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Vitamin D, Testosterone, Epigenetics and Pain an Evolving Concept of Neurosignaling, Neuroplasticity and Homeostasis

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Abstract

The thought of exploring a possible relationship between the broad systems of steroid hormone physiology (specifically vitamin D and testosterone) and nocioception was prompted by an unexpectedly frequent personal clinical observation. Patients with chronic pain syndromes or chronic musculoskeletal pain often have low serum levels of vitamin D and testosterone. Mining for relevant information in Pub Med, Medline and Cochrane Systems Review, three concepts repeatedly emerge that provide a common context for understanding the mechanics of these diverse systems—epigenetic, homeostasis and neuroplasticity. Viewing homeostasis within the framework of epigenetics allows reasoned speculation as to how various human systems interact to maintain integrity and function, while simultaneously responding in a plastic manner to external stimuli. Cell signaling supports normal function by regulating synaptic activity, but can also effect plastic change in the central and peripheral nervous system. This is most commonly achieved by post-translational remodeling of chromatin. There is thus persistent epigenetic change in protein synthesis with all the related downstream effects but without disruption of normal DNA sequencing. In itself, this may be considered an example of genomic homeostasis. Epigenetic mechanisms in nociception and analgesia are active in the paleospinothalamic and neospinothalamic tracts at all levels. Physiologic response to a nociceptive insult, whether mechanical, inflammatory or ischemic, is provided by cell signaling that is significantly enhanced through epigenetic mechanisms at work in nociceptors, Gamma-Aminobutyric Acid (GABA) and glutamate receptors, voltage gated receptors, higher order neurons in the various dorsal horn laminae and proximal nociceptive processing centers in the brainstem and cortex. The mediators of these direct or epigenetic effects are various ligands also active in signaling, such as free radicals, substance P, a variety of cytokines, growth factors and G proteins,

stress responsive proteins, matrix and structural proteins such as reelin and the Jmjd3 gene/enzyme. Calcitriol, the vitamin D receptor and vitamin D Responsive Elements collectively determine regulatory effects of this secosteroid hormone. Agents of homeostasis and plasticity include various D-system specific cytochrome enzymes (CYP 24, CYP 27 A1, B1), as well as more widely active enzymes and protein cell signalers (Jmjd3, Calbindin, BMP), many of which play a role in the nociceptive system. While the highlighted information represents an understanding of complex systems that is currently in its infancy, there are clear results from reliable research at a foundational level. These results are beginning to tell a compelling tale of the homeostasis and plasticity inherent in vitamin D and nociception systems.

Keywords

Gamma-Aminobutyric Acid (GABA), Jumonji (Jmjd3), Homer Gene, Cholecalciferol, Calcitriol, Neurosteroids, Glutamate, 5-HT

1. Introduction

A Possible Relationship between Nocioception, Vitamin D3 and Testosterone

The thought of exploring possible relationships between the broad content domains of vitamin D Metabolism, testosterone related physiology and nocioception as regulated by typical and epigenetic mechanics, within the context of homeostasis and neuroplasticity, was prompted by a personal and unexpectedly frequent clinical observation. I have seen that many patients with chronic pain syndromes or chronic musculoskeletal pain have low serum levels of vitamin D and testosterone. Admittedly, this personal observation is inconsistently supported by high-quality scientific investigations, and understandable by a host of plausible mechanisms. Nonetheless the following thought process draws upon an exploding body of knowledge as it pertains to the not so disparate steroids vitamin D and testosterone, and the emerging concepts that are elucidating the processes of nociception and epigenetics.

A body of knowledge, as identified through Cochran systems review, Pubmed and Medline, exists that supports the idea that epigentics plays a significant role in the nociceptive process, vitamin D dependent systems and testosterone metabolism. The epigenetic influences that affect steroid hormones are also being identified.

At this time, a link between vitamin D function and the nociceptive system seems probable, while a link with testosterone more tenuous. Much of the highlighted information represents an understanding of complex systems that is currently in its infancy, but there are clear results from reliable research at a foundational level and these results are beginning to tell a compelling tale of the homeostasis and plasticity inherent in human physiology.

2. Background

2.1. Epigenetics

Epigenetics is the study of changes gene expression (phenotype) without change in the underlying DNA sequence [1]. This term is used for processes which are both heritable and transient (in a generational sense). In general, the focus of epigenetics is on stable changes maintained across several cell generations [2].

The geneticist Dr. A. Riggs provides a useful operational definition of epigenetics as "the study of mitotically and/or meiotically heritable changes in gene function that cannot be explained by changes in DNA sequence" [3]. In 2008, the consensus definition of epigenetic process was written by a consortium of geneticists as a "stabley heritable phenotype resulting from changes in a chromosome without alterations in the DNA sequence" [4]. For the purpose of this paper, epigenetic processes are the non-genomic mechanisms that result in this type of change.

Epigenetics is typically divided into; predetermined and probabilistic epigenesis [5]. Predetermined epigenesis is a one way progression from DNA structure to functional maturation of a protein that is predictable [6]. Probabilistic epigenesis allows a bidirectional structure-function relationship between DNA and protein, with extrinsic factors being key determinants of gene expression.

With epigenetic mechanics, there is no change in the nucleotide sequence. They allow the genome to display both plastic and homeostatic properties, adapting to change across cell generations while maintaining genetic integrity. The resulting ability to differentially express protein synthesis is an integral part of human development and adaptation [7].

Epigenetic change in response to an environmental trigger or stimulus can have tremendous impact on an organism. For example, mice fed supplements that alter expression of the agouti gene show variability in fur color, weight, and propensity to develop cancer [8]. In humans, identical twins with different environmental exposures develop clear differences in the content and genomic distribution of 5-methylcytosine DNA and histone acetylation that is reflected in differences in their medical histories [9].

Epigenetic change is achieved through diverse strategies including DNA methylation and histone modification therefore altering genetic expression and without changing the DNA sequence [10] (**Figure 1**).

Chromatin (**Figure 2**) is activated or silenced at either the transcriptional or post-transcriptional level. Transcriptional activation may occur through histone modification and transcriptional silencing is more commonly due to DNA methylation and epigenetic "crosstalk" between the processes may add yet another dimension to epigenetically directed plasticity and homeostasis [11] [12]. Post-transcriptional gene silencing is the result of blocking the mRNA of a particular gene, generally by a small non-coding RNA [13]. The destruction of the mRNA prevents gene-directed protein synthesis.

Chromatin is fundamentally DNA wrapped around histone proteins. The

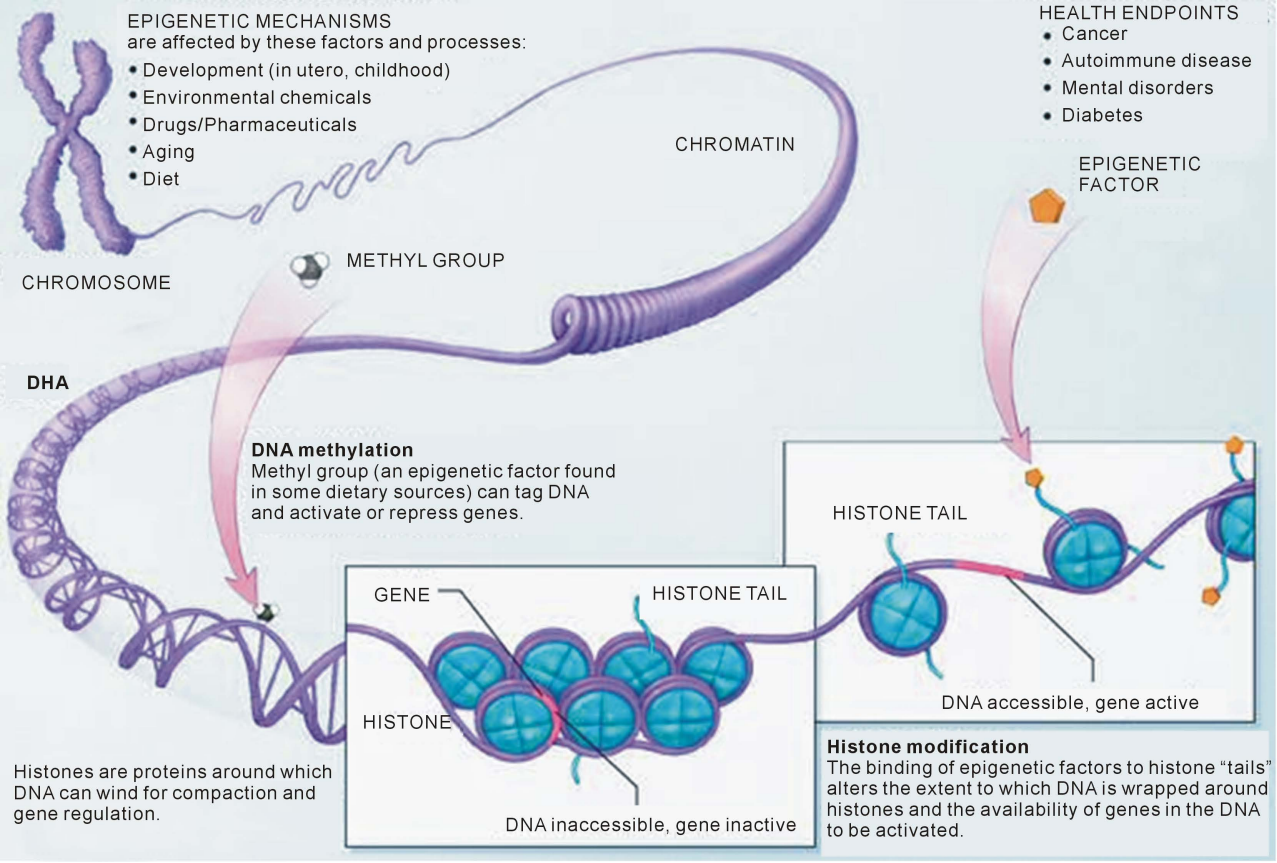

Figure 1. A scientific illustration of how epigenetic mechanisms can affect health (https://commonfund.nih.gov/epigenomics/figure).

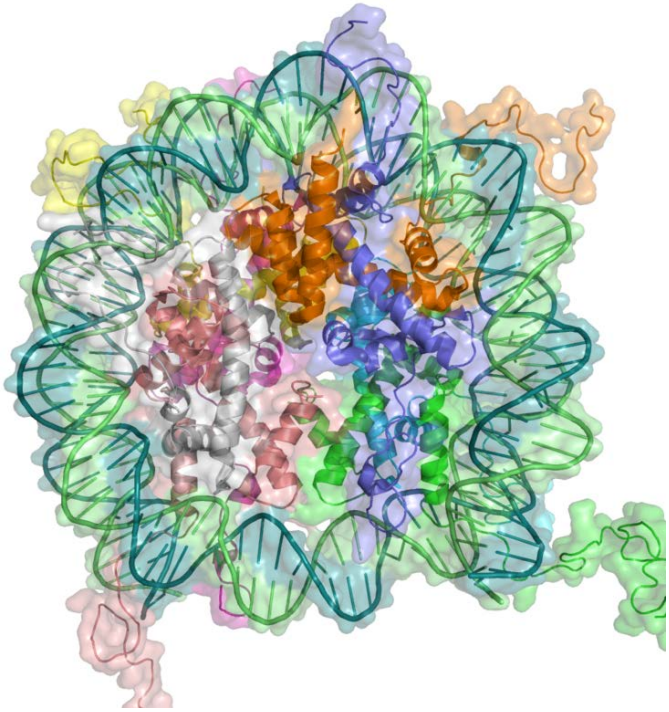

Figure 2. Chromatin-DNA plus histone complex (https://commons.wikimedia.org/wiki/File%3ANucleosome_1KX5_2.png). By Richard Wheeler (Zephyris) 2005.

confirmation of this complex determines a transcription state. Remodelling of the chromatin results in epigenetic, or heritable, transcription states. The change in gene expression is related to the way that histone, or the mated DNA, changes.

Change in the histone component of chromatin, or remodeling change, occurs by post-translational change in the long amino acid chains that form histone proteins. This alters the configuration of the histone, and the histone-DNA complex such that protein synthesis is altered by the change in available DNA loci. In a similar manner, during replication the altered configuration may act as a new template and the altered chromatin is carried forward, although the reality of this heritability is a matter of current controversy.

The way that histone proteins change is through acetylation, methylation, phosphorylation, enzyme binding of the respiratory protein ubiquitin (ubiquitylation) or similar addition of a related protein (small ubiquitin-like modifier; sumoylation) [13] [14] [15]. This is meaningful at the N-terminus (tail) where it impacts transcriptional competence, perhaps by removing the "native" positive charge. This affects the DNA (negative phosphate backbone)-histone (positive nitrogen tail) bond and provides DNA access to transcriptional factors—the cis model of epigenetic function where changes in the histone tail directly affects DNA expression [16] [17]. Histone tail changes may also indirectly impact DNA. In the trans model, the histone modification creates a binding site for chromatin modifying enzymes [18].

The various possible histone modifications appear to produce a variable response that is dependent upon the specific modification, the site of modifcation and the combination of multiple and varied modifications [19]. This produces a very broad and dynamic range of differnential gene expressions (the result of a putative histone code). In most cases, histone modification is most commonly transcriptional activating [20].

Epigenetic change in DNA results from Methylation. Generally this occurs at the sites where cytosine and guanine are bound by a phosphate group, the CpG site [21]. Methylation converts cytosine to 5-methylcytosine. DNA methylation helps to suppress the expression and mobility of transposons (the "jumping genes" or transposable elements within a gene) [22] [23] [24] [25]. This highly methylated areas of the genetic base pairs tend to be less transcriptionally active. DNA methylation occurs in response to an external stimulus through the mediation of multiple independent DNA methyltransferases.

Methylation of histone may also provide a site for transcriptional factor binding. These are most typically inactivating and may silence the involved gene [26] [27]. Methylation may also, incidentally, reveal the lineage of a chromosome as the additional methyl group is passed in a germ cell line, an aspect of genetic imprinting, due to enzymatic preferential affinity for methylated cytosine [28] [29] [30].

Histones H3 and H4 can also be demethylated by histone lysine demethylase

(KDM) at the active site—the Jumonji domain (JmjC). JmjC is involved in demethylating mono-, di-, tri-methylated substrates [31] [32].

Yet another mechanism of chromatin change is via non-coding RNA. From a mechanical perspective these changes have generally been thought to silence gene expression, although their role in gene activation is increasingly identified [33].

Approximately 60% of genes coding for human protein are regulated by micro RNA (miRNAs) and many miRNAs are epigenetically regulated. miRNA genes are repressed by epigenetic methylation. Other miRNAs are epigenetically regulated by histone modifications or by combined DNA methylation and histone modification. Such potentially sophisticated epigenetic regulation of an epigenetic agent is an example of how homeostasis and plasticity interact at a fundamental level in biological systems [34] [35] [36].

More than 2000 miRNAs have been identified to date within humans [37]. And as each miRNA expressed in a cell may down-regulate over one hundred messenger RNAs, the potential impact of epigenetic modification at this level is immense.

Ribozymes, alternately, are catalytic RNA molecules that do not down-regulate messenger RNAs, but rather cleave them, thus silencing gene expression [38] [39] [40].

Additionally, small interfering RNA's represent a vast array of small non-coding RNA's that are involved in epigenetic gene regulation [41].

Epigenetic change can also be effected by various mechanisms that play a significant role in the developing organism. A list of the most common such processes includes paramutation, bookmarking, imprinting and transvection [42] [43]. These processes are largely predetermined, at times latent, epigenetic events. Another mechanism, DNA damage, is epigenetically probabilistic [44].

DNA damage which is frequent, approximately 10,000 times a day per cell due to oxidative damage alone, can produce epigenetic changes at the site of a DNA repair [45] [46].

The most significant damage, a double strand break in DNA, can promote DNA methylation or histone modification that most commonly silences the involved gene [47]. Enzymatic mediators of DNA repair can accumulate and promote chromatin remodeling at adjacent undamaged sites.

Subsequent changes may even affect expression of DNA repair genes and this might have a significant downstream effect, perhaps resulting in an escape from genetic homeostasis [48]. Inflammatory conditions that stimulate nociception can damage DNA and produce epigenetic change.

Free radical generation with the potential to damage DNA results from exposure to many environmental toxins as well as from constitutive processes such as aging [49] [50]. High levels of free radicals foster reaction with DNA. The product of this reaction is 8-hydroxy-2'-deoxyguanosine (8OHdG). This can be used as a biomarker for oxidative DNA damage. Levels of 8OHdG increase following

inflammatory processes [51].

2.2. Nociception

Nociception is the perception of noxious stimuli—stimuli with the potential to effect tissue damage. In a broader sense it is the perception of pain. As such the nociceptive system is a complex sensory neural network that communicates widely with motor, affective, cognitive and other sensory systems. Evidence, subsequently presented, supports the concept of nociceptive homeostatic control, as well as a robust intrinsic ability of the system to provide a plastic response in pathological states, and that this plasticity is realized through epigenetic mechanisms.

Stimulation of a peripheral nociceptor, generally a free nerve ending that reaches firing threshold in response to mechanical, thermal or chemical changes. In turn, the receptor initiates signal propagation along the peripheral nerve which synapses in the posterior horn of the spinal cord. The signal is modified in specific cord laminae by segmental and suprasegemental factors prior to proximal transmission, ultimately to processing centers in the thalamus and cortex [52]. Nociception can also trigger a variety of autonomic mediated responses (diaphoresis, nausea, tachycardia, hypertension and so on) [53].

The nociceptive system is constructed to provide a wide range of variable response, generally subserved through two afferent pathways and multiple efferent pathways or systems [54] [55]. These pathways correlate with specific identification in response to a painful stimulus. There are a wide variety of neurotransmitters involved in the afferent and efferent limbs of the nociceptive system [56].

In the periphery, nociceptors stimulate either fast, myelinated sensory fibers (A delta fibers) or slow, unmyelinated ones (C fibers). These fibers synapse with the second order neuron (of types that vary in electrical sensitivity, inhibiting or facilitating interneuron connections and firing thresholds) located in various laminae of the spinal cord (largely laminae II, IV, and V). At this point the signal enters the spinothalamic tract. Before reaching the brain the spinothalamic tract divides into the lateral neospinothalamic tract in the medial paleospinothalamic tract. Neospinothalamic fibers terminate at the synapse with ventrobasal thalamic neurons. Signals are then projected to the somatosensory cortex. The paleospinothalamic tract receives unmyelinated C fiber which generated signals that are projected to neurons in the reticular formation, thalamus, medulla, pons and periaqueductal gray matter and subsequently are transmitted to many areas of the brain for processing.

The efferent limb of the system incorporates an endogenous analgesia system that regulates nociception and pain responses [57]. Peripheral signal generation can be inhibited prior to central nervous system propagation, and signal transmission can also be inhibited or facilitated centrally (in the spinal cord, periaqueductal gray matter and thalamus) [58] [59].

Analgesia, or inhibition of nociceptive neurons, occurs when opioid receptors

are activated. There are multiple types of opioid receptors and a great deal of genetic, and possibly epigenetic, variability in their characteristics [60].

2.3. Epigenetics of Pain

The impact of epigenetic regulation of the pain system is apparent in the context of both homeostasis and plasticity. The previously described mechanisms, promoting DNA or histone methylation, histone modification and DNA damage, function in homeostatic pain modulation within the nociceptive system. This is particularly evident in cases of neuropathic or chronic pain where sensory neural activity continues in the absence of a peripheral stimulus [61] [62].

2.4. Epigenetics in the Afferent Nociceptive System

Inflammation, ischemia and nerve injury produce local tissue acidosis and are potent inducers of nociceptive activity, possibly via of the mediation of extracellular proteoglycans and G protein ligands [63] [64]. The byproducts of tissue damage (inflammatory cells, histamines, bradykinin, prostaglandins, ATP, nitrous oxide, cytokines and growth factors) electrically active a peripheral nociceptors. They produce local tissue acidosis, the magnitude of which correlates well with the degree of perceived pain [65] [66]. The production of free radicals, with their potential for DNA damage and gene silencing, is one example probabilistic epigenetic processing in the nociceptive system. Inflammatory products can also contribute to histone modification [67] [68] [69].

Inflammation and pain transduction are associated with the production of multiple peripheral factors such as nerve growth factor (NGF) and brain derived neurotrophic factor (BDNF) that signal transcription of ligands, particularly excitatory nociceptive peptides such as substance P, which is located within the cell body and within the posterior horn [70] [71]. Substance P also activates proton-sensing G-proteins (of these, GTPγS has been most studied) which in turn potentiate sodium channels (specifically Tetrodotoxin (TTX)-resistant voltage-gated Na (Na (V)) channels) in the paleospinothalamic nociceptive tract [72].

The G proteins however are also resultant in phosphorylative histone remodeling through several intermediaries [73] [74]. The reconfigured chromatin transcribes proteins which lower the threshold for ion-gated nociceptor activation [75]. This is an epigenetically produced bias that favors pain signal transmission. It is also a potential point of departure from a homeostatic to a plastic nociceptive process where changes in protein transcription result in an altered nociception phenotype. The resultant gene expression promotes a lasting decrease in activation threshold and increase in signal duration such as occurs in chronic, deafferented pain states [76].

The stress responsive protein deacytlases (SIRT) are encoded by SIRT genes and function in diverse molecular pathways including inflammatory and DNA repair pathways [77]. Sirtuin-6 (SIRT6) specifically has been identified as a me-

diator of inflammation-induced epigenetic change affecting both histone and DNA [78].

2.5. Epigenetic Mechanisms in the Efferent Limb of the Nociceptive System

DNA methylation can affect the opiate receptor. Endorphins are produced in response to painful stimuli [79]. These peptides bind opioid receptors in the brain (mu, kappa and delta), and dorsal horn of the spinal cord (mu and kappa receptors) to activate GABAergic, glutaminergic, cholinergic, substance P and serotonergic releasing neurons [80]. There is evidence for predetermined (developmental) epigenetic modification of opiate receptors through DNA methylation, and this modification may correlate with individual variations in pain perception, tolerance and related behaviours [81].

In the endogenous inhibitory arm of the nociceptive system, analgesic can be produced by activating the μ-opioid receptor (MOR), a member of the G-protein coupled receptor (GPCR) superfamily. Under homeostatic conditions, signaling is later terminated by intrinsic GTPase activity and a regulator of G protein signaling (RGS) protein [82] [83].

RGS proteins are GTPase-accelerating proteins (GAPs) and therefore reduce G protein mediated signal duration and intensity [84]. Loss of RGS activity results in enhanced endogenous opioid peptide signaling and inhibition of GABA release in periaqueductal gray matter. This introduces the potential of epigenetically mediated variability in the inhibitory arm of the nociceptive system.

DNA methylation can also affect the reelin protein. Reelin is a matrix protein that promotes glutamate receptor maturation [85] [86]. It is present in spinal cord nociceptive processing centers (laminae I, II and V pain) and involved in signal processing and opiate receptor sensitivity [87] [88]. DNA methylation of Reelin gene promoter sequence controls transcription of the protein, and changes in methylation result in altered transcription [89]. This is potentially another example of neuroplasticity and epigenetically directed change in the way that an afferent pain signal can be processed within the CNS [90].

2.6. Epigenetics and Descending Pathways

DNA methylation and resulting changes can affect the inhibitory limb of nociceptive pathways, most commonly through methylation of DNA transcribing neurotransmitters.

Dopamine is an inhibiting neurotransmitter that is present in the basal ganglia, spinal cord, thalamus and periaqueductal gray matter, Dysregulation of dopamine production is associated with chronic pain states and there is evidence that DNA methylation of promoter genes can lead to such dysregulation [91] [92].

There is also evidence of epigenetic control within the nociceptive system through DNA methylation of brain derived nerve growth factor (BDNF) [93].

This nerve factor and the related downstream proteins are essential for nerve cell viability and function. The BDNF-related gene has multiple promoter sites that are susceptible to chromatin alteration, most particularly DNA methylation [94] [95] [96]. Pain has the potential to downregulate BDNF DNA methylation and increase selective production of BDNF isoforms in the dorsal horn and dorsal root ganglion.

The glutamate receptor, NMDA (N methyl D aspartate), controls excitatory synaptic plasticity and memory through ion gated channel control. It is the primary agent of excitatory synapse plasticity within the nociceptive system in the brain and spinal cord [97] [98] [99]. DNA methylation of the promoter gene results in variation of the NMDA receptor expression and activity, favoring either the receptor subtype responsible for rapidly decaying or slowly decaying signals [100]. Receptor subunits have variable binding affinity's for the nociceptive neurotransmitter glutamine. There are further distinctions in the NMDA receptor, with subtypes of subunits, all of which contributes to a highly variability, or the ability to promote a differential response to an afferent pain signal [101] [102] [103] [104].

Epigenetic changes that result from nociceptive stimulation can also occur through the histone modification. Inflammatory conditions that increase IL-1, IL-6, IL-8, and metalloproteinases have demonstrated histone acetylation and phosphorylation [105] [106]. Such changes may impact NMDA receptors gene expression, altering subunits and the nociceptive bias in the brain and spinal cord, as noted above [107].

Histone modification can lead to stimulation of receptors in the posterior horn (brain derived neurotrophic factor (BDNF)/tropomyosin-related kinase receptor B (TrkB), glutamate/NMDA and substance P/NK-1) [108] and results in central sensitization, the ongoing pain response after removal of a painful stimulus.

Intracellular calcium will increase with activation of these receptors [109] and activation of calcium-dependent kinases that phosphorylate nuclear proteins. This is an illustration of the plastic response capability within the nociceptive system.

With the stimulation of C-fibers, activation of intranuclear factors occurs, which then promotes gene transcription V of the calcium-mediated cAMP responsive element binding protein (CREB) [110] [111]. Calcium mediated regulation occurs in the brain and spinal cord through acetylation of histone and phosphorylation of multiple transcription factors, changing chromatin and subsequent protein transcription.

Non-coding miRNA might be another post-translational epigenetic regulatory pathway within the nociceptive system. The mRNAs of inflammatory proteins can be exposed to miRNA modification [112]. With muscle inflammation, the levels of miRNA increase rapidly within a sensory ganglion [113] [114].

There is evidence from both animal and human studies that miRNA di-

rected epigenetic change plays a role in inflammatory pain [115] [116] [117] and that this change is upregulated by the inflammatory cytokines IL-1 and TNF-alpha [118] [119] [120]. Similarly, there is good evidence for miRNA mediated modulation of nociceptive pathways in chronic pain states, with differential signaling based on both nociceptive stimulus and the site of signal processing [121].

Another post-transcriptional epigenetic consideration in the nociceptive system that is a consequence of altered protein production via RNA alteration is the transformation of the protein Homer I to Homer IA [122].

Homer I is a structural protein that promotes pain transmission in the dorsal horn, binding glutamate receptors at the cell membrane. It is a constituent of the signaling complex that releases calcium from intracellular pools. Thus Homer I potentially initiates the downstream effects on ion gated channels in the nociceptive system [123] [124].

The native Homer gene transcription product is differentially deaminated (transforming adenosine into inosine [125] by adenosine deaminase to produce Homer IA, which results in different cellular functions. Homer IA lacks the ability to link with glutamate receptors and so its ligand activity inhibits, rather than facilitates, the transmission of a nociceptive signal.

With peripheral injury, Homer IA is produced in spinal neurons. This is an example of epigenetic homeostasis. If upregulation of the protein fails (failure to deaminate), pain transmission to proximal segments of the nociceptive system become more probable [126].

2.7. 5 Hydroxytryptamine (5-HT) Receptors

Another site where RNA deamination serves a nociceptive epigenetic function is at the serotonin (5-HT) receptor. The receptor receives suprasegmental stimulation. It is another G-protein coupled protein in the dorsal horn and inhibits signal transmission in ascending nociceptive fibers [127] [128] [129]. Alteration of the protein results in hypersensitivity to pain [130].

2.8. Steroids, Neurosteroids, Steroid Hormones, Secosteroids, Vitamin D and Testosterone

A steroid is an organic compound that contains four joined cycloalkane rings. A secosteroid is a steroid with a failure of bonding of B-ring carbon atoms, leaving one "broken ring". Neurosteroids are bioactive steroids that are transcribed in neurons and glial cells in both the central and peripheral nervous systems. The role that they play in nociception is an increasing focus of investigation related to Pain Medicine [131] [132] [133] [134] [135].

The neuroprotective effects of pregnenolone (**Figure 3**) and role of progesterone in neurogenesis within the nociceptive system have been well described [136] [137] [138]. The effect of dehydroepiandrosterone (DHEA) on peripheral and central receptors has also been well described [139]. This 5-alpha

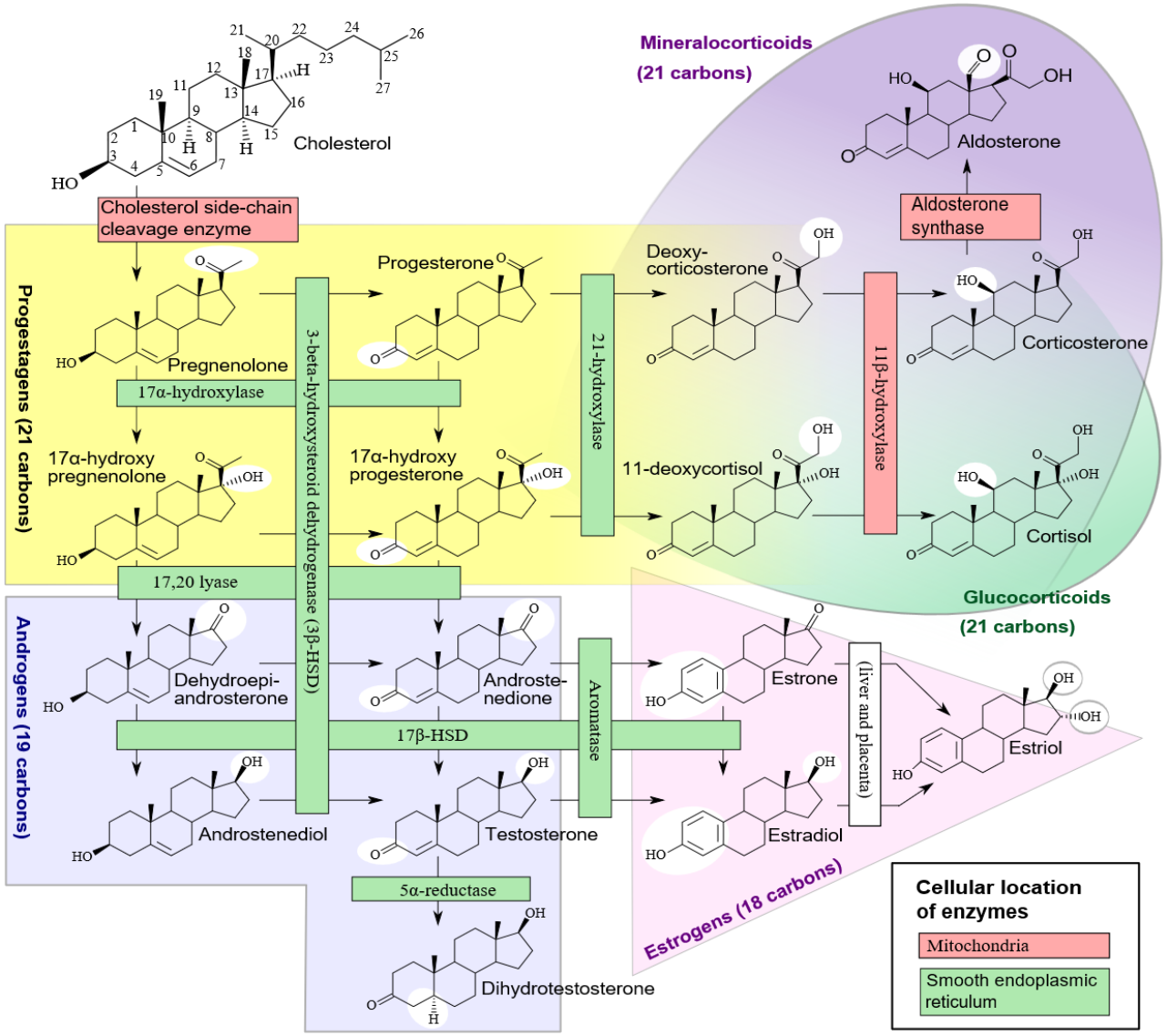

Figure 3. Steroidogenesis (https://en.wikiversity.org/wiki/WikiJournal_of_Medicine/Diagram_of_the_pathways_of_human_steroidogenesis) (https://doi.org/10.15347/wjm/2014.005).

reduced neurosteroid is produced in the neuron. At the present time it is apparent from the mammalian models that inflammatory pain upregulates GABA mediated inhibition of nociceptive transmission by signaling an increase in endogenous neurosteroid production [140].

Neurosteroid production within the nociceptive system can be either typical or nongenomic (epigenetic). These steroids rapidly modulate neurons within the nociceptive pathway through ionotropic (specific and nonspecific cation channel) receptors and by coupled G-protein, metabotropic neurosteroid receptors in peripheral nociceptor cell membrane [141] [142] [143] [144].

The gonadal and secosteroid hormones vitamin D and testosterone, most particularly vitamin D3, have wide ranging implications in health and disease [145].

Their function in multiple body systems is increasingly well understood. There is evidence from both clinical and foundational studies that they may also play a role as "pseudo-neurosteroids" within the nociceptive system, affecting transduction, transmission and modulation of pain signaling, either directly or through the regulation of calcium binding proteins [146] [147].

Steroid hormones, including vitamin D and testosterone, are cholesterol derivatives. The genes/enzymes responsible for regulation of the vitamin D hormone include several cytochrome P450 related enzymes, specifically CYP2R1, CYP27B1 and vitamin D 25 hydroxylase and hydroxyl vitamin D3 1-alpha-hydroxylase [148] [149] [150]. In the case of testosterone, regulatory genes/enzymes include CYP11A (mitochondrial cytochrome P450 oxidase), CYP 17A (a related oxidase in the endoplasmic reticulum), 3-beta hydroxysteroid dehydrogenase and 17-beta hydroxysteroid dehydrogenase [151].

3. Vitamin D

Vitamin D3 is a prohormone produced in skin through ultraviolet irradiation of 7-dehydrocholesterol. It is biologically inert and must be metabolized to 25-hydroxy vitamin D3 in the liver and then to 1*α*, 25-hydroxyvitamin D3 in the kidney before it becomes active [152] (**Figure 4**).

The hormonal form of vitamin D3, *i.e.*, 1*α*, 25-hydroxyvitamin D3, acts through a nuclear receptor to carry out its many functions, including calcium absorption, phosphate absorption in the intestine, calcium mobilization in bone, and calcium reabsorption in the kidney [153] [154]. It also has several noncalcemic functions in the body.

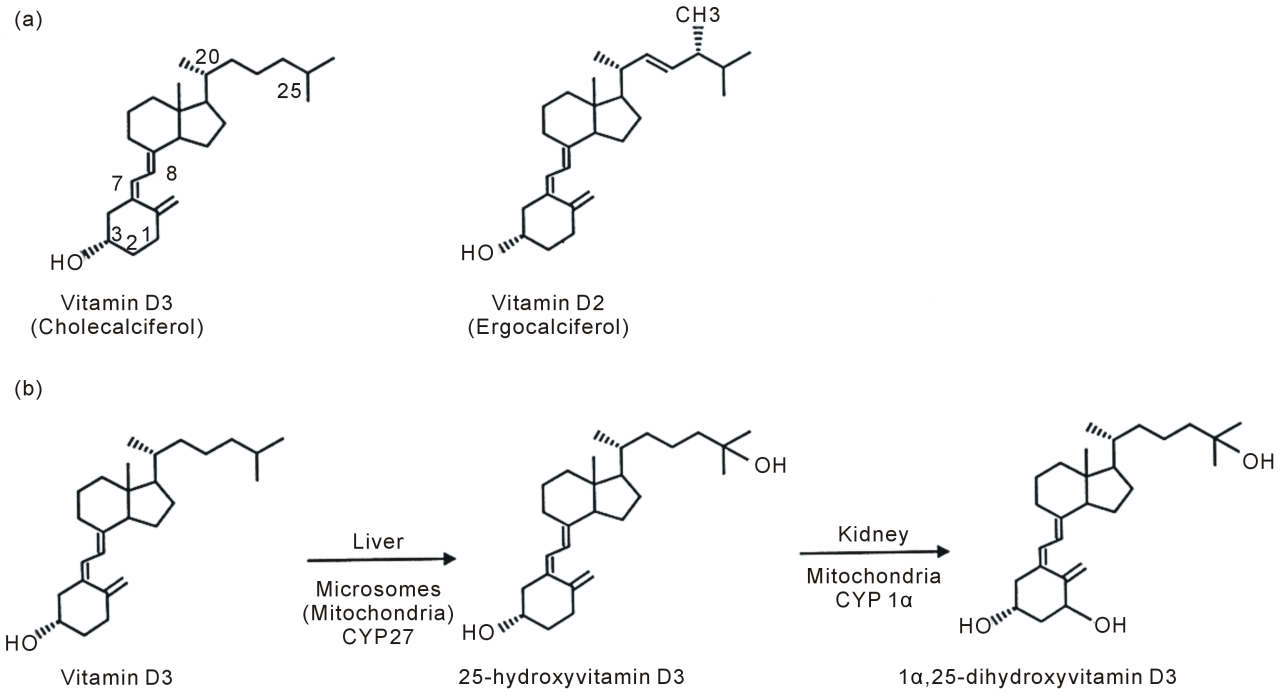

Figure 4. (From http://physrev.physiology.org/content/78/4/1193.figures-only).

The critical role of vitamin D in human physiologic systems is increasingly recognized as a key determinant in the homeostasis involved in health maintenance. This secosteroid has been identified as a key element in the normal development and operation of wide ranging metabolic pathways that impact, among others, the musculoskeletal and neurologic systems.

Vitamin D exists in several forms. The two predominant forms are vitamin D2 (ergocalciferol), and vitamin D3 (cholecalciferol) (**Figure 4**), known together as calciferol. All forms are fat-soluble lipids derived from cholesterol that function as hormones [155] [156]. The forms of vitamin D are secosteroids (steroids in which one of the bonds in the steroid rings is broken) [157]. The difference in structure and function between vitamin D forms relates to their respective side chains.

Vitamin D3 and vitamin D2 can be ingested. Vitamin D3 can also be synthesized from cholesterol by ultraviolet irradiated skin. In the epidermal basal and spinosum strata large quantities of vitamin D3, 7-Dehydrocholesterol can be produced rapidly by optimal ultraviolet light exposure, at wavelengths betweem 295 and 300 nm [158].

Activation of calciferol occurs through two separate hydroxylation stages. The first, conversion to calcidiol (25-hydroxyvitamin D3) occurs in the liver and the second, conversion of 25-hydroxyvitamin D3 to the active hormone calcitriol (1,25 dihydroxyvitamin D3 or 1,25$(OH)_2$D3) occurs in the kidney. These conversions are mediated by several cytochrome P450-related enzymes including CYP2R1 and CYP27B1 [159] [160].

Ingested or endogenously produced, cholecalciferol is hydroxylated in the liver at position 25 to form calcidiol by hepatocyte produced vitamin D 25 hydroxylase. There are two enzymes thought to be involved in the 25-hydroxylation step. They are most active in the liver but can be seen elsewhere in the body [161].

In the proximal renal tubules calcidiol is hydroxylated by 25-hydroxyvitamin D3 1-alpha-hydroxylase at the 1-α position to perform calcitriol.

In addition to renal hydroxylation, calcitriol is synthesized in the immune system by monocytes. Dihydroxyvitamin D3 in this instance acts not as a hormone, but rather as a cytokine, stimulating the innate immune system [162] [163].

Once made, the product is released into the plasma, where it is bound to α-globulin, vitamin D binding protein. This protein transports calcitriol to its target organs. Calcitriol circulates as a hormone, regulating the serum calcium and phosphate concentration and related growth and remodeling of bone as well as more diverse neuromuscular and immune functions [164].

Once at its target, calcitriol binds with the vitamin D receptor (VDR) in the cell nucleus. The VDR C-domain which is a DNA-binding domain, a calcitriol-binding domain called the E-domain, as well as an F-domain, which is one of the activating domains. When calcitriol activates this nuclear pathway, the

VDR/ligand collectively determines the specific transcriptional response and downstream physiologic effect [165].

Calcitriol can also activate calcium channels in the plasma membrane, in a signal transduction pathway [166].

The VDR modulates the gene expression of calcium and phosphorus transport proteins (including calbindin) [167]. The VDR is a cell nuclear receptor that can facilitate either the activation or suppression of its target genes [168]. The receptor acts via vitamin D-responsive elements (VDREs) which are located near the target gene. When the VDR binds with calcitriol, the receptor changes its confirmation. It concomitantly binds multiple protein coactivators when facilitating active transcription. In this instance the stereotactic change allows phosporylation of serine-205 and transcription is either suppressed or intiated, depending on the gene [169] [170]. There are multiple coactivators which might provide a highly variable dynamic phenotypic and transcriptional response in many different body systems [171].

This wide array of dynamic response is probably intimately related to calcitriol's role in both health and disease. It allows for homeostasis in multiple systems and escape from this homeostasis may have notable genomic plastic effects such as tumorgenesis[172] [173] [174].

Both an excess and a deficiency in vitamin D appear to cause abnormal functioning and premature aging [175] [176] [177]. The relationship between serum calcidiol level and all-cause mortality is parabolic [178].

Through activation of the VDR, calcitriol activates osteoblasts to secrete receptor activator nuclear factor-$\kappa\beta$ ligand (RANKL) [179] [180]. Osteoclastogenesis and bone resorption are activated by RANKL. Activated VDRs promote bone mineralization but also stimulate osteoclastic elevation of serum calcium and phosphorus concentrations. They regulate parathyroid hormone production and affect the function of pancreatic islet cells [181] [182] [183] [184]. VDRs are also present in monocytes and activated T and B cells and are important in immune system function [185] [186].

VDRs regulate expression of tyrosine hydroxylation via gene activation in the adrenal medullary cells. They are also involved in the biosynthesis of nitric oxide synthase [187] [188]. It is reasonable to thus propose a role for vitamin D receptors participating in, and possibly linking, the physiologic responses to both stress and pain.

3.1. Vitamin D Hormonal Homeostasis

The synthesis of cholecalciferol is generally adequate to maintain serum concentrations and toxicity is prevented through a negative feedback loop.

Cytochrome enzymes are agents of vitamin D homeostasis, as well as participants in the highly differential response of nuclear targets. They possibly provide a window through which a link between vitamin D/steroid hormone and nociceptive function can be seen under both homeostatic and plastic conditions.

1,25-dihydroxyvitamin D_3 is degraded by mitochondrial enzymes in the target cells CYP24A1 facilitates a series of catabolic steps that begins with 24 hydroxylation of $1,25(OH)_2D_3$ and ends with the production of calcitroic acid. Feedback regulation of both production in the kidney and degradation at peripheral targets maintains homeostasis of the active metabolite ($1,25(OH)_2D_3$).

Transmembrane serum calcium-sensing proteins in the parathyroid gland bind to G proteins when calcium concentrations fall. This stimulates the release of parathyroid hormone (PTH). Parathyroid hormone then stimulates proximal convoluted tubule cells and osteoblasts. Notably, PTH elevates 1α-hydroxylase concentrations in the convoluted tubule cells. This favors hydroxylation of calcidiol to calcitriol [189] [190].

Subsequently vitamin D signals cells to promote kidney reabsorption of calcium. When blood calcium levels exceed the threshold of the system, either through mobilization from bone, gastrointestinal absorption or kidney reabsorption, the cascade of events indcuced by the parathyroid gland will be inhibited. When serum calcium concentrations are excessively elevated, thyroid glands C-cells secrete calcitonin, which blocks bone calcium mobilization. CYP27B1 is also down-regulated by calcitriol itself, which then negatively signals CYP27B1 transcription, thus curbing vitamin D synthesis [191].

Calcitonin also stimulates the renal 1α-hydroxylation that provides vitamin D hormone for non-calcemic needs under normal calcium conditions [192]. Such non-calcemic needs are just beginning to be identified, but the potential impact of vitamin D, the VDR and VDREs are likely to prove to be immense.

One example of this is vitamin D hormone induction of 24-hydroxylase (CYP24). CYP24 is not only involved in the control of vitamin D production itself. It is also involved in the processing of nociceptive signals in the DRG (dorsal root ganglion), as well as testosterone production in the Leydig cells [193].

3.2. Epigenetics of Vitamin D

Epigenetic mechanisms are intrinsic to vitamin D signaling pathways. The effects are mediated both upstream at the activating or inactivating gene/enzymes CYP27A1, CYP27B1 or CYP24 as well as at the VDR [194]. Activity of VDRs can be modulated epigenetically by histone acetylation [195].

Calcitriol is an active participant in predetermined epigenetics. The effect of vitamin D on fetal programming and gene regulation are reflected in the normal operation of many physiologic systems throughout life. Another nongenomic, probabilistic, way in which vitamin D might enhance the ability of human systems to respond to stimuli likely exists.

A signal transduction mechanism that is VDR/VDRE dependent may operate in either the typical gene transcription pathway or through a signal transduction pathway that involves calcium channels located on the plasma membrane [196] [197] [198]. There is evidence that the nuclear VDR/VDREs ligand mediating genomic effects is different from the membrane VDR/VDREs that initiate the

nongenomic transduction pathway effects [199] [200] [201].

Vitamin D also has a more fundamental role in the epigenetic regulation-plastic or homeostatic physiological systems. Epigenetic regulation by calcitriol stilumates the expression of the JMJD3 gene which codes for a histone demethylase [201]. To date there have been relatively few reliably documented instances where this mechanism is active. Nonetheless vitamin D signaling might have a multisystem applicability through its regulatory action on the expression of genes coding for histone demethylases of the Jumonji C (JmjC) domain and lysine-specific demethylase (LSD) families, and subsequently on gene transcription and cell phenotype [202] [203].

3.3. Testosterone

Testosterone is a steroid hormone, and androgen rather than a secosteroid. It is synthesized by the Leydig cells in the testes, to a lesser extent in the ovaries, and minimally in the zona reticularis of the adrenal cortex and skin [204]. In the serum it is transported to target tissues by the sex-hormone-binding-globulin protein.

In men, testosterone is a determining epigenetic factor for reproductive tissue and secondary sexual characteristic development. It promotes increased muscle and bone mass through increased protein synthesis in these testosterone receptor-dense tissues [205].

The serum concentration of testosterone is 7 - 8 times greater in the adult male than in the adult female. Testosterone sensitivity is greater in the female. The daily production and consumption of testosterone is greater in the male.

The anabolic effects of testosterone include augmentation of muscle mass, bone density, linear growth, bone maturation and sex organ maturation [206] [207]. To do this it likely interacts at a molecular level with the vitamin D hormone system of signaling as previously described.

A developmental epigenetic effect of the hormone first occurs with gender formation during the second trimester, with feminization or masculinization of the fetus. Animal studies point to aromatase (an enzyme that converts testosterone into estradiol) as being responsible for male sexual differentiation of the cerebrum [208]. Serum levels of testosterone during this key developmental period are a better predictor of sex typed behavior than adult levels [209] [210].

Testosterone is a physiologic regulator of the hypothalamic-pituitary-adrenal axis and plays a role in both cognition and general health/physical energy [211]. It determines the density of thromboxane A2 receptors on platelets and megakaryocytes responsible for platelet aggregation [212] [213]. Testosterone levels decline gradually with age.

In addition to its well described positive trophic effects on skeletal and cardiac muscle and other organs, there is evidence that testosterone affects attention, memory and spatial orientation [214] [215] [216] [217].

A non-linear relationship between testosterone and its physiologic functions

is supported by the literature. There is a curvilinear relationship which exists between spatial performance and circulating testosterone. Both deficiency and excess of circulating hormone have a negative effect on cognition [218].

3.4. Testosterone Homeostasis

The amount of testosterone synthesized is regulated by the hypothalamic-pituitary-gonadal axis (**Figure 5**) through gonadotropin-releasing hormone produced in the hypothalamus. This stimulates the release of luteinizing and follicle stimulating hormones from the pituitary. These hormones stimulate the production of testosterone. Testosterone acts through a negative feedback loop on the hypothalamus and pituitary to inhibit the release of stimulatory hormones [219].

There are multiple extrinsic factors that can affect testosterone production. These include weight loss (increased synthesis as fat cell production of aromatase increases testosterone conversion into estradiol, lowering baseline serum testosterone levels and this signaling increased testosterone production), zinc deficiency (decreased synthesis), sleep (production increases during REM sleep), exercise and protein ingestion (increased production). Notably, there is a positive correlation between vitamin D and testosterone levels [220] [221] [222].

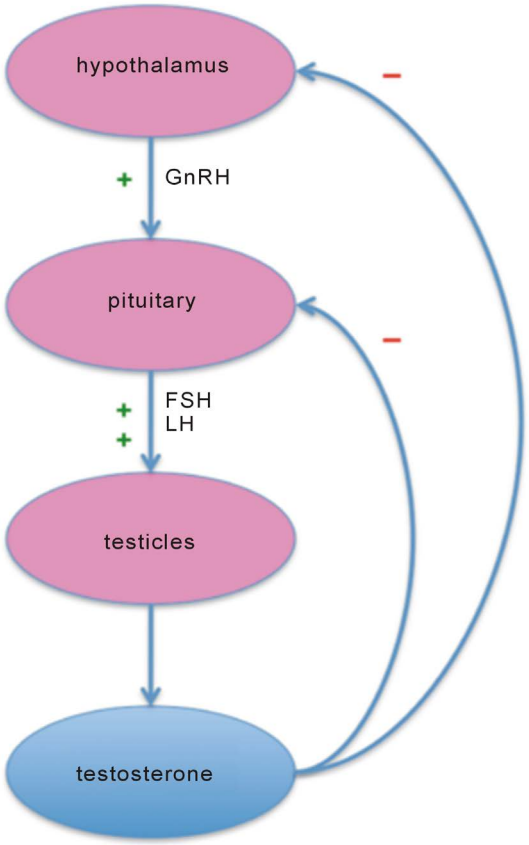

Figure 5. Hypothalamic-pituitary-testicular axis (http://www.wikilectures.eu/w/Hypothalamic-pituitary-gonadal_axis).

Free Testosterone is transported into the target cell cytoplasm where both the hormone and its reduced derivative 5-alpha dihydrotestosterone (via 5-alpha reductase) bind to the androgen receptor. This complex then enters the cell nucleus and binds nucleotide sequences of chromosomal DNA [223]. As with vitamin D, the mechanism involves hormone activity elements which can impact transcriptional activity thereby causing androgen effects.

3.5. Testosterone and Vitamin D

There is an association with vitamin D and testosterone levels. Some of the evidence is conflicting regarding vitamin D supplementation as a means to increase the production of testosterone in men [224] [225] [226]. It is unclear whether the response of testosterone to D3 supplementation is dependent upon serum levels of vitamin D. The association between calcitriol and total and free testosterone is linear at typical calcitriol concentrations and achieves a plateau at higher levels [227].

3.6. Testosterone and Epigenetics

Beyond its clear role in predetermined epigenetics, testosterone likely also plays a part in probabilistic epigenetic mechanics. The small amounts of testosterone produced in the adrenal glands may have implications for the nociceptive system.

Testosterone produced by the adrenal cortex participates with aldosterone and cortisol in the modulation of the stress response [228]. Testosterone and its estradiol derivative can also be considered as secondary neurosteroids that impact nociception [229] [230]. Adrenal testosterone and its derivatives are produced in the Zona Reticularis, the inner most cortical layer, along with androstenediones and the estrogen-precursor, dehydroepiandrosterone (DHEA) and its derivatives. There is evidence that stress related testosterone production is epigenetically modifiable through gene targets of several miRNAs [231]. It is reasonable to consider pain as a stressor that alters testosterone production that in turn modifies nociceptive processing. This thought is consistent with evidence that negatively correlate pain perception and certain chronic pain states with testosterone levels [232] [233] [234].

Testosterone mediated epigenetic change in the nervous system is well documented in motor pathways [235] [236] [237]. Its role in regulating the coupling mechanisms between calcium channels Ca (v2.2) and transmitter release at neuromuscular junctions has been demonstrated [238]. It seems that testosterone is one of the steroid hormones which produce both genomic and non-genomic effects in not just motor, but also possibly sensory, nervous system signaling [239] [240]. The non-genomic effects may operate through modulation of the plasma membrane proteins, including voltage- and ligand-operated ion channels or G-protein-coupled receptors [241] [242]. Seventeen beta-estradiol, testosterone, pregnenolone sulfate and dehyroepiandrosterone sulfate increase the hydrolytic

activity of plasma membrane Ca^{2+}-ATPase in a dose dependent manner. They decrease the stimulatory effect of the calcium pump activator, calmodulin [243].

The activity of both calcium channels and calmodulin has been amply demonstrated in the nociceptive system [244] [245]. This represents a possible epigenetic interface between testosterone and nociception.

3.7. Participating Gene/Enzymes, Signaling Proteins and Receptors

There are several genes/enzymes, signaling proteins and receptors that play a prominent role at the interface between vitamin D and testosterone hormone signaling, epigenetic mechanisms and the nociceptive systems. These elements are located within the nociceptive framework—sometimes discretely (such as in the brainstem, dorsal laminae or root ganglia) and at times diffusely. Included in this list are the emzymes Jumonji domain-containing 3 (Jmjd3) histone demethylase, CYP24A1 (25-hydroxyvitamin D_3-24-hydroxyalse), CYP17A1 (steroid 17-alpha-monooxygenase, or 17α-hydroxylase/17, 20 lyase/17,20 desmolase), and CYP27B1 (25-hydroxyvitamin D_3 1-alpha hydroxylase).

Proteins, such as calbindin-D28K, multiple bone morphogenetic proteins (BMPs) and the Homer/Homer 1 protein, have identifiable functions that intersect domains, as do various receptors such as the VDR receptor, the chemokine CC motif receptor 2 (CXCR2) and GABA, glutamate and G-protein receptors.

3.8. Jumonji Domain-Containing 3 (Jmjd3)

Jumonji domain-containing 3 (Jmjd3) is a histone demethylase present in the cell nucleus that specifically catalyzes the removal of trimethylation of histone H3 at lysine 27 (H3K27me3) [246]. Its role in cell differentiation and predetermined epigenetics has been well described. For example, it regulates the expressions of Bone sialoprotein BSP and Osteocalcin OCN via transcription factors Runx2 and Osterix and drives osteoblast differentiation [247]. It also is a notable epigenetic agent within the nervous system [248] [249]. Speculatively, it might prove to link such diverse processes as inflammation, nociception and cell differentiation [250].

Vitamin D induces the expression of the Jmjd3 gene via VDR signaling [251] [252]. There is also transcriptional regulation of Jmjd3 from the cell cytoplasm via the signal transducers and activators of transcription (STAT) proteins STAT1 and STAT3, and this is a crucial link between a painful stimulus and nociceptive response, in this case by inducing inflammation in microglia [253] [254]. Cytotoxic and inflammatory factors (particularly cytokines) are associated with microglial activation and signaling within the nociceptive system [255] [256].

The cytokines activate STAT proteins by binding with Janus kinase. This enzyme then phosphorylates the STAT protein which is subsequently transported into the cell nucleus. The STAT protein binds to the target gene and activates

transcription [257] [258]. In the instance of pain, it is reasonable to assume that this would include multiple activating and inhibitory or silencing nociceptive factors. Such relevance of STAT proteins, and Jmjd3, to the nociceptive system has been recently recognized [259].

Further evidence suggests that Jmjd3 may also serve an epigenetic regulatory function within the nociceptive system in non-inflammatory pain states, such as chronic bladder pain/interstitial cystitis, although the evidence for this is less compelling [260].

3.9. CYP24A1, 25-Hydroxyvitamin D_3-24-Hydroxyalse

CYP24A1, 25-hydroxyvitamin D_3-24-hydroxylase, is a mitochondrial enzyme that degrades the hormonal form of the vitamin (calcitriol). A homeostatic balance exists wherein calcitriol rapidly induces CYP24A1 expression and is thus the primary regulator of CYP24A1.

CYP24A1 expression is up-regulated by 1,25-dihydroxyvitamin D(3) via a vitamin D receptor (VDR)/retinoid X receptor (RXR) heterodimer that binds to two vitamin D response elements (VDREs) located near the proximal promoter. Along with calcitriol, co-regulators are responsible for an increase in RNA polymerase II and histone H4 acetylation, further enhacing the CYP24A1 up-regulation [261] [262].

The enzyme's primary function is to limit the extent and duration of vitamin D responsive target transcription by affecting the hormone's circulating levels in both normal physiological or pathological states, specific to an individual cell type/tissue [263].

The baseline level of CYP24A1 expression is determined in a cell type-specific manner by various factors including glucocorticoids, estrogens, testosterone, retinoid ligands and local growth factors [264] [265] [266].

3.10. CYP17A1

CYP17A1 is found in the zona reticularis of the adrenal cortex. It catalyzes synthesis of certain lipids, including cholesterol and its neurosteroid derivatives [267]. It has both 17alpha-hydroxyalse and 17,20-lyase activities, and is a key enzyme for the downstream production of neurosteroids, including testosterone [268] [269] [270].

Single nucleotide polymorphisms (SNPs) in CYP17A1 and VDR genes appear to be significantly associated with arthralgia. Interactions between CYP27B1 and both CYP17A1 and VDR SNPs may produce an additive effect on pain intensity. [271].

CYP27B1 and CYP24 expression in unmyelinated sensory neurons controls vitamin D metabolite concentrations [272]. Unmyelinated CGRP-positive neurons seem to have a separate and distinct vitamin D phenotype with hormonally-regulated ligand and receptor levels [273].

Within a neural cell population that is mainly nociceptive, these findings sug-

gest that vitamin D signaling may play a specialized role. The implication is, through nuclear or extranuclear signaling pathways, calcitriol may affect sensory neurons thereby affecting sensory processes including pain and proprioception.

3.11. Calcium Binding Proteins

Among the various calcium binding proteins, Calbindin D28K and Calretinin are present in sensorineural pathways [274]. Calbindin-D28K functions in the intestine, kidney and neuroendocrine cells. Animal studies have demonstrated its activity in laminae I and II of the dorsal horn and in spinal ganglia, in a distribution similar to that of substance P-containing primary afferent neurons [275].

This would suggest that calbindin has a regulatory role in pain transmission, likely through its binding of free calcium [276] [277]. Animal studies demonstrate that a lack of calbindin D28K is associated with deficient GABAergic neurotransmission and dysfunctional sensory processing of nociceptive stimuli [278]. This also supports the idea that the protein plays an active role in nociceptive sensory transmission.

Calbindin is encoded in humans by the CALB1 gene [279]. The protein is transcribed by this VDR regulated gene, although this vitamin D dependence may have exceptions, as perhaps in its synthesis in the brain [280] [281].

Calbindin thus provides a link between vitamin D and nociceptive systems. It might also provide a link to other steroid hormones such as testosterone. There is evidence that androgen hormones facilitate renal calcium transport which leads to a compensatory decrease in Calbindin-D28K expression [282]. Admittedly speculative, an indirect pathway for testosterone directed, homeostatic, regulation of Calbindin-D28K expression might exist.

Calbindin may play a significant role in central processing of nociceptive signals [283] [284]. The evidence for this being a Vitamin D dependent process is not clear.

Animal studies indicate that chronic deafferentation of skin and peripheral tissues that induce central pain states are associated with plasticity of nociceptive pathways. More specifically, central pain states are associated with an increase in activity of Calbindin cells at spinal, brainstem, and thalamic levels, specifically Calbindin-D28K regulation of gamma-aminobutyric acid type A receptors [285].

3.12. Bone Morphogenetic Protein-2 (BMP2)

Bone morphogenetic protein-2 (BMP2) is an important component of multiple signaling pathways. These pathways include the hedgehog pathway (note participation of the protein in predetermined epigenetics in this instance) and the transforming growth factor beta (TGFB) pathway [286].

BMP2 stimulates osteoblastic differentiation in concert with vitamin D (activated VDRs) and is itself epigenetically down-regulated by vitamin D through calcitriol induced transcriptional repression by DNA methylation and histone

modification [287].

BMPs, possibly including BMP2, might also play a signaling role in the developing nociceptive system [288]. It is unclear at this time whether BMP2 might act in adult neurosensory modulation, but seems to participate indirectly in the expression of ligands that direct sensory neuronal differentiation [289].

3.13. Interleukin-8 Receptor, Beta, or Chemokine CC Motif Receptor 2

Interleukin-8 receptor, beta, or chemokine cc motif receptor 2, or CXCR2 is a member of the G protein-coupled receptors. The ligand is interleukin-8, which it binds with high affinity, and transduces the signal through a G-protein activated second messenger system (G-coupled) [290] [291].

CXCR2 is a pro-inflammatory receptor that is active in processing (induction and sensitization) both neuropathic and inflammatory pain [292] [293] [294]. It is epigenetically regulated by histone modification as evidenced in animal studies [295], although its regulation is poorly understood and likely complex. In other systems where they coexist, VDR/vitamin D elements down-regulate CXCR2 expression [296].

3.14. Structural Proteins and the Metabotropic Glutamate Receptors

The neuronal protein Homer1 is encoded by the HOMER1 gene. It is a post-synaptic scaffold protein that is widely expressed in the central nervous system as well as in peripheral tissues, the list of which notably includes the kidney and ovary [297] [298].

The expression of Homer1 is induced by neuronal activity. Thus Homer1 expression increases after periods of activity or stress, injury, or other challenge. It binds several targets that affect both indirectly via G proteins (metabotropic glutamate receptor) and directly via the inositol triphosphate, IP_3Rs receptor [299] [300].

Homer1 is associated with the Shank scaffold protein and there is evidence that the interaction between the 2 scaffold proteins may be epigenetically modifiable and thus alter the response of the system [301] [302].

The Metabotropic glutamate receptor 5 regulates neuronal excitability in the spinal dorsal horn, promoting nociceptive transmission. Thus metabotropic glutamate (mGluR) receptors play important roles in the modulation of nociception [303].

Pain transmission is inhibited by stimulation of metabotropic glutamate 2 (mGlu2) receptors in the posterior horn of the spinal cord [304]. MGluRs can also be seen in pain regulatory centers of the brainstem, forebrain and in peripheral nociceptors [305] [306] [307]. mGlu2 receptor regulation occurs via the acetylation-promoted activation of the p65/RelA transcription factor [308] [309].

Homer1 binds receptors with the net effect of uncoupling glutamate receptors

from the excitatory pathway [310]. It also contracts the dendritic spine. It allows the CNS to self-down-regulate under conditions of excessive stimulation (CNS homeostatic plasticity) [311] [312] [313] [314].

Homer proteins also influence the function of their binding partners. Binding to Homer1 alters the function of cation non-sepcific (TRPC1) and calcium specific (IP3R) channels [315]. In this way, Homer proteins regulate localization of binding partners and affect neuronal signaling. While this mechanism is best described in the brain, the constituent elements of this hemostatic/plastic mechanism are present in the nociceptive system as well.

In short, metabotropic glutamate receptors (mGluRs) and Homer proteins play critical roles in neuronal functions including plasticity and nociception. Homer proteins regulate mGluR function by facilitating coupling to effectors such as the inositol triphosphate receptor as noted above. This modulation occurs postsynaptically. Thus, alteration of mGluR signaling by changes in Homer protein expression may confer sensitivity to the neuronal response to stimulation, such as a nociceptive stimulus.

The postsynaptic structural effect of the protein on the dendritic spine contains the rise in calcium ions released from intracellular stores, damping excessive stimulation [316] [317]. Along with the change in dendritic spine confirmation, the protein scaffold (including Homer1) undergoes translocation along with the intracellular Ca^{2+} channel inositol triphosphate receptor (IP3R) and endoplasmic reticulum (ER) proteins that are vitamin D dependent (Calreticulin and Calbindin) [318]. The induced change might have consequences for local Ca^{2+} homeostasis as well as overall neuronal signaling.

3.15. Inhibitory Neurosignaling by Gamma-Aminobutyric Acid (GABA) and G Proteins

Ubiquitous in central nervous system, GABA (**Figure 6**) regulates neuronal excitability. This includes nociceptive pathways [319]. GABA acts pre-and post synaptically. Its direct action occurs by binding to an ionotropic (direct) receptor-the GABA A receptor (a ligand-gated chloride ion channel receptor). It also acts indirectly through a metabotropic receptor—the GABA B receptor [320]. In this instance it regulates intermediary, coupled G proteins which open or close ion channels including calcium channels.

Voltage gated calcium channels modulate the function of peripheral and central pain pathways by influencing fast synaptic transmission and neuronal excitability [321]. Both the various subtypes of high voltage activated type Ca^{2+} channels and the low voltage activated or transient (T)-type Ca^{2+} channels (T-channels) participate in the modulation of nociception [322].

G proteins may also play an additional role in the processing of pain signals by modulating endogenous opioid supraspinal antinociception [323] [324]. Animal studies of thermal induced nociception suggest that they also function at a spinal level as regulators of opioid antinociceptive pathways [325].

Within DRG neurons, G protein coupled proton-sensing receptors have been

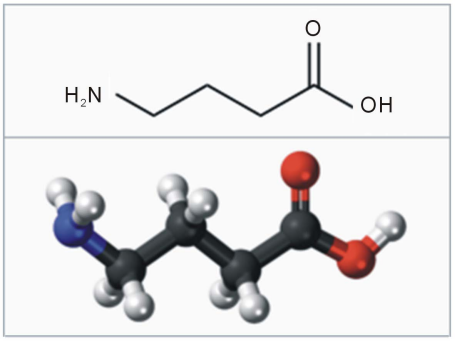

Figure 6. γ-Aminobutyric acid or GABA (https://commons.wikimedia.org/wiki/File:Gamma-Aminobutters%C3%A4ure_-_gamma-aminobutyric_acid.svg) (https://commons.wikimedia.org/wiki/File:GABA_3D_ball.png).

identified [326]. Such GABA-G-protein coupled receptors might have a role in epigenetic regulation of pain related to insults that produce local tissue acidosis, such as is seen with mechanical trauma, inflammation and ischemia.

Neurons that produce GABA as their output (GABAergic neurons) have mainly inhibitory action [327]. Loading of GABA and glycine into synaptic vesicles via the vesicular GABA transporter (VGAT) is an essential step in inhibitory neurotransmission [328] [329].

Alteration of GABAergic and/or glycinergic neurotransmission is linked to sensory processing in various pain disorders [330]. GABA, once released from vesicles, rapidly activates GABA type A receptors. This gives rise to an inhibitory, small and brief postsynaptic current, the phasic response [331]. Neurosteroids released from close proximity neurons or glia prolong the decay of this current. This enhances synaptic inhibition.

Neurons may also contain extrasynaptic receptors that are activated by low levels of ambient GABA, producing a tonic response—a background current that might influence neuronal firing [332]. Neurosteroids can also enhance this response, even at concentrations that have little effect on the phasic response. 3α5α-THPROG, 5α-pregnan-3α-ol-20-one; 5α-DHPROG, 5α-dihydroprogesterone have demonstrated this effect in animal studies [333].

Epigenetic modulation, through the phosphorylation of protein, can affect the sensitivity of GABA (A)-receptors to neurosteroids [334] [335].

4. Neurosteroids and Gaba Mediated Nociception

Neurosteroids represent a class of endogenous steroids that are synthesized in the brain, the adrenals and the gonads. They have potent and selective effects on the GABA A-receptor [336].

Neurosteroids act at an interface of homeostatic and epigenetic modulation of the nociceptive system and might be influenced through vitamin D dependent systems. In a non-genomic manner, reduced metabolites of testosterone, deoxycorticosterone and progesterone are positive modulators of GABA A-receptor.

5α-androstane-3α, 3α5α-tetrahydrodeoxycorticosterone (3α5α-THDOC), Al-

lopregnanolone (3α-OH-5α-pregnan-20-one) and 17α-diol(Adiol) augment the GABA-mediated Cl(-) currents acting on a site(s) through a pathway which is currently unidentified. This does not involve the GABA receptor [337] therefore further enhancing inhibition.

Other neurosteroids may decrease inhibitory neurosignaling. For example, acting as GABA A-receptor antagonists are 3β-OH pregnane steroids and pregnenolone sulfate(PS). This process, already identified in areas of the CNS which subserve cognition, memory and mood, almost certainly is altered in chronic pain states, and might be active in nociceptive anatomic substrates as well. This most likely would occur for inhibitory synaptic transmission in lamina II of the spinal cord.

Endogenous 5 alpha-reduced neurosteroids are produced locally in lamina II and modulate GABA A-receptor function during inflammatory pain [338]. The production of 5 alpha-reduced neurosteroids is controlled by the endogenous activation of the peripheral benzodiazepine receptor (PBR), which initiates the first step of neurosteroidogenesis by stimulating the translocation of cholesterol across the inner mitochondrial membrane.

Stimulation of the PBR prolongs GABAergic signaling, at least during inflammatory pain [339], perhaps indicating that neurosteroidogenesis plays a regulatory role in nociception at the spinal cord level.

The amount of inhibition caused by GABA A-receptors can be varied by tetrahydro-deoxycorticosterone, endogenous neurosteroids, and allopregnanolone.

5. Clinical Considerations

The above information, largely abstracted from animal studies, is of unclear meaning in a clinical context. The heterogeneity of studies in design, intent and quality limit the ability to accurately assess them for significance and size effect.

Epidemiological studies consistently demonstrate that gender differences, often correlated with hormonal modulation, in the incidence, prevalence and modulation of deep tissue and inflammatory pain [340] [341]. Gonadal hormones are thought to play a modulatory role at the level of the primary afferent, dorsal horn and supraspinal sites, and descending inhibitory pain pathways. Their function appears, at least in part, to depend upon the balance between estradiol and testosterone.

Multiple meta analysis of the use of vitamin D for the treatment of chronic pain yield contradictory conclusions [342] [343]. In the chronic pain literature, results are inconsistent. In vitamin D deficient patient's, pain threshold and tolerance, particularly in instances of acute pain, appear to be lower. In such cases there is moderate evidence supporting the use of vitamin D3 to reduce pain. There is evidence that vitamin D may provide improved pain control in patients undergoing treatment for rheumatoid arthritis, osteoporosis, and sickle cell disease [344] [345] [346]. There is currently an ongoing, large cohort, well-designed study exploring pain severity and cartilage integrity as affected by vitamin D

status and patients with osteoarthritis [347].

In part the action of vitamin D may be etiology dependent, being expressed differentially following mechanical as opposed to biochemical (trauma with tissue acidosis, inflammatory, ischemic) insults [348]. It may also depend upon overall health status [349].

6. Concluding Thoughts

The above review is based upon information currently available. The relative explosion in pertinent research across diverse disciplines that encompasses genetics, physiology and clinical sciences is adding to our understanding on an almost daily basis. The link between mechanisms that provide homeostatic balance and plasticity within the nociceptive system is concurrently becoming clear.

Neuronal signaling is mediated by mechanical means, neurotransmitters or a vast array of proteins, many of which have been described in detail. The wide variety of signaling elements are often common to both steroid, and specifically vitamin D, dependent systems and the nociceptive system. These particularly include the signaling mechanisms, the neurotransmitters and receptors that effect nociceptive function through transcription of proteins that function not just physiologically, but also in a homeostatic or plastic manner.

Cytokines, protein kinases, various receptors that are stimulated directly (VDR, ionotropic receptors as examples) or through secondary messaging (G protein coupled metabotropic receptors), neurosteroid/steroid hormone, cholinergic, dopaminergic, serotonergic, GABA, glutamate and other signaling systems, and structural proteins and neurotrophic factors all play an apparent role.

The operational mechanics are inconclusive of the epigenetic pathways utilized by many systems to respond to signaling. The speculation that steroid dependent sub-systems within the nociceptive system, under certain conditions, might be signaled by vitamin D3, and possibly in specific cases by testosterone, is a reasonable one given the current evidence base.

Viewing homeostasis within the framework of epigenetics allows reasoned speculation as to how various human systems interact to maintain integrity and function, while responding in a plastic manner to external stimuli. This holds promise in the clinical application of information in order to provide more evidence based, specific and targeting intervention in the management of human pain.

While this understanding of complex systems is in its infancy, it suggests potentially fruitful areas for both foundational and clinical research. It might be a seed for the future development of a concept of epigenetics, plasticity and homeostasis, and the role of vitamin D3, in the nociceptive system.

References

[1] Epigenetics. BioMedicine.

[2] Epigenetics. Wikipedia, the Free Encyclopediaen. https://en.wikipedia.org/wiki/Epigenetics

[3] Haig, D. (2004) The (Dual) Origin of Epigenetics. *Cold Spring Harbor Symposia on Quantitative Biology*, **69**, 67-70.

[4] Berger, S.L. Kouzarides, T., Shiekhattar, R. and Shilatifard. A. (2009) An Operational Definition of Epigenetics. *Cold Spring Harbor Symposia on Quantitative Biology*, **23**, 781-783.

[5] Gottlieb, G. (2007) Probabilistic Epigenesis. *Developmental Science*, **10**, 1-11. https://doi.org/10.1111/j.1467-7687.2007.00556.x

[6] Development—Epigenesis and the Epigenetic Cascade. https://notes.utk.edu/Bio/

[7] Mehler, M.F. (2008) Epigenetic Principles and Mechanisms Underlying Nervous System Functions in Health and Disease. *Progress in Neurobiology*, **86**, 305-341.

[8] Singh, S. and Li, S.S.-L. (2012) Epigenetic Effects of Environmental Chemicals Bisphenol and Phthalates. *International Journal of Molecular Sciences*, **13**, 10143-10153. https://doi.org/10.3390/ijms130810143

[9] Fraga, M.F., Ballestar, E., Paz, M.F., Ropero, S., Setien, F., Ballestar, M.L., Heine-Suner. D., Cigudosa, J.C., Urioste, M., Benitez, J., Boix-Chornet, M., Sanchez-Aguilera, A., Ling, C., Carlsson, E., Poulsen, P., Vaag, A., Stephan, Z., Spector, T.D., Wu, Y.Z., Plass, C. and Esteller, M. (2005) Epigenetic Differences Arise during the Lifetime of Monozygotic Twins. *Proceedings of the National Academy of Sciences of the United States of America*, **102**, 10604-10609.

[10] Meaney, M.J. and Szyf, M. (2005) Environmental Programming of Stress Responses through DNA Methylation: Life at the Interface between a Dynamic Environment and a Fixed Genome. *Dialogues in Clinical Neuroscience*, **7**, 103-123.

[11] Irvine, R.A. Lin, I.G. and Hsieh, C.-L. (2002) DNA Methylation Has a Local Effect on Transcription and Histone Acetylation. *Molecular and Cellular Biology*, **22**, 6689-6696. https://doi.org/10.1128/MCB.22.19.6689-6696.2002

[12] Vaissière, T., Sawan, C. and Herceg, Z. (2008) Epigenetic Interplay between Histone Modifications and DNA Methylation in Gene Silencing. *Mutation Research*, **659**, 40-48.

[13] Yang, X.J. and Chiang, C.M. (2013) Sumoylation in Gene Regulation, Human Disease, and Therapeutic Action. *F*1000*Prime Reports*, **5**, 45.

[14] Zhang, W. and Sidhu, S.S. (2013) Development of Inhibitors in the Ubiquitination Cascade. *FEBS Letters*, **588**, 356-356.

[15] Valin, A. and Gill, G. (2007) Regulation of the Dual-Function Transcription Factor Sp3 by SUMO. *Biochemical Society Transactions*, **35**, 1393-1396.

[16] Zalucki, Y.M., Power, P.M. and Jennings, M.P. (2007) Selection for Efficient Translation Initiation Biases Codon Usage at Second Amino Acid Position in Secretory Proteins. *Nucleic Acids Research*, **5**, 5748-5754.

[17] Schlick, T., Hayes, J. and Grigoryev, S. (2012) Toward Convergence of Experimental Studies and Theoretical Modeling of the Chromatin Fiber. *Journal of Biological Chemistry*, **287**, 5183-5191.

[18] Onder, T.T., Kara, N., Cherry, A., Sinha, A.U., Zhu, N., Bernt, K.M., Cahan, P., Marcarci, B.O., Unternaehrer, J., Gupta, P.B., Lander, E.S., Armstrong, S.A. and Daley, G.Q. (2012) Chromatin-Modifying Enzymes as Modulators of Reprogramming. *Nature*, **483**, 598-602.

[19] Iyer, L.M., Anantharaman, V., Wolf, M.Y. and Aravind, L. (2008) Comparative Genomics of Transcription Factors and Chromatin Proteins in Parasitic Protists and Other Eukaryotes. *International Journal for Parasitology*, **38**, 1-31.

[20] Sawicka, A. and Seiser, C. (2012) Histone H3 phosphorylation—A Versatile Chro-

matin Modification for Different Occasions. *Biochimie*, **94**, 2193-2201.

[21] Long, H.K., Blackledge, N.P. and Klose, R.J. (2013) ZF-CxxC Domain—Containing Proteins, CpG Islands and the Chromatin Connection. *Biochemical Society Transactions*, **41**, 727-740.

[22] Deaton, A.M. and Bird, A. (2011) CpG Islands and the Regulation of Transcription. *Genes & Development*, **5**, 1010-1022.

[23] Ahmad, I. and Rao, D.N. (1996) Chemistry and Biology of DNA Methyltransferases. *Critical Reviews in Biochemistry and Molecular Biology*, **31**, 361-380.

[24] He, X.J., Chen, T. and Zhu, J.K. (2011) Regualtion and Function of DNA Methylation in Plants and Animals. *Cell Research*, **21**, 442-465.

[25] Yoder, J.A., Walsh, C.P. and Bestor, T.H. (1997) Cytosine Methylation and the Ecology of Intragenomic Parasites. *Trends in Genetics*, **13**, 335-340.

[26] Ferrari, K.J. and Pasini, D. (2013) Regulation and Function of DNA and Histone Methylations. *Current Pharmaceutical Design*, **19**, 719-733.

[27] Hublitz, P., Albert, M. and Peters, A.H. (2009) Mechanisms of Transcriptional Repression by Histone Lysine Methylation. *International Journal of Developmental Biology*, **53**, 335-354.

[28] Brian, A. and Meshorer, E. (2012) Concise Review: Chromatin and Genome Organization in Reprogramming. *Stem Cells*, **30**, 1793-1799.

[29] Gill, M.E., Erkek, S. and Peters, A.H. (2012) Parental Epigenetic Control of Embryogenesis: A Balance between Inheritance and Reprogramming. *Current Opinion in Cell Biology*, **24**, 387-396.

[30] Scharf, A.N. and Imhof, A. (2011) Every Methyl Counts—Epigenetic Calculus. *FEBS Letters*, **585**, 2001-2007.

[31] Cheng, X. and Zhang, X. (2007) Structural Dynamics of Protein Lysine Methylation and Demethylation. *Mutation Research/Fundamental and Molecular Mechanisms of Mutagenesis*, **618**, 102-115.

[32] Agger, K., Cloos, P.A., Christensen, J., Pasini, D., Rose, S., Rappsilber, J., Issaeva, I., Canaani, E., Salcini, A.E. and Helin, K. (2007) UTX and JMJD3 Are Histone H3K27 Demethylases Involved in HOX Gene Regulation and Development. *Nature*, **449**, 731-734.

[33] Jiao, A.L. and Slack, F.J. (2013) RNA-Mediated Gene Activation. *Epigenetics*, **9**, 27-36.

[34] Liu, X., Chen, X., Yu, X., Tao, Y., Bode, A.M., Dong, Z. and Cao, Y.J. (2013) Regulation of MicroRNAs by Epigenetics and Their Interplay Involved in Cancer. *Journal of Experimental & Clinical Cancer Research*, **32**, 96.

[35] Schiffgen, M., Schmidt, D.H., Von Rucker, A., Muller, S.C. and Ellinger, J. (2013) Epigenetic Regulation of MicroRNA Expression in Renal Cell Carcinoma. *Biochemical and Biophysical Research Communications*, **436**, 79-84. https://doi.org/10.1016/j.bbrc.2013.05.061

[36] Liu, C., Teng, Z.Q., McQuate, A.L., Jobe, E.M., Christ, C.C., Von Hoyningen-Huene, S.J., Reyes, M.D., Polich, E.D., Xing, Y., Li, Y., Guo, W. and Zhao, X. (2013) An Epigenetic Feedback Regulatory Loop Involving MicroRNA-195 and MBD1 Governs Neural Stem Cell Differentiation. *PLoS ONE*, **8**, e51436.

[37] MicroRNA in Wikipedia.

[38] Bergeron, L.J., Ouellet, J. and Perreault, J.P. (2003) Ribozyme-Based Gene-Inactivation Systems Require a Fine Comprehension of Their Substrate Specificities; the Case of

Delta Ribozyme. *Current Medicinal Chemistry*, **10**, 2589-2597. https://doi.org/10.2174/0929867033456486

[39] Locke, S.M. and Martienssen, R.A. (2006) Slicing and Spreading of Heterochromatic Silencing by RNA Interference. *Cold Spring Harbor Symposia on Quantitative Biology*, **71**, 497-503. https://doi.org/10.1101/sqb.2006.71.062

[40] Kim, K. and Liu, F. (2007) Inhibition of Gene Expression in Human Cells Using RNase P-Derived Ribozymes and External Guide Sequences. *Biochimica et Biophysica Acta* (*BBA*)—*Gene Structure and Expression*, **1769**, 603-612. https://doi.org/10.1016/j.bbaexp.2007.09.001

[41] Hohjoh, H. (2013) Disease-Causing Allele-Specific Silencing by RNA Interference. *Pharmaceutical*s (*Basel*), **6**, 522-535

[42] Epigenetics in Wikipedia.

[43] Ostuni, R., Piccolo, V., Barozzi, I., Polletti, S., Termanini, A., Bonifacio, S., Curina, A., Prosperini, E., Ghisletti, S. and Natoli, G. (2013) Latent Enhancers Activated by Stimulation in Differentiated Cells. *Cell*, **152**, 157-171.

[44] Flottmann, M., Scharp, T. and Klipp, E. (2012) A Stochastic Model of Epigenetic Dynamics in Somatic Cell Programming. *Frontiers in Physiology*, **3**, 216. https://doi.org/10.3389/fphys.2012.00216

[45] Ames, B.N., Shigenaga, M.K. and Hagen, T.M. (1993) Oxidants, Antioxidants and the Degenerative Diseases of Aging. *Proceedings of the National Academy of Sciences of the United States of America*, **90**, 7915-7922. https://doi.org/10.1073/pnas.90.17.7915

[46] Lee, B., Morano, A., Porcellini, A. and Muller, M.T. (2012) GADD45$^{\alpha}$ Inhibition of DNMT1 Dependent DNA Methylation during Homology Directed DNA Repair. *Nucleic Acids Research*, **40**, 2481-2493. https://doi.org/10.1093/nar/gkr1115

[47] Teodoridis, J.M., Strathdee, G. and Brown, R. (2004) Epigenetic Silencing Mediated by CpG Island Mehtylation: Potential as a Therapeutic Target and as a Biomarker. *Drug Resistance Updates*, **7**, 267-278. https://doi.org/10.1016/j.drup.2004.06.005

[48] Orlando, L., Schiavone, P., Fedele, P., Calvani, N., Nacci, A., Cinefra, M., D'Amico, M., Mazzoni, E., Marino, A., Sponziello, F., Morelli, F., Lombardi, L., Silvestris, N. and Cinieri, S. (2012) Poly (ADP-Ribose) Polymerase (PARP): Rationale, Preclinical and Clinical Evidences of Its Inhibition as Breast Cancer Treatement. *Expert Opinion on Therapeutic Targets*, **16**, S83-S89.

[49] Wang, C.H., Wu, S.B., Wu, Y.T. and Wei, Y.H. (2013) Oxidative Stress Response Elicited by Mitochondrial Dysfunction: Implication in the Pathophysiology of Aging. *Experimental Biology and Medicine*, **238**, 450-460. https://doi.org/10.1177/1535370213493069

[50] Korkmaz, A., Rosales-Corral, S. and Reiter, R.J. (2012) Gene Regulation by Melatonin Linked to Epigenetic Phenomena. *Gene*, **503**, 1-11. https://doi.org/10.1016/j.gene.2012.04.040

[51] Lunec, J., Herbert, K., Blount, S., Griffiths, H.R. and Emery, P. (1994) 8-Hydroxydeoxyguanosine. A Marker of Oxidative DNA Damage in Systemic Lupus Erythematosus. *FEBS Letters*, **348**, 131-138. https://doi.org/10.1016/0014-5793(94)00583-4

[52] Nociception from Wikipedia.

[53] Drummond, P.D. (2010) Sensory Disturbances in Complex Regional Pain Syndrome: Clinical Observations, Autonomic Interactions, and Possible Mechanisms. *Pain Medicine*, **11**, 1257-1266.

[54] Willis, W.D. (1985) Nociceptive Pathways: Anatomy and Physiology of Nociceptive Ascending Pathways. *Philosophical Transactions of the Royal Society of London. Series B, Biological Sciences*, **308**, 253-270.

[55] Willis Jr., W.D. (1988) Anatomy and Physiology of Descending Control of Nociceptive Responses of Dorsal Horn Neurons: Comprehensive Review. *Progress in Brain Research*, **77**, 1-29.

[56] Fields, H.L., Heinricher, M.M. and Mason, P. (1991) Neurotransmitters in Nociceptive Modulatory Circuits. *Annual Review of Neuroscience*, 1**4**, 219-245.

[57] Holden, J.E., Jeong, Y. and Forrest, J.M. (2005) The Endogenous Opioid System and Clinical Pain Management. *AACN Clinical Issues*, **16**, 291-301.

[58] Stein, C., Clark, J.D., Oh, U., Vasko, M.R., Wilcoxe, G.L., Overland, A.C., Vanderah, T.W. and Spencerg, R.H. (2009) Peripheral Mechanisms of Pain and Analgesia. *Brain Research Reviews*, **60**, 90-113.

[59] Mayer, D.J. and Price, D.D. (1976) Central Nervous System Mechanisms of Analgesia. *PAIN*, **2**, 379-404.

[60] Sato, H., Droney, J., Ross, J., Olesen, A.E., Staahl, C., Andersen, T., Branford, R., Riley, J., Arendt-Nielsen, L. and Drewes, A.M. (2013) Gender, Variation in Opioid Receptor Genes and Sensitivity to Experimental Pain. *Molecular Pain*, **9**, 20.

[61] Doehring, A., Geisslinger, G. and Lotsch, J. (2011) Epigenetics in Pain and Analgesia: An Imminent Research Field. *European Journal of Pain*, **15**, 11-16. https://doi.org/10.1016/j.ejpain.2010.06.004

[62] Zhu, X.Y., Huang, C.S., Li, Q., Chang, R.M., Song, Z.B., Zou, W.Y. and Guo, Q.L. (2012) P300 Exerts an Epigenetic Role in Chronic Neuropathic Pain through Its Acetyltransferase Activity in Rats Following Chronic Constriction Injury (CCI). *Molecular Pain*, **8**, 84.

[63] Kubo, A., Katanosaka, K. and Mizumura, K. (2012) Extracellular Matrix Proteoglycan Plays a Pivotal Role in Sensitization by Low pH of Mechanosensitive Currents in Nociceptive Sensory Neurons. *Journal of Physiology*, **590**, 2995-3007.

[64] Huang, C.W., Tzeng, J.N., Chen, Y.J., Tsai, W.F., Chen, C.C. and Sun, W.H. (2007) Nociceptors of Dorsal Root Ganglion Express Proton-Sensing G-Protein-Coupled Receptors. *Molecular and Cellular Neuroscience*, **36**, 195-210.

[65] Birklein, F., Weber, M., Ernst, M., Riedl, B., Neundorfer, B. and Handwerker, H.O. (2000) Experimental Tissue Acidosis Leads to Increased Pain in Complex Regional Pain Syndrome (CRPS). *PAIN*, **87**, 227-234. https://doi.org/10.1016/s0304-3959(00)00286-4

[66] Garaughty, S., Chu, K.L., Brown, B.S., Zhu, C.Z., Zhong, C., Joshi, S.K., Honore, P., Faltynek, C.R. and Jarvis, M.F. (2008) Contribtions of Central and Peripheral TRPV1 Receptors to Mechanically Evoked and Spontaneous Firing of Spinal Neurons in Inflamed Rats. *Journal of Neurophysiology*, **100**, 3158-3166. https://doi.org/10.1152/jn.90768.2008

[67] Wieczorek, M., Ginter, T., Brand, P., Heinzel, T. and Kramer, O.H. (2012) Acetylation Modulates the STAT Signaling Code. *Cytokine & Growth Factor Reviews*, **23**, 293-305. https://doi.org/10.1016/j.cytogfr.2012.06.005

[68] Takeshima, H., Ikegmai, D., Wakabayashi, M., Niwa, T., Kim, Y.J. and Ushijima, T. (2012) Induction of Aberrant Trimethylation of Histone H_3 Lysine 27 by Inflammation in Mouse Colonic Epithelial Cells. *Carcinogenesis*, **33**, 2384-2390. https://doi.org/10.1093/carcin/bgs294

[69] Tian, Y., Ni, D., Yang, W., Zhang, Y., Zhao, K., Song, J., Mao, Q., Tian, Z., Van Vel-

kinburgh, J.C., Yang, D., Wu, Y. and Ni, B. (2013) Telbivudine Treatment Corrects HBV-Induced Epigenetic Alterations in Liver Cells of Patients with Chronic Hepatitis B. *Ciarcinogenesis*, **35**, 53-61.

[70] Taylor, B.K., Fu, W., Kuphal, K.E., Stiller, C.O., Winter, M.K., Chen, W., Corder, G.F., Urban, J.H., McCarson, K.E. and Marvizon, J.C. (2014) Inflammation Enhances Y1 Receptor Signaling, Neuropeptide Y-Mediated Inhibition of Hyepralgesia, and Substance P Release from Primary Afferent Neurons. *Neuroscience*, **256**, 178-194.

[71] Lin, Y.T., Ro, L.S., Wang, H.L. and Chen, J.C. (2011) Up Regulation of Dorsal Root Ganglia BDNF and trkB Receptor in Inflammatory Pain: An *in Vivo* and *in Vitro* Study. *Journal of Neuroinflammation*, **8**, 126. https://doi.org/10.1186/1742-2094-8-126

[72] Cang, C.L., Zhang, H., Zhang, Y.Q. and Zhao, Z.Q. (2009) PKCϵ-Dependent Potentiation of TTX-Resistant $Na_v1.8$ Current by Neurokinin-1 Receptor Activation in Rat Dorsal Root Ganglion Neurons. *Molecular Pain*, **5**, 33. https://doi.org/10.1186/1744-8069-5-33

[73] Chang, C.W., Lee, L., Yu, D., Dao, K., Bossuyt, J. and Bers, D.M. (2013) Acute β-Adrenergic Activation Triggers Nuclear Import of Histone Deacetylase 5 and Delays G_q-Induced Transcriptional Activation. *Journal of Biological Chemistry*, **288**, 192-204. https://doi.org/10.1074/jbc.M112.382358

[74] Ventura, C. and Mailoi, M. (2001) Protein Kinase C Control of Gene Expression. *Critical Reviews in Eukaryotic Gene Expression*, **11**, 243-267.

[75] Wu, D.F., Chandra, D., McMahon, T., Wang, D., Dadgar, J., Kharazia, V.N., Liang, Y.J., Waxman, S.G., Dib-Hajj, S.D. and Messing, R.O. (2012) PKCε Phosphorylation of the Sodium Channel $Na_V1.8$ Increases Channel Function and Produces Mechanical Hyperalgesia in Mice. *Journal of Clinical Investigation*, **122**, 1306-1315. https://doi.org/10.1172/JCI61934

[76] Zheng, J.H., Walters, E.T. and Song, X.J. (2007) Dissociation of Dorsal Root Ganglion Neurons Induces Hyperexcitability That Is Maintained by Increased Responsiveness to cAMP and cGMP. *Journal of Neurophysiology*, **97**, 15-25. https://doi.org/10.1152/jn.00559.2006

[77] Sirtuin in Wikipedia.

[78] Etchegaray, J.P., Zhong, L. and Mostoslavsky, R. (2013) The Histone Deacetylase SIRT6: At The Crossroads between Epigenetics, Metabolism and Disease. *Current Topics in Medicinal Chemistry*, **13**, 2991-3000.

[79] Adam, S., Sprouse-Blum, B.A., Greg Smith, B.S., Daniel Sugai, B.A. and Don Parsa, M.D.F. (2010) Understanding Endorphins and Their Importance in Pain Management. *Hawaii Medical Journal*, **69**, 70-71.

[80] Ossipov, M.H., Dussor, G.O. and Porreca, F. (2010) Central Modulation of Pain. *Journal of Clinical Investigation*, **120**, 3779-3787.

[81] Hunt, S.P. (2009) Genes and the Dynamics of Pain Control. *Functional Neurology*, **24**, 9-15.

[82] Wang, Q. and Traynor, J.R. (2013) Modulation of μ-Opioid Receptor Signaling by RGS19 in SH-SY5Y Cells. *Molecular Pharmacology*, **83**, 512-520. https://doi.org/10.1124/mol.112.081992

[83] Clark, M.J., Linderman, J.J. and Traynor, J.R. (2008) Endogenous Regulators of G Protein Signaling Differentially Modulate Full and Partial μ-Opioid agonists at Adenyl Cyclase as Predicted by a Collision Coupling Model. *Molecular Pharmacology*, **73**, 1538-1548. https://doi.org/10.1124/mol.107.043547

[84] Garzon, J., Rodriguez-Munoz, M., de la Torre-Madrid, E. and Sanchez-Blazquez, P. (2005) Effector Antagonism by the Regulators of G Protein Signaling (RGS) Proteins Causes Desensitization of μ-Opioid Receptors in the CNS. *Psychopharmacology* (*Berlin*), **180**, 1-11.

[85] Reelin in Wikipedia.

[86] Campo, C.G., Sinagra, M., Verrier, D., Manzoni, O.J. and Chavis, P. (2009) Reelin Secreted by GABAergic Neurons Regulates Glutamate Receptor Homeostasis. *PLoS ONE*, **4**, e5505.

[87] Villeda, S.A., Akopians, A.L., Babayan, A.H., Basbaum, A.I. and Phelps, P.E. (2006) Absence of Reelin Results in Altered Nociception and Aberrant Neuronal Positioning in the Dorsal Spinal Cord. *Neuroscience*, **139**, 1385-1396.

[88] Wang, X., Babayan, A.H., Basbaum, A.I. and Phelps, P.E. (2012) Loss of the Reelin-Signaling Pathway Differentially Disrupts Heat, Mechanical and Chemical Nociceptive Processing. *Neuroscience*, **226**, 441-450. https://doi.org/10.1016/j.neuroscience.2012.09.027

[89] Mitchell, C.P., Chen, Y., Kundakovic, M., Costa, E. and Grayson, D.R. (2005) Histone Deacetylase Inhibitors Decrease Reelin Promoter Methylation *in Vitro*. *Journal of Neurochemistry*, **93**, 483-492. https://doi.org/10.1111/j.1471-4159.2005.03040.x

[90] Ognibene, E., Adriani, W., Granstream, O., Pieretti, S. and Laviola, G. (2007) Impulsivity-Anxiety-Related Behaviour and Profiles of Morphine-Induced Analgesia in Heterozygous Reeler Mice. *Brain Research*, **1131**, 173-180.

[91] Jarcho, J.M., Mayer, E.A., Jiang, Z.K., Feier, N.A. and London, E.D. (2012) Pain, Affective Symptoms, and Cognitive Deficits in Patients with Cerebral Dopamine Dysfunction. *PAIN*, **153**, 744-754. https://doi.org/10.1016/j.pain.2012.01.002

[92] Lapirot, O., Melin, C., Modolo, A., Nicolas, C., Messaoudi, Y., Monconduit, L., Artola, A., Luccarini, P. and Dallel, R. (2011) Tonic and Phasic Descending Dopaminergic Controls of Nociceptive tRansmission in the Medullary Dorsal Horn. *PAIN*, **152**, 1821-1831. https://doi.org/10.1016/j.pain.2011.03.030

[93] Karpova, N.N. (2013) Role of BDNF Epigenetics in Activity-Dependent Neuronal Plasticity. *Neuropharmacology*, **76**, 709-718.

[94] Duric, V. and McCarson, K.E. (2007) Neurokinin-1 (NK-1) Receptor and Brain-Derived Neurotrophic Factor (BDNF) Gene Expressions Is Differentially Modulated in the Rat Spinal Dorsal Horn and Hippocampus during Inflammatory pain. *Molecular Pain*, **3**, 32.

[95] Duric, V. and McCarson, K.E. (2006) Effects of Analgesic or Antidepressant Drugs on Pain- or Stress-Evoked Hippocampal and Spinal Neurokinin-1 Receptor and Brain-Derived Neurotrophic Factor Gene Expression in the Rat. *Journal of Pharmacology and Experimental Therapeutics*, **319**, 1235-1243. https://doi.org/10.1124/jpet.106.109470

[96] Tian, F., Hu, X.Z., Wu, X., Jiang, H., Pan, H., Marini, A.M. and Lipsky, R.H. (2009) Dynamic Chromatin Remodeling Events in Hippocampal Neurons Are Associated with NMDA Receptor-Mediated Activation of *Bdnf* Gene Promoter 1. *Journal of Neurochemistry*, **109**, 1375-1388. https://doi.org/10.1111/j.1471-4159.2009.06058.x

[97] NMDA Receptor in Wikipedia.

[98] Aira, Z., Buesa, I., Garcia del Cano, G., Bilbao, J., Donate, F., Zimmermann, M. and Azkue, J.J. (2013) Transient, 5-HT2B Receptor-Mediated Facilitation in Neuropathic Pain: Up-Regulation of PKCγ and Engagement of the NMDA Receptor in Dorsal Horn Neurons. *PAIN*, **154**, 1865-1877.

https://doi.org/10.1016/j.pain.2013.06.009

[99] Fenselau, H., Heinke, B. and Sandkuhler, J. (2011) Heterosynaptic Long-Term Potentiation at GABAergic Synapses of Spinal Lamina I Neurons. *Journal of Neurochemistry*, **31**, 17383-17391. https://doi.org/10.1523/JNEUROSCI.3076-11.2011

[100] Jin, D.Z., Guo, M.L., Xue, B., Mao, L.M. and Wang, J.Q. (2013) Differential Regulation of CaMKIIα Interactions with mGluR5 and NMDA Receptors by Ca^{2+} in Neurons. *Journal of Neuroscience*, **127**, 620-631.

[101] Soares, C., Lee, K.F., Nassrallah, W. and Beique, J.C. (2013) Differential Subcellular Targeting of Glutamate Receptor Subtypes during Homeostatic Synaptic Plasticity. *Journal of Neuroscience*, **33**, 13547-13559. https://doi.org/10.1523/JNEUROSCI.1873-13.2013

[102] Yan, D., Yamasaki, M., Straub, C., Watanabe, M. and Tomita, S. (2013) Homeostatic Control of Synaptic Transmission by Distinct Glutamate Receptors. *Neuron*, **78**, 687-699. https://doi.org/10.1016/j.neuron.2013.02.031

[103] Flores-Soto, M.E., Chaparro-Huerta, V., Escoto-Delgadillo, M., Urena-Guerrero, M.E., Camins, A. and Beas-Zarate, C. (2013) Receptor to Glutamate NMDA-Type: The Functional Diversity of the NR1 Isoforms and Pharmacological Properties. *Current Pharmaceutical Design*, **19**, 6709-6719.

[104] Papouin, T., Ladepeche, L., Ruel, J., Sacchi, S., Labasque, M., Hanini, M., Groc, L., Pollegioni, L., Mothet, J.P. and Oliet, S.H. (2012) Synaptic and Extrasynaptic NMDA Receptors Are Gated by Different Endogenous Coagonists. *Cell*, **150**, 633-646. https://doi.org/10.1016/j.cell.2012.06.029

[105] Adenuga, D. and Rahman, I. (2010) Protein Kinase CK2-Mediated Phosphorylation of HDAC2 Regulates Co-Repressor Formation, Deacetylase Activity and Acetylation of HDAC2 by Cigarette Smoke and Aldehydes. *Archives of Biochemistry and Biophysics*, **498**, 62-73.

[106] Flood, S., Parri, R., Williams, A., Duance, V. and Mason, D. (2007) Modulation of Interleukin-6 and Matrix Metalloproteinase 2 Expression in Human Fibroblast-Like Synoviocytes by Functional Ionotropic Glutamate Receptors. *Arthritis & Rheumatology*, **56**, 2523-2534. https://doi.org/10.1002/art.22829

[107] Zhang, X., Xu, Y., Wang, J., Zhou, Q., Pu, S., Jiang, W. and Du, D. (2012) The Effect of Intrathecal Administration of Glial Activation Inhibitors on Dorsal Horn BDNF Overexpression and Hind Paw Mechanical Allodynia in Spinal Nerve Ligated Rats. *Journal of Neural Transmission*, **119**, 329-336. https://doi.org/10.1007/s00702-011-0713-7

[108] Zhou, L.-J., Yang, T., Wei, X., Liu, Y., Xin, W.J., Chen, Y., Pang, R.P., Zang, Y, Li, Y.Y. and Liu, X.G. (2011) Brain-Derived Neurotrophic Factor Contributes to Spinal Long-Term Potentiation and Mechanical Hypersensitivity by Activation of Spinal Microglia in Rat. *Brain, Behavior, and Immunity*, **25**, 322-334. https://doi.org/10.1016/j.bbi.2010.09.025

[109] Zhou, X., Lin, D.S., Zheng, F., Sutton, M.A. and Wang, H. (2010) Intracellular Calcium and Calmodulin Link Brain-Derived Neurotrophic Factor to p70S6 Kinase Phosphorylation and Dendritic Protein Synthesis. *Journal of Neuroscience Research*, **8**, 1420-1432.

[110] Xin, W.J., Gong, Q.J., Xu, J.T., Yang, H.W., Zang, Y., Zhang, T., Li, Y.Y. and Liu, X.G. (2006) Role of Phosphorylation of ERK in Induction and Maintenance of LTP of the C-Fiber Evoked Field Potentials in Spinal Dorsal Horn. *Journal of Neuroscience Research*, **84**, 934-943. https://doi.org/10.1002/jnr.21013

[111] Kawasaki, Y., Kohno, T., Zhuang, Z.Y., Brenner, G.J., Wang, H., Van der Meer, C.,

Befort, K., Woolf, C.J. and Ji, R.R. (2004) Ionotropic and Metabotropic Receptors, Protein Kinase A, Protein Kinase C, and Src Contribute to C-Fiber-Induced ERK Activation and Camp Response Element-Binding Protein Phosphorylation in Dorsal Horn Neurons, Leading to Central Sensitization. *Journal of Neuroscience*, **24**, 8310-8321. https://doi.org/10.1523/JNEUROSCI.2396-04.2004

[112] Gabay, O. and Sanchez, C. (2012) Epigenetics, Sirtuins and Osteoarthritis. *Joint Bone Spine*, **79**, 570-573. https://doi.org/10.1016/j.jbspin.2012.04.005

[113] Ni, J., Gao, Y., Gong, S., Guo, S., Hisamitsu, T. and Jiang, X. (2013) Regulation of μ-Opioid Type 1 Receptors by MicroRNA 134 in Dorsal Root Ganglion Neurons Following Peripheral Inflammation. *European Journal of Pain*, **17**, 313-323. https://doi.org/10.1002/j.1532-2149.2012.00197.x

[114] Tam Tam, S., Bastian, I., Zhou, X.F., Vander Hoek, M., Michael, M.Z., Gibbins, I.L. and Haberberger, R.V. (2011) MicroRNA-143 Expression in Dorsal Root Ganglion Neurons. *Cell and Tissue Research*, 163-173. https://doi.org/10.1007/s00441-011-1263-x

[115] Li, X., Kroin, J.S., Kc, R., Gibson, G., Chen, D., Corbett, G.T., Pahan, K., Fayyaz, S., Kim, J.S., van Wijnen, A.J., Suh, J., Kim, S.G. and Im, H.J. (2013) Altered Spinal MicroRNA-146a and the MicroRNA-183 Cluster Contribute to Osteoarthritic Pain in Knee Joints. *Journal of Bone and Mineral Research*, **28**, 2512-2522.

[116] Gheinani, A.H., Burkhard, F.C. and Monastyrskaya, K. (2013) Deciphering MicroRNA Code in Pain and Inflammation: Lessons from Bladder Pain Syndrome. *Cellular and Molecular Life Sciences*, **70**, 3773-3789. https://doi.org/10.1007/s00018-013-1275-7

[117] Kynast, K.L., Russe, O.Q., Moser, C.V., Geisslinger, G. and Niederberger, E. (2013) Modulation of Central Nervous System—Specific MicroRNA-124a Alters the Inflammatory Response in the Formalin Test in Mice. *PAIN*, **154**, 368-376. https://doi.org/10.1016/j.pain.2012.11.010

[118] McCoy, C.E. (2011) The Role of MiRNAs in Cytokine Signaling. *Frontiers in Bioscience*, **16**, 2161-2171. https://doi.org/10.2741/3845

[119] Kusuda, R., Cadetti, F., Ravanelli, M.I., Sousa, T.A., Zanon, S., DeLucca, F.L. and Lucas, G. (2011) Differential Expression of MicroRNAs in Mouse Pain Models. *Molecular Pain*, **7**, 17.

[120] Hori, Y., Goto, G., Arai-Iwasaki, M., Ishikawa, M. and Sakamoto, A. (2013) Differential Expression of Rat Hippocampal MicroRNAs in Two Rat Models of Chronic Pain. *International Journal of Molecular Medicine*, **32**, 1287-1292. https://doi.org/10.3892/ijmm.2013.1504

[121] Sakai, A., Saitow, F., Miyake, N., Miyake, K., Shimada, T. and Suzuki, H. (2013) MiR-7a Alleviates the Maintenance of Neuropathic Pain through Regulation of Neuronal Excitability. *Brain*, **136**, 2738-2750.

[122] Inoue, Y., Udo, H., Inokuchi, K. and Sugiyama, H. (2007) Homer1a Regulates the Activity-Induced Remodeling of Synaptic Structures in Cultured Hippocampal Neurons. *Neuroscience*, **150**, 841-852.

[123] Miletic, G., Driver, A.M., Miyabe-Nishiwaki, T. and Miletic, V. (2009) Early Changes in Homer1 Proteins in the Spinal Dorsal Horn Are Associated with Loose Ligation of the Rat Sciatic Nerve. *Anesthesia & Analgesia*, **109**, 2000-2007. https://doi.org/10.1213/ANE.0b013e3181beea9b

[124] Miyabe, T., Miletic, G. and Miletic, V. (2006) Loose Ligation of the Sciatic Nerve in Rats Elicits Transient Up-Regulation of Homer1a Gene Expression in the Spinal Dorsal Horn. *Neuroscience Letter*s, **398**, 296-299.

https://doi.org/10.1016/j.neulet.2006.01.011

[125] Shiraishi-Yamaguchi, Y. and Furuich, T. (2007) The Homer Family Proteins. *Genome Biology*, **8**, 206. https://doi.org/10.1186/gb-2007-8-2-206

[126] Tappe-Theodor, A., Fu, Y., Kuner, R. and Neugebauer, V. (2011) Homer1a Signaling in the Amygdala Counteracts Pain-Related Synaptic Plasticity, mGluR1 Function and Pain Behaviors. *Molecular Pain*, **7**, 38.

[127] Seeburg, P.H. and Hartner, J. (2003) Regulation of Ion Channel/Neurotransmitter Receptor Function by RNA Editing. *Current Opinion in Neurobiology*, **13**, 279-283. https://doi.org/10.1016/S0959-4388(03)00062-X

[128] Schmauss, C. and Howe, J.R. (2002) RNA Editing of Neurotransmitter Receptors in the Mammalian Brain. *Science's STKE*, **2002**, pe26.

[129] Carmel, L., Koonin, E.V. and Dracheva, S. (2012) Dependencies among Editing Sites in Serotonin 2C Receptor mRNA. *PLoS Computational Biology*, **8**, e1002663. https://doi.org/10.1371/journal.pcbi.1002663

[130] Miranda, A., Peles, S., McLean, P.G. and Sengupta, J.N. (2006) Effects of the 5-HT3 Receptor Antagonist, Alosetron, in a Rat Model of Somatic and Visceral Hyperalgesia. *PAIN*, **126**, 54-63.

[131] Charlet, A., Lasbennes, F., Darbon, P. and Poisbeau, P. (2008) Fast Non-Genomic Effects of Progesterone-Derived Neurosteroids on Nociceptive Thresholds and Pain Symptoms. *PAIN*, **139**, 603-609. https://doi.org/10.1016/j.pain.2008.06.016

[132] King, S.R. (2013) Neurosteroids and the Nervous System. Springer-Verlag New York Inc., New York, 70-71.

[133] Trevisan, G., Rossato, M.F., Walker, C.I., Klafke, J.Z., Rosa, F., Oliveira, S.M., Tonello, R., Guerra, G.P., Boligon, A.A., Zanon, R.B., Athayde, M.L. and Ferreira, J. (2012) Identification of the Plant Steroid α-Spinasterol as a Novel Transient Receptor Potential Vanilloid 1 Antagonist with Antinociceptive Properties. *Journal of Pharmacology and Experimental Therapeutics*, **343**, 258-269. https://doi.org/10.1124/jpet.112.195909

[134] Lu, Y., Li, Z., Li, H.J., Du, D., Wang, L.P., Yu, L.H., Burnstock, G. and Chen, A. (2012) A Comparative Study of the Effect of 17β-Estradiol and Estriol on Peripheral Pain Behavior in Rats. *Steroids*, **77**, 241-249. https://doi.org/10.1016/j.steroids.2011.11.011

[135] Ji, Y., Tang, B. and Traub, R.J. (2011) Spinal Estrogen Receptor Alpha Mediates Estradiol-Induced Pronociception in a Visceral Pain Model in the Rat. *PAIN*, **152**, 1182-1191. https://doi.org/10.1016/j.pain.2011.01.046

[136] Melcangi, R.C., Giatti, S., Calabrese, D., Pesaresi, M., Cermenati, G., Mitro, N., Viviani, B., Garcia-Segura, L.M. and Caruso, D. (2013) Levels and Actions of Progesterone and Its Metabolites in the Nervous System during Physiological and Pathological Conditions. *Progress in Neurobiology*, **113**, 56-69.

[137] Roglio, I., Bianchi, R., Gotti, S., Scurati, S., Giatti, S., Pesaresi, M., Caruso, D., Panzica, G.C. and Melcangi, R.C. (2008) Neuroprotective Effects of Dihydroprogesterone and Progesterone in an Experimental Model of Nerve Crush Injury. *Neuroscience*, **155**, 673-685. https://doi.org/10.1016/j.neuroscience.2008.06.034

[138] Wojtal, K., Trojnar, M.K. and Czuczwar, S.J. (2006) Endogenous Neuroprotective Factors: Neurosteroids. *Pharmacological Reports*, **58**, 335-340.

[139] Mizota, K., Yoshida, A., Uchida, H., Fujita, R. and Ueda, H. (2005) Novel Type of Gq/11 Protein-Coupled Neurosteroid Receptor Sensitive to Endocrine Disrupting Chemicals in Mast Cell Line (RBL-2H3). *British Journal of Pharmacology*, **145**,

545-550.

[140] Patte-Mensah, C., Kibaly, C., Boudard, D., Schaeffer, V., Begle, A., Saredi, S., Meyer, L. and Mensah-Nyagan, A.G. (2006) Neurogenic Pain and Steroid Synthesis in the Spinal Cord. *Journal of Molecular Neuroscience*, **8**, 17-31.

[141] Miyamoto, M., Tsuboi, Y., Honda, K., Kobayashi, M., Takamiya, K., Huganir, R.L., Kondo, M., Shinoda, M., Sessle, B.J., Katagiri, A., Kita, D., Suzuki, I., Oi, Y. and Iwata, K. (2012) Involvement of AMPA Receptor GluR2 and GluR3 Trafficking in Trigeminal Spinal Subnucleus Caudalis and C1/C2 Neurons in Acute-Facial Inflammatory Pain. *PLoS ONE*, **7**, e44055.

[142] Wang, S., Dai, Y., Kobayashi, K., Zhu, W., Kogure, Y., Yamanaka, H., Wan, Y., Zhang, W. and Noguchi, K. (2012) Potentiation of the P2X3 ATP Receptor by PAR-2 in Rat Dorsal Root Ganglia Neurons, through Protein Kinase-Dependent Mechanisms, Contributes to Inflammatory Pain. *European Journal of Neuroscience*, **36**, 2293-2301. https://doi.org/10.1111/j.1460-9568.2012.08142.x

[143] Gangadharan, V., Wang, R., Ulzhofer, B., Luo, C., Bardoni, R., Bali, K.K., Agarwal, N., Tegeder, I., Hildebrandt, U., Nagy, G.G., Todd, A.J., Ghirri, A., Haussler, A., Sprengel, R. and Seeburg, P.H. (2011) Peripheral Calcium-Permeable AMPA Receptors Regulate Chronic Inflammatory Pain in Mice. *Journal of Clinical Investigation*, **121**, 1608-1623. https://doi.org/10.1172/JCI44911

[144] Zhou, H.Y., Zhang, H.M., Chen, S.R. and Pan, H.L. (2007) Increased Nociceptive Input Rapidly Modulates Spinal GABAergic Transmission through Endogenously Released Glutamate. *Journal of Neurophysiology*, **97**, 871-882. https://doi.org/10.1152/jn.00964.2006

[145] Kulie, T., Groff, A., Redmer, J., Hounshell, J. and Shcrager, S. (2009) Vitamin D: An Evidence-Based Review. *Journal of the American Board of Family Medicine*, **22**, 698-706.

[146] Turner, M.K., Hooten, W.M., Schmidt, J.E., Kerkvliet, J.L., Townsend, C.O. and Bruce, B.K. (2008) Prevalence Clinical Correlates of Vitamin D Inadequacy among Patients with Chronic Pain. *Pain Medicine*, **9**, 979-984. https://doi.org/10.1111/j.1526-4637.2008.00415.x

[147] Harrington, A.M., Brierley, S.M., Issacs, N., Hughes, P.A., Castro, J. and Blackshaw, L.A. (2012) Sprouting of Colonic Afferent Central Terminals and Increased Spinal Mitogen-Activated Protein Kinase Expression in a Mouse Model of Chronic Visceral Hypersensitivity. *Journal of Comparative Neurology*, **520**, 2241-2255. https://doi.org/10.1002/cne.23042

[148] Dastani, Z., Li, R. and Richards, B. (2013) Genetic Regulation of Vitamin D Levels. *Calcified Tissue International*, **92**, 106-117. https://doi.org/10.1007/s00223-012-9660-z

[149] Takeyama, K. and Kato, S. (2011) The Vitamin D3 1Alpha-Hydroxylase Gene and Its Regulation by Active Vitamin D3. *Bioscience, Biotechnology, and Biochemistry*, **75**, 208-213. https://doi.org/10.1271/bbb.100684

[150] Jones, G., Prosser, D.E. and Kaufmann, M. (2014) Cytochrome P450-Mediated Metabolism of Vitamin D. *Journal of Lipid Research*, **55**, 13-31.

[151] Cytochrome P450 in Wikipedia.

[152] Vitamin D in Wikipedia.

[153] Haussler, M.R., Haussler, C.A., Jurutka, P.W., Thompson, P.D., Hsieh, J.C., Remus, L.S., Selznick, S.H. and Whitfield, G.K. (1997) The Vitamin D Hormone and Its Nuclear Receptor: Molecular Actions and Disease States. *Journal of Endocrinology*, **154**, S57-S73.

[154] Ebert, R., Schutze, N., Adamski, J. and Jakob, F. (2006) Vitamin D Signaling Is Modulated on Multiple Levels in Health and Disease. *Molecular and Cellular Endocrinology*, **248**, 149-159.

[155] Larkins, R.G. and Aust, N.Z. (1981) The Current Status of the Vitamin D Metabolites. *Internal Medicine Journal*, **11**, 28-32.

[156] Mazzaferro, S., Goldsmith, D., Larsson, T.E., Massy, Z.A. and Cozzolino, M. (2013) Vitamin D Metabolites and/or Analogs: Which D for Which Plant? *Current Vascular Pharmacology*, **12**, 339-349.

[157] Secosteroids in Wikipedia.

[158] MacLaughlin, J.A., Anderson, R.R. and Holick, M.F. (1982) Spectral Character of Sunlight Modulates Photosynthesis of Previtamin D3 and Its Photoisomers in Human Skin. *Science*, **216**, 1001-1003.

[159] Zehnder, D., Bland, R., Williams, M.C., McNinch, R.W., Howie, A.J., Stewart, P.M. and Hewison, M. (2001) Extrarenal Expression of 25-Hydroxyvitamin d(3)-1 Alpha-Hdroxylase. *Journal of Clinical Endocrinology & Metabolism*, **86**, 888-894.

[160] Chen, J.B., Levine, M.A., Bell, N.H., Mangelsdorf, D.J. and Russell, D.W. (2004) Genetic Evidence That the Human CYP2R1 Enzyme Is a Key Vitamin D 25-Hydroxylase. *Proceedings of the National Academy of Sciences of the United States of America*, **101**, 7711-7715.

[161] Omdahl, J.L., Morris, H.A. and May, B.K. (2002) Hydroxylase Enzymes of the Vitamin D Pathway: Expression, Function, and Regulation. *Annual Review of Nutrition*, **22**, 139-166.

[162] Shuler, F.D., Ilendrix, J., Ilodrogc, S. and Short, A. (2013) Antibiotic-Like Actions of Vitamin D. *West Virginia Medical Journal*, **109**, 22-25.

[163] Ikeuchi, T., Nakamura, T., Fukumoto, S. and Takada, H. (2013) A Vitamin D3 Analog Augmented Interleukin-8 Production by Human Monocytic Cells in Response to Various Microbe-Related Synthetic Ligands, Especially NOD2 Agonistic Muramyldipeptide. *International Immunopharmacology*, **15**, 15-22.

[164] Deluca, H.F. (2004) Overview of General Physiologic Features and Functions of Vitamin D. *American Journal of Clinical Nutrition*, **80**, 1689S-1696S.

[165] Jin, C.H., Kerner, S.A., Hong, M.H. and Pike, J.W. (1996) Transcriptional Activation and Dimerization Functions in the Human Vitamin D Receptor. *Molecular Endocrinology*, **10**, 945-957.

[166] Zhang, X.Y., Payal, B., Melissa, O. and Zanello, L.P. (2007) 1α,25$(OH)_2$-Vitamin D_3 Membrane-Initiated Calcium Signaling Modulates Exocytosis and Cell Survival. *Journal of Steroid Biochemistry and Molecular Biology*, **103**, 457-461.

[167] Ozono, K., Sone, T. and Pike, J.W. (1991) The Genomic Mechanism of Action of 1,25-Dihydroxyvitamin D3. *Journal of Bone and Mineral Research*, **6**, 1021-1027.

[168] Maier, C.J., Maier, R.H., Rid, R., Trost, A., Hundsberger, H., Eger, A., Hintner, H., Bauer, J.W. and Onder, K. (2012) PIM-1 Kinase Interacts with the DNA Binding Domain of the Vitamin D Receptor: A Further Kinase Implicated in 1,25-$(OH)_2D_3$ Signaling. *BMC Molecular Biology*, **13**, 18.

[169] Arriagada, G., Paredes, R., Olate, J., van Wijnen, A., Lian, J.B., Stein, G.S,. Stein, J.L., Onate, S. and Montecino, M. (2007) Phosphorylation at Serine 208 of the 1Alpha,25-Dihydroxy Vitamin D3 Receptor Modulates the Interaction with Transcriptional Coactivators. *Journal of Steroid Biochemistry and Molecular Biology*, **103**, 425-429.

[170] Asano, L., Ito, I., Kuwabara, N., Waku, T., Yanagisawa, J., Miyachi, H. and Shimizu, T.

(2013) Structural Basis for Vitamin D Receptor Agonism by Novel Non-Secosteroidal Ligands. *FEBS Letters*, **587**, 957-963.

[171] Sicinska, W. and Rotkiewicz, P. (2009) Structural Changes of Vitamin D Receptor Induced by 20-epi-1Alpha,25-(OH)2D3: An Insight from a Computational Analysis. *Journal of Steroid Biochemistry and Molecular Biology*, **113**, 253-258.

[172] Campbell, F.C., Xu, H., El-Tanani, M., Crowe, P. and Bingham, V. (2010) The Yin and Yang of Vitamin D Receptor (VDR) Signaling in Neoplastic Progression: Operational Networks and Tissue-Specific Growth Control. *Biochemical Pharmacology*, **79**, 1-9.

[173] Nemazannikova, N., Antonas, K. and Dass, C.R. (2013) Role of Vitamin D Metabolism in Cutaneous Tumour Formation and Progression. *Journal of Pharmacy and Pharmacology*, **65**, 2-10.

[174] Lopes, N., Paredes, J., Costa, J.L., Ylstra, B. and Schmitt, F. (2012) Vitamin D and the Mammary Gland: A Review on Its Role in Normal Development and Breast Cancer. *Breast Cancer Research*, **14**, 211.

[175] Tuohimaa, P., Keisala, T., Minasyan, A., Cachat, J. and Kalueff, A. (2009) Vitamin D, Nervous System and Aging. *Psychoneuroendocrinology*, **34**, S278-S286.

[176] Redwood, A.B., Gonzalez-Suarez, I. and Gonzalo, S. (2011) Regulating the Levels of Key Factors in Cell Cycle and DNA Repair: New Pathways Revealed by Lamins. *Cell Cycle*, **10**, 3652-3657. https://doi.org/10.4161/cc.10.21.18201

[177] Chen, T.H., Kuro, O.M., Chen, C.H., Sue, Y.M., Chen, Y.C., Wu, H.H. and Cheng, C.Y. (2013) The Secreted Klotho Protein Restores Phosphate Retention and Suppresses Accelerated Aging in Klotho Mutant Mice. *European Journal of Pharmacology*, **698**, 67-73.

[178] Sempos, C.T., Durazo-Arvizu, R.A., Dawson-Hughes, B., Yetley, E.A., Looker, A.C., Schleicher, R.L., Cao, G., Burt, V., Kramer, H., Bailey, R.L., Dwyer, J.T., Zhang, X., Gahche, J., Coates, P.M. and Picciano, M.F. (2013) Is There a Reverse J-Shaped Association between 25-Hydroxyvitamin D and All-Cause Mortality? Results from the U.S. Nationally Respresentative NHANES. *Journal of Clinical Endocrinology & Metabolism*, **98**, 3001-3009.

[179] Pike, J.W. and Meyer, M.B. (2010) The Vitamin D Receptor: New Paradigms for the Regulation of Gene Expression by 1,25-Dihydroxyvitamin D_3. *Endocrinology Metabolism Clinics of North America*, **39**, 255-269.

[180] St-Arnaud, R. (2008) The Direct Role of Vitamin D on Bone Homeostasis. *Archives of Biochemistry and Biophysics*, **473**, 255-230.

[181] Choi, M., Ozeki, J., Hashizume, M., Kato, S., Ishihara, H. and Makishima, M. (2012) Vitamin D Receptor Activation Induces Peptide YY Transcription in Pancreatic Islets. *Endocrinology*, **153**, 5188-5199.

[182] Wang, A.P., Li, X., Chao, C., Huang, G., Liu, B.L., Peng, J. and Zhou, Z.G. (2012) 1α,25(OH)(2)D(3) Protects Pancreatic β-Cell Line from Cytokine-Induced Apoptosis and Impaired Insulin Secretion. *National Medical Journal of China*, **92**, 695-699.

[183] Takiishi, T., Gysemans, C., Bouillon, R. and Mathieu, C. (2010) Vitamin D and Diabetes. *Endocrinology Metabolism Clinics of North America*, **39**, 419-446.

[184] Zierold, C., Ming, J.A. and DeLuc, H.F. (2003) Regulation of 25-Hydroxyvitamin D3-24-Hydroxylase mRNA by 1,25-Dihydroxyvitamin D3 and Parathyroid Hormone. *Journal of Cellular Biochemistry*, **88**, 234-237.

[185] Moran-Auth, Y., Penna-Martinez, M., Shoghi, F., Ramos-Lopez, E. and Badenhoop, K. (2013) Vitamin D Status and Gene Transcription in Immune Cells. *Journal*

of Steroid Biochemistry and Molecular Biology, **136**, 83-85. https://doi.org/10.1016/j.jsbmb.2013.02.005

[186] Carlberg, C., Seuter, S. and Heikkinen, S. (2012) The First Genome-Wide View of Vitamin D Receptor Locations and Their Mechanistic Implications. *Anticancer Research*, **32**, 271-282.

[187] Andrukohova, O., Slavic, S., Zeitz, U., Riesen, S.C., Heppelmann, M.S., Ambrisko, T.D., Markovic, M., Kuebler, W.M. and Erben, R.G. (2013) Vitamin D Is a Regulator of Endothelial Nitric Oxide Synthase and Arterial Stiffness in Mice. *Molecular Endocrinology*, **28**, 53-64.

[188] Uberti, F., Lattuada, D., Morsanuto, V., Bolis, G., Vacca, G., Squarzanti, D.F., Cisari, D. and Molinari, C. (2013) Vitamin D Protects Human Endothelial Cells from Oxidative Stress through the Autophagic and Survival Pathways. *Journal of Clinical Endocrinology & Metabolism*, **99**, 1367-1374.

[189] Harrison, M.T. (1964) Interrelationships of Vitamin D and Parathyroid Hormone in Calcium Homeostasis. *Postgraduate Medical Journal*, **40**, 497-505.

[190] Shaota, O., Mundey, M.K., San, P., Godber, I.M., Lawson, N. and Hosking, D.J. (2004) The Relationship between Vitamin D and Parathyroid Hormone: Calcium Homeostasis, Bone Turnover, and Bone Mineral Density in Postmenopausal Women with Established Osteoporosis. *Bone*, **35**, 312-319. https://doi.org/10.1016/j.jsbmb.2006.12.079

[191] Avila, E., Diaz, L., Barrera, D., Halhali, A., Mendez, I., Gonzalez, L., Zuegel, U., Steinmeyer, A. and Larrea, F. (2007) Regulation of Vitamin D Hydroxylases Gene Expression by 1,25-Dihydroxyvitamin D3 and Cyclic AMP in Cultured Human Syncytiotrophoblasts. *Journal of Steroid Biochemistry and Molecular Biology*, **103**, 90-96.

[192] Breslau, N.A. (1988) Normal and Abnormal Regulation of 1,25-(OH)2D Synthesis. *American Journal of the Medical Sciences*, **296**, 417-425.

[193] Kan, P.B., Hirst, M.A. and Feldman, D. (1985) Inhibition of Steroidogenic Cyctochrome P-450 Enzymes in Rat Testis by Ketoconazole and Related Imidazole Anti-Fungal Drugs. *Journal of Steroid Biochemistry*, **23**, 1023-1029.

[194] Karlic, H. and Varga, F. (2011) Impact of Vitamin D Metabolism on Clinical Epigenetics. *Clinical Epigenetics*, **2**, 55-61.

[195] Seuter, S., Heikkinen, S. and Carlberg, C. (2013) Chromatin Acetylation at Transcription Start Sites and Vitamin D Receptor Binding Regions Relates to Effects of 1α,25-Dihydroxyvitamin D3 and Histone Deacetylase Inhibitors on Gene Expression. *Nucleic Acids Research*, **41**, 110-124.

[196] Chow, E.C., Quach, H.P., Vieth, R. and Pang, K.S. (2013) Temporal Changes in Tissue 1α,25-Hydroxyvitamin D3, Vitamin D Receptor Target Genes, and Calcium and PTH Levels after 1,25(OH)2D3 Treatment in Mice. *American Journal of Physiology-Endocrinology and Metabolism*, **304**, E977-E989.

[197] Zannata, L., Goulart, P.B., Goncalves, R., Pierozan, P., Winkelmann-Duarte, E.C., Woehl, V.M., Pessoa-Pureur, R., Silva, F.R. and Zamoner, A. (2012) 1α,25-dihyrdroxyvitamin D_3 Mechanism of Action: Modulation of L-Type Calcium Channels Leading to Calcium Uptake and Intermediate Filament Phosphorylation in Cerebral Cortex of Young Rats. *Biochimica et Biophysica Acta* (*BBA*)—*Molecular Cell Research*, **1823**, 1708-1719.

[198] Wu, Y., Miyamoto, T., Li, K., Nakagomi, H., Sawada, N., Kira, S., Kobayashi, H., Zakohji, H., Tsuchida, T., Fukazwa, M., Araki, I. and Takeda, M. (2011) Decreased Expression of the Epithelial Ca^{2+} Channel TRPV5 and TRPV6 in Human Renal Cell

Carcinoma Associated with Vitamin D Receptor. *Journal of Urology*, **186**, 2419-2425.

[199] Mizwicki, M.T., Keidel, D., Bula, C.M., Bishop, J.E., Zanello, L.P., Wurtz, J.M., Moras, D. and Norman, A.W. (2004) Identification of an Alternative Ligand-Binding Pocket in the Nuclear Vitamin D Receptor and Its Functional Importance in 1Alpha,25(OH)2-Vitmain D3 Signaling. *Proceedings of the National Academy of Sciences of the United States of America*, **101**, 12876-12881.

[200] Kim, Y.S., MacDonald, P.N., Dedhar, S. and Hruska, K.A. (1996) Association of 1α,25-Dihydroxyvitamin D3-Occupied Vitamin D Receptors with Cellular Membrane Acceptance Sites. *Endocrinology*, **137**, 3649-3658.

[201] Carlberg, C. and Molnar, F. (2012) Current Status of Vitamin D Signaling and Its Therapeutic Applications. *Current Topics in Medicinal Chemistry*, **12**, 528-547.

[202] Ene, C.I., Edwards, L., Riddick, G., Baysan, M., Woolard, K., Kotliarova, S., Lai, C., Belova, G., Cam, M., Walling, J., Zhou, M., Stevenson, H., Kim, H.S., Killian, K., Veenstra, T., Bailey, R., Song, H., Zhang, W. and Fine, H.A. (2012) Histone Demethylase Jumonji D3 (*JMJD*3) as a Tumour Suppressor by Regulating p53 Protein Nuclear Stabilization. *PLoS ONE*, **7**, e51407.

[203] Kruidenier, L., Chung, C.W., Cheng, Z., Liddle, J., Che, K., Joberty, G., Bantscheff, M., Bountra, C., Bridges, A., Diallo, H., Eberhard, D., Hutchinson, S., Jones, E., Katso, R., Leveridge, M., Mander, P.K., Mosley, J., Ramirez-Molina, D., Rowland, P., Schofield, C.J., Sheppard, R.J., Smith, J.E., Swales, C., Tanner, R., Thomas, P., Tumber, A., Drewes, G., Oppermann, U., Patel, D.J., Lee, K. and Wislon, D.M. (2012) A Selective Jumonji H3K27 Demethylase Inhibitor Modulates the Proinflammatory Macrophage Response. *Nature*, **488**, 404-408.

[204] Testosterone in Wikipedia.

[205] Griggs, R.C., Kingston, W., Jozefowicz, R.F., Herr, B.E., Forbes, G. and Halliday, D. (1989) Effect of Testosterone on Muscle Mass and Muscle Protein Synthesis. *Journal of Applied Physiology* (1985), **66**, 498-503.

[206] Blomberg Jensen, M. (2012) Vitamin D Metabolism, Sex Hormones, and Male Reproductive Function. *Reproduction*, **144**, 135-152.

[207] Echchgadda, I., Song, C.S., Roy, A.K. and Chatterjee, B. (2004) Dehydroepiandrosterone Sulfotransferase Is a Target for Transcriptional Induction by the Vitamin D Receptor. *Molecular Pharmacology*, **65**, 720-729.

[208] Roselli, C.E., Liu, M. and Hurn, P.D. (2009) Brain Aromatization: Classic Roles and New Perspectives. *Seminars in Reproductive Medicine*, **27**, 207-217.

[209] Beyer, C., Morali, G., Larsson, K. and Sodersteing, P. (1976) Steroid Regulation of Sexual Behaviour. *Journal of Steroid Biochemistry*, **7**, 1171-1176.

[210] Roselli, C.E. and Stormshak, F. (2009) Prenatal Programming of Sexual Partner Preference: The Ram Model. *Journal of Neuroendocrinology*, **21**, 359-364.

[211] Durdiakova, J., Ostatnikova, D. and Celec, P. (2011) Testosterone and Its Metabolites—Modulators of Brain Functions. *Acta Neurobiologiae Experimentalis Journal* (*Wars*), **71**, 434-454.

[212] Ajayi, A.A. and Halushka, P.V. (2005) Castration Reduces Platelet Thromboxane A2 Receptor Density and Aggregability. *QJM*: *An International Journal of Medicine*, **98**, 349-356.

[213] Ajayi, A.A., Mathur, R. and Halushka, P.V. (1995) Testosterone Increases Human Platelet Thromboxane A2 Receptor Density and Aggregation Responses. *Circulation*, **91**, 2742-2747.

[214] Boss, L., Kang, D.H., Marcus, M. and Bergstrom, N. (2013) Endogenous Sex Hormones and Cognitivie Function in Older Adults: A Systematic Review. *Western Journal of Nursing Research*, **36**, 388-426.

[215] Morley, J.E. (2013) Scientific Overview of Hormone Treatment Used for Rejuvenation. *Fertility and Sterility*, **99**, 1807-1813.

[216] Williams, C.L. and Meck, W.H. (1991) The Organizational Effects of Gonadal Steroids on Sexually Dimorphic Spatial Ability. *Psychoneuroendocrinology*, **16**, 155-176.

[217] Tsujimura, A. (2013) The Relationship between Testosterone Deficiency and Men's Health. *The World Journal of Men's Health*, **31**, 126-135.

[218] Moffat, S.D. and Hampson, E. (1996) A Curvilinear Relationship between Testosterone and Spatial Cognition in Humans: Possible Influence of Hand Preference. *Psychoneuroendocrinology*, **21**, 323-337.

[219] Introduction to the Hypothalamo-Pituitary-Adrenal (HPA) Axis. http://biochemistry2.ucsf.edu/programs/ptf/mn%20links/HPA%20Axis%20Physio.pdf

[220] Lee, D.M., Tajar, A., Pye, S.R., Boonen, S., Vanderschueren, D., Bouillon, R., O'Neill, T.W., Bartfai, G., Casanueva, F.F., Finn, J.D., Forti, G., Giwercman, A., Han, T.S., Huhtaniemi, I.T., Kula, K., Lean, M.E., Pendleton, N., Punab, M., Wu, F.C. and EMAS Study Group (2012) Association of Hypogonadism with Vitamin D Status: The European Male Ageing Study. *European Journal of Endocrinology*, **166**, 77-85.

[221] Wehr, E., Pilz, S., Boehm, B.O., Marz, W. and Obermayer-Pietsch, B. (2010) Association of Vitamin D Status with Serum Androgen Levels in Men. *Clinical Endocrinology* (*Oxford*), **73**, 243-248.

[222] Gunnarsson, O., Indridason, O.S., Franzson, L. and Sigurdsson, G. (2009) Factors Associated with Elevated or Blunted PTH Response in Vitamin D Insufficient Adults. *Journal of Internal Medicine*, **265**, 488-495.

[223] Cell Signaling Tutorial—The Biology Project—University of Arizona. www.biology.arizona.edu

[224] Jorde, R., Grimnes, G., Hutchinson, M.S., Kjaergaard, M., Kamycheva, E. and Svartberg, J. (2013) Supplementation with Vitamin D Does Not Increase Serum Testosterone Levels in Healthy Males. *Hormone and Metabolic Research*, **45**, 675-681.

[225] Pilz, S., Frisch, S., Koertke, H., Kuhn, J., Dreier, J., Obermayer-Pietsch, B., Wehr, E. and Zittermann, A. (2011) Effect of Vitamin D Supplementation on Testosterone Levels in Men. *Hormone and Metabolic Research*, **43**, 223-225.

[226] Lerchbaum, E. and Obermayer-Pietsch, B. (2012) Vitamin D and Fertility: A Systemic Review. *European Journal of Endocrinology*, **166**, 765-778.

[227] Nimptsch, K., Platz, E.A., Willett, W.C. and Giovannucci, E. (2012) Asssociation between Plasma 25-OH Vitamin D and Testosterone Levels in Men. *Clinical Endocrinology* (*Oxford*), **77**, 106-112.

[228] Viau, V. and Meaney, M.J. (1996) The Inhibitory Effect of Testosterone on Hypothalamic-Pituitary-Adrenal Responses to Stress Is Mediated by the Medial Preoptic Area. *Journal of Neuroscience*, **16**, 1866-1876.

[229] Scarduzio, M., Panichi, R., Pettorossi, V.E. and Grassi, S. (2013) Synaptic Long-Term Potentiation and Depression in the Rat Medial Vestibular Nuclei Depend on Neural Activation of Estrogenic and Androgenic Signals. *PLoS ONE*, **8**,

e80792.

[230] Caruso, D., Pesaresi, M., Abbiati, F., Calabrese, D., Giatti, S., Garcia-Segura, L.M. and Melcangi, R.C. (2013) Comparison of Plasma and Cerebrospinal Fluid Levels of Neuroactive Steroids with Their Brain, Spinal Cord and Peripheral Nerve Levels in Male and Female Rats. *Psychoneuroendocrinology*, **38**, 2278-2290.

[231] Akinbami, M.A., Philip, G.H., Sridaran, R., Mahesh, V.B. and Mann, D.R. (1999) Expression of mRNA and Proteins for Testicular Steroidogenic Enzymes and Brain and Pituitary mRNA for Glutamate Receptors in Rats Exposed to Immobilization Stress. *Journal of Steroid Biochemistry and Molecular Biology*, **70**, 143-149.

[232] Vincent, K., Warnaby, C., Stagg, C.J., Moore, J., Kennedy, S. and Tracey, I. (2013) Brain Imaging Reveals That Engagement of Descending Inhibitory Pain Pathways in Healthy Women in a Low Endogenous Estradiol State Varies with Testosterone. *PAIN*, **154**, 515-524. https://doi.org/10.1016/j.pain.2012.11.016

[233] Fischer, L., Arthuri, M.T., Torres-Chavez, K.E. and Tambeli, C.H. (2009) Contribution of Endogenous Opioids to Gonadal Hormones-Induced Temporomandibular Joint Antinociception. *Behavioral Neuroscience*, **123**, 1129-1140. https://doi.org/10.1037/a0017063

[234] Sudhakar, H.H. and Venkatesh, D. (2003) Gender Differences in Predator Induced Pain Perception in Rats. *Indian Journal of Experimental Biology*, **41**, 270-272.

[235] Centenera, M.M., Harris, J.M., Tilley, W.D. and Butler, L.M. (2008) The Contribution of Different Androgen Receptor Domains to Receptor Dimerization and Signaling. *Molecular Endocrinology*, **22**, 2373-2382. https://doi.org/10.1210/me.2008-0017

[236] Bialek, M., Zaremba, P., Borowicz, K.K. and Czuczwar, S.J. (2004) Neuroprotective Role of Testosterone in the Nervous System. *Polish Journal of Pharmacology*, **56**, 509-518.

[237] Kinderman, N.B. and Jones, K.J. (1993) Testosterone Enhancement of the Nerve Cell Body Response to Injury: Evidence Using *in Situ* Hybridization and Ribosomal DNA Probes. *Journal of Neuroscience*, **13**, 1523-1532.

[238] Nudler, S.I., Pagani, M.R., Urbano, F.J., McEnery, M.W. and Uchitel, O.D. (2005) Testosterone Modulates Ca(v2.2) Calcium Channels' Functional Expression at Rat Levator Ani Neuromuscular Junction. *Neuroscience*, **134**, 817-826.

[239] Caruso, D., Scurati, S., Roglio, I., Nobbio, L., Schenone, A. and Melcangi, R.C. (2008) Neuroactive Steroid Levels in a Transgeneic Rat Model of CMT1A Neuropathy. *Journal of Molecular Neuroscience*, **34**, 249-253. https://doi.org/10.1007/s12031-007-9029-3

[240] Juerta-Ocampo, I., Mena, F., Barrios, F., Martinez, G., Gonzalez, L. and Larriva-Sahd, J. (2005) Perinatal Exposure to Androgen Suppresses Sexual Dimorphism in Nerve Trunk Diameter, Axon Number, and Fiber Size Spectrum: A Quantitative Ultrastructural Study of the Adult Rate Mammary Nerve. *Brain Research*, **1060**, 179-183. https://doi.org/10.1016/j.brainres.2005.07.051

[241] Vasconsuelo, A., Pronsato, L., Ronda, A.C., Boland, R. and Milanesi, L. (2011) Role of 17β-Estradiol and Testosterone in Apoptosis. *Steroids*, **76**, 1223-1231.

[242] Gonzalez-Montelongo, M.C., Marin, R., Marrero-Alonso, J. and Diaz, M. (2010) Androgens Induce Nongenomic Stimulation of Colonic Contractile Activity through Induction of Calcium Sensitization and Phosphorylation of LC_{20} and CPI_{-17}. *Molecular Endocrinology*, **24**, 1007-1023. https://doi.org/10.1210/me.2009-0472

[243] Zylinska, L., Gromadzinska, E. and Lachowicz, L. (1999) Short-Time Effects of

Neuroactive Steroids on Rat Cortical Ca^{2+}-ATPase Activity. *Biochimica et Biophysica Acta* (*BBA*)—*Molecular and Cell Biology of Lipids*, **1437**, 257-224. https://doi.org/10.1016/S1388-1981(99)00015-3

[244] Liang, R., Liu, X., Wei, L., Wang, W., Zheng, P., Yan, X., Zhao, Y., Liu, L. and Cao, X. (2012) The Modulation of the Excitability of Primary Sensory Neurons by Ca^{2+}-CaM-CaMKII Pathway. *Neurological Sciences*, **33**, 1083-1093. https://doi.org/10.1007/s10072-011-0907-7

[245] Harteneck, C. (2003) Proteins Modulating TRP Channel Function. *Cell Calcium*, **3**, 303-310. https://doi.org/10.1016/S0143-4160(03)00043-5

[246] Hubner, M.R. and Spector, D.L. (2010) Role of H3K27 Demethylase Jmjd3 and UTX in Transcriptional Regulation. *Cold Spring Harbor Symposia on Quantitative Biology*, **75**, 43-49. https://doi.org/10.1101/sqb.2010.75.020

[247] Yang, D., Okamura, H., Nakashima, Y. and Hnaeji, T. (2013) Histone Demethylase Jmjd3 Regulates Osteoblast Differentiation via Transcription Factors Runx2 and Osterix. *Journal of Biological Chemistry*, **288**, 33530-33541.

[248] Estaras, C., Akizu, N., Garcia, A., Beltran, S., de la Cruz, X. and Martinez-Balbas, M.A. (2012) Genome-Wide Analysis Reveals That Smad3 and JMJD3 HDM Co-Activate the Neural Development Program. *Development*, **139**, 2681-2691. https://doi.org/10.1242/dev.078345

[249] Burgold, T., Spreafico, F., De Santa, F., Totaro, M.G., Prosperini, E., Natoli, G. and Testa, G. (2008) The Histone H3 Lysine 27-Specific Demethylase Jmjd3 Is Required for Neural Commitment. *PLoS ONE*, **3**, e3034.

[250] De Santa, F., Totaro, M.G., Prosperini, E., Notarbartolo, S., Testa, G., and Natoli, G. (2007) The Histone H3 Lysine-27 Demethylase Jmjd3 Links Inflammation to Inhibition of Polycomb-Mediated Gene Silencing. *Cell*, **130**, 1083-1094.

[251] Pereira, F., Barbachano, A., Singh, P.K., Campbell, M.J., Munoz, A. and Larriba, M.J. (2012) Vitamin D Has Wide Regulatory Effects on Histone Demethylase Genes. *Cell Cycle*, **11**, 1081-1089. https://doi.org/10.4161/cc.11.6.19508

[252] Pereira, F., Barbachano, A., Silva, J., Bonilla, F., Campbell, M.J., Munoz, A. and Larriba, M.J. (2011) KDM6B/JMJD3 Histone Demethylase Is Induced by Vitamin D Andmodulates Its Effects in Colon Cancer Cells. *Human Molecular Genetics*, **20**, 4655-4665. https://doi.org/10.1093/hmg/ddr399

[253] Przanowski, P., Dabrowski, M., Ellert-Miklaszewska, A., Kloss, M., Mieczkowski, J., Kaza, B., Ronowicz, A., Hu, F., Piotrowski, A., Kettenmann, H., Komorowski, J. and Kaminska, B. (2013) The Signal Transducers Stat 1 and Stat 3 and Their Novel Target Jmjd3 Drive the Expression of Inflammatory Genes in Microglia. *Journal of Molecular Medicine* (*Berlin*), **92**, 239-254.

[254] Busch-Dienstfertig, M., Labuz, D., Wolfram, T., Vogel, N.N. and Stein, C. (2012) JAK-STAT1/3-Induced Expression of Signal Sequence-Encoding Proopiomelanocortin mRNA in Lymphocytes Reduces Inflammatory Pain in Rats. *Molecular Pain*, **8**, 83.

[255] Huang, H.C., Nakatsuka, M. and Iwai, Y. (2013) Activation of Microglial Cells in the Trigeminal Subnucleus Caudalis Evoked by Inflammatory Stimulation of the Oral Mucosa. *Okajimas Folia Anatomica Japonica*, **89**, 137-145.

[256] Del Valle, L., Schwartzman, R.J. and Alexander, G. (2009) Spinal Cord Histopathological Alterations in a Patient with Longstanding Complex Regional Pain Syndrome. *Brain, Behavior, and Immunity*, **23**, 85-91. https://doi.org/10.1016/j.bbi.2008.08.004

[257] Mohr, A., Chatain, N., Domoszlai, T., Rinis, N., Sommerauer, M., Vogt, M. and

Muller-Newen, G. (2012) Dynamics and Non-Canonical Aspects of JAK/STAT Signalling. *European Journal of Cell Biology*, **91**, 524-532.

[258] Aaronson, D.S. and Horvath, C.M. (2002) A Road Map for Those Who Don't Know JAK-STAT. *Science*, **296**, 1653-1655. https://doi.org/10.1126/science.1071545

[259] Pham, D., Yu, Q., Walline, C.C., Muthukrishnan, R., Blum, J.S. and Kaplan, M.H. (2013) Opposing Roles of STAT4 and Dnmt3a in Th1 Gene Regulation. *Journal of Immunology*, **191**, 902-911.

[260] Elgavish, A. (2009) Epigenetic Reprogramming: A Possible Etiological Factor in Bladder Pain Syndrome/Interstitial Cystitis? *Journal of Urology*, **181**, 980-984.

[261] Meyer, M.B., Goetsch, P.D. and Pike, J.W. (2010) A Downstream Intergenic Cluster of Regulatory Enhancers Contributes to the Induction of *CYP*24*A*1 Expression by 1α,25-Dihydroxyvitamin D_3. *Journal of Biological Chemistry*, **285** 15599-15610. https://doi.org/10.1074/jbc.M110.119958

[262] Pike, J.W. and Meyer, M.B. (2012) Regulation of Mouse CYP24a1 Expression via Promoter-Proximal and Downstream-Distal Enhancers Highlights New Concepts of 1,25-Dihydroxyvitamin D(3) Action. *Archives of Biochemistry and Biophysics*, **523**, 2-8.

[263] Jones, G., Prosser, D.E. and Kaufmann, M. (2012) 25-Hydroxyvitamin D-24-Hydroxylase (CYP24a1): Its Important Role in the Degradation of Vitamin D. *Archives of Biochemistry and Biophysics*, **523**, 9-18. https://doi.org/10.1016/j.abb.2011.11.003

[264] Bacchetta, J., Sea, J.L., Chun, R.F., Lisse, T.S., Wesseling-Perry, K., Gales, B., Adams, J.S., Salusky, I.B. and Hewison, M. (2013) Fibroblast Growth Factor 23 Inhibits Extrarenal Synthesis of 1,25-Dihydroxyvitamin D in Human Monocytes. *Journal of Bone and Mineral Research*, **28**, 46-55. https://doi.org/10.1002/jbmr.1740

[265] Hidalgo, A.A., Trump, D.L. and Johnson, C.S. (2010) Glucocorticoid Regulation of the Vitamin D Receptor. *Journal of Steroid Biochemistry and Molecular Biology*, **121**, 372-375.

[266] Choi, M., Yamada, S. and Makishima, M. (2011) Dynamic and Ligand-Selective Interactions of Vitamin D Receptor with Retinoid X Receptor and Cofactors in Living Cells. *Molecular Pharmacology*, **80**, 1147-1155.

[267] CYP17A1—Wikipedia, the Free Encyclopedia. https://en.wikipedia.org/wiki/CYP12A1

[268] Kushida, A. and Tamura, H. (2009) Retinoic Acids Induced Neurosteroid Biosynthesis in Human Glial GI_{-1} Cells via the Induction of Steroidogenic Genes. *Journal of Biochemistry*, **146**, 917-923. https://doi .org/10.1093/jb/mvp142

[269] Diotel, N., Do Rego, J.L., Anglade, I., Vaillant, C., Pellegrini, E., Gueguen, M.M., Mironov, S., Vaudry, H. and Kah, O. (2011) Activity and Expression of Steroidogenic Enzymes in the Brain of Adult Zebrafish. *European Journal of Neuroscience*, **34**, 45-56.

[270] Stojkov, N.J., Janjic, M.M., Baburski, A.Z., Mihajlovic, A.I., Drljaca, D.M., Sokanovic, S.J., Bjelic, M.M., Kostic, T.S. and Andric, S.A. (2013) Sustained *in Vivo* Blockade of α_1-Adrenergic Receptors Prevented Some of Stress-Triggered Effects on Steroidogenic Machinery in Leydig Cells. *American Journal of Physiology-Endocrinology and Metabolism*, **305**, E194-E204.

[271] Garcia-Giralt, N., Rodriguez-Sanz, M., Prieto-Alhambra, D., Servitja, S., Torres-Del Pliego, E., Balcells, S., Albanell, J., Grinberg, D., Diez-Perez, A., Tusquets, I. and Nogues, X. (2013) Genetic Determinants of Aromatase Inhibitor-Related Arthralgia: The B-ABLE Cohort Study. *Breast Cancer Research and Treatment*, **140**,

385-395.

[272] Tague, S.E. and Smith, P.G. (2011) Vitamin D Receptor and Enzyme Expression in Dorsal Root Ganglia of Adult Female Rats: Modulation by Ovarian Hormones. *Journal of Chemical Neuroanatomy*, **41**, 1-12. https://doi.org/10.1016/j.jchemneu.2010.10.001

[273] Calcium-Binding Protein in Wikipedia.

[274] Li, J.L., Li, Y.Q., Li, J.S., Kaneko, T. and Mizuno, N. (1999) Calcium-Binding Protein-Immunoreactive Projection Neurons in the Caudal Subnucleus of the Spinal Trigeminal Nucleus of the Rat. *Neuroscience Research*, **35**, 225-240. https://doi.org/10.1016/S0168-0102(99)00086-3

[275] Li, Y.N., Sakamoto, H., Kawate, T., Cheng, C.X., Li, Y.C., Shimada, O. and Atsumi, S. (2005) An Immunocytochemical Study of Calbindin-D28K in Laminae I and II of the Dorsal Horn and Spinal Ganglia in the Chicken with Special Reference to the Relation to Substance P-Containing Primary Afferent Neurons. *Archives of Histology and Cytology*, **68**, 57-70.

[276] Albuquerque, C., Lee, C.J., Jackson, A.C. and MacDermott, A.B. (1999) Subpopulations of GABAergic and Non-GABAergic Rat Dorsal Horn Neurons Express Ca^{2+}-Permeable AMPA Receptors. *European Journal of Neuroscience*, **11**, 2758-2766. https://doi.org/10.1046/j.1460-9568.1999.00691.x

[277] Yoshida, S., Senba, E., Kubota, Y., Hagihira, S., Yoshiya, I., Emson, P.C. and Tohyama, M. (1990) Calcium-Binding Proteins Calbindin and Parvalbumin in the Superficial Dorsal Horn of the Rat Spinal Cord. *Neuroscience*, **37**, 839-848.

[278] Egea, J., Malmierca, E., Rosa, A.O., del Barrio, L., Negredo, P., Nunez, A. and Lopez, M.G. (2012) Participation of Calbindin-D28K in Nociception: Results from Calbindin-D28K Knockout Mice. *Pflugers Arch*, **463**, 449-458.

[279] Calbindin in Wikipedia.

[280] Christakos, S., Dhawan, P., Peng, X., Obukhov, A.G., Nowycky, M.C., Benn, B.S., Zhong, Y., Liu, Y. and Shen, Q. (2007) New Insights into the Function and Regulation of Vitamin D Target Proteins. *Journal of Steroid Biochemistry and Molecular Biology*, **103**, 405-410.

[281] Matsumoto, K., Ieda, T., Saito, N., Ono, T. and Shimada, K. (1998) Role of Retinoic Acid in Regulation of mRNA Expression of CaBP-D28k in the Cerebellum of the Chicken. *Comparative Biochemistry and Physiology Part A*: *Molecular & Integrative Physiology*, **120**, 237-242.

[282] Hsu, Y.J., Dimke, H., Schoeber, J.P., Hsu, S.C., Lin, S.H., Chu, P., Hoenderop, J.G. and Bindels, R.J. (2010) Testosterone Increases Urinary Calcium Excretion and Inhibits Expression of Renal Calcium Transport Proteins. *Kidney International*, **77**, 601-608.

[283] Antal, M., Petko, M., Polgar, E., Heizmann, C.W. and Storm-Mathisen, J. (1996) Direct Evidence of an Extensive GABAergic Innervation of the Spinal Dorsal Horn by Fibers Descending from the Rostral Ventromedial Medulla. *Neuroscience*, **73**, 509-518.

[284] Ren, K. and Ruda, M.A. (1994) A Comparative Study of the Calcium-Binding Proteins Calbindin-D28K, Calretinin, Calmodulin and Parvalbumin in the Rat Spinal Cord. *Brain Research Reviews*, **19**, 163-179.

[285] Rausell, E., Cusick, C.G., Taub, E. and Jones, E.G. (1992) Chronic Deafferentation in Monkeys Differentially Affects Nociceptive and Non-Nociceptive Pathways Distinguished by Specific Calcium-Binding Proteins and Down-Regulates Gamma-Aminobutyric Acid Type A Receptors at Thalamic Levels. *Proceedings of the*

National Academy of Sciences of the United States of America, **89**, 2571-2575.

[286] Bone Morphogenetic Protein 2 in Wikipedia.

[287] Fu, B., Wang, H., Wang, J., Barouhas, I., Liu, W., Shuboy, A., Bushinsky, D.A., Zhou, D. and Favus, M.J. (2013) Epigenetic Regulation of *BMP*2 by 1,25-Dihydroxyvitamin D_3 through DNA Methylation and Histone Modification. *PLoS ONE*, **8**, e61423.

[288] Lantero, A., Tramullas, M., Diaz, A. and Hurle, M.A. (2011) Transforming Growth Factor-β in Normal Nociceptive Processing and Pathological Pain Models. *Molecular Neurobiology*, **45**, 76-86.

[289] Hall, A.K., Burke, R.M., Anand, M. and Dinsio, K.J. (2002) Activin and Bone Morphogenetic Proteins Are Present in Perinatal Sensory Neuron Target Tissues That Induce Neuropeptides. *Journal of Neurobiology*, **52**, 52-60.

[290] Interleukin 8 Receptor, Beta in Wikipedia.

[291] CXCR2 Chemokine(C-X-C Motif) Receptor 2 [*Homo sapiens* (Human)]. https://www.ncbi.nlm.nih.gov/gene/3579

[292] Zhang, Z.J., Cao, D.L., Zhang, X., Ji, R.R. and Gao, Y.J. (2013) Chemokine Contribution to Neuropathic Pain: Respective Induction of CXCL1 and CXCR2 in Spinal Cord Astrocytes and Neurons. *PAIN*, **154**, 2185-2197.

[293] Dornelles, F.N., Andrade, E.L., Campos, M.M. and Calixto, J.B. (2014) Role of CXCR2 and TRPV1 in Functional, Inflammatory and Behavioural Changes in the Rat Model of Cyclophosphamide-Induced Hemorrhagic Cystitis. *British Journal of Pharmacology*, **171**, 452-467.

[294] Carreira, E.U., Carregaro, V., Teixeria, M.M., Moriconi, A., Aramini, A., Verri Jr., W.A., Ferreira, S.H., Cunha, F.Q. and Cunha, T.M. (2013) Neutrophils Recruited by CXCR1/2 Signalling Mediate Post-Incisional Pain. *European Journal of Pain*, **17**, 654-663.

[295] Sun, Y., Sahbaie, P., Liang, D.Y., Li, W.W., Li, X.Q., Shi, X.Y. and Clark, J.D. (2013) Epigenetic Regulation of Spinal CXCR2 Signaling in Incisional Hypersensitivity in Mice. *Anesthesiology*, **119**, 1198-1208.

[296] Tang, L., Yu, Y., Chen, J., Li, Q., Yan, M. and Guo, Z. (2003) The Inhibitory Effect of VitD3 on Proliferation of Keratinocyte Cell Line HACAT Is Mediated by Down-Regulation of CXCR2 Expression. *Clinical and Experimental Dermatology*, **28**, 416-419.

[297] Homer1 in Wikipedia.

[298] Homer1. Homer Homolog1 (Drosophilia) [*Homo sapiens* (Human)]. https://www.ncbi.nlm.nih.gov/gene/9456

[299] Beqollari, D. and Kammermeier, P.J. (2013) The Interaction between mGluR1 and the Calcium Channel $Cav_{2.1}$ Preserves Coupling in the Presence of Long Homer Proteins. *Neuropharmacology*, **66**, 302-310.

[300] Chen, T., Fei, F., Jiang, X.F., Zhang, L., Qu, Y., Huo, K. and Fei, Z. (2012) Down-Regulation of Homer1b/c Attenuates Glutamate-Mediated Excitotoxicity through endoplasmic Reticulum and Mitochondria Pathways in Rat Cortical Neurons. *Free Radical Biology & Medicine*, **52**, 208-217.

[301] Jardin, I., Lopez, J.J., Berna-Erro, A., Salido, G.M. and Rosado, J.A. (2013) Homer Proteins in Ca^{2+} Entry. *IUBMB Life*, **65**, 497-504

[302] Hayashi, M.K., Tang, C., Verpelli, C., Narayanan, R., Stearns, M.H., Xu, R.M., Li, H., Sala, C. and Hayashi, Y. (2009) The Postsynaptic Density Proteins Homer and Shank form a Polymeric Network Structure. *Cell*, **137**, 159-171.

[303] Metabotropic Glutamate Receptor in Wikipedia.

[304] Zhou, H.Y., Chen, S.R., Chen, H. and Pan, H.L. (2011) Functional Plasticity of Group II Metabotropic Gltutamate Receptors in Regulating Spinal Excitatory and Inhibitory Synaptic Input in Neuropathic Pain. *Journal of Pharmacology and Experimental Therapeutics*, **336**, 254-264.

[305] Jin, Y.H., Takemura, M., Furuyama, A. and Yonehara, N. (2012) Peripheral Glutamate Receptors Are Required for Hyperalgesia Induced by Capsaicin. *Pain Research and Treatment*, **2012**, Article ID: 915706.

[306] Carlton, S.M., Zhou, S., Govea, R. and Du, J. (2011) Group II/III Metabotropic Glutamate Receptors Exert Endogenous Activity-Dependent Modulation of TRPV1 Receptors on Peripheral Nociceptors. *Journal of Neuroscience*, **31**, 12727-12737.

[307] Onozawa, K., Yagasaki, Y., Izawa, Y., Abe, H. and Kawakami, Y. (2011) Amygdala-Prefrontal Pathways and the Dopamine System Affect Nociceptive Responses in the Prefrontal Cortex. *BMC Neuroscience*, **12**, 115.

[308] Chiechio, S., Copani, A., Zammataro, M., Battaglia, G. and Gereau 4th, R.W. and Nicoletti, F. (2010) Transcriptional Regulation of Type-2 Metabotropic Glutamate Receptors: An Epigenetic Path to Novel Treatments for Chronic Pain. *Trends in Pharmacological Sciences*, **31**, 153-160.

[309] Chiechio, S., Zammataro, M., Morales, M.E., Busceti, C.L., Drago, F., Gereau 4th, R.W., Copani, A. and Nicoletti, F. (2009) Epigenetic Modulation of mGlu2 Receptors by Histone Deacetylase Inhibitors in the Treatment of Inflammatory Pain. *Molecular Pharmacology*, **75**, 1014-1020.

[310] Kammermeier, P.J. (2008) Endogenous Homer Proteins Regulate Metabotropic Glutamate Receptor Signaling in Neurons. *Journal of Neuroscience*, **28**, 8560-8567.

[311] Sala, C., Roussignol, G., Meldolesi, J. and Fagni, L. (2005) Key Role of the Postsynaptic Density Scaffold Proteins Shank and Homer in the Functional Architecture of Ca^{2+} Homeostasis at Dendritic Spines in Hippocampal Neurons. *Journal of Neuroscience*, **25**, 4587-4592.

[312] Sala, C., Futai, K., Yamamoto, K., Worley, P.F., Hayashi, Y. and Sheng, M. (2003) Inhibition of Dendritic Spine Morphogenesis and synaptic Transmission by Activity-Inducible Protein Homer1a. *Journal of Neuroscience*, **23**, 6327-6337.

[313] Tappe, A., Klugmann, M., Luo, C., Hirlinger, D., Agarwal, N., Benrath, J., Ehrengruber, M.U., During, M.J. and Kuner, R. (2006) Synaptic Scaffolding Protein Homer1a Protects against Chronic Inflammatory Pain. *Nature Medicine*, **12**, 677-681. https://doi.org/10.1038/nm1406

[314] Hayashi, Y., Okamoto, K., Bosch, M. and Futai, K. (2012) Roles of Neuronal Activity-Induced Gene Products in Hebbian and Homeostatic Synaptic Plasticity, Tagging, and Capture. *Advances in Experimental Medicine and Biology*, **970**, 335-354.

[315] Yuan, J.P., Kiselyov, K., Shin, D.M., Chen, J., Shcheynikov, N., Kang, S.H., Dehoff, M.H., Schwarz, M.K., Saaburg, P.H., Muallem, S. and Worley, P.F. (2003) Homer Binds TRPC Family Channels and Is Required for Gating of TRPC1 by IP3 Receptors. *Cell*, **114**, 777-789.

[316] Biess, A., Korkotian, E. and Holcman, D. (2011) Barriers to Diffusion in Dendrites and Estimation of Calcium Spread Follfowing Synaptic Inputs. *PLoS Computational Biology*, **7**, e1002182.

[317] Lewis, S. (2013) Synaptic Transmission: Keeping Calcium Contained. *Nature Reviews Neuroscience*, **14**, 456.

[318] Magalhaes, A.C., Dunn, H. and Ferguson, S.S. (2012) Regulation of GPCR Activity, Trafficking and Localization by GPCR-Interacting Proteins. *British Journal of Pharmacology*, **165**, 1717-1736.

[319] Gamma-Aminobutyric Acid in Wikipedia, the Free Encyclopedia. https://en.wikipedia.org/wiki/GABA_receptor

[320] GABA Receptor in Wikipedia, the Free Encyclopedia. https://en.wikipedia.org/wiki/GABA_receptor

[321] Adams, D.J. and Berecki, G. (2013) Mechanisms of Conotoxin Inhibition of N-Type (Ca(v)2.2) Calcium Channels. *Biochimica et Biophysica Acta*, **1828**, 1619-1628.

[322] Todorovic, S.M. and Jevtovic-Todorovic, V. (2011) T-Type Voltage-Gated Calcium Channels as Targets for the Development of the Novel Pain Therapies. *British Journal of Pharmacology*, **163**, 484-495.

[323] Sánchez-Blázquez, P., Rodríguez-Díaz, M., DeAntonio, I. and Garzóon, J. (1999) Endomorphin-1 and Endomorphin-2 Show Differences in Their Activation of Mu Opioid Receptor-Requlated G Proteins in Supraspinal Antinociception in Mice. *Journal of Pharmacology and Experimental Therapeutics*, **291**, 12-18.

[324] Mizoguchi, H., Narita, M., Oji, D.E., Suganuma, C., Nagase, H., Sora, I., UhI, G.R., Cheng, E.Y. and Tseng, L.F. (1999) The Mu-Opioid Receptor Gene-Dose Dependent Reductions in G-Protein Activation in the Pons/Medulla and Antinociception Induced by Endomorphins in Mu-Opioid Receptor Knockout Mice. *Neuroscience*, **94**, 203-207.

[325] Lamberts, J.T., Smith, C.E., Li, M.H., Ingram, S.L., Neubig, R.R. and Traynor, J.R. (2013) Differential Control of Opioid Antinociception to Thermal Stimuli in a Knock-In Mouse Expressing Regulator of G-Protein Signaling-Insensitive $G\alpha_o$ Protein. *Journal of Neuroscience*, **33**, 4369-4377.

[326] Chen, Y.J., Huang, C.W., Lin, C.S., Chang, W.H. and Sun, W.H. (2009) Expression and Function of Proton-Sensing G-Protein-Coupled Receptors in Inflammatory Pain. *Molecular Pain*, **5**, 39.

[327] GABA and Glutamate Physiology. www.fsas.org/pdfs/gaba_and_glutamate.pdf

[328] Apostolides, P.F. and Trussell, L.O. (2013) Rapid, Activity-Independent Turnover of Vesicular Transmitter Content at a Mixed Glycine/GABA Synapse. *Journal of Neuroscience*, **33**, 4768-4781.

[329] Wojcik, S.M., Katsurabayashi, S., Guillemin, I., Friauf, E., Rosenmund, C., Brose, N. and Rhee, J.S. (2006) A Shared Vesicular Carrier Allows Synaptic Corelease of GABA and Glycine. *Neurology*, **50**, 575-587.

[330] Yamada, M.H., Nishikawa, K., Kubo, K., Yanagawa, Y. and Saito, S. (2012) Impaired Glycinergic Synaptic Transmission and Enhanced Inflammatory Pain in Mice with Reduced Expression of Vesicular GABA Transporter (VGAT). *Molecular Pharmacology*, **81**, 610-619.

[331] Wlodarczyk, A.I., Sylantyev, S., Herd, M.B., Kersanté, F., Lambert, J.J., Rusakov, D.A., Linthorst, A.C., Semyanov, A., Belelli, D., Pavlov, I. and Walker, M.C. (2013) GABA-Independent GABAA Receptor Openings Maintain Tonic Currents. *Journal of Neuroscience*, **33**, 3905-3914.

[332] Herb, M.B., Belelli, D. and Lambert, J.J. (2007) Neurosteroid Modulation of Synaptic and Extrasynaptic GABA(A) Receptors. *Pharmacology & Therapeutics*, **116**, 20-34.

[333] Nani, F., Bright, D.P., Revilla-Sanchez, R., Tretter, V., Moss, S.J. and Smart, T.G. (2013) Tyrosine Phosphorylation of $GABA_A$ Receptor γ2-Subunit Regulates Tonic and Phasic Inhibition in the Thalamus. *Journal of Neuroscience*, **33**, 12718-12727.

[334] Mitchell, E.A., Herd, M.B., Grunn, B.G., Lambert, J.J. and Belelli, D. (2008) Neurosteroid Modulation of GABAA Receptors: Molecular Determinants and Signific-

ance in Health and Disease. *Neurochemistry International*, **52**, 588-595.

[335] Vithlani, M. and Moss, S.J. (2009) The Role of $GABA_AR$ Phosphorylation in the Construction of Inhibitory Synapses and the Efficacy of Neuronal Inhibition. *Biochemical Society Transactions*, **37**, 1355-1358.

[336] Wang, M. (2011) Neurosteroids and GABA-A Receptor Function. *Frontiers in Endocrinology* (*Lausanne*), **2**, 44.

[337] Ueda, H., Inoue, M., Yoshida, A., Mizuno, K., Yamamoto, H., Maruno, J., Matsuno, K. and Mita, S. (2001) Metabotropic Neurosteroid/Sigma-Receptor Involved in Stimulation of Nociceptor Endings in Mice. *Journal of Pharmacology and Experimental Therapeutics*, **298**, 703-710.

[338] Schlichter, R., Keller, A.F., De Roo, M., Berton, J.D., Inquimbert, P. and Poisbeau, P. (2006) Fast Nongenomic Effects of Steroids on Synaptic Transmission and Role of Endogenous Neurosteroids in Spinal Pain Pathways. *Journal of Molecular Neuroscience*, **28**, 33-51.

[339] Poisbeau, P., Patte-Mensah, C., Keller, A.F., Barrot, M., Breton, J.D., Luis-Delgado, O.E., Freund-Mercier, M.J., Mensah-Nyagan, A.G. and Schlichter, R. (2005) Inflammatory Pain Upregulates Spinal Inhibition via Endogenous Neurosteroid Production. *Journal of Neuroscience*, **25**, 11768-11776.

[340] Mason, J.E. (2010) Pain: Sex Differences and Implications for Treatment. *Metabolism*, **59**, S16-S20.

[341] Traub, R.J. and Ji, Y. (2013) Sex Differences and Hormonal Modulation of Deep Tissue Pain. *Frontiers in Neuroendocrinology*, **34**, 350-366.

[342] Straube, S., Derry, S., Moore, R.A. and McQuay, H.J. (2010) Vitamin D for the Treatment of Chronic Painful Conditions in Adults. *The Cochrane Database of Systematic Reviews*, No. 1, CD007771.

[343] Chlebowski, R.T., Pettinger, M., Johnson, K.C., Wallace, R., Womack, C., Mossacar-Rahmani, Y., Stefanick, M., Wactawski-Wende, J., Carbone, L., Lu, B., Eaton C., Walitt, B. and Kooperberg, C.L. (2013) Calcium plus Vitamin D Supplementation and Joint Symptoms in Postmenopausal Women in the Women's Health Initiative Randomized Trial. *Journal of the Academy of Nutrition and Dietetics*, **113**, 1302-1310.

[344] Catalano, A., Morabito, N., Atteritano, M., Basile, G., Cucinotta, D. and Lasco, A. (2012) Vitamin D Reduces Musculoskeletal Pain after Infusion of Zoledronic Acid for Postmenopausal Osteoporosis. *Calcified Tissue International*, **90**, 279-285.

[345] Gopinath, K. and Danda, D. (2011) Supplementation of 1,25 Dihydroxy Vitamin D3 in Patients with Treatment Naïve Early Rheumatoid Arthritis: A Randomized Controlled Trail. *International Journal of Rheumatic Diseases*, **14**, 332-339.

[346] Osunkwo, I., Ziegler, T.R., Alvarez, J., McCraken, C., Cherry, K., Osunkwo, C.E., Ofori-Acquah, S.F., Ghosh, S., Ogunbobode, A., Rhodes, J., Eckman, J.R., Dampier, C. and Tangpricha, V. (2012) High Dose Vitamin D Therapy for Chronic Pain in Children and Adolescents with Sickle Cell Disease: Results of a Randomized Double Blind Pilot Study. *British Journal of Haematology*, **159**, 211-215.

[347] Cao, Y., Jones, G., Cicuttini, F., Winzenberg, T., Wluka, A., Sharman, J., Nguo, K. and Ding, C. (2012) Vitamin D Supplementation in the Managent of Knee Osteoarthritis: Study Protocol for a Randomized Controlled Trail. *Trails*, **13**, 131.

[348] Kim, T.H., Lee, B.H., Lee, H.M., Lee, S.H., Park, J.O., Kim, H.S., Kim, S.W. and Moon, S.H. (2013) Prevalence of Vitamin D Deficiency in Patients with Lumbar Spinal Stenosis and Its Relationship with Pain. *Pain Physician*, **16**, 165-176.

[349] Glover, T.L., Goodin, B.R., Horgas, A.L., Kindler, L.L., King, C.D., Sibille, K.T., Peloquin, C.A., Riley 3rd, J.L., Staud, R., Bradley, L.A. and Fillingin, R.B. (2012) Vitamin D Race and Experimental Pain Sensitivity in Older Adults with Knee Osteoarthritis. *Arthritis & Rheumatology*, **64**, 3926-3935.