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REVIEW

Vertebrate Cranial Placodes I. Embryonic Induction

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Cranial placodes are focal regions of thickened ectoderm in the head of vertebrate embryos that give rise to a wide variety of cell types, including elements of the paired sense organs and neurons in cranial sensory ganglia. They are essential for the formation of much of the cranial sensory nervous system. Although relatively neglected today, interest in placodes has recently been reawakened with the isolation of molecular markers for different stages in their development. This has enabled a more finely tuned approach to the understanding of placode induction and development and in some cases has resulted in the isolation of inducing molecules for particular placodes. Both morphological and molecular data support the existence of a preplacodal domain within the cranial neural plate border region. Nonetheless, multiple tissues and molecules (where known) are involved in placode induction, and each individual placode is induced at different times by a different combination of these tissues, consistent with their diverse fates. Spatiotemporal changes in competence are also important in placode induction. Here, we have tried to provide a comprehensive review that synthesises the highlights of a century of classical experimental research, together with more modern evidence for the tissues and molecules involved in the induction of each placode. © 2001 Academic Press

Key Words: **placode; olfactory; hyophyseal; adenohypophysis; lens; trigeminal; lateral line; otic; epibranchial; zebrafish; *Xenopus*; axolotl; chick; mouse.**

INTRODUCTION

Cranial placodes are transient, discrete regions of thickened columnar epithelium that form in characteristic positions in the head of vertebrate embryos. They make vital contributions to the paired sense organs (nose, eyes, ears, and lateral line) and to cranial sensory ganglia (Ariëns Kappers, 1941; Le Douarin *et al.*, 1986, 1992; Vogel, 1992; Webb and Noden, 1993; Northcutt, 1996; Graham and Begbie, 2000). They are essential for the formation of the sensory nervous system in the head. They form a wide variety of cell types, including ciliated sensory receptors, sensory neurons, neuroendocrine and endocrine cells, glia, and other supporting cells. Like the neural crest, which forms the rest of the peripheral nervous system together with pigment cells and much of the cranial skeleton (Hall, 1999; Le Douarin and Kalcheim, 1999), cranial placodes form at the border of the neural plate and epidermis and give rise to migratory cells. It has been suggested that neural crest and placodes may have evolved from a common ancestral population present in vertebrate ancestors (Gans and Northcutt, 1983; Baker and Bronner-Fraser, 1997b).

Despite their importance for the development of the vertebrate sensory nervous system, and a research tradition stretching back well over a century (van Wijhe, 1883; Froriep, 1885; Beard, 1886; von Kupffer, 1891), placodes are often ignored today. In recent years, the availability of molecular markers for different placodes at various stages of their development has reawakened an interest in their induction and development and has led to the beginnings of a molecular characterisation of placode induction. The aim of this review is to provide a comprehensive synthesis of past and current research on placode induction; an accompanying review will address the more speculative issue of placode evolution (C. V. H. Baker and M. Bronner-Fraser, in preparation). After a general introduction to the different placodes and the evidence for a preplacodal domain at the border of the cranial neural plate, we take each placode in

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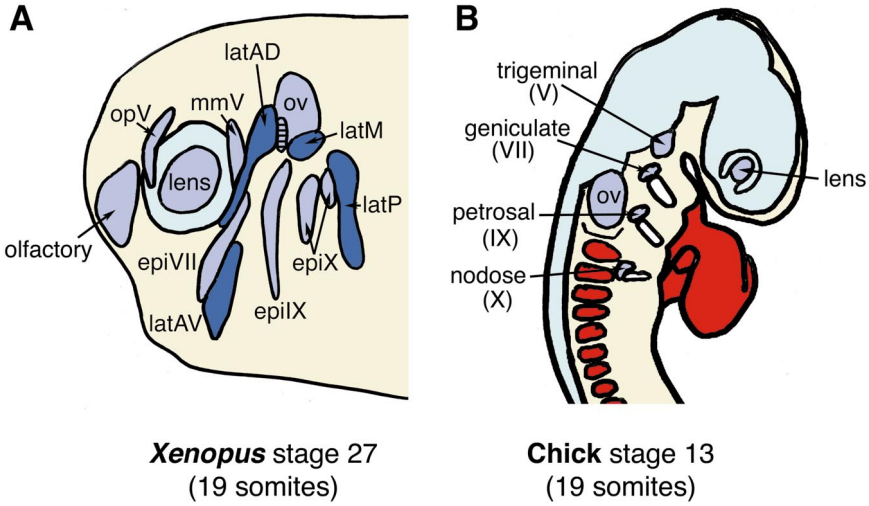

FIG. 1. Location of cranial placodes in amphibian and chick embryos. (A) Cranial placodes in a 19-somite stage *Xenopus* embryo. Hatched area represents remaining otic placode. epi, epibranchial placode; epiVII, facial/geniculate placode; epiIX, glossopharyngeal/petrosal placode; epiX, vagal/nodose placodes; lat, lateral line placode; latAD, anterodorsal lateral line placode; latM, middle lateral line placode; latP, posterior lateral line placode; mmV, maxillomandibular trigeminal placode; opV, ophthalmic trigeminal placode; ov, otic vesicle. After Schlosser and Northcutt (2000). (B) Cranial placodes in a 19-somite stage chick embryo. After Le Douarin *et al.* (1986).

turn and describe the available experimental evidence pertaining to its induction.

Location, Function, and Derivatives

The location of most of the cranial placodes in 19-somite stage *Xenopus* and chick embryos is shown in Fig. 1. Although the focal ectodermal thickenings that give rise to hairs, feathers, and teeth are also called placodes, we refer here solely to the cranial placodes associated with the nervous system that arise from ectoderm at the border between the neural plate/neural crest and epidermis. Fate maps for cranial placodes in neural plate and neurula stage amphibian and chick embryos are shown in Figs. 2 and 3. In anteroposterior order, the cranial placodes include the hypophyseal, olfactory, lens, trigeminal, otic, lateral line, epibranchial, and presumably also the recently discovered hypobranchial placodes (Schlosser *et al.*, 1999; Schlosser and Northcutt, 2000). In a review of the first 60 years of placode research, Ariëns Kappers defined placodes as "local ectodermal thickenings of the embryonic head that show extensive differentiation and can participate in the formation of sense organs and elements of the peripheral nervous system" (Ariëns Kappers, 1941). He included in his definition all the placodes described here except the lens placode, which was excluded owing to its nonneural fate. We include the lens placode here by virtue of its origin from ectoderm at the neural plate border and its intimate association with the sensory nervous system. The olfactory, lens, otic, and lateral line placodes form or contribute to the paired sense organs, while the trigeminal and epibranchial placodes form sensory neurons in cranial sensory ganglia (Fig. 4).

The *olfactory* placodes form the olfactory neurons and supporting cells of the paired nasal epithelia, as well as neuroendocrine cells and glia that migrate into the brain. The *hypophyseal* placode gives rise to the endocrine adenohypophysis ("anterior" or "epithelial" pituitary). The *lens* placodes form the lenses of the paired eyes. The *trigeminal* placodes (ophthalmic and maxillomandibular) form primary sensory neurons in the distal portions of their respective trigeminal lobes or ganglia (Fig. 4) that transmit somatosensory stimuli (touch, pain, temperature) from the skin of the face and jaws and the teeth. The *dorsolateral series* of placodes includes the *otic* and, in anamniotes, *lateral line* placodes, which give rise to the hair cells of the paired inner ear and lateral line sense organs, together with their epithelial supporting cells and afferent innervation. The inner ear provides balance and auditory information, while the lateral line system includes both mechano- and electroreceptors and senses disturbances in the surrounding water and electrical fields. The *epibranchial* placodes form immediately caudodorsal to where the pharyngeal pouches contact the ectoderm, i.e., above the branchial clefts. These placodes (the geniculate, petrosal, and nodose) give rise to sensory neurons in the eponymous distal ganglia of cranial nerves VII (facial), IX (glossopharyngeal), and X (vagal) (Fig. 4). They provide afferent innervation for taste buds and for a broad spectrum of receptors in the heart and other visceral organs, transmitting information concerning, for example, heart rate, blood pressure, bronchial irritation, and gut

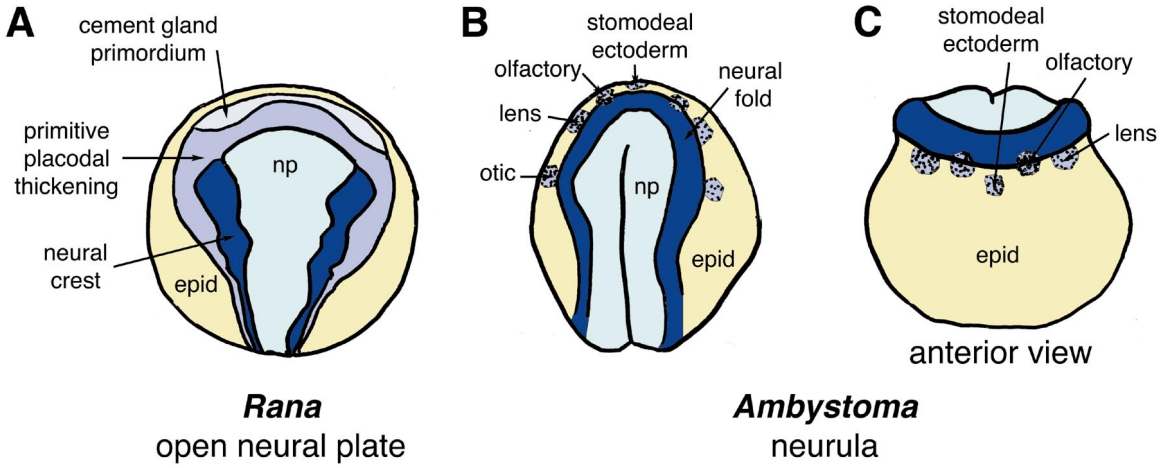

FIG. 2. Cranial placodes fate map to the anterior neural plate border in amphibian embryos. (A) Fate map of open neural plate stage *Rana* embryo (dorsal view), showing primitive placodal thickening. After Knouff (1935). (B) Dorsal view and (C) ventral view of early neurula stage *Ambystoma* embryo fate map. Stomodeal ectoderm represents hypophyseal placode. After Carpenter (1937). epid, epidermis; np, neural plate.

distension. A more detailed description of the function and derivatives of each placode is given in later sections.

Evidence for a Preplacodal Ectoderm Field at the Neural Plate Border

Fate-mapping experiments in gastrula and neurula stage embryos have shown that cranial placodes originate from ectoderm at the border of the neural plate/neural crest and future epidermis (Röhlich, 1931; Knouff, 1935; Carpenter, 1937; Fautrez, 1942; Rudnick, 1944; Jacobson, 1959; van Oostrom and Verwoerd, 1972; Verwoerd and van Oostrom, 1979; Rosenquist, 1981; D'Amico-Martel and Noden, 1983; Couly and Le Douarin, 1985, 1990; Tam, 1989; Garcia-Martinez *et al.*, 1993; Kozlowski *et al.*, 1997; Schlosser and Northcutt, 2000). Figures 2 and 3 show fate maps for the cranial placodes in amphibian and chick embryos. The ectoderm that will form the olfactory and hypophyseal placodes is incorporated into the anterior neural folds (anterior neural ridge), on the outer side of the fold (Carpen-

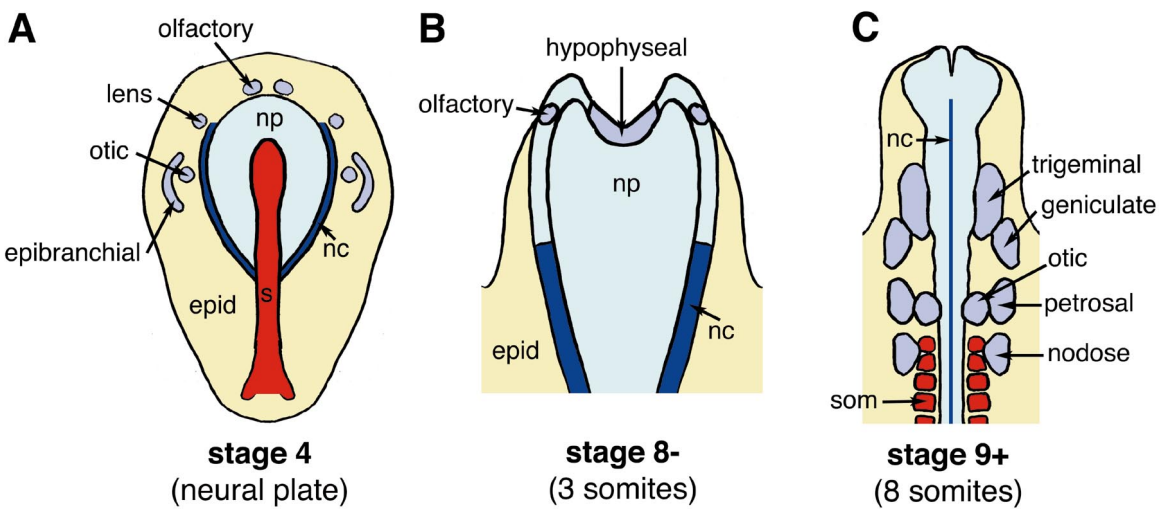

FIG. 3. Cranial placodes fate map to the anterior neural plate border in chick embryos. (A) Neural plate stage fate map. After Rudnick (1944). (B) Neurula stage fate map of the anterior neural folds. After Couly and Le Douarin (1987). (C) Fate map at the 8-somite stage. After D'Amico-Martel and Noden (1983). epid, epidermis; nc, neural crest; np, neural plate; s, primitive streak; som, somites.

ter, 1937; Couly and Le Douarin, 1985; Eagleson *et al*., 1986; Rubenstein *et al*., 1998; although see Whitlock and Westerfield, 2000). This region of the neural plate border does not usually form neural crest cells. All remaining placodes form from ectoderm immediately lateral to the cranial neural crest-forming region. Parts of the prospective trigeminal and otic placode ectoderm may also be incorporated into the lateral ectoderm of the neural folds in the chick (Couly and Le Douarin, 1990; Stark *et al*., 1997; Baker *et al*., 1999).

Placodes are often discussed as completely separate entities. However, multiple placodes may form from discrete ectodermal thickenings, especially in fish and amphibians. For example, in the lamprey embryo, the hypophyseal and olfactory placodes form from different regions of a large placode (Braun, 1996). Similarly, a common hypophyseal and olfactory placode can be seen in *Rana* (Knouff, 1935). In the cod embryo at neural keel stages, a continuous dorsal ectodermal thickening is observed that later segregates into the lens placode, a midbrain-level placode, and a hindbrain level placode that includes otic and lateral line placodes (Miyake *et al*., 1997). The thickening continues caudal to the otic vesicle but disappears towards the trunk (Miyake *et al*., 1997), suggesting that it may also include the postotic epibranchial placodes. In zebrafish, the homeobox gene *Prox1* is expressed in a broad region of head ectoderm at the 10-somite stage from the eye to the otic vesicle and is later expressed in the trigeminal ganglion, otic vesicle, and lateral line primordium (Glasgow and Tomarev, 1998). In *Xenopus*, a dorsolateral placode area at neurula stages ultimately gives rise to the otic placode and most lateral line placodes (Schlosser and Northcutt, 2000).

Furthermore, there are substantial morphological and molecular data from several different species to support the existence of a general preplacodal domain within the cranial neural plate border at gastrula and neurula stages. As the origin of placodes from a common anlage is debated (see, e.g., Graham and Begbie, 2000), this evidence is given below in some detail.

Fish. In the zebrafish (*Danio rerio*) at midgastrula (50% epiboly; 5.5–6 h; Kimmel *et al*., 1995), fate-mapping data indicate that precursors of all the different placodes are arranged already in the expected anteroposterior order, in an overlapping territory at the border of the prospective anterior neural plate (Kozlowski *et al*., 1997). The homeobox genes *dlx3* and *dlx7* are both expressed in late gastrula stage embryos (8 h) in a stripe corresponding to the cells of the future neural plate border (Akimenko *et al*., 1994; Ellies *et al*., 1997). By the end of gastrulation, this stripe has converged to the edge of the developing neural keel (Akimenko *et al*., 1994; Ellies *et al*., 1997). Expression is then restricted at the 6-somite stage (12 h) to the cells of the future olfactory and otic placodes and is also seen much later, at 24 h, in the trigeminal ganglia (Akimenko *et al*., 1994; Ellies *et al*., 1997). The homeodomain transcription factor *six4.1* and the transcription cofactor *eya1* are both expressed at the end of gastrulation (10 h) in a horseshoe-shaped domain surrounding the anterior half of the developing neural keel (Kobayashi *et al*., 2000; Sahly *et al*., 1999). Expression of both genes is progressively restricted to the olfactory, hypophyseal, otic, and lateral line placodes; neither gene is expressed in the trigeminal or lens placode, although expression of *six4.1* is seen at 24 h in the trigeminal ganglia (Kobayashi *et al*., 2000; Sahly *et al*., 1999). A similar expression pattern of *Eya1* is seen in the medaka (*Oryzias latipes*), while the Sry-related HMG box transcription factor *Sox3* in this species is expressed in presumptive placodal ectoderm and in all placodes including the lens, as well as in the central nervous system (Köster *et al*., 2000).

Amphibians. In amphibians, placodes form as thickenings in the inner (sensorial) layer of the ectoderm (see, e.g., Northcutt and Brändle, 1995; Schlosser and Northcutt, 2000). (The hatching gland and cement gland, which both form at the anterior neural plate border, are not considered here as they primarily arise from surface ectoderm; Drysdale and Elinson, 1992). In the urodele *Necturus*, the ectoderm at the lateral edge of the neural folds thickens from anterior to posterior as the neural folds develop, forming a wide band of deep ectoderm that later forms all the placodes (Platt, 1896). In the frog *Rana*, a primitive placodal thickening is seen at early neural plate stages around the anterior neural plate (where no neural crest forms) and around the neural crest; this primitive thickening later gives rise to all the placodes including the hypophyseal placode (Knouff, 1935; Fig. 3A). Knouff felt that the trigeminal placode, despite bordering the primitive placodal thickening, was not a part of it (Knouff, 1935). Also, although he described the lens placode as arising from ectoderm that once formed part of the primitive placodal thickening, he did not feel it was derived from the thickening as this ectoderm was thin in the intervening stages before the lens placode could be identified (Knouff, 1935). The primitive placodal thickening is broad in the head but abruptly narrows at the head/trunk interface, although it can still be traced back into the trunk until it disappears at the 3–4 somite stage (Knouff, 1935).

A preplacodal thickening incorporating all future placodes has not been observed in the frog *Xenopus* or the urodele *Ambystoma* (axolotl), although as described above, several placodes are initially part of discrete multiplacodal areas (Schlosser and Northcutt, 2000; Landacre, 1926; Northcutt and Brändle, 1995). The homeobox gene *Six1* is expressed in *Xenopus* at the late neural fold to 4-somite stage (stage 17/18; Nieuwkoop and Faber, 1967) in a continuous band surrounding the anterior neural plate (Pandur and Moody, 2000). The band of expression is separated from the prospective olfactory placode region in the anterior neural ridge, which also expresses *Six1* (Pandur and Moody, 2000). This expression domain is very similar to the primitive placodal thickening in *Rana*, where a common olfactory and hypophyseal placodal thickening is already separate from the remainder of the primitive placodal thickening at the 1–2 somite stage (Knouff, 1935). The lens placode does not express *Six1* (Pandur and Moody, 2000).

Also in *Xenopus*, *Sox2* and *Sox3* are expressed from gastrula stages in dorsal ectoderm including prospective neural plate and a subset of the neural crest; they are maintained at all stages in at least some placodes (Mizuseki *et al*., 1998; Penzel *et al*., 1997).

Chick. At the neural plate stage, placodes fate-map to discrete regions around the anterior neural plate border outside the region fated to form neural crest, except for the olfactory placode, which lies adjacent to the prospective forebrain at the very anterior (Rudnick, 1944; Fig. 3A). Bone morphogenetic protein-4 (BMP4) expression in neural plate and neurula stages demarcates the neural plate border region, while *Sox2* marks the neural plate (Streit and Stern, 1999a; Rex *et al*., 1997). The BMP4 expression domain in early neurula stage embryos (stage 5; Hamburger and Hamilton, 1951) corresponds well to the fate map of placodes and neural crest in definitive primitive streak embryos (Rudnick, 1944). *Sox3* is initially expressed in the entire embryonic ectoderm and is maintained in the central nervous system and in all placodes except the trigeminal and possibly the hypophyseal (Rex *et al*., 1997; Dr. Paul Scotting, personal communication). *Six4* is expressed in neurula stage embryos (stage 6) in a horseshoe-shaped crescent surrounding the developing anterior neural plate, corresponding precisely to the placodal fate map (Esteve and Bovolenta, 1999). *Six4* is subsequently expressed in each placode as it develops, although expression is not apparent in the trigeminal placodes (despite later expression in the trigeminal ganglia) and was not described in the hypophyseal placode (Esteve and Bovolenta, 1999).

Mammals. In the rat, a carbohydrate differentiation antigen (CDA-3C2) is expressed at the open neural plate stage (E9) in the ectoderm lateral to the neural plate border that gives rise to all the placodes (Prouty and Levitt, 1993). Weaker staining is seen in the neural folds and no staining in the neural plate itself (Prouty and Levitt, 1993). By E11, only the otic vesicles and olfactory placodes express CDA-3C2, together with selected regions of the CNS; no expression was ever seen in neural crest derivatives (Prouty and Levitt, 1993).

In the mouse, placode and neural crest cells arise from lateral ectoderm in the more proximal areas of the primitive streak (gastrula) embryo, at the border of the prospective neural plate (Tam, 1989). At neurula stages, a lateral area of thick ectoderm is continuous with the cranial neural folds, and nonplacodal ectoderm subsequently thins to leave the placodes in relief, which thicken further (van Oostrom and Verwoerd, 1972; Verwoerd and van Oostrom, 1979). The winged helix transcription factor *BF1* (*Foxg1*; Kaestner *et al*., 2000) is expressed in head surface ectoderm from the 6-somite stage and is subsequently maintained in all placodes and placode-derived cells, including the lens and hypophyseal placodes (Hatini *et al*., 1999). Using a *lac*Z-*BF1* transgene, expression was detected in the anterior ectodermal ridge at the 1-somite stage; expression subsequently spreads to the surface ectoderm, reaching the level of the posterior hindbrain by the 8-somite stage (E8.5). By E9.5, ectodermal expression is restricted to the placodes and placode-derived cells (Hatini *et al*., 1999). The initial uniform expression in head ectoderm may reflect the initial thickened state of the head ectoderm (van Oostrom and Verwoerd, 1972; Verwoerd and van Oostrom, 1979).

In human embryos, a continuous horseshoe-shaped area of thickened ectoderm is seen at the 4–20 somite stage that includes all the placodes (O'Rahilly and Müller, 1985). At the 25-somite stage, this apparently forms the rostral and pharyngeal part of an "ectodermal ring" that extends all the way around the embryo and includes the apical ectodermal ridges of the limb buds (O'Rahilly and Müller, 1985).

Establishment of the Neural Plate Border

Given the existence of a preplacodal ectoderm field at the border of the neural plate, as attested by morphological and molecular markers, the first step in placode induction would seem to be the formation of this field. Therefore, the establishment of the neural plate border region, which also gives rise to the neural crest, is of prime importance in considering placode induction. It is important to remember, however, that placodes are only generated from the cranial neural plate border.

Neural determination in vertebrate embryos begins during or even before gastrula stages, as shown by explant and grafting experiments and marker expression in amphibian, chick, mouse, and zebrafish (Jones and Woodland, 1989; Sive *et al*., 1989; Roberts *et al*., 1991; Storey *et al*., 1992; Ang *et al*., 1994; Neave *et al*., 1995; Grinblat *et al*., 1998; Kuo *et al*., 1998; Darnell *et al*., 1999; Wilson *et al*., 2000; Streit *et al*., 2000). Current models of neural plate induction are reviewed in Lee and Jessell (1999), Streit and Stern (1999b), and Weinstein and Hemmati-Brivanlou (1999). Primarily based on studies in *Xenopus*, the prevailing model suggests that the organiser induces neural tissue through its secretion of various antagonists of BMP signalling such as noggin, chordin, and follistatin. However, experiments in the chick suggest that BMP inhibition may act in association with, or downstream of, other organiser-derived factors that may include fibroblast growth factors (FGFs; Streit *et al*., 1998, 2000; Streit and Stern, 1999b; Wilson *et al*., 2000).

The balance between BMPs and their antagonists is important in establishing and/or maintaining the prospective neural plate border, as shown by experiments in chick, zebrafish, and *Xenopus* (Streit and Stern, 1999a; Neave *et al*., 1997; Nguyen *et al*., 1998; LaBonne and Bronner-Fraser, 1998b). The neural plate border in *Xenopus* may be set even before gastrulation begins, through planar signalling from the forming dorsal lip of the blastopore or through signals from the endoderm before the dorsal lip is a signalling centre (London *et al*., 1988; Savage and Phillips, 1989; Zhang and Jacobson, 1993). It is possible that the entire neural plate transiently acquires a neural border identity in response to planar signals from the organiser that is subsequently restricted to the lateral edges of the neural plate by midline signals from the involuting mesoderm (see La-

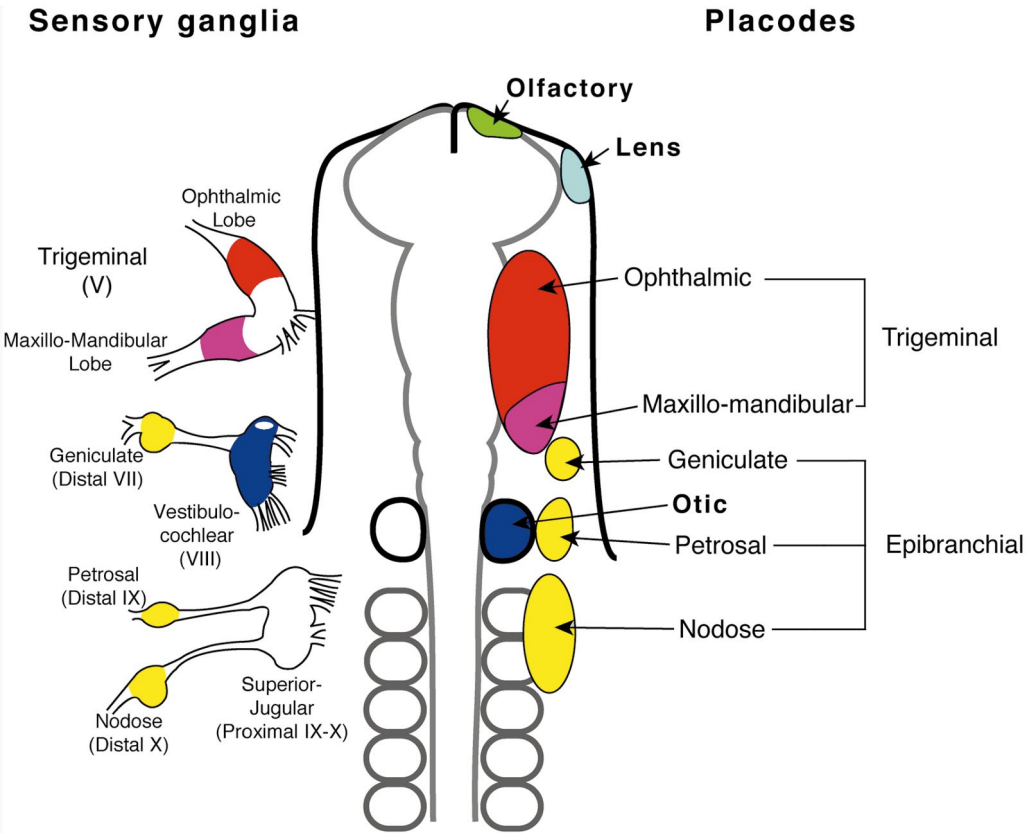

FIG. 4. In the chick embryo, trigeminal and epibranchial placodes (8-somite stage fate map shown on right side) contribute sensory neurons to the distal lobes of the trigeminal ganglion (V) and the distal ganglia of cranial nerves VII, IX, and X (shown on left side). The neural crest contributes sensory neurons to the proximal trigeminal ganglion (V) and the proximal ganglia of cranial nerves VII, IX, and X. fb, forebrain; hb, hindbrain; mb, midbrain. After Webb and Noden (1993).

Bonne and Bronner-Fraser, 1999; Streit and Stern, 1999b). This may be reflected in the expression throughout the early neural plate of genes whose later expression is restricted to the neural plate border, such as *Pax3* and *Zic* family members (see LaBonne and Bronner-Fraser, 1999). Interestingly, given the anterior restriction of placode-forming ectoderm, mechanisms of neural plate border formation may differ in cranial and more caudal regions of the neural plate (Streit and Stern, 1999a).

The regionalisation of preplacodal ectoderm may begin during gastrula stages. It has been suggested from fate-mapping data that olfactory placode territory may be established even in pregastrula stages in the chick, as it seems to move differently from other surrounding cell fates (Hatada and Stern, 1994). This may reflect the early specification of the forebrain region: in zebrafish, forebrain markers are specified by planar signals from the organiser just before or at the onset of gastrulation (Grinblat *et al.*, 1998), although they are not committed at this stage (Woo and Fraser, 1997). *Pax8* is expressed in prospective otic placode ectoderm from late gastrula stages in the zebrafish (Pfeffer *et al.*, 1998). Most known markers of particular placodes, however, begin to be expressed from neural plate or neurula stages. This is discussed in detail in the sections below on the induction of individual placodes.

Neural Crest Induction

Both placodes and neural crest cells are induced at the neural plate border, in distinct but overlapping domains. Prospective placodal ectoderm forms lateral to the neural crest. At the most rostral levels, however, it abuts the neural plate directly and becomes incorporated into the outer folds of the anterior neural ridge, where it will give rise to the olfactory and hypophyseal placodes. The rostral forebrain, including the anterior neural ridge, does not form neural crest cells, except possibly for a few in the mouse (Adelmann, 1925; Knouff, 1935; Jacobson, 1959; Chibon, 1967; Nichols, 1981; Couly and Le Douarin, 1985; Sadaghiani and Thiébaud, 1987; Osumi-Yamashita *et al.*, 1994). As placodes form from the most rostral regions of the neural plate border and are absent at trunk levels, the formation of

the placode-forming domain must be different at least in some respects from the formation of the neural crest domain. Nonetheless, it seems likely that many of the signals involved in neural crest induction will also be involved in placode induction, given their joint location at the anterior neural plate border and their overlapping sets of derivatives (see Baker and Bronner-Fraser, 1997b).

Neural crest induction has been extensively reviewed in recent years (Baker and Bronner-Fraser, 1997a; LaBonne and Bronner-Fraser, 1998a, 1999; García-Castro and Bronner-Fraser, 1999; Lee and Jessell, 1999) and is briefly summarised here. Perhaps the first step is the establishment of a neural crest-forming domain within the neural plate border by signals from nonaxial mesoderm (Muhr *et al*., 1997; Bang *et al*., 1997, 1999). Nonaxial mesoderm is both necessary and sufficient to induce neural crest cells and markers in amphibian and chick embryos (Raven and Kloos, 1945; Selleck and Bronner-Fraser, 1995; Bang *et al*., 1997, 1999; Muhr *et al*., 1997; Bonstein *et al*., 1998; Marchant *et al*., 1998). However, interactions between neural plate and nonneural ectoderm are sufficient to generate neural crest cells *in vitro* (note that placode derivatives have not been looked for in these experiments; Dickinson *et al*., 1995; Selleck and Bronner-Fraser, 1995; Mancilla and Mayor, 1996). This was also proposed from the results of *in vivo* grafting experiments (Rollhäuser-ter Horst, 1979, 1980; Moury and Jacobson, 1990; Dickinson *et al*., 1995; Selleck and Bronner-Fraser, 1995; Mancilla and Mayor, 1996), although mesoderm involvement cannot be ruled out in any of these *in vivo* experiments. Hence, after or in addition to the establishment of the neural crest-forming domain within the neural plate border, signals from the nonneural ectoderm, such as BMPs (Liem *et al*., 1995; Muhr *et al*., 1997), may dorsalise this domain to induce neural crest cell fate. Wnt family members can induce neural crest cells from neuralised ectoderm (Saint-Jeannet *et al*., 1997; Chang and Hemmati-Brivanlou, 1998; LaBonne and Bronner-Fraser, 1998b) and Wnt signalling is required for neural crest cell induction *in vivo* (Saint-Jeannet *et al*., 1997; LaBonne and Bronner-Fraser, 1998b). FGFs are also able to induce neural crest cells from neuralised ectoderm (Kengaku and Okamoto, 1993; Mayor *et al*., 1997; LaBonne and Bronner-Fraser, 1998b), although their action may be Wnt-dependent (LaBonne and Bronner-Fraser, 1998b). In summary, neural crest induction is a multistep process, involving signals from the nonaxial mesoderm and nonneural ectoderm, but there is much left to learn before a fully coherent model can be assembled. It will be interesting to discover whether any relationship can be uncovered between neural crest induction and placode induction.

Cardiac Mesoderm Induction: A Model for Induction of the Preplacodal Domain?

The restriction of placodal fate to anterior regions of the neural plate border is not due to restrictions in competence, although changing patterns of competence, as will be seen, can be important in restricting the induction of particular placodes to particular regions. At gastrula stages, all regions of the ectoderm are competent to form placodes, and this competence is maintained in trunk ectoderm even into tailbud stages for some placodes. Instead, the cranial restriction of general placodal fate must be explained in terms of local induction. Prospective cardiac mesoderm is present in a similar anterior horseshoe-shaped domain, at least in chick and zebrafish (for reviews of cardiac mesoderm development, see Jacobson and Sater, 1988; Fishman and Chien, 1997; Mohun and Sparrow, 1997). It is possible that the mechanism of cardiac mesoderm induction might therefore shed some light on, or even be related to, the induction of the preplacodal domain.

In the chick, cardiac mesoderm is induced where unidentified signals from anterior endoderm overlap with endodermal BMP2 and ectodermal BMP4 signals in the periphery (Schultheiss *et al*., 1995, 1997; Ladd *et al*., 1998). It is possible that the anterior restriction of the preplacodal domain is also due to induction only where signals from anterior endoderm overlap with BMP signals at the border of the neural plate. There is as yet no direct evidence for this model, although both anterior endoderm and cardiac mesoderm have some placode-inducing ability, as will be seen in later sections.

The Pax/Six/Eya/Dach Genetic Regulatory Network and Specific Placode Induction

All placodes express one or more members of the *Pax* family of paired class homeobox genes at a relatively early stage in their development (see later sections). *Pax8* is the earliest known marker of the otic placode; *Pax6* is a well-known and essential marker of the lens placode and olfactory placodes; *Pax3* is the earliest known marker for the ophthalmic trigeminal placode, while *Pax2* is an early marker for the otic and epibranchial placodes.

It has become clear in recent years that Pax family transcription factors, which comprise one of the most ancient homeodomain transcription factor families (reviewed in Galliot *et al*., 1999) may function in an evolutionarily conserved regulatory network. In *Drosophila*, eye development is regulated by the Pax6 homologues Eyeless (Ey) and Twin of eyeless (Toy), which function synergistically in a complex regulatory network with another homeodomain transcription factor, Sine oculis (So), and two nuclear proteins, Eyes absent (Eya) and Dachshund (Dac; reviewed in Wawersik and Maas, 2000). Eya and Dac contain transcriptional activation domains but lack DNA-binding domains, and Eya can physically interact with both So and Dac (Chen *et al*., 1997; Pignoni *et al*., 1997). This suggests a model in which So and Eya form a transcription factor complex that binds to specific DNA sequences that function throughout retinal development. Dac is only required for a subset of steps in retinal development, so Dac may bind to Eya as a coactivator for efficient activation of target genes specific to these steps (Chen *et al*., 1997;

Pignoni *et al*., 1997). It is important to note that So and Eya are not obligate partners, being expressed in different tissues in *Drosophila* and having different mutant phenotypes (Pignoni *et al*., 1997).

Vertebrate *Six*, *Eya*, and *Dach* genes are homologues of *Drosophila so*, *eya*, and *dac* (reviewed in Wawersik and Maas, 2000). The conservation of the genetic regulatory network involving *Pax*, *Six*, *Eya*, and *Dach* genes is suggested by the synergistic regulation of myogenesis in the chick by *Pax3*, *Six1*, *Eya2*, and *Dach2* (Heanue *et al*., 1999). As in *Drosophila*, vertebrate Eya physically interacts with Six and Dach proteins (Heanue *et al*., 1999; Ohto *et al*., 1999). Overlapping expression of different combinations of *Pax*, *Six*, *Eya*, and *Dach* family members is seen in all vertebrate placodes at various stages in their development, as will be seen below in the sections on individual placodes, suggesting the existence of specific *Pax*/*Six*/*Eya*/*Dach* genetic networks in each placode. Which network is activated may depend not only on the particular signals present but also on the state of competence of the ectoderm. In the medaka fish, overexpression at the two-cell stage of the general placodal marker *Sox3* leads to ectopic *Pax6* expression and lens formation in ventrolateral head ectoderm, to ectopic *Pax6* and *Eya1* expression (and possibly olfactory placode formation) in anterior head ectoderm, and to ectopic *Pax2* and *Eya1* expression and otic vesicle formation in trunk ectoderm (Köster *et al*., 2000).

The induction of ectopic eyes by *ey*/*Pax6* in *Drosophila* only occurs where the fly BMP homologue *decapentaplegic* (*dpp*) is expressed (Chen *et al*., 1999). Although *dpp* does not itself induce *ey* expression, *dpp* and *ey* cooperatively regulate *so*, *eya*, and *dac* expression, while *dpp*, *eya*, and *so* also cooperatively regulate *dac* expression (Chen *et al*., 1999; Curtiss and Mlodzik, 2000). There is therefore an essential synergistic and reiterative interaction between *dpp* and the genetic network controlling eye development that may restrict high levels of expression of this network to the source of *dpp* expression through a complex series of positive feedback loops (Chen *et al*., 1999). This model also suggests how a general signalling pathway can be integrated locally with a group of tissue-specific transcription factors (Chen *et al*., 1999; Curtiss and Mlodzik, 2000). In vertebrates, BMPs are essential for, or have been implicated in, the development of the hypophyseal, lens, and epibranchial placodes, in addition to their early expression at the neural plate border where the preplacodal ectoderm domain is formed.

In myogenesis, Wnt family members are probably involved in inducing *Pax3* and presumably therefore the *Pax3*/*Six1*/*Eya2*/*Dach2* myogenic network (Heanue *et al*., 1999). In the *Drosophila* leg imaginal disc, *dac* expression is induced by the combined action of the Wnt homologue Wingless (Wg) and low levels of Dpp (Lecuit and Cohen, 1997). Many Wnt signalling pathway genes are expressed in placodes. Further, Wg and high levels of Dpp act jointly to induce *distalless* expression in the leg imaginal disc (Lecuit and Cohen, 1997), and vertebrate *distalless* (*dlx*) homologues are also expressed in different placodes.

Taken together, these results are consistent with a scenario in which Wnt signals are likely to be important for the induction of different *Pax/Six/Eya/Dach* networks, which then cooperatively interact with BMP signals to induce downstream genes. In placode induction, therefore, BMPs are likely to be involved at multiple steps, beginning with their initial involvement in the formation of the neural plate border. Different states of competence in different regions of ectoderm are also likely to be important in the induction of different networks.

LAYOUT AND DEFINITIONS

In the following sections, we take each placode in turn and examine the embryological and molecular evidence for the mechanisms underlying its induction. For each placode, we present (1) its function, development, and derivatives, including a table of early molecular markers in different species; (2) embryological evidence for the location of competent ectoderm, time of specification and/or commitment, and inducing tissues; and (3) molecules involved or implicated in its induction.

Competence for a given developmental fate is defined as ability to adopt that fate. Competence remains essentially uncharacterised at the molecular level, although it may relate to the distribution of receptors or downstream effector molecules. Spatiotemporal changes in competence may be important in refining an inductive response. Competent ectoderm can be used in assays to define the location, duration, and nature of inducing tissues.

Specification and commitment are defined experimentally. A tissue is *specified* to follow a particular developmental pathway if it does so when isolated from the embryo and cultured in a neutral medium, i.e., in the absence of other signals. Experiments in which tissue is cultured in the presence of serum or embryonic extract are not tests of specification, as multiple unknown signals are present. A tissue is *committed* to follow a particular developmental pathway if it does so regardless of its environment, i.e., in the presence of other signals. Commitment is usually tested by grafting the tissue elsewhere within the embryo. Culturing tissue in the presence of serum could also be interpreted in some cases as showing commitment. Ideally, multiple environments should be tested and the most challenging used as a read-out of commitment. Specification does not necessarily imply commitment, as a specified tissue may adopt other fates when placed in a different environment. The molecular basis of specification and commitment is still obscure. The timing of specification and commitment provides insight into the time of action of inducing signals. Specification indicates when the tissue is starting to respond, and commitment indicates when a given molecular response is complete.

As described above, we feel there is strong evidence to

support the existence of a general preplacodal domain at the cranial neural plate border. Nonetheless, as will be clear from the evidence presented below, individual placodes are induced from ectoderm within this domain by widely differing mechanisms. Given the very different fates of the different placodes and their separate evolutionary origins (Graham and Begbie, 2000; Shimeld and Holland, 2000), these differences are not unexpected.

THE ANTERIOR NEURAL RIDGE: OLFACTORY AND HYPOPHYSEAL PLACODES

The anterior neural ridge defines the rostral boundary of the neural plate (see Papalopulu, 1995). It acts as a local organising centre that regulates gene expression in the anterolateral neural plate (Shimamura and Rubenstein, 1997; Ermakova *et al.*, 1999; Houart *et al.*, 1998), at least partly through its production of FGF8 (Shimamura and Rubenstein, 1997). The anterior neural ridge gives rise to the olfactory and hypophyseal placodes as well as to some forebrain tissues such as the olfactory bulbs (Knouff, 1935; Carpenter, 1937; Jacobson, 1959; Chibon, 1967; van Oostrom and Verwoerd, 1972; Klein and Graziadei, 1983; Couly and Le Douarin, 1985, 1987; Eagleson *et al.*, 1986; Eagleson and Harris, 1990; Kawamura and Kikuyama, 1992; Osumi-Yamashita *et al.*, 1994; Houart *et al.*, 1998; Whitlock and Westerfield, 2000; reviewed in Papalopulu, 1995; Rubenstein *et al.*, 1998). Future olfactory placode and olfactory bulb tissues are contiguous, as are future adenohypophysis and hypothalamus tissues (Couly and Le Douarin, 1985, 1987; Eagleson *et al.*, 1986; Whitlock and Westerfield, 2000). It is possible that a common progenitor cell population in the anterior neural ridge may give rise both to placodes and to forebrain tissues (Papalopulu, 1995). However, recent fate-map data from the zebrafish at the 4–5 somite stage suggest that the olfactory placode and telencephalon develop from distinct cellular fields: single cells in the anterior neural plate at this time never give rise to progeny in both the olfactory organ and the telencephalon (Whitlock and Westerfield, 2000). These data also suggest that the olfactory placode develops by the convergence of a large field of cells within the lateral anterior neural plate (Whitlock and Westerfield, 2000). A close relationship between olfactory placode-derived cells and the central nervous system is suggested nonetheless by a report that dissociated olfactory epithelial cells, when transplanted into different regions of the developing rat brain, will differentiate into neuronal or glial cells with a central phenotype (Magrassi and Graziadei, 1996). Similarly, the rostral part of the hypophyseal primordium in the toad incorporates into the diencephalic floor during normal development (Kawamura and Kikuyama, 1992).

The close spatial relationship between the hypophyseal and olfactory placodes at early embryonic stages is also interesting in light of the essential link between olfaction and reproduction. Gonadotropin-releasing hormone (GnRH), which acts on the adenohypophysis, is primarily produced by olfactory placode-derived neurons in vertebrates, while hormones secreted from the adenohypophysis in response to GnRH coordinate most reproductive activities. If the olfactory placode primordium is ablated at neurula stages in the toad, it is regenerated by presumptive hypophyseal tissue (Kawamura and Kikuyama, 1996), although the reverse does not seem to occur, at least in the chick (El Amraoui and Dubois, 1993b).

Various transcription factors are useful early markers for the anterior neural ridge and its derivatives. In the mouse, the homeobox genes *Pax6* and *Six3*, together with *Dach1*, are initially expressed specifically in the anterior neural ridge and subsequently also in the adjacent neural plate (Grindley *et al.*, 1995; Oliver *et al.*, 1995a; Caubit *et al.*, 1999; Davis *et al.*, 1999). *Dlx5* is expressed in the anterior neural ridge and expression is later maintained in the olfactory placode and the forebrain (Yang *et al.*, 1998; Depew *et al.*, 1999). Members of the *Anf* family of paired-like homeobox genes (*Hesx1/Rpx*, *XANF*, *GANF*) are specifically expressed in the anterior neural folds, and later expressed exclusively in the adenohypophysis (Kazanskaya *et al.*, 1997).

Forebrain Induction

Given that the olfactory placodes and the adenohypophysis ultimately derive from the anterior neural ridge, it is likely that signals that induce forebrain are important for the induction of these two placodes. Conversely, abrogation of forebrain development is likely to result in loss of these placodes. There are several recent reviews on forebrain induction (Bouwmeester and Leyns, 1997; Beddington and Robertson, 1998, 1999; Brewster and Dahmane, 1999; Niehrs, 1999). Briefly, BMP- and Wnt-inhibiting signals from the organiser and organiser-derived anterior mesendoderm, and also from the anterior visceral endoderm in mice and the dorsal yolk syncytial layer in fish, are essential for forebrain induction and patterning. This is reflected in a requirement for these tissues in olfactory and hypophyseal placode induction, as will be seen in the following sections. For example, *Hesx1* function in the anterior visceral endoderm is necessary for normal development of the olfactory placodes and pituitary gland, as well as the forebrain; its mutation in humans causes septo-optic dysplasia (Dattani *et al.*, 1998).

OLFACTORY PLACODES

Derivatives and Function

The olfactory placodes give rise to the ciliated sensory receptor cells of the olfactory (odorant-sensing) and vomeronasal (pheromone-sensing) epithelia (reviewed in Buck, 2000), whose axons project into the brain to form the olfactory, vomeronasal, and terminal nerves (Demski,

1993). The placodes also form nonneuronal supporting cells and glandular cells in these epithelia (Farbman, 1994). In addition, they form two cell types that delaminate and migrate into the brain along the olfactory and terminal nerves. First, they give rise to the glial cells that line the olfactory and vomeronasal nerves and the olfactory nerve layer of the olfactory bulb (Couly and Le Douarin, 1985; Marin-Padilla and Amieva B, 1989; Chuah and Au, 1991; Norgren *et al*., 1992; Ramón-Cueto and Avila, 1998; Ramón-Cueto and Valverde, 1995). The olfactory placode is the only placode known to produce glia: all other peripheral glial cells are derived from the neural crest. Second, the olfactory placodes give rise to neurons secreting gonadotropin-releasing hormone (Schwanzel-Fukuda and Pfaff, 1989; Wray *et al*., 1989; Murakami *et al*., 1992; El Amraoui and Dubois, 1993a; Yamamoto *et al*., 1996; Dellovade *et al*., 1998; reviewed in Muske, 1993; Tarozzo *et al*., 1995; Daikoku, 1999). The olfactory epithelium continuously generates neurons throughout adult life (Farbman, 1994).

GnRH neurons. Olfactory placode-derived GnRH neurons migrate along the terminal and vomeronasal nerves into the forebrain and diencephalon. Here, they form the terminal nerve–septo-preoptic GnRH system, which is the main regulator of gonadotropin release in most vertebrates (Muske, 1993). In the hypothalamus, GnRH neurons mostly terminate on the vasculature or cerebrospinal fluid and thus are involved in endocrine aspects of reproduction. GnRH (reviewed in King and Millar, 1991) usually reaches the hypophysis (pituitary gland) via the circulation; here, it stimulates cells of the adenohypophysis to release gonadotropins (luteinising hormone and follicle-stimulating hormone), which are essential for reproduction. These hormones control reproductive periods, ovarian cycles, sperm production, and many aspects of reproductive behaviour including sexual and parental behaviour and territoriality. Outside the hypothalamus, GnRH neurons project to neurons in various different brain areas, including the olfactory bulbs, most of which have been implicated in some aspect of reproduction (Muske, 1993). GnRH can act directly on neuronal targets in the brain, as a neurotransmitter or neuromodulator. It also acts as a neuromodulator on olfactory receptor cells, possibly increasing their sensitivity to odorants during the breeding season (Eisthen *et al*., 2000).

Kallmann's syndrome. The importance of the link between the olfactory and reproductive systems is highlighted in humans by Kallmann's syndrome, which is characterised by anosmia, due to the lack of olfactory bulbs and tracts, and hypogonadism, due to GnRH deficiency (reviewed in Rugarli, 1999). In Kallmann's syndrome, olfactory neurons extend towards but do not reach the brain, and GnRH neurons form but fail to migrate into the brain (Schwanzel-Fukuda *et al*., 1989; Hardelin *et al*., 1999). The X-linked form of this syndrome is due to mutations in the *KAL-1* gene, which encodes an extracellular matrix glycoprotein named anosmin-1 (Franco *et al*., 1991; Legouis *et al*., 1991; Soussi-Yanicostas *et al*., 1996; Hardelin *et al*., 1999). Anosmin-1 is expressed in the olfactory bulbs and is suggested to act as a guidance cue for olfactory neurons (Rugarli *et al*., 1993; Legouis *et al*., 1994; Lutz *et al*., 1994; Hardelin *et al*., 1999). The GnRH neuron migration failure may be indirect, as these neurons migrate along olfactory nerves. Indeed, GnRH neurons will migrate along ectopic olfactory nerves growing into the metencephalon after transplantation of the olfactory placode to the trigeminal placode (Gao *et al*., 2000).

Requirement for telencephalic development. The olfactory placode is also important for the development of the olfactory bulbs and indeed the telencephalon as a whole (reviewed in Graziadei and Monti-Graziadei, 1992; Dryer and Graziadei, 1994). Olfactory neurons are the first peripheral input to reach the brain during development (see references in Graziadei and Monti-Graziadei, 1992; Gong and Shipley, 1995). Ablation of the olfactory placodes in frog embryos results in the failure of development of the telencephalon, even when the surgery is performed before the olfactory nerves have reached the brain (see Graziadei and Monti-Graziadei, 1992). In *Small-eye* mice, which lack olfactory placodes owing to mutation in the *Pax6* gene, the olfactory bulbs are also missing (Schmahl *et al*., 1993; Dellovade *et al*., 1998). If the olfactory placode is grafted heterotopically, olfactory neurons will establish ectopic connections with specific different regions of the central nervous system and are associated with marked hyperplasia of the contacted region (Burr, 1924a,b; May, 1927; Stout and Graziadei, 1980; Magrassi and Graziadei, 1985). In the rat, the arrival of pioneer olfactory axons in the telencephalon correlates with a slowing in the cell cycle and an increased rate of cell differentiation in the olfactory bulb primordium that leads to the morphogenesis of the bulb (Gong and Shipley, 1995). In the zebrafish, pioneer olfactory neurons, originating from a more anterior part of the neural plate than sensory olfactory neurons, establish initial contact with the *emx1*-expressing region of the telencephalon and subsequently die (Whitlock and Westerfield, 1998). These pioneer neurons are necessary to target incoming sensory axons to the developing olfactory bulb (Whitlock and Westerfield, 1998).

Later in development, the olfactory epithelium is also required for induction of the neural crest-derived cartilaginous nasal capsule (Schmalhausen, 1939; Corsin, 1971).

Early Events and Markers

A summary of early events and molecular marker expression in the olfactory placodes of different vertebrates is given in Table 1.

Competence

Experiments on amphibian embryos have shown that competence to form the olfactory placode is initially widespread but is lost in nonhead ectoderm by neurula stages. In *Rana*, middle gastrula-stage presumptive belly epidermis is

competent to form nose when transplanted to the head region of gastrula or neural plate stage hosts, or in response to induction by the archenteron roof (Zwilling, 1940). Gill or flank ectoderm from older neurulae or tailbud embryos is not competent, however (Zwilling, 1940). In *Triturus* (newt), late gastrula stage presumptive epidermis can be induced to form nose (Kawakami, 1950, 1952). In *Ambystoma* (axolotl), anterior flank ectoderm from very early neurula is competent to form nose when grafted to the prospective nose region, while the same ectoderm from neurula or tailbud stage embryos is not competent (Haggis, 1956). In *Taricha* (newt), ventral epidermis from early neurula stage embryos is competent to form nose when grafted to the placodal region of a host of the same stage (Jacobson, 1963a).

In the chick, the olfactory placode can be induced from surrounding head ectoderm after ablation of the forebrain and presumptive olfactory placode as late as the 15-somite stage (Waddington and Cohen, 1936). Hence, competence is retained in rostral head ectoderm until relatively late stages.

Specification/Commitment

In amphibian embryos, grafting of prospective olfactory placode ectoderm to different embryonic regions shows commitment to an olfactory fate from relatively early stages. In grafts to the flank of urodele host embryos, *Rana* olfactory ectoderm begins to be committed from neural plate stages (Zwilling, 1940). In grafts to different regions of the head, *Ambystoma* olfactory ectoderm showed commitment from the early neurula stage (Carpenter, 1937; Reyer, 1962). In explant experiments, however, *Ambystoma* olfactory ectoderm was not found to be specified until the 4-somite stage (stage 21; Bordzilovskaya *et al*., 1989; Haggis, 1956). Likewise, in grafts to the anterior flank, commitment was only seen from the 4-somite stage (Haggis, 1956). These differences are almost certainly due to a more permissive environment for olfactory development in the head than is found in the trunk or in culture.

Prospective olfactory placode ectoderm from early neurula stage *Taricha* embryos is not yet specified to form nose when isolated in culture (Jacobson, 1963a). However, some commitment is shown at late neurula stages, since reversing the anteroposterior polarity of presumptive placodal ectoderm at late neurula stages leads to the formation of noses and ears both rostrally and caudally (Jacobson, 1963c). These results suggest the existence of both committed and labile cell populations within the ectoderm at the late neurula stage (Jacobson, 1963c).

In contrast, chick olfactory placode ectoderm seems to be committed only at late stages. Grafts of isolated olfactory placode ectoderm to the chorioallantoic membrane only form olfactory epithelium after the 24-somite stage, when the placode is already thickened *in vivo* (Street, 1937). These large species-specific differences could be explained by the presence of permissive factors in the amphibian embryonic graft sites that are not present at the chorioallantoic membrane in the chick, highlighting the operational nature of the definition of commitment and the need for further investigation. It is also essential to examine the specification and commitment of olfactory placode ectoderm towards the expression of specific molecules, rather than assaying the formation of a complex olfactory epithelium.

Inducing Tissues

Anterior mesendoderm. The early commitment of *Ambystoma* olfactory placode ectoderm led Reyer to suggest that the endoderm or prechordal mesoderm of the rostrodorsal archenteron wall is the source of the induction (Reyer, 1962). In *Triturus*, early gastrula stage prechordal mesoderm can induce nose from late gastrula stage presumptive epidermis, as can late gastrula stage prechordal mesoderm and prospective notochord (Kawakami, 1950, 1952). In *Rana*, archenteron roof implanted into the blastocoel of gastrula (yolk-plug stage) embryos induces a small olfactory organ, with no associated brain tissue, in host belly ectoderm (Zwilling, 1940). Other explant and grafting experiments also suggested that the olfactory organ is induced by the archenteron roof (Emerson, 1945). After endoderm ablation in neurula stage embryos, nose formation is reduced more than that of lens or ear, suggesting some requirement for endoderm (Jacobson, 1963b). Reversing the anteroposterior polarity of the endoderm has little effect, suggesting that the inducing activity is not regionalised (Jacobson, 1963b). Reversing the anteroposterior polarity of the neural plate leads to relatively normal positioning of noses, lenses, and ears (Jacobson, 1963c), highlighting the importance of the underlying tissues. Yntema (1955) and Denis (1959) (in the newt *Pleurodeles*) thought that head mesoderm and forebrain/neural crest act successively to induce the olfactory organ.

Anterior neural plate. Unspecified prospective olfactory placode ectoderm from early neurula stage *Taricha* embryos forms nose when cocultured with either anterior endoderm or anterior neural plate and fold (Jacobson, 1963a). However, differentiation is most complete when both neural and nonneural inducers are present in the explant (Jacobson, 1963a). Jacobson concluded that the nose is first induced by underlying endoderm in late gastrula and neurula stages, and subsequently by the forebrain (Jacobson, 1963c). The presence of forebrain was found to be important to maintain olfactory placode development in flank grafts or explants of prospective olfactory ectoderm from neurula-stage *Ambystoma* embryos (Haggis, 1956). Likewise, in the chick, presumptive olfactory placode ectoderm from donors younger than the 24-somite stage forms olfactory epithelium in chorioallantoic grafts only if forebrain tissue is included in the graft (Street, 1937). Nonetheless, olfactory placodes develop if the forebrain (and presumptive placode ectoderm) is ablated as late as the 15-somite stage (Waddington and Cohen, 1936; Orts Llorca and Ferrol, 1961),

TABLE 1
Molecular Markers and Early Events in Olfactory Placode Development

		Zebrafish		
8 h	Late gastrula	*Zcoe2; dlx3; dlx7*	Transcription factors	Bally-Cuif *et al.*, 1998; Akimenko *et al.*, 1994; Ellies *et al.*, 1997
10 h	End of gastrulation	*eya1*	Transcription cofactor	Sahly *et al.*, 1999
10 h	End of gastrulation	*six4.1*	Transcription factor	Kobayashi *et al.*, 2000
14 h	10 somites	Placode cells aggregate		Hansen and Zeiske, 1993
17 h	16 somites	Placodes thicken		Whitlock and Westerfield, 1998
18–19 h	18–20 somites	*runxa*	Transcription factor	Kataoka *et al.*, 2000
22 h	26 somites	L1.1	Neuronal adhesion molecule	Tongiorgi *et al.*, 1995
24 h	30 somites	Axons grow out		Whitlock and Westerfield, 1998
24 h	30 somites	*dlx4* (=mouse *Dlx5*)	Transcription factor	Akimenko *et al.*, 1994
32 h		Olfactory pits form		Whitlock and Westerfield, 1998
		Xenopus		
Stage 10.5	Early gastrula	*Sox2; Sox3*	Transcription factors	Mizuseki *et al.*, 1998; Penzel *et al.*, 1997
Stage 12	Late gastrula	*Pax6*	Transcription factor	Hirsch and Harris, 1997; Li *et al.*, 1997
Stage 14	Neural plate	*ngnr1*	Transcription factor	Schlosser and Northcutt, 2000
Stage 14–15	Neural plate	*Otx2*	Transcription factor	Zygar *et al.*, 1998
Stage 16	Midneural fold	X*Dll3* (=mouse *Dlx5*)	Transcription factor	Papalopulu and Kintner, 1993
Stage 17	Late neural fold	*XCoe2; Six1*	Transcription factors	Dubois *et al.*, 1998; Pandur and Moody, 2000
Stage 18	3–4 somites	*XIF3*	Cytoskeletal protein	Goldstone and Sharpe, 1998
Stage 23	12 somites	Placodes thicken		Nieuwkoop and Faber, 1967
Stage 23	12 somites	*eomesodermin; Otx1; Otx4*	Transcription factors	Ryan *et al.*, 1998; Kablar *et al.*, 1996
Stage 24	15 somites	*Emx2*	Transcription factor	Pannese *et al.*, 1998
Stage 25/26	16–17 somites	BMP2	Ligand	Clement *et al.*, 1995
Stage 30	25 somites	Olfactory axons grow out		Klein and Graziadei, 1983
Stage 40		Olfactory pits form		Klein and Graziadei, 1983
		Chick		
Stage 3	Gastrula	*Sox3*	Transcription factor	Rex *et al.*, 1997; Dr. Paul Scotting, personal communication
Stage 6	Neurula	*Six3; Six4*	Transcription factors	Bovolenta *et al.*, 1998; Esteve and Bovolenta, 1999
Stage 8	4 somites	*frizzled-7*	Wnt receptor	Stark *et al.*, 2000
Stage 9	7 somites	*Dlx3*	Transcription factor	Pera and Kessel, 1999
Stage 9	7 somites	*frizzled-2*	Wnt receptor	Stark *et al.*, 2000
Stage 13	19 somites	*Eya2*	Transcription cofactor	Mishima and Tomarev, 1998
Stage 14	23 somites	Placodes thicken		Street, 1937
Stage 15	24–27 somites	*Sfrp2*	Wnt antagonist	Ladher *et al.*, 2000
≤Stage 17	≤29–32 somites	*Dlx5*	Transcription factor	Pera *et al.*, 1999
≤Stage 18	≤30–36 somites	*Serrate1*	Notch ligand	Myat *et al.*, 1996
Stage 18–20	30–42 somites	Olfactory pits form		Romanoff, 1960
Stage 18–20	30–42 somites	*Deltex2*	Notch signal transduction pathway	Frolova and Beebe, 2000
Stage 18	30–36 somites	Hu	Early neuronal marker	Wakamatsu and Weston, 1997
Stage 19	37–40 somites	GnRH	Hormone	Yamamoto *et al.*, 1996; Mulrenin *et al.*, 1999
Stage 21–22	E3.5	GnRH neurons begin to migrate		Yamamoto *et al.*, 1996

TABLE 1—*Continued*

		Mouse		
E8	1–7 somites	*Pax6; Six3; Otx2; Pitx1 (Ptx1); Dlx5; BF1 (Foxg1)*	Transcription factors	Grindley *et al.*, 1995; Oliver *et al.*, 1995a; Mallamaci *et al.*, 1996; Lanctôt *et al.*, 1997; Yang *et al.*, 1998; Hatini *et al.*, 1999
E8.5	8–10 somites	*Sox2*	Transcription factor	Wood and Episkopou, 1999
E8.75	12 somites	*Fgf8*	Ligand	Heikinheimo *et al.*, 1994; Mahmood *et al.*, 1995a
E9	13–20 somites	*mtll*	Orphan nuclear receptor	Monaghan *et al.*, 1995
E9	13–20 somites	Activin receptor 1	Receptor	Yoshikawa *et al.*, 2000
E9.5	21–29 somites	Placodes thicken		Tamarin and Boyde, 1977; Kaufman, 1992
E9.5	21–29 somites	*Msx1; Sox3; HES6*	Transcription factors	Grindley *et al.*, 1995; Collignon *et al.*, 1996; Vasiliauskas and Stern, 2000
E9.5	21–29 somites	*Eya1; Eya2; Eya4*	Transcription cofactors	Xu *et al.*, 1997; Borsani *et al.*, 1999
E9.5	21–29 somites	*RALDH3*	Retinaldehyde dehydrogenase	Mic *et al.*, 2000
E9.5	21–29 somites	sFRP1	Wnt antagonist	Leimeister *et al.*, 1998
E9.5	21–29 somites	c-*kit*	Stem cell factor receptor	Orr-Urtreger *et al.*, 1990
E9.5	21–29 somites	*Delta1*	Notch ligand	Bettenhausen *et al.*, 1995
E9.5	24–25 somites	*Mash1; ngn1; NeuroD*	Transcription factors	Cau *et al.*, 1997
E9.5	27 somites	Class IIIβ-tubulin; *SCG10*	Neuronal differentiation markers	Cau *et al.*, 1997
E9.75		*Emx2*	Transcription factor	Simeone *et al.*, 1992; Mallamaci *et al.*, 1998
E10	30–34 somites	Olfactory pits form		Kaufman, 1992
≤E10.5	≤35–39 somites	*Six1; Six6; Hes1; Hes5*	Transcription factors	Oliver *et al.*, 1995b; Jean *et al.*, 1999; Cau *et al.*, 2000
E10.5	35–39 somites	*Hey1*	Transcription factor	Leimeister *et al.*, 1999
E10.5	35–39 somites	*Notch2*	Receptor	Williams *et al.*, 1995

suggesting that placode induction can occur in the absence of forebrain tissue. Further, the size and differentiation of the olfactory organs after forebrain ablation correlates with the amount of prechordal mesoderm present, suggesting that this tissue is responsible for the induction (Orts Llorca and Ferrol, 1961).

Summary. Taken together, these results suggest that anterior mesendoderm is the most important source of olfactory placode inducing signals, while the forebrain provides reinforcing signals. Clearly, it is important to examine the induction of olfactory placode-specific genes by these tissues in order to begin to understand the nature of this complex induction.

Molecules

Transcription factors. Otx2 is required in the visceral endoderm for induction of the anterior neural plate, and olfactory placodes are missing in *Otx2* mutant mouse embryos (Matsuo *et al*., 1995; Rhinn *et al*., 1998). In the mouse, the paired domain transcription factor *Pax6* (mouse *Small eye*, human *Aniridia*), which is first expressed in the anterior neural ridge, is required cell autonomously in head ectoderm for olfactory placode and olfactory bulb development (Hogan *et al*., 1986; Hill *et al*., 1991; Grindley *et al*., 1995; Quinn *et al*., 1996). It is required for *Eya1* and *Eya2* expression in the olfactory placodes just before they thicken (Xu *et al*., 1997). The homeobox transcription factor *Dlx5* (*Dll3/Dlx3* in *Xenopus*; Bendall and Abate-Shen, 2000), which is expressed in the anterior neural ridge and its derivatives, including the olfactory placodes (Papalopulu and Kintner, 1993; Yang *et al*., 1998), is important for olfactory epithelium development and for induction of the nasal capsule (Depew *et al*., 1999). In *Emx2*-null mice, the olfactory bulbs are disorganised and the olfactory epithelium fails to project to the olfactory bulb (Yoshida *et al*., 1997). The proneural bHLH transcription factor *Mash1* is required for the development of most olfactory receptor neurons (Guillemot *et al*., 1993).

Signalling molecules. There may be a role for retinoic acid in the development of both the olfactory placodes and the olfactory bulbs: the frontonasal mesenchyme between the olfactory placode and the ventrolateral forebrain is the source of retinoids that may activate retinoic acid receptors in both these structures (LaMantia *et al*., 1993). Further, in *Small eye* (*Pax6*) mutant mice, which lack olfactory placodes and olfactory bulbs (Hogan *et al*., 1986; Hill *et al*., 1991; Grindley *et al*., 1995; Quinn *et al*., 1996), the fronto-

nasal mesenchyme does not produce retinoic acid, and a subpopulation of frontonasal mesenchyme cells is missing (Anchan *et al.*, 1997; Enwright and Grainger, 2000). It is possible that this missing population of cells corresponds to a subpopulation of mesencephalic neural crest cells that normally migrates to the frontonasal region between the 1–7 somite stage (E8; time of labelling) and the 13–20 somite stage (E9; time of analysis), before the onset of retinoid signalling (Osumi-Yamashita *et al.*, 1994). A similar population of mesencephalic neural crest cells fails to reach the frontonasal region in *Pax6* mutant rats (Matsuo *et al.*, 1993), probably owing to a defect in the migration pathway to the frontonasal mass (Osumi-Yamashita *et al.*, 1997). Hence the retinoid source in the frontonasal mesenchyme may be neural crest-derived (Anchan *et al.*, 1997). However, RALDH3, a retinaldehyde dehydrogenase that catalyses retinoic acid formation, is expressed in the olfactory epithelium at the 21–29 somite stage (E9.5), raising the possibility that at least one retinoid source is the olfactory epithelium itself (Mic *et al.*, 2000). As *Pax6* is expressed initially in head ectoderm and subsequently in the olfactory placodes, but not in the neural crest, this may suggest the existence of a *Pax6*-dependent signal from head ectoderm that is necessary for neural crest cell migration to the frontonasal mass. A requirement for neural crest cells for olfactory placode development, however, seems unlikely given that nose can be induced from gastrula ectoderm by prechordal mesoderm and notochord in amphibian embryos (Kawakami, 1950, 1952).

Summary

The olfactory placodes are derived from ectoderm originally located in the anterior neural ridge. Competence is widespread in gastrula and neural plate stages, but is lost by neurula stages, at least in nonhead ectoderm, in amphibians. Specification and commitment in the axolotl are seen from the 4-somite stage, but chick olfactory ectoderm seems to be committed much later, once the placode has thickened *in vivo*. These large species-specific differences may reflect the differences in the graft sites used to test commitment. A two-step model of induction is suggested by the implication of both anterior mesendoderm and forebrain in olfactory placode induction. No signalling molecules have as yet been shown to be necessary or sufficient for olfactory placode induction. It is essential to begin examining olfactory placode induction using specific molecular markers so that this complex induction may be analysed in more detail.

HYPOPHYSEAL PLACODE

The induction of the hypophyseal placode, out of all the different placodes, is perhaps best characterised at the molecular level. A relatively brief account is given here, as this induction has been extensively reviewed (Treier and Rosenfeld, 1996; Watkins-Chow and Camper, 1998; Dasen and Rosenfeld, 1999; Jeremy and Michael, 1999; Kioussi *et al.*, 1999a; Sheng and Westphal, 1999).

Derivatives and Function

The *adenohypophysis* of the pituitary gland is derived from *Rathke's pouch*, a dorsal outpocketing from a placode (the hypophyseal placode) in midline oral ectoderm in the roof of the oral cavity, ventral to the posterior diencephalon. The *neurohypophysis* is derived from the *infundibulum*, a ventral evagination of the posterior diencephalon. The terms "anterior pituitary" and "posterior pituitary" are best avoided because they refer to anatomical divisions that do not hold across all vertebrates; adenohypophysis and neurohypophysis divide the pituitary according to its embryonic origins (Kardong, 1998). Figure 5 summarises the development of Rathke's pouch into the adenohypophysis.

Fate-map data over the last 15 years from chick, amphibian, and mouse embryos have revealed that both "epithelial" (adenohypophysis) and "neural" (neurohypophysis) parts of the pituitary gland are derived from adjacent anlagen in the anterior neural ridge and anterior neural plate, respectively (Eagleson *et al.*, 1986, 1995; Kawamura and Kikuyama, 1992; Couly and Le Douarin, 1985; Osumi-Yamashita *et al.*, 1994; reviewed in Kawamura and Kikuyama, 1998; Rubenstein *et al.*, 1998). In the chick, presumptive hypophyseal ectoderm probably lies in the rostral-most (outer) ectoderm of the ridge rather than in the neural plate proper (Rubenstein *et al.*, 1998). Presumptive hypophyseal tissue in the midanterior neural ridge is progressively internalised by the closure and overgrowth of the forebrain. It elongates along the body axis and makes contact caudally with the rostral foregut, then flattens along the diencephalic floor and makes contact with the forming infundibulum (Eagleson *et al.*, 1986; Kawamura and Kikuyama, 1992). Here, it is induced to form a placode and evaginates towards the ventral diencephalon to form Rathke's pouch (Fig. 5). After it detaches from the oral ectoderm to form the adenohypophysis, different endocrine cells are generated within it in a precise temporal and spatial order. Hormones secreted from the adenohypophysis include gonadotropins (follicle-stimulating hormone and luteinising hormone), prolactin, adrenocorticotropin, thyroid-stimulating hormone, melanophore-stimulating hormone, and growth hormone.

Part of the grafted anterior neural ridge tissue that gives rise to the adenohypophysis is attached to, and maybe even incorporates into, the foregut endoderm (Kawamura and Kikuyama, 1992). This may be important when considering the observation that in hagfish embryos, the adenohypophysis seems to arise from an outgrowth of the foregut endoderm (Gorbman, 1983; Gorbman and Tamarin, 1985). Similarly, the most rostral part of the grafted anterior neural ridge incorporates into the diencephalic floor, in the preoptic region of the hypothalamus (Kawamura and Kikuyama, 1992).

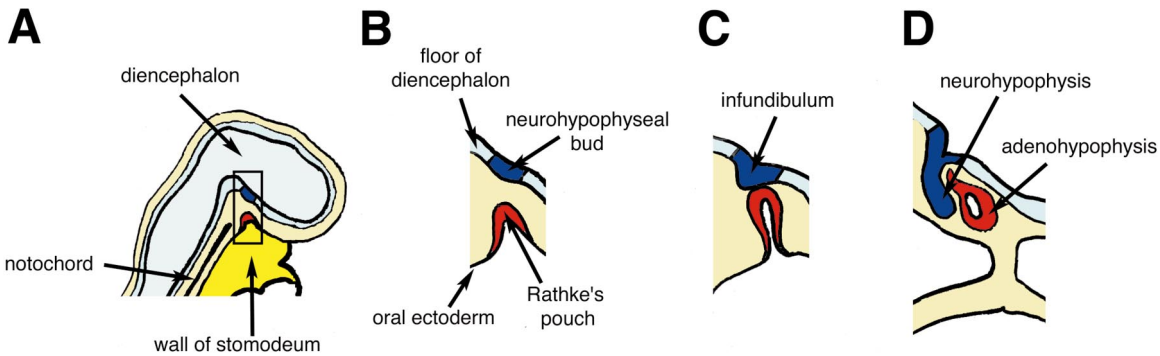

FIG. 5. Development of the pituitary gland. (A) Sagittal section of young embryo showing formation of Rathke's pouch and infundibulum. (B–D) Development of Rathke's pouch and the neurohypophysis. After Kardong (1998).

Teleosts, unlike more basal fish and most tetrapods, do not develop an equivalent structure to Rathke's pouch; rather, the hypophyseal anlage is a solid structure (Bond, 1996). In the zebrafish, Lim3 (Lhx3) protein expression in prospective hypophyseal placode cells is initially asymmetric (Glasgow *et al.*, 1997). At the 21-somite stage (19.5 h), a few Lim3-positive cells appear on the left side, adjacent to the anterior ventral diencephalon, followed by cells on the right side of the anterior ventral diencephalon (Glasgow *et al.*, 1997). By 28 h, these initially bilateral clusters have moved to the midline and fused to form a single pituitary cluster (Glasgow *et al.*, 1997).

Early Events and Markers

Most of the molecular information on the induction of the adenohypophysis comes from studies on mouse embryos. Some early markers and events in development of the hypophyseal placode in mouse and other vertebrates are shown in Table 2. The *Anf* (*a*nterior *n*eural *f*old) family of transcription factors (*Hesx1*/*Rpx*; *XANF*, *GANF*) are all expressed in the anterior neural ridge and then maintained in the adenohypophysis, making these excellent markers for the forming hypophyseal placode (Kazanskaya *et al.*, 1997).

Competence

At the 3-somite stage in the chick, presumptive hypophyseal placode ectoderm lies within the outer ectoderm of the midanterior neural ridge (Couly and Le Douarin, 1985; Rubenstein *et al.*, 1998). If this region is ablated at the 2–4 somite stage, the adenohypophysis fails to form (El Amraoui and Dubois, 1993b), suggesting that the adjacent tissue within the ridge is no longer competent to form adenohypophysis. In the 10–13 somite stage chick embryo, however, head ectoderm rostral to the otic vesicle can be induced to form hypophyseal cell types by ventral diencephalon in culture, if cephalic or somitic mesoderm from embryos older than the 5-somite stage is also present (Fedtsova and Barabanov, 1990; Gleiberman *et al.*, 1999). Otic placode ectoderm and trunk ectoderm are not competent to respond, while competence is retained in head ectoderm until the 21-somite stage (Fedtsova and Barabanov, 1990; Gleiberman *et al.*, 1999). These results show that competence is spatially restricted and changes with time.

Specification/Commitment

In urodeles, the adenohypophysis is not committed before the 17–18 somite stage (stage 30; Blount, 1932). Interestingly, chick hypophyseal tissue was recorded as showing some commitment in chorioallantoic grafts even from early neurula (primitive streak) stages (Rudnick, 1932), although this requires further investigation. When Rathke's pouch from avian embryos is placed in culture, it only differentiates if a source of mesenchyme is present, and even then only after the 25-somite stage (Le Douarin *et al.*, 1967; Ferrand, 1969a; Frémont and Ferrand, 1979). In the mouse, Rathke's pouch does not form endocrine cell lineages until the 35–39 somite stage (E10.5), when cultured in the presence of serum (Treier *et al.*, 1998).

Inducing Tissues

Signals from the diencephalon are essential for induction of the hypophyseal placode and its subsequent morphogenesis to form Rathke's pouch and its derivatives (Stein, 1929; Blount, 1930, 1932; Burch, 1938; Hillemann, 1943; Le Douarin *et al.*, 1967; Ferrand, 1969a,b, 1972; Daikoku *et al.*, 1982; Treier *et al.*, 1998). It is possible that the notochord may be involved in pouch morphogenesis (Gleiberman *et al.*, 1999). There is also some evidence that signals from foregut endoderm are required for development of the adenohypophysis (see Kawamura and Kikuyama, 1998). Signals from the adenohypophysis, conversely, are required for full development of the neurohypophysis (Smith, 1920; quoted in Hillemann, 1943; Kawamura and Kikuyama, 1998).

TABLE 2
Molecular Markers and Early Events in Hypophyseal Placode Development

		Zebrafish		
8.5 h	Late gastrula	*six3*	Transcription factor	Kobayashi *et al.*, 1998
10 h	End of gastrulation	*Danf1;six4.1*	Transcription factors	Kazanskaya *et al.*, 1997; Kobayashi *et al.*, 2000
10 h	End of gastrulation	*eya1*	Transcription cofactor	Sahly *et al.*, 1999
19.5 h	21 somites	Lim3 (Lhx3)	Transcription factor	Glasgow *et al.*, 1997
26–27 7		*nk2.2*	Transcription factor	Barth and Wilson, 1995; Karlstrom *et al.*, 1999
28 h		Single pituitary cluster formed		Glasgow *et al.*, 1997
≤32 h		*six6*	Transcription factor	Seo *et al.*, 1998
		Xenopus		
Stage 11	Early gastrula	*Xanf2*	Transcription factor	Mathers *et al.*, 1995
Stage 12	Late gastrula	*Pitx1; Pitx2*	Transcription factors	Hollemann and Pieler, 1999
Stage 14	Neural plate	*Lim3 (Lhx3)*	Transcription factor	Taira *et al.*, 1993
Stage 20	6–7 somites	Placode thickens		Nieuwkoop and Faber, 1967; Nyholm, 1977
Stage 25/26		BMP2	Ligand	Clement *et al.*, 1995
		Chick		
Stage 5	Early neurula	*Ganf1*	Transcription factor	Kazanskaya *et al.*, 1997
Stage 6	Neurula	*Six3*	Transcription factor	Bovolenta *et al.*, 1998
Stage 10	10 somites	*Pitx1 (Ptx1)*	Transcription factor	Lanctôt *et al.*, 1997
Stage 11	13 somites	Rathke's pouch evaginates		Venzke, 1942; Ferrand, 1969b
Stage 13	19 somites	*SFrp1*	Wnt antagonist	Esteve *et al.*, 2000
		Mouse		
E7.5	Neural plate	*Hesx1 (Rpx; Anf family)*	Transcription factor	Hermesz *et al.*, 1996
E8	1–7 somites	*Pax6; BF1 (Foxg1); Pitx1 (Ptx1)*	Transcription factors	Walther and Gruss, 1991; Hatini *et al.*, 1999; Lanctôt *et al.*, 1997
≤E8.5	≤8–12 somites	*Pitx2*	Transcription factor	Mucchielli *et al.*, 1996; Gage and Camper, 1997
E8.5	10 somites	*Islet1*	Transcription factor	Ericson *et al.*, 1998
E8.5	8–12 somites	Placode thickens		Kaufman, 1992; Dasen and Rosenfeld, 1999
E9	13–20 somites	Rathke's pouch evaginates		Kaufman, 1992; Dasen and Rosenfeld, 1999
E9	13–20 somites	Activin receptor I	Receptor	Yoshikawa *et al.*, 2000
E9.5	20 somites	*Lhx3 (Lim3)*	Transcription factor	Ericson *et al.*, 1998
E9.5	21–29 somites	*Six1; Six2*	Transcription factors	Oliver *et al.*, 1995b; Ohto *et al.*, 1998
E9.5	21–29 somites	*Eya1*	Transcription cofactor	Xu *et al.*, 1997
≤E10.5	≤35–39 somites	*Notch2*	Receptor	Williams *et al.*, 1995
E10.5	35–39 somites	*Six6*	Transcription factor	Jean *et al.*, 1999
E11.5		*Msx1*	Transcription factor	MacKenzie *et al.*, 1991

Molecules

Many of the signalling molecules and transcription factors involved in the development of the adenohypophysis in the mouse have been identified in recent years (for reviews, see Dasen and Rosenfeld, 1999; Jeremy and Michael, 1999; Kioussi *et al.*, 1999a; Sheng and Westphal, 1999; Watkins-Chow and Camper, 1998; Treier and Rosenfeld, 1996).

Transcription factors. The homeobox genes *Six3*, *Pax6*, and the *Anf* family are all expressed in the anterior neural ridge and later in the adenohypophysis (Oliver *et al.*, 1995a; Walther and Gruss, 1991; Kazanskaya *et al.*, 1997; Bovolenta *et al.*, 1998). *Six3* overexpression in zebrafish leads to enlargement of the rostral forebrain, suggesting a primary role in anterior neural plate formation (Kobayashi *et al.*,

1998). In *Hesx1* mutants, Rathke's pouch fails to form, again probably owing to *Hesx1* function in anterior neural plate formation (Dattani *et al*., 1998). In contrast, *Pax6* function is required later, during the differentiation of different cell types in the adenohypophysis (Kioussi *et al*., 1999b).

The paired-class transcription factors *Pitx1* (*Ptx1*, *P-OTX*) and *Pitx2* are expressed in the oral ectoderm before Rathke's pouch formation and are subsequently maintained in Rathke's pouch and the adenohypophysis (Lamonerie *et al*., 1996; Mucchielli *et al*., 1996; Szeto *et al*., 1996; Lanctôt *et al*., 1997). They may be redundantly required for Rathke's pouch formation, as neither has any effect when mutated alone, but each is required later, for target gene activation in different pituitary cell types (Gage *et al*., 1999; Lin *et al*., 1999; Szeto *et al*., 1999).

The *Nkx2.1* gene (*T/ebp*, *Ttf1*), which is expressed in the ventral diencephalon, is essential for pituitary development (Kimura *et al*., 1996).

Signalling molecules. BMP4, secreted by the presumptive infundibulum, is required for induction of the placode and initial evagination of the oral ectoderm to form the pouch rudiment (Takuma *et al*., 1998). Later, FGF8 from the posterior diencephalon induces expression of the LIM family member *Lhx3* in the developing pouch (Ericson *et al*., 1998). FGF8 expression in the diencephalon (Takuma *et al*., 1998) and *Lhx3*/*Lhx4* function in the placode (Sheng *et al*., 1997) are required for pouch morphogenesis. The IIIb isoform of the *Fgfr 2* gene is required for maintenance of Rathke's pouch and the formation of the adenohypophysis (De Moerlooze *et al*., 2000).

Sonic hedgehog is initially expressed throughout the oral ectoderm but is restricted from Rathke's pouch as soon as it begins to form; a ventral–dorsal BMP2 gradient later develops from the ectodermal boundary thus created that specifies different cell types within the adenohypophysis (Treier *et al*., 1998). In zebrafish, mutations in the Hedgehog target gene *Gli2* (*you-too*), whose expression overlaps with that of *sonic hedgehog* in the ventral diencephalon, disrupt formation of the adenohypophysis as well as the adjacent diencephalon, with concomitant down-regulation of *nk2.2*, *lim3*, and *Six3* in the adenohypophysis (Karlstrom *et al*., 1999). Treatment of *Xenopus* animal caps with Hedgehog family members induces the *Anf* family member *XANF-2*, while if mesoderm is present (in activin-treated caps), it also induces a late pituitary-specific marker, pro-opiomelanocortin (Ekker *et al*., 1995; Lai *et al*., 1995). *Wnt5a*, which is broadly expressed in the ventral diencephalon, can induce some endocrine cell types in Rathke's pouch in culture (Treier *et al*., 1998).

Current understanding of the coordination of endocrine cell type specification, which involves signals both from the ventral diencephalon and from the mesenchyme just ventral to the pituitary (Ericson *et al*., 1998), has been extensively reviewed (Dasen and Rosenfeld, 1999; Jeremy and Michael, 1999; Kioussi *et al*., 1999a; Sheng and Westphal, 1999; Watkins-Chow and Camper, 1998; Treier and Rosenfeld, 1996).

Summary

The hypophyseal placode is derived from ectoderm originally located in the anterior neural ridge. Competence is widespread within preotic head ectoderm until the 21-somite stage in the chick. Commitment occurs during late neural tube stages. The placode is induced by signals from the diencephalon, including BMP4 (essential for placode induction) and FGF8 (essential for pouch morphogenesis). Wnt5a and Sonic Hedgehog are involved in the formation of specific cell types within the adenohypophysis.

LENS PLACODES

Derivatives and Function

The lens placodes fate-map to the ectoderm just outside the anterior neural plate. They thicken after contact with the forming optic vesicles and give rise to the lenses of the paired eyes. Lens transparency depends on the concentrated presence of highly stable, soluble proteins called crystallins, which have been recruited through evolution from diverse metabolic enzymes and stress-protective proteins (Cvekl and Piatigorsky, 1996; Piatigorsky, 1998). Crystallins are differentially accumulated in successive layers of lens fibre cells to give a gradient of refractive index that decreases smoothly from the centre to the periphery. Accumulated metabolic enzymes also contribute to transparency in the cornea (Piatigorsky, 1998; Jester *et al*., 1999). Lens induction has been extensively reviewed (Jacobson, 1966; Jacobson and Sater, 1988; Saha *et al*., 1989; Grainger, 1992, 1996; Cvekl and Piatigorsky, 1996; Oliver and Gruss, 1997; Kondoh, 1999; Treisman, 1999; Ogino and Yasuda, 2000; Wawersik and Maas, 2000). For reviews of the factors involved in later stages of lens differentiation, which include FGFs and TGFβ, see Wride (1996) and McAvoy *et al*. (1999).

Early Events and Markers

A time-course of lens placode development together with molecular markers of lens placode ectoderm is given in Table 3. Molecules involved in lens placode induction and development have been reviewed recently by several authors (Cvekl and Piatigorsky, 1996; Oliver and Gruss, 1997; Kondoh, 1999; Treisman, 1999; Ogino and Yasuda, 2000; Wawersik and Maas, 2000).

Competence

In gastrula stage amphibian embryos, all nonneural ectoderm is competent to form lens when grafted to the lens-forming region of open neural plate stage hosts. During neurulation, this competence becomes restricted to lens-forming and nearby head ectoderm (Liedke, 1951, 1955;

Reyer, 1958a,b; Henry and Grainger, 1987). In contrast, in the chick, trunk ectoderm is competent to form lens (in cocultures with optic vesicle or prospective cardiac mesoderm, in the presence of serum) until at least the 14-somite stage (Karkinen-Jääskeläinen, 1978; Jorquera *et al*., 1989).

Temporal changes in lens competence are also seen within the same tissue in *Xenopus*. Explanted ventral ectoderm from gastrula stage *Xenopus* embryos autonomously gains (mid to late gastrula) and then loses (late gastrula to early neurula) lens competence, as assayed by grafting it to the presumptive lens region of open neural plate stage hosts (Servetnick and Grainger, 1991). Further, nonlens head ectoderm from neural tube stage embryos (including otic placode ectoderm) is more competent to form lens when grafted to the optic vesicle than is nonlens head ectoderm from neural plate stage embryos (Grainger *et al*., 1997), suggesting a general increase in lens competence in head ectoderm during neurulation.

Pax6 expression is required in head ectoderm to enable response to a lens-inducing signal from the optic vesicle in mice (see below; Fujiwara *et al*., 1994; Quinn *et al*., 1996; Collinson *et al*., 2000). Hence, *Pax6* expression is necessary for lens-forming competence. This is one of the few instances in which placode-forming competence can be related to the expression of a particular molecule. Clearly, not all lens-competent ectoderm expresses *Pax6* (e.g., nonneural gastrula stage ectoderm). It is probable, therefore, that lens competence reflects competence to express *Pax6*. This competence changes both spatially and temporally during development in response to as-yet unknown signals.

Lens regeneration from diencephalic tissues. Although the lens is derived from surface ectoderm in normal development, various diencephalic-derived tissues have the ability to transdifferentiate to form lens cells. In larval and adult urodele amphibians, the dorsal iris epithelium will regenerate a lens after lentectomy ("Wolffian lens regeneration"); for an historical review, see Reyer (1954). Lenslike structures or lentoids containing crystallins also develop from a variety of diencephalic-derived tissues in culture, including retina and epiphysis, from birds and mammals as well as amphibians (reviewed in McAvoy, 1980). As pointed out by Grindley *et al*. (1995), all these tissues express *Pax6*. Indeed, *Pax6* upregulation correlates with lens transdifferentiation of avian retinal pigmented epithelium cells in culture (Kosaka *et al*., 1998) and with lens regeneration *in vivo*, both from the iris in the newt and from the cornea in *Xenopus* (Mizuno *et al*., 1999). Here again, therefore, competence to form lens seems to reflect either *Pax6* expression or competence to express *Pax6* in response to lens-inducing signals.

Specification/Commitment

In *Xenopus*, lens specification occurs when contact with the optic vesicle is established, at the 8–9 somite stage (stage 21; Henry and Grainger, 1990). In chick and mouse, specification has not been tested rigorously. When cultured in the presence of serum, prospective lens ectoderm from chick and mouse shows some lens differentiation before contact with the optic vesicle (Karkinen-Jääskeläinen, 1978). Indeed, ectoderm rostral to the head process from neural plate to early neurula stage chick embryos (stage 4–5) will form lens in culture in the presence of serum, as will presumptive lens ectoderm, lateral head ectoderm, and stomodeal ectoderm from 10–13 somite stage embryos (stage 10–11; Barabanov and Fedtsova, 1982). The presence of serum in these cultures makes them hard to interpret, however. Interestingly, δ-crystallin is transiently expressed in the adenohypophysis and surrounding oral ectoderm in normal chick development (Barabanov, 1977; Fedtsova *et al*., 1981; Ueda and Okada, 1986; Sullivan *et al*., 1998). These results illustrate the important point that a given region of ectoderm may be exposed to multiple inducing signals and show some response to those signals, but the eventual fate adopted depends on which signals first have sufficient cumulative effects to trigger differentiation (Jacobson, 1966).

Inducing Tissues

Optic cup. The earliest experiments on lens induction were performed a century ago by Spemann (1901; quoted in Saha *et al.*, 1989), who showed that the lens failed to form following ablation of the optic cup in *Rana*. However, this was not the case in all species tested (reviewed in Saha *et al*., 1989), casting doubt on the necessity of the optic vesicle for lens formation. Various experimenters showed that the optic cup could induce lens formation in epidermis from other regions of the body (reviewed in Saha *et al*., 1989), suggesting that the optic cup was both necessary and sufficient for lens induction. However, the lack of donor-host marking in many of these experiments suggested possible artefacts due to contamination with residual donor lens tissue (reviewed in Saha *et al*., 1989). More recent experiments in *Xenopus* and *Rana* showed that the optic cup is a weak inducer in these species (Henry and Grainger, 1987; Grainger *et al*., 1988, 1997; Saha *et al*., 1989), also casting doubt on the sufficiency of the optic cup for lens induction.

However, in avian embryos, quail-chick experiments showed that the optic cup can induce lens from trunk ectoderm (Karkinen-Jääskeläinen, 1978). In zebrafish, overexpression of *Six6* leads to ectopic retina formation in the midbrain and, occasionally, to associated ectopic lens formation (Bernier *et al*., 2000). In *Xenopus*, lenses do not form when the optic vesicles fail to evaginate after abrogation of the function of the *tailless* (*Xtll*) orphan nuclear receptor (Hollemann *et al*., 1998). The optic cup is necessary for lens formation in mice, as in *Rx* mutants, the optic vesicles do not form and no eye structures are visible (Mathers *et al*., 1997). In *Lhx2* mutants, in which the optic vesicle fails to contact the surface ectoderm, *Pax6* is not expressed in presumptive lens ectoderm and neither lens placodes nor lenses form (Porter *et al*., 1997). Similarly, lens develop-

TABLE 3
Molecular Markers and Early Events in Lens Placode Development

		Zebrafish		
10 h	End of gastrulation	*Pax6.2*	Transcription factor	Nornes *et al.*, 1998
≤14 h	≤10 somites	*Pax6.1*	Transcription factor	Püschel *et al.*, 1992
14 h	10 somites	Prox1	Transcription factor	Glasgow and Tomarev, 1998
		Xenopus		
Stage 10.5	Early gastrula	*Sox2; Sox3*	Transcription factors	Mizuseki *et al.*, 1998; Penzel *et al.*, 1997; Zygar *et al.*, 1998; Schaefer *et al.*, 1999
Stage 12	Late gastrula	*Pax6; Pitx1*	Transcription factors	Hirsch and Harris, 1997; Li *et al.*, 1997; Hollemann and Pieler, 1999
Stage 13	Late gastrula/early neural plate	*Xlens1* (probably *FoxE1*)	Transcription factor	Kenyon *et al.*, 1999
Stage 14–15	Neural plate	*Otx2*	Transcription factor	Zygar *et al.*, 1998
Stage 21	8–9 somites	Optic vesicles contact ectoderm		Henry and Grainger, 1990
Stage 27	19 somites	Placodes thicken		Nieuwkoop and Faber, 1967
		Chick		
Stage 3	Gastrula	*Sox3*	Transcription factor	Rex *et al.*, 1997; Dr. Paul Scotting, personal communication
Stage 6	Neurula	*Pax6; Six4*	Transcription factors	Li *et al.*, 1994; Esteve and Bovolenta, 1999
Stage 8	4 somites	NCAM	Adhesion molecule	Thiery *et al.*, 1982
Stage 10	9–11 somites	Optic vesicles contact ectoderm		Karkinen-Jääskeläinen, 1978
Stage 11	12 somites	Placodes thicken		McKeehan, 1951
Stage 11	13 somites	*Six3; L-Maf; Gata3*	Transcription factors	Bovolenta *et al.*, 1998; Ogino and Yasuda, 1998; Sheng and Westphal, 1999
Stage 12	16 somites	*Sox2; Sox3*	Transcription factors	Kamachi *et al.*, 1998
Stage 12	16 somites	*Deltex2*	Notch pathway	Frolova and Beebe, 2000
Stage 13	19 somites	δ-crystallin	Lens protein	Kamachi *et al.*, 1998
Stage 13/14	21 somites	Lens invagination		McKeehan, 1951
≤Stage 14	≤22 somites	*Serrate1*	Notch ligand	Myat *et al.*, 1996
≤Stage 14	≤22 somites	*SFrp1*	Wnt antagonist	Esteve *et al.*, 2000
Stage 14	22 somites	*Prox1; Sox1*	Transcription factors	Tomarev *et al.*, 1996; Kamachi *et al.*, 1998
Stage 16	27–29 somites	Lens vesicle forms		Karkinen-Jääskeläinen, 1978
		Mouse		
E8	1–7 somites	*Pax6; BF1 (Foxg1)*	Transcription factors	Walther and Gruss, 1991; Grindley *et al.*, 1995; Hatini *et al.*, 1999
E9	13–20 somites	*Sox2*	Transcription factor	Furuta and Hogan, 1998; Wawersik *et al.*, 1999
E9	13–20 somites	*sFRP2*	Wnt antagonist	Wawersik *et al.*, 1999
E9.5	19–25 somites	Optic vesicles contact ectoderm; placodes thicken		Karkinen-Jääskeläinen, 1978; Kaufman, 1992
E9.5	22 somites	*Msx2*	Transcription factor	Monaghan *et al.*, 1991; Furuta and Hogan, 1998
E9.5	21–29 somites	*Prox1; FoxE3; AP2α; AP2β*	Transcription factors	Oliver *et al.*, 1993; Wigle *et al.*, 1999; Blixt *et al.*, 2000; West-Mays *et al.*, 1999
E9.5	21–29 somites	*Eya1*	Transcription cofactor	Xu *et al.*, 1997
E9.5	21–29 somites	Activin receptor I	Receptor	Yoshikawa *et al.*, 2000
E10	32 somites	Lens vesicle forms		Karkinen-Jääskeläinen, 1978
≤E10.5	≤35–39 somites	*Sox1*	Transcription factor	Nishiguchi *et al.*, 1998
E10.5	35–39 somites	*Six3; Pitx3; Maf*	Transcription factors	Oliver *et al.*, 1995a; Semina *et al.*, 1997, 1998; Kawauchi *et al.*, 1999; Kim *et al.*, 1999; Ring *et al.*, 2000

ment is severely disturbed in *HES1* mutant mice, in which the optic vesicles fail to interact properly with the surface ectoderm (Tomita *et al.*, 1996). If the optic vesicle is removed as late as the 20-somite stage (early E9.5) in mice, the early lens marker *Sox2* is not induced and lenses do not form, again suggesting a requirement for the optic cup in lens development (Furuta and Hogan, 1998).

In summary, therefore, there is substantial evidence to suggest that the optic cup is necessary for lens formation, and in at least some species it is also sufficient for lens induction. The molecular nature of some optic cup-derived signals involved in lens induction is described below.

Anterior mesendoderm and anterior neural plate. Other tissues besides the optic vesicle have also been shown to be important in lens placode induction. In normal amphibian development, the first tissue to underlie presumptive lens ectoderm is the endoderm of the future pharynx (foregut). As gastrulation proceeds, future heart tissue extends to the posterior margin of the lens ectoderm. Mangold (1931) (quoted in Twitty, 1955) suggested that the archenteron roof might act directly on lens epidermis. Indeed, prechordal mesoderm from the archenteron roof of early (but not middle or late) gastrula stage *Triturus* embryos can induce lens from late gastrula presumptive trunk ectoderm, in the absence of retinal tissue (Kawakami, 1952). In the chick, presumptive cardiac mesoderm can induce lens from trunk ectoderm (Jorquera *et al.*, 1989). Also in the chick, foregut endoderm can induce lens from oral ectoderm (which includes the anlage of Rathke's pouch) from 10–19 somite stage embryos (Fedtsova and Barabanov, 1978). In *Xenopus*, however, coculture studies show that neither dorsolateral mesoderm nor endoderm can induce lens, whether from lens-competent ventral gastrula ectoderm or from neural plate stage (unspecified) presumptive lens ectoderm (Henry and Grainger, 1990). Furthermore, it was suggested that mesoderm is not required for lens induction, since direct neural induction of *Xenopus* animal caps by BMP inhibitors or cell dissociation, which results in anterior neural specification, also leads to activation of *Pax6* and lens-specific crystallins (Altmann *et al.*, 1997). However, anterior neural plate alone cannot induce lens from lens-competent gastrula-stage ectoderm (Henry and Grainger, 1990), but does so when combined with underlying mesendoderm (as assayed by the induction of lens placode markers and crystallin immunoreactivity; Zygar *et al.*, 1998).

Lens induction from unspecified prospective lens ectoderm, by tissues other than the optic cup, is usually most successful when anterior neural plate is combined with anterior mesendoderm. Anterior endoderm or anterior lateral plate mesoderm can induce lens from unspecified (early neurula stage) lens ectoderm, but not from trunk epidermis, in *Ambystoma* and *Taricha* (Jacobson, 1955, 1958). In *Xenopus*, anterior neural plate-mediated induction of lens from unspecified (neural plate stage) lens ectoderm is enhanced by dorsolateral mesoderm (Henry and Grainger, 1990; Servetnick *et al.*, 1996). Neural plate or prospective heart mesoderm can induce lens from prospective placode ectoderm (the common preplacodal primordium) from early neural plate stage *Taricha* embryos (Jacobson, 1963a). Again, lens induction is most successful when neural plate and prospective heart mesoderm are both present, while anterior endoderm was also implicated to some extent (Jacobson, 1963a,b; reviewed in Jacobson, 1966). In *Ambystoma*, good lens differentiation was obtained when prospective lens ectoderm was grafted to the stomodeal region, close to prospective foregut endoderm (Carpenter, 1937).

Summary. In summary, there is substantial evidence to support a role for anterior mesendoderm and anterior neural plate in lens induction, in addition to that of the optic cup. A more molecular analysis, exemplified by that of Zygar *et al.* (1998), will help to clarify the role of these tissues in lens induction.

Molecules

Transcription factors. Perhaps the most important transcription factor in lens placode development is *Pax6*, the vertebrate homologue of the *Drosophila eyeless* and *twin of eyeless* genes (Quiring *et al.*, 1994; Czerny *et al.*, 1999). *Pax6* is implicated in eye and anterior head development in many different animal phyla (see Wawersik and Maas, 2000; Gehring and Ikeo, 1999; Callaerts *et al.*, 1997; Oliver and Gruss, 1997; Cvekl and Piatigorsky, 1996). *Pax6* expression defines the eye field in both the neuroectoderm and surface ectoderm of the head. *Pax6* can directly regulate lens *crystallin* gene expression (reviewed in Callaerts *et al.*, 1997; Cvekl and Piatigorsky, 1996), but is also necessary for much earlier stages of lens placode induction. A lens-specific *Pax6* transcriptional control element has been identified in the mouse that directs reporter gene expression from the 12-somite stage (E8.5) specifically in presumptive lens and corneal ectoderm (Williams *et al.*, 1998; Kammandel *et al.*, 1999). This will prove a very useful tool in the analysis of lens placode induction.

Pax6 is essential for lens formation, as seen in mouse *Small-eye* and human *Aniridia* mutations (Hogan *et al.*, 1986; Hill *et al.*, 1991; Quiring *et al.*, 1994; Grindley *et al.*, 1995; Ton *et al.*, 1991; Glaser *et al.*, 1992; Jordan *et al.*, 1992). *Pax6* may autoregulate (Grindley *et al.*, 1995), and it can cell-autonomously induce lens-specific markers in animal caps and ectopic lenses (and eyes) *in vivo* in *Xenopus*, albeit only in the head (Altmann *et al.*, 1997; Chow *et al.*, 1999). *Pax6* is required cell-autonomously in mouse head ectoderm for lens placode formation (Quinn *et al.*, 1996) and for competence to respond to a lens-inducing signal from the optic vesicle (Fujiwara *et al.*, 1994; Collinson *et al.*, 2000). *Pax6* is therefore both necessary and sufficient for lens formation, at least in head ectoderm.

It has been suggested that contact between the optic vesicle and lens shields the lens-forming region from inhibitory signals produced by migrating neural crest cells. In *Pax6* mutant rats, which lack lenses, mesencephalic neural crest cells that normally migrate to the frontonasal mass

accumulate around the optic cup (Matsuo *et al*., 1993). However, tissue recombination experiments show that *Pax6* mutant ectoderm cannot form lenses in response to wild-type optic vesicle (Fujiwara *et al*., 1994). Further, *Pax6* can cell-autonomously induce lens-specific markers in animal caps and ectopic lenses *in vivo* in *Xenopus* head ectoderm (Altmann *et al*., 1997; Chow *et al*., 1999). These results suggest that the lack of lenses in *Pax6* mutants is due to a direct requirement for lens formation, rather than an indirect effect on neural crest cell migration.

In *Xenopus*, the winged-helix transcription factor *Xlens1* (probably *Xenopus FoxE1*; Kaestner *et al*., 2000) is expressed from late gastrula stages in a stripe of ectoderm at the anterior border of the neural plate, including the presumptive lens and olfactory ectoderm (Kenyon *et al*., 1999). It is restricted to presumptive lens ectoderm by neural tube stages, and is maintained in the lens placode and vesicle; it is downregulated in differentiating lens cells (Kenyon *et al*., 1999). *Pax6* can induce *Xlens1* expression and *crystallin* gene expression *in vitro*, but *Xlens1* is not sufficient to induce *crystallin* gene expression. *Xlens1* misexpression promotes proliferation and suppresses differentiation of lens-forming ectoderm (Kenyon *et al*., 1999).

Microophthalmia and holoprosencephaly are observed in humans with mutations in the *Six3* gene (Wallis *et al*., 1999). In the mouse, *Six3* is expressed at the 21–29 somite stage (E9.5) in the optic vesicles, and only in the lens placode after its formation (Oliver *et al*., 1995a). Overexpression of murine *Six3* in medaka fish embryos induces ectopic lenses in the otic vesicle in a noncell autonomous fashion, suggesting that *Six3* expression may induce a secreted factor that alters cell fate specification (Oliver *et al*., 1996). Together with its expression in the optic vesicles, this raises the possibility of *Six3* involvement in the production of a lens-forming signal from the optic vesicles (see next section).

Various members of the *Sox* family of HMG-box transcription factors are expressed in the presumptive lens placode of chick, mouse, and *Xenopus* embryos after the optic vesicle contacts the overlying ectoderm, before the placode can be morphologically detected (Kamachi *et al*., 1995, 1998; Furuta and Hogan, 1998; Zygar *et al*., 1998). These genes can directly activate *crystallin* gene expression and are essential for lens differentiation (Kamachi *et al*., 1995; Nishiguchi *et al*., 1998). In the medaka fish, as described in the previous section, *Sox3* overexpression leads to ectopic expression of *Pax6* and ectopic lens formation in ventral and lateral head ectoderm (Köster *et al*., 2000).

The *paired*-like homeobox gene *Prox1* and the winged-helix transcription factor gene *FoxE3* are required for late stages in lens morphogenesis (Wigle *et al*., 1999; Blixt *et al*., 2000). Mutation in the *paired*-like homeobox gene *Pitx3* causes the *aphakia* phenotype in mice, characterised by small eyes without lenses; in these mice, the lens cup forms normally but subsequent development is impaired (Semina *et al*., 1997). *Pitx3* mutation is associated with cataracts in humans, again suggesting involvement in later stages of lens differentiation (Semina *et al*., 1998). The bZIP transcription factor gene *Maf* is expressed in the lens vesicle after invagination, regulates *crystallin* gene expression, and is required for terminal lens fibre differentiation (Ogino and Yasuda, 1998; Kawauchi *et al*., 1999; Kim *et al*., 1999; Ring *et al*., 2000). *Eya1* expression in the lens placode is missing in *Pax6* mutant mice (Xu *et al*., 1997). *Eya1* mutation in humans is associated with cataracts, suggesting a late function in lens development (Azuma *et al*., 2000).

Signalling molecules. BMP7, which is expressed in the optic vesicle and overlying head ectoderm at the 21–29 somite stage (E9.5), is required for lens placode formation in mice (Wawersik *et al*., 1999). BMP7-specific antagonists inhibit lens formation in optic rudiment cultures, while in *Bmp7* mutants, *Sox2* is absent from the presumptive lens placode at the 13–20 somite stage (E9) and *Pax6* is no longer expressed at the 21–29 somite stage (E9.5) in the ectoderm of the lens-forming region (Wawersik *et al*., 1999). *Bmp7* expression is unaffected in *Pax6* mutant mice, suggesting that BMP7 acts upstream of *Pax6* in lens placode ectoderm and that it is required to maintain ectodermal *Pax6* expression (Wawersik *et al*., 1999). The authors suggest that BMP7 is required for a late stage of preplacode formation and thus acts to maintain rather than initiate placode formation.

BMP4 from the optic vesicle is also required for a late phase of lens induction in mice (Furuta and Hogan, 1998). It is first expressed in the distal part of the forming optic vesicle and overlying ectoderm at the 14–16 somite stage (E8.5). Towards the stage of lens placode formation at the 21–29 somite stage (E9.5), its expression becomes restricted to the dorsal tip of the optic vesicle and it is no longer seen in the lens placode. In *Bmp4* mutant embryos, the lens placode does not form, and this can be rescued in explants by exogenous BMP4 protein. *Msx2* expression is lost, but *Pax6* and *Six3* expression are unaffected, suggesting a defect in a later phase of lens determination than seen in *Bmp7* mutants (Wawersik *et al*., 1999). This also suggests that *Pax6* is not sufficient for lens formation in *Bmp4* mutants. BMP4 alone cannot substitute for the optic vesicle *in vivo* at the 16–20 somite stage in wild-type embryos, confirming that other signals from the optic vesicle are also required for lens induction (Furuta and Hogan, 1998).

Retinoic acid has also been implicated in lens induction. Retinoids are produced by the optic vesicle and head ectoderm, and this retinoid production is reduced in *Pax6* mutant mice (Enwright and Grainger, 2000). Retinoic acid production by the frontonasal mesenchyme is also affected in *Pax6* mutant mice (Anchan *et al*., 1997). Transgene reporters containing retinoic acid response elements (RAREs) are active in the optic vesicles from the 8–12 somite stage (E8.5) and in presumptive lens ectoderm from the 10-somite stage (E8.75; Rossant *et al*., 1991; Balkan *et al*., 1992; Enwright and Grainger, 2000). RALDH2 and RALDH3, retinaldehyde dehydrogenases that catalyse retinoic acid formation, are expressed in the optic vesicle from the 8–12 somite stage (E8.5; Mic *et al*., 2000). Further, retinoic acid receptors can directly activate crystallin genes

(Tini *et al.*, 1993; Li *et al.*, 1997; Gopal-Srivastava *et al.*, 1998), and lenses sometimes do not form in mice null for retinoic acid receptors α and γ (Lohnes *et al.*, 1994). The retinoic acid-responsive transcription factors *AP-2α* and *AP-2β* are expressed in the lens placode; *AP-2α* is required for morphogenesis of the lens vesicle, and it is possible that these two genes act redundantly in earlier stages of placode formation (West-Mays *et al.*, 1999).

In the rat, FGF1 (aFGF) is transiently expressed in anterior optic vesicle cells directly apposed to the lens placode (de Iongh and McAvoy, 1993). In the chick, *Fgf8* is expressed in the distal optic vesicle contacting the head ectoderm at the 13-somite stage (stage 11), and FGF8 ectopically applied to the eye region can induce the lens marker *L-Maf* (Vogel-Höpker *et al.*, 2000). FGFs seem to be important in lens regeneration both after lentectomy *in vivo* (Del Rio-Tsonis *et al.*, 1997, 1998) and in lens transdifferentiation *in vitro* (Hyuga *et al.*, 1993; Bosco *et al.*, 1997; Sakaguchi *et al.*, 1997). They are also important for lens fibre differentiation during normal lens development (reviewed in McAvoy *et al.*, 1999).

Summary

Competence to form lens placodes is widespread at gastrula and neural plate stages but is restricted to (and indeed increases in) head ectoderm during neurulation, at least in amphibian embryos. Lens placode specification or commitment occurs before or after contact with the optic vesicles, depending on the species. Neural plate and anterior mesendoderm are sufficient for lens placode induction in some species, while the optic cup is both necessary and sufficient. This suggests a model in which anterior neural plate and mesendoderm are the initial source of inducing signals that are subsequently reinforced by signals from the optic cup. Optic cup-derived signals include BMP4, which is necessary but not sufficient for placode induction, and BMP7, which is essential for placode maintenance.

TRIGEMINAL PLACODES

Derivatives and Function

The trigeminal ganglion complex of cranial nerve V in all craniates develops from two separate ganglia, the ophthalmic (opV; sometimes called profundal) and maxillomandibular (mmV; sometimes called gasserian or trigeminal, the latter used when "profundal" is used for the ophthalmic trigeminal ganglion). In most craniates (hagfish, lampreys, lungfish, and tetrapods) the two ganglia fuse during development to form a single complex. In *Xenopus*, the ganglia are separate distally but fused at their proximal end as they condense at the 15-somite stage (stage 24; Schlosser and Northcutt, 2000). In the axolotl, the two ganglia are completely separate until some time after the 25-somite stage (stage 35), when they fuse (Northcutt and Brändle, 1995). In elasmobranchs and some basal actinopterygians, such as *Polypterus*, the two ganglia and even the nerve roots remain completely separate throughout development (Northcutt and Bemis, 1993; Piotrowski and Northcutt, 1996; also see discussion in Schlosser and Northcutt, 2000).

The trigeminal ganglion is of mixed origin, containing neurons derived both from neural crest and from placodes (all supporting cells in the ganglion are derived from the neural crest). An exception may be seen in urodeles, where neural crest cells do not seem to contribute neurons to the opV lobe and a mmV placode was not seen, suggesting that the neurons of the mmV lobe are all derived from neural crest cells (Northcutt and Brändle, 1995). However, an earlier report suggested the existence of a small mmV placode that did contribute neurons to the mmV lobe (Stone, 1922). Separate or bipartite opV and mmV placodes contribute neurons to their respective ganglia in fish, frogs, birds, and mice (Schilling and Kimmel, 1994; Knouff, 1935; Schlosser *et al.*, 1999; Schlosser and Northcutt, 2000; Hamburger and Hamilton, 1951; D'Amico-Martel and Noden, 1983; Ma *et al.*, 1998).

In anamniotes, trigeminal neurons are born very early and form part of the primary nervous system that mediates swimming reflexes. In mammals, the ophthalmic branch of the ophthalmic ganglion innervates the skin of the head region, the eyeball and eye muscles, and the nose; the maxillary branch innervates the upper jaw, including the upper teeth, while the mandibular branch innervates the lower jaw, including the lower teeth and the tongue. Similar innervation patterns are seen in other vertebrates. Trigeminal ganglion neurons mediate touch, pain, temperature, and proprioception, although placode-derived trigeminal neurons may not mediate proprioception (see Noden, 1980). In addition, many proprioceptive trigeminal afferents from muscles associated with jaw movement originate from the mesencephalic nucleus of the trigeminal nerve in the midbrain. This nucleus comprises the only primary sensory neurons in the brain and it has been suggested to be derived from the neural crest (Narayanan and Narayanan, 1978). Independently evolved trigeminal-mediated sensory modalities also include infrared reception in snakes (e.g., Tan and Gopalakrishnakone, 1988) and possibly vampire bats (Kishida *et al.*, 1984), electroreception in monotremes (reviewed in Pettigrew, 1999), and thermal taste sensation in mammals (Cruz and Green, 2000). Ophthalmic trigeminal axons also mediate magnetoreception in fish and birds (Beason and Semm, 1996; Walker *et al.*, 1997; Diebel *et al.*, 2000), possibly by innervating magnetoreceptors in the olfactory lamellae (Walker *et al.*, 1997; Diebel *et al.*, 2000).

In birds, both opV and mmV lobes of the ganglion contain small neural crest-derived neurons proximally, and large placode-derived neurons distally (Hamburger and Hamilton, 1951; D'Amico-Martel and Noden, 1983; Fig. 4). Placode-derived neurons can be distinguished from neural crest-derived neurons by their expression of *Brn3* (Artinger *et al.*, 1998). The placode-derived neurons differentiate before the neural crest-derived neurons (D'Amico-Martel

and Noden, 1980). The first condensation of the trigeminal ganglion is composed only of neural crest-derived cells that are later joined by placode-derived neurons (Covell and Noden, 1989), but neural crest cells are not required for placode formation (Stark *et al*., 1997) or for gangliogenesis and target-finding by placode-derived neurons (Hamburger, 1961). Indeed, ectopic opV ganglia sometimes differentiate within the epidermis in the chick (Kuratani and Hirano, 1990). The main difference seen in neural crest-ablated embryos is that the placode-derived neurons tend to form two separate ganglia and disperse more than in the presence of neural crest cells, suggesting the latter act as an aggregation centre (Hamburger, 1961). Also after neural crest ablation, development of placode-derived trigeminal ganglia and their central projections is delayed, but not halted (Moody and Heaton, 1983b). Placode-derived trigeminal neurons, in contrast, are necessary for the establishment of normal peripheral projections by neural crest-derived trigeminal neurons (Hamburger, 1961). Further, placode-derived neuronal projections to the metencephalon are essential for trigeminal motor neuron migration and axonal projection (Moody and Heaton, 1983a,b).

Some interesting parallels may be drawn between the large, placode-derived distal neurons of the trigeminal ganglia and the large, ventrolateral neurons of the entirely neural crest-derived trunk dorsal root ganglia. In both mouse and chick dorsal root ganglia, the large ventrolateral neurons are born earlier than the small dorsomedial neurons (Carr and Simpson, 1978; Lawson and Biscoe, 1979). In the mouse, *ngn2* is required exclusively during the early phase of neurogenesis for development of large neurons, while *ngn1* is required during both phases, for a subset of large neurons and for the later generation of small neurons (Ma *et al*., 1999). *Ngn2* is expressed in neural crest cells early in their migration (Gradwohl *et al*., 1996; Sommer *et al*., 1996; Perez *et al*., 1999; Ma *et al*., 1999), and *ngn2*-positive neural crest cells in explant cultures are committed to a sensory fate (Greenwood *et al*., 1999). In the chick, forced expression of *ngn2* in premigratory neural crest cells can bias them to a sensory fate (Perez *et al*., 1999). These results suggest the existence of a population of neural crest cells that is committed to a sensory fate early in their migration by expression of *ngn2*; these give rise to the early-born large ventrolateral neurons that could then provide a population of pioneer neurons (Ma *et al*., 1999). In the trigeminal ganglion, therefore, it would appear that the large placode-derived neurons are equivalent, at least in their pioneer function, to the large neurons of the dorsal root ganglia. In contrast to the requirement for *ngn2* in large dorsal root ganglion neurons, *ngn1* expression is required in the trigeminal placode for ganglion formation, although *ngn1* activates *ngn2* expression in these cells (Fode *et al*., 1998; Ma *et al*., 1998).

Early Events and Markers

A time course of trigeminal placode development and marker expression in different vertebrates is given in Table 4. In the chick, the trigeminal placodes are detectable morphologically only as scattered foci of neuroblast delamination (D'Amico-Martel and Noden, 1983), although *Pax3* is a useful early marker for prospective chick opV ectoderm from the 4-somite stage (Stark *et al*., 1997). However, in most vertebrates, including turtles (Brachet, 1914), the trigeminal placodes are clearly distinct as thickened patches of ectoderm. Neurogenesis often begins within prospective trigeminal placode ectoderm well before the placode itself can be detected morphologically (see Table 4).

Competence

In *Pleurodeles*, belly ectoderm from neurula stage embryos can contribute to all placode-derived ganglia, including the trigeminal, when substituted for ectoderm lateral to the neural folds in same-stage host embryos (Chibon, 1967). Székely (1959) exchanged the trigeminal and vagal (nodose) placodes between late neurula stage *Triturus* and *Pleurodeles* embryos and used a behavioural response as his assay for neuronal fate. In the larval animal, the vagal nerve (cranial nerve X) mediates a gill depression reflex. After metamorphosis, the trigeminal nerve mediates the afferent pathway of a corneal (lid-closure) reflex. When the vagal placode was grafted in place of the trigeminal placode, a gill reflex could be evoked in the larval animal by touching the cornea. This suggests that the graft-derived neurons had established functional central connections with vagal motor nuclei, appropriate to the donor fate. After metamorphosis, however, a corneal reflex could be evoked normally, suggesting that normal trigeminal-type connections with the sixth motor nucleus (which mediates the efferent component of the corneal reflex) had also been made. These results show that while some cells in the late neurula stage vagal placode were already committed to a vagal-type fate, other cells were competent to adopt a trigeminal fate. They also show that neuronal subtype specificity (vagal versus trigeminal) is determined to some extent before axon outgrowth occurs.

In the chick, competence to express Pax3 in the opV placode area was mapped by grafting different regions of quail ectoderm to the opV placode of chick hosts (Baker *et al*., 1999). Competence is present in lateral epiblast taken from gastrula and neurula stage embryos. At the 3-somite stage, head ectoderm rostral to the first somite, including the presumptive otic placode, is competent to express Pax3 when grafted to the opV placode region. Competence is highest in head ectoderm rostral to the otic placode region, and competence in otic placode ectoderm is lost after the 3-somite stage, presumably reflecting specification towards an otic fate. Ectoderm from more caudal regions, including the presumptive nodose placode region (vagal region), was not competent to express Pax3 at any stage tested, perhaps suggesting an earlier loss of competence in this ectoderm than is seen in the amphibian embryo (Baker *et al*., 1999). The Wnt receptors *frizzled-2* and *frizzled-7* show a fairly broad expression in rostral head ectoderm at these stages, correlating with competence to express Pax3 (Stark *et al*.,

TABLE 4
Molecular Markers and Early Events in Trigeminal Placode Development

		Zebrafish		
10 h	End of gastrulation	Islet-1	Transcription factor	Korzh *et al.*, 1993
11 h	2 somites	*Zcoe2; ngn1*	Transcription factors	Bally-Cuif *et al.*, 1998
11⅔	5 somites	Placodes visible		Dr. Tom Schilling, personal communication; Kimmel *et al.*, 1995
14 h	10 somites	*Tbx2*	Transcription factor	Ruvinsky *et al.*, 2000
16 h	14 somites	HNK1 surface labelling	Neuronal marker	Metcalfe *et al.*, 1990
16 h	14 somites	L1.1	Neuronal adhesion molecule	Tongiorgi *et al.*, 1995
16.5 h	15 somites	Axons grow out		Metcalfe *et al.*, 1990
		Xenopus		
Stage 10.5–12	Gastrula	Final mitosis of trigeminal neurons		Lamborghini, 1980
Stage 10.5	Early gastrula	*Sox2; Sox3*	Transcription factors	Mizuseki *et al.*, 1998; Penzel *et al.*, 1997
Stage 12	Late gastrula	*Delta1*	Notch ligand	Chitnis *et al.*, 1995
Stage 13	Early neural plate	*XCoe2*	Transcription factor	Dubois *et al.*, 1998
Stage 13.5	Neural plate	*N-tubulin*	Cytoskeletal protein	Chitnis *et al.*, 1995
Stage 14	Neural plate	*Xngnr1; NeuroD*	Transcription factors	Schlosser and Northcutt, 2000
Stage 15	Early neural fold	XIF3	Cytoskeletal protein	Goldstone and Sharpe, 1998
Stage 20	6–7 somites	Islet1	Transcription factor	Sharpe and Goldstone, 2000
Stage 21	8–9 somites	Bipartite opV–mmV placodes visible; neuroblasts/neurons delaminate		Schlosser and Northcutt, 2000
Stage 24	15 somites	opV and mmV ganglia form		Schlosser and Northcutt, 2000
Stage 27	19 somites	opV and mmV placodes are smaller and separate		Schlosser and Northcutt, 2000
		Chick		
Stage 8	4–5 somites	*Pax3* (opV)	Transcription factor	Stark *et al.*, 1997
Stage 10	10 somites	*FREK* (opV)	FGF receptor	Marcelle *et al.*, 1995; Stark *et al.*, 1997
Stage 11	13 somites	Neurofilament (NF-M) (opV)	Cytoskeletal protein	J. Sechrist and M. Bronner-Fraser, unpublished data
Stage 11	13 somites	Neuroblasts/neurons delaminate (opV)		Stark *et al.*, 1997
Stage 12	16 somites	*GFRα2* (opV and mmV)	GDNF receptor	Homma *et al.*, 2000
≤Stage 13	≤19 somites	Neuronal β-tubulin	Cytoskeletal protein	Moody *et al.*, 1989
Stage 14	22 somites	*GFRα1* (opV and mmV)	GDNF receptor	Homma *et al.*, 2000
≤Stage 15	≤24–27 somites	*Notch1*	Receptor	Myat *et al.*, 1996
Stage 15	24–27 somites	c-*ret* (opV ganglion)	GDNF receptor	Homma *et al.*, 2000
Stage 15	24–27 somites	*Tlx1; Tlx3; Brn3.0* (opV and mmV placode-derived neurons)	Transcription factors	Logan *et al.*, 1998; Artinger *et al.*, 1998
Stage 18	30–36 somites	*GFRα1* (mmV lobe); *GFRα4* (opV and mmV lobes)	GDNF receptors	Homma *et al.*, 2000
Stage 18	30–36 somites	*SFrp1*	Wnt antagonist	Esteve *et al.*, 2000
		Mouse		
E8	1–7 somites	*BF1 (Foxg1)*	Transcription factor	Hatini *et al.*, 1999
E8	>8 somites	*Notch1*	Receptor	Reaume *et al.*, 1992
E8.25	9 somites	*ngn1*	Transcription factor	Ma *et al.*, 1998
≤E8.5	≤10 somites	*Pax3*	Transcription factor	Stark *et al.*, 1997

TABLE 4—*Continued*

		Mouse		
E8.5	12 somites	*Delta1*	Notch ligand	Ma *et al.*, 1998
E8.5	10–12 somites	Neuroblasts delaminate		Verwoerd and van Oostrom, 1979; Nichols, 1986
E9	13–20 somites	*ngn2; NeuroD*	Transcription factors	Ma *et al.*, 1998
≤E9.5	≤21–29 somites	*Eya2*	Transcription cofactor	Xu *et al.*, 1997
E9.5	21–29 somites	*HES6*	Transcription factor	Pissarra *et al.*, 2000; Vasiliauskas and Stern, 2000
≤E10.5	≤35–39 somites	*Dach1* in ganglion	Transcription cofactor	Hammond *et al.*, 1998; Davis *et al.*, 1999

2000). However, neither is expressed specifically in the opV placode, while *frizzled-2* is expressed in the otic placode, which loses competence at early stages to express Pax3. Nonetheless, it is possible that the distribution of competence is related to the distribution of receptor molecules.

Specification/Commitment

Using the same behavioural assay system described in the preceding section, Székely (1959) found that when late neurula stage trigeminal placode was substituted for the vagal placode (between *Triturus* and *Pleurodeles* embryos), there was a normal gill reflex after gill stimulation. Hence, the trigeminal placode is apparently uncommitted at the late neurula stage. The opV placode in 10–12 somite stage *Ambystoma* embryos (stage 26) is already committed to a neuronal fate, as it forms a ganglion when transplanted to the flank (Stone, 1924, 1928a). It is able to substitute for the gasserian (mmV) placode, forming a ganglion that seems entirely to replace the gasserian in form and function (Stone, 1924). However, when substituted for various lateral line placodes, it forms a small ganglion whose nerve fibres often follow along with those of lateral line and other cranial nerves, but never actually innervate lateral line organs (Stone, 1928a).

In the chick, presumptive opV placode ectoderm is specified and committed to express Pax3 essentially concomitant with Pax3 expression in the explanted ectoderm (Baker *et al.*, 1999). Further, Pax3-positive cells in explanted opV ectoderm are specified and committed to form Pax3-positive neurons and are not competent to form epibranchial-type neurons (Baker and Bronner-Fraser, 2000). Cells that are not already expressing Pax3 seem to be competent to form epibranchial placode-type neurons (Baker and Bronner-Fraser, 2000). Thus, individual cells in opV ectoderm seem to be committed to a trigeminal neuron fate once they begin to express Pax3 (Baker and Bronner-Fraser, 2000).

Inducing Tissues

In the chick, the signal(s) that induces Pax3 in presumptive opV placode ectoderm is a diffusible activity derived from the neural tube (Stark *et al.*, 1997). Neural fold ablation shows that neural crest cells are not required for Pax3 expression in opV placode ectoderm, although the opV ganglion that forms is misshapen and displaced (Stark *et al.*, 1997). Blocking interactions between the neural tube and presumptive opV ectoderm with an impermeable barrier leads to the absence of Pax3 expression in the ectoderm and of neurofilament-positive cells in the mesenchyme (Stark *et al.*, 1997). Permeable barriers, including those of small pore size, have no effect on Pax3 expression, showing that a diffusible signal from the neural tube is necessary for opV placode formation and differentiation (Stark *et al.*, 1997). By grafting presumptive opV placode ectoderm (before it is specified to express Pax3) adjacent to the neural tube at different axial levels, Pax3-inducing activity was found to extend the entire length of the neuraxis, with a possible difference at the r2,3 level, where the caudal boundary of Pax3 expression normally lies (Baker *et al.*, 1999). It appears therefore that changes in competence in head ectoderm may be important in the localisation of Pax3 expression to the opV placode itself during normal development. The Pax3-inducing activity from the neural tube was shown in coculture experiments to be direct, rather than being mediated through adjacent mesoderm (Baker *et al.*, 1999).

Molecules

Pax3 is expressed in chick opV ectoderm from the 4-somite stage and is maintained in the distal placode-derived neurons of the opV lobe of the trigeminal ganglion (Stark *et al.*, 1997). *Pax3* is also expressed at low levels in the proximal (neural crest-derived) opV lobe and in the mmV lobe of the ganglion (Stark *et al.*, 1997). A reduction in the opV nerve was reported in *Pax3*-mutant *Splotch* mice (Tremblay *et al.*, 1995), although cranial neural crest migration and contribution to the trigeminal ganglion appear to be normal (Serbedzija and McMahon, 1997). This suggests that the opV defect in the mutant mice is due to the loss of placodal *Pax3* expression and that *Pax3* function is indeed necessary for the formation of the placodal component of the opV ganglion.

Also in the mouse, *ngn1* is essential for *Delta1* expression in the trigeminal placode and for formation of the trigemi-

nal ganglion (Ma *et al.*, 1998). The opV ganglion is also missing in *Otx2* mutant mice, although this was attributed to defects in mesencephalic neural crest cells (Matsuo *et al.*, 1995).

Summary

Competence to form the trigeminal placodes is widespread at gastrula and neurula stages in amphibians; in the chick, competence is restricted to preotic head ectoderm by the 4-somite stage. Commitment occurs during tailbud stages in amphibians, while in the chick, opV placode commitment occurs concomitant with Pax3 expression. A widespread, diffusible neural tube-derived signal is necessary and sufficient for Pax3 expression in unspecified opV placode ectoderm in the chick and for opV ganglion formation. No signalling molecules have as yet been implicated in trigeminal placode induction.

LATERAL LINE PLACODES

Derivatives and Function

The lateral line system of fish and aquatic amphibians comprises lines of secondary sensory cells (i.e., cells that lack centrally projecting axons and must be secondarily innervated) on the surface of the head and trunk (reviewed in Coombs *et al.*, 1989). There are two types of lateral line organs: mechanoreceptive neuromasts that respond to disturbances in the water (Fig. 6) and electroreceptive ampullary organs that respond to weak electric fields. The lateral line system functions in various different behaviours, including surface feeding, schooling behaviour, obstacle avoidance, and subsurface prey detection (reviewed in Coombs *et al.*, 1989). In fish, lateral line organs can be free neuromasts on the skin or in shallow pits or grooves, or entirely recessed in tubular, fluid-filled canals in the skin or bony tissue of the skull or scales; the canals communicate with the surface via pores (reviewed in Münz, 1989). Lateral line organs are thought to be involved in inducing the formation of the canals that enclose them (reviewed in Webb and Noden, 1993). Amphibians do not have canal structures in the lateral line. Many amphibians lose the lateral line system at metamorphosis (reviewed in Fritzsch, 1990). Amphibians with aquatic adult forms, such as *Xenopus*, often retain the lateral line system, while some terrestrial forms, such as the salamander, cover it with epidermis (reviewed in Fritzsch, 1989).

Lateral line organs are derived from a series of dorsolateral placodes on the head of the embryo (Fig. 1A). Phylogenetic surveys of the lateral line system suggest that the earliest gnathostomes possessed at least six pairs of dorsolateral placodes: three preotic and three postotic (Northcutt, 1997). These placodes undergo a remarkable migration that results in the deposition of lines of lateral line organs over the head and along the length of the body. It should be pointed out that essentially nothing is known about the differentiation of specific placodes within the lateral line placode series, although some information is available on lateral line placode induction in general, as will be seen below.

There is some evidence from DiI labelling that neural crest cells may make a small contribution to both hair cells and supporting cells in lateral line neuromasts in *Xenopus* and teleosts (Collazo *et al.*, 1994). Neural crest cells are not required for formation of the lateral line system, however (Stone, 1922).

Lateral line placode migration. The migration of lateral line placodes was first demonstrated in a remarkable experiment by Harrison (1904), who exchanged whole heads between embryos of differently pigmented species of *Rana*. He saw, in living embryos, the outgrowth and migration of the pigmented body line primordium through the unpigmented host epidermis and its conversion into a row of lateral line organs. Stone demonstrated the same phenomenon by transplanting vital dye-stained head epidermis onto unstained *Ambystoma* hosts (Stone, 1933, 1937). An historical review of lateral line experiments in the first half of the century can be found in Wright (1951) and in Lannoo and Smith (1989). A more recent general review of lateral line structure and development in different phyla is given in Blaxter (1987).

A lateral line placode gives rise both to the neuromasts of a particular line and to the neurons that innervate that line (Stone, 1922). Each placode is polarised such that one pole gives rise to the cells of the migrating primordium, while the other pole contributes neuroblasts to the ganglion for that primordium (Stone, 1922; Sahly *et al.*, 1999). The neuroblasts delaminate and aggregate adjacent to the placodes to form the neurons of the lateral line ganglia. As the primordium migrates, it deposits clusters of cells at intervals from its trailing end, possibly due to or aided by epidermal growth (Winklbauer and Hausen, 1983); these will form the primary neuromasts of the line. Additional accessory neuromasts are generated from primary neuromasts such that a single plaque or stitch in a late larva or adult is derived from a single primary neuromast. Maximal speeds reached by migrating primordial cells in *Xenopus* are 20–30 μm/h (Winklbauer and Hausen, 1983). The migrating primordium is tightly encased between the outer epidermal layer, the basal lamina, and the surrounding inner epidermal cells; it displaces the latter as it migrates (Stone, 1933).

Recently, considerable insight has been gained into the molecular nature of the guidance of the posterior lateral line primordium in zebrafish (Shoji *et al.*, 1998). The posterior lateral line placode migrates along the trunk at the level of the horizontal myoseptum of the axial muscles; the myoseptum divides the myotomes into dorsal and ventral halves. In zebrafish, the homologue of the growth-cone repulsive molecule semaphorin III/D/collapsin 1, Sema Z1a, is expressed in the dorsal and ventral portions of the myotome but not in the horizontal myoseptum (Shoji *et al.*, 1998). The lateral line primordium and the comigrating lateral line axons (Metcalfe, 1985) therefore migrate along a

Sema Z1a-free zone. Sema Z1a is also expressed by cells at the leading edge of the migrating lateral line primordium, in front of the growth cones of the lateral line axons that migrate with it (Shoji *et al*., 1998). This expression suggests that it may prevent the growth cones from overtaking the primordium, thus keeping the two in register (Shoji *et al*., 1998). In zebrafish mutants lacking the horizontal myoseptum (*floating head* and *you-too*), Sema Z1a is uniformly expressed by the entire myotome. In these mutants, the lateral line primordium migrates aberrantly ventral to the myotomes, while the lateral line growth cones are defasciculated and follow the aberrant migration pathway of the primordium (Shoji *et al*., 1998). These results suggest that a Sema Z1a-free zone along the horizontal myoseptum delimits the migration pathway of the posterior lateral line placode and its associated axons. It will be interesting to discover whether similar mechanisms determine the migration pathways of cranial lateral lines and lateral line placodes in other species.

Some information is available on the guidance of the lateral line placodes that migrate onto the body in the axolotl. If the ectoderm from over the somites in the pathway of the middle body line primordium is rotated 90°, the migrating primordium either stops at the rostral edge of the graft or follows an abnormal, irregular course over the body (Smith *et al*., 1990). Removal of somites in the path of the primordium also caused it to stop or be diverted, suggesting that cues from both the somites and the overlying ectoderm are necessary for proper migration (Smith *et al*., 1990).

In the axolotl, a postotic lateral line placode transplanted dorsal to the optic vesicle will only begin to migrate when the host supraorbital primordium reaches it, and then it follows the normal supraorbital pathway (Stone, 1929). When preotic and postotic placodes are interchanged, however, they migrate normally without any dependence on host primordia (Stone, 1928b). When a lateral line placode from tailbud stage embryos is grafted to the belly, it will differentiate to form a ganglion but it will not migrate (Stone, 1929). The polarity of the placode, with one pole forming neurons and the other the migrating primordium, and hence its direction of migration, seems already to be established at tailbud stages (Stone, 1928c).

Innervation. Harrison (1904) first showed that axons from the lateral line ganglia extend into the migratory placode. In the development of the zebrafish midbody line, the most advanced growth cone of lateral line axons was deep among the cells of the migrating placode, and growth cones were never seen ahead of the migrating placode (Metcalfe, 1985). Innervation is not required for placode migration or neuromast differentiation, however (Tweedle, 1977). Neural crest-derived Schwann cells are present along the lateral line nerves as the placode migrates (Stone, 1933). It has been suggested that lateral line placodes can also give rise to glia along the lateral line nerves (Collazo *et al*., 1994; Sahly *et al*., 1999). The same lateral line placode can give rise both to mechanosensory neuromasts and to electroreceptors in axolotls (Northcutt *et al*., 1995). Electroreceptors and neuromasts are innervated by primary afferent fibres in the same lateral line nerves, from the same lateral line ganglia (see Bodznick, 1989). Efferent innervation, provided by cells in the rhombencephalic reticular formation near the facial motor nucleus (reviewed in Fritzsch, 1989; Roberts and Meredith, 1989), affords inhibitory input to the neuromast hair cells and suppresses self-stimulation from the animal's own movements (see Bodznick, 1989).

Electroreception. For a general review of vertebrate electroreception, see Bullock (1982). Electroreception in monotreme mammals, which is mediated by trigeminal neurons, is unrelated to lateral line electroreception (for a recent review, see Pettigrew, 1999). The same lateral line placode can give rise both to mechanosensory neuromasts and to electroreceptors in axolotls (Northcutt *et al*., 1995). Electroreceptors and neuromasts are innervated by primary afferent fibres in the same lateral line nerves, from the same lateral line ganglia (Bodznick, 1989). Electroreception is rare in teleosts, but present in all nonteleost fish (except for holostean fish and hagfish) and in many amphibians (except anurans; Bodznick, 1989). The phylogenetic distribution of electroreception indicates that it is a very ancient vertebrate sensory system probably present in the common vertebrate ancestor that later evolved separately in some teleosts (Bodznick, 1989). It is unclear whether lateral line neuromasts evolved from lateral line electroreceptors, or vice versa, or whether both evolved separately from a common ancestor such as the ciliated receptors found in invertebrate chordates (Bodznick, 1989; Budelmann, 1989; Jørgensen, 1989).

Early Events and Markers

A time course of lateral line placode development and marker expression in different anamniotes is given in Table 5.

Competence

Ventral belly ectoderm from early neurula to early tailbud stage axolotl embryos (stages 13–22) is competent to form lateral line organs when grafted to the lens and surrounding head region of neurula stage hosts (Liedke, 1955). Indeed, ventral belly ectoderm retains lateral line competence at least until late tailbud stages (stage 30), while it loses competence to form the otic placode much earlier in development, by midneurula stages (Dr. Gerhard Schlosser, personal communication). Prospective gill ectoderm apparently retains competence to form lateral line organs for longer than it retains otic competence when grafted to the ear region; however, this is complicated by the potential presence of lateral line organs in the gill region in older embryos (Yntema, 1950).

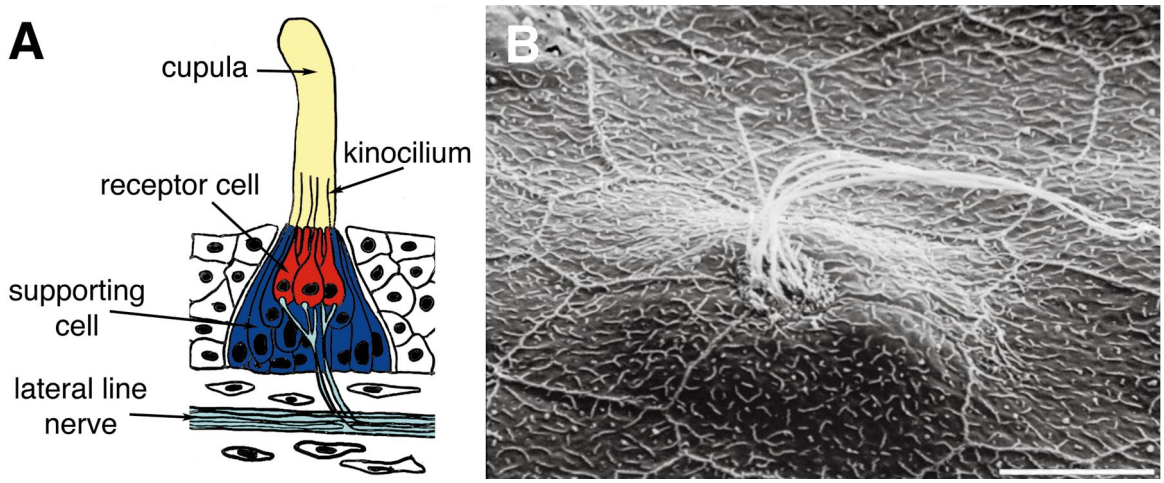

FIG. 6. Lateral line neuromast structure. (A) Schematic section through a neuromast. After Winklbauer (1989). (B) Scanning electron micrograph of a lateral line neuromast on the skin of a stage 49 *Xenopus* tadpole.

Specification/Commitment

In the axolotl, grafting of prospective pigmented lateral line placode ectoderm to the belly of albino host embryos has shown that lateral line placodes are already committed to forming neuromasts and ganglia at neural fold stages (Dr. Gerhard Schlosser, personal communication). Grafting of lateral line placodes to the belly of host embryos shows that at the 9–11 somite stage (stage 25–26), lateral line placodes are committed to ganglion formation (Stone, 1929). Preotic and postotic lateral line placodes can substitute for one another at the 7–12 somite stage (stages 23–27; Stone, 1928b), but not for the ophthalmic trigeminal placode, also showing commitment to a lateral line fate (Stone, 1929).

Inducing Tissues

Lateral line placodes are part of the dorsolateral series of placodes that includes the otic placode. However, their induction does seem to be separate from that of the otic placode, as differences exist in the duration of both competence of nonplacodal ectoderm to form otic or lateral line placodes and inductive activity. Ectoderm from the prospective gill region of middle neurula stage axolotl embryos (stage 16) can be induced to form lateral line organs and ganglia when grafted to the ear region of host embryos at least until after the 25-somite stage (stage 35; Yntema, 1950). This suggests that despite the early commitment of presumptive lateral line ectoderm, the inducing activity persists until late stages. In contrast, ear-inducing activity is lost by the 20-somite stage (stage 32; Yntema, 1950). Further, operations that induce otic vesicles do not always induce lateral line organs (e.g., Stone, 1931; see otic placode section).

In the axolotl, lateral mesoderm from the region bordering the lateral margin of the neural plate is able to induce lateral line sense organs (Holtfreter, 1933). When medial archenteron roof from early neurula stages is grafted into the ventromedial blastocoel of gastrula stage hosts, where it differentiates into notochord, it induces lateral line organs and ganglia, as well as otic vesicles and neural crest derivatives, in the ventral part of the head (Raven and Kloos, 1945). Lateral line organs (and otic vesicles) can be induced in the absence of neural induction, but are always induced in association with neural crest derivatives (Raven and Kloos, 1945). The authors interpreted this as showing that neural crest cells themselves induce both lateral line organs and otic vesicles; however, it could equally be a direct action of mesoderm on the host ectoderm to produce both tissues. Lateral archenteron roof grafts are also able to induce lateral line, otic vesicles, and neural crest derivatives, but with lower frequency than medial grafts (Raven and Kloos, 1945). If the hindbrain is ablated unilaterally in the neurula stage axolotl embryo, lateral line organs can be induced at a distance from the regenerated hindbrain, separated from it by mesenchyme and therefore also suggesting that the mesoderm is involved in the induction (Harrison, 1945).

There is also some evidence for the involvement of the neural plate in the induction of lateral line placodes (Mangold, 1929). When neural plate from early or late neurula stage *Triton* (newt) embryos is grafted into the blastocoel of a gastrula stage host, it induces lateral line organs in the overlying ventral epidermis (Mangold, 1929).

Molecules

Although no molecules have as yet been implicated in lateral line placode induction, retinoic acid affects the number of neurons in lateral line ganglia. Retinoic acid treatment for 1 h at dome stage (late blastula; beginning of epiboly) in zebrafish leads to reduced numbers of neurons in

TABLE 5
Molecular Markers and Early Events in Lateral Line Placode Development

		Zebrafish		
10 h	End of gastrulation	*eya1*	Transcription cofactor	Sahly *et al.*, 1999
10 h	End of gastrulation	*six4.1*	Transcription factor	Kobayashi *et al.*, 2000
11.3 h	4 somites	*Nkx5.1*	Transcription factor	Adamska *et al.*, 2000
13 h	8 somites	Islet-1	Transcription factor	Korzh *et al.*, 1993
16 h	14 somites	*Tbx2*	Transcription factor	Ruvinsky *et al.*, 2000
16.5 h	15 somites	*Kal1.1* (posterior primordium)	Extracellular matrix glycoprotein	Ardouin *et al.*, 2000
18 h	18 somites	Posterior placode visible		Metcalfe, 1985, 1989; Sahly *et al.*, 1999
18 h	18 somites	Hu (posterior ganglion)	Neuronal marker	Raible and Kruse, 2000
18–19 h	18–20 somites	*runxb* (posterior primordium)	Transcription factor	Kataoka *et al.*, 2000
22 h	26 somites	*Kal1.2* (posterior primordium)	Extracellular matrix glycoprotein	Ardouin *et al.*, 2000
24 h	30 somites	*Eya1* (anterodorsal and anteroventral primordia)	Transcription cofactor	Sahly *et al.*, 1999
24 h	30 somites	Hu (anterodorsal ganglion)	Neuronal marker	Raible and Kruse, 2000
≤28 h		Prox1 (migrating primordia)	Transcription factor	Glasgow and Tomarev, 1998
30 h		Hu (middle ganglion)	Neuronal marker	Raible and Kruse, 2000
34 h		*erm; pea3* (migrating primordia)	Transcription factors	Münchberg *et al.*, 1999
40 h		Hu (anteroventral ganglion)	Neuronal marker	Raible and Kruse, 2000
		Xenopus		
Stage 10.5	Early gastrula	*Sox2; Sox3*	Transcription factors	Mizuseki *et al.*, 1998; Penzel *et al.*, 1997
Stage 17	Late neurula	*Six1*	Transcription factor	Pandur and Moody, 2000
Stage 21	8–9 somites	Dorsolateral placode area visible (anterodorsal; anteroventral; middle; plus otic)		Schlosser and Northcutt, 2000
Stage 21	8–9 somites	*Delta1* in dorsolateral placode area prefigures placodes	Notch ligand	Schlosser and Northcutt, 2000
Stage 21	8–9 somites	*NeuroD* (neurogenic regions of placodes)	Transcription factor	Schlosser and Northcutt, 2000
Stage 23	12 somites	*ngnr1* (weak) (neurogenic regions of placodes)	Transcription factor	Schlosser and Northcutt, 2000
Stage 24	15 somites	Anterodorsal placode visible within dorsolateral placode area; anterodorsal ganglion forms; dorsolateral placode area extends to posterior placode		Schlosser and Northcutt, 2000
Stage 27	19 somites	Anteroventral, middle and posterior placodes visible within dorsolateral placode area; anteroventral, middle and posterior ganglia begin to form		Schlosser and Northcutt, 2000
Stage 31	22–23 somites	c-*kit-related receptor 1* (migrating primordia)	Receptor tyrosine kinase	Baker *et al.*, 1995
Stage 31	22–23 somites	*Tbx2; Tbx3* (migrating primordia)	Transcription factors	Takabatake *et al.*, 2000
≤Stage 35/36	≤36 somites	p21-activated kinase 1 (migrating primordia and neuromasts)	Cytoskeletal effector protein	Islam *et al.*, 2000
Stage 35/36	36 somites	*cadherin6* (posterior placode)	Adhesion molecule	David and Wedlich, 2000
Stage 35/36	36 somites	Supratemporal placode visible		Schlosser and Northcutt, 2000

TABLE 5—*Continued*

		Axolotl		
Stage 23	7 somites	Anterodorsal and posterior placodes visible		Northcutt and Brändle, 1995
Stage 27	12 somites	Neurogenesis begins in anterodorsal and posterior placodes		Northcutt and Brändle, 1995
Stage 29	16 somites	Anteroventral placode visible		Northcutt and Brändle, 1995
≤Stage 30	≤17 somites	*Dlx3; Msx2*	Transcription factors	Metscher *et al.*, 1997
Stage 34	24–25 somites	Middle placode visible		Northcutt and Brändle, 1995
Stage 35		*Hoxb3* (middle placode only)	Transcription factor	Metscher *et al.*, 1997
Stage 35		Supratemporal placode visible		Northcutt and Brändle, 1995

the anterior lateral line ganglion and conversely to a striking increase in the number of neurons in the posterior lateral line ganglion (Holder and Hill, 1991). The *premature death* mutation in the axolotl, as well as compromising the ability of cranial neural crest cells to differentiate into cartilage (Graveson and Armstrong, 1990, 1994, 1996), also compromises the ability of lateral line placodes to differentiate into neuromasts (Smith *et al.*, 1994).

Summary

Competence to form lateral line placodes is widespread and retained at least until late tailbud stages in the axolotl. Commitment occurs at neurula/early tailbud stages. Axial and nonaxial mesoderm, and neural plate, are each sufficient to induce lateral line placodes. No signalling molecules have yet been shown to be necessary or sufficient to induce lateral line placodes.

OTIC PLACODES

The otic placode gives rise to the entire inner ear, including the mechanosensory hair cells that transmit balance and auditory information, all supporting cells, and the biomineralised otoliths or otoconia that assist in perception. It also gives rise to the neurons of the vestibuloacoustic (vestibulocochlear) ganglion of cranial nerve VIII, which provide afferent innervation for the inner ear hair cells. The development of the inner ear has been extensively reviewed (Fekete, 1996, 1999; Fritzsch *et al.*, 1997, 1999; Rubel *et al.*, 1997; Whitfield *et al.*, 1997; Torres and Giraldez, 1998). Experiments from the first half of the century are also reviewed in Yntema (1955).

Early Events and Markers

A description of early markers and events in otic placode formation in different species is given in Table 6. The earliest specific marker for the otic placode in all vertebrates seems to be *Pax8*, which is expressed in prospective otic placode ectoderm from late gastrula/neurula stages in zebrafish, *Xenopus*, and the mouse (see Table 6). In the zebrafish, the oval group of cells that makes up the placode thickens and forms an ovoid solid ball just beneath the surface ectoderm, which develops a lumen through cavitation, rather than invaginating through a cup stage as in other vertebrates (Haddon and Lewis, 1996).

Competence

In the newt, ventral ectoderm from early neurula stages is competent to form the otic vesicle when grafted into the otic region of a host of the same stage (Jacobson, 1963a). In the axolotl, prospective olfactory placode ectoderm is competent to form the ear, as shown by rotation of the placodal epidermis at early and late neurula stages (Jacobson, 1963c). Also in the axolotl, the ectoderm surrounding the ear region has the capacity to regenerate the vesicle after extirpation, although potency is markedly decreased after invagination of the otic cup (Kaan, 1926; Yntema, 1933). Hence both head and trunk ectoderm are initially competent to form the otic placode. Ventral belly ectoderm loses competence to form the otic placode by midneurula stages (Dr. Gerhard Schlosser, personal communication). However, prospective limb ectoderm from 5–6 somite stage embryos (stage 22) is competent to form the ear when grafted in place of presumptive ear ectoderm of same-stage hosts (Kaan, 1926). Indeed, limb ectoderm can contribute to the ear when grafted in place of specific regions of the otic cup even when taken from 13–15 somite embryos (stage 28; Kaan, 1926). This interesting retention of otic competence in limb ectoderm should be investigated further. In *Xenopus*, early neural plate stage trunk ectoderm forms otic vesicles in response to beads soaked with FGF2 or 3 implanted into presomitic mesoderm (Lombardo and Slack, 1998; Lombardo *et al.*, 1998). By midneural fold stages, however, trunk epidermis has lost competence to respond.

In the chick, anterior epiblast (presumptive extraembryonic ectoderm) from late gastrula/neural plate stage embryos (stage 3+ to 4) is competent to form an otic vesicle and neurons when grafted to the otic placode region (Groves

and Bronner-Fraser, 2000). Preotic and trunk ectoderm are competent to contribute to the otic vesicle until the 6-somite stage, but retain competence to form Pax2-positive cells until the 10-somite stage (Groves and Bronner-Fraser, 2000). Similarly, presumptive ophthalmic trigeminal placode (midbrain-level) ectoderm is competent to contribute to the otic placode at least up to the 7-somite stage (Baker *et al*., 1999). Preotic head ectoderm extending to the level of the rostral hindbrain, and postotic ectoderm extending to the level of the sixth somite (vagal ectoderm), but not more posterior trunk ectoderm, is competent to form otic vesicles in response to FGF3 overexpressed *in vivo* (Vendrell *et al*., 2000). Since widespread expression of the HSV-1-derived amplicon viral vector was achieved 8 h after infection, this competence seems to persist until at least the 15-somite stage (Vendrell *et al*., 2000).

In summary, otic placode-forming competence is initially widespread but becomes restricted to a relatively broad area of hindbrain-level and vagal ectoderm at neural tube stages. Specific regions of ectoderm, such as limb ectoderm in the axolotl, may retain competence until later stages.

Specification/Commitment

In *Xenopus*, presumptive otic placode ectoderm is specified to form otic vesicles by the early neurula stage (Gallagher *et al*., 1996). In *Taricha*, in contrast, placodal ectoderm explanted at early neurula stages is not yet specified to form the ear (Jacobson, 1963a). In the axolotl, commitment to otic vesicle formation has been tested by grafting otic ectoderm to three different sites: preotic head ectoderm, the ventral body wall, and the limb (Yntema, 1933, 1939; Ginsburg, 1946, 1995). Different results were obtained in each case, highlighting the operational nature of the definition of commitment and the difficulty of arriving at a final analysis. The limb clearly provided the most challenging environment for otic vesicle formation, as it was only seen from donors with four or more somites (stage 20 and older), while sensory areas (hair cells) only formed if ectoderm was grafted after the 7-somite stage (stage 23; Yntema, 1933). Vesicles formed from younger otic ectoderm in grafts to preotic head ectoderm (Yntema, 1939) or to the ventral body wall (Ginsburg, 1946, 1995). Two vesicles often formed in close proximity in the latter grafts, leading the author to suggest that there are two morphogenetic centres in the otic placode that direct the formation of two independent vesicles when the placode is in a heterotopic location (Ginsburg, 1995). A zebrafish mutant, *quadro*, also splits the otic placode into two fields (Malicki *et al*., 1996).

In *Rana*, presumptive otic placode ectoderm grafted to the flank shows commitment to otic vesicle formation, ganglion formation, and sometimes also to sensory patch (hair cell) formation, depending on the species, at the late gastrula/early neural plate stage (Zwilling, 1941; Ginsburg, 1995). In *Triturus*, grafts to the flank showed significant commitment to otic vesicle and sensory patch formation at the 4–6 somite stage (stages 21–22; Ginsburg, 1946, 1995).

Several investigators have cultured prospective otic placode ectoderm from the chick in the presence of serum. Under these conditions, 4–6 somite stage otic ectoderm does not form neurons, while neurons develop from 22–27 somite stage otic vesicles (intermediate stages were not tested; Vogel and Davies, 1993). In other experiments, 7-somite stage otic ectoderm formed neurons (Adam *et al*., 1998) and also rudimentary otic vesicles with localised *lmx1* expression (Giraldez, 1998). 19-somite stage otic placode ectoderm forms hair cells in the presence of serum, although earlier stages were not tested (Adam *et al*., 1998). The presence of serum in these cultures makes them difficult to interpret.

In the chick, specification of otic placode ectoderm towards Pax2 expression is seen at the 5–6 somite stage, while specification towards *BMP7* expression is seen from the 7–8 somite stage; for both genes, specification occurs essentially concomitantly with expression *in vivo* (Groves and Bronner-Fraser, 2000). Commitment of prospective otic placode ectoderm to an otic fate in the chick has been tested by grafting it to four different locations: preotic head ectoderm (at the level of the midbrain or rostral hindbrain), postotic head ectoderm (the prospective nodose placode), the lateral trunk, and the wingbud. As for the axolotl, the wingbud is evidently the most challenging environment for otic placode development. In such grafts, otic vesicles only form when the donor is older than the 10-somite stage, i.e., after the otic placode has thickened (Herbrand *et al*., 1998). Hair cells and some other inner ear cell types are committed in grafts to the wingbud from the 18–22 somite stage (stage 13–14), while complete differentiation of the otic epithelium is seen in vesicles from 30-somite stage embryos (stage 18; Swanson *et al*., 1990). In contrast, in grafts to the preotic region or to the lateral trunk, 5–6 somite stage otic placode ectoderm shows commitment both to otic vesicle formation and to *Pax2* expression, with some commitment to *Nkx5.1* expression (Herbrand *et al*., 1998; Groves and Bronner-Fraser, 2000). In grafts to the nodose placode, 4–6 somite stage otic ectoderm showed no commitment to form vestibular neurons, instead forming nodose neurons (Vogel and Davies, 1993). These results again show that the time of commitment varies depending on the test environment. Eventually, these differences may enable the isolation of permissive or inhibitory factors for the induction that are present in one location but not in another.

Inducing Tissues

Location and duration. There is considerable evidence to support the existence of ear-inducing signals from both mesendoderm and the neural tube; this is given in detail in the following sections. In the following paragraphs, we review the available evidence on the location and duration

TABLE 6
Molecular Markers and Early Events in Otic Placode Development

		Zebrafish		
8 h	Late gastrula	*Dlx3; Dlx7*	Transcription factors	Akimenko *et al.*, 1994; Ellies *et al.*, 1997
8.5 h	Late gastrula (80% epiboly)	*Pax8*	Transcription factor	Pfeffer *et al.*, 1998
10 h	End of gastrulation	*eya1*	Transcription cofactor	Sahly *et al.*, 1999
10 h	End of gastrulation	*six4.1*	Transcription factor	Kobayashi *et al.*, 2000
11 h	3 somites	*Pax2.1; Tbx2*	Transcription factors	Krauss *et al.*, 1991; Ruvinsky *et al.*, 2000
12 h	6 somites	*Pax2.2*	Transcription factor	Pfeffer *et al.*, 1998
12 h	6 somites	*scyba*	CXC-type chemokine	Long *et al.*, 2000
13 h	8 somites	Islet-1	Transcription factor	Korzh *et al.*, 1993
14 h	10 somites	Placodes visible		Haddon and Lewis, 1996
14 h	10 somites	*deltaA; deltaB; deltaD; serrateB*	Notch ligands	Haddon *et al.*, 1998
16 h	14 somites	*Nkx5.1*	Transcription factor	Adamska *et al.*, 2000
16.5 h	15 somites	*Kal1.1*	Extracellular matrix glycoprotein	Ardouin *et al.*, 2000
18.5 h	19 somites	*Fgf8*	Ligand	Reifers *et al.*, 1998
19 h	21 somites	Otoliths form		Haddon and Lewis, 1996
22 h	26 somites	Neuroblasts delaminate		Haddon and Lewis, 1996
24 h	30 somites	Hair cells differentiate		Haddon and Lewis, 1996
		Xenopus		
Stage 10.5	Early gastrula	*Sox2; Sox3*	Transcription factors	Mizuseki *et al.*, 1998; Penzel *et al.*, 1997
Stage 12/13	Late gastrula/ neural plate	*Pax8*	Transcription factor	Heller and Brändli, 1999
Stage 14	Neural plate	*Tbx2*	Transcription factor	Takabatake *et al.*, 2000
Stage 21	8–9 somites	Common dorsolateral placode area visible (otic + lateral line)		Schlosser and Northcutt, 2000
Stage 21	8–9 somites	*Pax2*	Transcription factor	Heller and Brändli, 1997
Stage 21	8–9 somites	*Delta1*	Notch ligand	Schlosser and Northcutt, 2000
Stage 22	9–10 somites	*groucho-related genes 4* and *5*	Transcriptional repressors (interact with Wnt effector XTcf3)	Roose *et al.*, 1998; Molenaar *et al.*, 2000
Stage 22	9–10 somites	p21-activated kinase 1	Cytoskeletal effector protein	Islam *et al.*, 2000
Stage 22–23	9–12 somites	Placodes invaginate		Schlosser and Northcutt, 2000
Stage 27	19 somites	Otic vesicle formed		Schlosser and Northcutt, 2000
Stage 28	20–22 somites	Neuroblasts delaminate		Schlosser and Northcutt, 2000
		Chick		
Stage 3	Gastrula	*Sox3*	Transcription factor	Rex *et al.*, 1997; Dr. Paul Scotting, personal communication
Stage 6	Neurula	*Six4*	Transcription factor	Esteve and Bovolenta, 1999
Stage 8	4–5 somites	Large region of thickened ectoderm visible		Waddington, 1937
Stage 8	4–5 somites	*Pax2*	Transcription factor	Groves and Bronner-Fraser, 2000
Stage 8	5 somites	*lmx1*	Transcription factor	Giraldez, 1998
Stage 9	6 somites	*sprouty2*	FGF antagonist	Chambers and Mason, 2000
Stage 9	7 somites	*frizzled1*	Wnt receptor	Stark *et al.*, 2000
Stage 9	7 somites	*Bmp7*	Ligand	Groves and Bronner-Fraser, 2000
Stage 9	7–8 somites	Placodes thicken		Romanoff, 1960
≤Stage 9	≤8 somites	*frzb1*	Wnt antagonist	Duprez *et al.*, 1999; Baranski *et al.*, 2000

TABLE 6—*Continued*

		Chick		
Stage 10	9–10 somites	*Notch1*	Receptor	Groves and Bronner-Fraser, 2000
Stage 10	10 somites	*Nkx5.1; Gbx2; Dlx3*	Transcription factors	Herbrand *et al.*, 1998; Hidalgo-Sánchez *et al.*, 2000; Pera and Kessel, 1999
Stage 10	10 somites	*Fgf8*	Ligand	Hidalgo-Sánchez *et al.*, 2000
≤Stage 11	≤13 somites	*Wnt5a*	Ligand	Baranski *et al.*, 2000
Stage 11	13 somites	Placodes invaginate		Romanoff, 1960
Stage 11	13 somites	*Gata3*	Transcription factor	Sheng and Stern, 1999
Stage 11	13 somites	*Bmp4; Delta1; Serrate1*	Ligands	Wu and Oh, 1996
Stage 11	13 somites	*frizzled2*	Wnt receptor	Stark *et al.*, 2000
Stage 11	13 somites	*SFrp1; SFrp2*	Wnt antagonists	Esteve *et al.*, 2000; Ladher *et al.*, 2000
Stage 12	16 somites	*frizzled7*	Wnt receptor	Stark *et al.*, 2000
Stage 12	16 somites	*lunatic fringe*	Notch modifying enzyme	Cole *et al.*, 2000
Stage 12	16 somites	*NCAM*	Cell adhesion molecule	Thiery *et al.*, 1982
≤Stage 14	≤22 somites	*SOHo1*	Transcription factor	Deitcher *et al.*, 1994
Stage 14	22 somites	Neuroblasts delaminate		Adam *et al.*, 1998
Stage 15	24–27 somites	*Tlx1; Tlx3* (ganglion)	Transcription factors	Logan *et al.*, 1998
≤Stage 17	≤29–32 somites	*Dlx5*	Transcription factor	Pera *et al.*, 1999
Stage 17	29–32 somites	*Brn3.0*	Transcription factor	Artinger *et al.*, 1998
		Mouse		
E8	Presomitic	*Pax8*	Transcription factor	Pfeffer *et al.*, 1998
E8	1–7 somites	*BF1 (Foxg1); Dlx5*	Transcription factors	Hatini *et al.*, 1999; Yang *et al.*, 1998; Depew *et al.*, 1999
E8	1–7 somites	*Fgf3; Bmp7*	Ligands	McKay *et al.*, 1996; Solloway and Robertson, 1999
E8.5	8–10 somites	*Sox2*	Transcription factor	Wood and Episkopou, 1999
E8.5	9–11 somites	Placodes visible		Kaufman, 1992
E8.5	10–12 somites	c-*kit*	Stem cell factor receptor	Orr-Urtreger *et al.*, 1990
E8.5	10–12 somites	*Hmx3 (Nkx5.1)*	Transcription factor	Rinkwitz-Brandt *et al.*, 1995; Wang *et al.*, 1998
E8.5	12 somites	*Notch1*	Receptor	Reaume *et al.*, 1992
E9	13–20 somites	Placodes invaginate; neuroblasts delaminate		Kaufman, 1992; Ma *et al.*, 1998
E9	13–20 somites	*ngn1; NeuroD*	Transcription factors	Ma *et al.*, 1998
E9	13–20 somites	Activin receptor I	Receptor	Yoshikawa *et al.*, 2000
E9	13–20 somites	*Bmp4; Delta1*	Ligands	Morsli *et al.*, 1998; Ma *et al.*, 1998; Morrison *et al.*, 1999
E9	13–20 somites	*lunatic fringe*	Notch modifying enzyme	Morsli *et al.*, 1998
E9.5	25–30 somites	Otic vesicles close		Kaufman, 1992
E9.5	21–29 somites	*Pax2; Six1; Hey1; Hey2; Tbx1; Tbx2; HES6*	Transcription factors	Nornes *et al.*, 1990; Oliver *et al.*, 1995b; Leimeister *et al.*, 1999; Chapman *et al.*, 1996; Pissarra *et al.*, 2000; Vasiliauskas and Stern, 2000
E9.5	21–29 somites	*Eya1; Eya4; Dach1*	Transcription cofactors	Xu *et al.*, 1997; Borsani *et al.*, 1999; Caubit *et al.*, 1999; Davis *et al.*, 1999
E9.5	21–29 somites	*RhoB*	Signal transduction	Henderson *et al.*, 2000
E9.5	21–29 somites	*RALDH3*	Retinaldehyde dehydrogenase	Mic *et al.*, 2000
≤E10	≤30–34 somites	*Otx1; Otx2*	Transcription factors	Morsli *et al.*, 1999
≤E10.5	≤35–39 somites	*Gata3*	Transcription factor	George *et al.*, 1994

of otic-inducing activity that does not specifically implicate a particular tissue or tissues.

In the axolotl, experiments in which prospective gill ectoderm is grafted into the otic region of progressively older hosts suggested that ear-inducing activity is lost by the 20-somite stage (stage 32; Yntema, 1950). Interestingly, late gastrula/early neurula gill ectoderm is not competent to respond to ear-inducing activity in older hosts, while late neurula and older gill ectoderm is competent (Yntema, 1950). Yntema proposed that an early mesodermal phase of otic induction is followed by a later neural phase and that the younger gill ectoderm is not competent to respond to the neural phase alone (Yntema, 1950). However, an alternative explanation could be that the otic-inducing activity acts on both prospective otic and gill ectoderm (which lies directly beneath the prospective otic placode) during early neurula stages but is subsequently lost. Hence, younger gill ectoderm grafted to older hosts will not respond, as the signal is no longer present. Older gill ectoderm has already received the otic-inducing signal and will follow an otic fate when grafted to the otic placode. In this regard, it is noteworthy that some genes expressed in the otic placode in the chick show a fairly broad initial distribution, suggesting a response of adjacent nonotic ectoderm to the same signals (Groves and Bronner-Fraser, 2000). Since even the older gill ectoderm will not form otic vesicles when grafted to 20-somite stage hosts, however, additional signals must be necessary for otic induction or maintenance.

In chorioallantoic membrane grafts of early neurula (head process stage) chick blastoderm, the only region that forms otic vesicles and associated ganglia is a transverse strip just rostral to the node (Rawles, 1936). This region also forms hindbrain tissue, heart, and endoderm derivatives (Rawles, 1936). These results show that all the signals necessary for otic vesicle induction and ganglion formation are present at the head process stage in this region of the embryo. In the 3–10 somite stage chick embryo, otic placode marker- and vesicle-inducing activity maps to the axial level of the hindbrain, from rhombomere 2/3 to at least as far caudally as the first two somites (Groves and Bronner-Fraser, 2000). Intriguingly, epithelial vesicles, but not molecular markers of the otic placode, can be induced in more rostral regions (Groves and Bronner-Fraser, 2000). Pax2- and *Notch*-inducing activity is progressively lost after the 11-somite stage, while vesicle-inducing activity *BMP7*- and *Sox3*-inducing activity are maintained at least until the 21-somite stage (see Table 6), showing that different aspects of otic placode differentiation can be uncoupled experimentally (Groves and Bronner-Fraser, 2000).

Although some investigators thought that the migrating neural crest induced the otic placode (Trampusch, 1941), it should be clear from Table 6 that otic placode induction begins long before neural crest cell emigration from the neural tube.

Mesendoderm. Zebrafish mutants that lack prechordal mesoderm show delayed formation of the otic placode and vesicle and later patterning defects; mutants that completely lack cephalic mesendoderm show a longer delay but otic vesicles still form (Mendonsa and Riley, 1999). The extent of the delay correlates with a delayed upregulation of *dlx3* and reduced *pax2.1* expression in the presumptive otic placode (Mendonsa and Riley, 1999). These results support a role for cephalic mesendoderm in the normal induction of the otic placode and also in later patterning events. They also show that other signals, presumably derived from the hindbrain (see next section), are sufficient to rescue otic placode induction.

In trout embryos, the middle (not the most anterior) portion of the archenteron roof (axial mesoderm) of mid-gastrula stage embryos induces brainlike vesicles with adjacent otic vesicles from extraembryonic gastrula ectoderm (Eakin, 1939). In the teleost *Fundulus*, otic vesicles and brain tissue form when germ ring tissue (future mesendoderm), taken either 90 or 180° away from the dorsal lip of the blastopore of young and middle gastrulae, is grafted to the head region of host embryos (Oppenheimer, 1938). This was interpreted as showing that gastrula-stage germ ring (mesendoderm) is competent to form otic vesicles in fish (Oppenheimer, 1938). However, in more recent experiments where labelled zebrafish germ ring tissue was grafted to the animal pole (future forebrain) region, the ectopic otic vesicles and hindbrain tissue are seen to be host-derived and must therefore have been induced by the grafted mesendodermal progenitors (Woo and Fraser, 1997). However, these experiments do not distinguish between direct induction of otic vesicles and secondary induction by neural tissue.

Various experiments on amphibian embryos have shown that mesendoderm can induce otic vesicles in the absence of neural tissue. In *Triton*, when the mesendoderm underlying the prospective hindbrain of early neurula embryos is grafted beneath the ventral ectoderm of early neurula hosts (in which the ventral ectoderm has lost the capacity to form neural tissue), otic vesicles are induced (Kohan, 1944). Similarly, in the experiments described previously in the lateral line section, medial archenteron roof (axial mesendoderm) from early neurula stage axolotl embryos can induce otic vesicles from ventral head ectoderm in the absence of neural induction (Raven and Kloos, 1945). Lateral archenteron roof grafts (nonaxial mesoderm) are also able to induce otic vesicles, but with lower frequency than medial grafts (Raven and Kloos, 1945). In *Ambystoma*, if the prospective hindbrain and ear ectoderm are ablated bilaterally and abdominal ectoderm grafted instead, the underlying mesoderm can induce an otic vesicle on both sides, again showing mesoderm action in the absence of neural tissue (Harrison, 1945). Also in the axolotl, lateral mesoderm from the region bordering the lateral margin of the neural plate is able to induce otic vesicles (Holtfreter, 1933). These experiments show that mesendoderm is able to induce otic vesicles directly, in the absence of neural tissue.

In amphibian embryos, the prospective otic ectoderm overlies the future cardiac mesoderm at the open neural plate stage (Jacobson, 1963a). In the newt, endoderm and cardiac mesoderm together are able very occasionally to

induce an ear from unspecified placodal ectoderm (Jacobson, 1963a). Endoderm together with posterior neural plate is able to induce ears more frequently, while posterior neural plate and fold is able to induce ears in all cases (Jacobson, 1963a).

In the chick, extirpation of the mesendoderm of Hensen's node in neural plate–early neurula stage embryos (stage 4–5) leads either to complete lack of formation of the otic placode or to unilateral loss or delayed development of the right-side placode without affecting the hindbrain (Orts-Llorca and Jimenez-Collado, 1971). The mesoderm invaginating through Hensen's node, which invaginates more quickly on the left side (hence the greater effect of the ablation on the right side of the embryo), contributes to the head mesoderm below the presumptive otic placode (Rosenquist, 1966). These results suggest that head mesendoderm is essential for otic placode induction in the chick (Orts-Llorca and Jimenez-Collado, 1971). If the hindbrain neural fold is ablated at a stage before the otic placode is specified, an ear may nevertheless form, also suggesting an influence of mesoderm on ear induction (Waddington, 1937). Recent molecular evidence from the chick suggests that FGF19 from paraxial mesoderm cooperates with Wnt8c from the hindbrain to induce some otic placode markers and thickening of the placode ectoderm (Ladher *et al.*, 2000; see section on signalling molecules, below).

Neural tube. In zebrafish, hindbrain progenitors, but not fore- or midbrain progenitors, from late gastrula embryos (80% epiboly) can induce ectopic otic vesicles from host ventral ectoderm (Woo and Fraser, 1998). This also suggests a restriction in otic-inducing activity along the neuraxis. Hindbrain progenitors are not able to induce otic vesicles when grafted within presumptive forebrain territory (Woo and Fraser, 1998), although this region is competent to form otic vesicles in response to mesendoderm progenitors (Woo and Fraser, 1997). This suggests a limitation in hindbrain inductive ability relative to mesendodermal inductive ability (Woo and Fraser, 1998). As described above, otic vesicle formation is delayed, but not prevented, in zebrafish mutants lacking prechordal mesoderm or cephalic mesendoderm, suggesting that signals from the hindbrain are sufficient for otic vesicle induction (Mendonsa and Riley, 1999).

In the axolotl, neural plate rotation can reverse the anteroposterior axis of the ear, suggesting an influence on ear patterning (Harrison, 1936). Neural plate-stage prospective hindbrain tissue can induce an incomplete ear and its ganglion when grafted to the flank (Stone, 1931). Neurula-stage hindbrain grafted beneath early neurula stage ventral ectoderm (which has lost the capacity to form neural tissue) will induce otic vesicles (Kohan, 1944). Hindbrain tissue grafted to other regions of the body can induce small, abnormal otic vesicles (Harrison, 1945). However, donor-host labels were not used in all these grafting experiments; hence it is possible that the otic vesicles are derived from contaminating donor ectoderm. If the hindbrain is replaced at the 4-somite stage (stage 21) with either the mesencephalon or the anterior spinal cord, ears form in the normal location but do not visibly differentiate, although the otic ganglion forms in some cases (Detwiler, 1948; Detwiler and van Dyke, 1950). This suggests a requirement for the hindbrain for normal patterning of the ear. However, as the otic placode is committed to form a vesicle at early neurula stages (Ginsburg, 1995), these results do not give any useful information on otic placode induction. In *Taricha*, posterior neural plate and fold induce ears from unspecified (early neurula stage) placodal ectoderm in all cases (Jacobson, 1963a). This may suggest that some otic-inducing activity is widespread along the neuraxis at neural plate/fold stages.

In the chick, ablation of the neural tube before the 7-somite stage abrogates otic vesicle formation and ectodermal *lmx1* expression (see Table 6), suggesting that the neural tube is necessary for some aspects of otic placode morphogenesis (Giraldez, 1998). Both dorsal and ventral regions of the neural tube support otic vesicle formation, but the dorsal neural tube is necessary to maintain *lmx1* expression (Giraldez, 1998). Similarly, in vitamin A-deficient quail embryos that lack posterior rhombomeres, the otic placodes form normally and in an apparently normal location but they fail to invaginate to form vesicles, suggesting an important role for the hindbrain in placode morphogenesis and subsequent development (Maden *et al.*, 1996). These results differ somewhat from those reported by Waddington, in which ears still formed after ablation of the neural tube before the 9-somite stage (Waddington, 1937). Small, badly developed otic vesicles are occasionally seen when hindbrain neural fold is grafted beneath nonotic anterior head ectoderm (Waddington, 1937), although it is possible that these originated from donor ectoderm associated with the graft. After rhombomere rotation or grafting, small ectopic otic vesicles are associated with r4 in rostral head ectoderm (Kuratani and Eichele, 1993; Sechrist *et al.*, 1994). However, ectopic otic vesicles seen after rhombomere rotation using quail donors are all donor-derived (Dr. Andrew Groves, personal communication). Recent molecular evidence from the chick suggests that Wnt8c from the hindbrain cooperates with FGF19 from the paraxial mesoderm to induce some otic placode markers and thickening of the placode ectoderm (Ladher *et al.*, 2000; see section on signalling molecules, below).

In mice that are double mutant for retinoic acid receptor genes α and β, r5 is enlarged, and an enlarged otic pit forms that gives rise to small ectopic otic vesicles, together with a much larger otic vesicle located further rostrally than in wild-type (Dupé *et al.*, 1999). However, it is also possible that there is an effect on the mesoderm in these mice. Conversely, the cochlea and vestibular apparatus are missing in mice double mutant for *Hoxa1* and *Hoxb1*, in which r5 is severely reduced or absent (Gavalas *et al.*, 1998). Again, these results suggest a role for the hindbrain in otic placode morphogenesis and differentiation.

Summary. In summary, there is substantial evidence to support a role for mesendoderm in otic placode induction. Both axial and nonaxial mesendoderm seem to have otic

placode-inducing activity. It is also clear that neural tube-derived signals are involved at least in later aspects of otic placode development, such as otic vesicle formation and morphogenesis. This supports a two-step model of otic placode induction in which initial induction occurs at late gastrula/early neurula stages via signals from mesendoderm, with later signals, probably emanating from the hindbrain, that serve to reinforce the early induction and pattern the ear. Recent molecular evidence from the chick, described below, supports this model (Ladher *et al.*, 2000).

Molecules

For a comprehensive recent review of mutations affecting inner ear development, see Fekete (1999).

Transcription factors. Mice mutant for *Pax2* have severe inner ear defects, lacking the entire cochlea and a large part of the vestibuloacoustic ganglion, although the vestibular apparatus is normal (Favor *et al.*, 1996; Torres *et al.*, 1996). *Pax2* mutation in humans leads to renal-coloboma syndrome, which includes high-frequency hearing loss (reviewed in Eccles and Schimmenti, 1999). *Eya1* mutation in humans leads to branchio-oto-renal syndrome, which includes inner ear defects (Abdelhak *et al.*, 1997), while ear development in *Eya1*-deficient mice arrests at the otic vesicle stage (Xu *et al.*, 1999). *Eya1* is not required for *Pax8* and *Pax2* expression but is required for *Six1* and *Fgf3* expression in the otic vesicle (Xu *et al.*, 1999). In the medaka fish, overexpression of *Sox3* occasionally leads to the formation of ectopic *Pax2*-positive, *Eya1*-positive otic vesicles in trunk ectoderm adjacent to the neural tube (Köster *et al.*, 2000).

Ngn1 is required for neurogenesis within the otic placode: its mutation in mice eliminates the vestibulocochlear ganglion (Ma *et al.*, 1998). *Gata3* mutation in humans leads to deafness in the HDR syndrome (hypoparathyroidism, sensorineural deafness, and renal anomaly; Van Esch *et al.*, 2000). *Otx1* and *Otx2* have roles in the later morphogenesis of the inner ear (Morsli *et al.*, 1999), as does *Hmx3* (*Nkx5.1*; Wang *et al.*, 1998). The bHLH gene *Math1*, a homologue of the *Drosophila atonal* gene, is expressed in the inner ear sensory epithelia and is essential for hair cell differentiation (Bermingham *et al.*, 1999).

Signalling molecules. FGF3 is expressed at certain stages in r5 and r6, exactly adjacent to the future otocyst, in *Xenopus*, chick, and mice (Wilkinson *et al.*, 1988; Tannahill *et al.*, 1992; Mahmood *et al.*, 1995b; Lombardo *et al.*, 1998). Chick *sprouty2*, a homologue of a *Drosophila* intracellular antagonist of EGF and FGF signalling (Casci *et al.*, 1999), is expressed in the otic placode and mesenchyme from the 6-somite stage, precisely adjacent to the *Fgf3* expression domain within the hindbrain (Mahmood *et al.*, 1995b). Some evidence from antisense oligonucleotide and blocking antibody expression in chick explants *in vitro* suggested an involvement of FGF3 in otocyst formation (Represa *et al.*, 1991); however, the experimental design and controls of these experiments have been questioned, and they could not be repeated *in vivo* (Mahmood *et al.*, 1995b). FGF2 and FGF3 will both induce small otic vesicles from *Xenopus* trunk ectoderm if FGF-soaked beads are implanted into presomitic mesoderm at early neural plate stages (Lombardo and Slack, 1998; Lombardo *et al.*, 1998). Similarly, overexpression of FGF3 (but not FGF2) in chick embryos leads to ectopic otic vesicle formation in the head and the vagal region (Vendrell *et al.*, 2000). However, the temporal pattern of *Fgf3* expression in the hindbrain does not always correlate well with otic placode induction (Tannahill *et al.*, 1992; Mahmood *et al.*, 1995b, 1996; McKay *et al.*, 1996). Furthermore, otic vesicles form normally in mice with a targeted disruption of the *Fgf3* gene (Mansour *et al.*, 1993). Similarly, otic vesicles form normally in the *Fgf3*-deficient *kreisler* mutation, which lack r5 (Deol, 1964; Frohman *et al.*, 1993; McKay *et al.*, 1994, 1996; Manzanares *et al.*, 1999). Instead, these mice show defects in late stages of otic morphogenesis and differentiation, suggesting that FGF3 is involved in patterning the otic vesicle, rather than in otic vesicle induction.

In zebrafish, inactivation of the *Fgf8* gene by the *acerebellar* mutation leads to a reduction of both *Pax2.1* and *Nkx5.1* expression in the otic placode, although both the otocyst and vestibuloacoustic ganglion still form (Reifers *et al.*, 1998; Adamska *et al.*, 2000).

Recent evidence from the chick implicates a different FGF family member, *Fgf19*, together with *Wnt8c*, in otic placode induction (Ladher *et al.*, 2000). *Fgf19* is transiently expressed in a restricted area of paraxial mesoderm adjacent to the hindbrain from late neurula (stage 6) to the 7-somite stage (stage 9) and transiently in the hindbrain itself from the 6–9 somite stage (stage 9− to 9+; Ladher *et al.*, 2000). The *Fgf19*-positive region colocalises with the otic placode along the rostrocaudal axis, but is not initially subjacent to prospective otic placode ectoderm, which lies lateral to the neural plate at the 1-somite stage (stage 7). By about the 10-somite stage it is in close proximity to the placode (Ladher *et al.*, 2000), although this is well after initial expression of early molecular markers of otic placode ectoderm such as Pax2, first seen at the 4-somite stage (Groves and Bronner-Fraser, 2000). *Fgf19*-expressing mesoderm from 1-somite stage embryos can induce the otic placode markers *Pax2*, *Dlx5*, *Nkx5.1*, the otic cup marker *SOHo1* (Deitcher *et al.*, 1994), and a hair cell antigen (Richardson *et al.*, 1990; Bartolami *et al.*, 1991) in neural ectoderm from neurula (stage 5) embryos (Ladher *et al.*, 200). The use of neural rather than nonneural ectoderm as the responding tissue in this assay means that it is not possible to conclude that the mesodermal signal alone is sufficient for otic induction. Further, some of the molecular markers (*Pax2*, *Dlx5*, *SOHo1*) are expressed in the central nervous system at some stage, making their induction rather hard to interpret. However, chick *Nkx5.1* and the hair cell antigen are specific to the otic placode. Human FGF19 can induce these markers, but not hair cells, in neural ectoderm from neurula (stage 5) or 1-somite stage embryos, but has no effect on 1-somite stage presumptive otic placode ectoderm, suggest-

ing that an interaction with a neural signal is required (Ladher *et al.*, 2000). *Wnt8c* is specifically expressed at the 1-somite stage in the neural plate overlying *Fgf19*-expressing mesoderm, and it is expressed in the caudal hindbrain until the 10-somite stage (Ladher *et al.*, 2000). FGF19 can induce *Wnt8c* in neural tissue from neurula embryos (stage 5), but not in presumptive otic ectoderm from 1-somite stage embryos, suggesting that FGF19 may initiate *Wnt8c* expression in the neural plate (Ladher *et al.*, 2000). Wnt8c alone can induce *Fgf3* from prospective otic placode ectoderm from 1-somite stage embryos, while Wnt8c and FGF19 in combination induce *Pax2*, *Dlx5*, *Nkx5.1*, and *SOHo* expression and thickening of the ectoderm (Ladher *et al.*, 2000). Hair cells are not induced, however, suggesting that additional signals are required for cell type differentiation (Ladher *et al.*, 2000). These results suggest a model in which mesodermal FGF19 induces *Wnt8c* expression in the neural plate; these two signals then cooperate to induce some otic placode marker genes and also otic placode thickening in prospective otic placode ectoderm.

Enlarged and ectopic otic vesicles form in mice that are double mutant for retinoic acid receptor genes α and β, in which r5 is enlarged (Dupé *et al.*, 1999). Supernumerary otic vesicles are also found in retinoid-deficient rat embryos, and it was suggested that postotic neuroepithelium in these embryos retains a more anterior-like fate, leading to induction of otic vesicles from more caudal ectoderm than in normal development (White *et al.*, 1998). Conversely, the otic placodes apparently form normally in vitamin A-deficient quail embryos that lack detectable retinoids (and posterior rhombomeres), although they fail to invaginate (Maden *et al.*, 1996). RALDH3, a retinaldehyde dehydrogenase that catalyses retinoic acid formation, is expressed in the mouse otic vesicle at E9.5 (Mic *et al.*, 2000), while retinoic acid treatment suppresses cell proliferation and induces premature differentiation of the otic vesicle in the chick (Represa *et al.*, 1990). These data may suggest a later role for retinoic acid in otic vesicle morphogenesis and differentiation, although further investigation is required.

Summary

Competence to form the otic vesicle is widespread at gastrula and early neurula stages, but is lost from trunk ectoderm by midneurula stages in the axolotl. Limb ectoderm retains otic competence until much later, however. In the chick, competence to form the otic vesicle is lost from preotic and trunk ectoderm by the 6-somite stage, while competence to express some molecular markers is retained a little longer. In *Xenopus*, otic placode ectoderm is specified to form vesicles by the early neurula stage, while in the axolotl, it is committed to form vesicles by the 4-somite stage (in grafts to the limb) and to form hair cells by the 7-somite stage. In the chick, otic placode ectoderm is specified to express some early molecular markers from the 5–6 somite stage. In grafts to the wingbud, it is committed to form otic vesicles from the 10-somite stage and to form hair cells by the 18-somite stage. Mesendoderm is necessary and sufficient for otic placode induction, while the neural tube (hindbrain) may be sufficient and is clearly involved in morphogenesis and patterning of the otic placode. A two-step model for otic placode formation suggests that initial induction by mesendoderm during gastrula/early neurula stages is reinforced and maintained by signals from the hindbrain. Members of the FGF family, including FGF3, may be involved in aspects of otic placode induction and development. In the chick, a model has emerged in which FGF19, expressed in paraxial mesoderm at the axial level of the otic placode from neurula stages, induces Wnt8c in the prospective caudal hindbrain. These two signals then cooperate to induce several otic placode markers and placodal thickening in prospective otic placode ectoderm. However, they are clearly not sufficient for all aspects of otic placode differentiation and morphogenesis, and much work remains to be done to complete a molecular picture of otic placode induction.

EPIBRANCHIAL PLACODES

The epibranchial placodes form in a rostrocaudal sequence, immediately dorsocaudal to the area of branchial cleft formation, shortly before (in *Xenopus*; Schlosser and Northcutt, 2000) or concomitant with contact between the outpocketing endoderm of the pharyngeal pouches and the overlying ectoderm. The area of ectoderm that thickens near the region of pharyngeal pouch contact is more extensive than the neurogenic region of the placodes in the chick (D'Amico-Martel and Noden, 1983). In the mouse, the entire branchial region is covered with thick ectoderm until the 10-somite stage, when the ectoderm outside the future epibranchial placodes begins to thin, leaving the placodes in relief (Verwoerd and van Oostrom, 1979).

The first epibranchial placode (facial or *geniculate*) forms above the first branchial cleft and gives rise to neurons in the distal (geniculate) ganglion of cranial nerve VII (facial), which primarily innervate taste buds. The second epibranchial placode (glossopharyngeal or *petrosal*) forms above the second branchial cleft and gives rise to neurons in the distal (petrosal) ganglion of cranial nerve IX (glossopharyngeal), which innervate taste buds, the heart, and other visceral organs. The third epibranchial placode (vagal or *nodose*) forms above the third branchial cleft and gives rise to neurons in the distal (nodose) ganglion of cranial nerve X (vagal), which primarily innervates the heart and other visceral organs (see Zhuo *et al.*, 1997). In fish, amphibia, and birds, additional vagal epibranchial placodes form above more posterior branchial clefts and contribute neurons to the nodose ganglion or ganglia (Landacre, 1912; Yntema, 1937, 1943; Schlosser and Northcutt, 2000; D'Amico-Martel and Noden, 1983). Six vagal epibranchial placodes have been described in the lamprey (Damas, 1951).

Although distal (placode-derived) and proximal (neural

TABLE 7
Molecular Markers and Early Events in Epibranchial Placode Development

		Xenopus		
Stage 10.5	Early gastrula	*Sox2; Sox3*	Transcription factors	Mizuseki *et al.*, 1998; Penzel *et al.*, 1997
Stage 22	9–10 somites	Facial/geniculate placodes visible; *ngnr1*	Transcription factor	Schlosser and Northcutt, 2000
Stage 23	12 somites	Glossopharyngeal/petrosal placodes visible; *ngnr1*	Transcription factor	Schlosser and Northcutt, 2000
Stage 24	15 somites	1st and 2nd pharyngeal pouches contact ectoderm of facial and glossopharyngeal placodes respectively; neuroblasts delaminate; *NeuroD; Delta1*	Transcription factor; Notch ligand	Schlosser and Northcutt, 2000
Stage 24	15 somites	1st vagal/nodose placode visible; neuroblasts delaminate; *ngnr1; NeuroD; Delta1*	Transcription factors; Notch ligand	Schlosser and Northcutt, 2000
Stage 27	19 somites	3rd pharyngeal pouch contacts 1st vagal placode ectoderm; 2nd vagal placode visible; *ngnr1; NeuroD; Delta1*	Transcription factors; Notch ligand	Schlosser and Northcutt, 2000
Stage 32	26 somites	4th pharyngeal pouch contacts 2nd vagal placode ectoderm; 3rd vagal placode visible; *ngnr1; NeuroD; Delta1*	Transcription factors; Notch ligand	Schlosser and Northcutt, 2000
Stage 33/34	32 somites	5th pharyngeal pouch contacts 3rd vagal placode ectoderm		Schlosser and Northcutt, 2000
Stage 35/36	36 somites	*cadherin6* (all vagal placodes)	Adhesion molecule	David and Wedlich, 2000
		Chick		
Stage 3	Gastrula	*Sox3*	Transcription factor	Rex *et al.*, 1997; Dr. Paul Scotting, personal communication
Stage 6	Neurula	*Six4* (all)	Transcription factor	Esteve and Bovolenta, 1999
Stage 9	7 somites	*c-ret*	GDNF receptor	Homma *et al.*, 2000
Stage 9	7 somites	*Sox3*	Transcription factor	Dr. Paul Scotting, personal communication
Stage 10	10 somites	*Pax2*	Transcription factor	Hidalgo-Sánchez *et al.*, 2000
Stage 10	10 somites	*Fgf8*	Ligand	Hidalgo-Sánchez *et al.*, 2000
Stage 12–13	17–18 somites	Butyrylcholinesterase activity (all)		Layer and Kaulich, 1991
≤Stage 12	≤16 somites	*Deltex2* (all)	Notch pathway	Frolova and Beebe, 2000
Stage 13–14	19–22 somites	Neuroblasts delaminate (geniculate)		D'Amico-Martel and Noden, 1983
Stage 14	22 somites	*Serrate1*	Notch ligand	Myat *et al.*, 1996
Stage 15	24–27 somites	*Sfrp2* (all)	Wnt antagonist	Ladher *et al.*, 2000
Stage 15–16	24–28 somites	Neuroblasts delaminate (petrosal)		D'Amico-Martel and Noden, 1983
≤Stage 16	≤26–28 somites	*Phox2a* (all; mainly in neuroblasts)	Transcription factor	Begbie *et al.*, 1999
Stage 16	26–28 somites	*frizzled1* (all)	Wnt receptor	Stark *et al.*, 2000
Stage 16–17	26–32 somites	Neuroblasts delaminate (nodose)		D'Amico-Martel and Noden, 1983
≤Stage 17	≤29–32 somites	*Notch1*	Receptor	Myat *et al.*, 1996
Stage 17	29–32 somites	*Tlx3* (all epibranchial ganglia)	Transcription factor	Logan *et al.*, 1998
Stage 18	30–36 somites	Neural crest cells (prospective glia) penetrate ganglia		Ayer-Le Lièvre and Le Douarin, 1982
Stage 18	30–36 somites	*frizzled7* (all)	Wnt receptor	Stark *et al.*, 2000
Stage 18	30–36 somites	*Frzb* (all)	Wnt antagonist	Ladher *et al.*, 2000
Stage 18	30–36 somites	*GFRα1* (geniculate and petrosal ganglia); *GFRα2* (petrosal ganglion)	GDNF receptors	Homma *et al.*, 2000
≤Stage 19		Hoxb5 (nodose placode and ganglion)	Transcription factor	Kuratani and Wall, 1992; Baker and Bronner-Fraser, 2000
≤Stage 20		Myb (all)	Transcription factor	León *et al.*, 1992

TABLE 7—*Continued*

		Mouse		
E8	1–7 somites	*BF1 (Foxg1)*	Transcription factor	Hatini *et al.*, 1999
E8.5	8–12 somites	c-*kit* (all)	Stem cell factor receptor	Orr-Urtreger *et al.*, 1990
E8.5	11–13 somites	*ngn2* then *ngn1* (geniculate)	Transcription factors	Fode *et al.*, 1998; Ma *et al.*, 1998
E9	13–20 somites	*Notch1* (all)	Receptor	Reaume *et al.*, 1992
E9	17 somites	Neuroblasts delaminate (geniculate); *Math3* then *NeuroD* in neuroblasts	Transcription factors	Nichols, 1986; Fode *et al.*, 1998
E9	18–20 somites	*ngn2* then *ngn1* (petrosal)	Transcription factors	Fode *et al.*, 1998; Ma *et al.*, 1998
E9	18–20 somites	Hoxb5 in postotic pharyngeal ectoderm then restricted to nodose	Transcription factor	Kuratani and Wall, 1992; Wall *et al.*, 1992
E9	20 somites	Neuroblasts delaminate (petrosal); *Math3* then *NeuroD* in neuroblasts	Transcription factors	Nichols, 1986; Fode *et al.*, 1998
≤E9.5	≤20–24 somites	*Phox2a* (all); *Phox2b* (all); *HES6* (all)	Transcription factors	Valarché *et al.*, 1993; Tiveron *et al.*, 1996; Morin *et al.*, 1997; Fode *et al.*, 1998; Pattyn *et al.*, 1997; Pissarra *et al.*, 2000; Vasiliauskas and Stern, 2000
≤E9.5	≤20–24 somites	*Eya1; Eya2* (all)	Transcription cofactors	Xu *et al.*, 1997
≤E9.5	≤20–24 somites	*Delta1* (all)	Notch ligand	Bettenhausen *et al.*, 1995
E9.5	20–24 somites	*sFRP1* (all)	Wnt antagonist	Leimeister *et al.*, 1998
E9.5	20–24 somites	*ngn2* then *ngn1* (nodose)	Transcription factors	Fode *et al.*, 1998; Ma *et al.*, 1998
E9.5	23 somites	Neuroblasts delaminate (nodose); *Math3* then *NeuroD* in neuroblasts	Transcription factors	Nichols, 1986; Fode *et al.*, 1998

crest-derived) ganglia for cranial nerves VII, IX, and X are clearly distinct in amniotes (Fig. 4), they cannot be recognised in *Xenopus*, presumably owing to early fusion of the ganglionic primordia (Schlosser and Northcutt, 2000). Further, throughout *Xenopus* development, the facial ganglion is intimately fused with the anteroventral lateral line ganglion, while the glossopharyngeal ganglion is intimately fused with the middle lateral line ganglion (Schlosser and Northcutt, 2000).

The supporting cells of the epibranchial ganglia in the chick are all derived from neural crest cells (Narayanan and Narayanan, 1980). In the axolotl, some evidence suggests that supporting cells in these ganglia may also derive from placodes (Yntema, 1937, 1943); this may be worth revisiting.

The geniculate placode in nonteleost fish has been described as giving rise to the spiracular organ (Vitali, 1925, 1926; Ranzi, 1926). The spiracular organ, which was lost in teleosts, is usually considered to be a specialised lateral line organ (reviewed in Barry and Bennett, 1989). Similarly, the geniculate placode of birds gives rise to the paratympanic organ in the middle ear (Vitali, 1925; Yntema, 1944; D'Amico-Martel and Noden, 1983). Both the spiracular organ and the paratympanic organ contain neuromasts with typical mechanosensory hair cells; they are thought to be homologous (Vitali, 1925, 1926; von Bartheld, 1990). The spiracular organ is innervated by a branch of the anterior lateral line nerve (Barry and Boord, 1984; Song and Northcutt, 1991), while the paratympanic organ is innervated by the geniculate ganglion and its lateral or "paratympanic" extension (Vitali, 1911; Yntema, 1944; von Bartheld, 1990). The origin of these organs from the geniculate placode is perhaps surprising given their close resemblance to lateral line organs. It is interesting to note that both in fish and in *Xenopus*, the anteroventral lateral line ganglion is intimately fused with the facial (geniculate) ganglion from the beginning of its development and is never recognisable as a distinct structure (Landacre, 1912; Schlosser and Northcutt, 2000). Furthermore, in *Xenopus*, the anteroventral lateral line placode is so closely apposed to the posterior and caudal border of the facial (geniculate) epibranchial placode throughout development that it is difficult to be sure of the exact boundary between them (Schlosser and Northcutt, 2000). It seems possible, therefore, that the spiracular organ (and perhaps the paratympanic organ?) could be derived from a lateral line placode closely associated with the first epibranchial placode; however, this question should be reinvestigated. If the spiracular and paratympanic organs indeed arise from the geniculate placode, then it must be accepted that the geniculate placode is able to form not only sensory neurons, but also mechanosensory hair cells and nonneural epithelium.

Early Events and Markers

A description of early molecular markers and events in epibranchial placode formation in different species is given in Table 7. Little or no published information exists on the epibranchial placodes in zebrafish, although recently, zebrafish transgenic for GFP under the control of an Islet-1 promoter were shown to express GFP in epibranchial ganglia (Higashijima *et al*., 2000).

Competence

In *Pleurodeles*, belly ectoderm from neurula stage embryos can contribute to all epibranchial placode-derived ganglia when substituted for ectoderm lateral to the neural folds in same-stage host embryos (Chibon, 1967). Similarly, in *Xenopus*, belly ectoderm from neural fold embryos (stage 16) is competent to contribute to epibranchial placodes and form epibranchial placode-derived neurons (Dr. Gerhard Schlosser, personal communication). As described in the section on trigeminal placodes, Székely (1959) exchanged the trigeminal and vagal (nodose) placodes between late neurula stage *Triturus* and *Pleurodeles* embryos, using a behavioural response as his assay for neuronal fate. In the larval animal, the vagal nerve mediates the gill depression reflex. When the trigeminal placode was substituted for the vagal placode, there was a normal gill reflex after gill stimulation. Hence, the heterotopically grafted trigeminal placode was competent to form connections appropriate to its new location.

In the chick, both trunk ectoderm from 10–16 somite stage donors and presumptive otic placode ectoderm from 4–7 somite stage donors are competent to form nodose neurons when grafted to the nodose placode region of 4–10 somite stage hosts (Vogel and Davies, 1993). Cells in presumptive ophthalmic trigeminal placode ectoderm that are not yet expressing the opV marker Pax3 are competent to express the epibranchial placode marker Pax2 and to form neurons expressing the epibranchial neuron marker *Phox2a* when grafted to the nodose region (Baker and Bronner-Fraser, 2000). As in Székely's experiments, this suggests that unspecified cells within trigeminal placode ectoderm are competent to contribute to the nodose placode. However, opV ectoderm, unlike otic-level ectoderm, is not competent to express the nodose placode marker Hoxb5 when grafted to the nodose placode region, suggesting that there are some differences between opV and more caudal ectoderm (Baker and Bronner-Fraser, 2000).

Cardiac neural crest cells (at the level of somites 1–3) can form nodose neurons if the nodose placode is ablated in the chick before the 5-somite stage, but not if the ablation is performed at later stages (Harrison *et al*., 1995). The compensation presumably reflects the neuronal differentiation of neural crest cells that would normally form Schwann cells in the nodose ganglion (see, e.g., Wakamatsu *et al*., 2000 for neuron-glial fate determination in dorsal root ganglia). However, it is hard to explain why this does not occur if the placode is ablated after the 5-somite stage. The authors point out that host-derived neurons are present in the nodose ganglia after the ablations, suggesting some level of regeneration from surrounding ectoderm, and that this may somehow be related to the lack of neural crest-derived neurons after later ablations (Harrison *et al*., 1995). Conversely, nodose placode-derived cells can compensate for ablated cardiac neural crest in the formation of cholinergic neurons in cardiac ganglia (parasympathetic) and ectomesenchyme cells in the heart (Kirby, 1988a,b).

Specification/Commitment

As described in the preceding section, Székely (1959) exchanged the trigeminal and vagal (nodose) placodes between late neurula stage *Triturus* and *Pleurodeles* embryos. In the larval animal, the vagal nerve mediates a gill depression reflex. After metamorphosis, the trigeminal nerve mediates the afferent pathway of a corneal (lid-closure) reflex. When the vagal placode is grafted in place of the trigeminal placode, a gill reflex can be evoked in the larval animal by touching the cornea. This result suggests that the graft-derived neurons established functional central connections with vagal motor nuclei, suggesting commitment of late neurula stage vagal ectoderm to a vagal fate. After metamorphosis, however, as described previously, a corneal reflex can be evoked normally, suggesting that normal trigeminal-type connections with the sixth motor nucleus (which mediates the efferent component of the corneal reflex) have also been made. These results show that some but not all cells in the late neurula stage vagal placode are already committed to a vagal-type fate.

In the chick, presumptive otic and nodose placodes at the 4–6 somite stage, which are not yet specified to form neurons, can be interchanged (Vogel and Davies, 1993). In both cases, the grafted ectoderm forms neurons appropriate to its new location, in terms of their hindbrain innervation pattern, neurite growth, and cell survival characteristics (Vogel and Davies, 1993). This shows that nodose placode ectoderm at the 4–6 somite stage is not yet committed to a nodose fate. However, presumptive nodose placode ectoderm is not competent at the 3–6 somite stage to express the ophthalmic trigeminal placode marker Pax3 when grafted at the level of the midbrain (Baker *et al*., 1999). Although these results do not necessarily speak to specification towards a nodose fate, they suggest at least that this ectoderm is already qualitatively different from more rostral head ectoderm, which is competent to express Pax3 (Baker *et al*., 1999).

Inducing Tissues

The epibranchial placodes appear relatively late in development in comparison to the other placodes, and they form in close spatial and temporal association with both migrating neural crest streams (see, e.g., Schlosser and Northcutt, 2000) and pharyngeal pouch endoderm. Neither neural tube nor neural crest are required for formation of the epi-

branchial placodes, however, as shown in ablation experiments in the chick (Yntema, 1944; Begbie *et al*., 1999). In contrast, there is substantial evidence to suggest that pharyngeal pouch endoderm is both necessary and sufficient for epibranchial placode induction.

The close temporal association between epibranchial ganglion formation and pharyngeal endoderm contact with the overlying ectoderm was noted in the axolotl by Landacre, who suggested a possible influence of pharyngeal endoderm on epibranchial placode induction (Landacre, 1931). In lamprey embryos reared in daylight, which have various defects, the epibranchial ganglia are missing when the pharyngeal pouches do not make contact with the ectoderm, suggesting that pharyngeal endoderm is necessary for epibranchial placode induction (Damas, 1951). In mouse embryos double mutant for *Hoxa1* and *Hoxb1*, the second branchial arch does not develop and the petrosal and nodose ganglia are significantly reduced (Gavalas *et al*., 1998). Recent *in vitro* coculture experiments in the chick also showed that pharyngeal endoderm is sufficient to induce epibranchial (*Phox2a*-positive) neurons in nonplacode-forming r3-level cranial ectoderm, although not in trunk ectoderm (Begbie *et al*., 1999). Since trunk ectoderm is competent to form nodose neurons when grafted to the nodose region in the embryo (Vogel and Davies, 1993), trunk ectoderm must receive an additional signal in the cranial environment that allows it to respond to epibranchial placode-inducing signals *in vivo* (Begbie *et al*., 1999).

Molecules

Transcription factors. The basic helix-loop-helix transcription factor *ngn2* is not required for epibranchial placode formation; however, it is essential for delamination, migration, and differentiation of geniculate and petrosal placode-derived cells (although the geniculate ganglion is compensated for later, possibly by neural crest cells; Fode *et al*., 1998). *Ngn1* and *ngn2* expression in the nodose ganglion (Ma *et al*., 1998) might be redundant, as the nodose ganglion is unaffected in either *ngn1* or *ngn2* mutant mice (Fode *et al*., 1998; Ma *et al*., 1998). In all three epibranchial placodes, *ngn2* is required for *Delta-like 1* expression (Fode *et al*., 1998).

In *Phox2a* mutant mice, the petrosal and nodose ganglia are severely atrophic, with the geniculate ganglion being affected to a lesser degree (Morin *et al*., 1997). *Phox2a* is not required for delamination or aggregation of epibranchial placode-derived cells or for their expression of some neuronal markers (Morin *et al*., 1997). However, *Phox2a* is required for transient dopamine β-hydroxylase (DBH; reviewed in Katz *et al*., 1987) expression (Morin *et al*., 1997). It is also required for expression of the glial cell line-derived neurotrophic factor (GDNF) receptor subunit Ret (Morin *et al*., 1997), and hence probably for neuron survival in response to GDNF (Buj-Bello *et al*., 1995; Trupp *et al*., 1995; Moore *et al*., 1996; Morin *et al*., 1997). The epibranchial placode cells that express Phox2a also express the highest levels of *ngn2* transcripts, suggesting that the same signal might be involved in the induction of both genes, coupling the generic and neuronal identity subprograms (Lo *et al*., 1999). *Phox2b* mutant mice show severe apoptotic atrophy of all three epibranchial placode-derived ganglia at E11.5, together with loss of DBH and *Ret* expression, suggesting *Phox2b* is a downstream effector of *Phox2a* (Pattyn *et al*., 1999).

Although Hoxb5 is expressed in the nodose placode and ganglion, it does not seem to be required for formation of nodose neurons (Rancourt *et al*., 1995). In *Eya1* mutant mice, the petrosal ganglia are absent (Xu *et al*., 1999).

Signalling molecules. A major molecular player in the induction of epibranchial placode neurons was recently identified. BMP7, which is expressed in chick pharyngeal pouch endoderm and ectoderm at the site of endoderm/ectoderm contact at least from the 19-somite stage (stage 13), is sufficient to induce *Phox2a*-positive neurons from r3-level ectoderm, which does not normally contribute to placodes (Begbie *et al*., 1999). Also, the BMP7- and activin-inhibitor follistatin (Liem *et al*., 1997) reduces the neuronal induction by pharyngeal endoderm in cocultures, suggesting a requirement for BMP7 in pharyngeal endoderm-mediated induction (Begbie *et al*., 1999). In the mouse, high levels of *Bmp7* and *Bmp5* are seen in branchial arch ectoderm at the 8–12 somite stage (E8.25–E8.5), and branchial arch outgrowth is dramatically reduced in *Bmp5; Bmp7* double mutants (Solloway and Robertson, 1999).

Other potential signals expressed in pharyngeal pouch endoderm in the chick include *Fgf3*, which is expressed here from the 5-somite stage (Mahmood *et al*., 1995b). Increased *Fgf3* levels are observed in the first pouch at the 14-somite stage and in the second and third pouches at later stages (Mahmood *et al*., 1995b). In the mouse, *Fgf8* is expressed in pharyngeal pouch endoderm from the 1–7 somite stage (E8), and by the 21–29 somite stage (E9.5) is localised to the extreme lateral regions of each pouch and the overlying surface ectoderm (Crossley and Martin, 1995; Mahmood *et al*., 1995a). This *Fgf8* expression is unaffected in *Bmp5;Bmp7* double mutants (Solloway and Robertson, 1999). Similar expression of *Fgf8* is seen in the chick, at the anterior endodermal margin of each arch together with the overlying ectoderm, at least as early as the 10-somite stage (Wall and Hogan, 1995; Mahmood *et al*., 1995a; Veitch *et al*., 1999; Hidalgo-Sánchez *et al*., 2000). Neural crest ablation does not affect expression of either *Bmp7* or *Fgf8* in the pharyngeal pouches (Veitch *et al*., 1999).

Chick GDNF, the ligand for c-Ret, is also specifically expressed from the 7-somite stage (stage 9) in the pharyngeal pouch endoderm that will contact the placodes (Homma *et al*., 2000).

Retinoic acid is essential for correct patterning of pharyngeal pouch endoderm (see Wendling *et al*., 2000). In mouse embryos double null for retinoic acid receptors α and β, the third branchial pouch fails to contact the ectoderm and the fourth pouch is very small; the petrosal and nodose nerves

and ganglia are fused in these embryos (Dupé *et al*., 1999). Retinoid deficiency in rat embryos results in the absence of postotic pharyngeal arches and petrosal and nodose ganglia (White *et al*., 1998). Treatment of 2–4 somite stage mouse embryos with a pan-retinoic acid receptor antagonist leads to complete absence of the third and fourth pharyngeal pouches, apoptosis in the petrosal placode, and fusion of petrosal and nodose placodes and hypoplasia of their respective ganglia (Wendling *et al*., 2000). Conversely, retinoic acid treatment of 13–20 somite stage (E9) mouse embryos leads to increased cell death in epibranchial placodes and placode-derived ganglia (Sulik *et al*., 1987).

Bisdiamine treatment during E9 and E10 of embryonic rat development leads to specific loss of the nodose ganglion and also, variably, the petrosal ganglion, without affecting the proximal neural crest-derived ganglia of their respective nerves (Kuratani and Bockman, 1992). Bisdiamine treatment leads to a spectrum of defects similar to those seen in various human syndromes, including DiGeorge syndrome, fetal alcohol syndrome, and retinoic acid embryopathy (see Kuratani and Bockman, 1992).

Nothing is known about the differentiation of specific placodes within the epibranchial placode series, although some molecular markers show differential expression within epibranchial placode-derived ganglia (see Table 7).

Summary

Competence to form epibranchial placodes is widespread at neurula stages and even later; trunk ectoderm in the chick retains competence until at least the 16-somite stage. Some commitment to a vagal fate is seen by late neurula stages in the axolotl, while in the chick commitment must occur later, some time after the 6-somite stage. Pharyngeal pouch endoderm is necessary and sufficient for epibranchial placode induction. In the chick, BMP7 from pharyngeal pouch endoderm is necessary and sufficient for induction of epibranchial placode neurons from head ectoderm in coculture experiments, though not for induction of neurons from trunk ectoderm.

HYPOBRANCHIAL PLACODES

In addition to the epibranchial placodes, two hypobranchial placodes are found in *Xenopus* and the direct-developing frog *Eleutherodactylus coqui* (Schlosser *et al*., 1999; Schlosser and Northcutt, 2000). In *Xenopus*, the more rostral of the two placodes can be recognised at the 32-somite stage (stage 33/34), immediately caudoventral to the second pharyngeal pouch, and so ventral to the petrosal (IX) placode (Schlosser and Northcutt, 2000). The second pharyngeal pouch appears at the 36-somite stage (stage 35/36), immediately caudoventral to the third pharyngeal pouch, and so ventral to the first vagal (X) placode. Also at this stage, *ngnr1* and *NeuroD* are expressed in both hypobranchial placodes. It was not possible to determine whether these placodes are derived from the ventral-most tip of the respective epibranchial placodes or whether they arise *de novo*. They are located immediately adjacent to the developing heart, and cells leaving the placodes maintain *NeuroD* expression and remain close to the pericard until at least stage 40 (Schlosser and Northcutt, 2000). At stage 42, *Six1* expression is also seen in the hypobranchial placodes (Pandur and Moody, 2000). The fate of these cells is unknown. It is also unclear whether hypobranchial placodes have been overlooked in other vertebrates, which seems possible given their late development and ventral location, or whether they are specific to frogs. No information is available on their induction.

GENERAL CONCLUSIONS

Despite their origin from a common anlage that can be defined molecularly and, in some species, morphologically, it is clear from the preceding sections that different placodes are induced by different mechanisms and at different times (see also Graham and Begbie, 2000). Signals from endoderm, mesoderm, and neural tissue have all been implicated in placode induction; the precise combination, however, is often entirely different for each distinct placode. This complexity is not surprising given the diverse fates of the different placodes, from those generating only sensory neurons (trigeminal and epibranchial) to the complex array of cell types formed within the otic vesicle and olfactory epithelium.

It is also evident that much remains to be learned about placode induction. Molecules involved in induction have been identified only for the hypophyseal, lens, otic, and epibranchial placodes, and even here it is certain that many more such players remain to be discovered. A common theme thus far is the involvement of BMPs, probably at multiple steps, and FGFs, while the expression of Wnt signalling pathway genes in multiple placodes strongly suggests that Wnt family members will also be implicated in their induction and development. Different *Pax*/*Six*/*Eya*/*Dach* signalling networks are activated in different placodes. There is certainly no single "placode induction" step, unless it be the establishment of the preplacodal domain within the cranial neural plate border, and even this is likely to be a complex process. Although placode and neural crest derivatives can be induced simultaneously in some experimental situations (e.g., Raven and Kloos, 1945), the relationship between placodes and neural crest cells is still unclear and should be investigated more thoroughly. Furthermore, like the various derivatives of the neural crest, different placodes probably evolved at different times (Baker and Bronner-Fraser, 1997b; Graham and Begbie, 2000; Shimeld and Holland, 2000).

Placode induction can serve to illustrate several general features of embryonic induction (Jacobson and Sater, 1988). The induction of different placodes in different regions results not only from localised inducing activities, but also

from local restrictions in competence and suppressive interactions. Competence to form placodes may be related to the expression of particular genes, such as *Sox* family members (e.g., Köster *et al*., 2000). Although at gastrula stages all ectoderm is competent to form all placodes, this competence is rapidly restricted, probably as the ectoderm becomes specified towards particular fates. However, this is not a general placode restriction, as a particular region of ectoderm at a given time can be competent to form one placode, but not another, again highlighting the differences between different placodes. A large proportion of cranial ectoderm forms placodes (e.g., D'Amico-Martel and Noden, 1983; Schlosser and Northcutt, 2000; Fig. 1A), and a given region can be exposed to multiple placode-inducing signals. The fate ultimately adopted by a particular region depends on its history of cumulative exposure to different signals, including its changing competence as a result of such signals (see, e.g., Baker *et al*., 1999).

As predicted over a decade ago (Gurdon, 1987), the recent explosion in molecular information on placode induction has been made possible primarily by the use of specific molecular markers, rather than complex morphological readouts, in induction assays. The constant stream of newly identified markers for different stages of placode induction not only adds new layers of complexity to the known molecular underpinnings of placode induction and development, but enables us to dissect these processes at a molecular level. It is certain that the next few years will bring great strides in the molecular characterisation of placode induction.

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REFERENCES

Abdelhak, S., Kalatzis, V., Heilig, R., Compain, S., Samson, D., Vincent, C., Weil, D., Cruaud, C., Sahly, I., Leibovici, M., Bitner-Glindzicz, M., Francis, M., Lacombe, D., Vigneron, J., Charachon, R., Boven, K., Bedbeder, P., Van Regemorter, N., Weissenbach, J., and Petit, C. (1997). A human homologue of the *Drosophila eyes absent* gene underlies branchio-oto-renal (BOR) syndrome and identifies a novel gene family. *Nat. Genet.* **15,** 157–164.

Adam, J., Myat, A., Le Roux, I., Eddison, M., Henrique, D., Ish-Horowicz, D., and Lewis, J. (1998). Cell fate choices and the expression of Notch, Delta and Serrate homologues in the chick inner ear: Parallels with *Drosophila* sense-organ development. *Development* **125,** 4645–4654.

Adamska, M., Léger, S., Brand, M., Hadrys, T., Braun, T., and Bober, E. (2000). Inner ear and lateral line expression of a zebrafish *Nkx5–1* gene and its downregulation in the ears of FGF8 mutant, *ace. Mech. Dev.* **97,** 161–165.

Adelmann, H. B. (1925). The development of the neural folds and cranial ganglia of the rat. *J. Comp. Neurol.* **39,** 19–171.

Akimenko, M. A., Ekker, M., Wegner, J., Lin, W., and Westerfield, M. (1994). Combinatorial expression of three zebrafish genes related to *Distal-less*: Part of a homeobox gene code for the head. *J. Neurosci.* **14,** 3475–3486.

Altmann, C. R., Chow, R. L., Lang, R. A., and Hemmati-Brivanlou, A. (1997). Lens induction by Pax-6 in *Xenopus laevis. Dev. Biol.* **185,** 119–123.

Anchan, R. M., Drake, D. P., Haines, C. F., Gerwe, E. A., and LaMantia, A. S. (1997). Disruption of local retinoid-mediated gene expression accompanies abnormal development in the mammalian olfactory pathway. *J. Comp. Neurol.* **379,** 171–184.

Ang, S. L., Conlon, R. A., Jin, O., and Rossant, J. (1994). Positive and negative signals from mesoderm regulate the expression of mouse *Otx2* in ectoderm explants. *Development* **120,** 2979–2989.

Ardouin, O., Legouis, R., Fasano, L., David-Watine, B., Korn, H., Hardelin, J., and Petit, C. (2000). Characterization of the two zebrafish orthologues of the *KAL-1* gene underlying X chromosome-linked Kallmann syndrome. *Mech. Dev.* **90,** 89–94.

Ariëns Kappers, J. (1941). Kopfplakoden bei Wirbeltieren. *Rev. Anat. Embryol. Cell. Biol.* **33,** 370–412.

Artinger, K. B., Fedtsova, N., Rhee, J. M., Bronner-Fraser, M., and Turner, E. (1998). Placodal origin of Brn-3-expressing cranial sensory neurons. *J. Neurobiol.* **36,** 572–585.

Ayer-Le Lièvre, C. S., and Le Douarin, N. M. (1982). The early development of cranial sensory ganglia and the potentialities of their component cells studied in quail-chick chimeras. *Dev. Biol.* **94,** 291–310.

Azuma, N., Hirakiyama, A., Inoue, T., Asaka, A., and Yamada, M. (2000). Mutations of a human homologue of the *Drosophila eyes absent gene* (*EYA1*) detected in patients with congenital cataracts and ocular anterior segment anomalies. *Hum. Mol. Genet.* **9,** 363–366.

Baker, C. V. H., and Bronner-Fraser, M. (1997a). The origins of the neural crest. Part I: Embryonic induction. *Mech. Dev.* **69,** 3–11.

Baker, C. V. H., and Bronner-Fraser, M. (1997b). The origins of the neural crest. Part II: An evolutionary perspective. *Mech. Dev.* **69,** 13–29.

Baker, C. V. H., and Bronner-Fraser, M. (2000). Establishing neuronal identity in vertebrate neurogenic placodes. *Development* **127,** 3045–3056.

Baker, C. V. H., Sharpe, C. R., Torpey, N. P., Heasman, J., and Wylie, C. C. (1995). A *Xenopus* c-*kit*-related receptor tyrosine kinase expressed in migrating stem cells of the lateral line system. *Mech. Dev.* **50,** 217–228.

Baker, C. V. H., Stark, M. R., Marcelle, C., and Bronner-Fraser, M. (1999). Competence, specification and induction of Pax-3 in the trigeminal placode. *Development* **126,** 147–156.

Balkan, W., Klintworth, G. K., Bock, C. B., and Linney, E. (1992). Transgenic mice expressing a constitutively active retinoic acid receptor in the lens exhibit ocular defects. *Dev. Biol.* **151,** 622–625.

Bally-Cuif, L., Dubois, L., and Vincent, A. (1998). Molecular cloning of *Zcoe2*, the zebrafish homolog of *Xenopus Xcoe2* and mouse *EBF-2*, and its expression during primary neurogenesis. *Mech. Dev.* **77,** 85–90.

Bang, A. G., Papalopulu, N., Kintner, C., and Goulding, M. D. (1997). Expression of *Pax-3* is initiated in the early neural plate by

posteriorizing signals produced by the organizer and by posterior non-axial mesoderm. *Development* **124,** 2075–2085.

Bang, A. G., Papalopulu, N., Goulding, M. D., and Kintner, C. (1999). Expression of *Pax-3* in the lateral neural plate is dependent on a *Wnt*-mediated signal from posterior nonaxial mesoderm. *Dev. Biol.* **212,** 366–380.

Barabanov, V. M. (1977).Detection of δ-crystallins in the adenohypophysis of chick embryos. *Dokl. Akad. Nauk. SSSR* **234,** 195–198.

Barabanov, V. M., and Fedtsova, N. G. (1982). The distribution of lens differentiation capacity in the head ectoderm of chick embryos. *Differentiation* **21,** 183–190.

Baranski, M., Berdougo, E., Sandler, J. S., Darnell, D. K., and Burrus, L. W. (2000). The dynamic expression pattern of *frzb-1* suggests multiple roles in chick development. *Dev. Biol.* **217,** 25–41.

Barry, M. A., and Bennett, M. V. L. (1989). Specialised lateral line receptor systems in elasmobranchs: The spiracular organs and vesicles of Savi. *In* "The Mechanosensory Lateral Line. Neurobiology and Evolution" (S. Coombs, P. Görner, and H. Münz, Eds.), pp. 591–606. Springer-Verlag, New York.

Barry, M. A., and Boord, R. L. (1984). The spiracular organ of sharks and skates: Anatomical evidence indicating a mechanoreceptive role. *Science* **226,** 990–992.

Barth, K. A., and Wilson, S. W. (1995). Expression of zebrafish *nk2.2* is influenced by *sonic hedgehog/vertebrate hedgehog-1* and demarcates a zone of neuronal differentiation in the embryonic forebrain. *Development* **121,** 1755–1768.

Bartolami, S., Goodyear, R., and Richardson, G. (1991). Appearance and distribution of the 275 kD hair-cell antigen during development of the avian inner ear. *J. Comp. Neurol.* **314,** 777–788.

Beard, J. (1886). The system of branchial sense organs and their associated ganglia in Ichthyopsida. A contribution to the ancestral history of Vertebrates. *Q. J. Microsc. Sci.* **26,** 95–156.

Beason, R., and Semm, P. (1996). Does the avian ophthalmic nerve carry magnetic navigational information? *J. Exp. Biol.* **199,** 1241–1244.

Beddington, R. S., and Robertson, E. J. (1998). Anterior patterning in mouse. *Trends Genet.* **14,** 277–284.

Beddington, R. S., and Robertson, E. J. (1999). Axis development and early asymmetry in mammals. *Cell* **96,** 195–209.

Begbie, J., Brunet, J.-F., Rubenstein, J. L. R., and Graham, A. (1999). Induction of the epibranchial placodes. *Development* **126,** 895–902.

Bendall, A. J., and Abate-Shen, C. (2000). Roles for Msx and Dlx homeoproteins in vertebrate development. *Gene* **247,** 17–31.

Bermingham, N. A., Hassan, B. A., Price, S. D., Vollrath, M. A., Ben-Arie, N., Eatock, R. A., Bellen, H. J., Lysakowski, A., and Zoghbi, H. Y. (1999). *Math1*: An essential gene for the generation of inner ear hair cells. *Science* **284,** 1837–1841.

Bernier, G., Panitz, F., Zhou, X., Hollemann, T., Gruss, P., and Pieler, T. (2000). Expanded retina territory by midbrain transformation upon overexpression of *Six6* (*Optx2*) in *Xenopus* embryos. *Mech. Dev.* **93,** 59–69.

Bettenhausen, B., Hrabe de Angelis, M., Simon, D., Guénet, J. L., and Gossler, A. (1995). Transient and restricted expression during mouse embryogenesis of *Dll1*, a murine gene closely related to *Drosophila Delta*. *Development* **121,** 2407–2418.

Blaxter, J. H. S. (1987). Structure and development of the lateral line. *Biol. Rev.* **62,** 471–514.

Blixt, A., Mahlapuu, M., Aitola, M., Pelto-Huikko, M., Enerback, S., and Carlsson, P. (2000). A forkhead gene, FoxE3, is essential for lens epithelial proliferation and closure of the lens vesicle. *Genes Dev.* **14,** 245–254.

Blount, R. F. (1930). The implantation of additional hypophyseal rudiments in urodele embryos. *Proc. Natl. Acad. Sci. USA* **16,** 218–222.

Blount, R. F. (1932). Transplantation and extirpation of the pituitary rudiment and the effects upon pigmentation in the urodele embryo. *J. Exp. Zool.* **63,** 113–141.

Bodznick, D. (1989). Comparisons between electrosensory and mechanosensory lateral line systems. *In* "The Mechanosensory Lateral Line. Neurobiology and Evolution" (S. Coombs, P. Görner, and H. Münz, Eds.), pp. 655–678. Springer-Verlag, New York.

Bond, C. E. (1996). "Biology of Fishes." Saunders College Publishing, New York.

Bonstein, L., Elias, S., and Franks, D. (1998). Paraxial-fated mesoderm is required for neural crest induction in *Xenopus* embryos. *Dev. Biol.* **193,** 156–168.

Bordzilovskaya, N. P., Dettlaff, T. A., Duhon, S. T., and Malacinski, G. M. (1989). Developmental-stage series of axolotl embryos. *In* "Developmental Biology of the Axolotl" (J. B. Armstrong and G. M. Malacinski, Eds.), pp. 176–186. Oxford Univ. Press, Oxford, UK.

Borsani, G., DeGrandi, A., Ballabio, A., Bulfone, A., Bernard, L., Banfi, S., Gattuso, C., Mariani, M., Dixon, M., Donnai, D., Metcalfe, K., Winter, R., Robertson, M., Axton, R., Brown, A., van Heyningen, V., and Hanson, I. (1999). EYA4, a novel vertebrate gene related to *Drosophila eyes absent*. *Hum. Mol. Genet.* **8,** 11–23.

Bosco, L., Venturini, G., and Willems, D. (1997). In vitro lens transdifferentiation of *Xenopus laevis* outer cornea induced by fibroblast growth factor (FGF). *Development* **124,** 421–428.

Bouwmeester, T., and Leyns, L. (1997). Vertebrate head induction by anterior primitive endoderm. *Bioessays* **19,** 855–863.

Bovolenta, P., Mallamaci, A., Puelles, L., and Boncinelli, E. (1998). Expression pattern of *cSix3*, a member of the Six/sine oculis family of transcription factors. *Mech. Dev.* **70,** 201–203.

Brachet, A. (1914). Sur l'origine des ganglions du trijumeau chez *Chrysemys marginata* (Syn. *Chr. picta* Schn.). *Ann. Soc. R. Zool. Malacol. Belg.* **48,** 31–49.

Braun, C. B. (1996). The sensory biology of the living jawless fishes: A phylogenetic assessment. *Brain Behav. Evol.* **48,** 262–276.

Brewster, R., and Dahmane, N. (1999). Getting a-head of the organizer: Anterior–posterior patterning of the forebrain. *Bioessays* **21,** 631–636.

Buck, L. B. (2000). The molecular architecture of odor and pheromone sensing in mammals. *Cell* **100,** 611–618.

Budelmann, B.-U. (1989). Hydrodynamic receptor systems in invertebrates. *In* "The Mechanosensory Lateral Line. Neurobiology and Evolution" (S. Coombs, P. Görner, and H. Münz, Eds.), pp. 605–631. Springer-Verlag, New York.

Buj-Bello, A., Buchman, V. L., Horton, A., Rosenthal, A., and Davies, A. M. (1995). GDNF is an age-specific survival factor for sensory and autonomic neurons. *Neuron* **15,** 821–828.

Bullock, T. H. (1982). Electroreception. *Ann. Rev. Neurosci.* **5,** 121–170.

Burch, A. B. (1938). Suppression of *pars intermedia* of pituitary body in *Hyla regilla* by operations upon the gastrula. *Proc. Soc. Exp. Biol. Med.* **38,** 608–610.

Burr, H. S. (1924a). Hyperplasia in the brain of *Amblystoma*. *Proc. Soc. Exp. Biol. NY* **21,** 472–474.

Burr, H. S. (1924b). Some experiments on the transplantation of the olfactory placode in *Amblystoma*. *J. Comp. Neurol.* **37,** 455–479.

Callaerts, P., Halder, G., and Gehring, W. J. (1997). PAX-6 in development and evolution. *Annu. Rev. Neurosci.* **20,** 483–532.

Carpenter, E. (1937). The head pattern in *Amblystoma* studied by vital staining and transplantation methods. *J. Exp. Zool.* **75,** 103–129.

Carr, V. M., and Simpson, S. B., Jr. (1978). Proliferative and degenerative events in the early development of chick dorsal root ganglia. I. Normal development. *J. Comp. Neurol.* **182,** 727–739.

Casci, T., Vinós, J., and Freeman, M. (1999). Sprouty, an intracellular inhibitor of Ras signalling. *Cell* **96,** 655–665.

Cau, E., Gradwohl, G., Fode, C., and Guillemot, F. (1997). Mash1 activates a cascade of bHLH regulators in olfactory neuron progenitors. *Development* **124,** 1611–1621.

Cau, E., Gradwohl, G., Casarosa, S., Kageyama, R., and Guillemot, F. (2000). *Hes* genes regulate sequential stages of neurogenesis in the olfactory epithelium. *Development* **127,** 2323–2332.

Caubit, X., Thangarajah, R., Theil, T., Wirth, J., Nothwang, H. G., Ruther, U., and Krauss, S. (1999). Mouse Dac, a novel nuclear factor with homology to *Drosophila* dachshund shows a dynamic expression in the neural crest, the eye, the neocortex, and the limb bud. *Dev. Dyn.* **214,** 66–80.

Chambers, D., and Mason, I. (2000). Expression of *sprouty2* during early development of the chick embryo is coincident with known sites of FGF signalling. *Mech. Dev.* **91,** 361–364.

Chang, C., and Hemmati-Brivanlou, A. (1998). Neural crest induction by *Xwnt7B* in *Xenopus*. *Dev. Biol.* **194,** 129–134.

Chapman, D. L., Garvey, N., Hancock, S., Alexiou, M., Agulnik, S. I., Gibson-Brown, J. J., Cebra-Thomas, J., Bollag, R. J., Silver, L. M., and Papaioannou, V. E. (1996). Expression of the T-box family genes, *Tbx1-Tbx5*, during early mouse development. *Dev. Dyn.* **206,** 379–390.

Chen, R., Amoui, M., Zhang, Z., and Mardon, G. (1997). Dachshund and eyes absent proteins form a complex and function synergistically to induce ectopic eye development in *Drosophila*. *Cell* **91,** 893–903.

Chen, R., Halder, G., Zhang, Z., and Mardon, G. (1999). Signalling by the TGFb homolog *decapentaplegic* functions reiteratively within the network of genes controlling retinal cell fate determination in *Drosophila*. *Development* **126,** 935–943.

Chibon, P. (1967). Marquage nucléaire par la thymidine tritiée des dérivés de la crête neurale chez l'Amphibien Urodèle *Pleurodeles waltlii* Michah. *J. Embryol. Exp. Morphol.* **18,** 343–358.

Chitnis, A., Henrique, D., Lewis, J., Ish-Horowicz, D., and Kintner, C. (1995). Primary neurogenesis in *Xenopus* embryos regulated by a homologue of the *Drosophila* neurogenic gene *Delta*. *Nature* **375,** 761–766.

Chow, R. L., Altmann, C. R., Lang, R. A., and Hemmati-Brivanlou, A. (1999). Pax6 induces ectopic eyes in a vertebrate. *Development* **126,** 4213–4222.

Chuah, M. I., and Au, C. (1991). Olfactory Schwann cells are derived from precursor cells in the olfactory epithelium. *J. Neurosci. Res.* **29,** 172–180.

Clement, J. H., Fettes, P., Knochel, S., Lef, J., and Knochel, W. (1995). Bone morphogenetic protein 2 in the early development of *Xenopus laevis*. *Mech. Dev.* **52,** 357–370.

Cole, L. K., Le Roux, I., Nunes, F., Laufer, E., Lewis, J., and Wu, D. K. (2000). Sensory organ generation in the chicken inner ear: Contributions of *bone morphogenetic protein 4*, *serrate1*, and *lunatic fringe*. *J. Comp. Neurol.* **424,** 509–520.

Collazo, A., Fraser, S. E., and Mabee, P. M. (1994). A dual embryonic origin for vertebrate mechanoreceptors. *Science* **264,** 426–430.

Collignon, J., Sockanathan, S., Hacker, A., Cohen-Tannoudji, M., Norris, D., Rastan, S., Stevanovic, M., Goodfellow, P. N., and Lovell-Badge, R. (1996). A comparison of the properties of *Sox-3* with *Sry* and two related genes *Sox-1* and *Sox-2*. *Development* **122,** 509–520.

Collinson, J. M., Hill, R. E., and West, J. D. (2000). Different roles for *Pax6* in the optic vesicle and facial epithelium mediate early morphogenesis of the murine eye. *Development* **127,** 945–956.

Coombs, S., Görner, P., and Münz, H. (1989). "The Mechanosensory Lateral Line. Neurobiology and Evolution." Springer-Verlag, New York.

Corsin, J. (1971). Influence des placodes olfactives et des ébauches optiques sur la morphogenèse du squelette crânien chez *Pleurodeles waltlii* Michah. *Ann. Embryol. Morphol.* **1,** 41–48.

Couly, G. F., and Le Douarin, N. M. (1985). Mapping of the early neural primordium in quail-chick chimeras I. *Dev. Biol.* **110,** 422–439.

Couly, G. F., and Le Douarin, N. M. (1987). Mapping of the early neural primordium in quail-chick chimeras II. *Dev. Biol.* **120,** 198–214.

Couly, G., and Le Douarin, N. M. (1990). Head morphogenesis in embryonic avian chimeras: Evidence for a segmental pattern in the ectoderm corresponding to the neuromeres. *Development* **108,** 543–558.

Covell, D. A., and Noden, D. M. (1989). Embryonic development of the chick primary trigeminal sensory-motor complex. *J. Comp. Neurol.* **286,** 488–503.

Crossley, P. H., and Martin, G. R. (1995). The mouse *Fgf8* gene encodes a family of polypeptides and is expressed in regions that direct outgrowth and patterning in the developing embryo. *Development* **121,** 439–451.

Cruz, A., and Green, B. G. (2000). Thermal stimulation of taste. *Nature* **403,** 889–892.

Curtiss, J., and Mlodzik, M. (2000). Morphogenetic furrow initiation and progression during eye development in *Drosophila*: The roles of *decapentaplegic*, *hedgehog* and *eyes absent*. *Development* **127,** 1325–1336.

Cvekl, A., and Piatigorsky, J. (1996). Lens development and crystallin gene expression: Many roles for Pax-6. *Bioessays* **18,** 621–630.

Czerny, T., Halder, G., Kloter, U., Souabni, A., Gehring, W. J., and Busslinger, M. (1999). *twin of eyeless*, a second *Pax-6* gene of *Drosophila*, acts upstream of *eyeless* in the control of eye development. *Mol. Cell.* **3,** 297–307.

D'Amico-Martel, A., and Noden, D. M. (1980). An autoradiographic analysis of the development of the chick trigeminal ganglion. *J. Embryol. Exp. Morphol.* **55,** 167–182.

D'Amico-Martel, A., and Noden, D. M. (1983). Contributions of placodal and neural crest cells to avian cranial peripheral ganglia. *Am. J. Anat.* **166,** 445–468.

Daikoku, S. (1999). The olfactory origin of luteinizing hormone-releasing hormone (LHRH) neurons. A new era in reproduction physiology. *Arch. Histol. Cytol.* **62,** 107–117.

Daikoku, S., Chikamori, M., Adachi, T., and Maki, Y. (1982). Effect of the basal diencephalon on the development of Rathke's pouch in rats: A study in combined organ cultures. *Dev. Biol.* **90,** 198–202.

Damas, H. (1951). Observations sur le developpement des ganglions craniens chez *Lampetra fluviatilis* (L.). *Arch. Biol.* **62,** 65–95.

Darnell, D. K., Stark, M. R., and Schoenwolf, G. C. (1999). Timing and cell interactions underlying neural induction in the chick embryo. *Development* **126,** 2505–2514.

Dasen, J. S., and Rosenfeld, M. G. (1999). Signalling mechanisms in pituitary morphogenesis and cell fate determination. *Curr. Opin. Cell. Biol.* **11,** 669–677.

Dattani, M. T., Martinez-Barbera, J. P., Thomas, P. Q., Brickman, J. M., Gupta, R., Martensson, I. L., Toresson, H., Fox, M., Wales, J. K., Hindmarsh, P. C., Krauss, S., Beddington, R. S., and Robinson, I. C. (1998). Mutations in the homeobox gene *HESX1/Hesx1* associated with septo-optic dysplasia in human and mouse. *Nat. Genet.* **19,** 125–133.

David, R., and Wedlich, D. (2000). *Xenopus* cadherin-6 is expressed in the central and peripheral nervous system and in neurogenic placodes. *Mech. Dev.* **97,** 187–190.

Davis, R. J., Shen, W., Heanue, T. A., and Mardon, G. (1999). Mouse *Dach*, a homologue of *Drosophila dachshund*, is expressed in the developing retina, brain and limbs. *Dev. Genes Evol.* **209,** 526–536.

de Iongh, R., and McAvoy, J. W. (1993). Spatio-temporal distribution of acidic and basic FGF indicates a role for FGF in rat lens morphogenesis. *Dev. Dyn.* **198,** 190–202.

De Moerlooze, L., Spencer-Dene, B., Revest, J., Hajihosseini, M., Rosewell, I., and Dickson, C. (2000). An important role for the IIIb isoform of fibroblast growth factor receptor 2 (FGFR2) in mesenchymal-epithelial signalling during mouse organogenesis. *Development* **127,** 483–492.

Deitcher, D. L., Fekete, D. M., and Cepko, C. L. (1994). Asymmetric expression of a novel homeobox gene in vertebrate sensory organs. *J. Neurosci.* **14,** 486–498.

Del Rio-Tsonis, K., Jung, J. C., Chiu, I. M., and Tsonis, P. A. (1997). Conservation of fibroblast growth factor function in lens regeneration. *Proc. Natl. Acad. Sci. USA* **94,** 13701–13706.

Del Rio-Tsonis, K., Trombley, M. T., McMahon, G., and Tsonis, P. A. (1998). Regulation of lens regeneration by fibroblast growth factor receptor 1. *Dev. Dyn.* **213,** 140–146.

Dellovade, T. L., Pfaff, D. W., and Schwanzel-Fukuda, M. (1998). The gonadotropin-releasing hormone system does not develop in *Small-Eye* (*Sey*) mouse phenotype. *Brain Res. Dev. Brain Res.* **107,** 233–240.

Demski, L. S. (1993). Terminal nerve complex. *Acta Anat.* **148,** 81–95.

Denis, H. (1959). Sur l'induction des capsules nasales chez les Amphibiens. *Arch. Biol.* **70,** 851–873.

Deol, M. S. (1964). The abnormalities of the inner ear in *kreisler* mice. *J. Embryol. Exp. Morphol.* **12,** 475–490.

Depew, M. J., Liu, J. K., Long, J. E., Presley, R., Meneses, J. J., Pedersen, R. A., and Rubenstein, J. L. (1999). *Dlx5* regulates regional development of the branchial arches and sensory capsules. *Development* **126,** 3831–3846.

Detwiler, S. R. (1948). Further quantitative studies on locomotor capacity of larval *Amblystoma* following surgical procedures upon embryonic brain. *J. Exp. Zool.* **108,** 45–74.

Detwiler, S. R., and van Dyke, R. H. (1950). The role of the medulla in the differentiation of the otic vesicle. *J. Exp. Zool.* **113,** 179–199.

Dickinson, M. E., Selleck, M. A., McMahon, A. P., and Bronner-Fraser, M. (1995). Dorsalization of the neural tube by the non-neural ectoderm. *Development* **121,** 2099–2106.

Diebel, C. E., Proksch, R., Green, C. R., Neilson, P., and Walker, M. M. (2000). Magnetite defines a vertebrate magnetoreceptor. *Nature* **406,** 299–302.

Dryer, L., and Graziadei, P. P. (1994). Influence of the olfactory organ on brain development. *Perspect. Dev. Neurobiol.* **2,** 163–174.

Drysdale, T. A., and Elinson, R. P. (1992). Cell migration and induction in the development of the surface ectodermal pattern of the *Xenopus laevis* tadpole. *Dev. Growth Differ.* **34,** 51–59.

Dubois, L., Bally-Cuif, L., Crozatier, M., Moreau, J., Paquereau, L., and Vincent, A. (1998). XCoe2, a transcription factor of the Col/Olf-1/EBF family involved in the specification of primary neurons in *Xenopus*. *Curr. Biol.* **8,** 199–209.

Dupé, V., Ghyselinck, N. B., Wendling, O., Chambon, P., and Mark, M. (1999). Key roles of retinoic acid receptors alpha and beta in the patterning of the caudal hindbrain, pharyngeal arches and otocyst in the mouse. *Development* **126,** 5051–5059.

Duprez, D., Leyns, L., Bonnin, M. A., Lapointe, F., Etchevers, H., De Robertis, E. M., and Le Douarin, N. (1999). Expression of *Frzb-1* during chick development. *Mech. Dev.* **89,** 179–183.

Eagleson, G. W., and Harris, W. A. (1990). Mapping of the presumptive brain regions in the neural plate of *Xenopus laevis*. *J. Neurobiol.* **21,** 427–440.

Eagleson, G. W., Jenks, B. G., and Van Overbeeke, A. P. (1986). The pituitary adrenocorticotropes originate from neural ridge tissue in *Xenopus laevis*. *J. Embryol. Exp. Morphol.* **95,** 1–14.

Eagleson, G. W., Ferreiro, B., and Harris, W. A. (1995). Fate of the anterior neural ridge and the morphogenesis of the *Xenopus* forebrain. *J. Neurobiol.* **28,** 146–158.

Eakin, R. M. (1939). Regional determination in the development of the trout. *Roux's Arch. EntwMech.* **139,** 274–281.

Eccles, M. R., and Schimmenti, L. A. (1999). Renal-coloboma syndrome: A multi-system developmental disorder caused by *PAX2* mutations. *Clin. Genet.* **56,** 1–9.

Eisthen, H. L., Delay, R. J., Wirsig-Wiechmann, C. R., and Dionne, V. E. (2000). Neuromodulatory effects of gonadotropin releasing hormone on olfactory receptor neurons. *J. Neurosci.* **20,** 3947–3955.

Ekker, S. C., McGrew, L. L., Lai, C. J., Lee, J. J., von Kessler, D. P., Moon, R. T., and Beachy, P. A. (1995). Distinct expression and shared activities of members of the *hedgehog* gene family of *Xenopus laevis*. *Development* **121,** 2337–2347.

El Amraoui, A., and Dubois, P. M. (1993a). Experimental evidence for an early commitment of gonadotropin-releasing hormone neurons, with special regard to their origin from the ectoderm of nasal cavity presumptive territory. *Neuroendocrinology* **57,** 991–1002.

El Amraoui, A., and Dubois, P. M. (1993b). Experimental evidence for the early commitment of the presumptive adenohypophysis. *Neuroendocrinology* **58,** 609–615.

Ellies, D. L., Stock, D. W., Hatch, G., Giroux, G., Weiss, K. M., and Ekker, M. (1997). Relationship between the genomic organization and the overlapping embryonic expression patterns of the zebrafish *dlx* genes. *Genomics* **45,** 580–590.

Emerson, H. S. (1945). The development of late gastrula explants of *Rana pipiens* in salt solution. *J. Exp. Zool.* **100,** 497–521.

Enwright, J. F., 3rd, and Grainger, R. M. (2000). Altered retinoid signalling in the heads of *Small eye* mouse embryos. *Dev. Biol.* **221,** 10–22.

Ericson, J., Norlin, S., Jessell, T. M., and Edlund, T. (1998). Integrated FGF and BMP signalling controls the progression of progenitor cell differentiation and the emergence of pattern in the embryonic anterior pituitary. *Development* **125,** 1005–1015.

Ermakova, G. V., Alexandrova, E. M., Kazanskaya, O. V., Vasiliev, O. L., Smith, M. W., and Zaraisky, A. G. (1999). The homeobox

gene, *Xanf-1*, can control both neural differentiation and patterning in the presumptive anterior neurectoderm of the *Xenopus laevis* embryo. *Development* **126,** 4513–4523.

Esteve, P., and Bovolenta, P. (1999). *cSix4*, a member of the six gene family of transcription factors, is expressed during placode and somite development. *Mech. Dev.* **85,** 161–165.

Esteve, P., Morcillo, J., and Bovolenta, P. (2000). Early and dynamic expression of *cSfrp1* during chick embryo development. *Mech. Dev.* **97,** 217–221.

Farbman, A. I. (1994). Developmental biology of olfactory sensory neurons. *Semin. Cell. Biol.* **5,** 3–10.

Fautrez, J. (1942). La signification de la partie céphalique du bourrelet de la plaque médullaire chez les Urodèles. Localisation des ébauches présomptives des microplacodes des nerfs crâniens et de la crête ganglionnaire de la tête au stade neurula. *Bull. Cl. Sci. Acad. R. Belg.* **28,** 391–403.

Favor, J., Sandulache, R., Neuhauser-Klaus, A., Pretsch, W., Chatterjee, B., Senft, E., Wurst, W., Blanquet, V., Grimes, P., Sporle, R., and Schughart, K. (1996). The mouse *Pax2*^{1Neu} mutation is identical to a human *PAX2* mutation in a family with renal-coloboma syndrome and results in developmental defects of the brain, ear, eye, and kidney. *Proc. Natl. Acad. Sci. USA* **93,** 13870–13875.

Fedtsova, N. G., and Barabanov, V. M. (1978). Lens and adenohypophysis differentiation in ectoderm of the oral region of chick embryos *in vitro*. *Ontogenez* **9,** 530–536.

Fedtsova, N. G., and Barabanov, V. M. (1990). Distribution of competence for adenohypophyseal development in chick embryo ectoderm. *Sov. J. Dev. Biol.* **21,** 183–188.

Fedtsova, N. G., Minina, T. A., and Barabanov, V. M. (1981). Synthesis of a lens-specific antigen (δ-crystallin) in rudimentary chicken adenohypophysis. *Biull. Eksp. Biol. Med.* **92,** 314–316.

Fekete, D. M. (1996). Cell fate specification in the inner ear. *Curr. Opin. Neurobiol.* **6,** 533–541.

Fekete, D. M. (1999). Development of the vertebrate ear: Insights from knockouts and mutants. *Trends Neurosci.* **22,** 263–269.

Ferrand, R. (1969a). Différenciation en greffe chorio-allantoïdienne de l'épithélium de la poche de Rathke d'embryons de Caille et de Poulet. *C. R. Soc. Biol.* **163,** 2669–2672.

Ferrand, R. (1969b). Influence inductrice exercée par le plancher encéphalique sur l'ébauche adénohypophysaire aux jeunes stades du développement de l'embryon de poulet. *C. R. Acad. Sci. Sér. D* **268,** 550–553.

Ferrand, R. (1972). Etude expérimentale des facteurs de la differenciation cytologique de l'adénohypophyse chez l'embryon de poulet. *Arch. Biol.* **83,** 297–371.

Fishman, M. C., and Chien, K. R. (1997). Fashioning the vertebrate heart: Earliest embryonic decisions. *Development* **124,** 2099–2117.

Fode, C., Gradwohl, G., Morin, X., Dierich, A., LeMeur, M., Goridis, C., and Guillemot, F. (1998). The bHLH protein NEUROGENIN 2 is a determination factor for epibranchial placode-derived sensory neurons. *Neuron* **20,** 483–494.

Franco, B., Guioli, S., Pragliola, A., Incerti, B., Bardoni, B., Tonlorenzi, R., Carrozzo, R., Maestrini, E., Pieretti, M., Taillon-Miller, P., *et al.* (1991). A gene deleted in Kallmann's syndrome shares homology with neural cell adhesion and axonal path-finding molecules. *Nature* **353,** 529–536.

Frémont, P. H., and Ferrand, R. (1979). In vitro studies on the self-differentiating capacities of quail adenohypophysis epithelium. *Anat. Embryol. (Berl.)* **156,** 255–67.

Fritzsch, B. (1989). Diversity and regression in the amphibian lateral line and electrosensory system. *In* "The Mechanosensory Lateral Line. Neurobiology and Evolution" (S. Coombs, P. Görner, and H. Münz, Eds.), pp. 99–114. Springer-Verlag, New York.

Fritzsch, B. (1990). The evolution of metamorphosis in amphibians. *J. Neurobiol.* **21,** 1011–1021.

Fritzsch, B., Barald, K. F., and Lomax, M. I. (1997). Early embryology of the vertebrate ear. *In* "Development of the Auditory System" (E. W. Rubel, A. N. Popper, and R. R. Fay, Eds.). Springer Verlag, New York.

Fritzsch, B., Pirvola, U., and Ylikoski, J. (1999). Making and breaking the innervation of the ear: Neurotrophic support during ear development and its clinical implications. *Cell. Tissue Res.* **295,** 369–382.

Frohman, M. A., Martin, G. R., Cordes, S. P., Halamek, L. P., and Barsh, G. S. (1993). Altered rhombomere-specific gene expression and hyoid bone differentiation in the mouse segmentation mutant *kreisler* (*kr*). *Development* **117,** 925–936.

Frolova, E., and Beebe, D. (2000). The expression pattern of a novel *Deltex* homologue during chicken embryogenesis. *Mech. Dev.* **92,** 285–289.

Froriep, A. (1885). Ueber Anlagen von Sinnesorganen am Facialis, Glossopharyngeus und Vagus, über die genetische Stellung des Vagus zum Hypoglossus, und über die Herkunft der Zungenmusculatur. *Arch. Anat. Physiol.* 1–55.

Fujiwara, M., Uchida, T., Osumi-Yamashita, N., and Eto, K. (1994). Uchida rat (*rSey*): A new mutant rat with craniofacial abnormalities resembling those of the mouse *Sey* mutant. *Differentiation* **57,** 31–38.

Furuta, Y., and Hogan, B. L. M. (1998). BMP4 is essential for lens induction in the mouse embryo. *Genes Dev.* **12,** 3764–3775.

Gage, P. J., and Camper, S. A. (1997). Pituitary homeobox 2, a novel member of the *bicoid*-related family of homeobox genes, is a potential regulator of anterior structure formation. *Hum. Mol. Genet.* **6,** 457–464.

Gage, P. J., Suh, H., and Camper, S. A. (1999). Dosage requirement of *Pitx2* for development of multiple organs. *Development* **126,** 4643–4651.

Gallagher, B. C., Henry, J. J., and Grainger, R. M. (1996). Inductive processes leading to inner ear formation during *Xenopus* development. *Dev. Biol.* **175,** 95–107.

Galliot, B., de Vargas, C., and Miller, D. (1999). Evolution of homeobox genes: Q_{50} Paired-like genes founded the Paired class. *Dev. Genes Evol.* **209,** 186–197.

Gans, C., and Northcutt, R. G. (1983). Neural crest and the origin of vertebrates: A new head. *Science* **220,** 268–274.

Gao, C., Noden, D. M., and Norgren, R. B., Jr. (2000). LHRH neuronal migration: Heterotypic transplantation analysis of guidance cues. *J. Neurobiol.* **42,** 95–103.

García-Castro, M., and Bronner-Fraser, M. (1999). Induction and differentiation of the neural crest. *Curr. Opin. Cell. Biol.* **11,** 695–698.

Garcia-Martinez, V., Alvarez, I. S., and Schoenwolf, G. C. (1993). Locations of the ectodermal and nonectodermal subdivisions of the epiblast at stages 3 and 4 of avian gastrulation and neurulation. *J. Exp. Zool.* **267,** 431–446.

Gavalas, A., Studer, M., Lumsden, A., Rijli, F. M., Krumlauf, R., and Chambon, P. (1998). *Hoxa1* and *Hoxb*1 synergize in patterning the hindbrain, cranial nerves and second pharyngeal arch. *Development* **125,** 1123–1136.

Gehring, W. J., and Ikeo, K. (1999). *Pax6*: Mastering eye morphogenesis and eye evolution. *Trends Genet.* **15,** 371–377.

George, K. M., Leonard, M. W., Roth, M. E., Lieuw, K. H., Kioussis, D., Grosveld, F., and Engel, J. D. (1994). Embryonic expression and cloning of the murine GATA-3 gene. *Development* **120,** 2673–2686.

Ginsburg, A. S. (1946). Specific differences in the determination of the internal ear and other ectodermal organs in certain *Urodela. C. R. (Dokl.) Acad. Sci. URSS* **54,** 557–560.

Ginsburg, A. S. (1995). Determination of the labyrinth in different amphibian species and its correlation with determination of the other ectoderm derivatives. *Roux's Arch. Dev. Biol.* **204,** 351–358.

Giraldez, F. (1998). Regionalized organizing activity of the neural tube revealed by the regulation of *lmx1* in the otic vesicle. *Dev. Biol.* **203,** 189–200.

Glaser, T., Walton, D. S., and Maas, R. L. (1992). Genomic structure, evolutionary conservation and aniridia mutations in the human PAX6 gene. *Nat. Genet.* **2,** 232–239.

Glasgow, E., and Tomarev, S. I. (1998). Restricted expression of the homeobox gene *prox 1* in developing zebrafish. *Mech. Dev.* **76,** 175–178.

Glasgow, E., Karavanov, A. A., and Dawid, I. B. (1997). Neuronal and neuroendocrine expression of *lim3*, a LIM class homeobox gene, is altered in mutant zebrafish with axial signalling defects. *Dev. Biol.* **192,** 405–419.

Gleiberman, A. S., Fedtsova, N. G., and Rosenfeld, M. G. (1999). Tissue interactions in the induction of anterior pituitary: role of the ventral diencephalon, mesenchyme, and notochord. *Dev. Biol.* **213,** 340–353.

Goldstone, K., and Sharpe, C. R. (1998). The expression of *XIF3* in undifferentiated anterior neuroectoderm, but not in primary neurons, is induced by the neuralizing agent noggin. *Int. J. Dev. Biol.* **42,** 757–762.

Gong, Q., and Shipley, M. T. (1995). Evidence that pioneer olfactory axons regulate telencephalon cell cycle kinetics to induce the formation of the olfactory bulb. *Neuron* **14,** 91–101.

Gopal-Srivastava, R., Cvekl, A., and Piatigorsky, J. (1998). Involvement of retinoic acid/retinoid receptors in the regulation of murine αB-crystallin/small heat shock protein gene expression in the lens. *J. Biol. Chem.* **273,** 17954–17961.

Gorbman, A. (1983). Early development of the hagfish pituitary gland: Evidence for the endodermal origin of the adenohypophysis. *Amer. Zool.* **23,** 639–654.

Gorbman, A., and Tamarin, A. (1985). Early development of oral, olfactory and adenohypophyseal structures of agnathans and its evolutionary implications. *NATO ASI Ser. Ser. A, Life Sci.* **103,** 165–185.

Gradwohl, G., Fode, C., and Guillemot, F. (1996). Restricted expression of a novel murine *atonal*-related bHLH protein in undifferentiated neural precursors. *Dev. Biol.* **180,** 227–241.

Graham, A., and Begbie, J. (2000). Neurogenic placodes: a common front. *Trends Neurosci.* **23,** 313–316.

Grainger, R. M. (1992). Embryonic lens induction: Shedding light on vertebrate tissue determination. *Trends Genet.* **8,** 349–355.

Grainger, R. M. (1996). New perspectives on embryonic lens induction. *Semin. Cell. Dev. Biol.* **7,** 149–155.

Grainger, R. M., Herry, J. J., and Henderson, R. A. (1988). Reinvestigation of the role of the optic vesicle in embryonic lens induction. *Development* **102,** 517–526.

Grainger, R. M., Mannion, J. E., Cook, T. L., and Zygar, C. A. (1997). Defining intermediate stages in cell determination: Acquisition of a lens-forming bias in head ectoderm during lens determination. *Dev. Genet.* **20,** 246–257.

Graveson, A. C., and Armstrong, J. B. (1990). The *premature death* (*p*) mutation of *Ambystoma mexicanum* affects a subpopulation of neural crest cells. *Differentiation* **45,** 71–75.

Graveson, A. C., and Armstrong, J. B. (1994). In vivo evidence that the *premature death* (*p*) mutation of *Ambystoma mexicanum* affects an early segregating subpopulation of neural crest cells. *J. Exp. Zool.* **269,** 327–335.

Graveson, A. C., and Armstrong, J. B. (1996). *Premature death* (*p*) mutation of *Ambystoma mexicanum* affects the ability of ectoderm to respond to neural induction. *J. Exp. Zool.* **274,** 248–254.

Graziadei, P. P., and Monti-Graziadei, A. G. (1992). The influence of the olfactory placode on the development of the telencephalon in *Xenopus laevis. Neuroscience* **46,** 617–629.

Greenwood, A. L., Turner, E. E., and Anderson, D. J. (1999). Identification of dividing, determined sensory neuron precursors in the mammalian neural crest. *Development* **126,** 3545–3559.

Grinblat, Y., Gamse, J., Patel, M., and Sive, H. (1998). Determination of the zebrafish forebrain: induction and patterning. *Development* **125,** 4403–4416.

Grindley, J. C., Davidson, D. R., and Hill, R. E. (1995). The role of *Pax-6* in eye and nasal development. *Development* **121,** 1433–1442.

Groves, A. K., and Bronner-Fraser, M. (2000). Competence, specification and commitment in otic placode induction. *Development* **127,** 3489–3499.

Guillemot, F., Lo, L. C., Johnson, J. E., Auerbach, A., Anderson, D. J., and Joyner, A. L. (1993). Mammalian *achaete-scute* homologue 1 is required for the early development of olfactory and autonomic neurons. *Cell* **75,** 463–476.

Gurdon, J. B. (1987). Embryonic induction—molecular prospects. *Development* **99,** 285–306.

Haddon, C., and Lewis, J. (1996). Early ear development in the embryo of the zebrafish, *Danio rerio. J. Comp. Neurol.* **365,** 113–128.

Haddon, C., Jiang, Y. J., Smithers, L., and Lewis, J. (1998). Delta-Notch signalling and the patterning of sensory cell differentiation in the zebrafish ear: Evidence from the *mind bomb* mutant. *Development* **125,** 4637–4644.

Haggis, A. J. (1956). Analysis of the determination of the olfactory placode in *Amblystoma punctatum*. *J. Embryol. Exp. Morphol.* **4,** 120–138.

Hall, B. K. (1999). "The Neural Crest in Development and Evolution." Springer-Verlag, New York.

Hamburger, V. (1961). Experimental analysis of the dual origin of the trigeminal ganglion in the chick embryo. *J. Exp. Zool.* **148,** 91–117.

Hamburger, V., and Hamilton, H. L. (1951). A series of normal stages in the development of the chick embryo. *J. Morphol.* **88,** 49–92.

Hammond, K. L., Hanson, I. M., Brown, A. G., Lettice, L. A., and Hill, R. E. (1998). Mammalian and *Drosophila dachshund* genes are related to the *Ski* proto-oncogene and are expressed in eye and limb. *Mech. Dev.* **74,** 121–131.

Hansen, A., and Zeiske, E. (1993). Development of the olfactory organ in the zebrafish, *Brachydanio rerio. J. Comp. Neurol.* **333,** 289–300.

Hardelin, J. P., Julliard, A. K., Moniot, B., Soussi-Yanicostas, N., Verney, C., Schwanzel-Fukuda, M., Ayer-Le Lièvre, C., and Petit, C. (1999). Anosmin-1 is a regionally restricted component of basement membranes and interstitial matrices during organogenesis: Implications for the developmental anomalies of X chromosome-linked Kallmann syndrome. *Dev. Dyn.* **215,** 26–44.

Harrison, R. G. (1904). Experimentelle Untersuchung über die Entwicklung der Sinnesorgane der Seitenlinie bei den Amphibien. *Arch. Mikrosk. Anat. EntwMech.* **63,** 35–149.

Harrison, R. G. (1936). Relations of symmetry in the developing ear of *Amblystoma punctatum*. *Proc. Natl. Acad. Wash.* **22,** 238–247.

Harrison, R. G. (1945). Relations of symmetry in the developing embryo. *Trans. Conn. Acad. Arts. Sci.* **36,** 277–330.

Harrison, T. A., Stadt, H. A., Kumiski, D., and Kirby, M. L. (1995). Compensatory responses and development of the nodose ganglion following ablation of placodal precursors in the embryonic chick (*Gallus domesticus*). *Cell. Tissue Res.* **281,** 379–385.

Hatada, Y., and Stern, C. D. (1994). A fate map of the epiblast of the early chick embryo. *Development* **120,** 2879–2889.

Hatini, V., Ye, X., Balas, G., and Lai, E. (1999). Dynamics of placodal lineage development revealed by targeted transgene expression. *Dev. Dyn.* **215,** 332–343.

Heanue, T. A., Reshef, R., Davis, R. J., Mardon, G., Oliver, G., Tomarev, S., Lassar, A. B., and Tabin, C. J. (1999). Synergistic regulation of vertebrate muscle development by *Dach2*, *Eya2*, and *Six1*, homologs of genes required for *Drosophila* eye formation. *Genes Dev.* **13,** 3231–3243.

Heikinheimo, M., Lawshe, A., Shackleford, G. M., Wilson, D. B., and MacArthur, C. A. (1994). *Fgf-8* expression in the postgastrulation mouse suggests roles in the development of the face, limbs and central nervous system. *Mech. Dev.* **48,** 129–138.

Heller, N., and Brändli, A. W. (1997). *Xenopus Pax-2* displays multiple splice forms during embryogenesis and pronephric kidney development. *Mech. Dev.* **69,** 83–104.

Heller, N., and Brändli, A. W. (1999). *Xenopus Pax-2/5/8* orthologues: Novel insights into *Pax* gene evolution and identification of *Pax-8* as the earliest marker for otic and pronephric cell lineages. *Dev. Genet.* **24,** 208–219.

Henderson, D. J., Ybot-Gonzalez, P., and Copp, A. J. (2000). RhoB is expressed in migrating neural crest and endocardial cushions of the developing mouse embryo. *Mech. Dev.* **95,** 211–214.

Henry, J. J., and Grainger, R. M. (1987). Inductive interactions in the spatial and temporal restriction of lens-forming potential in embryonic ectoderm of *Xenopus laevis*. *Dev. Biol.* **124,** 200–214.

Henry, J. J., and Grainger, R. M. (1990). Early tissue interactions leading to embryonic lens formation in *Xenopus laevis*. *Dev. Biol.* **141,** 149–163.

Herbrand, H., Guthrie, S., Hadrys, T., Hoffmann, S., Arnold, H. H., Rinkwitz-Brandt, S., and Bober, E. (1998). Two regulatory genes, *cNkx5–1* and *cPax2*, show different responses to local signals during otic placode and vesicle formation in the chick embryo. *Development* **125,** 645–654.

Hermesz, E., Mackem, S., and Mahon, K. A. (1996). *Rpx*: A novel anterior-restricted homeobox gene progressively activated in the prechordal plate, anterior neural plate and Rathke's pouch of the mouse embryo. *Development* **122,** 41–52.

Hidalgo-Sánchez, M., Alvarado-Mallart, R., and Alvarez, I. S. (2000). *Pax2*, *Otx2*, *Gbx2* and *Fgf8* expression in early otic vesicle development. *Mech. Dev.* **95,** 225–229.

Higashijima, S., Hotta, Y., and Okamoto, H. (2000). Visualization of cranial motor neurons in live transgenic zebrafish expressing green fluorescent protein under the control of the *islet-1* promoter/enhancer. *J. Neurosci.* **20,** 206–218.

Hill, R. E., Favor, J., Hogan, B.L.M., Ton, C. T., Saunders, G. F., Hanson, I. M., Prosser, J., Jordan, T., Hastie, N. D., and van Heyningen, V. (1991). Mouse *Small eye* results from mutations in a paired-like homeobox-containing gene. *Nature* **354,** 522–525.

Hillemann, H. H. (1943). An experimental study of the development of the pituitary gland in chick embryos. *J. Exp. Zool.* **93,** 347–373.

Hirsch, N., and Harris, W. A. (1997). *Xenopus* Pax-6 and retinal development. *J. Neurobiol.* **32,** 45–61.

Hogan, B. L. M., Horsburgh, G., Cohen, J., Hetherington, C. M., Fisher, G., and Lyon, M. F. (1986). *Small eyes* (*Sey*): A homozygous lethal mutation on chromosome 2 which affects the differentiation of both lens and nasal placodes in the mouse. *J. Embryol. Exp. Morphol.* **97,** 95–110.

Holder, N., and Hill, J. (1991). Retinoic acid modifies development of the midbrain-hindbrain border and affects cranial ganglion formation in zebrafish embryos. *Development* **113,** 1159–1170.

Hollemann, T., Bellefroid, E., and Pieler, T. (1998). The *Xenopus* homologue of the *Drosophila* gene *tailless* has a function in early eye development. *Development* **125,** 2425–2432.

Hollemann, T., and Pieler, T. (1999). *Xpitx-1*: A homeobox gene expressed during pituitary and cement gland formation of *Xenopus* embryos. *Mech. Dev.* **88,** 249–252.

Holtfreter, J. (1933). Der Einfluss von Wirtsalter und verschiedenen Organbezirken auf die Differenzierung von angelagertem Gastrulaektoderm. *Roux's Arch. EntwMech.* **127,** 619–775.

Homma, S., Oppenheim, R. W., Yaginuma, H., and Kimura, S. (2000). Expression pattern of GDNF, *c-ret*, and GFRas suggests novel roles for GDNF ligands during early organogenesis in the chick embryo. *Dev. Biol.* **217,** 121–137.

Houart, C., Westerfield, M., and Wilson, S. W. (1998). A small population of anterior cells patterns the forebrain during zebrafish gastrulation. *Nature* **391,** 788–792.

Hyuga, M., Kodama, R., and Eguchi, G. (1993). Basic fibroblast growth factor as one of the essential factors regulating lens transdifferentiation of pigmented epithelial cells. *Int. J. Dev. Biol.* **37,** 319–326.

Islam, N., Poitras, L., and Moss, T. (2000). The cytoskeletal effector xPAK1 is expressed during both ear and lateral line development in *Xenopus*. *Int. J. Dev. Biol.* **44,** 245–248.

Jacobson, A. G. (1955). The roles of the optic vesicle and other head tissues in lens induction. *Proc. Natl. Acad. Sci. USA* **41,** 522–555.

Jacobson, A. G. (1958). The roles of neural and non-neural tissues in lens induction. *J. Exp. Zool.* **139,** 525–557.

Jacobson, A. G. (1963a). The determination and positioning of the nose, lens and ear. I. Interactions within the ectoderm, and between the ectoderm and underlying tissues. *J. Exp. Zool.* **154,** 273–283.

Jacobson, A. G. (1963b). The determination and positioning of the nose, lens and ear. II. The role of the endoderm. *J. Exp. Zool.* **154,** 285–291.

Jacobson, A. G. (1963c). The determination and positioning of the nose, lens and ear. III. Effects of reversing the antero-posterior axis of epidermis, neural plate and neural fold. *J. Exp. Zool.* **154,** 293–303.

Jacobson, A. G. (1966). Inductive processes in embryonic development. *Science* **152,** 25–34.

Jacobson, A. G., and Sater, A. K. (1988). Features of embryonic induction. *Development* **104,** 341–359.

Jacobson, C.-O. (1959). The localization of the presumptive cerebral regions in the neural plate of the axolotl larva. *J. Embryol. Exp. Morphol.* **7,** 1–21.

Jean, D., Bernier, G., and Gruss, P. (1999). *Six6* (*Optx2*) is a novel murine *Six3*-related homeobox gene that demarcates the presumptive pituitary/hypothalamic axis and the ventral optic stalk. *Mech. Dev.* **84,** 31–40.

Jeremy, S. D., and Michael, G. D. (1999). Signalling mechanisms in pituitary morphogenesis and cell fate determination. *Curr. Opin. Cell. Biol.* **11,** 669–677.

Jester, J. V., Moller-Pedersen, T., Huang, J., Sax, C. M., Kays, W. T., Cavangh, H. D., Petroll, W. M., and Piatigorsky, J. (1999). The cellular basis of corneal transparency: Evidence for "corneal crystallins". *J. Cell. Sci.* **112,** 613–622.

Jones, E. A., and Woodland, H. R. (1989). Spatial aspects of neural induction in *Xenopus laevis*. *Development* **107,** 785–791.

Jordan, T., Hanson, I., Zaletayev, D., Hodgson, S., Prosser, J., Seawright, A., Hastie, N., and van Heyningen, V. (1992). The human PAX6 gene is mutated in two patients with aniridia. *Nat. Genet.* **1,** 328–332.

Jørgensen, J. M. (1989). Evolution of octavolateralis sensory cells. *In* "The Mechanosensory Lateral Line. Neurobiology and Evolution" (S. Coombs, P. Görner, and H. Münz, Eds.), pp. 115–145. Springer-Verlag, New York.

Jorquera, B., Goicoechea, O., and Molinari, E. (1989). *In vitro* studies of lens formation in chicken embryo in the absence of hypoblast and optic vesicle. *Anat. Histol. Embryol.* **18,** 32–44.

Kaan, H. W. (1926). Experiments on the development of the ear of *Amblystoma punctatum*. *J. Exp. Zool.* **46,** 13–61.

Kablar, B., Vignali, R., Menotti, L., Pannese, M., Andreazzoli, M., Polo, C., Giribaldi, M. G., Boncinelli, E., and Barsacchi, G. (1996). *Xotx* genes in the developing brain of *Xenopus laevis*. *Mech. Dev.* **55,** 145–158.

Kaestner, K. H., Knochel, W., and Martinez, D. E. (2000). Unified nomenclature for the winged helix/forkhead transcription factors. *Genes Dev.* **14,** 142–146.

Kamachi, Y., Sockanathan, S., Liu, Q., Breitman, M., Lovell-Badge, R., and Kondoh, H. (1995). Involvement of SOX proteins in lens-specific activation of crystallin genes. *EMBO J.* **14,** 3510–3519.

Kamachi, Y., Uchikawa, M., Collignon, J., Lovell-Badge, R., and Kondoh, H. (1998). Involvement of Sox1, 2 and 3 in the early and subsequent molecular events of lens induction. *Development* **125,** 2521–2532.

Kammandel, B., Chowdhury, K., Stoykova, A., Aparicio, S., Brenner, S., and Gruss, P. (1999). Distinct *cis*-essential modules direct the time-space pattern of the *Pax6* gene activity. *Dev. Biol.* **205,** 79–97.

Kardong, K. V. (1998). "Vertebrates: Comparative Anatomy, Function, Evolution." WCB/McGraw-Hill, Boston.

Karkinen-Jääskeläinen, M. (1978). Permissive and directive interactions in lens induction. *J. Embryol. Exp. Morphol.* **44,** 167–179.

Karlstrom, R. O., Talbot, W. S., and Schier, A. F. (1999). Comparative synteny cloning of zebrafish *you-too*: Mutations in the Hedgehog target *gli2* affect ventral forebrain patterning. *Genes Dev.* **13,** 388–393.

Kataoka, H., Ochi, M., Enomoto, K., and Yamaguchi, A. (2000). Cloning and embryonic expression patterns of the zebrafish Runt domain genes, *runxa* and *runxb*. *Mech. Dev.* **98,** 139–143.

Katz, D. M., Adler, J. E., and Black, I. B. (1987). Catecholaminergic primary sensory neurons: autonomic targets and mechanisms of transmitter regulation. *Fed. Proc.* **46,** 24–29.

Kaufman, M. H. (1992). "The Atlas of Mouse Development." Academic Press, London.

Kawakami, I. (1950). Inducing effects of the killed archenteron roof on late gastrula ectoderm. *Mem. Coll. Sci., Kyoto Imp. Univ., Ser. B* **19,** 1–5.

Kawakami, I. (1952). Relation of inductive effect to embryonic field, with special reference to the developmental mechanism of the cephalic sensory organs in amphibians. *Annot. Zool. Jpn.* **25,** 97–105.

Kawamura, K., and Kikuyama, S. (1992). Evidence that hypophysis and hypothalamus constitute a single entity from the primary stage of histogenesis. *Development* **115,** 1–9.

Kawamura, K., and Kikuyama, S. (1996). Origine de l'adénohypophyse et du système olfactif chez le crapaud: Démonstration par la méthode des chimères. *C. R. Soc. Biol.* **190,** 311–316.

Kawamura, K., and Kikuyama, S. (1998). Morphogenesis of the hypothalamus and hypophysis: Their association, dissociation and reassociation before and after "Rathke". *Arch. Histol. Cytol.* **61,** 189–198.

Kawauchi, S., Takahashi, S., Nakajima, O., Ogino, H., Morita, M., Nishizawa, M., Yasuda, K., and Yamamoto, M. (1999). Regulation of lens fiber cell differentiation by transcription factor c-Maf. *J. Biol. Chem.* **274,** 19254–19260.

Kazanskaya, O. V., Severtzova, E. A., Barth, K. A., Ermakova, G. V., Lukyanov, S. A., Benyumov, A. O., Pannese, M., Boncinelli, E., Wilson, S. W., and Zaraisky, A. G. (1997). Anf: a novel class of vertebrate homeobox genes expressed at the anterior end of the main embryonic axis. *Gene* **200,** 25–34.

Kengaku, M., and Okamoto, H. (1993). Basic fibroblast growth factor induces differentiation of neural tube and neural crest lineages of cultured ectoderm cells from *Xenopus* gastrula. *Development* **119,** 1067–1078.

Kenyon, K. L., Moody, S. A., and Jamrich, M. (1999). A novel *fork head* gene mediates early steps during *Xenopus* lens formation. *Development* **126,** 5107–5116.

Kim, J. I., Li, T., Ho, I. C., Grusby, M. J., and Glimcher, L. H. (1999). Requirement for the c-Maf transcription factor in crystallin gene regulation and lens development. *Proc. Natl. Acad. Sci. USA* **96,** 3781–3785.

Kimmel, C. B., Ballard, W. W., Kimmel, S. R., Ullmann, B., and Schilling, T. F. (1995). Stages of embryonic development of the zebrafish. *Dev. Dyn.* **203,** 253–310.

Kimura, S., Hara, Y., Pineau, T., Fernandez-Salguero, P., Fox, C. H., Ward, J. M., and Gonzalez, F. J. (1996). The *T/ebp* null mouse: Thyroid-specific enhancer-binding protein is essential for the organogenesis of the thyroid, lung, ventral forebrain, and pituitary. *Genes Dev.* **10,** 60–69.

King, J. A., and Millar, R. P. (1991). Gonadotropin-releasing hormones. *In* "Vertebrate Endocrinology: Fundamentals and Biomedical Implications" (P. K. T. Pang and M. P. Schreibman, Eds.), Vol. 4, Part B, pp. 1–31. Academic Press, New York.

Kioussi, C., Carriere, C., and Rosenfeld, M. G. (1999a). A model for the development of the hypothalamic-pituitary axis: Transcribing the hypophysis. *Mech. Dev.* **81,** 23–35.

Kioussi, C., O'Connell, S., St-Onge, L., Treier, M., Gleiberman, A. S., Gruss, P., and Rosenfeld, M. G. (1999b). *Pax6* is essential for establishing ventral-dorsal cell boundaries in pituitary gland development. *Proc. Natl. Acad. Sci. USA* **96,** 14378–14382.

Kirby, M. L. (1988a). Nodose placode contributes autonomic neurons to the heart in the absence of cardiac neural crest. *J. Neurosci.* **8,** 1089–1095.

Kirby, M. L. (1988b). Nodose placode provides ectomesenchyme to the developing chick heart in the absence of cardiac neural crest. *Cell. Tissue Res.* **252,** 17–22.

Kishida, R., Goris, R. C., Terashima, S., and Dubbeldam, J. L. (1984). A suspected infrared-recipient nucleus in the brainstem of the vampire bat, *Desmodus rotundus. Brain. Res.* **322,** 351–355.

Klein, S. L., and Graziadei, P. P. (1983). The differentiation of the olfactory placode in *Xenopus laevis*: A light and electron microscope study. *J. Comp. Neurol.* **217,** 17–30.

Knouff, R. A. (1935). The developmental pattern of ectodermal placodes in *Rana pipiens. J. Comp. Neurol.* **62,** 17–71.

Kobayashi, M., Toyama, R., Takeda, H., Dawid, I. B., and Kawakami, K. (1998). Overexpression of the forebrain-specific homeobox gene *six3* induces rostral forebrain enlargement in zebrafish. *Development* **125,** 2973–2982.

Kobayashi, M., Osanai, H., Kawakami, K., and Yamamoto, M. (2000). Expression of three zebrafish *Six4* genes in the cranial sensory placodes and the developing somites. *Mech. Dev.* **98,** 151–155.

Kohan, R. (1944). The chordomesoderm as an inductor of the ear vesicle. *C. R. (Dokl.) Acad. Sci. URSS* **45,** 39–41.

Kondoh, H. (1999). Transcription factors for lens development assessed *in vivo. Curr. Opin. Genet. Dev.* **9,** 301–308.

Korzh, V., Edlund, T., and Thor, S. (1993). Zebrafish primary neurons initiate expression of the LIM homeodomain protein Isl-1 at the end of gastrulation. *Development* **118,** 417–425.

Kosaka, M., Kodama, R., and Eguchi, G. (1998). *In vitro* culture system for iris-pigmented epithelial cells for molecular analysis of transdifferentiation. *Exp. Cell Res.* **245,** 245–251.

Köster, R. W., Kühnlein, R. P., and Wittbrodt, J. (2000). Ectopic *Sox3* activity elicits sensory placode formation. *Mech. Dev.* **95,** 175–187.

Kozlowski, D. J., Murakami, T., Ho, R. K., and Weinberg, E. S. (1997). Regional cell movement and tissue patterning in the zebrafish embryo revealed by fate mapping with caged fluorescein. *Biochem. Cell. Biol.* **75,** 551–562.

Krauss, S., Johansen, T., Korzh, V., and Fjose, A. (1991). Expression of the zebrafish paired box gene *pax*[*zf-b*] during early neurogenesis. *Development* **113,** 1193–1206.

Kuo, J. S., Patel, M., Gamse, J., Merzdorf, C., Liu, X., Apekin, V., and Sive, H. (1998). Opl: A zinc finger protein that regulates neural determination and patterning in *Xenopus. Development* **125,** 2867–2882.

Kuratani, S. C., and Bockman, D. E. (1992). Inhibition of epibranchial placode-derived ganglia in the developing rat by bisdiamine. *Anat. Rec.* **233,** 617–624.

Kuratani, S. C., and Eichele, G. (1993). Rhombomere transplantation repatterns the segmental organization of cranial nerves and reveals cell-autonomous expression of a homeodomain protein. *Development* **117,** 105–117.

Kuratani, S. C., and Hirano, S. (1990). The appearance of trigeminal ectopic ganglia within the surface ectoderm in the chick embryo. *Arch. Histol. Cytol.* **53,** 575–583.

Kuratani, S. C., and Wall, N. A. (1992). Expression of Hox 2.1 protein in restricted populations of neural crest cells and pharyngeal ectoderm. *Dev. Dyn.* **195,** 15–28.

LaBonne, C., and Bronner-Fraser, M. (1998a). Induction and patterning of the neural crest, a stem cell-like precursor population. *J. Neurobiol.* **36,** 175–189.

LaBonne, C., and Bronner-Fraser, M. (1998b). Neural crest induction in *Xenopus*: Evidence for a two-signal model. *Development* **125,** 2403–2414.

LaBonne, C., and Bronner-Fraser, M. (1999). Molecular mechanisms of neural crest formation. *Annu. Rev. Cell. Dev. Biol.* **15,** 81–112.

Ladd, A. N., Yatskievych, T. A., and Antin, P. B. (1998). Regulation of avian cardiac myogenesis by activin/TGFb and bone morphogenetic proteins. *Dev. Biol.* **204,** 407–419.

Ladher, R. K., Church, V. L., Allen, S., Robson, L., Abdelfattah, A., Brown, N. A., Hattersley, G., Rosen, V., Luyten, F. P., Dale, L., and Francis-West, P. H. (2000). Cloning and expression of the Wnt antagonists *Sfrp-2* and *Frzb* during chick development. *Dev. Biol.* **218,** 183–198.

Ladher, R. K., Anakwe, K. U., Gurney, A. L., Schoenwolf, G. C., and Francis-West, P. H. (2000). Identification of synergistic signals initiating inner ear development. *Science* **290,** 1965–1968.

Lai, C. J., Ekker, S. C., Beachy, P. A., and Moon, R. T. (1995). Patterning of the neural ectoderm of *Xenopus laevis* by the amino-terminal product of hedgehog autoproteolytic cleavage. *Development* **121,** 2349–2360.

LaMantia, A. S., Colbert, M. C., and Linney, E. (1993). Retinoic acid induction and regional differentiation prefigure olfactory pathway formation in the mammalian forebrain. *Neuron* **10,** 1035–1048.

Lamborghini, J. E. (1980). Rohon-Beard cells and other large neurons in *Xenopus* embryos originate during gastrulation. *J. Comp. Neurol.* **189,** 323–333.

Lamonerie, T., Tremblay, J. J., Lanctôt, C., Therrien, M., Gauthier, Y., and Drouin, J. (1996). *Ptx1*, a *bicoid*-related homeobox transcription factor involved in transcription of the pro-opiomelanocortin gene. *Genes Dev.* **10,** 1284–1295.

Lanctôt, C., Lamolet, B., and Drouin, J. (1997). The bicoid-related homeoprotein Ptx1 defines the most anterior domain of the embryo and differentiates posterior from anterior lateral mesoderm. *Development* **124,** 2807–2817.

Landacre, F. L. (1912). The epibranchial placodes of *Lepidosteus osseus* and their relation to the cerebral ganglia. *J. Comp. Neurol.* **22,** 1–69.

Landacre, F. L. (1926). The primitive lines of *Amblystoma jeffersonianum. J. Comp. Neurol.* **40,** 471–495.

Landacre, F. L. (1931). Data on the relative time of formation of the cerebral ganglia of *Amblystoma jeffersonianum. J. Comp. Neurol.* **53,** 205–224.

Lannoo, M. J., and Smith, S. C. (1989). The lateral line system. *In* "Developmental Biology of the Axolotl" (J. B. Armstrong and G. M. Malacinski, Eds.), pp. 176–186. Oxford Univ. Press, Oxford, UK.

Lawson, S. N., and Biscoe, T. J. (1979). Development of mouse dorsal root ganglia: An autoradiographic and quantitative study. *J. Neurocytol.* **8,** 265–274.

Layer, P. G., and Kaulich, S. (1991). Cranial nerve growth in birds is preceded by cholinesterase expression during neural crest cell migration and the formation of an HNK-1 scaffold. *Cell. Tissue Res.* **265,** 393–407.

Le Douarin, N. M., and Kalcheim, C. (1999). "The Neural Crest." Cambridge Univ. Press, Cambridge, UK.

Le Douarin, N., Ferrand, R., and Le Douarin, G. (1967). La différenciation de l'ébauche épithéliale de l'hypophyse séparée du plancher encéphalique et placée dans des mésenchymes hétérologues. *C. R. Acad. Sci., Ser. D* **264,** 3027–3029.

Le Douarin, N. M., Fontaine-Perus, J., and Couly, G. (1986). Cephalic ectodermal placodes and neurogenesis. *Trends Neurosci.* **9,** 175–180.

Le Douarin, N. M., Kalcheim, C., and Teillet, M.-A. (1992). The cellular and molecular basis of early sensory ganglion development. *In* "Sensory Neurons: Diversity, Development, and Plasticity" (S. A. Scott, Ed.), pp. 143–170. Oxford Univ. Press, Oxford, UK.

Lecuit, T., and Cohen, S. M. (1997). Proximal-distal axis formation in the *Drosophila* leg. *Nature* **388,** 139–145.

Lee, K. J., and Jessell, T. M. (1999). The specification of dorsal cell fates in the vertebrate central nervous system. *Annu. Rev. Neurosci.* **22,** 261–294.

Legouis, R., Hardelin, J. P., Levilliers, J., Claverie, J. M., Compain, S., Wunderle, V., Millasseau, P., Le Paslier, D., Cohen, D., Caterina, D., *et al.* (1991). The candidate gene for the X-linked Kallmann syndrome encodes a protein related to adhesion molecules. *Cell* **67,** 423–435.

Legouis, R., Hardelin, J. P., Petit, C., and Ayer-Le Lievre, C. (1994). Early expression of the *KAL* gene during embryonic development of the chick. *Anat. Embryol. (Berl.)* **190,** 549–562.

Leimeister, C., Bach, A., and Gessler, M. (1998). Developmental expression patterns of mouse sFRP genes encoding members of the secreted frizzled related protein family. *Mech. Dev.* **75,** 29–42.

Leimeister, C., Externbrink, A., Klamt, B., and Gessler, M. (1999). *Hey* genes: A novel subfamily of *hairy*- and *Enhancer of split*-related genes specifically expressed during mouse embryogenesis. *Mech. Dev.* **85,** 173–177.

León, Y., Miner, C., Represa, J., and Giraldez, F. (1992). Myb p75 oncoprotein is expressed in developing otic and epibranchial placodes. *Dev. Biol.* **153,** 407–410.

Li, H. S., Yang, J. M., Jacobson, R. D., Pasko, D., and Sundin, O. (1994). *Pax-6* is first expressed in a region of ectoderm anterior to the early neural plate: Implications for stepwise determination of the lens. *Dev. Biol.* **162,** 181–194.

Li, X., Cvekl, A., Bassnett, S., and Piatigorsky, J. (1997). Lens-preferred activity of chicken δ1- and δ2-crystallin enhancers in transgenic mice and evidence for retinoic acid-responsive regulation of the δ1-crystallin gene. *Dev. Genet.* **20,** 258–266.

Liedke, K. B. (1951). Lens competence in *Amblystoma punctatum*. *J. Exp. Zool.* **117,** 573–592.

Liedke, K. B. (1955). Studies on lens induction in *Amblystoma punctatum*. *J. Exp. Zool.* **130,** 353–379.

Liem, K. F., Jr., Tremml, G., Roelink, H., and Jessell, T. M. (1995). Dorsal differentiation of neural plate cells induced by BMP-mediated signals from epidermal ectoderm. *Cell* **82,** 969–979.

Liem, K. F., Jr., Tremml, G., and Jessell, T. M. (1997). A role for the roof plate and its resident TGFb-related proteins in neuronal patterning in the dorsal spinal cord. *Cell* **91,** 127–138.

Lin, C. R., Kioussi, C., O'Connell, S., Briata, P., Szeto, D., Liu, F., Izpisua-Belmonte, J. C., and Rosenfeld, M. G. (1999). Pitx2 regulates lung asymmetry, cardiac positioning and pituitary and tooth morphogenesis. *Nature* **401,** 279–282.

Lo, L., Morin, X., Brunet, J. F., and Anderson, D. J. (1999). Specification of neurotransmitter identity by Phox2 proteins in neural crest stem cells. *Neuron* **22,** 693–705.

Logan, C., Wingate, R. J., McKay, I. J., and Lumsden, A. (1998). *Tlx-1* and *Tlx-3* homeobox gene expression in cranial sensory ganglia and hindbrain of the chick embryo: Markers of patterned connectivity. *J. Neurosci.* **18,** 5389–5402.

Lohnes, D., Mark, M., Mendelsohn, C., Dolle, P., Dierich, A., Gorry, P., Gansmuller, A., and Chambon, P. (1994). Function of the retinoic acid receptors (RARs) during development (I). Craniofacial and skeletal abnormalities in RAR double mutants. *Development* **120,** 2723–2748.

Lombardo, A., and Slack, J. M. (1998). Postgastrulation effects of fibroblast growth factor on *Xenopus* development. *Dev. Dyn.* **212,** 75–85.

Lombardo, A., Isaacs, H. V., and Slack, J. M. (1998). Expression and functions of FGF-3 in *Xenopus* development. *Int. J. Dev. Biol.* **42,** 1101–1107.

London, C., Akers, R., and Phillips, C. (1988). Expression of Epi 1, an epidermis-specific marker in *Xenopus laevis* embryos, is specified prior to gastrulation. *Dev. Biol.* **129,** 380–389.

Long, Q., Quint, E., Lin, S., and Ekker, M. (2000). The zebrafish *scyba* gene encodes a novel CXC-type chemokine with distinctive expression patterns in the vestibulo-acoustic system during embryogenesis. *Mech. Dev.* **97,** 183–186.

Lutz, B., Kuratani, S., Rugarli, E. I., Wawersik, S., Wong, C., Bieber, F. R., Ballabio, A., and Eichele, G. (1994). Expression of the Kallmann syndrome gene in human fetal brain and in the manipulated chick embryo. *Hum. Mol. Genet.* **3,** 1717–1723.

Ma, Q., Chen, Z., del Barco Barrantes, I., de la Pompa, J. L., and Anderson, D. J. (1998). *neurogenin1* is essential for the determination of neuronal precursors for proximal cranial sensory ganglia. *Neuron* **20,** 469–482.

Ma, Q., Fode, C., Guillemot, F., and Anderson, D. J. (1999). NEUROGENIN1 and NEUROGENIN2 control two distinct waves of neurogenesis in developing dorsal root ganglia. *Genes Dev.* **13,** 1717–1728.

MacKenzie, A., Ferguson, M. W., and Sharpe, P. T. (1991). *Hox-7* expression during murine craniofacial development. *Development* **113,** 601–611.

Maden, M., Gale, E., Kostetskii, I., and Zile, M. (1996). Vitamin A-deficient quail embryos have half a hindbrain and other neural defects. *Curr. Biol.* **6,** 417–426.

Magrassi, L., and Graziadei, P. P. (1985). Interaction of the transplanted olfactory placode with the optic stalk and the diencephalon in *Xenopus laevis* embryos. *Neuroscience* **15,** 903–921.

Magrassi, L., and Graziadei, P. P. (1996). Lineage specification of olfactory neural precursor cells depends on continuous cell interactions. *Brain Res. Dev.* **96,** 11–27.

Mahmood, R., Bresnick, J., Hornbruch, A., Mahony, C., Morton, N., Colquhoun, K., Martin, P., Lumsden, A., Dickson, C., and Mason, I. (1995a). A role for FGF-8 in the initiation and maintenance of vertebrate limb bud outgrowth. *Curr. Biol.* **5,** 797–806.

Mahmood, R., Kiefer, P., Guthrie, S., Dickson, C., and Mason, I. (1995b). Multiple roles for FGF-3 during cranial neural development in the chicken. *Development* **121,** 1399–1410.

Mahmood, R., Mason, I. J., and Morriss-Kay, G. M. (1996). Expression of *Fgf-3* in relation to hindbrain segmentation, otic pit position and pharyngeal arch morphology in normal and retinoic acid-exposed mouse embryos. *Anat. Embryol. (Berl.)* **194,** 13–22.

Malicki, J., Schier, A. F., Solnica-Krezel, L., Stemple, D. L., Neuhauss, S. C., Stainier, D. Y., Abdelilah, S., Rangini, Z., Zwartkruis, F., and Driever, W. (1996). Mutations affecting development of the zebrafish ear. *Development* **123,** 275–283.

Mallamaci, A., Di Blas, E., Briata, P., Boncinelli, E., and Corte, G. (1996). OTX2 homeoprotein in the developing central nervous system and migratory cells of the olfactory area. *Mech. Dev.* **58,** 165–178.

Mallamaci, A., Iannone, R., Briata, P., Pintonello, L., Mercurio, S., Boncinelli, E., and Corte, G. (1998). EMX2 protein in the developing mouse brain and olfactory area. *Mech. Dev.* **77,** 165–172.

Mancilla, A., and Mayor, R. (1996). Neural crest formation in *Xenopus laevis*: Mechanisms of *Xslug* induction. *Dev. Biol.* **177,** 580–589.

Mangold, H. (1931). Das Determinationsproblem. III. Das Wirbeltierauge in der Entwicklung und Regeneration. *Ergeb. Biol.* **7,** 193–403.

Mangold, O. (1929). Die Induktionsfähigkeit der Medullarplatte und ihrer Bezirke. *Verh. Dtsch. Zool. Ges.* **33,** 166–173.

Mansour, S. L., Goddard, J. M., and Capecchi, M. R. (1993). Mice homozygous for a targeted disruption of the proto-oncogene *int-2* have developmental defects in the tail and inner ear. *Development* **117,** 13–28.

Manzanares, M., Trainor, P. A., Nonchev, S., Ariza-McNaughton, L., Brodie, J., Gould, A., Marshall, H., Morrison, A., Kwan, C. T., Sham, M. H., Wilkinson, D. G., and Krumlauf, R. (1999). The role of *kreisler* in segmentation during hindbrain development. *Dev. Biol.* **211,** 220–237.

Marcelle, C., Wolf, J., and Bronner-Fraser, M. (1995). The *in vivo* expression the the FGF receptor FREK mRNA in avian myoblasts suggests a role in muscle growth and differentiation. *Dev. Biol.* **172,** 100–114.

Marchant, L., Linker, C., Ruiz, P., Guerrero, N., and Mayor, R. (1998). The inductive properties of mesoderm suggest that the neural crest cells are specified by a BMP gradient. *Dev. Biol.* **198,** 319–329.

Marin-Padilla, M., and Amieva B, M. R. (1989). Early neurogenesis of the mouse olfactory nerve: Golgi and electron microscopic studies. *J. Comp. Neurol.* **288,** 339–352.

Mathers, P. H., Miller, A., Doniach, T., Dirksen, M. L., and Jamrich, M. (1995). Initiation of anterior head-specific gene expression in uncommitted ectoderm of *Xenopus laevis* by ammonium chloride. *Dev. Biol.* **171,** 641–654.

Mathers, P. H., Grinberg, A., Mahon, K. A., and Jamrich, M. (1997). The *Rx* homeobox gene is essential for vertebrate eye development. *Nature* **387,** 603–607.

Matsuo, I., Kuratani, S., Kimura, C., Takeda, N., and Aizawa, S. (1995). Mouse *Otx2* functions in the formation and patterning of rostral head. *Genes Dev.* **9,** 2646–2658.

Matsuo, T., Osumi-Yamashita, N., Noji, S., Ohuchi, H., Koyama, E., Myokai, F., Matsuo, N., Taniguchi, S., Doi, H., Iseki, S., Ninomiya, Y., Fujiwara, M., Watanabe, T., and Eto, K. (1993). A mutation in the *Pax-6* gene in rat *small eye* is associated with impaired migration of midbrain crest cells. *Nat. Gen.* **3,** 299–304.

May, R. M. (1927). Modifications des centres nerveux dues à la transplantation de l'oeil et de l'organe olfactif chez les embryons d'Anoures. *Arch. Biol.* **37,** 335–396.

Mayor, R., Guerrero, N., and Martínez, C. (1997). Role of FGF and noggin in neural crest induction. *Dev. Biol.* **189,** 1–12.

McAvoy, J. W. (1980). Induction of the eye lens. *Differentiation* **17,** 137–149.

McAvoy, J. W., Chamberlain, C. G., de Iongh, R. U., Hales, A. M., and Lovicu, F. J. (1999). Lens development. *Eye* **13,** 425–437.

McKay, I. J., Muchamore, I., Krumlauf, R., Maden, M., Lumsden, A., and Lewis, J. (1994). The *kreisler* mouse: A hindbrain segmentation mutant that lacks two rhombomeres. *Development* **120,** 2199–2211.

McKay, I. J., Lewis, J., and Lumsden, A. (1996). The role of FGF-3 in early inner ear development: An analysis in normal and *kreisler* mutant mice. *Dev. Biol.* **174,** 370–378.

McKeehan, M. S. (1951). Cytological aspects of embryonic lens induction in the chick. *J. Exp. Zool.* **117,** 31–64.

Mendonsa, E. S., and Riley, B. B. (1999). Genetic analysis of tissue interactions required for otic placode induction in the zebrafish. *Dev. Biol.* **206,** 100–112.

Metcalfe, W. K. (1985). Sensory neuron growth cones comigrate with posterior lateral line primordial cells in zebrafish. *J. Comp. Neurol.* **238,** 218–224.

Metcalfe, W. K. (1989). Organization and development of the zebrafish posterior lateral line. *In* "The Mechanosensory Lateral Line. Neurobiology and Evolution" (S. Coombs, P. Görner, and H. Münz, Eds.), pp. 147–159. Springer-Verlag, New York.

Metcalfe, W. K., Myers, P. Z., Trevarrow, B., Bass, M. B., and Kimmel, C. B. (1990). Primary neurons that express the L2/HNK-1 carbohydrate during early development in the zebrafish. *Development* **110,** 491–504.

Metscher, B. D., Northcutt, R. G., Gardiner, D. M., and Bryant, S. V. (1997). Homeobox genes in axolotl lateral line placodes and neuromasts. *Dev. Genes Evol.* **207,** 287–295.

Mic, F. A., Molotkov, A., Fan, X., Cuenca, A. E., and Duester, G. (2000). RALDH3, a retinaldehyde dehydrogenase that generates retinoic acid, is expressed in the ventral retina, otic vesicle and olfactory pit during mouse development. *Mech. Dev.* **97,** 227–230.

Mishima, N., and Tomarev, S. (1998). Chicken *Eyes absent 2* gene: Isolation and expression pattern during development. *Int. J. Dev. Biol.* **42,** 1109–1115.

Miyake, T., von Herbing, I. H., and Hall, B. K. (1997). Neural ectoderm, neural crest, and placodes: Contribution of the otic placode to the ectodermal lining of the embryonic opercular cavity in Atlantic cod (Teleostei). *J. Morphol.* **231,** 231–252.

Mizuno, N., Mochii, M., Yamamoto, T. S., Takahashi, T. C., Eguchi, G., and Okada, T. S. (1999). *Pax-6* and *Prox 1* expression during lens regeneration from *Cynops* iris and *Xenopus* cornea: Evidence for a genetic program common to embryonic lens development. *Differentiation* **65,** 141–149.

Mizuseki, K., Kishi, M., Matsui, M., Nakanishi, S., and Sasai, Y. (1998). *Xenopus* Zic-related-1 and Sox-2, two factors induced by chordin, have distinct activities in the initiation of neural induction. *Development* **125,** 579–587.

Mohun, T., and Sparrow, D. (1997). Early steps in vertebrate cardiogenesis. *Curr. Opin. Genet. Dev.* **7,** 628–633.

Molenaar, M., Brian, E., Roose, J., Clevers, H., and Destrée, O. (2000). Differential expression of the Groucho-related genes 4 and 5 during early development of *Xenopus laevis*. *Mech. Dev.* **91,** 311–315.

Monaghan, A. P., Davidson, D. R., Sime, C., Graham, E., Baldock, R., Bhattacharya, S. S., and Hill, R. E. (1991). The *Msh*-like homeobox genes define domains in the developing vertebrate eye. *Development* **112,** 1053–1061.

Monaghan, A. P., Grau, E., Bock, D., and Schutz, G. (1995). The mouse homolog of the orphan nuclear receptor *tailless* is expressed in the developing forebrain. *Development* **121,** 839–853.

Moody, S. A., and Heaton, M. B. (1983a). Developmental relationships between trigeminal ganglia and trigeminal motoneurons in chick embryos. I. Ganglion development is necessary for motoneuron migration. *J. Comp. Neurol.* **213,** 327–343.

Moody, S. A., and Heaton, M. B. (1983b). Developmental relationships between trigeminal ganglia and trigeminal motoneurons in chick embryos. II. Ganglion axon ingrowth guides motoneuron migration. *J. Comp. Neurol.* **213,** 344–349.

Moody, S. A., Quigg, M. S., and Frankfurter, A. (1989). Development of the peripheral trigeminal system in the chick revealed by

an isotype-specific anti-b-tubulin monoclonal antibody. *J. Comp. Neurol.* **279,** 567–580.

Moore, M. W., Klein, R. D., Farinas, I., Sauer, H., Armanini, M., Phillips, H., Reichardt, L. F., Ryan, A. M., Carver-Moore, K., and Rosenthal, A. (1996). Renal and neuronal abnormalities in mice lacking GDNF. *Nature* **382,** 76–79.

Morin, X., Cremer, H., Hirsch, M.-R., Kapur, R. P., Goridis, C., and Brunet, J.-F. (1997). Defects in sensory and autonomic ganglia and absence of locus coeruleus in mice deficient for the homeobox gene *Phox2a*. *Neuron* **18,** 411–423.

Morrison, A., Hodgetts, C., Gossler, A., Hrabé de Angelis, M., and Lewis, J. (1999). Expression of *Delta1* and *Serrate1* (*Jagged1*) in the mouse inner ear. *Mech. Dev.* **84,** 169–172.

Morsli, H., Choo, D., Ryan, A., Johnson, R., and Wu, D. K. (1998). Development of the mouse inner ear and origin of its sensory organs. *J. Neurosci.* **18,** 3327–3335.

Morsli, H., Tuorto, F., Choo, D., Postiglione, M. P., Simeone, A., and Wu, D. K. (1999). *Otx1* and *Otx2* activities are required for the normal development of the mouse inner ear. *Development* **126,** 2335–2343.

Moury, J. D., and Jacobson, A. G. (1990). The origins of neural crest cells in the axolotl. *Dev. Biol.* **141,** 243–253.

Mucchielli, M. L., Martinez, S., Pattyn, A., Goridis, C., and Brunet, J. F. (1996). *Otlx2*, an *Otx*-related homeobox gene expressed in the pituitary gland and in a restricted pattern in the forebrain. *Mol. Cell. Neurosci.* **8,** 258–271.

Muhr, J., Jessell, T. M., and Edlund, T. (1997). Assignment of early caudal identity to neural plate cells by a signal from caudal paraxial mesoderm. *Neuron* **19,** 487–502.

Mulrenin, E. M., Witkin, J. W., and Silverman, A. J. (1999). Embryonic development of the gonadotropin-releasing hormone (GnRH) system in the chick: A spatio-temporal analysis of GnRH neuronal generation, site of origin, and migration. *Endocrinology* **140,** 422–433.

Münchberg, S. R., Ober, E. A., and Steinbeisser, H. (1999). Expression of the Ets transcription factors *erm* and *pea3* in early zebrafish development. *Mech. Dev.* **88,** 233–236.

Münz, H. (1989). Functional organization of the lateral line periphery. *In* "The Mechanosensory Lateral Line. Neurobiology and Evolution" (S. Coombs, P. Görner, and H. Münz, Eds.), pp. 285–297. Springer-Verlag, New York.

Murakami, S., Kikuyama, S., and Arai, Y. (1992). The origin of the luteinizing hormone-releasing hormone (LHRH) neurons in newts (*Cynops pyrrhogaster*): The effect of olfactory placode ablation. *Cell. TissueRes.* **269,** 21–27.

Muske, L. E. (1993). Evolution of gonadotropin-releasing hormone (GnRH) neuronal systems. *Brain Behav. Evol.* **42,** 215–230.

Myat, A., Henrique, D., Ish-Horowicz, D., and Lewis, J. (1996). A chick homologue of *Serrate* and its relationship with *Notch* and *Delta* homologues during central neurogenesis. *Dev. Biol.* **174,** 233–247.

Narayanan, C. H., and Narayanan, Y. (1978). Determination of the embryonic origin of the mesencephalic nucleus of the trigeminal nerve in birds. *J. Embryol. Exp. Morphol.* **43,** 85–105.

Narayanan, C. H., and Narayanan, Y. (1980). Neural crest and placodal contributions in the development of the glossopharyngeal-vagal complex in the chick. *Anat. Rec.* **196,** 71–82.

Neave, B., Rodaway, A., Wilson, S. W., Patient, R., and Holder, N. (1995). Expression of zebrafish GATA 3 (*gta3*) during gastrulation and neurulation suggests a role in the specification of cell fate. *Mech. Dev.* **51,** 169–182.

Neave, B., Holder, N., and Patient, R. (1997). A graded response to BMP-4 spatially coordinates patterning of the mesoderm and ectoderm in the zebrafish. *Mech. Dev.* **62,** 183–195.

Nguyen, V. H., Schmid, B., Trout, J., Connors, S. A., Ekker, M., and Mullins, M. C. (1998). Ventral and lateral regions of the zebrafish gastrula, including the neural crest progenitors, are established by a *bmp2b/swirl* pathway of genes. *Dev. Biol.* **199,** 93–110.

Nichols, D. H. (1981). Neural crest formation in the head of the mouse embryo as observed using a new histological technique. *J. Embryol. Exp. Morphol.* **64,** 105–120.

Nichols, D. H. (1986). Mesenchyme formation from the trigeminal placodes of the mouse embryo. *Am. J. Anat.* **176,** 19–31.

Niehrs, C. (1999). Head in the WNT: The molecular nature of Spemann's head organizer. *Trends Genet.* **15,** 314–319.

Nieuwkoop, P. D., and Faber, J. (1967). "Normal Table of *Xenopus laevis* (Daudin)." North-Holland, Amsterdam.

Nishiguchi, S., Wood, H., Kondoh, H., Lovell-Badge, R., and Episkopou, V. (1998). *Sox1* directly regulates the γ-crystallin genes and is essential for lens development in mice. *Genes Dev.* **12,** 776–781.

Noden, D. M. (1980). Somatotopic and functional organization of the avian trigeminal ganglion: An HRP analysis in the hatchling chick. *J. Comp. Neurol.* **190,** 405–428.

Norgren, R. B., Ratner, N., and Brackenbury, R. (1992). Development of olfactory nerve glia defined by a monoclonal antibody specific for Schwann cells. *Dev. Dyn.* **194,** 231–238.

Nornes, H. O., Dressler, G. R., Knapik, E. W., Deutsch, U., and Gruss, P. (1990). Spatially and temporally restricted expression of *Pax2* during murine neurogenesis. *Development* **109,** 797–809.

Nornes, S., Clarkson, M., Mikkola, I., Pedersen, M., Bardsley, A., Martinez, J. P., Krauss, S., and Johansen, T. (1998). Zebrafish contains two *pax6* genes involved in eye development. *Mech. Dev.* **77,** 185–196.

Northcutt, R. G. (1996). The origin of craniates: Neural crest, neurogenic placodes, and homeobox genes. *Isr. J. Zool.* **42,** S273–S313.

Northcutt, R. G. (1997). Evolution of gnathostome lateral line ontogenies. *Brain Behav. Evol.* **50,** 25–37.

Northcutt, R. G., and Bemis, W. E. (1993). Cranial nerves of the coelacanth, *Latimeria chalumnae* [Osteichthyes: Sarcopterygii: Actinistia], and comparisons with other craniata. *Brain Behav. Evol.* **42,** 1–76.

Northcutt, R. G., and Brändle, K. (1995). Development of branchiomeric and lateral line nerves in the axolotl. *J. Comp. Neurol.* **355,** 427–454.

Northcutt, R. G., Brändle, K., and Fritzsch, B. (1995). Electroreceptors and mechanosensory lateral line organs arise from single placodes in axolotls. *Dev. Biol.* **168,** 358–373.

Nyholm, N. E. I. (1977). Some morphogenetic features of the adenohypophysial primordium of early *Xenopus laevis* tadpoles. *Cell. Tissue Res.* **180,** 223–230.

O'Rahilly, R., and Müller, F. (1985). The origin of the ectodermal ring in staged human embryos of the first 5 weeks. *Acta Anat.* **122,** 145–157.

Ogino, H., and Yasuda, K. (1998). Induction of lens differentiation by activation of a bZIP transcription factor, L-Maf. *Science* **280,** 115–118.

Ogino, H., and Yasuda, K. (2000). Sequential activation of transcription factors in lens induction. *Dev. Growth Differ.* **42,** 437–448.

Ohto, H., Takizawa, T., Saito, T., Kobayashi, M., Ikeda, K., and Kawakami, K. (1998). Tissue and developmental distribution of *Six* family gene products. *Int. J. Dev. Biol.* **42,** 141–148.

Ohto, H., Kamada, S., Tago, K., Tominaga, S. I., Ozaki, H., Sato, S., and Kawakami, K. (1999). Cooperation of Six and Eya in activation of their target genes through nuclear translocation of Eya. *Mol. Cell. Biol.* **19,** 6815–6824.

Oliver, G., and Gruss, P. (1997). Current views on eye development. *Trends Neurosci.* **20,** 415–421.

Oliver, G., Sosa-Pineda, B., Geisendorf, S., Spana, E. P., Doe, C. Q., and Gruss, P. (1993). *Prox 1*, a *prospero*-related homeobox gene expressed during mouse development. *Mech. Dev.* **44,** 3–16.

Oliver, G., Mailhos, A., Wehr, R., Copeland, N. G., Jenkins, N. A., and Gruss, P. (1995a). *Six3*, a murine homologue of the *sine oculis* gene, demarcates the most anterior border of the developing neural plate and is expressed during eye development. *Development* **121,** 4045–4055.

Oliver, G., Wehr, R., Jenkins, N. A., Copeland, N. G., Cheyette, B. N., Hartenstein, V., Zipursky, S. L., and Gruss, P. (1995b). Homeobox genes and connective tissue patterning. *Development* **121,** 693–705.

Oliver, G., Loosli, F., Köster, R., Wittbrodt, J., and Gruss, P. (1996). Ectopic lens induction in fish in response to the murine homeobox gene. *Six3. Mech. Dev.* **60,** 233–239.

Oppenheimer, J. M. (1938). Potencies for differentiation in the teleostean germ ring. *J. Exp. Zool.* **79,** 185–212.

Orr-Urtreger, A., Avivi, A., Zimmer, Y., Givol, D., Yarden, Y., and Lonai, P. (1990). Developmental expression of *c-kit*, a proto-oncogene encoded by the *W* locus. *Development* **109,** 911–923.

Orts Llorca, F., and Ferrol, N. M. (1961). Determination und Differenzierung der Reichplakoden beim Huhnerembryo. *Roux's Arch. EntwMech.* **153,** 434–442.

Orts-Llorca, F., and Jimenez-Collado, J. (1971). Regulation of the embryo after the extirpation of Hensen's node: Consequences on the differentiation of the otic placode. *Arch. Anat. Histol. Embryol.* **54,** 1–11.

Osumi-Yamashita, N., Ninomiya, Y., Doi, H., and Eto, K. (1994). The contribution of both forebrain and midbrain crest cells to the mesenchyme in the frontonasal mass of mouse embryos. *Dev. Biol.* **164,** 409–419.

Osumi-Yamashita, N., Kuratani, S., Ninomiya, Y., Aoki, K., Iseki, S., Chareonvit, S., Doi, H., Fujiwara, M., Watanabe, T., and Eto, K. (1997). Cranial anomaly of homozygous *rSey* rat is associated with a defect in the migration pathway of midbrain crest cells. *Dev. Growth Differ.* **39,** 53–67.

Pandur, P. D., and Moody, S. A. (2000). *Xenopus Six1* gene is expressed in neurogenic cranial placodes and maintained in the differentiating lateral lines. *Mech. Dev.* **96,** 253–257.

Pannese, M., Lupo, G., Kablar, B., Boncinelli, E., Barsacchi, G., and Vignali, R. (1998). The *Xenopus Emx* genes identify presumptive dorsal telencephalon and are induced by head organizer signals. *Mech. Dev.* **73,** 73–83.

Papalopulu, N. (1995). Regionalization of the forebrain from neural plate to neural tube. *Perspect. Dev. Neurobiol.* **3,** 39–52.

Papalopulu, N., and Kintner, C. (1993). *Xenopus Distal-less* related homeobox genes are expressed in the developing forebrain and are induced by planar signals. *Development* **117,** 961–975.

Pattyn, A., Morin, X., Cremer, H., Goridis, C., and Brunet, J.-F. (1997). Expression and interactions of the two closely related homeobox genes *Phox2a* and *Phox2b* during neurogenesis. *Development* **124,** 4065–4075.

Pattyn, A., Morin, X., Cremer, H., Goridis, C., and Brunet, J. F. (1999). The homeobox gene *Phox2b* is essential for the development of autonomic neural crest derivatives. *Nature* **399,** 366–370.

Penzel, R., Oschwald, R., Chen, Y., Tacke, L., and Grunz, H. (1997). Characterization and early embryonic expression of a neural specific transcription factor *xSOX3* in *Xenopus laevis. Int. J. Dev. Biol.* **41,** 667–677.

Pera, E., and Kessel, M. (1999). Expression of DLX3 in chick embryos. *Mech. Dev.* **89,** 189–193.

Pera, E., Stein, S., and Kessel, M. (1999). Ectodermal patterning in the avian embryo: Epidermis versus neural plate. *Development* **126,** 63–73.

Perez, S. E., Rebelo, S., and Anderson, D. J. (1999). Early specification of sensory neuron fate revealed by expression and function of neurogenins in the chick embryo. *Development* **126,** 1715–1728.

Pettigrew, J. D. (1999). Electroreception in monotremes. *J. Exp. Biol.* **202,** 1447–1454.

Pfeffer, P. L., Gerster, T., Lun, K., Brand, M., and Busslinger, M. (1998). Characterization of three novel members of the zebrafish *Pax2/5/8* family: Dependency of *Pax5* and *Pax8* expression on the *Pax2.1* (*noi*) function. *Development* **125,** 3063–3074.

Piatigorsky, J. (1998). Multifunctional lens crystallins and corneal enzymes. More than meets the eye. *Ann. N. Y. Acad. Sci.* **842,** 7–15.

Pignoni, F., Hu, B., Zavitz, K. H., Xiao, J., Garrity, P. A., and Zipursky, S. L. (1997). The eye-specification proteins So and Eya form a complex and regulate multiple steps in *Drosophila* eye development. *Cell* **91,** 881–891.

Piotrowski, T., and Northcutt, R. G. (1996). The cranial nerves of the Senegal bichir, *Polypterus senegalus* [Osteichthyes: Actinopterygii: Cladistia]. *Brain Behav. Evol.* **47,** 55–102.

Pissarra, L., Henrique, D., and Duarte, A. (2000). Expression of *hes6*, a new member of the Hairy/Enhancer-of-split family, in mouse development. *Mech. Dev.* **95,** 275–278.

Platt, J. B. (1896). Ontogenetic differentiation of the ectoderm in *Necturus*. Study II. On the development of the peripheral nervous system. *Q. J. Microsc. Sci.* **38**, 485–547.

Porter, F. D., Drago, J., Xu, Y., Cheema, S. S., Wassif, C., Huang, S. P., Lee, E., Grinberg, A., Massalas, J. S., Bodine, D., Alt, F., and Westphal, H. (1997). *Lhx2*, a LIM homeobox gene, is required for eye, forebrain, and definitive erythrocyte development. *Development* **124,** 2935–2944.

Prouty, S. M., and Levitt, P. (1993). Immunocytochemical analysis of a novel carbohydrate differentiation antigen (CDA-3C2) associated with olfactory and otic systems during embryogenesis in the rat. *J. Comp. Neurol.* **332,** 444–470.

Püschel, A. W., Gruss, P., and Westerfield, M. (1992). Sequence and expression pattern of *pax-6* are highly conserved between zebrafish and mice. *Development* **114,** 643–651.

Quinn, J. C., West, J. D., and Hill, R. E. (1996). Multiple functions for *Pax6* in mouse eye and nasal development. *Genes Dev.* **10,** 435–446.

Quiring, R., Walldorf, U., Kloter, U., and Gehring, W. J. (1994). Homology of the *eyeless* gene of *Drosophila* to the *Small eye* gene in mice and *Aniridia* in humans. *Science* **265,** 785–789.

Raible, D. W., and Kruse, G. J. (2000). Organization of the lateral line system in embryonic zebrafish. *J. Comp. Neurol.* **421,** 189–198.

Ramón-Cueto, A., and Avila, J. (1998). Olfactory ensheathing glia: Properties and function. *Brain Res. Bull.* **46,** 175–187.

Ramón-Cueto, A., and Valverde, F. (1995). Olfactory bulb ensheathing glia: A unique cell type with axonal growth- promoting properties. *Glia* **14,** 163–173.

Rancourt, D. E., Tsuzuki, T., and Capecchi, M. R. (1995). Genetic interaction between *hoxb-5* and *hoxb-6* is revealed by nonallelic noncomplementation. *Genes Dev.* **9,** 108–122.

Ranzi, S. (1926). L'organo di senso spiracolare dei Selaci. *Pubbl. Stn. Zool. Napoli* **7,** 37–76.

Raven, C. P., and Kloos, J. (1945). Induction by medial and lateral pieces of the archenteron roof with special reference to the determination of the neural crest. *Acta Néerl. Morphol.* **5,** 348–362.

Rawles, M. E. (1936). A study in the localization of organ-forming areas in the chick blastoderm of the head-process stage. *J. Exp. Zool.* **72,** 271–315.

Reaume, A. G., Conlon, R. A., Zirngibl, R., Yamaguchi, T. P., and Rossant, J. (1992). Expression analysis of a *Notch* homologue in the mouse embryo. *Dev. Biol.* **154,** 377–387.

Reifers, F., Böhli, H., Walsh, E. C., Crossley, P. H., Stainier, D. Y., and Brand, M. (1998). *Fgf8* is mutated in zebrafish *acerebellar* (*ace*) mutants and is required for maintenance of midbrain-hindbrain boundary development and somitogenesis. *Development* **125,** 2381–2395.

Represa, J., Sanchez, A., Miner, C., Lewis, J., and Giraldez, F. (1990). Retinoic acid modulation of the early development of the inner ear is associated with the control of c-*fos* expression. *Development* **110,** 1081–1090.

Represa, J., León, Y., Miner, C., and Giraldez, F. (1991). The *int-2* proto-oncogene is responsible for induction of the inner ear. *Nature* **353,** 561–563.

Rex, M., Orme, A., Uwanogho, D., Tointon, K., Wigmore, P. M., Sharpe, P. T., and Scotting, P. J. (1997). Dynamic expression of chicken *Sox2* and *Sox3* genes in ectoderm induced to form neural tissue. *Dev. Dyn.* **209,** 323–332.

Reyer, R. W. (1954). Regeneration of the lens in the amphibian eye. *Q. Rev. Biol.* **29,** 1–46.

Reyer, R. W. (1958a). Studies on lens induction in *Amblystoma punctatum* and *Triturus viridescens viridescens*. I. Transplants of prospective belly ectoderm. *J. Exp. Zool.* **138,** 505–555.

Reyer, R. W. (1958b). Studies on lens induction in *Amblystoma punctatum* and *Triturus viridescens viridescens*. II. Transplants of prospective lateral head ectoderm. *J. Exp. Zool.* **139,** 137–179.

Reyer, R. W. (1962). Differentiation and growth of the embryonic nose, lens and corneal anlagen implanted into the larval eye or dorsal fin in *Amblystoma punctatum*. *J. Exp. Zool.* **151,** 123–153.

Rhinn, M., Dierich, A., Shawlot, W., Behringer, R. R., Le Meur, M., and Ang, S. L. (1998). Sequential roles for *Otx2* in visceral endoderm and neuroectoderm for forebrain and midbrain induction and specification. *Development* **125,** 845–856.

Richardson, G. P., Bartolami, S., and Russell, I. J. (1990). Identification of a 275-kD protein associated with the apical surfaces of sensory hair cells in the avian inner ear. *J. Cell. Biol.* **110,** 1055–1066.

Ring, B. Z., Cordes, S. P., Overbeek, P. A., and Barsh, G. S. (2000). Regulation of mouse lens fiber cell development and differentiation by the *Maf* gene. *Development* **127,** 307–317.

Rinkwitz-Brandt, S., Justus, M., Oldenettel, I., Arnold, H. H., and Bober, E. (1995). Distinct temporal expression of mouse *Nkx-5.1* and *Nkx-5.2* homeobox genes during brain and ear development. *Mech. Dev.* **52,** 371–381.

Roberts, B. L., and Meredith, G. E. (1989). The efferent system. *In* "The Mechanosensory Lateral Line. Neurobiology and Evolution" (S. Coombs, P. Görner, and H. Münz, Eds.), pp. 445–459. Springer-Verlag, New York.

Roberts, C., Platt, N., Streit, A., Schachner, M., and Stern, C. D. (1991). The L5 epitope: An early marker for neural induction in the chick embryo and its involvement in inductive interactions. *Development* **112,** 959–970.

Röhlich, K. (1931). Gestaltungsbewegungen der präsumptiven Epidermis während der Neurulation und Kopfbildung bei *Triton taeniatus*. *Arch. EntwMech. Org. (Wilhelm Roux)* **124,** 66–81.

Rollhäuser-ter Horst, J. (1979). Artificial neural crest formation in amphibia. *Anat. Embryol.* **157,** 113–120.

Rollhäuser-ter Horst, J. (1980). Neural crest replaced by gastrula ectoderm in Amphibia. *Anat. Embryol.* **160,** 203–211.

Romanoff, A. L. (1960). "The Avian Embryo. Structural and Functional Development." Macmillan, New York.

Roose, J., Molenaar, M., Peterson, J., Hurenkamp, J., Brantjes, H., Moerer, P., van de Wetering, M., Destree, O., and Clevers, H. (1998). The *Xenopus* Wnt effector XTcf-3 interacts with Groucho-related transcriptional repressors. *Nature* **395,** 608–612.

Rosenquist, G. C. (1966). A radioautographic study of labeled grafts in the chick blastoderm: development from primitive streak stages to stage 12. *Contrib. Embryol.* **38,** 71–110.

Rosenquist, G. C. (1981). Epiblast origin and early migration of neural crest cells in the chick embryo. *Dev. Biol.* **87,** 201–211.

Rossant, J., Zirngibl, R., Cado, D., Shago, M., and Giguere, V. (1991). Expression of a retinoic acid response element-*hsplacZ* transgene defines specific domains of transcriptional activity during mouse embryogenesis. *Genes Dev.* **5,** 1333–1344.

Rubel, E. W., Popper, A. N., and Fay, R. R. (1997). Development of the Auditory System. Springer–Verlag, New York.

Rubenstein, J. L., Shimamura, K., Martinez, S., and Puelles, L. (1998). Regionalization of the prosencephalic neural plate. *Annu. Rev. Neurosci.* **21,** 445–477.

Rudnick, D. (1932). Thyroid-forming potencies of the early chick blastoderm. *J. Exp. Zool.* **62,** 287–317.

Rudnick, D. (1944). Early history and mechanics of the chick blastoderm. *Q. Rev. Biol.* **19,** 187–212.

Rugarli, E. I. (1999). Kallmann syndrome and the link between olfactory and reproductive development. *Am. J. Hum. Genet.* **65,** 943–948.

Rugarli, E. I., Lutz, B., Kuratani, S. C., Wawersik, S., Borsani, G., Ballabio, A., and Eichele, G. (1993). Expression pattern of the Kallmann syndrome gene in the olfactory system suggests a role in neuronal targeting. *Nat. Genet.* **4,** 19–26.

Ruvinsky, I., Oates, A. C., Silver, L. M., and Ho, R. K. (2000). The evolution of paired appendages in vertebrates: T-box genes in the zebrafish. *Dev. Genes Evol.* **210,** 82–91.

Ryan, K., Butler, K., Bellefroid, E., and Gurdon, J. B. (1998). *Xenopus* eomesodermin is expressed in neural differentiation. *Mech. Dev.* **75,** 155–158.

Sadaghiani, B., and Thiébaud, C. H. (1987). Neural crest development in the *Xenopus laevis* embryo, studied by interspecific transplantation and scanning electron microscopy. *Dev. Biol.* **124,** 91–110.

Saha, M. S., Spann, C. L., and Grainger, R. M. (1989). Embryonic lens induction: More than meets the optic vesicle. *Cell. Differ. Dev.* **28,** 153–171.

Sahly, I., Andermann, P., and Petit, C. (1999). The zebrafish *eya1* gene and its expression pattern during embryogenesis. *Dev. Genes Evol.* **209,** 399–410.

Saint-Jeannet, J.-P., He, X., Varmus, H. E., and Dawid, I. B. (1997). Regulation of dorsal fate in the neuraxis by *Wnt-1* and *Wnt-3a*. *Proc. Natl. Acad. Sci. USA* **94,** 13713–13718.

Sakaguchi, D. S., Janick, L. M., and Reh, T. A. (1997). Basic fibroblast growth factor (FGF-2) induced transdifferentiation of retinal pigment epithelium: Generation of retinal neurons and glia. *Dev. Dyn.* **209,** 387–398.

Savage, R., and Phillips, C. R. (1989). Signals from the dorsal blastopore lip region during gastrulation bias the ectoderm toward a nonepidermal pathway of differentiation in *Xenopus laevis*. *Dev. Biol.* **133,** 157–168.

Schaefer, J. J., Oliver, G., and Henry, J. J. (1999). Conservation of gene expression during embryonic lens formation and cornea-lens transdifferentiation in *Xenopus laevis*. *Dev. Dyn.* **215,** 308–318.

Schilling, T. F., and Kimmel, C. B. (1994). Segment and cell type lineage restrictions during pharyngeal arch development in the zebrafish embryo. *Development* **120,** 483–494.

Schlosser, G., and Northcutt, R. G. (2000). Development of neurogenic placodes in *Xenopus laevis*. *J. Comp. Neurol.* **418,** 121–146.

Schlosser, G., Kintner, C., and Northcutt, R. G. (1999). Loss of ectodermal competence for lateral line placode formation in the direct developing frog *Eleutherodactylus coqui*. *Dev. Biol.* **213,** 354–369.

Schmahl, W., Knoedlseder, M., Favor, J., and Davidson, D. (1993). Defects of neuronal migration and the pathogenesis of cortical malformations are associated with *Small eye* (*Sey*) in the mouse, a point mutation at the *Pax-6*-locus. *Acta Neuropathol.* **86,** 126–135.

Schmalhausen, O. I. (1939). Role of the olfactory sac in the development of the cartilage capsule of the olfactory organ in Urodela. *C. R. Acad. Sci. URSS* **23,** 395–398.

Schultheiss, T. M., Xydas, S., and Lassar, A. B. (1995). Induction of avian cardiac myogenesis by anterior endoderm. *Development* **121,** 4203–4214.

Schultheiss, T. M., Burch, J. B., and Lassar, A. B. (1997). A role for bone morphogenetic proteins in the induction of cardiac myogenesis. *Genes Dev.* **11,** 451–462.

Schwanzel-Fukuda, M., Bick, D., and Pfaff, D. W. (1989). Luteinizing hormone-releasing hormone (LHRH)-expressing cells do not migrate normally in an inherited hypogonadal (Kallmann) syndrome. *Brain Res. Mol. Brain Res.* **6,** 311–326.

Schwanzel-Fukuda, M., and Pfaff, D. W. (1989). Origin of luteinizing hormone-releasing hormone neurons. *Nature* **338,** 161–164.

Sechrist, J., Scherson, T., and Bronner-Fraser, M. (1994). Rhombomere rotation reveals that multiple mechanisms contribute to the segmental pattern of hindbrain neural crest migration. *Development* **120,** 1777–1790.

Selleck, M. A., and Bronner-Fraser, M. (1995). Origins of the avian neural crest: The role of neural plate-epidermal interactions. *Development* **121,** 525–538.

Semina, E. V., Reiter, R. S., and Murray, J. C. (1997). Isolation of a new homeobox gene belonging to the *Pitx*/*Rieg* family: Expression during lens development and mapping to the aphakia region on mouse chromosome 19. *Hum. Mol. Genet.* **6,** 2109–2116.

Semina, E. V., Ferrell, R. E., Mintz-Hittner, H. A., Bitoun, P., Alward, W. L., Reiter, R. S., Funkhauser, C., Daack-Hirsch, S., and Murray, J. C. (1998). A novel homeobox gene *PITX3* is mutated in families with autosomal- dominant cataracts and ASMD. *Nat. Genet.* **19,** 167–170.

Seo, H. C., Drivenes, Ellingsen, S., and Fjose, A. (1998). Expression of two zebrafish homologues of the murine *Six3* gene demarcates the initial eye primordia. *Mech. Dev.* **73,** 45–57.

Serbedzija, G. N., and McMahon, A. P. (1997). Analysis of neural crest cell migration in *Splotch* mice using a neural crest-specific LacZ reporter. *Dev. Biol.* **185,** 139–147.

Servetnick, M., and Grainger, R. M. (1991). Changes in neural and lens competence in *Xenopus* ectoderm: Evidence for an autonomous developmental timer. *Development* **112,** 177–188.

Servetnick, M. D., Cook, T. L., Jr., and Grainger, R. M. (1996). Lens induction in axolotls: Comparison with inductive signalling mechanisms in *Xenopus laevis*. *Int. J. Dev. Biol.* **40,** 755–761.

Sharpe, C., and Goldstone, K. (2000). The control of *Xenopus* embryonic primary neurogenesis is mediated by retinoid signalling in the neurectoderm. *Mech. Dev.* **91,** 69–80.

Sheng, G., and Stern, C. D. (1999). *Gata2* and *Gata3*: Novel markers for early embryonic polarity and for non-neural ectoderm in the chick embryo. *Mech. Dev.* **87,** 213–216.

Sheng, H. Z., Moriyama, K., Yamashita, T., Li, H., Potter, S. S., Mahon, K. A., and Westphal, H. (1997). Multistep control of pituitary organogenesis. *Science* **278,** 1809–1812.

Sheng, H. Z., and Westphal, H. (1999). Early steps in pituitary organogenesis. *Trends Genet.* **15,** 236–240.

Shimamura, K., and Rubenstein, J. L. (1997). Inductive interactions direct early regionalization of the mouse forebrain. *Development* **124,** 2709–2718.

Shimeld, S. M., and Holland, P. W. (2000). Vertebrate innovations. *Proc. Natl. Acad. Sci. USA* **97,** 4449–4452.

Shoji, W., Yee, C. S., and Kuwada, J. Y. (1998). Zebrafish semaphorin Z1a collapses specific growth cones and alters their pathway in vivo. *Development* **125,** 1275–1283.

Simeone, A., Gulisano, M., Acampora, D., Stornaiuolo, A., Rambaldi, M., and Boncinelli, E. (1992). Two vertebrate homeobox genes related to the *Drosophila empty spiracles* gene are expressed in the embryonic cerebral cortex. *EMBO J.* **11,** 2541–2550.

Sive, H. L., Hattori, K., and Weintraub, H. (1989). Progressive determination during formation of the anteroposterior axis in *Xenopus laevis*. *Cell* **58,** 171–180.

Smith, P. E. (1920). The pigmentary, growth, and endocrine disturbance induced in the anuran tadpole by the early ablation of the pars buccalis of the hypophysis. *Am. Anat. Memoirs* **11,** 1–151.

Smith, S. C., Lannoo, M. J., and Armstrong, J. B. (1990). Development of the mechanoreceptive lateral-line system in the axolotl: Placode specification, guidance of migration, and the origin of neuromast polarity. *Anat. Embryol.* **182,** 171–180.

Smith, S. C., Graveson, A. C., and Hall, B. K. (1994). Evidence for a developmental and evolutionary link between placodal ectoderm and neural crest. *J. Exp. Zool.* **270,** 292–301.

Solloway, M. J., and Robertson, E. J. (1999). Early embryonic lethality in *Bmp5*;*Bmp7* double mutant mice suggests functional redundancy within the 60A subgroup. *Development* **126,** 1753–1768.

Sommer, L., Ma, Q., and Anderson, D. J. (1996). *neurogenins*, a novel family of *atonal*-related bHLH transcription factors, are putative mammalian neuronal determination genes that reveal progenitor cell heterogeneity in the developing CNS and PNS. *Mol. Cell. Neurosci.* **8,** 221–241.

Song, J. K., and Northcutt, R. G. (1991). Morphology, distribution and innervation of the lateral-line receptors of the Florida gar, *Lepisosteus platyrhincus*. *Brain Behav. Evol.* **37,** 10–37.

Soussi-Yanicostas, N., Hardelin, J. P., Arroyo-Jimenez, M. M., Ardouin, O., Legouis, R., Levilliers, J., Traincard, F., Betton, J. M., Cabanie, L., and Petit, C. (1996). Initial characterization of anosmin-1, a putative extracellular matrix protein synthesized by definite neuronal cell populations in the central nervous system. *J. Cell. Sci.* **109,** 1749–1757.

Spemann, H. (1901). Uber Korrelationen in der Entwicklung des Auges. *Verh. Anat. Ges. Vers. Bonn* **15,** 61–79.

Stark, M. R., Sechrist, J., Bronner-Fraser, M., and Marcelle, C. (1997). Neural tube-ectoderm interactions are required for trigeminal placode formation. *Development* **124,** 4287–4295.

Stark, M. R., Biggs, J. J., Schoenwolf, G. C., and Rao, M. S. (2000). Characterization of avian *frizzled* genes in cranial placode development. *Mech. Dev.* **93,** 195–200.

Stein, K. F. (1929). Early embryonic differentiation of the chick hypophysis as shown in chorio-allantoic grafts. *Anat. Rec.* **43,** 221–237.

Stone, L. S. (1922). Experiments on the development of the cranial ganglia and the lateral line sense organs in *Amblystoma punctatum*. *J. Exp. Zool.* **35,** 421–496.

Stone, L. S. (1924). Experiments on the transplantation of placodes of the cranial ganglia in the amphibian embryo. I. Heterotopic transplantations of the ophthalmic placode upon the head of *Amblystoma punctatum*. *J. Comp. Neurol.* **38,** 73–105.

Stone, L. S. (1928a). Experiments on the transplantation of placodes of the cranial ganglia in the amphibian embryo. II. Heterotopic transplantations of the ophthalmic placode upon the head and body of *Amblystoma punctatum*. *J. Comp. Neurol.* **47,** 91–116.

Stone, L. S. (1928b). Experiments on the transplantation of placodes of the cranial ganglia in the amphibian embryo. III. Preauditory and postauditory placodal materials interchanged. *J. Comp. Neurol.* **47,** 117–154.

Stone, L. S. (1928c). Primitive lines in *Amblystoma* and their relation to the migratory lateral line primordia. *J. Comp. Neurol.* **45,** 169–190.

Stone, L. S. (1929). Experiments on the transplantation of placodes of the cranial ganglia in the amphibian embryo. IV. Heterotopic transplantations of the postauditory placodal material upon the head and body of *Amblystoma punctatum*. *J. Comp. Neurol.* **48,** 311–330.

Stone, L. S. (1931). Induction of the ear by the medulla and its relation to experiments on the lateralis system in Amphibia. *Science* **74,** 577.

Stone, L. S. (1933). The development of lateral-line sense organs in amphibians observed in living and vital-stained preparations. *J. Comp. Neurol.* **57,** 507–540.

Stone, L. S. (1937). Further experimental studies of the development of lateral-line sense organs in amphibians observed in living preparations. *J. Comp. Neurol.* **68,** 83–115.

Storey, K. G., Crossley, J. M., De Robertis, E. M., Norris, W. E., and Stern, C. D. (1992). Neural induction and regionalisation in the chick embryo. *Development* **114,** 729–741.

Stout, R. P., and Graziadei, P. P. (1980). Influence of the olfactory placode on the development of the brain in *Xenopus laevis* (Daudin). I. Axonal growth and connections of the transplanted olfactory placode. *Neuroscience* **5,** 2175–2186.

Street, S. F. (1937). The differentiation of the nasal area of the chick embryo in grafts. *J. Exp. Zool.* **77,** 49–80.

Streit, A., and Stern, C. D. (1999a). Establishment and maintenance of the border of the neural plate in the chick: involvement of FGF and BMP activity. *Mech. Dev.* **82,** 51–66.

Streit, A., and Stern, C. D. (1999b). Neural induction. A bird's eye view. *Trends Genet.* **15,** 20–24.

Streit, A., Lee, K. J., Woo, I., Roberts, C., Jessell, T. M., and Stern, C. D. (1998). Chordin regulates primitive streak development and the stability of induced neural cells, but is not sufficient for neural induction in the chick embryo. *Development* **125,** 507–519.

Streit, A., Berliner, A. J., Papanayotou, C., Sirulnik, A., and Stern, C. D. (2000). Initiation of neural induction by FGF signalling before gastrulation. *Nature* **406,** 74–78.

Sulik, K. K., Johnston, M. C., Smiley, S. J., Speight, H. S., and Jarvis, B. E. (1987). Mandibulofacial dysostosis (Treacher Collins syndrome): A new proposal for its pathogenesis. *Am. J. Med. Genet.* **27,** 359–372.

Sullivan, C. H., Marker, P. C., Thorn, J. M., and Brown, J. D. (1998). Reliability of δ-crystallin as a marker for studies of chick lens induction. *Differentiation* **64,** 1–9.

Swanson, G. J., Howard, M., and Lewis, J. (1990). Epithelial autonomy in the development of the inner ear of a bird embryo. *Dev. Biol.* **137,** 243–257.

Székely, G. (1959). Functional specificity of cranial sensory neuroblasts in Urodela. *Acta Biol. Acad. Sci. Hung.* **10,** 107–115.

Szeto, D. P., Ryan, A. K., O'Connell, S. M., and Rosenfeld, M. G. (1996). P-OTX: A PIT-1-interacting homeodomain factor expressed during anterior pituitary gland development. *Proc. Natl. Acad. Sci. USA* **93,** 7706–7710.

Szeto, D. P., Rodriguez-Esteban, C., Ryan, A. K., O'Connell, S. M., Liu, F., Kioussi, C., Gleiberman, A. S., Izpisua-Belmonte, J. C., and Rosenfeld, M. G. (1999). Role of the *Bicoid*-related homeodomain factor *Pitx1* in specifying hindlimb morphogenesis and pituitary development. *Genes Dev.* **13,** 484–494.

Taira, M., Hayes, W. P., Otani, H., and Dawid, I. B. (1993). Expression of LIM class homeobox gene *Xlim-3* in *Xenopus* development is limited to neural and neuroendocrine tissues. *Dev. Biol.* **159,** 245–256.

Takabatake, Y., Takabatake, T., and Takeshima, K. (2000). Conserved and divergent expression of *T-box* genes *Tbx2-Tbx5* in. *Xenopus. Mech. Dev.* **91,** 433–437.

Takuma, N., Sheng, H. Z., Furuta, Y., Ward, J. M., Sharma, K., Hogan, B. L., Pfaff, S. L., Westphal, H., Kimura, S., and Mahon, K. A. (1998). Formation of Rathke's pouch requires dual induction from the diencephalon. *Development* **125,** 4835–4840.

Tam, P. P. (1989). Regionalisation of the mouse embryonic ectoderm: allocation of prospective ectodermal tissues during gastrulation. *Development* **107,** 55–67.

Tamarin, A., and Boyde, A. (1977). Facial and visceral arch development in the mouse embryo: a study by scanning electron microscopy. *J. Anat.* **124,** 563–580.

Tan, C. K., and Gopalakrishnakone, P. (1988). Infrared sensory neurons in the trigeminal ganglia of the python (*Python reticulatus*)—a horseradish peroxidase study. *Neurosci. Lett.* **86,** 251–256.

Tannahill, D., Isaacs, H. V., Close, M. J., Peters, G., and Slack, J. M. (1992). Developmental expression of the Xenopus int-2 (FGF-3) gene: Activation by mesodermal and neural induction. *Development* **115,** 695–702.

Tarozzo, G., Peretto, P., and Fasolo, A. (1995). Cell migration from the olfactory placode and the ontogeny of the neuroendocrine compartments. *Zool. Sci.* **12,** 367–383.

Thiery, J. P., Duband, J. L., Rutishauser, U., and Edelman, G. M. (1982). Cell adhesion molecules in early chicken embryogenesis. *Proc. Natl. Acad. Sci. USA* **79,** 6737–6741.

Tini, M., Otulakowski, G., Breitman, M. L., Tsui, L. C., and Giguere, V. (1993). An everted repeat mediates retinoic acid induction of the γF-crystallin gene: Evidence of a direct role for retinoids in lens development. *Genes Dev.* **7,** 295–307.

Tiveron, M. C., Hirsch, M. R., and Brunet, J. F. (1996). The expression pattern of the transcription factor Phox2 delineates synaptic pathways of the autonomic nervous system. *J. Neurosci.* **16,** 7649–7660.

Tomarev, S. I., Sundin, O., Banerjee-Basu, S., Duncan, M. K., Yang, J. M., and Piatigorsky, J. (1996). Chicken homeobox gene *Prox 1* related to *Drosophila prospero* is expressed in the developing lens and retina. *Dev. Dyn.* **206,** 354–367.

Tomita, K., Ishibashi, M., Nakahara, K., Ang, S. L., Nakanishi, S., Guillemot, F., and Kageyama, R. (1996). Mammalian *hairy* and *Enhancer of split* homolog 1 regulates differentiation of retinal neurons and is essential for eye morphogenesis. *Neuron* **16,** 723–734.

Ton, C. C., Hirvonen, H., Miwa, H., Weil, M. M., Monaghan, P., Jordan, T., van Heyningen, V., Hastie, N. D., Meijers-Heijboer, H., Drechsler, M., *et al.* (1991). Positional cloning and characterization of a paired box- and homeobox-containing gene from the *aniridia* region. *Cell* **67,** 1059–1074.

Tongiorgi, E., Bernhardt, R. R., and Schachner, M. (1995). Zebrafish neurons express two L1-related molecules during early axonogenesis. *J. Neurosci. Res.* **42,** 547–561.

Torres, M., and Giraldez, F. (1998). The development of the vertebrate inner ear. *Mech. Dev.* **71,** 5–21.

Torres, M., Gómez-Pardo, E., and Gruss, P. (1996). *Pax2* contributes to inner ear patterning and optic nerve trajectory. *Development* **122,** 3381–3391.

Trampusch, H.A.L. (1941). On ear induction. *Acta Neerl. Morph. Norm. Path.* **4,** 195–213.

Treier, M., and Rosenfeld, M. G. (1996). The hypothalamic-pituitary axis: Co-development of two organs. *Curr. Opin. Cell. Biol.* **8,** 833–843.

Treier, M., Gleiberman, A. S., O'Connell, S. M., Szeto, D. P., McMahon, J. A., McMahon, A. P., and Rosenfeld, M. G. (1998). Multistep signalling requirements for pituitary organogenesis in vivo. *Genes Dev.* **12,** 1691–1704.

Treisman, J. E. (1999). A conserved blueprint for the eye? *Bioessays* **21,** 843–850.

Tremblay, P., Kessel, M., and Gruss, P. (1995). A transgenic neuroanatomical marker identifies cranial neural crest deficiencies associated with the *Pax3* mutant. *Splotch. Dev. Biol.* **171,** 317–329.

Trupp, M., Ryden, M., Jornvall, H., Funakoshi, H., Timmusk, T., Arenas, E., and Ibanez, C. F. (1995). Peripheral expression and biological activities of GDNF, a new neurotrophic factor for avian and mammalian peripheral neurons. *J. Cell. Biol.* **130,** 137–148.

Tweedle, C. D. (1977). Ultrastructure of lateral line organs in aneurogenic amphibian larvae (*Ambystoma*). *Cell. Tissue Res.* **185,** 191–197.

Twitty, V. (1955). Eye. *In* "Analysis of Development" (B. H. Willier, P. A. Weiss, and V. Hamburger, Eds.), pp. 402–414. W. B. Saunders, Philadelphia.

Ueda, Y., and Okada, T. S. (1986). Transient expression of a "lens-specific" gene, δ-crystallin, in the embryonic chicken adenohypophysis. *Cell. Differ.* **19,** 179–185.

Valarché, I., Tissier-Seta, J. P., Hirsch, M. R., Martinez, S., Goridis, C., and Brunet, J. F. (1993). The mouse homeodomain protein Phox2 regulates *Ncam* promoter activity in concert with Cux/CDP and is a putative determinant of neurotransmitter phenotype. *Development* **119,** 881–896.

Van Esch, H., Groenen, P., Nesbit, M. A., Schuffenhauer, S., Lichtner, P., Vanderlinden, G., Harding, B., Beetz, R., Bilous, R. W., Holdaway, I., Shaw, N. J., Fryns, J. P., Van de Ven, W., Thakker, R. V., and Devriendt, K. (2000). GATA3 haploinsufficiency causes human HDR syndrome. *Nature* **406,** 419–422.

van Oostrom, C. G., and Verwoerd, C. D. A. (1972). The origin of the olfactory placode. *Acta Morphol. Neerl. Scand.* **9,** 160.

van Wijhe, J. W. (1883). Uber die Mesodermsegmente und die Entwicklung der Nerven des Selachierkopfes. *Verh. Acad. Setensch. (Amsterdam)* **22**(E), 1–50.

Vasiliauskas, D., and Stern, C. D. (2000). Expression of mouse *HES-6*, a new member of the Hairy/Enhancer of split family of bHLH transcription factors. *Mech. Dev.* **98,** 133–137.

Veitch, E., Begbie, J., Schilling, T. F., Smith, M. M., and Graham, A. (1999). Pharyngeal arch patterning in the absence of neural crest. *Curr. Biol.* **9,** 1481–1484.

Vendrell, V., Carnicero, E., Giraldez, F., Alonso, M. T., and Schimmang, T. (2000). Induction of inner ear fate by FGF3. *Development* **127,** 2011–2019.

Venzke, W. G. (1942). The embryological development and physiology of the endocrine glands of the common fowl (*Gallus domesticus*). *Iowa State Coll. J. Sci.* **17,** 145–148.

Verwoerd, C. D. A., and van Oostrom, C. G. (1979). Cephalic neural crest and placodes. *Adv. Anat. Embryol. Cell. Biol.* **58,** 1–75.

Vitali, G. (1911). Di un interessante derivato della prima fessura branchiale nel passero. *Anat. Anz.* **39,** 219–224.

Vitali, G. (1925). Il comportamento dell'organo della prima fessura branchiale (placode epibranchiale) nei Selaci. *Monit. Zool. Ital.* **36,** 122–130.

Vitali, G. (1926). La façon de se comporter du placode de la première fente branchiale (placode épibranchial) dans la série des vertébrés. *Arch. Ital. Biol.* **76,** 94–106.

Vogel, K. S. (1992). Origins and early development of vertebrate cranial sensory neurons. *In* "Sensory Neurons: Diversity, Development, and Plasticity" (S. A. Scott, Ed.), pp. 171–193. Oxford Univ. Press, Oxford, UK.

Vogel, K. S., and Davies, A. M. (1993). Heterotopic transplantation of presumptive placodal ectoderm changes the fate of sensory neuron precursors. *Development* **119,** 263–276.

Vogel-Höpker, A., Momose, T., Rohrer, H., Yasuda, K., Ishihara, L., and Rapaport, D. H. (2000). Multiple functions of fibroblast growth factor-8 (FGF-8) in chick eye development. *Mech. Dev.* **94,** 25–36.

von Bartheld, C. S. (1990). Development and innervation of the paratympanic organ (Vitali organ) in chick embryos. *Brain Behav. Evol.* **35,** 1–15.

von Kupffer, C. (1891). The development of the cranial nerves of vertebrates. *J. Comp. Neurol.* **1,** 246–264; 315–332.

Waddington, C. H. (1937). The determination of the auditory placode in the chick. *J. Exp. Biol.* **14,** 232–239.

Waddington, C. H., and Cohen, A. (1936). Experiments on the development of the head of the chick embryo. *J. Exp. Biol.* **13,** 219–236.

Wakamatsu, Y., and Weston, J. A. (1997). Sequential expression and role of Hu-binding proteins during neurogenesis. *Development* **124,** 3449–3460.

Wakamatsu, Y., Maynard, T. M., and Weston, J. A. (2000). Fate determination of neural crest cells by NOTCH-mediated lateral inhibition and asymmetrical cell division during gangliogenesis. *Development* **127,** 2811–2821.

Walker, M. M., Diebel, C. E., Haugh, C. V., Pankhurst, P. M., Montgomery, J. C., and Green, C. R. (1997). Structure and function of the vertebrate magnetic sense. *Nature* **390,** 371–390.

Wall, N. A., and Hogan, B. L. (1995). Expression of bone morphogenetic protein-4 (BMP-4), bone morphogenetic protein-7 (BMP-7), fibroblast growth factor-8 (FGF-8) and sonic hedgehog (SHH) during branchial arch development in the chick. *Mech. Dev.* **53,** 383–392.

Wall, N. A., Jones, C. M., Hogan, B. L. M., and Wright, C. V. E. (1992). Expression and modification of Hox 2.1 protein in mouse embryos. *Mech. Dev.* **37,** 111–120.

Wallis, D. E., Roessler, E., Hehr, U., Nanni, L., Wiltshire, T., Richieri-Costa, A., Gillessen-Kaesbach, G., Zackai, E. H., Rommens, J., and Muenke, M. (1999). Mutations in the homeodomain of the human *SIX3* gene cause holoprosencephaly. *Nat. Genet.* **22,** 196–198.

Walther, C., and Gruss, P. (1991). *Pax-6*, a murine paired box gene, is expressed in the developing CNS. *Development* **113,** 1435–1449.

Wang, W., Van De Water, T., and Lufkin, T. (1998). Inner ear and maternal reproductive defects in mice lacking the *Hmx3* homeobox gene. *Development* **125,** 621–634.

Watkins-Chow, D. E., and Camper, S. A. (1998). How many homeobox genes does it take to make a pituitary gland? *Trends Genet.* **14,** 284–290.

Wawersik, S., and Maas, R. L. (2000). Vertebrate eye development as modeled in *Drosophila. Hum. Mol. Genet.* **9,** 917–925.

Wawersik, S., Purcell, P., Rauchman, M., Dudley, A. T., Robertson, E. J., and Maas, R. (1999). BMP7 acts in murine lens placode development. *Dev. Biol.* **207,** 176–188.

Webb, J. F., and Noden, D. M. (1993). Ectodermal placodes: Contributions to the development of the vertebrate head. *Am. Zool.* **33,** 434–447.

Weinstein, D. C., and Hemmati-Brivanlou, A. (1999). Neural induction. *Annu. Rev. Cell. Dev. Biol.* **15,** 411–433.

Wendling, O., Dennefeld, C., Chambon, P., and Mark, M. (2000). Retinoid signalling is essential for patterning the endoderm of the third and fourth pharyngeal arches. *Development* **127,** 1553–1562.

West-Mays, J. A., Zhang, J., Nottoli, T., Hagopian-Donaldson, S., Libby, D., Strissel, K. J., and Williams, T. (1999). AP-2a transcription factor is required for early morphogenesis of the lens vesicle. *Dev. Biol.* **206,** 46–62.

White, J. C., Shankar, V. N., Highland, M., Epstein, M. L., DeLuca, H. F., and Clagett-Dame, M. (1998). Defects in embryonic hindbrain development and fetal resorption resulting from vitamin A deficiency in the rat are prevented by feeding pharmacological levels of all-trans-retinoic acid. *Proc. Natl. Acad. Sci. USA* **95,** 13459–13464.

Whitfield, T., Haddon, C., and Lewis, J. (1997). Intercellular signals and cell-fate choices in the developing inner ear: Origins of global and of fine-grained pattern. *Sem. Cell. Dev. Biol.* **8,** 239–247.

Whitlock, K. E., and Westerfield, M. (1998). A transient population of neurons pioneers the olfactory pathway in the zebrafish. *J. Neurosci.* **18,** 8919–8927.

Whitlock, K. E., and Westerfield, M. (2000). The olfactory placodes of the zebrafish form by convergence of cellular fields at the edge of the neural plate. *Development* **127,** 3645–3653.

Wigle, J. T., Chowdhury, K., Gruss, P., and Oliver, G. (1999). *Prox1* function is crucial for mouse lens-fibre elongation. *Nat. Genet.* **21,** 318–322.

Wilkinson, D. G., Peters, G., Dickson, C., and McMahon, A. P. (1988). Expression of the FGF-related proto-oncogene *int-2* during gastrulation and neurulation in the mouse. *EMBO J.* **7,** 691–695.

Williams, R., Lendahl, U., and Lardelli, M. (1995). Complementary and combinatorial patterns of *Notch* gene family expression during early mouse development. *Mech. Dev.* **53,** 357–368.

Williams, S. C., Altmann, C. R., Chow, R. L., Hemmati-Brivanlou, A., and Lang, R. A. (1998). A highly conserved lens transcriptional control element from the *Pax-6* gene. *Mech. Dev.* **73,** 225–229.

Wilson, S. I., Graziano, E., Harland, R., Jessell, T. M., and Edlund, T. (2000). An early requirement for FGF signalling in the acquisition of neural cell fate in the chick embryo. *Curr. Biol.* **10,** 421–429.

Winklbauer, R. (1989). Development of the lateral line system in *Xenopus. Prog. Neurobiol.* **32,** 181–206.

Winklbauer, R., and Hausen, P. (1983). Development of the lateral line system in *Xenopus laevis*. I. Normal development and cell movement in the supraorbital system. *J. Embryol. Exp. Morphol.* **76,** 265–281.

Woo, K., and Fraser, S. E. (1997). Specification of the zebrafish nervous system by nonaxial signals. *Science* **277,** 254–257.

Woo, K., and Fraser, S. E. (1998). Specification of the hindbrain fate in the zebrafish. *Dev. Biol.* **197,** 283–296.

Wood, H. B., and Episkopou, V. (1999). Comparative expression of the mouse *Sox1*, *Sox2* and *Sox3* genes from pre-gastrulation to early somite stages. *Mech. Dev.* **86,** 197–201.

Wray, S., Grant, P., and Gainer, H. (1989). Evidence that cells expressing luteinizing hormone-releasing hormone mRNA in the mouse are derived from progenitor cells in the olfactory placode. *Proc. Natl. Acad. Sci. USA* **86,** 8132–8136.

Wride, M. A. (1996). Cellular and molecular features of lens differentiation: A review of recent advances. *Differentiation* **61,** 77–93.

Wright, M. R. (1951). The lateral line system of sense organs. *Q. Rev. Biol.* **26,** 264–280.

Wu, D. K., and Oh, S. H. (1996). Sensory organ generation in the chick inner ear. *J. Neurosci.* **16,** 6454–6462.

Xu, P. X., Woo, I., Her, H., Beier, D. R., and Maas, R. L. (1997). Mouse *Eya* homologues of the *Drosophila eyes absent* gene require *Pax6* for expression in lens and nasal placode. *Development* **124,** 219–231.

Xu, P. X., Adams, J., Peters, H., Brown, M. C., Heaney, S., and Maas, R. (1999). *Eya1*-deficient mice lack ears and kidneys and show abnormal apoptosis of organ primordia. *Nat. Genet.* **23,** 113–117.

Yamamoto, N., Uchiyama, H., Ohki-Hamazaki, H., Tanaka, H., and Ito, H. (1996). Migration of GnRH-immunoreactive neurons from the olfactory placode to the brain: A study using

avian embryonic chimeras. *Brain Res. Dev. Brain Res.* **95,** 234–244.

Yang, L., Zhang, H., Hu, G., Wang, H., Abate-Shen, C., and Shen, M. M. (1998). An early phase of embryonic *Dlx5* expression defines the rostral boundary of the neural plate. *J. Neurosci.* **18,** 8322–8330.

Yntema, C. L. (1933). Experiments on the determination of the ear ectoderm in the embryo of *Amblystoma punctatum*. *J. Exp. Zool.* **65,** 317–357.

Yntema, C. L. (1937). An experimental study of the origin of the cells which constitute the VIIth and VIIIth cranial ganglia and nerves in the embryo of *Amblystoma punctatum*. *J. Exp. Zool.* **75,** 75–101.

Yntema, C. L. (1939). Self-differentiation of heterotopic ear ectoderm in the embryo of *Amblystoma punctatum*. *J. Exp. Zool.* **80,** 1–17.

Yntema, C. L. (1943). An experimental study on the origin of the sensory neurones and sheath cells of the IXth and Xth cranial nerves in *Amblystoma punctatum*. *J. Exp. Zool.* **92,** 93–119.

Yntema, C. L. (1944). Experiments on the origin of the sensory ganglia of the facial nerve in the chick. *J. Comp. Neurol.* **81,** 147–167.

Yntema, C. L. (1950). An analysis of induction of the ear from foreign ectoderm in the salamander embryo. *J. Exp. Zool.* **113,** 211–244.

Yntema, C. L. (1955). Ear and nose. *In* "Analysis of Development" (B. H. Willier, P. A. Weiss, and V. Hamburger, Eds.), pp. 415–428. Saunders, Philadelphia.

Yoshida, M., Suda, Y., Matsuo, I., Miyamoto, N., Takeda, N., Kuratani, S., and Aizawa, S. (1997). *Emx1* and *Emx2* functions in development of dorsal telencephalon. *Development* **124,** 101–111.

Yoshikawa, S., Aota, S., Shirayoshi, Y., and Okazaki, K. (2000). The ActR-I activin receptor protein is expressed in notochord, lens placode and pituitary primordium cells in the mouse embryo. *Mech. Dev.* **91,** 439–444.

Zhang, J., and Jacobson, A. G. (1993). Evidence that the border of the neural plate may be positioned by the interaction between signals that induce ventral and dorsal mesoderm. *Dev. Dyn.* **196,** 79–90.

Zhuo, H., Ichikawa, H., and Helke, C. J. (1997). Neurochemistry of the nodose ganglion. *Prog. Neurobiol.* **52,** 79–107.

Zwilling, E. (1940). An experimental analysis of the development of the anuran olfactory organ. *J. Exp. Zool.* **84,** 291–324.

Zwilling, E. (1941). The determination of the otic vesicle in *Rana pipiens*. *J. Exp. Zool.* **86,** 333–342.

Zygar, C. A., Cook, T.L.J., and Grainger, R. M. (1998). Gene activation during early stages of lens induction in *Xenopus laevis*. *Development* **125,** 3509–3519.

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