The role of Hypothalamus in Regulating Hormonal Signaling, and Transcription Factors as potential Target on Cancer, and Metabolic disorders Treatment and Prevention(Part one)

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Abstract: The hypothalamus is located in the medio-basal region of the brain and acts as a gateway between the endocrine and nervous systems. The hypothalamus consists of a highly diverse collection of Inter-connected neurons and supporting glial cells that allow this region of the brain to sense and respond to a diverse range of hormonal and metabolic signals

From an endocrinology perspective, the parvi-cellular neurons of the hypothalamus are of particular interest as they function as a control centre for several critical physiological processes including growth, metabolism and reproduction by regulating hormonal signaling from target cognate cell types in the anterior pituitary.

There is evidence suggesting that the newly generated hypothalamic neurons may involve in regulation of metabolism, energy balance, body weight, and social behavior as well.

In recent years, studies have shed light on the interactions between signaling molecules and activation of transcription factors that regulate hypothalamic cell fate commitment and terminal differentiation.

In this article, I discuss Structure of the Hypothalamus, Development of the Hypothalamus, Functional anatomy of the neuroendocrine hypothalamus, Hypothalamic induction and the role of signaling pathways Patterning the hypothalamic primordium

Key Word: Hypothalamus,Neuropeptides,Neurogenesis, Metabolic disorders and Cancer

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1. Introduction

The brain is crucial in regulating metabolism (1).Within the brain, a number of anatomical regions are recognised to play a role in metabolic homeostasis (2),(3). However, the hypothalamus in particular is critical in sensing and integrating signals from the periphery and effecting appropriate physiological changes to maintain homeostasis. The hypothalamus influences a broad spectrum of physiological functions, including pituitary hormone synthesis and secretion, autonomic nervous system activity, energy intake and expenditure, body temperature, reproduction and behavior. The hypothalamic parvicellularneuro-secretory neurons are of particular interest due to their role in controlling anterior pituitary (AP) hormone secretion. While much of the early research into hypothalamic development and function has been conducted in rats, recent advances in murine transgenesis and mutagenesis techniques have established mice as the principal model for analysis of central nervous system (CNS) development.While much of the early research into hypothalamic development and function has been conducted in rats, recent advances in murine transgenesis and mutagenesis techniques have established mice as the principal model for analysis of central nervous system (CNS) development. Therefore, in this article I have focused the role of Hypothalamus in regulating hormonal signaling, transcription factors and Neurogenesis.

2. Neuropeptides

Neuropeptides represent large and diverse group of molecules responsible for communication among cells in the central nervous system (CNS). Although neuropeptides may be located in the periphery and also play a role in controlof peripheral functions, their major effects are within the CNS, by taking part in the regulation of thermoregulation, food and water intake, circadian rhythms, and sexual and reproductive behavior. A molecule can be considered as a neuropeptide, when it possesses distinct properties:(4),it is a small-size protein molecule,(4),it is produced and secreted by cells of the nervous system, and(5),it plays a specific role in the regulation of neuronal cells(6).At least in mammals, neuropeptides are encoded by over 70 genes(6).The size of the molecule is usually between 3 and 100 amino acids (AC), while more than 75% of known neuropeptides have a molecule of less than 30 AC(7).Neuropeptide synthesis takes place mostly in a physiologically inactive form as the pre-pro-molecules. The precursor, before being stored or released from the cell, is typically degraded to short chain of

AC through endo-peptidases in the Golgi apparatus or directly in the secretory vesicles(8),(9).Nevertheless, some neuropeptide molecules undergo further posttranslational modifications, necessary to ensure their stability and full biological activity, such as phosphorylation, acetylation, sulphonation, or removal of their terminal part. Metabolic changes necessary to achieve a fully active form of the neuropeptide are sometimes so intensive that the result leads to an extreme shortening of the peptide chain. Particularly, thyrotropin releasing hormone (TRH) consists only of three amino acids, compared to its much larger pre-pro-form (10).Neuro-peptides are secreted from large dense core vesicles by regulated secretion. They may be stored in vesicles together with other low molecular weight neuropeptides or even with other neurotransmitters(11).Their secretion is not necessarily limited to the synaptic cleft; however it usually occurs in the close vicinity(10),(12),(13).There are also reported cases of secretion from the cell body or from dendritic spines(14).Neuropeptides play a crucial role in cell-to-cell communication by affecting gene expression(15),synaptogenesis (16),and modulation of membrane excitability(17).Some neuropeptides even may act as neurotransmitters(18).Despite the often generalized physiological effects of many neuropeptides, time of their biological activity in the circulation is significantly limited. For instance, oxytocin has a half-life in blood of approximately 120 seconds, compared to the half-life in the CNS extracellular space, which is about 20 minutes(19).Diffusion through the extracellular space and binding to membrane receptors are in a case of robust neuropeptide much slower, however, from the physical-chemical point of view, more solid(20).Slower modulatory effect on the potential of the postsynaptic membrane is linked to the mechanism of the neuropeptide receptor pathway. Most neuropeptides have their own specific receptor coupled with G-protein. Although the size of neuropeptide molecules is relatively large compared to classical neurotransmitters, affinity to the specific receptors is approximately 1000-fold higher than that of the neurotransmitter, thus being capable of eliciting a biological response at lower concentrations(17).

3. Classification of Neuropeptides

Up to date, the different databases cover over 5900 neuropeptides divided into large groups(6),(7),(8).While the number of neuropeptides in vertebrates reaches nearly 2500(7),it can be expected that the list is still not complete. The division into families may be based on similarities in the gene structure (e.g., calcitonin gene family, F- and Y-amide gene family), molecule structure (e.g., oxytocin/vasopressin family, insulin/insulin-like growth factor (IGF) family), function (e.g., opioid neuropeptide family, adipose neuropeptide family), or localization of neurons producing each neuropeptide (hypothalamic neuropeptide family, hypophysealneuropeptide family). Many novel neuropeptides remain unclassified. One given peptide is often localized to different brain areas and it is involved in more than one biological function. Neuropeptides expressed in hypothalamic neurons form a large group of well-described peptides with a variety of peripheral (endocrine) and central functions.

4. Structure of the Hypothalamus

4.1. Hypothalamic Neuronal Populations

The hypothalamus is an ancient and conserved forebrain area, traditionally divided to lateral, medial, and periventricular part and furthermore to the distinct functional

nuclei(21).Hypothalamic nuclei contain diverse cell populations(22),which can be defined by specific patterns of gene expression, such as ion channels, transcription factors, and neuropeptides. Populations of neurons secreting various neuropeptides located in the lateral hypothalamus play a major role in food intake. In the arcuate nucleus, neurons express orexigenic agouti-related peptide (AgRP), Neuropeptide Y (NPY), and anorexigenic peptides proopio-melanocortin (POMC). Another group of neurons produce peptides promoting food Intake orexin and melanin-concentrating hormone (MCH)(23).Nevertheless, arcuate hypothalamic neurons that produce proopiomelanocortin (POMC) secrete an anorexic neuropeptide melanocyte-stimulating hormone (α-MSH), a proteolytic product of POMC. Another endogenous peptide product of the POMC represents adrenocorticotropic hormone (ACTH), β- and γ-melanocyte-stimulating hormones (β- and γ-MSH), and β- endorphin. Located lateral to the arcuate nucleus, the ventromedial nucleus is the major constituent of the mediobasal hypothalamus. Ventromedial nucleus is important in the regulation of sexual behavior and analgesia(24).Large amount of neuropeptides, such as Substance P, enkephalins, and NPY, is synthesized in the ventromedial nucleus(24).The periventricular part of the hypothalamus is responsible for secretion of NPY, TRH, somatostatin, leptin, gastrin, and gonadotropin-releasing hormone. Paraventricular and supraoptic nuclei of the hypothalamus contain neurons producing corticotrophinreleasing hormone (CRH), TRH, oxytocin, and vasopressin

4.2. Hypothalamic Neuronal Connections

The hypothalamussends information directly to other brain areas andto periphery by neural projections and indirectly to the blood stream; neuro-endocrine regulation ismediatedmostly via hypothalamic-pituitary-adrenal, hypothalamic-pituitarythyroideal, and hypothalamic-pituitary-gonadal axes. Proper transmission of neural signals from periphery to the hypothalamus ismediated by visceral and somatosensory inputs. Furthermore, control of the autonomic nervous systemis assured by direct outputs to the brain stem. It is verywell documented that neural projections originating or terminating in the hypothalamus are involved in regulation of food, energy, and heat balance. The hypothalamo-neurohypophyseal system plays a fundamental role in the control of fluid and electrolyte balance forming complex neural network responsible for an integrated response(25).It is also known that olfactory receptor neurons formcircuitswith hypothalamic sub-regions(26).Next, pathways from retina to the suprachiasmatic nucleus of the hypothalamus are involved in regulation of circadian rhythms and light-dark cycle. Projections from the hypothalamus to the cerebral cortex participate in the control of sexual, reproductive, and social behavior(21),(22),(27).

5. Development of the Hypothalamus

Therelevant information on functional organization of intraand inter-hypothalamic circuits has dramatically increased in the last decades. The hypothalamus has complex connections with other brain regions ranging from retina to cortex. These connections are formed during embryonic development; however they are further rearranged later in life under conditions of nutritional state, stress, or lactation(28),(29),(30).Hypothalamic circuits, connections, and pathways are thus dynamically regulated resulting in marked changes of brain plasticity manifested by enhanced neurogenesis and neuritogenesis. It is known that hypothalamic neurogenic niche (hypothalamic proliferating zone) lining the ventral portion of the third

ventricle consists of cells with high proliferative activity even in the adult age(31),(32).Precursor cells lining the third ventricle are able to receive diverse molecular signals, for example, neuropeptides, and growth factors present in the cerebrospinal fluid.Mounting evidence suggest that hypothalamic neurogenic capacities can be affected in the adult mammalian brain(33).In addition to production of neurons, shift from neurogenesis to gliogenesis has been shown in the developing hypothalamus(34).Traditionally, it was believed that most of the hypothalamus is formed in three neurogenetic stages producing neurons that progressively accumulate; however recent studies suggest that hypothalamic progenitor cells have common origin(34).Nevertheless, it is known that the hypothalamus develops from the rostral diencephalon and cells from various origins migrate to the hypothalamic region during development. Hypothalamic neuron populations are under the control of many intracellular transcriptional factors. The most known are sonic hedgehog protein (Shh)(34),(35),and a group of proteins belonging to wingless family (Wnt), which has been long known to be involved in patterning during development(32).Shh is considered as a morphogen that regulates the dorsoventral patterning of central nervous system. Recent study has demonstrated that Shh coordinates anteroposterior and dorsoventral patterning in the hypothalamus(36),moreover it has been reported that chemorepulsive effect of Shh repels hypothalamic axons from the ventricular zone of the hypothalamus and results in their growth in fascicules(37).Differentiated neurons or glia cells cease to express Shh(38).Wnt signaling is required for neurogenesis and eventually for anterior patterning, including the region that gives rise to the hypothalamus(32),(39).Newborn cells have been described in the adult hypothalamus, suggesting constitutive neurogenic and cell proliferation responsive to mitogen action(40).The development of hypothalamic tissue is under control of other morphogenes, namely, bone morphogenetic proteins and fibroblast growth factors (FGF).Neurites in the hypothalamus are guided to their targets by many attractive and repulsive guidance molecules netrins, slits, semaphorins, and ephrins that have been reviewed in the context of autism elsewhere(41).Complex molecular interactions, including the action of neuropeptide oxytocin, occur at the origin of the hypothalamic region and generation of hypothalamic cell types during development (42),(43).

6. Functional anatomy of the neuroendocrine hypothalamus

The vertebrate hypothalamus is located ventral to the thalamus and dorsal to the pituitary gland, at the mediobasal region of the CNS. It extends from the optic chiasm (located anteriorly) to the mammillary body (located posteriorly) and is organized into four distinct rostral-to-caudal regions: preoptic, anterior, tuberal, and mammillary. It is also divided into three medial-to-lateral areas: periventricular, medial and lateral. The periventricular hypothalamus contains four distinct cell clusters: the paraventricular nucleus (PVN), arcuate nucleus (ARC), supra-chiasmatic nucleus (SCN), and the periventricular nucleus (PeVN; The medial hypothalamic zone is comprised of the medial preoptic nucleus, the anterior hypothalamus (AH), the dorsomedial nucleus, the ventromedial nucleus (VMN) and the mammillary nuclei. The lateral hypothalamus consists of the preoptic area (POA) and hypothalamic area. Located throughout hypothalamus are hypothalamic neurosecretory cells that are divided into two populations: the parvicellular and magnocellularneurosecretory systems. The former consists of neurons controlling the release of specific AP neurohormones: thyrotropin-releasing hormone (TRH;

located within the medial part of the medial parvicellular subdivision of the PVN), corticotropin-releasing hormone (CRH; located within the lateral part of the medial parvicellular subdivision of the PVN), growth hormone-releasing hormone (GHRH; located within the lateral part of the ARC), somatostatin (SS; located within the PeVN), gonadotropin-releasing hormone (GnRH; located within the medial POA), dopamine (DA; located within the medial part of the ARC and detected by the enzymatic activity of tyrosine hydroxylase) and, the recently discovered gonadotropin-inhibiting hormone (GnIH; located within the dorsomedial nuclei in rodents; The magnocellularneurosecretory system consists of neuronal cells secreting two neurohormones, vasopressin (AVP) and oxytocin (OT), whose axons project directly into the posterior pituitary (neurohypophysis) and release peptides systemically in response to various homeostatic cues (osmotic, cardiovascular and reproductive). The primary focus of this review is the development and function of the parvicellular neurons. For in-depth information and discussion on the magnocellularneurosecretory system we refer the reader to the paper by(44).

7. Hypothalamic induction and the role of signaling pathways

The hypothalamus develops from the ventral region of the diencephalon(45),and, in the mouse, its primordium is morphologically evident from approximately 9.5 days post coitum (dpc; where 0.5 dpc is defined as noon of the day on which a copulation plug is present). Developmental studies performed in mice, chick and zebrafish indicate that sonic hedgehog (SHH) signaling plays an important role in the induction and early patterning of the hypothalamus(46),(47),(36).of SHH from the murine axial mesendoderm, from 7.5 dpc, is essential for correct patterning of the anterior midline. In humans as well as in mice, mutations in the *SHH/Shh*gene (and several other components of this pathway) result in holopro-sencephaly due to a failure of hypothalamic anlagen induction and optic field separation(48),(49).Increased SHH activity leads to ectopic expression ofhypothalamic markers in zebrafish, suggesting that SHH signaling has an instructive rather than a permissive role in shaping the hypothalamus(50),(51),(52).Studies in chick have shown that once the hypo-thalamic primordium is established, down-regulation of *Shh*is critical for the progression of ventral cells into proliferating hypothalamic progenitors, at least within the ventral tubero-mammillary region(46).In addition, *Shh*down-regulation is mediated, to some extent, by local production of Bone Morphogenetic Proteins (BMPs), which belong to the transforming growth factor-beta (TGFβ) super family of signaling proteins(46).This antagonism between SHH (ventral gradient morphogen) and BMP (dorsal gradient morphogen) in the hypothalamus is reminiscent of their opposing actions in dorsal-ventral patterning of the neural tube. However, in the developing hypothalamus this incorporates a temporal aspect (SHH early - BMP late) that appears necessary for establishing region-specific transcriptional profiles(53),(54).Although axial secretion of another member of the TGFβ super-family, NODAL, is also necessary for hypothalamic induction, the early lethality of *Nodal* mutants has precluded detailed assessment of its role in hypothalamic development in mice(55),(56).Genetic studies in zebrafish have shown that the Wnt signaling pathway is required for specification of the hypothalamic anlagen, its regionalization and neurogenesis(57),(39).Together, these studies have shown that hypothalamic induction and pattern formation depends on the activities of major protein signaling pathways involved in patterning, regional identity and cell fate determination.

8. Patterning the hypothalamic primordium

Embryonic neurogenesis in vertebrates follows a stereotypical progression that begins with the generation of the neural tube, which is composed of a pseudostratified columnar epithelium of cycling stem cells. As a general rule, these neuronal precursors acquire distinct positional identities, commit to a neuronal fate, exit mitosis, migrate away from the periluminalprogenitor zone and terminally differentiate. A large body of evidence, gained principally from mouse and chick embryos, has established that transcription factors belonging to the homeo-domain and basic Helix-Loop Helix (bHLH) families play a major role in neurogenesis (58).Regionally restricted expression of these factors is induced in response to local signaling cues (see above), establishing a transcription factor "code" that directs the generation of distinct neuronal cell types at each neuroaxial level. Mouse mutagenesis has identified several transcription factor pathways critical for the development of the parvicellular neurons in the POA, PVN, PeVN, VMN and ARC, which together provide the foundation for a rudimentary "hypothalamic transcription factor code" and are outlined below.

8.1. Sim1/Arnt2-Brn2 pathway: The bHLH-PAS transcription factor SIM1 is expressed in the incipient PVN, supraoptic nucleus (SON), and anterior PeVN (aPeVN) from 10.5 dpc and is maintained in these regions into postnatal development(44),(59).Homozygous *Sim1* mutants die shortly after birth and exhibit significant hypoplasia of the anterior hypothalamus. Histological and molecular marker analysis has revealed that these mutants lack virtually all neurons of the SON and PVN, including those expressing TRH and CRH. SS neurons in the aPeVN and other populations of TRH neurons in the lateral hypothalamus and in the POA region are also missing. Interestingly, mutant mice lacking the *Sim1* dimerisation partner ARNT2 have a strikingly similar phenotype, indicating that these proteins function co-operatively in the AH(60),(61).A key downstream target of SIM1/ARNT2 is *Brn2,* which encodes a POU domain transcription factor and is required for the differentiation of CRH (as well as OT and AVP) neurons of the PVN/SON. *Brn2* expression in the prospective PVN/SON region is absent in *Sim1* and *Arnt2* mutants, indicating that *Brn2* is regulated by SIM1/ARNT2, although it is not currently known if this is a direct or indirect interaction.

8.2. Otp:The homeobox gene *Orthopedia*(*Otp*) is expressed in neurons giving rise to the PVN, SON, aPeVN and ARC throughout their development. *Otp*mutants die as neonates and fail to generate the parvicellular and magnocellular neurons of the anterior PeVN, PVN, SON, and ARC(62),(63). These defects are associated with reduced cell proliferation, abnormal cell migration, and failure of terminal differentiation. Like the *Sim1* and *Arnt2* mutants, *Otp*null embryos fail to maintain *Brn2* expression. However, OTP does not appear to directly interact with SIM1 or ARNT2(44),and SIM1 and OTP donot regulate each other's expression, suggesting that OTP and SIM1/ARNT2 operate in parallel or convergent pathways.

8.3. Nkx2.1: During early development of the CNS, signals produced from the anterior axial mesendoderm induce expression of the homeodomain transcription factor gene *Nkx2.1* (also known as T/ebp) in the overlying presumptive hypothalamus(64),(65).*Nkx2.1* mutant mice die at birth and, in addition to lung and thyroid defects, exhibit profound abnormalities in the ventral hypothalamus, including agenesis of the ARC and VMN. Interestingly, null mutants also fail to generate the Rathke's pouch (which does not express *Nkx2.1*), confirming the ventral diencephalon/infundibular recess is essential for induction of the AP(65),(66).

8.4. Sf1: The *Sf1* gene encodes an orphan nuclear hormone receptor that is required for normal development of the gonads and adrenals and function of pituitary gonadotropes(67),(68).Within the CNS, *Sf1* is specifically expressed within the VMN and is required for multiple phases of VMN development. Analysis of *Sf1* null embryos indicates that this transcription factor is initially involved in the survival and migration of VMN precursors from the ventricular zone and at later stages is required for aggregation and condensation of the VMN nucleus and terminal differentiation.

8.5. Hmx2/Hmx3: Two closely related homeobox genes, *Hmx2* and *Hmx3,* are expressed in overlapping domains of the ventral hypothalamus from 10.5 dpc(69).While single gene mutants do not have any discernable hypothalamic phenotype (although it bears noting that ear development is affected), *Hmx2*;*Hmx3* null mice exhibit postnatal dwarfism and a severe deficiency of GHRH neurons in the ARC, but not the VMN(69).Expression of the homeobox gene *Gsh1*, which overlaps with *Hmx2* and *Hmx3* and is required for *Ghrh*expression, is also absent in *Hmx2;Hmx3* null embryos. Neuronal cell numbers in the ARC are not significantly different in double mutants indicating that, despite their widespread expression, *Hmx2* and *Hmx3* are not required for early determination of neuro-progenitors in this region of the hypothalamus.

8.6. Mash1:MASH1 is a proneural protein that belongs to the bHLH family of transcription factors and is required for neurogenesis and subtype specification in many regions of the CNS(70).*Mash1* is expressed throughout the ventral retrochiasmaticneuroepithelium from 10.5-12.5 dpc. *Mash1* null embryos exhibit hypoplasia of the ARC and VMN nuclei due to neurogenic failure and increased apoptosis(71).Using a knock-in strategy,(71),elegantly showed that this phenotype could be rescued by ectopic expression of *Ngn2*, which is also a member of the bHLHproneural gene family. *Mash1* also appears to have a role in subtype specification (that cannot be rescued by *Ngn2*), and is absolutely required for expression of *Gsh1* and the subsequent generation of *Ghrh*-expressing neurons.

8.7. Sox3:*Sox3*is a member of the *SOX* (Sry-related HMG box) family of transcription factor genes and is located on the X chromosome (72).This gene was initially implicated in hypothalamic development from clinical and genetic studies of families with the male-specific congenital disorder X-linked Hypopituitarism (XH). XH males have GH deficiency and, in some cases, additional pituitary hormone deficiencies as well as intellectual disability(73).Magnetic resonance imaging analysis of affected males has revealed abnormalities of the hypothalamic region including ectopic posterior pituitary and thin pituitary stalk, indicating that XH results primarily from a hypothalamic defect(74).Interestingly, XH is associated with duplications and mutations in *SOX3*, suggesting that over-expression and loss-of-function mutations result in a similar developmental defect(73),(74).Although the mechanism by which altered *SOX3* dosage causes XH is not fully understood, genetic studies in mice have provided some clues. *Sox3* null animals exhibit multiple pituitary hormone deficiency, variable dwarfism and CNS abnor-malities, indicating that SOX3 function is broadly conserved in mice and humans(75),(76).Importantly, *Sox3* is expressed in the developing hypothalamus but has minimal expression in the AP, suggesting that hypothalamic (and not AP) dysfunction is the primary cause of pituitary hormone deficiency in *Sox3* mutants. Studies from our laboratory have shown that *Sox3* is expressed in the hypothalamus from inception to maturity suggesting that it may have multiple roles in hypothalamicdevelopment and function. Analysis of *Sox3*

mutants has indicated that early expression in the ventral diencephalon/infundibular recess (at 10.5 dpc) is required for normal induction and morphogenesis of the AP, but, remarkably, not AP function(75).From approximately 12.5 dpc, *Sox3* expression is restricted to multiple hypothalamic regions/nuclei including the hypothalamic neuroepithelium, median eminence, ARC, PVN, medial POA and VMN. Interestingly, all of these nuclei contain parvicellular neuronal subtypes. It is therefore possible that the multiple pituitary hormone deficiencies in *Sox3* null mice (and some XH patients) may reflect a specific requirement for *SOX3* in the generation and/or maintenance of some, if not all, parvicellular neuronal subtypes. Alternatively, or in addition, defective development of the median eminence, which also expresses *Sox3 (75),* and our unpublished data), may compromise the functional connection to the portal vasculature, resulting in altered regulation of AP hormone synthesis and secretion by parvicellular neuronal factors.

9. Conclusion

It is important to understand the role of hypothalamic neuropeptides in neurogenesis and neuritogenesis. Particularly, small-size neuropeptides may play a role in neuronal proliferation and differentiation influencing growth and guidance of neurites and participating in the formation of neural circuits in early development. It is clear that distinct sets of transcription factors play a role in the differentiation of hypothalamic progenitor cells into neurons and the commitment of subsets of neurons into cells that secrete hypophysiotropic factors. These factors provide an important framework for further functional studies that may lead to the generation of a transcriptional code for hypothalamic development. This process will likely be informed by parallel studies of other brain regions where knowledge of neuronal subtype specification and differentiation is further advanced. While parallel studies will provide useful intellectual synergy, it will also be necessary to focus on the discovery of novel hypo-physiotropic cell molecules and pathways. This will be facilitated by recent advances in molecular and cellular biology including the identification of hypothalamic transcription factor gene targets using ChIP sequencing analysis, directed differentiation of ES cells into hypothalamic neuronal fates, and characterization of novel mouse models using N-ethyl-N-nitrosourea (ENU) mutagenesis. As the role of new hypothalamic genes is deciphered it may become possible to detect patterns that will lead to a clearer understanding of brain development and evolution of the neuroendocrine system. Furthermore, further studies should focus on neuropeptide pathways and their changes during development. Conditional formation of neuronal circuits is extremely important. Maintenance of balance in the orexigenic and anorexigenic hypothalamic neuropeptides is especially important in the context of maximized energy intake and mass gain during early stages of development.

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