) and the normally located ilium; on the right side the acetabulum is absent and the right pelvic process grows from the caudo-ventral part of the pelvic plate (Fig. 2c). The left ilium is slightly shorter than the right one, but since the last one grows from the caudal part of the pelvis, its anterior margin is located more caudal



Fig. 3. Ectromelia in larvae of *Bufo bufo: a*, View from above; *b*, *c*, structure of the pelvic arch and femur from the right (*b*); *epT*, epiphysis of tibia; other designations are the same as in Fig. 1.

than that in the left one (Fig. 2*a*). Both pelvic processes are covered with a bony cuff.

There is a little twist on top of the left fifth – sixth segments of the axial skeleton. There are only eight vertebral anlages in the trunk. The structure of the neural arches of the ninth vertebra resembles that of the first caudal ones; the left arch is fused with the urostyle. The eighth vertebra is the last in the trunk and similar to the common sacral vertebra, but in contrast to the latter it has a pair of postzygapophyses. The neural arches of the eighth and ninth segments bear increasing transverse processes, but the right diapophysis on the ninth neural arch is slightly expanded and does not reach ilium. However, since only those processes are called sacral, which reach the pelvis and have the distal edge not narrower than their base (Kovalenko, 1994), we consider that this backbone has three sacral diapophyses (two in the eighth and one in the ninth segments).

Case 3 (AG-B.b.-1, Fig. 3). The larva is at the end of metamorphosis. One incomplete hind limb is present which is located practically on the midline (Fig. 3a). This is a left leg because it articulates with the pelvis on the left side.

The pelvis consists of one oval gristly plate and two iliac processes. On its left the plate has the acetabulum, but on the right it is absent. The left pelvic process is located normally; on the right side the ilium grows from the caudo-ventral part of the pelvic plate (Fig. 3b). The right ilium is appreciably shorter than the left one. Both processes are cartilaginous. The only leg consists of a one lever (thigh). There is a self-dependent epiphysis on the distal end of the femur. It can be suggested that it belonged to the tibia and arose due to life-time trauma. The femoral head closely adjoins the hollow acetabulum by its lateral surface (dysplasia of the thurl) and the thigh is directed practically backwards (Fig. 3a).

The axial skeleton is practically normal in many details of its structure: it consists of nine trunk vertebrae and the last of them has two sacral processes, which are formed as usual at this stage of ontogeny.

Case 4 (AG-B.b.-2, Fig. 4). The larva had finished its metamorphosis. There are three abnormal hind limbs (one on the left and two on the right side) and two pelvic arches in this specimen. The first pelvis is complete and two legs (right and left) articulate with it; the second pelvis is represented by only right half and one leg (right) is connected with it.

The complete pelvis is represented by two cartilaginous plates (the right is bigger than the left one) and by two iliac processes which arise from the dorso-anterial parts of the pelvic plates. The pelvic axis is turned to the right relatively to the body axis (Fig. 4b). The left part of this pelvis has the normal structure and there is a small hollow acetabulum on the right part. Both pelvic processes are nearly of an equal length, they are covered with a bony cuff and reach the transverse processes of the eighth vertebra, but the right ilium of this pelvis cannot be seen from above (Fig. 4a), because here lies a process of the second pelvis over it (Fig. 4c).

The left limb (Fig. 4b, I) consists of three levers. The left femur has nearly a normal articulation with the pelvis and is directed sideways at a normal angle. The second lever (shin) is single. The knee joint is maldeveloped (dysplasia) and its movement is limited. The tibial head closely adjoins the femur by its lateral surface and the shin is directed dorsally from the knee-joint (upwards, but not parallel to the body). The third lever (autopodium) is abortive; it consists of one element (possibly it is the underdeveloped tarsal). The skeletal element of the third lever has one epiphysis (proximal) and its distal part is tapered and fully covered with a bony cuff (Fig. 4a, I-3).

The right limb of the complete pelvis (Fig. 4c, II) consists of the thigh, shin and foot; the structure of



Fig. 4. Polymelia in larvae of *Bufo bufo: a*, View from above; *b*, *c*, structure of the pelvic arch and femur from below (*b*) and from the right (*c*); *I*, scheme of the first and second lever of leg *II*; *dig III*, toe of leg III; *il-II*, *il-II*, the number of iliac process; *l-I*, *l-III*, *l-III*, *number* of leg; *I-1*, *I-2*, etc., number of leg lever; dotted line, body axis (*b*) and ventral line of chorda (*c*); other designations are the same as in Fig. 1.

the first two levers is considerably modified. The haunch is represented by the skeletal element which has a triangular shape; its two cartilaginous epiphyses are intimately drawn together and so that the femur is shaped like V. The corpus of this element is covered with a bony cuff, its apex has a latero-ventral direction from the epiphysis (Fig. 4c, II-1). The proximal epiphysis enters the hollow acetabulum and the distal one joins with the shin. The shin consists of two elements, each of which is V-shaped. One of them forms articulation with the femur and therefore we suppose it is the tibia (Fig. 4b, c). The proximal and distal epiphyses of the second (fibula) element are strongly drawn together and located between the branches of the first one (Fig. 41). The structure of the autopodium of this leg is nearly normal, except not straightened digits.

The second pelvis consists practically of a single ilium (right) which forms junctions with the sacrum process in front and with the abnormal limb (Fig. 4*a*, *III*; *c*, *III*) behind. There is a small gristly plate in the distal end of this ilium. The third hind limb is located above the second hind limb and consists of the one distinct formed lever (femur) and weakly differentiated distal part. The proximal epiphysis of the femur is weakly developed, it closely adjoins the cartilaginous pelvic plate by its lateral surface and the thigh is directed practically backwards. The distal epiphysis is increased and its outlines are vague, the border between it and the next lever is not clear. The femur demonstrates a small V-shaped curve and has a triangular fold in the prominence. The corpus of the femur is covered by with a bony cuff. The shin is weakly differentiated, but two gristly elements in this part can be supposed. There are two small skeletal elements on the distal edge of the shin; they are covered with a bony cuff and directed practically perpendicular to the axis of the shin and so they resemble spurs. There is a single toe on the distal end of this limb; it is weakly differentiated and two or three gristly phalanges can be supposed.

There are eight trunk vertebrae, the structure of the ninth neural arch resembles the caudal ones and is fused with the urostyle. There are an expanded transverse process (sacral) on the ninth neural arch on the right and a very small process on the left. The left sacral diapophysis is formed by the eighth neural arch; the right transverse process of this vertebra is not increased although it is joined with the ilium. A dorsal part of the first neural arch is bifurcated on the left. The backbone demonstrates some asymmetry, but without signs of the "DS syndrome."

CONCLUSION

There are some models of the causal relationships in the development of the sacrum, pelvis and hind limbs (see Borkhvardt, 1995), including a notion about the mediated influence of the iliac spine (Kovalenko and Anisimova, 1987; Kovalenko, 1992) or of the hind limbs (Borkhvardt, 1995) on the sacrum shaping. The former version suggests that the process of the iliac spine changes the primary conditions of the transverse process forming in the last trunk segment; it widens the space in which the transversal process forms, and therefore more mesenchyme cells are attracted. The pelvic process like this favors the formation of a transverse process larger than those on other vertebrae. The second standpoint attaches the leading role to the hind limbs; their formation in ontogeny "promotes degeneration of sacrum myomeres" by "creation deficiency or absence of innervation" (Borkhvardt, 1995).

Investigations of the normal development of the pelvic-sacral complex of Tetrapoda and the experimental data (see Kovalenko, 1992; Borkhvardt, 1995) disprove none of these models but some of them confirm the former version, and others the second one. The experiments on the variance of the ontogenetic way by the surgical middling always leave doubts about the correctness of interpretation of the results (see Kovalenko, 1992), because a disorder of the organism's integrity creates a number of collateral effects and it is difficult to take them into account. In this respect natural anomalies, as a result of a more rare (a less probable) way of ontogeny, are the valuable material for the decision of such debatable questions, because these structure variants demonstrate the natural potentiality of ontogenetic machinery.

For instance, certain pelvic-sacral anomalies from our collection cast doubt on the correctness of the former model. In one series of the laboratory raising of common platanna (*Xenopus laevis*) we obtained 79 juveniles (in the last stages of metamorphosis), in which the pelvic arch is absent (the materials are prepared for the press); eight specimens from this series have both the sacrum process and five specimens one process (left or right). The sacrum diapophysis in all these specimens were underdeveloped (Kovalenko and Bordukova, 1995), nevertheless the possibility of their formation considerably reduces the role of the pelvic processes in sacrum development. V. G. Borkhvardt (1995) proposed a version about the mediated influence of the hind limb on sacrum development and formulated several consequences the first of which is as follows: "the sacrum is not formed in the absence of the hind limbs." The above data (cases 1-3, Figs. 1-3) adulterate this thesis and cast doubt on the leading role of the hind limbs in sacrum formation. Sacral processes are quite normally formed with the absence of one leg (Figs. 1 and 2) and even when a single limb is not fully developed (Fig. 3). Moreover, the number of sacral processes is increased in one specimen (Fig. 2). The pelvic structure gives no grounds to suppose that trauma was the cause of the absence of the hind limb.

Other variants of the abnormal structure contradict both the first and the second models. These are the variants with aplasia of one or both sacrum diapophyses, while the pelvic arch and the limbs are formed quite normally on both sides (Kovalenko, 1995).

So, the true relationships in the development of the pelvic-sacral area allow the following variants of structure:

— the sacrum is present in the presence of the pelvic processes and limbs (norm);

— the sacrum is absent in the absence of the ilium and hind limb on the same side (Kovalenko and Danilevskaya, 1994);

— the sacrum is absent in the presence of the pelvis and limbs (Kovalenko, 1995);

— the sacrum is present in the absence of the pelvic process and in the presence of the hind limbs (Kovalenko and Bordukova, 1995; Borkhvardt, 1995);

— the sacrum is present in the presence of the pelvis and the absence of one hind limb (present work);

— the presence of two sacral diapophysis in the presence of the supernumerary leg and supernumerary ilium (present work).

The structure variants described draw attention to the fact that the hind limbs and the pelvic arch are not connected between themselves in development. The pelvis or at least the iliac process can develop in the absence of the hind limb on the same side (Figs. 1-3). The legs can develop despite that fact that the pelvis is rudimentary or even absolutely absent (series I-93 from our collection, see above). In the last case, the femoral bones do not articulate with anything and such tadpoles have many restriction in movement, but nevertheless motion is still possible (the materials are being prepared for press).

The sacrum of Anura demonstrates a sufficient quantity of structure variants in the presence of the normally formed pelvis and legs (Kovalenko, 1996). There are different variants in the location of sacral diapophysis (on the eighth, ninth, and tenth vertebrae), including their asymmetrical location on two different vertebrae (see Figs. 3, 6; Kovalenko, 1994). This phenomena was connected with small changes in the location or in the length of the left and right ilium (Kovalenko, 1992). But it is very difficult to register such small changes in froglings (by the length of one segment), especially on total preparations, because at this time (after metamorphosis) the length of the ilium much more exceeds that of the segment. In the specimens described, the left and the right pelvic processes have obvious distinctions not only in length but also in location (Figs. 1 - 4). However, no correlation is observed in the location of the sacral diapophyses.

Thus, neither the pelvic arch nor the hind limbs are the single and sufficient factor for the sacrum formation and therefore we cannot consider them as a direct cause of these morphogenetic process.

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