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Somitomeres: mesodermal segments of vertebrate embryos

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Summary

Well before the somites form, the paraxial mesoderm of vertebrate embryos is segmented into somitomeres. When newly formed, somitomeres are patterned arrays of mesenchymal cells, arranged into squat, bilaminar discs. The dorsal and ventral faces of these discs are composed of concentric rings of cells. Somitomeres are formed along the length of the embryo during gastrulation, and in the segmental plate and tail bud at later stages. They form in strict cranial to caudal order. They appear in bilateral pairs, just lateral to Hensen's node in the chick embryo. When the nervous system begins to form, the brain parts and neuromeres are in a consistent relationship to the somitomeres. Somitomeres first appear in the head, and the cranial somitomeres do not become somites, but disperse to contribute to the head the same cell types contributed by somites in the trunk region. In the trunk and tail, somitomeres gradually condense and epithelialize to become somites. Models of vertebrate segmentation must now take into account the early presence of these new morphological units, the somitomeres.

Somitomeres were discovered in the head of the chick embryo (Meier, 1979), with the use of stereo scanning electron microscopy. The old question of whether the heads of the craniates are segmented is now settled, at least for the paraxial mesoderm. Somitomeres have now been identified in the embryos of a chick, quail, mouse, snapping turtle, newt, anuran (*Xenopus*) and a teleost (the medaka). In all forms studied, the first pair of somitomeres abut the prosencephalon, but caudal to that, for each tandem pair of somitomeres in the amniote and teleost, there is but one somitomere in the amphibia. The mesodermal segments of the shark embryo are arranged like those of the amphibia.

Key words: somitomere, paraxial mesoderm, chick, head, quail, mouse, amphibia.

Introduction

It is now known that the paraxial mesoderm of vertebrate embryos is segmented from the tip of the head to the tip of the tail. Ten years ago, in the course of a study of the induction of the otic placode in chick embryos, Stephen Meier removed the head ectoderm and examined the underlying paraxial head mesoderm with stereo scanning electron microscopy, his intent being to visualize the material that he thought was inducing the otic placode. He serendipitously discovered that the paraxial head mesoderm beneath the prospective otic placode is organized into segmental units, which he named 'somitomeres' (Meier, 1979).

These units are composed of loose mesenchymal cells organized into squat bilaminar discs. On the dorsal and ventral faces of each somitomere, the cells are arranged in concentric arrays around a central point that is often a cell. These faces are usually concave or convex with the central point of the somitomere at the deepest point of the concavity, or the highest point of the convexity. The cells composing the dorsal aspect of the somitomeres, facing the ectoderm, send up cell processes from the boundaries of the concentric rings of cells toward the ectoderm (Fig. 1). The three-dimensional arrangement of the cell processes helps emphasize the pattern, and somitomeres are most easily recognized in stereo. Often alignments of cells and cellular processes perpendicular to the long axis and traversing the midline of the somitomere divide the somitomere into anterior and posterior compartments. The borders between successive somitomeres are especially apparent because the outermost concentric array of cells of each somitomere curves away from that of its neighbour. Borders between somitomeres are also more apparent in stereo because they are ridges between success-

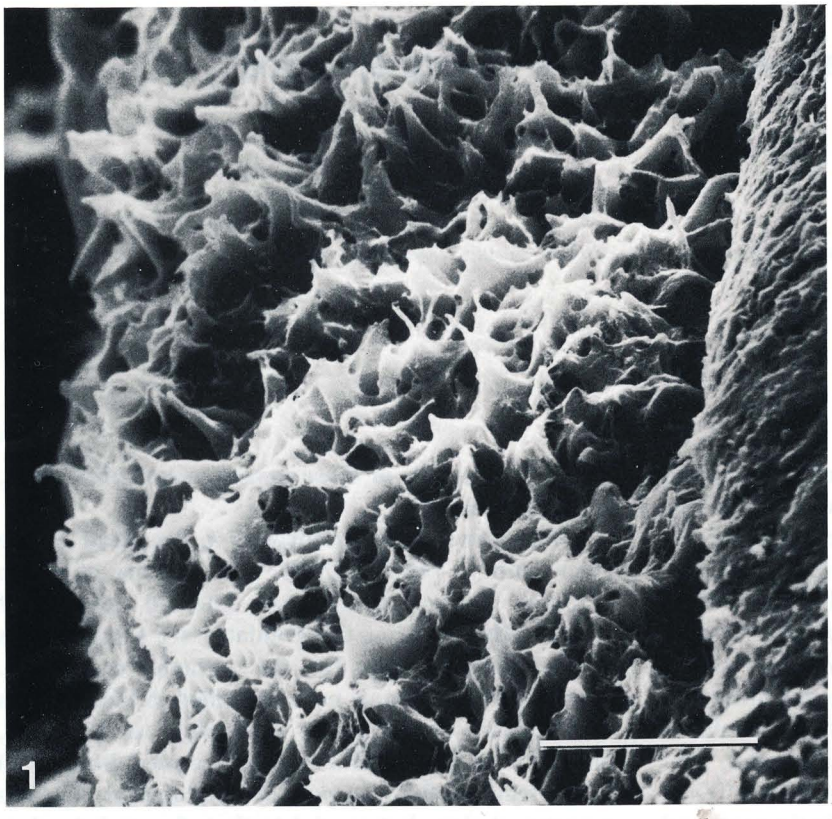

Fig. 1. SEM dorsal view of a somitomere in the segmental plate of a quail embryo. This oblique view facing caudally accentuates the concentric arrangement of cellular processes that are reaching toward the epidermis that has been removed. The centre of the somitomere is in the centre of the photograph. The neural tube runs down the right side. (From Jacobson & Meier, 1986.) Bar, 50 μm.

ive concavities or valleys between successive convexities and, at the borders, cell processes are thrown up in ridges. The ventral face of the somitomere, which apposes the endoderm, usually has a patch of more densely packed cells at the centre of the somitomere, and these cells contact the endoderm by means of a tuft of cellular processes (Fig. 2). The endoderm has a similar tuft of cellular processes that sticks out towards the yolk. The pattern of cell arrangement in the endoderm beneath the somitomere mimics that in the somitomere and the tufts on the endoderm appear to centre on the somitomere that lies, or that will lie, above it (Meier, 1979).

The patterned cell arrangement that constitutes a somitomere is subtle. In some preparations, the extracellular matrix obscures the pattern. Removal of much of this matrix by washing in buffer after dissecting away the ectoderm (or endoderm) is an important step in preparing specimens to be examined for somitomeres by scanning electron microscopy (SEM). Somitomeres are found in the same places at the same stages in different embryos, and a somitomere visualized on one side of the axis has a mirror-image mate on the other side. They are always found in consistent relationships to the parts of the brain and cranial neural crest migrates on the cranial somitomeres in a characteristic pattern (Jacobson, 1987).

Somitomeres were discovered in the head paraxial mesoderm and their discovery makes it clear that there is an early segmentation in the head mesoderm of a craniate, settling an old controversy.

There is another part of the paraxial mesoderm in somewhat older embryos that is not obviously segmented when viewed with light microscopy. This is the segmental plate, a stretch of paraxial mesoderm reaching from the last-formed somite to the retreating primitive streak in the bird embryo. The segmental plate cleaves somites from its rostral end and adds more paraxial mesoderm at its caudal end, either from the anterior end of the streak, or later from the tail bud. The segmental plate is thus a constantly changing population of cells moving caudally in the embryo. Packard & Jacobson (1976) have shown that the isolated segmental plate of the chick embryo will form ten to twelve somites, and concluded that a covert segmentation must exist in the segmental plate. Meier (1979) examined the segmental plate of the chick with stereo SEM and found that it is organized into ten to twelve somitomeres. Segmentation, therefore, is not occurring in the segmental plate, which is already segmented, but must occur before or during formation of the somitomeres.

Since somitomeres are circular, they do not occupy all the space in the band of paraxial mesoderm. Some cells that may not be part of the somitomeres are left in the crotches at the lateral and medial borders between adjacent tandem circular somitomeres. Meier (personal communication) thought that these cells probably contribute to vascular tissue. Vascular

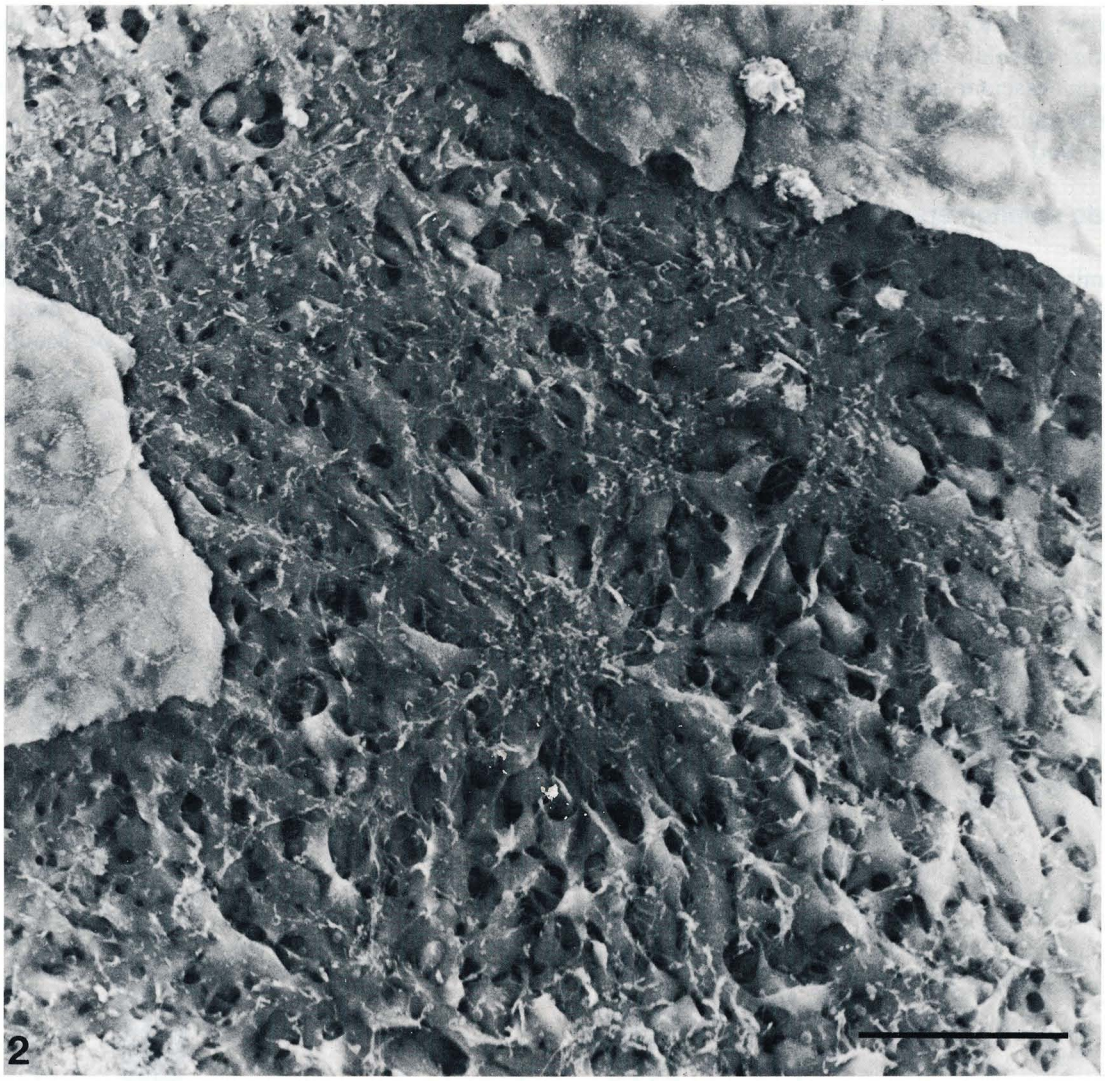

Fig. 2. SEM ventral view of the most recently formed somitomere of a stage-5 chick embryo. A central tuft of cell processes marks the centre of the circular somitomere. The top of the figure is caudal. The fragment of endoderm in the upper right has on its ventral surface (toward the viewer) a tuft of cell processes that predicts the centre of the next somitomere that will form. (From Meier, 1979.) Bar, 50 μm.

elements appear early, especially on the ventral surface of the mesoderm, and these cells are active in migration. Meier (1980) showed an anastomosing vascular network covers the ventral surface of the mesoderm, but appears to have a border that coincides with the lateral border of the somitomeres at stage 10. However, England (1986) has shown by SEM of dissected chick embryos and ink injections into the vascular system that vascular openings from the aorta penetrate intersegmentally between somitomeres. Early vascular elements from the paraxial mesoderm, already in position, may contribute to these intersegmental vessels.

It has also been suggested that prospective vascular elements migrate from the paraxial mesoderm into the lateral plate region. At stages when the intermediate mesoderm can be viewed dorsally with SEM (before the lateral plate folds over the intermediate mesoderm and abuts the somites) there are many cells that lie on the intermediate mesoderm perpendicular to the long axis and the somite border. These cells were shown to be vascular by their binding with a quail vascular endothelial marker, QH1 (Solursh *et al.* 1987). Jacob *et al.* (1978) implanted quail somites into chick embryos, then found quail cells migrating across the intermediate mesoderm and suggested that such cells were migrating myogenic cells, but probably they were vascular elements and the myogenic cells migrate later and may follow the pathways of the vascular tissue. Meier (1980) pointed out that the lateral plate mesoderm has grooves perpendicular to the long axis and coincident with intersomitomeric

boundaries. Meier & Burton (1986) suggest that migrating vascular elements exploit these grooves and form vascular pathways that later migrating myoblasts follow from the somites as they move to the wing bud.

When do somitomeres form?

The somitomeres of the bird embryo are formed in a strict cranial to caudal order, starting at the tip of the head during early gastrulation. During stage 3 (stages by Hamburger & Hamilton, 1951), while the primitive streak is still elongating, the first pair of somitomeres is formed to each side of the anterior tip of the primitive streak, and the prechordal plate, which resembles a somewhat condensed somitomere, is formed in the midline anterior to the streak and between the two somitomeres (Fig. 3) (Triplett & Meier, 1982). By the time stage 4 is reached, when the streak has attained its greatest elongation, the second pair of somitomeres has formed to each side of Hensen's node (Fig. 3) (Triplett & Meier, 1982).

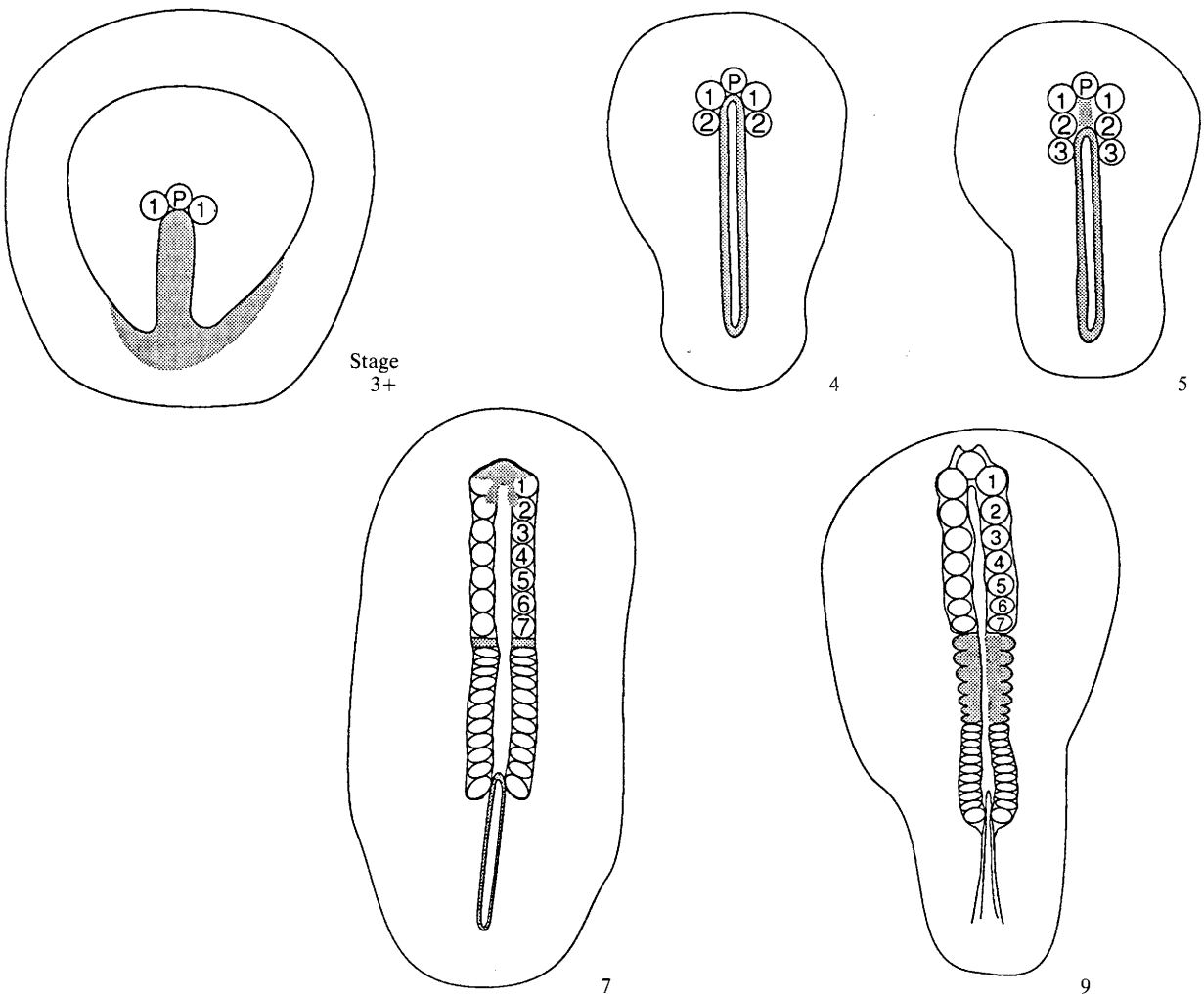

Fig. 3. In this drawing, the somitomeres are placed on outlines of normal stages of the chick embryo. The first pair of somitomeres (numbered 1 in circle), and the prechordal plate (P) at the midline, appear during formation of the primitive streak (shaded) at stage 3+. When the primitive streak has reached its greatest length at stage 4, the second pair of somitomeres (2) has formed just lateral to Hensen's node at the anterior end of the streak. By stage 5, the streak has regressed, laying down notochord along the midline between the somitomeres (shaded column), and to each side of Hensen's node the third pair (3) of somitomeres has formed. This process continues until about 19 somitomeres have formed (the number of caudal somitomeres at this stage is an estimate, perhaps an overestimate). The first somite (shaded bar) condenses from the eighth somitomere. The 11 units without numbers that lie between the somite and Hensen's node compose the segmental plate. Somitomeres continue to form lateral to Hensen's node as the streak regresses, and somites condense from the somitomeres at the cranial end of the segmental plate. By stage 9, seven somites (shaded) have condensed and a sufficient number of somitomeres have formed at the caudal end to keep the numbers of units at the segmental plates at 11.

Spratt (1955) had identified, with defect experiments, a region lateral and slightly caudal to Hensen's node that appeared necessary for somite formation and which he called the 'somite-forming center'. This is the precise region in which newly formed somitomeres appear after stage 4. By stage 5 (Fig. 3), another pair of somitomeres has formed as the streak and node have regressed farther caudad. All subsequent somitomeres are laid down in a sequential manner next to Hensen's node as the streak regresses until the streak is incorporated into the tail bud, after which the tail bud continues to spawn somitomeres. After stage 4, the regressing primitive node lays down notochord along the midline in segmented units that more or less match the somitomeric units (Triplett & Meier, 1982). After a fair number of somitomeres are established, probably about 19 or less, as shown at stage 7 in Fig. 3, the borders separate between the eighth somitomere and its neighbours and the first somite is formed. From then on, additional somites are formed in cranial caudal order following the first one, and new pairs of somitomeres are added tandemly as the streak and node regress caudally down the embryo.

The first three pairs of somites initially have an appearance different from the subsequent somites that form. As Lipton & Jacobson (1976*a*) described, these first three somites develop beneath the neural plate while it is open flat. Their peculiar chevron appearance persists until the plate begins to close into a tube, at which time the somites condense along with the neural plate toward the midline and lift with it as it rolls up. They then assume a more typical cuboidal appearance. The fourth and the subsequent more caudal somites initially form when the neural plate above them has begun to roll into a tube, and their appearance is cuboidal from the beginning. Jacobson (1984) has found this region of forming neural tube that overlies the forming somites to coincide with a wave of elongation that passes down the neural plate as it rolls into a tube. In contrast to the somites, the site of formation of somitomeres is beneath the open neural plate and the lateral edges of somitomeres exactly coincide with the lateral edge of the overlying neural plate.

After several somites have formed, as at stage 9 in Fig. 2, a segmental plate regularly contains ten to twelve somitomeres. New somites condense from the cranial end of the segmental plate, and new somitomeres are added at the caudal end as explained above. Meier (1979) has described the gradual conversion of somitomeres of the segmental plate into somites. At the caudal end of the segmental plate, the newly formed somitomere looks much like a cranial somitomere. Its dorsal surface is about the same size (175–185 μm in diameter), and it consists of a bilaminar, squat cylinder of loose mesenchymal cells arranged in concentric circles. Going cranially, the maturing somitomeres are more condensed. The next three somitomeres are 145–165 μm in diameter, the three cranial to them are 112–130 μm in diameter and are wedge shaped in cross section because they are shaping around the neural tube as it begins to form here. The next more cranial three somitomeres are 85–95 μm in diameter and are cuboidal in cross section. The most cranial of these will be the next somite to form. The three most recently formed somites are 95–105 μm in diameter. There are clefts discernible with SEM between somitomeres, especially as they approach transformation into somites. When the cleft becomes wide enough to be seen with light microscopy, the segmental unit is somewhat arbitrarily called a somite. (Elsdale & Davidson (1983) commented that the line between segmented and unsegmented mesoderm in the *Xenopus* embryo depends on how closely you look.) From the node to the last-formed somite, the somitomeres are progressively more compacted and more epithelial. The space between the two layers of the newly formed bilaminar somitomere is gradually converted into the myocoel of the somite as the mesenchymal cells become epithelial with their apical ends towards this central lumen. Junctional complexes progressively form, tying the apical ends of the cells together (Meier, 1979).

Growth (increase in volume) undoubtedly occurs during and after somitomere transformation into somites, at least in amniote embryos. Packard & Jacobson (1979) measured the increase in volume through time of the sixteenth somite of the chick embryo. The volume doubled about every five to eight hours. Meier (1979) observed mitotic cells (especially at the incipient luminal surface) in the transforming somitomeres of the segmental plate, but I know of no direct measurements of volume change in these somitomeres, and such measurements would be complicated by the compaction of the cells with loss of extracellular space that is occurring.

Somitomeres of the segmental plate become somites

Besides the observational evidence given above, implying that somitomeres of the segmental plate transform into somites, there is confirming experimental evidence. Using both chick and quail embryos, Packard & Meier (1983) made parallel cuts across the axis, one cut just posterior to the last-formed somite, and the other just anterior to the primitive streak. A cut was then made down the midline to form two explants with equal amounts of

segmental plate established by the parallel cuts. One segmental plate was processed immediately for examination by stereo SEM to determine the total number of somitomeres in the excised segmental plate. The other explant was cultured for five to eight hours (during which time several somites formed), then fixed for SEM. The total number of somites plus somitomeres was then determined in this explant. There was excellent agreement of the total numbers of segmental units in these paired explants. Of five cases, three had identical numbers of units, in one case the cultured explant had 11 units and the other 10·5, and in the final case the cultured explant had 7·5 units and the other had 8. Similar studies with similar results have been done on the embryos of the snapping turtle, *Chelydra serpentina* (Packard & Meier, 1984), and the mouse (Tam *et al.* 1982; Tam & Beddington, 1986). These studies give quite clear evidence that somitomeres of the segmental plate convert into somites.

Bellairs (1986) reviews and describes the events at the rear end of the chick embryo after Hensen's node becomes incorporated into the tail bud. She illustrates somitomeres in the tail bud as far back as its terminal portion, but somites never extend to the tip of the tail bud. She raises the interesting question of why the embryo stops making somites. She suggests one factor in the failure to form somites from the somitomeres at the tip of the tail bud may be that these terminal somitomeres are overtaken by cell death.

The fates of the head somitomeres

The most cranial seven somitomeres of the head of the chick embryo do not ordinarily form into somites. Rather, some grow larger and they all ultimately disperse (Meier, 1981). The first somitomere forms during stage 3 and, like most somitomeres, is initially about 175 μm in diameter. By stage 8 it is 240 μm in diameter. The second and third somitomeres have formed by stage 5, and by stage 8 they too are 240 μm in diameter. The fourth somitomere is more closely packed and is 170 μm in diameter at stage 8. The fifth somitomere is even more compact and is 160 μm in diameter at stage 8. The sixth somitomere has condensed to 135 μm in diameter by stage 8. All the somitomeres of the head make rather complex three-dimensional changes of shape following their formation as simple bilaminar discs. The seventh head somitomere is condensed like the sixth. It does not have a sharp boundary with the sixth somitomere, but its caudal border with the eighth segment (the first somite) is sufficiently separated to be visible with light microscopy. The seventh somitomere has its cells arranged more like those of the first somite than of its neighbouring sixth somitomere (Meier, 1981), and in SEM is easily mistaken for a somite.

Patterson (1907) examined the question of whether 'rudimentary somites' form cranial to the first somite in the chick embryo. He marked the mesoderm just anterior to the first somite by cautery or by pinning with a small glass pin. Later, after several more somites had formed, he found the cautery mark or the pin was still in position just anterior to the first somite, so no new somites had separated in front of the one first seen. During this study he did note the near-somite nature of what we now know to be the seventh somitomere, and other 'shallow transitory depressions' at regular intervals along the cranial paraxial mesoderm. He was probably observing the shallow intersegmental gaps between successive somitomeres.

The seven pairs of head somitomeres ultimately contribute the voluntary muscles of the head, some bone and dermal and meningeal tissue (Noden, 1982, 1983*a*). These contributions to the head were established by Noden with quail to chick transplants of somitomeric material and use of the quail nucleolar marker to identify the products of the somitomeres. The somitomeres essentially make in the head the same products that the myotomes, sclerotomes and dermatomes of the somites make in the trunk region. In the head, though, the pattern of muscle formation is controlled by the cranial neural crest that contributes the connective tissue of the head muscles. Noden (1983*b*) used transplantations to establish that this ability of the cranial neural crest to pattern the head muscles is programmed prior to the onset of neural crest migration when the precursors of the crest cells are still part of the neuroepithelium. So the cells that form the voluntary muscles of the head arise from the head somitomeres, but the pattern of muscle condensation is controlled by the cranial neural crest. This is just one way that the cranial neural crest has modified the development of the vertebrate head.

The relatedness of head somitomeres and trunk somites is suggested by more than the gradual gradation of one into the other. Noden (1986) has transplanted quail trunk somites or somitomeres in place of chick head somitomeres and found that the transplanted units produce myoblasts that are incorporated into voluntary muscles of the head. In the same paper, Noden reported preliminary results with transplantation of head somitomeres into the trunk region where they contributed to the appendicular muscles.

In some studies, somitomeres of the head appear to have converted into somites. Lipton & Jacobson (1974*b*) did some experiments with the initial goal of identifying the mechanisms by which somites are

elicited in the chick embryo. Stage-4 embryos were transected just caudal to Hensen's node and the cranial part was discarded. The posterior fragment of area pellucida contained primitive streak, but lacked Hensen's node and Spratt's 'somite-forming centers'. If the posterior fragment was cut in two down the midline of the primitive streak, both right and left halves formed rows of somites on the cut edges after 15 h more of incubation. If the posterior fragment were not split in two, no somites formed. We suggested that a prepattern of segmentation exists in these fragments, and development of somite pairs is dependent upon the shearing action of the regressing node, or could be released by cutting in the absence of the node. These experiments were done several years before somitomeres were discovered.

Meier & Jacobson (1982) repeated these and additional experiments and also examined the fragments with stereo SEM for the presence of somitomeres. The stage-4 embryo already has the first two pairs of somitomeres in place (Fig. 3) and our transverse cut posterior to Hensen's node removed these already-formed units. The posterior fragment should form five more head somitomeres, then somitomeres that become the anterior somites. When severed down the midline, the posterior fragments form their first somites after 4 h of incubation, and seven to eight somites are formed by 15 h. After 15 h of incubation, the posterior fragments were fixed and examined with stereo SEM. The whole medial edge of each isolate was composed into somites (cranially) and somitomeres (caudally). There should have been five head somitomeres at the anterior ends of these split fragments before the site of the first somite, but somites are present all the way to the cranial extent of the cut edge. This implies that prospective head somitomeres have become somites under these conditions.

In posterior fragments treated the same way, but not split down the midline, no somites formed after 15 h of incubation, but examination with stereo SEM of the dorsal aspects of the paraxial mesoderm of these fragments (after removal of the ectoderm) revealed two paraxial rows of somitomeres. Since fate maps show that the prospective paraxial mesoderm is located in the epiblast and streak, we marked the surface of both classes of posterior fragments (split or unsplit) with carbon, and found that epiblast converges toward the midline and ingresses into the streak. Gastrulation is thus continuing in the explants even though the node is missing. It appears that the cut edges heal and that epiblast and streak cells continue to ingress to form paraxial mesoderm. These studies are reviewed and amplified in Jacobson & Meier, 1986.

Packard (1986) has used tritiated thymidine and the quail nucleolar marker to identify the products of small implants placed in the epiblast near the primitive streak of chick embryos of Hamburger & Hamilton stages 3+ to 9+, that is, from early gastrula stages to when the embryo has eight somites. His experiments demonstrate that paraxial mesoderm cells continue to arise from the epiblast at least until the eight somite stage. The paraxial mesoderm has been mapped in the epiblast near the anterior third to fourth of the length of the primitive streak (Rosenquist, 1966; Nicolet, 1971), and Bellairs & Veini (1984) and others have found that when the embryo is cut across the primitive streak posterior to this presumptive paraxial mesodermal area, somites never form in the posterior piece. Tam & Beddington (1987), using labelled grafts, have demonstrated that the anterior streak is the origin of paraxial mesoderm also in the mouse embryo.

Where does segmentation of the mesoderm take place?

Now that we know that the segmental plate is full of somitomeres (Meier, 1979), and that the stage-4 embryo, posterior to Hensen's node, is also covertly segmented (Lipton & Jacobson, 1974*b*; Meier & Jacobson, 1982), the question of where segmentation of the paraxial mesoderm occurs has a new perspective. The segmental plate is already segmented, so segmentation must occur as a new somitomere is added lateral to Hensen's node, or there is covert segmentation into prospective somitomeres in the prospective paraxial mesoderm while it is still in the streak and epiblast.

Another interesting possibility has been raised by experiments of Bellairs & Veini (1984). They isolated various fragments of the area pellucida, some with and some without the primitive streak. Based on the presence, numbers and sizes of somites that formed in these pieces, they have suggested that a population of somite precursor cells is generated as the primitive streak forms and these become located in the presumptive somite regions by stage 4. Later, cells leave the primitive streak and interact with the precursor cells to form somites. Regression rearranges precursor cells along the axis and somitomeres form from these as more cells are added to them from the regressing streak. Eventually the somitomeres form somites.

Another interpretation of the experiments of Bellairs & Veini (1984) is possible. Prospective paraxial mesoderm lies in the primitive streak and also in the epiblast to each side that has not yet entered the streak. When Bellairs and Veini cut along side the primitive streak to create fragments with and without streak, the fragment with streak would have both the

part of the paraxial mesoderm that is already in the streak and the part still in the epiblast, while the fragment lacking the primitive streak would have only that part of the paraxial mesoderm population that is in the epiblast. In the fragment lacking streak, ingression of the paraxial mesoderm in the epiblast could occur around the cut edge to form the smaller somites seen in such fragments. The fragment containing both streak and epiblast would have more than a full complement of paraxial mesoderm (streak complement for both sides and epiblast complement for one side). Ingression of this population could account for the larger, greater numbers, and occasional double rows of somites that form in such fragments.

Bellairs and Veini could also be correct in their interpretation that a population of somite precursor cells is in position in prospective somite regions by stage 4. There is certainly mesoderm in those regions at this stage, but it has generally been thought that this is prospective lateral mesoderm that will be displaced laterally when the paraxial mesoderm ingresses. Meier & Jacobson (1982) noted that the mesoderm that occupies the paraxial region posterior to Hensen's node in the stage-4 chick embryo, visualized after removal of the epiblast, appears ribbed with seams of cellular alignment that run nearly perpendicular to the long axis at intervals that give a metameric appearance (their fig. 3). Possibly, the troughs between these ribs could help collect ingressing streak cells into somitomeres. It is even possible that the intercalated embryonic endoderm, which is put into position before stage 4, has some role in organizing the mesoderm above it. As pointed out above, Meier (1979) noted that the endodermal cells that underlie the somitomeres mimic the somitomeric cellular arrangement and 'buttons' of cellular processes appear in the endoderm where a somitomere will centre.

Bellairs (1986) has critically evaluated the various models of segmentation, some of which do not incorporate the presence of the already segmented somitomeres.

However the segmentation of the paraxial mesoderm is accomplished, it is done in regions that are active in gastrulation processes. Presomitomeric cells and their neighbours are actively moving about and it seems difficult to accommodate to this situation models that require gradients of morphogens or such. The segmentation process must involve epiblast cells that are loosing themselves from the epithelial epiblast, ingressing through the compacted primitive streak, then abruptly spreading in patterned array in the paraxial region. A newly formed somitomere has the appearance of an expansion figure. Mechanical processes may have a considerable role in a transformation such as this.

The cells of forming or newly formed somitomeres must in some manner be in communication since they are motile mesenchymal cells that have a coordinated group behaviour. One way they could be in communication is by forming transient gap junctions within the populations behaving as units. Warner (1986), in *Xenopus* and axolotl embryos, has shown by the presence of electrical coupling that cells of the somitomeric mesoderm about to form the somites are linked by gap junctions. It is not clear from her paper whether or not she has tested to see if successive somitomeres are isolated.

Experimental methods such as heat shock can result in disruption of somite formation, but there is a typical lag period before the effects are seen, and the effects continue for a discrete number of somites depending on the duration and intensity of the shock. In amphibia (Elsdale *et al.* 1976; Cooke, 1978; Elsdale & Pearson, 1979; Pearson & Elsdale, 1979; Cooke & Elsdale, 1980; Elsdale & Davidson, 1986; Armstrong & Graveson, 1988), and in the chick (Veini & Bellairs, 1986), the numbers of unaffected somites that appear after a heat shock fall within the numbers of somitomeres in the segmental plate of the embryo. The heat shock affects only the more recently formed somitomeres and prospective paraxial mesoderm not yet formed into somitomeres.

Somitomeres have been described in five classes of vertebrates

As discussed above, somitomeres were first discovered and described in chick embryos (Meier, 1979, 1980, 1981, 1982*a*,*b*, 1984; Meier & Jacobson, 1982; Anderson & Meier, 1981; Packard & Meier, 1983; Triplett & Meier, 1982). Somitomeres whose developmental history and appearance closely resemble those of the chick have been described in another avian species, in the embryos of the Japanese quail, *Coturnix coturnix japonica* (Meier, 1982*b*; Packard & Meier, 1983; Triplett & Meier, 1982).

Somitomeres have also been described in representatives of the mammals and the reptiles. In embryos of the laboratory mouse (*Mus domesticus*, CF-1 albino strain), somitomeres are formed during gastrulation. They are formed lateral to the primitive streak in the same order as described for the chick, and the patterning of the cells that compose a somitomere face is very similar in chick and mouse. There are seven head somitomeres that do not form somites in the mouse, and their relationships to brain parts are the same as in the chick embryo (Meier & Tam, 1982; Tam & Meier, 1982). In the mouse embryo, the segmental plate is called the presomitic mesoderm.

Where the avian embryos had ten to twelve somitomeres in the segmental plate, the mouse embryo has but six somitomeres in the presomitic mesoderm (Tam *et al.* 1982).

As a representative of the reptiles, embryos of the snapping turtle (*Chelydra serpentina*) have been examined for somitomeres (Packard & Meier, 1984; Meier & Packard, 1984). Somitomeres form during gastrulation in this species much as they do in the bird and mouse embryos. There are seven head somitomeres and about six somitomeres in the segmental plate, as in the mouse.

Since amniotes gastrulate with primitive streaks, Meier and I thought that some anamniote embryos that gastrulate in entirely different ways should be examined for somitomeres. We looked at a newt (*Taricha torosa*) as a representative of the urodeles (Jacobson & Meier, 1984), and Meier had nearly completed a study of somitomeres in an anuran (*Xenopus laevis*) when he died in January of 1986 (Jacobson & Meier, 1986). Both of these species form somitomeres during gastrulation, and they form in the usual cranial to caudal order. By the end of gastrulation (stages 13 and 14), the newt embryo paraxial mesoderm is composed of seven to eight somitomeres, so the embryo is already segmented from head to tailbud before the appearance of the first somite. The tailbud continues to add additional somitomeres. At stage 17 (a midneurula stage), the first somite forms that is visible by light microscopy. Stereo SEM reveals that this somite has formed from the seventh somitomere. I have re-examined our file of SEM prints of this stage to estimate the number of somitomeres present posterior to the first somite. At least four, and probably six, somitomeres lie caudal to this first somite. Four are clearly visible, then the segmental plate makes a turn. I estimate two more somitomeres are in the turn. The stage-17 embryo has, therefore, eleven to thirteen mesodermal segments along its axis when the seventh one condenses into a somite. At this 'one-somite' stage, both segments 5 and 6 can be seen to have deep clefts at their borders if viewed form the ventral side. These two segments soon separate dorsally as well and then can be seen to be somites. In the newt, therefore, the first somite to form (the seventh segment) is not the most anterior somite (which is the fifth segment). Newt somites appear in the order 7, 6, 5, 8, 9, 10, etc. Four somitomeres remain cranial to the most anterior somite, and these do not normally form somites.

In all the forms studied, the pair of first somitomeres is associated with the prosencephalon. The more caudal somitomeres that follow the first differ in the amphibia and the amniotes. The rule followed is that for each somitomere found in the newt or *Xenopus*, there are two somitomeres in the amniote

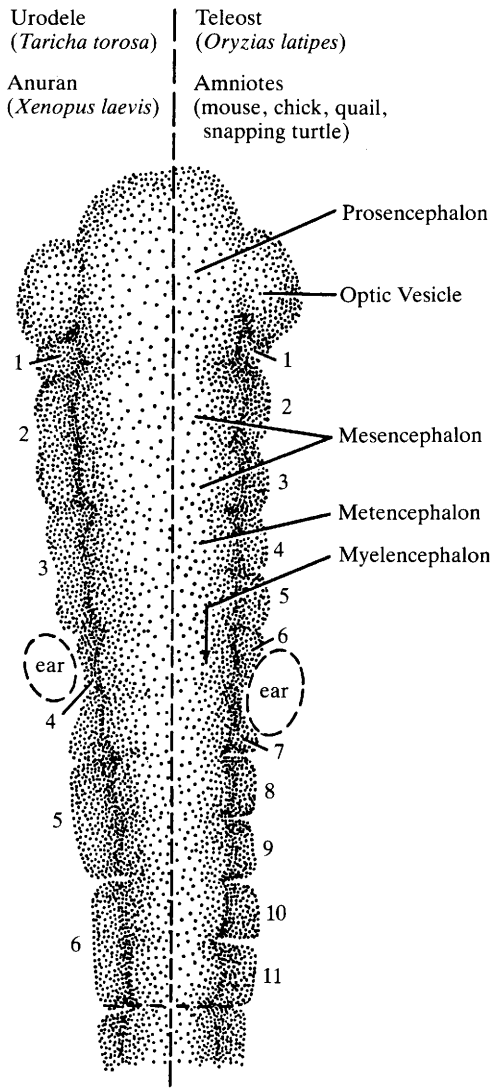

Fig. 4. Somitomeres (numbered) in amphibia are compared to those in amniotes and a teleost. The position of the ear vesicle is noted. In all forms, the first somitomere is next to the prosencephalon. Caudal to the first somitomere, wherever the amphibian has one somitomere, the amniote or teleost has two. The arrangement of mesodermal segments in the heads of shark embryos is similar to that of the amphibia. Note relationships of somitomeres to the brain parts and neuromeres.

embryos (Fig. 4). For example, one somitomere spans the length of the mesencephalon in the amphibian embryo, but the same region is occupied by two somitomeres in the amniote embryo. The same rule continues into the trunk regions and this may help account for the generally more anterior (by count of spinal nerves) innervation of the forelimb in amphibia as compared to the amniote embryos.

After some somites have formed, there is a region of mesoderm between the last-formed somite and the

Table 1. *Characteristics of somitomeres in different vertebrate embryos*

Example	No. of somitomeres in head	No. of somitomeres in segmental plate	Size of head somitomere	No. of cells on somitomere face	Reference
Laboratory mouse	7	5–7	80 μm	100	a
Chick and quail	7	10–12	175 μm	200–300	b
Snapping turtle (*Cheledra serpentina*)	7	6–7	90 μm	230	c
Taricha torosa (newt) (Urodele)	4	5–7	285 μm	38	d
Xenopus laevis (Anuran)	4	5–7	207 μm	35	e
Oryzias latipes (Teleost)	7	10	32 μm	56	f

a, Meier & Tam, 1982; b, Meier, 1979, 1982*b*; c, Meier & Packard, 1984; d, Jacobson & Meier, 1984; e, Jacobson & Meier, 1986; f, Martindale, Meier & Jacobson, 1987.

tailbud, in embryos of the newt and *Xenopus*, that is similar to the segmental plate of the bird embryo. This strip of mesoderm contains five to seven somitomeres in each of these forms.

The arrangement of the somitomeres in the head of the newt resembles closely the appearance of mesodermal segments in the head of a shark embryo (Goodrich, 1930; Jacobson & Meier, 1984). Like the amphibia, embryos of sharks have three mesodermal segments anterior to the otocyst and the fourth segment lies beneath the otocyst. One might have concluded from this that ancestral vertebrates had the lesser numbers of somitomeres seen in sharks and amphibia and the line that led to the amniotes had doubled their somitomere number (posterior to the first one), perhaps by splitting the units in two. We have now examined a teleost (the medaka, *Oryzias latipes*) and find that this form contains somitomeres in the arrangement seen in the chick embryo. The medaka has seven somitomeres in the head anterior to the first somite, and it has ten somitomeres in its segmental plate (Martindale, Meier & Jacobson, 1987). This finding opens the possibility that the ancestral form may have had seven head somitomeres, and this number has been converted to four twice, once in the shark line and once in the amphibian line. Such a conversion could have been by a fusing of pairs of segments. One would expect that somitomere number is a primordial trait, but there is not sufficient information to judge its history yet. It would be quite interesting to see how somitomeres are arranged in lungfish.

Data for the forms that have been studied so far are summarized in Fig. 4 and in Table 1.

When a somitomere first forms as a bilaminar disc, the numbers of cells that constitute the top of the cylinder can be counted or estimated (Table 1). A surprising observation emerges. A newly formed somitomere of a chick embryo has from 200–300 cells constituting the face of a somitomere that is about 175 μm in diameter. A newly formed somitomere of the newt, in contrast, is 285 μm in diameter, but its face is composed of just 38 cells. The much larger cells of the amphibian embryo have organized a somitomere pattern out of what must be about the minimum number of cells that can form such a pattern. It seems possible that amphibian embryos, which are composed of large, yolk-filled cells, may be restricted by this feature to having fewer and larger somitomeres.

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