# Forebrain gene expression domains and the evolving prosomeric model

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**The prosomeric model attributes morphological meaning to gene expression patterns and other data in the forebrain. It divides this territory into the same transverse segments (prosomeres) and longitudinal zones in all vertebrates. The axis and longitudinal zones of this model are widely accepted but controversy subsists about the number of prosomeres and their nature as segments. We describe difficulties encountered in establishing continuity between prosomeric limits postulated in the hypothalamus and intra-telencephalic limits. Such difficulties throw doubt on the intersegmental nature of these limits. We sketch a simplified model, in which the secondary prosencephalon (telencephalon plus hypothalamus) is a complex protosegment not subdivided into prosomeres, which exhibits patterning singularities. By contrast, we continue to postulate that prosomeres p1–p3 (i.e. the pretectum, thalamus and prethalamus) are the caudal forebrain.**

Topographic comparison of diverse gene expression patterns in the forebrain requires a comprehensive interpretive paradigm. Scientists working on gene expression patterns rely on a particular brain model, although many select this pragmatically, by authority or apparent convenience, and few have a rationale for their choice. It is, therefore, important to explain and discuss the properties of given models. For the past ten years, we have been developing the prosomeric model for the vertebrate forebrain [1–6]. At its core, this model recognizes the bent longitudinal axis of the forebrain relative to midbrain and hindbrain, and defines the primary anteroposterior (AP) and dorsoventral (DV) divisions – transverse neuromeres and longitudinal zones, respectively (Box 1). The model contemplates explicitly the existence of additional subdivisions of the main AP and DV zones (Figs 1,2) and recognizes the optic and telencephalic vesicles as specialized neural fields with patterning properties that are in part independent from the rest of the neural tube. It should be noted that the prosomeric model is primarily a morphological instrument (paradigm) that emphasizes evolutionarily conserved topological and molecular expression relationships in the neural tube. It is neither a hypothesis nor a theory of the development of brain parts. Such a model is useful for furthering progress as it provides a conceptual framework within which preconceived assumptions and deduced predictions can be tested.

The prosomeric model has been useful for classifying topologically numerous novel molecular expression patterns and associated histogenetic data. Molecularly specified domains of the neural wall can be delimited across species, and postulated gene functions can be tested subsequently in terms of regional and cellular fates. The topological definition of DV and AP subdivisions within prosomeres, each one capable in principle of producing particular types of neurons or glia, facilitates interpretation of observations in embryonic and adult brains in terms of the sequence of DV and AP patterning mechanisms that organize the neural tube (Figs 1,2). This paradigm has indeed helped causal thinking in terms of AP and DV patterning processes in the complex forebrain of vertebrates.

The model was supported importantly by its capacity to provide morphological meaning to numerous gene expression patterns; many expression domains were found to respect a subset of the boundaries postulated in the model [3–8]. The prosomeric model was conceived to be useful for all vertebrates, as was duly corroborated in several comparative morphological studies in organisms as diverse as lampreys and humans [9–11]. Knowledge about gene expression patterns in non-mammalian vertebrates has expanded considerably in recent years. Genes homologous to those with established molecular boundaries in the mouse forebrain have been mapped in the chick [12], *Xenopus* [13] and zebrafish or medaka [14–19]. Comparable expression of some of these genes was also located in the forebrain of agnathans [20,21]. In this field, the anatomically detailed prosomeric model seems helpful for interspecies comparisons. This approach is facilitating an emerging, novel perspective on the evolutionary origin of the vertebrate neural 'Bauplan' [22–24].

The prosomeric model seems now widely known and some of its novel premises, including the redefinition of the axial dimension of the forebrain and the proposed longitudinal zones, with their prechordal and epichordal parts, have been widely accepted. By contrast, the number, limits and nature as neural compartments of the prosomeres remain partially controversial [25,26] (Box 1). Here, we propose a simplification of the model, eliminating some of the controversial aspects without losing the

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 0166-2236/$ - see front matter  doi:10.1016/S0166-2236(03)00234-0

**Box 1. Definition of conceptual terms used**

**Segmentation**
For animal forms having bilateral symmetry and a length axis, segmentation involves patterning of the form into distinct and complete transverse parts (segments) aligned serially along the longitudinal (anteroposterior, or AP) axis. 'Distinct' implies that there are detectable intersegmental boundaries but does not preconceive their nature or how we visualize them; during development, boundaries can change their status from non-overt to overt, and then change again to a hidden status, thus requiring experimental demonstration at given stages. 'Complete' implies that only a complete cross-sectional (transverse) part of the form can be a segment.

**Metamery**
A set of segments of a form have the topological property of metamery if their fundamental structure is comparable; that is, if they are serially homologous in terms of basic components [57]. Bilateral symmetry and length axis, as well as any distinct segments, come to our attention before we can discover whether the latter are metameres, or whether their limits are comparable. In principle, an animal form can have non-metameric segments, or metameric segments separated by non-identical boundaries. Continuous metamery throughout the body axis is not obligatory. However, the definition of 'fundamental structure' can be re-examined until a common denominator is found for all observed segments.

**Neural segment**
We apply the general principle of segmentation (above) to the vertebrate neural tube. Neural segments are distinct and complete transverse parts of the neural tube, itself clearly a bilaterally symmetric and AP-elongated form. Transverse neural subdivisions are already apparent as molecularly distinct parts of the neural plate (protosegments), and these areas can have a characteristic fate [58]. After closure of the neural tube, complete transverse subdivisions known as neuromeres transiently appear as serial bulges separated by transverse constrictions of the neural wall [59]. This owes to differential proliferative rates, in addition to various secondarily acquired properties of the neuroepithelial cells at the boundaries (i.e. clonal restriction, gap-junctional isolation and differential expression of cell adhesion and matrix molecules). Bulging varies across species and therefore is not strictly required for neural segment definition. By contrast, differential molecular specification along the AP dimension is evolutionarily conserved and required for definition purposes, because it leads to the segment-boundary and segment-identity properties [60]. Modern methods thus allow earlier and more fundamental delimitation of brain segments than older, purely morphological methods. Neural segments later suffer morphogenetic deformation, and the intersegmental boundaries are effaced by the progressive radial expansion of the neural wall. Experimental fate mapping nevertheless supports the persistence of the primitive boundaries, now hidden, which delimit segment-derived domains of the brain wall even in the adult brain – irrespective of assorted 'violations' of these limits by tangentially migrating neurons [61]. The notion of 'transient' neural segments erroneously holds they exist only while they are visible as bulges separated by constrictions.

**Neuromere**
All distinct neural segments postulated by us share a fundamental dorsoventral (DV) structural pattern composed of four longitudinal zones (roof, alar plate, basal plate and floor). We therefore see them as generally metameric; hence, our term 'neuromere' refers to the common pattern of fundamental DV zones, whereas our term 'neural segment' is less restrictive and does not require this or any shared pattern, just distinct and complete transverse boundaries. Particular regions of the neural tube can be conceived as 'tagmata', in which local sets of segments share regional characters. This leads to the classic distinction of forebrain, midbrain, hindbrain and spinal neuromeres, called prosomeres, mesomeres, rhombomeres and myelomeres, respectively [59]. Depending on the 'fundamental structure' selected for comparisons, prosomeres and rhombomeres might or might not be mutually metameric. Note that there exist alternative concepts of neuromeres, in which metamery is postulated as a necessary condition for any neural segment. This combines with a definition of segments according to their boundary properties, rather than their internal structure, often disregarding dorsoventral completeness [26,62]. This viewpoint accepts only a limited number of 'transient brain segments' and leaves other transverse brain parts as morphological non-entities waiting for a concept. This is of limited value for most applied (e.g. causal, comparative or functional) purposes.

**Compartment**
A compartment is a self-contained developmental unit in terms of cell populations. The primary precursors at the inception of the compartment generate a spatially restricted polyclone of derivatives. Boundaries are, thus, clonal restriction limits, which are supposed to be absent inside the compartment, where cells should freely intermix. In the 1990 s it was conjectured that brain segments or neuromeres might actually be compartments, at least with regard to neuroepithelial cells [63]. Some authors have chosen to think that, if clonal restriction is not detected, 'other segments' do not exist. However, it remains conceivable that further research might eliminate false negative results, or that clonal restriction might after all not be a necessary criterion for intersegmental boundaries. It has not yet been proven that the clonal restriction boundaries separating adjacent rhombomeres in the hindbrain satisfy the property of completeness (they are absent across the floor plate); restriction might be absent in some distinct complete transverse limits and is not uniquely present throughout development. Indeed, a multiplicity of transverse clonal boundaries is established progressively over time, even inside the segments [63].

**Segmental or neuromeric subdivisions**
Segmental or neuromeric subdivisions are non-complete parts distinguished within segments or neuromeres; they can be variously disposed relative to the axial dimension of the segment.

**Longitudinal zones**
Longitudinal zones result from comparable dorsoventral patterning across the neural primordium [64].

**Zonal divisions**
Zonal divisions are subdivisions that form locally along the DV dimension of a primary longitudinal zone. Advancing DV patterning generates such subdivisions owing to step-like recruiting of differential gene expression patterns in localized groups of neural precursors at various distances from locally efficient signal sources, leading to the generation of specific neuronal or glial subtypes in a spatially graded fashion [65]. These derivatives usually adopt a stereotyped position in the corresponding mantle layer, unless they migrate elsewhere.

essential elements that have already been found to be of wide utility.

## Reasons for changing the model

The initial model we proposed [2–4] was tentative in several aspects; we subsequently modified some details, particularly of the ventral thalamus and mammillary areas, as our knowledge of developing structures increased [5,6,12,27]. The secondary prosencephalon and the adjoining part of the diencephalon represented the most difficult areas, owing to their deformation during telencephalic evagination and morphogenesis. Moreover, patterning effects apparently occur here that are not found elsewhere. Our efforts to analyze how the neuromeric boundaries initially postulated

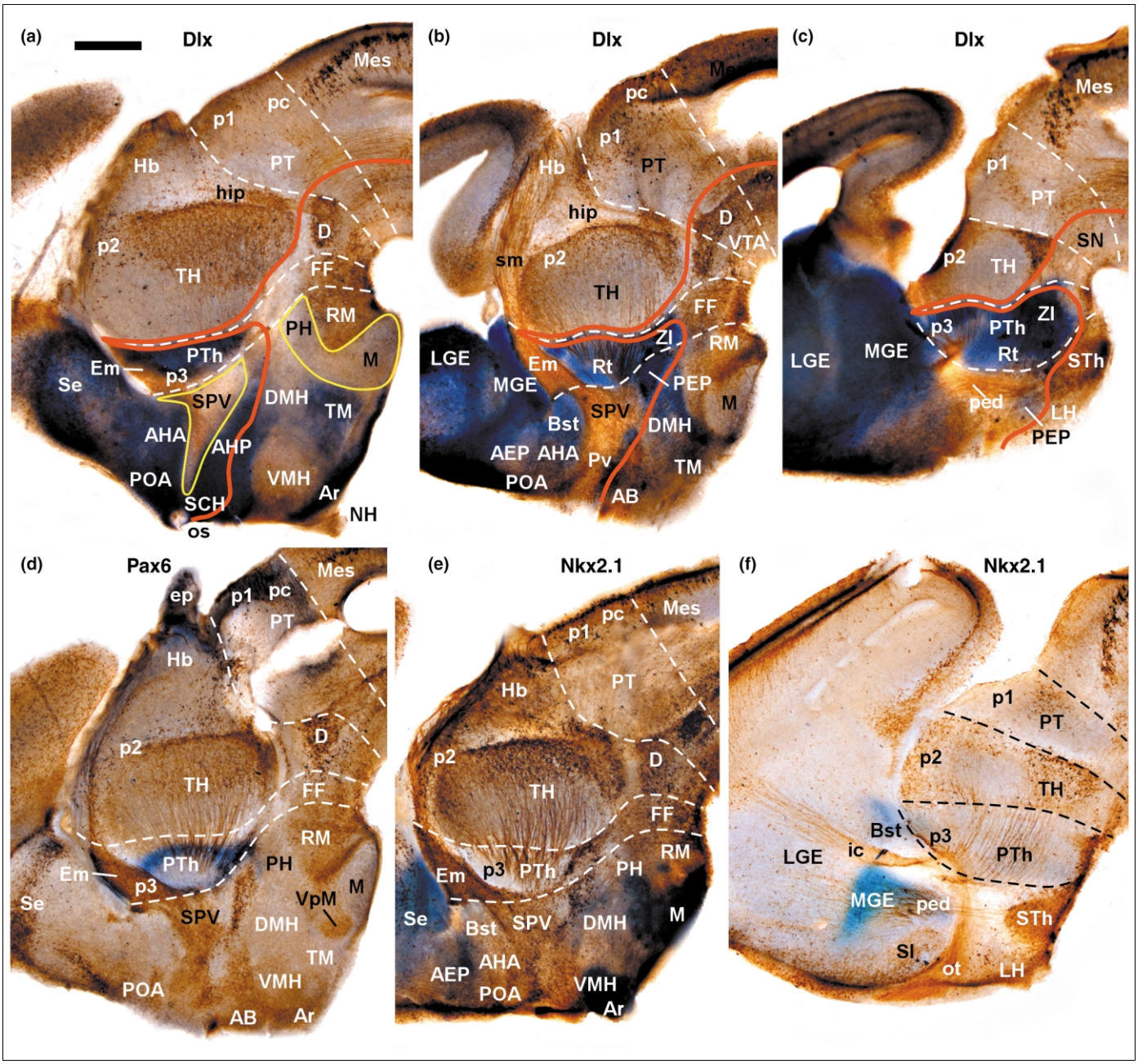


**Fig. 1**. The major extra-telencephalic forebrain subdivisions are illustrated by six sagittal vibratome sections (each 100 μm thick) of embryonic-day 15.5 mouse embryonic brain, in which immunohistochemistry for calretinin (brown) was combined with *in situ* hybridization (blue) using mRNA probes for *Dlx5* (a–c), *Pax6* (d) or *Nkx2.1* (e,f). Sections (a–c) proceed from medial to lateral, and sections (d–f) were chosen to match approximately the overlying panels. Postulated prosomeric boundaries are drawn in as white broken lines. The postulated alar–basal boundary is the red line in a–c, based on the *Nkx2.2* signal (note dorsal deflection at the interthalamic zona limitans). Various neural subdivisions and tracts are identified (see list of abbreviations). The *Otp* expression domains tracing the p3–hypothalamic boundary are outlined in yellow in (a). The section in (f) is slightly oblique; it shows favourably the calretinin-stained thalamocortical fibers as they cross the prethalamus and bend just across the hypothalamic boundary into the internal capsule (note also the position of the bed nucleus of stria terminalis, expressing *Nkx2.1*, dorsal to the internal capsule). The relative topography of the peduncular tract as it crosses the basal ganglia and approaches the subthalamic nucleus is also well represented in (c) and (f). Scale bar in (a), 4 mm in all panels. Abbreviations: AB, anterobasal nucleus; AEP, anterior entopeduncular area; AHA, anterior hypothalamus, anterior area; AHP, anterior hypothalamus, posterior area; Ar, arcuate nucleus; Bst, bed nucleus of stria terminalis; D, nucleus of Darkschewitsch; DMH, dorsomedial hypothalamic nucleus; Em, eminentia thalami; ep, epiphysis; FF, Forel fields; Hb, habenula (epithalamus); hip, habenulo-interpeduncular tract; ic, internal capsule; LGE, lateral ganglionic eminence; LH, lateral hypothalamus; M, mammillary complex; Mes, mesencephalon; MGE, medial ganglionic eminence; os, optic stalk; ot, optic tract; p1–p3, prosomeres 1–3; pc, posterior commissure; ped, telencephalic peduncle; PEP, posterior entopeduncular area; PH, posterior hypothalamus; POA, preoptic area; PT, pretectum; PTh, prethalamus (previously known as ventral thalamus); Pv, anterior periventricular nucleus; RM, retromammillary area; Rt, reticular nucleus; Se, septum; SI, substantia innominata; sm, stria medullaris; SN, substantia nigra; SPV; supraopto-paraventricular area; STh, subthalamic nucleus; TH, thalamus (previously known as dorsal thalamus); TM, tuberomammillary area; VMH, ventromedial hypothalamic nucleus; VpM, ventral premammilary nucleus; VTA, ventral tegmental area; ZI, zona incerta.

within the hypothalamus continued into the telencephalic roof, to establish the required segmental property of completeness (Box 1), met with various difficulties.

A fate map of the late neural plate in the chick [28] showed the subpallium and septum to lie topologically rostral to the telencephalic pallium, agreeing with fate-mapping data at the closed neural tube stage [29]. In principle, the pallium–subpallium boundary seemed a good candidate for continuing one of the interrupted transverse hypothalamic limits [27,30]. However, we were

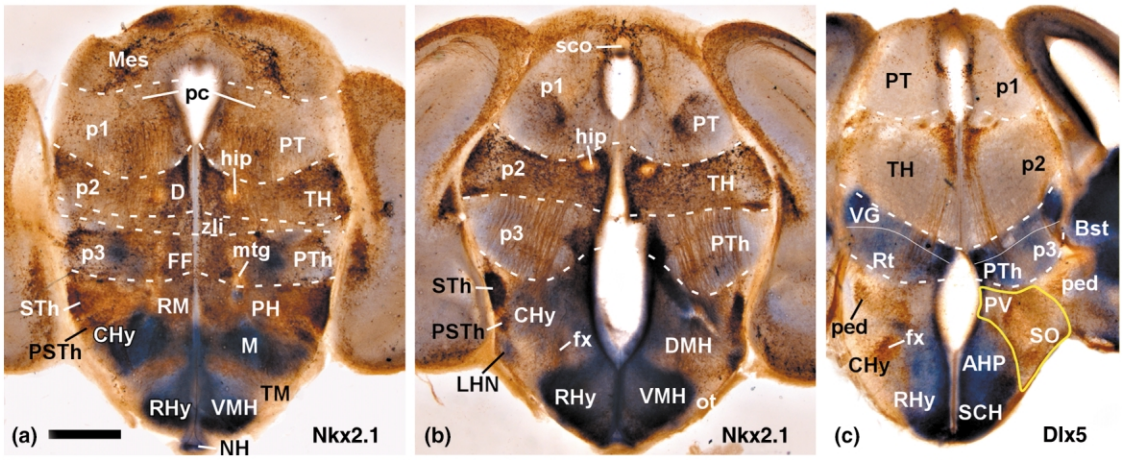


**Fig. 2**. Transverse forebrain boundaries in three slightly oblique horizontal vibratome sections (100 μm thick) of embryonic-day 15.5 mouse embryonic brain, in which immunohistochemistry for calretinin (brown) was combined with *in situ* hybridization (blue) using mRNA probes for *Nkx2.1* (a,b) or *Dlx5* (c). These sections proceed ventro-dorsally [compare section plane of (a) – through neurohypophysis and rostral mesencephalon – with Fig. 1a; (b) intersects the strongly calretinin-positive part of the thalamus; (c) passes through the supraopto-paraventricular area (yellow outline) and more dorsal parts of the thalamus and prethalamus; note the position of the bed nucleus of stria terminalis in (c)]. Various neural subdivisions and tracts are identified (see list of abbreviations). These horizontal sections also illustrate the caudal and rostral parts of the hypothalamus. Scale bar in (a), 4 mm in all panels. Abbreviations: AHP, anterior hypothalamus, posterior area; Bst, bed nucleus of stria terminalis; CHy, caudal hypothalamus; D, nucleus of Darkschewitsch; DMH, dorsomedial hypothalamic nucleus; FF, Forel fields; fx, fornix tract; hip, habenulo-interpeduncular tract; LHN, lateral hypothalamic nucleus; M, mammillary complex; Mes, mesencephalon; mtg, mammillotegmental tract; NH, neurohypophysis; ot, optic tract; p1–p3, prosomeres 1–3; ped, telencephalic peduncle; PH, posterior hypothalamus; PSTh, parasubthalamic nucleus; PT, pretectum; PTh, prethalamus (previously known as ventral thalamus); PV, paraventricular nucleus; RHy, rostral hypothalamus; RM, retromammillary area; Rt, reticular nucleus; SCH, suprachiasmatic area; sco, subcommissural organ; SO, supraoptic nucleus; STh, subthalamic nucleus; TH, thalamus (previously known as dorsal thalamus); TM, tuberomammillary area; VG, ventral geniculate nucleus; VMH, ventromedial hypothalamic nucleus; zli, zona limitans intrathalamica.

not satisfied that this boundary, or any other telencephalic limit [8], is continuous with the hypothalamic boundaries. The optoeminential domain (Figs 1a–1d, 3), a roughly longitudinal area under the hemispheric stalk that was defined initially as negative for *Dlx* gene expression [2–4], seems to interrupt potential continuities. Moreover, genes expressed in the optoeminential domain, such as *Sim-1* or *Otp*, show a long intra-telencephalic spike continuing along the terminal sulcus into the pallial amygdala [31,32] (Fig. 3); this feature blocks the rostral boundary of the prethalamus (ventral thalamus) from making contact with the pallium–subpallium limit. In addition, special patterning mechanisms that subdivide the telencephalon seem to generate criss-crossing roof signals originating either rostrally at the anterior neural ridge (the end of the roof plate [24]) or caudally at the cortical hem [33–36]. The resulting curved intra-telencephalic boundaries (Fig. 3; see also Fig. 11 in Ref. [12]) appear to be unrelated to the overall AP and DV dimensions of the neural tube (Fig. 3). Furthermore, induction of 'ventral' molecular properties in the telencephalon (i.e. *Shh* and *Nkx-2.1* expression in the basal ganglia; Fig. 3) takes place in rostral areas via inductive processes that lack continuity with the axial mesendoderm [37] and differ in part mechanistically from ventral patterning of the hypothalamus [38]. Thus, the telencephalon is a derivative of the alar plate that, during its evagination, secondarily becomes patterned into several pallial and subpallial subdivisions with boundaries that might not be directly related to any boundaries in the underlying rostral diencephalon. Figure 3 displays these topological patterning singularities, represented semi-schematically as observed in sections (Figs 1,2).

Because the definition of intersegmental boundaries between prosomeres requires that these be transverse and complete from roof to floor of the neural tube, our analysis inclined us more and more to the conclusion that observable hypothalamic boundaries could be secondary ones that develop inside the hypothalamus owing to various local influences and, thus, that do not reveal formal segmental elements of the forebrain.

## The modified model

Our present proposal, which also includes convenient terminological simplifications, reduces the number of postulated prosomeres to the three caudal diencephalic ones, p1–p3. These contain in their alar regions the pretectum (p1), the thalamus (previously known as the dorsal thalamus) plus the habenula or epithalamus (p2), and the prethalamus (previously known as the ventral thalamus) plus the eminentia thalami (p3) (Figs 1–3). The present proposal represents just two major changes upon our initial model [2–4]. First, the eminentia thalami – previously considered a dorsal part of p4 – was recognized as a domain dorsal to the prethalamus, occupying there a position analogous to that of the habenula relative to the thalamus, traversed longitudinally by the stria medullaris (sm in Fig. 3); the dorsal position of the eminentia thalami relative to the prethalamus is supported by diverse gene markers expressed in it, such as *Tbr-1* [12,27], *PLZF* [39], *R-cadherin* [40,41], *Ebf3* [42] and *Lhx9* [43]. The entire alar p3, including the eminentia thalami, expresses the gene *Arx* [44], whereas the eminentia is excluded from the p3 domain expressing *Dlx* genes (Figs 1a,b, 3). There are mouse and chick data suggesting that some neurons might migrate tangentially out of the eminentia thalami into the adjoining amygdala [12,32,45]. Second, present changes affect the old p4–p6 prosomeres, which are reinterpreted as secondary subdivisions

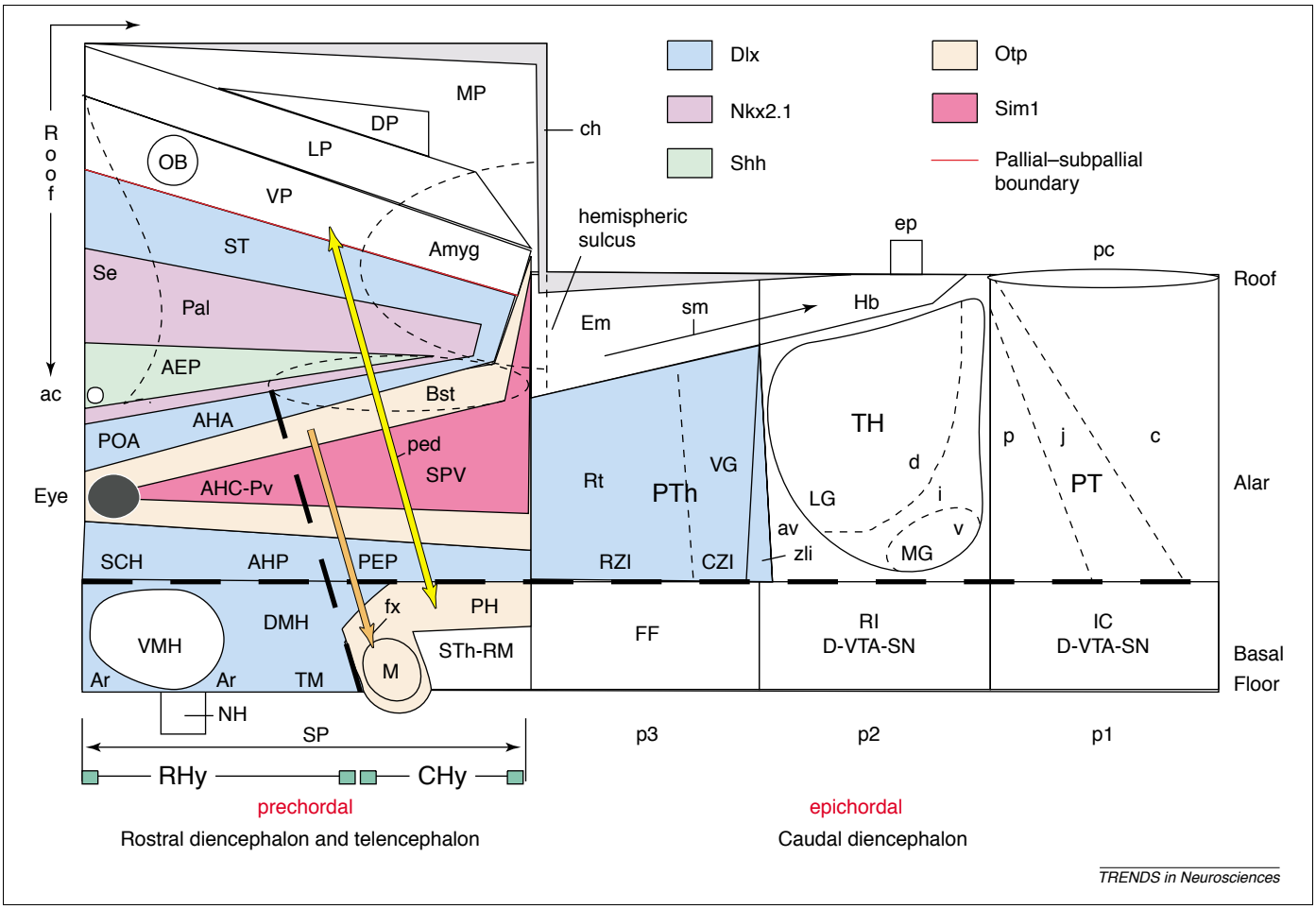


**Fig. 3.** Our modified prosomeric model, some relevant gene expression patterns, and secondary subdivisions emerging from recent research. The primary anteroposterior (AP) divisions are identified below. Primary dorsoventral (DV) zones are marked at the caudal end (the alar–basal boundary appears as large dashes superposed on a thin line; note the roof longitudinal zone ends beyond the choroid plexus and commissural septal areas at the anterior commissure [28]). Telencephalic subdivisions agree with gene patterns [12] and a recent fate map [28]; the pallial–subpallial boundary is drawn as a red line. The septal, amygdaloid and stria terminalis regions are histogenetic complexes (small dashes), thus explaining their adult structural diversity. The optoeminential domain under the telencephalic stalk shows tentative internal subdivisions and has at its caudal end spike-like dorsal expansions into the amygdala [31,32,45]. The hypothalamus appears secondarily divided into rostral and caudal parts with distinct derivatives (the mutual limit is marked as medium-large dashes; note the parallel-coursing fornix and peduncular tracts). We repositioned the retromammillary area, where the subthalamic nucleus arises, and redefined the posterior hypothalamus. Two AP subdivisions of the prethalamus are indicated (i.e. with higher *Dlx5* signal caudally and higher *Pax6* signal rostrally [12], forming two distinct parts of the zona incerta [53,54]; see Fig. 1d and thin white lines dividing the prethalamus in Fig. 2c); remarkably, the reticular nucleus forms in the rostral prethalamus, dorsal to the rostral zona incerta, whereas the ventral geniculate nucleus lies in caudal prethalamus, dorsal to the caudal zona incerta. The p3 tegmentum corresponds to the classic Forel fields. Subdivisions shown in the thalamus are limited to its main three histogenetic tiers (dorsal, intermediate and ventral) and the lateral and medial geniculate primordia [10,11,55]. Core and shell domains of the zona limitans intrathalamica are not indicated. The pretectum shows three AP subdivisions, described as precommissural, juxtacommissural and commissural domains [10,55]). Tegmental structures of p1 and p2 include iterated parts of the substantia nigra and the ventral tegmental area, a dopaminergic complex that clearly extends beyond the diencephalon–mesencephalon boundary [56]. Abbreviations: ac, anterior commissure; AEP, anterior entopeduncular area; AHA, anterior hypothalamus, anterior area; AHC, anterior hypothalamus, central part; AHP, anterior hypothalamus, posterior area; Amyg, amygdala; Ar, arcuate nucleus; av, anteroventral area of thalamus; Bst, bed nucleus of stria terminalis; c, commissural pretectum; ch, choroidal tela; CHy, caudal hypothalamus; CZI, caudal zona incerta; D, nucleus of Darkschewitsch; d, dorsal tier of thalamus; DMH, dorsomedial hypothalamic nucleus; DP, dorsal pallium; Em, eminentia thalami; FF, Forel fields; fx, fornix tract; Hb, habenula (epithalamus); i, intermediate tier of thalamus; IC, interstitial nucleus of Cajal; j, juxtacommissural pretectum; LG, lateral geniculate nucleus; LP, lateral pallium; M, mammillary complex; MG, medial geniculate nucleus; MP, medial pallium; NH, neurohypophysis; OB, olfactory bulb; p, precommissural pretectum; p1–p3, prosomeres 1–3; Pal, pallidum; pc, posterior commissure; ped, telencephalic peduncle; PEP, posterior entopeduncular area; PH, posterior hypothalamus; POA, preoptic area; PT, pretectum; PTh, prethalamus (previously known as ventral thalamus); Pv, anterior periventricular nucleus; RHy, rostral hypothalamus; RI, rostral interstitial nucleus; RM, retromammillary area; Rt, reticular nucleus; RZI, rostral zona incerta; SCH, suprachiasmatic area; Se, septum; sm, stria medullaris; SN, substantia nigra; SP, secondary prosencephalon; SPV; supraopto-paraventricular area; ST, striatum; STh, subthalamic nucleus; TH, thalamus (previously known as dorsal thalamus); TM, tuberomammillary area; v, ventral tier of thalamus; VG, ventral geniculate nucleus; VMH, ventromedial hypothalamic nucleus; VP, ventral pallium; VTA, ventral tegmental area; zli, zona limitans intrathalamica.

restricted to the hypothalamus within the secondary prosencephalon. The secondary prosencephalon is the entire prechordal (rostral-most) portion of the neural tube. Seeking terminological continuity, we divide the secondary prosencephalon into telencephalon and rostral diencephalon or hypothalamus [6] (Fig. 3). We define rostral and caudal parts of the hypothalamus (RHy and CHy, respectively, in Figs 2,3). The longitudinal or DV zones of this large domain remain unchanged, so that alar and basal parts of the classic hypothalamus can be distinguished.

The rostral hypothalamus correlates topographically with early expression of *Six3*, which extends from the septum to neurohypophysis [46]; this was the best evidence so far for defining molecularly the old p6 prosomere; however, analysis of the *Six3* or *Hesx1* knockout mice [47,48] revealed a loss of the whole secondary prosencephalon which, thus, seems to behave as a single

histogenetic field. The alar portion of the rostral hypothalamus contains the preoptic region plus the rostral optoeminential (anterior hypothalamic) and suprachiasmatic DV subdivisions (Fig. 3); the corresponding basal portion, characteristically *Shh*- and *Nkx2.1*-positive at early stages, contains the ventromedial and dorsomedial hypothalamic nuclei, which later partially downregulate *Nkx2.1* expression (Figs 1e, 2b); the rostral hypothalamic floor plate includes the median eminence, the neurohypophysis and the arcuate nucleus.

The caudal hypothalamus (the subthalamus or peduncular region in some literature) is aligned with the superficially coursing telencephalic peduncle (yellow arrow in Fig. 3; ped in Figs 1c,f, 2c, 3). Underneath it, the fornix tract (fx and orange arrow in Fig. 3) also traverses dorsoventrally the caudal hypothalamus, approaching the mammillary body. The alar part of the caudal hypothalamus contains the supraopto-paraventricular (caudal optoeminential) area and the posterior entopeduncular area (origin of the migrated entopeduncular nucleus; PEP in Fig. 1b,c). The basal plate is represented by the posterior hypothalamic area, in addition to the mammillary and retromammillary regions [often, part of the posterior hypothalamus is prolongated into the p3 tegmentum, but we characterize here the latter as 'Forel fields' (FF in Fig. 3)]. The subthalamic nucleus arises from the retromammillary region and migrates to the deep surface of the peduncle (Figs 1b,c,f, 2a,b, 3).

The boundary between caudal hypothalamus and p3 is well defined by the expression of several genes. In the alar plate, it coincides with the sharp caudal limit of the *Sim-1*, *Otp* and *Brn-2* genes, which are expressed in the optoeminential area, and with the sharp rostral limit of *Arx*, *Dlx* and *Pax6* transcripts in the prethalamus (Figs 1a–d, 2c, 3). In the mouse, the dorsal end of this limit surrounds the eminentia thalami along the stria terminalis into the vicinity of the amygdala, apparently to end at the fissura choroidea in the caudomedial wall of the telencephalon [31]. In the basal plate, the posterior hypothalamus of our model shows a distinct boundary with the Forel fields [i.e. in expression of *Otp* [49] (outlined in yellow in Fig. 1a; Fig. 3)]. There are specialized radial glia at this boundary [50]. Observations in frogs [7,51] reveal a marked cell-poor gap at this boundary, analogous to the aspect of the zona limitans intrathalamica (p2–p3 limit). Although studies aiming to detect clonal restriction properties of chicken interprosomeric boundaries failed to identify this boundary [28,29], this forebrain area might not have been sampled sufficiently. We therefore expect that additional studies might clarify whether or not there is a boundary of clonal expansion within the neuroepithelium at this limit, although we do not believe that clonal restriction uniquely defines intersegmental limits (Box 1). Figdor and Stern [25] did not separate p3 from the hypothalamus, thus postulating their large D1 neuromere (D2 corresponds to p2 and D3–D4 seem to represent secondary AP subdivisions in p1, in so far as they are described only in the alar plate [25,26]; compare with p1 in Fig. 3). Figdor and Stern and Larsen *et al*. [25,26] reported p3 cell clones that partially extended into the adjacent telencephalon as evidence of lack of a p3–p4 clonal restriction boundary. These conclusions need to be re-examined considering that p3 includes an eminential portion that partially evaginates beyond the hemispheric sulcus and thereby encroaches upon the amygdala (the hemispheric sulcus accordingly is not the rostral boundary of p3; Fig. 3).

## Concluding remarks

Comparison of the final schema of Larsen *et al*. [26] with our model suggests fundamental agreement with our p1–p3 prosomeres and a non-segmented secondary prosencephalon; we distinctly converge in our views on fundamental forebrain subdivisions. The proposal to reduce terminological confusion by calling the classically defined dorsal thalamus simply 'thalamus' and the classically defined ventral thalamus 'prethalamus' seems apropos at this point in time [52]. Finally, the present model should be a useful template for future fate-mapping experiments, gene expression analyses, gene and cellular function investigations and cross-species anatomical comparisons, all of which will contribute towards establishing more definitive information about the topological organization of the prosencephalon.

### Acknowledgements

The histological material shown was prepared by G. González (laboratory of L.P.). Our work is supported by grants PI-64/00862/FS-01 and BFI2000–03668 (L.P.) and from Nina Ireland and K02MH01046 (J.L.R.R.). Valuable discussions took place during C. Larsen's short stay in Murcia to learn the prosomeric model.

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