Minireview

MOLECULAR METABOLISM

Embryonic development of the hypothalamic feeding circuitry: Transcriptional, nutritional, and hormonal influences

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ABSTRACT

Background: Embryonic neurogenesis and differentiation in the hypothalamic feeding circuitry is under the control of a variety of diffused morphogens and intrinsic transcription factors, leading to the unique structural and functional characteristics of each nucleus.
Scope of review: The transcriptional regulation of the development of feeding neuroendocrine systems during the period of embryonic neurogenesis and differentiation will be reviewed here, with a special emphasis on genetic and environmental manipulations that yield an adverse metabolic phenotype.
Major conclusions: Emerging data suggest that developmental mechanisms can be perturbed not only by genetic manipulation, but also by manipulations to maternal nutrition during the gestational period, leading to long-lasting behavioral, neurobiological, and metabolic consequences. Leptin is neurotrophic in the embryonic brain, and given that it varies in proportion to maternal energy balance, may mediate these effects through an interaction with the mechanisms of hypothalamic development.

Keywords Leptin; Developmental origins of health and disease; Hypothalamic neurogenesis; Basic helix-loop helix

Homeostatic control over feeding and energy balance is regulated in large part by the hypothalamus, specifically a network of nuclei comprising the arcuate nucleus (ARC), ventromedial hypothalamus (VMH), paraventricular nucleus (PVN) and the lateral hypothalamus (LH). Work over the past decade has made it clear that the perinatal life is a critical period for the organization and development of the feeding circuitry. Genetic disruptions to leptin signaling, as well as altered maternal nutrition can permanently disrupt this organizational process leading to an obesity-prone phenotype [1]. Prenatal life is also a critical period during which a wide range of genetic and environmental manipulations can alter the structure and function of the feeding circuitry, leading to adverse metabolic outcomes. This review is intended to summarize our current understanding of embryonic hypothalamic neurogenesis and differentiation, and apply this to the context of developmental programming. Understanding the various genetic and transcriptional mechanisms that govern normal development may point to novel directions in the study of developmental programming and the role of hormonal and environmental influences in development.

1. NEUROGENESIS AND DIFFERENTIATION IN THE DEVELOPING HYPOTHALAMUS

Neural tube development can be divided into stages, the first of which is regionalization. During the period prior to embryonic day 10 (E10), the diencephalic region is separated from surrounding regions by the influence of organizing signals such as Wingless/integrins (Wnts), Sonic hedgehog (Shh), Bone morphogenetic proteins (Bmps), and fibroblast growth factors (Fgfs), culminating ultimately in the induction of the NK2 homeobox transcription factor Nkx2.1 — a key marker of hypothalamic tissue (for review, see Ref. [2]). The subsequent neurogenic period encompasses E12-16 [3,4]. During the neurogenic period, neural stem cells in the ventricular zone give rise to neural precursors which, under the influence of factors discussed in this review, go on to assume their mature phenotype and position. Neurons, astrocytes, and oligodendrocytes all derive from the same progenitor pool, so the proneural transcription factors that characterize the neurogenic period simultaneously drive neurogenesis and suppress gliogenesis (for review, see Ref. [5]). This review will begin by covering

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medially situated nuclei, though it should be noted that neurogenesis in the hypothalamus proceeds in an 'outside-in' fashion, with laterally situated nuclei being born earlier [6] (Figure 1).

2. ARCUATE NUCLEUS

The ARC feeding circuitry is characterized by two non-coextensive neural populations expressing either Pro-opiomelanocortin (POMC) or Neuropeptide Y (NPY) and Agouti-related peptide (AgRP). Neural progenitors destined to partake in feeding circuitry may become either orexigenic NPY/AgRP neurons or anorexigenic POMC neurons, and the opposing nature of these populations brings the issue of cell fate decisions into sharp relief. POMC mRNA is first detected in the ventral hypothalamus at E10.5, and NPY expression between E13.5-14.5 [7]. In the period following E13.5, there is a drastic decrease in the number of POMC neurons [7]. This decrease cannot be accounted for by apoptosis, rather it seems that these cells begin to assume a NPY/AgRP phenotype [7]. It is not known what governs this change in cell fate, but it is clear that these two populations are intimately linked during development (Table 1).

The ARC develops in the context of a number of homeobox and basic helix-loop-helix (bHLH) transcription factors with temporally distinct patterns of expression. Retinal and anterior neural fold homeobox (Rax), a homeobox transcription factor traditionally defined by its involvement in early retinal development, is critical for the formation of the ventral neuroendocrine hypothalamus [8]. Rax is expressed in the ventricular zone medial to POMC expressing cells at E12.5, the expected location and time of a transcription factor involved in the generation of these cells [9]. Using Cre-mediated lineage tracing, Lu et al., found evidence that Nkx2.1-expressing neurons in the ARC derived from Rax-expressing lineages [10]. Mice lacking Rax in SIX homeobox 3 (Six3)-expressing cells do not show ventral hypothalamic Nkx2.1 expression, nor do they ever express POMC [10]. This appears to be due to a reassignment of cell fate, as the VMH comes to ectopically express GABAergic neural marker Gad67 in lieu of the expected terminal markers. When Rax is specifically knocked out in Shh-expressing cells, the formation of the VMH is uniquely affected — these animals retain approximately normal patterns of POMC expression [10].

The Oligodendrocyte transcription factor family (Olig1 & Olig2) are bHLH transcription factors expressed medially in the developing CNS, with the highest densities found in the periventricular regions [11,12]. Contrary to what is suggested by their names, Olig1 and Olig2 have been implicated not only in oligodendrogenesis but also in neurogenesis [11,13]. Lineage tracing experiments indicate that a number of POMC and NPY cells derive from Olig1 progenitors, and that the majority of Olig1 expressing cells also express Bone morphogenetic protein receptor 1A (Bmpr1A) [14]. When Bmpr1A is knocked out in Olig1 progenitors, affected mice fail to gain weight when transitioning to solid food [15]. These mice have significantly fewer POMC neurons, and yet significantly greater numbers of AgRP neurons [14]. Neither of these outcomes depend on a difference in neurogenesis, leaving changes in cell fate as a possible mechanism.

The Neurogenins (Ngn1-3) are another family of bHLH transcription factors important in ARC development. Ngn3 expression begins as early as E9.5 and extinguishes by E17.5 [16,17]. Cells co-expressing Ngn3 and POMC are not seen during development, but fate mapping studies show that many of the earliest born (ca. E10.5) POMC neurons arose from Ngn3-expressing progenitors — later born POMC neurons arise from non-Ngn3 expressing progenitors [17]. A large fraction of NPY neurons also appear to have descended from Ngn3-expressing progenitors [17]. Ngn3$^{-/-}$ mice show a substantial reduction in the number of POMC neurons born between E10.5-13.5, though this number rebounds somewhat by E17.5 [17]. On the other hand, NPY neuron counts are dramatically increased in Ngn3$^{-/-}$ mice, a difference that appears as soon as NPY begins to be expressed [18,17]. Mutant mice do not show any changes in overall cell count, implying a change in cell fate as opposed to altered rates of neurogenesis or apoptosis [17].

This work has recently been elaborated upon with the generation of a conditional mutant strain of mice in which Ngn3 is knocked out only in cells that express the ventral diencephalic marker Nkx2.1 [19]. In contrast to Ngn3$^{-/-}$ mice, the conditional mutants survive until adulthood, however they develop early-onset obesity and hyperphagia

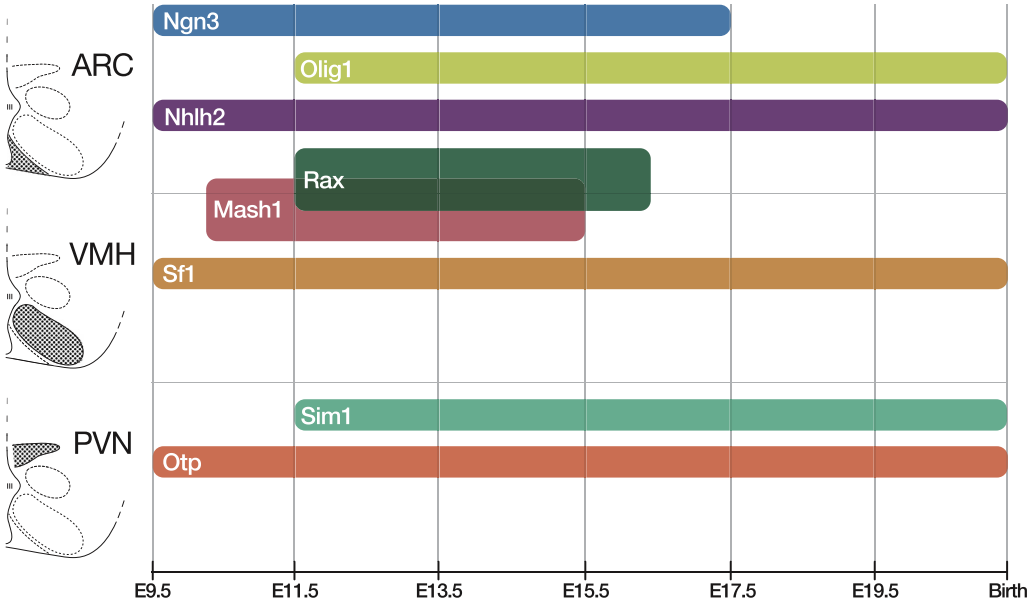

Figure 1: Expression timeline of transcription factors directly implicated in the generation of the hypothalamic feeding nuclei.

