strongly dependent on another sensory modality, but they became more difficult to see as the brain size increased and we suggest this may explain their clarity in the possum compared with the tammar.

S2, which lies caudolateral to S1, contains a second non-inverted, small representation of the contralateral body surface. Electrophysiological mapping has identified this area in all species examined: brush-tailed possum (Adey and Kerr, 1954; Coleman *et al.*, 1999; Elston and Manger, 1999; Huffman *et al.*, 1999a), and northern quoll and striped possum (Figure 10.2d) (Huffman *et al.*, 1999a), as well as in American opossums (Huffman *et al.*, 1999a; Karlen and Krubitzer, 2007), although in the quoll it could not be separated from the parietal ventral area. Huffman *et al.* (1999a) reported only a partial representation of the body surface in the species they examined, whereas Elston and Manger (1999) believed it probably contained a complete representation in the brush-tailed possum. The face and oral structures are adjacent to these representations in S1 (Figure 10.2d). While neurons in both S1 and S2 respond to cutaneous stimulation, those in S2 often habituate and their receptive fields are larger compared to those in S1 (Elston and Manger, 1999; Huffman *et al.*, 1999a). S2 is also architectonically distinct, with broad layers 3Cx and 5Cx and an ill-defined layer 4Cx (Adey and Kerr, 1954). In the quoll and the striped possum it stains moderately to darkly for myelin (Huffman *et al.*, 1999a), whereas in the brush-tailed possum there is sparse myelination (Elston and Manger, 1999). Coleman *et al.* (1999) have shown in the brush-tailed possum that inputs can reach S2 directly from the thalamus and are not dependent on a pathway through S1 (Figure 10.2h). That is to say, S1 and S2 are organised in parallel, as they are in a range of placental mammals.

A separate, somatotopically organised field, PV, rostral to S2, has been identified in the brush-tailed possum (Elston and Manger, 1999; Huffman *et al.*, 1999a) and the striped possum (Huffman *et al.*, 1999a). Neurons have larger receptive fields like those of S2 and often habituate (Elston and Manger, 1999). Two additional fields rostral and caudal to S1, R and C, have also been identified in the brush-tailed possum (Elston and Manger, 1999), the quoll and the striped possum (Huffman *et al.*, 1999a). Neurons in these areas respond to high-threshold cutaneous or deep receptors and may correspond to somatosensory areas identified in other mammals, including the Virginia opossum (Beck *et al.*, 1996). R has been proposed to correspond to area 3a of flying foxes and primates and the rostral somatosensory area of rodents, whereas C may correspond to the posterior parietal area of squirrels and posterior parietal cortex of rodents (Karlen and Krubitzer, 2007).

As well as receiving connections from somatosensory thalamus, outlined in the previous section, somatosensory cortex has reciprocal connections with thalamic nuclei. For example, after a deposit of tracer in the whisker area of somatosensory cortex of the tammar, anterograde label coincident with retrogradely labelled cells is seen in VPM and extends into Po (Figure 10.2c) (Marotte *et al.*, 1997). Large lesions of the cortex in the brush-tailed possum, which include somatosensory cortex, result in anterograde degeneration in VPM, VPL and Po (Rockel *et al.*, 1972). The somatosensory cortex has also been shown to receive projections from a number of other thalamic nuclei in the brush-tailed possum and northern quoll, including the ventrolateral, ventromedial, mediodorsal, anteromedial, ventroanterior and extrageniculate nuclei, as well as parts of the centro-intralaminar complex (Ward and Watson, 1973; Haight and Neylon, 1979; Haight and Neylon, 1981a, 1981b).

Corticocortical connections have been studied in the brush-tailed possum. S1 is connected ipsilaterally with other regions of S1, with S2, PV, R, C, posterior parietal cortex, motor cortex and perirhinal cortex (Elston and Manger, 1999). This is similar to that described in the Virginia opossum, except for the connection with the motor cortex (Beck *et al.*, 1996). Connections with other somatosensory areas are largely homotypic. Contralateral connections are similar, but less dense (Heath and Jones, 1971; Elston and Manger, 1999).

## Development

The most notable difference from eutherians is that metatherians are born in an immature state and thus much of development takes place postnatally in the pouch (see Chapters 3 and 14). As for the adult, most information on the developing somatosensory system is known for the trigeminal system and the species for which most is known is the tammar.

### Cutaneous receptors

Merkel cells have been described around the mouth in a number of newborn Australian species: northern quoll *Dasyurus hallucatus* (Figure 10.3a), brush-tailed possum, northern brown bandicoot (Gemmell *et al.*, 1988) and stripe-faced dunnart (Gemmell and Selwood, 1994), as well as in the intervibrissal skin of the newborn tammar (Waite *et al.*, 1994). They are also seen in the glabrous snout skin of the newborn opossum (Jones and Munger, 1985), but were not reported in the body skin of the newborn possum (Lyne *et al.*, 1970). These studies suggest that these sensitive touch receptors assist in the journey from the urinogenital sinus to the pouch and the locating of the teat.

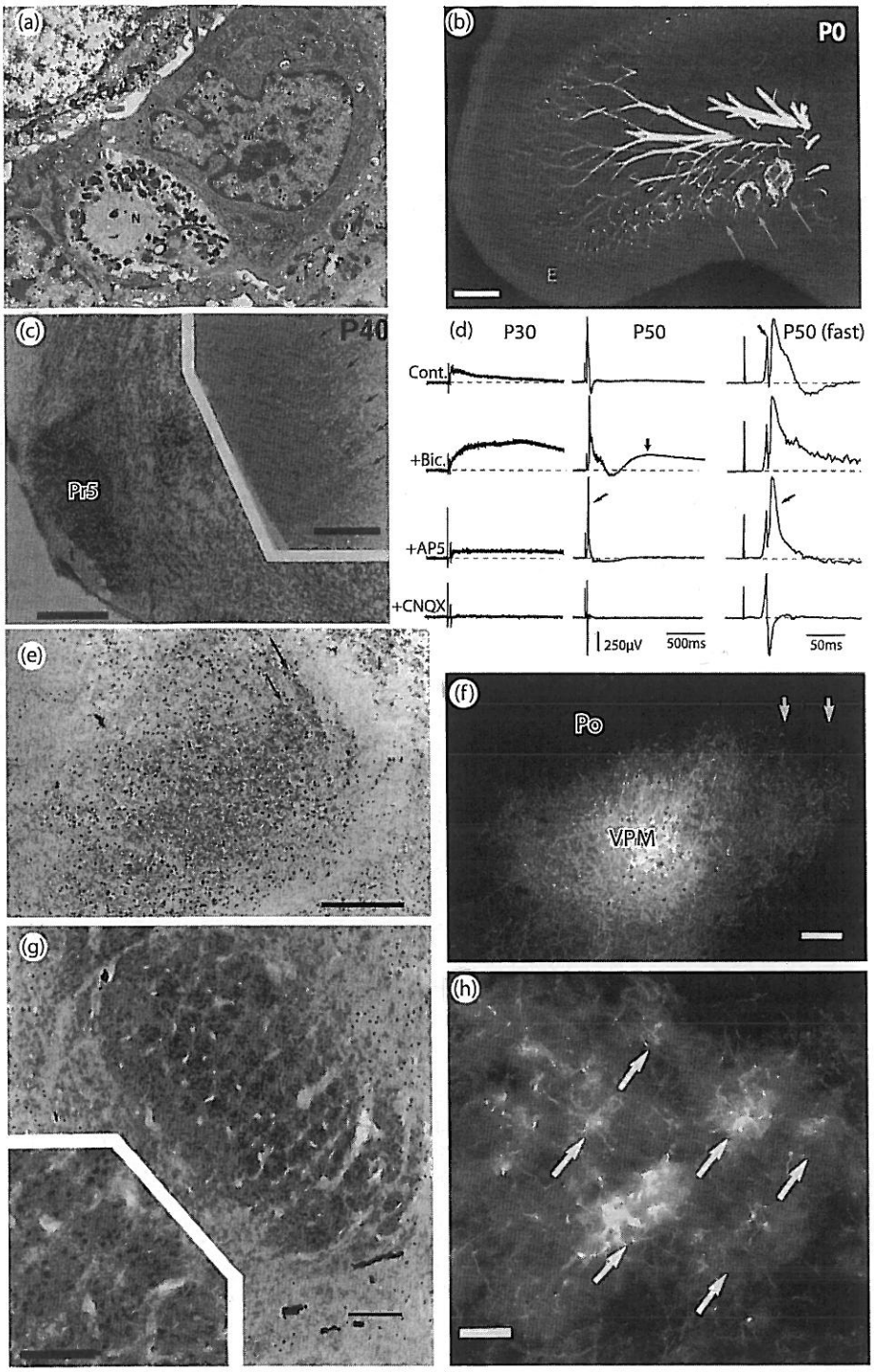


**Figure 10.3** (a) A large sensory nerve (N) in close apposition to a Merkel cell (M) in the newborn quoll (*Dasyurus hallucatus*); x18 500. From Gemmell *et al.* (1988), Figure 3, copyright Springer-Verlag 1988, with the kind permission of Springer Science and Business Media. (b) to (h) tammar wallaby (*Macropus eugenii*). (b) Confocal image (four scans at 25 μm intervals) of a 100 μm section through the maxillary process on postnatal day P0, following 1,1′- dioctadecyl-3,3,3′,3′- tetramethylindocarbocyanine perchlorate (DiI) application to the trigeminal ganglion. Labelled nerve fibres can be seen branching to supply the epidermis (E) and several developing vibrissal follicles (arrows); bar = 200 μm. From Waite *et al.* (1994), Figure 3A, with permission, copyright 1994 Wiley-Liss, Inc. (c) Transverse sections through Pr5 on P40. The characteristic almond shape is apparent; bar = 0.5 mm. Inset: higher-power view of Pr5 reacted for cytochrome oxidase (CO); five oblique lines of increased reactivity (arrows) can be seen; bar = 250 μm. From Waite *et al.* (1994), Figure 12E, with permission, copyright 1994 Wiley-Liss, Inc. (d) Responses recorded in VPM following electrical stimulation of Pr5 in *in vitro* preparations of the brainstem and thalamus

Development of the vibrissal follicles and pelage hairs and their innervation has been described for the tammar (Waite *et al.*, 1994) and there is some information on follicular development in the skin of the brush-tailed possum (Lyne, 1970) and the bandicoot (Lyne, 1957). In the tammar at birth, follicles consist of a solid epidermal peg surrounded by dermal condensations. They and surrounding skin are already innervated by a dense array of trigeminal afferents (Figure 10.3b). Occasional putative Merkel cells can be seen in the epidermis. By P10, a deep vibrissal nerve is recognisable and the follicle contains a dermal papilla. By P30, a hair cone is present and by P35 hairs are seen on the skin surface. The first receptors seen in the follicle are Merkel cells around the waist region, at P48. A blood sinus can first be recognised by P63, but it is not until P119 that the inner conical body and lanceolate and lamellated receptors supplying the mesenchymal sheath and waist region are apparent and the follicle resembles the adult structure. This rate of development is much slower than that seen in rodents, where in the rat the follicle is mature in three weeks (Munger and Rice, 1986). In the tammar this takes 18 to 20 weeks, although the sequence of development is similar.

The epidermal pegs of facial pelage hairs are first seen at P30 and by P48 have increased in number with sparse axon bundles among them. By 63 days, guard hairs can be distinguished from vellus hairs and by 83 days dermal papillae are well developed and hair shafts are common. By P119, pelage hairs have increased, as have axon bundles ramifying among them and at this time innervation of guard hairs is first seen in the form of circular arrays of nerves and lanceolate endings. Innervation of vellus hairs is not obvious as it is in the adult. Development of pelage hair is not complete until around P200 and animals only start leaving the pouch for short periods around P190.

Although not compared in the same species, the development of trigeminal innervation around the mouth and mystacial skin probably precedes that of the rest of the body. Thus, in the brush-tailed possum (Lyne, 1970), the hair follicles on body skin first appear about two days after birth. Groups are formed with a single, central primary follicle, lateral primary follicles and one or two secondary follicles. Nerves are first noted in the dermis at P21 and are not seen around the primary follicles until P54. The features of follicle development are similar in the bandicoot, but occur more rapidly (Lyne, 1957). In the opossum (*Monodelphis domestica*), neurites are present in the glabrous forepaw dermis at birth, but are not seen in the epidermis until P20. From P21 to P30, innervation density increases and Merkel cells appear (Brenowitz *et al.*, 1980).

As in the tammar (Waite *et al.*, 1994), epidermal Merkel cells are also the first receptors to appear between developing vibrissae in the rat and ferret, and precede other receptors in the follicles (Munger and Rice, 1986; Mosconi and Rice, 1993). Similarly, the lag in development and innervation between vibrissal follicles and intervibrissal hairs is also seen in rats and ferrets, although there is more overlap in ferrets (Munger and Rice, 1986; Mosconi *et al.*, 1993). Overall, despite the different developmental time-courses between diverse mammalian species, they show similar sequences of maturation of follicles and

**Caption for Figure 10.3** (Cont.)
at P30 and P50. Under control conditions (Cont.) at P30 (left) the post-synaptic response is prolonged, lasting in excess of 1 s. Addition of bicuculline (+Bic.) to block GABA-A receptors increases the amplitude of the response. Much of this response is NMDA mediated, as shown by the marked decrease in amplitude following addition of 2-amino-5-phosphovaleric acid (+AP5). The remainder of the post-synaptic response is blocked following addition of 6-cyano-7-nitroquinoxaline-2,3-dione (+CNQX) to block non-NMDA-mediated glutamatergic responses, leaving just the presynaptic volley. By P50 (middle, right) the control response is of much shorter duration and is dominated by the fast component. This can be distinguished from the presynaptic volley in the traces shown at a higher temporal resolution on the right. Addition of bicuculline prolonged the tail of the response, but did not affect the initial fast peak. Blockade of NMDA-mediated responses with AP5 reduced the amplitude of the slow component, but had little impact on the fast peak. Addition of CNQX completely blocked the fast peak, leaving just the presynaptic volley. This indicates that there is a change from slow, NMDA-dominated responses in the thalamus at P30 to fast, non-NMDA glutamatergic responses by P50. From Leamey *et al.* (1998), Figure 9, with permission, copyright 1998 Wiley-Liss, Inc. (e) Coronal section through VPM in a P52 wallaby stained for Nissl substance. A hint of segmentation is appearing at the lateral border of VPM (arrows); bar = 400 μm. From Leamey *et al.* (1996), Figure 5b, with permission, copyright 1996 Wiley-Liss, Inc. (f) Coronal section from the caudal half of VPM following the transport of DiI from Pr5 to the thalamus from a P50 animal. Dorsal is at the top, and medial is to the left. The fibres fill and define the borders of the VPM with no label present in the posterior nu. (Po). The fibres are no longer distributed uniformly across the nucleus as at younger ages, but rather show evidence of segregation into fingers at the lateral border (indicated by arrows); bar = 100 μm. Inset shows high power view; bar = 100 μm. From Leamey *et al.* (1998), Figure 2A, with permission, copyright 1998 Wiley-Liss, Inc. (g) Horizontal section through VPM at P75 reacted for CO. VPM is characterised by the presence of circular clusters of highly CO-positive cells; bar = 200 μm. From Leamey *et al.* (1996), Figure 6d, with permission, copyright 1996 Wiley-Liss, Inc. (h) Horizontal section through the VPM from a P75 animal following transport of DiI from Pr5. The afferents have become segregated into circular clusters (arrows). Rostral is at the top, and lateral is to the right; bar = 50 μm. From Leamey *et al.* (1998), Figure 3c, with permission, copyright 1998 Wiley-Liss, Inc.

intervibrissal skin, innervation and pattern of receptor development.

### Muscle receptors

Immature muscle spindles can be recognised in forelimb and cervical musculature of the red kangaroo (*Macropus rufus*) at birth (Kubota *et al.*, 1989), whereas in the tammar they cannot be recognised by routine histology in either the fore- or hindlimbs until around P30 (Harrison, 1991; Harrison and Porter, 1992). In contrast, in the neonatal opossum (*Monodelphis domestica*) they can be seen with immunostaining for spindle fibre types in jaw, forelimb and thoracic muscle (Sciote and Rowlerson, 1998), and those authors speculated that using this method they would be recognised at birth in the tammar. The sequence of spindle development in the tammar (Harrison, 1991) is similar to that of eutherians, but occurs postnatally. Spindles are mature by around P100, prior to the time when pouch young can stand unaided, indicating that they form in the absence of feedback produced by adult forms of posture and locomotion.

### Dorsal root ganglia and dorsal column nuclei

Large dorsal root ganglia are present at all levels in the newborn tammar (Harrison and Porter, 1992), and afferents, labelled from the brachial plexus or sciatic nerve, extend into the dorsal horn (Ho and Stirling, 1998). Dorsal-horn development is more advanced at cervical levels than in the lumbosacral cord (Harrison and Porter, 1992), probably relating to the greater maturity of the forelimbs needed for reaching the pouch, compared with the relatively immature hindlimbs. A rostrocaudal gradient of cord maturation has been described in the opossum (Hughes, 1973) and also occurs in eutherian species such as the rat. Distinct gracile and cuneate fasciculi are present by P17. While the histological development in the tammar is similar to that in eutherians such as the rat, it is more protracted and the tammar cord is far less mature at birth, corresponding at brachial levels to approximately E15 to E16 in the rat (Harrison and Porter, 1992).

Growth of dorsal root afferents into the cord has been studied in the South American opossum, where the first fibres, labelled by *Griffonia simplicifolia* isolectin B4, reach the lateral edge of the dorsal horn at P5 and then spread medially to reach an adult pattern of innervation by P20 (Kitchener *et al.*, 2006). CGRP-labelled afferents lag behind these slightly, reaching the lateral dorsal horn by P7 and then extending medially to give an adult pattern of labelling by P30.

### Trigeminal ganglia and brainstem sensory nuclei

In the tammar, the development of the trigeminal ganglia and nuclei stretches over many months, reflecting the slow, overall pace of development (see Figure 3.6, Chapters 3 and 14). At birth, the trigeminal ganglion is well developed with peripheral processes extending to the facial skin (see above) and central afferents distributed throughout the rostrocaudal extent of the sensory trigeminal nuclear complex (Waite *et al.*, 1994). This coincides with the trigeminal root showing strong immunoreactivity for GAP-43 (Hassiotis *et al.*, 2002), a phosphoprotein which is high in growth cones and developing nerves during axon elongation (Meiri *et al.*, 1986). Reflexes involving trigeminal inputs and jaw musculature are also present at birth and are presumably involved in suckling (Waite, personal observations). All the developing nuclei except Sp5O have higher levels of SDH and CO activity, like the adult. By P40, clear rostrocaudal subdivisions within the complex have emerged (Waite *et al.*, 1994), GAP-43 expression remains high in the trigeminal root and spinal trigeminal tract (Hassiotis *et al.*, 2002) and in Pr5, five lines of CO reactivity extend obliquely across the nucleus, reminiscent of the rows of mystacial vibrissae (Figure 10.3c; Waite *et al.*, 1994). By P147, small patches of CO activity are seen ventrolaterally in Pr5, presumably corresponding to individual vibrissae, but an entire representation is not apparent (Waite *et al.*, 1994). These patches are less clear than in rats, although in both species they are more obvious in juveniles than in adult animals (Belford and Killackey, 1979b). High CO reactivity has been suggested to correlate with synaptogenesis in developing animals (Wong-Riley, 1989) and, presumably, in the rat, synaptogenesis extends over a much shorter and concentrated period of time than in the tammar. This may explain the clearer appearance of patches in the developing rat (Waite *et al.*, 1994).

### Somatosensory thalamus

In the brush-tailed possum, birth of neurons in VPL/M is completed by P12, with substantial cell birth in the first postnatal week (Sanderson and Weller, 1990b). There is a gradient of neurogenesis from lateral (earlier) to medial (later), as in rodents (Angevine, 1970). Such information is not available for the tammar but, based on morphology, cell birth also appears to be primarily postnatal. At birth the diencephalon is composed of a neuroepithelial layer overlaid by a mantle zone and the boundaries of VPM cannot be clearly distinguished from VPL and surrounding nuclei on histological grounds until P47 (Leamey *et al.*, 1996), about the same time as in the brush-tailed possum (Sanderson and Weller, 1990b). The first trigeminothalamic axons from Pr5, tipped with

growth cones, enter the developing VPL/M complex at P15 (Leamey *et al.*, 1998), when it is seen as a dense cellular mass separated from the dorsal lateral geniculate by the external medullary lamina (Leamey *et al.*, 1996). Afferents continue to grow into and densely fill VPM up to P40 (Leamey *et al.*, 1998). The first segmentation within the nucleus is visible in Nissl- (Figure 10.3e) and CO-reacted material laterally in VPM at P52, and fingers of reactivity similar to the whisker-related patterns seen in the adult are clear by P73–75 (Figure 10.3g; Leamey *et al.*, 1996). This segregation occurs simultaneously with the segregation of the ingrowing afferents (Figure 10.3f and h) (Leamey *et al.*, 1998). A parallel electron-microscopic and electrophysiological study (Leamey *et al.*, 1998) showed that evoked synaptic responses can be recorded in VPM from the time of afferent ingrowth; prior to P50 excitatory responses are dominated by *N*-methyl-D-aspartate (NMDA)- mediated responses (Figure 10.3d). Such responses have been shown to be important for the formation of whisker-related patterns in mice (Li *et al.*, 1994). Coincident with the onset of synaptogenesis at P40, there is a decrease in duration of the response. Synaptic transmission prior to this is presumably via transmitter release from growth cones. At P50, the response is dominated by a fast non-NMDA-mediated potential, coincident with the onset of whisker-related patterns (Figure 10.3d). The presence of functional connections prior to the onset of whisker-related patterns suggests that synaptic interactions may play a role in this pattern formation.

Projections to the diencephalon from trigeminal sensory and dorsal column nuclei also develop postnatally in the North American opossum (Martin *et al.*, 1987). In contrast, ascending connections to VPM in the rat form prenatally and the formation of whisker-related patterns takes place over a few days (Belford and Killackey, 1979a), rather than a few weeks, as seen in the tammar (Leamey *et al.*, 1996).

### Somatosensory cortex

Neurogenesis in the somatosensory cortex of the brush-tailed possum is protracted and takes place in the first two postnatal months, with neurogenesis in the body region of the dorsolateral cortex lagging behind that of the head region in lateral cortex (Sanderson and Weller, 1990a). Neurogenesis finishes just before whisker-related barrels can be recognised in layer 4Cx at P65 (Sanderson and Weller, 1990a).

In the tammar, which has a similarly protracted developmental time-course to the possum, ascending input from the thalamus first reaches the cortex at P15 (Figure 10.4a, b) and afferents proceed to grow into and are widely distributed in the depth of the developing cortex without any sign of a waiting period (Figure 10.4c, d) (Marotte *et al.*, 1997). Pre- and postsynaptic cortical responses to thalamic stimulation in a thalamocortical slice preparation can be recorded from the time of afferent arrival, but are very slow and completely mediated by NMDA receptors. Thalamocortical synapses are first seen at P28 in the cortical plate when cortical responses remain slow and are still dominated by an NMDA component (Figure 10.4e) (Leamey *et al.*, 2007). The first corticothalamic projections are seen much later, at P60, from layer 5Cx cells (Marotte *et al.*, 1997). Whisker-related patterns typified by the first hint of CO patches (Figure 10.4g) in the just-formed layer 4Cx (Figure 10.4f) (Mark *et al.*, 2002), the clustering of afferents in layer 4Cx, and the clustering of the dendrites and somata of cells in layers 5Cx and 6Cx projecting to the thalamus (Figure 10.4h) (Marotte *et al.*, 1997) all become apparent at around the same time, at P72 to P76. This is slightly earlier than the patterns are first detected with SDH staining (Waite *et al.*, 1991) and coincides with the appearance of synapses (Leamey *et al.*, 2007) and synaptic responses in layer 4Cx in response to whisker stimulation *in vivo* (Figure 10.4i; Mark *et al.*, 2002) and the domination of thalamocortical responses by a fast non-NMDA-mediated component (Figure 10.4e) (Leamey *et al.*, 2007). This component may drive pattern formation and firing of cortical neurons (Leamey *et al.*, 2007). At older ages, the responses are longer lasting with more complex sequences of activity at different depths (Mark *et al.*, 2002).

Despite the very different time-course of development of the pathway between the rat and tammar (days compared to months), afferents in the rat are also in the cortex (Catalano *et al.*, 1991, 1996) for a relatively long time prior to the onset of evoked activity *in vivo* at P2 (Verley and Axelrad, 1975). Like in the tammar, the earliest evoked activity is temporally close to the appearance of layer 4Cx (Rice *et al.*, 1985), clustering of thalamocortical afferents (Catalano *et al.*, 1996) and whisker-related patterns demonstrated histochemically (Killackey and Belford, 1979).

## The whisker pathway, pattern development and plasticity

Experiments in the rodent whisker pathway have shown that the periphery is important for the development of whisker-related patterns centrally (Van der Loos and Woolsey, 1973; Weller and Johnson, 1975; Belford and Killackey, 1979b). Lesions to the whiskers or their nerve supply result in the loss of the corresponding whisker-related patch or patches and there is a critical period for this effect. Lesions made after the barrels in the cortex have formed have no effect, and the critical period for each level of the pathway correlates with the time of pattern formation at that level. Lesioning of the

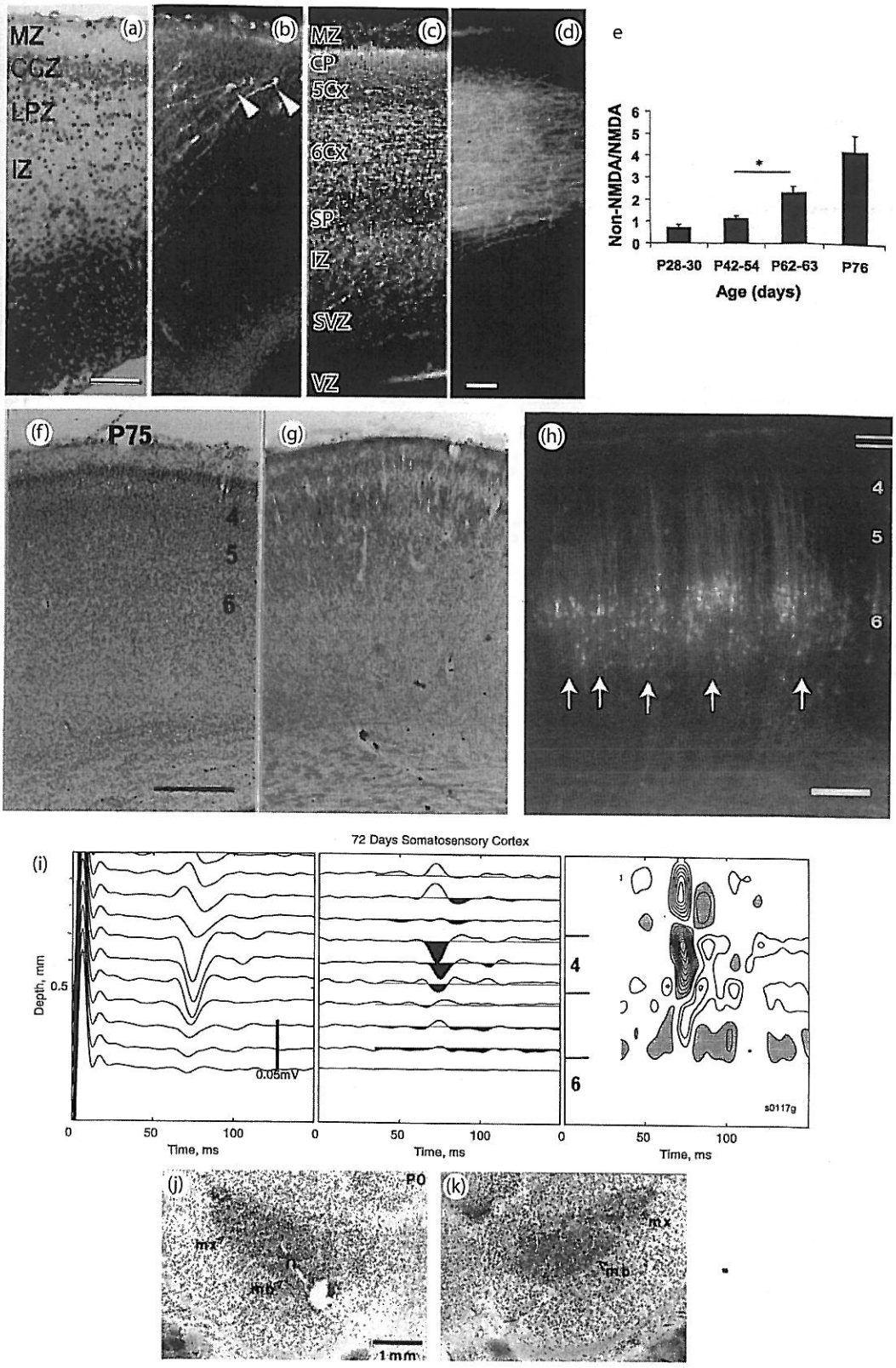


**Figure 10.4** (a) to (i) tammar wallaby (*Macropus eugenii*). (a) and b) Coronal sections through the somatosensory cortex at P15. Toluidine-blue-stained plastic section showing the developing cortical layers (a) and an adjacent section showing DiI-filled thalamocortical axons (b). Axons tipped with growth cones (arrowheads) reach to the base of the CCZ of the cortical plate. MZ – marginal zone; LPZ – loose-packed zone of the cortical plate; IZ – intermediate zone; SVZ – subventricular zone; VZ – ventricular zone; bar in (a) = 100 μm also applies to b). (c) and (d) Fluorescence views of a coronal section through the somatosensory cortex at P51 exposed for bisbenzimide counterstain to show the cortical layers (c) and DiI-filled thalamocortical axons (d). Layers 5Cx and 6Cx can now be recognised at the base of the cortical

whiskers in the brush-tailed possum also prevents the formation of thalamic (Figure 10.4j and k) and cortical patterns during a postnatal critical period (Weller, 1979; Waite and Weller, 1997). The closely timed sequence of pattern formation in the brainstem, thalamus and cortex in rodents led to the suggestion that a peripherally derived signal carried by afferent fibres swept along the pathway initiating pattern formation at each level (Killackey, 1985). The periphery clearly provides an important signal for pattern formation, but the target tissue also appears to play a role. This is supported by the time-course of development of the patterns in the tammar, where the protracted development in comparison to rodents clearly separates, in time, the formation of patterns at the different levels. Thus, the onset of pattern formation is separated by weeks rather than a few days as seen in rodents (brainstem – P40, thalamus – P52, cortex – P76). There is also a long delay between the arrival of afferents (brainstem – prenatal, thalamus – P15, cortex – P15) and the onset of functional activity (Waite *et al.*, 1994; Leamey *et al.*, 1996, 1998; Marotte *et al.*, 1997) and pattern formation at each level. These findings suggest that the pattern formation is independently controlled at each level rather than by a single peripheral signal and that maturation of target cells may play a role (Waite *et al.*, 1998). This is supported by findings that an increase in the non-NMDA-mediated glutamatergic response, a measure of target maturation, correlates well with pattern formation in both thalamus and cortex (Leamey *et al.*, 1998, 2007). For example, in the cortex, despite afferents arriving from P15, timing is controlled by the maturation of layer 4Cx cells, which are not in place until around P72 to P76.

The slow, primarily postnatal development of the whisker pathway in the tammar has been used to investigate the effects of lesions of the infraorbital nerve supplying the whiskers at various developmental times (Waite *et al.*, 2006). Lesions prior to innervation of the thalamus and cortex result in an absence of patterns at both levels, suggesting that any reinnervation of the follicles is inadequate to rescue normal pattern formation. Interestingly, lesions at times when thalamic or cortical patterns are forming or have formed indicate a single critical period for thalamus and cortex. This is despite the fact that the thalamic pattern first starts to appear around three weeks before that in the cortex and suggests a close relationship between the stabilisation of patterns in thalamus and cortex.

## What remains unknown?

This chapter indicates that much about the organisation and function of somatosensory pathways in marsupials is similar to that in eutherians. However, where differences

**Caption for Figure 10.4** (Cont.)

plate (CP); abbreviations as for (a). SP – subplate; bar in (d) = 100μm, also applies to c). (e) Contributions of *N*-methyl-D-aspartate (NMDA) and non-NMDA components of the glutamatergic postsynaptic response during development. Bar graph shows the increase in the non-NMDA to NMDA ratio during development. The ratio remains stable from P28 to P54, but increases significantly (*$P$ <0.05) by P63. There is a further increase by P76, although this is not statistically significant. (a) to (e) from Leamey *et al.* (2007) with permission, European Journal of Neuroscience, Federation of European Neuroscience Societies and Blackwell Publishing. (f) and (g) Coronal sections through the somatosensory cortex of an animal aged P75 stained with cresyl violet (f) and cytochrome oxidase (CO) (g). (f) Layers 4Cx to 6Cx are now recognisable cytoarchitectonically beneath the compact cell zone (ccz); bar = 500 μm. (g) CO reactivity is increased in layer 4Cx and the first hints of patches are seen; bar as for (f). From Mark *et al.* (2002), Figure 1c and d. (h) Low-power fluorescent view of DiI labelling in the somatosensory cortex at P76 after a deposit of DiI in the VPM. The pair of white bars mark the boundaries of the CCZ. At this age, the first sign of clustering of retrogradely labelled cells in layers 6Cx and 5Cx and their processes is seen (arrows); bar = 200 μm. From Marotte *et al.* (1997) Figure 7d, with permission, copyright 1997 Wiley-Liss, Inc. (i) Current source density (CSD) analysis of evoked potentials recorded in the cortex of an animal aged P72. The three plots are from a single data set. Each is on the space/time domain of the recording depth in the cortex and the time after a shock to the infraorbital branch of the trigeminal nerve. The plot on the left shows averaged evoked potentials. Each trace is the average of 10 sweeps. Positive is upwards. The centre plot shows the corresponding CSD waveforms. Where currents are negative, the waveforms are filled in black. The plot on the right shows the same CSD data plotted as contours on the depth latency domain. Current sinks, indicative of excitatory synaptic activity, are shaded, giving the depth and latency. Corresponding current sources are plotted as unshaded contours. Contour levels are at 10% steps relative to the maximum sink magnitude. Boundaries of cortical laminae were determined from histology. The short latency sink indicates synaptic activity is located in layer 4Cx. From Mark *et al.* (2002), Figure 3 upper panel. (j) and (k) Results of removing all large follicles from one side of the snout of a newborn brush-tailed possum on the whisker-related pattern in the thalamus. (j) A section through the somatosensory thalamus (control side). The cell-dense part (h) represents the possum's head. Some of the cells are arranged in clusters and rows (mx). They receive sensory information from the large mystacial whisker follicles. The others (mb) receive information from the chin; Nissl stain (thionin), 100 μm thick. (k) A section through the experimental side of the somatosensory thalamus. The rows and groups of cells are missing (mx) and the region receiving information from the chin is enlarged. Nissl stain (thionin), 100 μm thick. From Waite and Weller (1997) with permission from the University of New South Wales Press.

exist, research in marsupials is frequently disadvantaged by the lack of a systematic approach in many areas. Results are often described for just one species or neural area, so that it is impossible to assess the extent to which this reflects a general difference between marsupials and eutheria or a species peculiarity. For example, the interesting observation that Pacinian corpuscles in the wallaby hindlimb are less rapidly adapting than in eutheria could reflect a significant difference in response properties related to the different locomotor style. However, as this is the only observation of hindlimb Pacinian responses in marsupials, its significance is hard to evaluate. Similarly, the extent to which whisker-related specialisations, described in the possum and tammar, are present in other species would benefit from a more systematic approach. They are not seen in Didelphidae or Dasyuridae, but their presence in Diprotodontia has only been investigated for some gliders (present), brush-tailed possum (present) and tammar (present). They may well be more widespread, especially in the smaller species such as potoroos, and the smaller possums and macropods.

There are large gaps in our understanding of function in marsupial somatosensory systems. While receptor morphology has been relatively well described, especially for facial skin, there are few studies on response properties of mechanoreceptors and none for joint or thermal receptors or nociceptors. Descriptions of responses in the dorsal column nuclei are limited to the opossum, while we know nothing about dorsal-horn physiology. The roles of cutaneous and proprioceptive inputs in co-ordinating hindlimb reflexes have not been studied, despite the synchronous activation of right and left hindlimbs needed for jumping. Similarly for VPL/M thalamus, studies have focussed on projections and somatotopy; responses of single cells have not been described. The somatosensory cortical areas have fared rather better, both for the breadth of species investigated and the possible functions of different areas. However, the level of understanding of marsupial cortical physiology lags well behind that for eutherians.

Finally, despite the obvious ease of access to pouch young, few researchers have taken advantage of this for functional studies on development or plasticity after injury. The whisker pathway remains the only example of studies on onset of activity and the effects of early lesions. It would be of interest to know how the extended period of time in the pouch affects the development of muscle spindles, joint receptors and proprioceptive pathways. Furthermore, access to pouch young would allow studies on nerve lesions or amputations, before central connections and pathways have developed, not easily undertaken at corresponding times in utero. Similarly the potential to introduce changes within the pouch environment have not been exploited. For instance, modifications in the pouch temperature for short periods might provide interesting insights into the neural mechanisms which establish core temperature and thermoregulation. As discussed more fully in Chapter 14, access to pouch young provides unique opportunities for early functional and plasticity studies.

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