SOME ASPECTS OF EPILEPSY

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Epilepsy and Bone Metabolism

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Abstract

Epilepsy is one of the major causes of morbidity in India. Epilepsy is itself a neurological disorder but it has a major impact on bone health either directly or through adverse effects of antiepileptic drugs through various mechanisms. Social stigma is the most commonly experienced adverse effect by the patients with epilepsy around the world. It can affect people economically, socially and culturally. Bone metabolism is altered in such patients mostly due its effect on vitamin D and related hormones. Fractures resulting from reduction in bone mineral density are an important cause of morbidity and mortality in these patients. Antiepileptic drugs also produce an adverse effect on bone metabolism.

Keywords: Epilepsy; Bone metabolism; Anti-epileptic drugs.

Introduction

Epilepsy is defined as a group of neurological disorders characterized by any of the following conditions:

- At least two unprovoked (or reflex) seizures occurring greater than 24 hours apart.
- One unprovoked (or reflex) seizure and a probability of further seizures similar to the general recurrence risk (at least 60%) after two unprovoked seizures, occurring over the next 10 years.
- Diagnosis of an epilepsy syndrome.

Two or more unprovoked seizures may be defined as epilepsy and as per this definition, incidence of epilepsy is 0.3-0.5% in different population throughout the world and prevalence has been estimated as 5-10 person per 1000 [1]. However in India incidence has been estimated as 38-49.3 per 100,000 populations per year from two community based studies and prevalence stands at around 5 per thousand populations [2].

Epilepsy is considered to be resolved for individuals who had an age-dependent epilepsy syndrome but are now past that age or those who have remained seizure-free for the last 10 years, with no seizure medicines for the last 5 years. This is considered as one of the most common serious neurological disorder. In India, 90% of the estimated 5.5 million people with epilepsy are from rural population and around three fourth of them are deprived of treatment. Incidence of epilepsy is nearly half a million each year, which is clearly more than its prevalence. The lifetime risk of seizure is around 5% and the incidence tends to be higher at the extremes of age. Prevalence may be higher in developing countries because of the prevalence of parasitic illness such as cysticercosis [3].

Epileptogenesis means transformation of normal neuronal network into one that is chronically hyperexcitable. Evidences in epilepsy research have shown some genetic mutations affecting ion channel function and chromosomal microdeletions.
Thus, there is relatively long lasting depolarization of neuronal membrane due to influx of extracellular Calcium (Ca\(^{2+}\)) which leads to opening of voltage gated Sodium (Na\(^{+}\)) channels causing influx of sodium which makes neurons hyperexcitable due to release of glutamate [1].

Social stigma is the most commonly experienced adverse effect by the patients with epilepsy around the world. It can affect people economically, socially and culturally. Fractures resulting from reduction in bone mineral density are an important cause of morbidity and mortality in these patients. The costs of these fractures are tremendous including loss of daily functioning and wages, visits to doctors/ hospitals, hospitalizations and visits to physiotherapists.

The World Health Organization (WHO) has recognized this growing concern and developed an initiative to expand awareness and research in this field. Epilepsy is controllable, but not cured, with medication in about 70 % of cases.

Epilepsy can occur as a result of some genetic and acquired causes or there may be an interaction between the two. Some of the acquired causes include serious brain trauma, stroke, tumors and problems in the brain due to infections.

### Pathophysiology of epilepsy

For normal brain functioning, a balance between the excitatory signals (mediated by glutamate, aspartate) and inhibitory signals (mediated by \(\gamma\)-Amino Butyric Acid [GABA]) is required. The inhibitory Neurotransmitter (NT), GABA acts on ion channels and enhances chloride inflow leading to reduced action potential generation. The excitatory NTs act on sodium and calcium channels producing the opposite effect. This imbalance is the likely cause of seizure episodes [3].

### Seizure initiation and propagation

Initiation phase is characterized by high frequency bursts of action potentials and hyper synchronisation. This is due to long lasting depolarization of neuronal membrane due to influx of extracellular Calcium (Ca\(^{2+}\)), leading to voltage gated Sodium ion (Na\(^{+}\)) channel opening, and generation of repetitive action potentials. Then hyperpolarization occurs due to GABA receptors or Potassium ion (K\(^{+}\)) channels.

### Causes of epilepsy

- **Idiopathic**
- Genetic causes-tuberous Sclerosis, von-Hippel Landau Disease, neurofibromatosis
- Infantile hemiplegia
- Dysembyronic-cortical dysgenesis and sturge-weber Syndrome
- Mesial temporal sclerosis
- Cerebrovascular diseases
- Tumors
- Trauma
- Infections-cerebral abscess, toxoplasmosis, cysticercosis, tuberculoma. encephalitis
- Inflammatory-sarcoidosis, vasculitis
- Generalization of focal seizures
- Genetic causes-inborn errors of metabolism, storage diseases
- Cerebral birth injury
- Hydrocephalous
- Certain Drugs-some antimalarials, antibiotics, antiarrythmics, amphetamines
- Alcohol or its withdrawal
- Toxins
- Metabolic diseases-hypocalcemia, hyponatremia, hypoglycemia, hypagnesemia, liver and renal failure.

### Genetic basis of epilepsy

Almost all of the mutant genes encode voltage or ligand gated ion channels which are responsible for most of the inherited forms of epilepsies. Mutations have been identified in Na\(^{+}\), K\(^{+}\), Ca\(^{2+}\), chloride channels and in channels gated by GABA, acetylcholine, and most recently, in intracellular calcium release channels, Ryanodine Receptors (RYR\(_{2}\)) activated by calcium [4].

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**Table 1:** Classification of seizures (ILAE classification, 2010) [3].

<table>
<thead>
<tr>
<th>Classification of seizures</th>
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<tbody>
<tr>
<td><strong>A. Generalized seizures</strong></td>
</tr>
<tr>
<td>a) Tonic Clonic (in any combination)</td>
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<tr>
<td>b) Absence-</td>
</tr>
<tr>
<td>i. Typical</td>
</tr>
<tr>
<td>ii. Atypical</td>
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<tr>
<td>iii. Absence with special features</td>
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<tr>
<td>c) Myoclonic absence</td>
</tr>
<tr>
<td>d) Eyelid myoclonia</td>
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<tr>
<td>e) Myoclonic-</td>
</tr>
<tr>
<td>i. Myoclonic</td>
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<tr>
<td>ii. Myoclonic atonic</td>
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<tr>
<td>iii. Myoclonic tonic</td>
</tr>
<tr>
<td>f) Clonic</td>
</tr>
<tr>
<td>g) Tonic</td>
</tr>
<tr>
<td>h) Atonic</td>
</tr>
<tr>
<td><strong>B. Focal seizures</strong></td>
</tr>
<tr>
<td>a) Without impairment of consciousness or awareness (was ‘simple partial’)</td>
</tr>
<tr>
<td>i. Focal motor</td>
</tr>
<tr>
<td>ii. Focal sensory</td>
</tr>
<tr>
<td>b) With impairment of consciousness or awareness (was ‘complex partial’)</td>
</tr>
<tr>
<td>c) Evolving to a bilateral, convulsive seizures (was ‘secondarily generalized seizures’)</td>
</tr>
<tr>
<td>i. Tonic</td>
</tr>
<tr>
<td>ii. Clonic</td>
</tr>
<tr>
<td>iii. Tonic-clonic</td>
</tr>
<tr>
<td>C. Unknown</td>
</tr>
<tr>
<td>i. Epileptic spasms</td>
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Some Aspects of Epilepsy

Some molecular studies in animals have uncovered a number of genetic abnormalities that predispose to seizure activity. Mice harboring a mutation in the gene encoding the α1A-subunit of the voltage-dependent calcium channel are seizure prone. Similarly, a mutation in the sodium-hydrogen exchanger (Nhe-1) gene, which encodes a sodium-hydrogen exchanger, is responsible for seizures in the slow-wave epilepsy mutant mouse. The Nhe-1 exchanger has a prominent homeostatic role in virtually all cells of the Central Nervous System (CNS) by extruding a hydrogen ion in exchange for sodium, thereby maintaining intracellular pH and cell volume. Molecular genetic studies of human epilepsy have unveiled a number of inherited defects, predominantly in genes coding ion channel subunits. In the absence of epilepsy, mutations or polymorphisms in these or other human genes that regulate neuronal homeostasis may produce a subclinical predisposition to seizures that may only manifest during the stress of an acute illness.

**Diagnosis of epilepsy**

**General scheme to investigate a patient with epilepsy includes**

- EEG-standard, sleep or EEG with special electrodes in foramen ovale or subdural.
- To find out structural lesion - Computerized Tomography (CT scan) or Magnetic Resonance Imaging (MRI).
- For metabolic abnormality - biochemical tests for urea and electrolytes, blood glucose, liver function tests, calcium and magnesium.
- Inflammatory or infective disorder - full blood count, erythrocyte sedimentation rate, C-reactive protein, chest X-ray, Cerebrospinal Fluid (CSF) examination and serology for infections.
- Ambulatory EEG or Video telemetry.

**Management of epilepsy**

1. First aid

2. Lifestyle modification: Triggering factors for seizures should be identified and patients should be motivated to avoid such precipitating factors. Some of the precipitating factors include sleep deprivation, alcohol use or withdrawal, flickering lights, drugs misuse, music, reading and hot baths.

3. Dietary modification: The Ketogenic Diet (KD) is a high-fat, moderate-protein, low-carbohydrate diet earlier used to treat intractable epilepsy, primarily in the pediatric population. But later on KD was used to treat intractable epilepsies in all age groups and has gained popularity worldwide, with many centers having started their own programs. The classic KD is a 4:1 or 3:1 ratio of fat to combined protein and carbohydrate, and is individually modified to typically provide 75% to 80% of the recommended daily allowance of calories and 80% of estimated daily allowance of fluids. In large studies, the efficacy of the diet is independent of the type of seizure and has been effective for both generalized and partial seizures.

4. Surgery

5. Other treatment modalities: Vagal nerve stimulation or deep brain stimulation.

6. Antiepileptic drugs act: Medical management is the mainstay of treatment of epilepsy. Antiepileptic drugs acts through modification of activity of ion channels or neurotransmitters. The first AED used was bromide in 19th century. Later on, phenobarbitalone was introduced which was first synthesized organic agent for epilepsy treatment.

To minimize toxicity, treatment with a single drug should be preferred. Multiple therapies are needed when same patient develops 2 or more types of seizures or some complication like status epilepticus.

Phenytoin and carbamazepine are the two most commonly prescribed drugs in epilepsy and are more frequently associated with altered bone health including fractures, bone demineralization and reduced bone formation. Phenytoin also causes bone loss due to elevated bone resorption.

Toxic effects of chronic therapy are primarily dose related Central Nervous System (CNS) effects, behavioral changes, increased seizure frequency, gastrointestinal symptoms, gingival hyperplasia, osteomalacia and megaloblastic anemia. These can be controlled with proper dose adjustments. But there are some serious side effects on skin, bone marrow, liver that might be due to serious drug allergy. Endocrine side effects include inhibition of Antidiuretic Hormone (ADH) in patients of inappropriate ADH secretion and inhibition of insulin secretion leading to hyperglycemia and glycosuria.

Osteomalacia, with hypocalcemia and elevated alkaline phosphatase activity due to altered metabolism of vitamin D and defective calcium absorption are the common side effects. AEDs also reduce the concentrations of vitamin K dependent protein by increasing the metabolism of vitamin K leading to defective calcium metabolism in bone. Hypersensitivity reactions, systemic lupus erythematosus, fatal hepatic necrosis, neutropenia, leucopenia, red-cell aplasia, and lymphadenopathy are some of the rare side effects. There is some similarity between the genetic origin of the Na+/Ca²⁺ exchanger in brain and intestine, and phenytoin being a sodium-channel inhibitor can disturb the normal exchange of these ions, hence, leading to decreased calcium absorption in intestine.

Therefore, monitoring of serum AEDs levels is very important for proper dosing schedule. Serum drug levels measure the total drug, however, it is the amount of free drug that reflects the extracellular levels in brain and is related to the efficacy of the drug. Thus, patients with renal diseases or with impaired liver function who have low albumin levels show an increased ratio of free to bound drug, requiring drug dose modification titrated to clinical symptoms.

**Bone metabolism**

Bone is a dynamic tissue that is under continuous turnover or remodeling throughout life. Osteoclasts and osteoblasts are the two types of bone cells located on bone surface and are responsible for bone resorption and formation respectively.

Osteoblasts form bone by synthesizing the organic matrix, including type I collagen, and also synthesize new bone matrix by mineralization. The organic matrix is mineralized by inorganic calcium and phosphate with small amounts of carbonate, magnesium, sodium and potassium. Around 10-30% of the skeleton is remodeled each year, influenced mainly by PTH and 1,25 dihydroxy vitamin D.

On the other hand, osteoclast resorbs bone by secreting a mixture of acid and neutral proteases that acts sequentially to degrade collagen fibrils into small molecular weight fragments.
The active form of vitamin D, calcitriol, initiates biological responses via binding to the VDR. When ligand binds to VDR, it interacts with the Retinoid X Receptor (RXR) to form a heterodimer which then binds to VDREs in the region of genes which are directly controlled by vitamin D. Due to this binding, some conformational changes occur in the position of H12 at the C terminus of VDR which is responsible for binding either coactivators or corepressors. Ligand-activated VDR-RXR modulates the transcription of genes encoding proteins that are responsible for various actions of vitamin D which includes signalling intestinal calcium and phosphate absorption to affect skeletal and calcium homeostasis. Major biological networks influenced by VDR are bone, minerals, detoxification process, cell life (proliferation, differentiation, migration, and death), immune system, and metabolism (amino acid, lipid, and carbohydrate).

**Action of vitamin D on bone**

Ligand-VDR binding regulates the expression of genes in bone cells, that codes for some bone remodelling effectors which can either be catabolic or anabolic. Osteocytes and osteoblasts respond to this complex with bone cells expressing and releasing fibroblast growth factor 23 (FGF23) to control phosphate levels repressing CYP27B1 and inducing CYP24A1 for feedback reduction in 1,25 (OH)D levels. These complexes also enhance expression of Receptor Activator Nuclear factor-κ B Ligand (RANKL) to stimulate bone resorption by osteoclastogenesis. Osteoprotegerin, which is a decoy receptor for RANKL is repressed by vitamin D in osteoblasts. Vitamin D also represses Runx2 expression, thus, blunting the osteoblastic differentiation through the Bone Morphogenetic Protein (BMP) pathway. At higher concentrations, vitamin D predisposes bone towards resorption while at physiological concentrations chances of bone formation are greater [9].

**Mechanism of action of AEDs on vitamin D**

Antiepileptic drugs, particularly cytochrome P450 enzyme inducers like phenytoin, carbamazepine and phenobarbital can interfere in the balance between 25 (OH)D and 1,25 dihydroxy vitamin D through the activation of Pregnancy X Receptors (PXR) also called as Steroid and Xenobiotic Receptor (SXR). In 1998, PXR of mouse was first identified as a member of Nuclear Receptors (NR) super family on the basis of sequence homology. PXR is an intracellular receptor which resembles around 60% to DNA binding domain of VDR, present in gastrointestinal tract, kidneys and liver. Thus instead of VDR, PXR binds to VDRE at DNA and thereby affects the gene expression which is normally regulated by Vitamin D [9].

One study found that xenobiotics through PXR upregulate 24-hydroxylases, and induce cytochrome P450 enzymes which are involved in biotransformation of numerous active substances (CYP2C9, CYP3A4). This catalyzes the conversion of active vitamin D to more polar inactive metabolite. Some investigators found that xenobiotic activation of PXR did not upregulate CYP24 but increase the expression of a different isoenzyme, CYP3A4, in liver and small intestine [9].

**Mechanism of action of vitamin D**

Vitamin D hormone functions through a Vitamin D Receptor (VDR), a 427 amino acid peptide, which is a member of class II steroid hormones and is closely related to retinoic acid and thyroid hormone receptor. It has got a C-domain, E-domain which is ligand binding domain, and an activating F-domain. This receptor acts through VDREs (vitamin D responsive elements) which are repeat sequences of 6 nucleotides separated by 3 specified bases. The S’ arm of this sequence binds the retinoic acid X receptor and 3’ end binds to VDR. Action of 1,25 (OH)D$_3$ is mediated through binding to this nuclear receptor, which then regulates transcription of DNA to RNA [8].

**Antiepileptic drugs, particularly**

**Some Aspects of Epilepsy**

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Vitamin D and calcium metabolism

Vitamin D has a major role in mineral ion homeostasis. Several studies suggest that vitamin D deficiency is the most common cause of hypocalcaemia in primary care depending on population demographics. It may have prevalence as high as 50% [10].

Various proposed mechanisms are:

1. Vitamin D induces calbindin 9K, which is a calcium binding protein in intestine which helps in transport of calcium across the enterocytes. In small intestine, vitamin D also induces TRPV5 and TRPV6 (transient potential receptor vanilloid) genes resulting in improved calcium absorption efficiency [11].

2. VDRs are also present on osteoblasts and regulate several genes including bone matrix, osteocalcin and osteopontin which are up regulated and also down regulate type I collagen. Both vitamin D and PTH induce RANK ligand promoting osteoclastic differentiation, hence stimulating osteoclasto genesis and activating resting osteoclasts for bone resorption which mobilizes calcium from bone [11].

3. The distal renal tubule is responsible for reabsorption of the 1% of the filtered load of calcium. Vitamin D acts on receptors in Distal Convoluted Tubule (DCT) and stimulates calcium absorption.

Hypocalcemia and seizures

Extracellular calcium levels have been found to have an inverse relationship with neuronal excitability.

- External Ca2+ inhibits small conducting Na+ channel leaks (NALCNs), thereby shifting the voltage dependency of voltage gated Na+ channels and stabilizing Cyclic Nucleotide-Gated ion channels (CNG).
- Also external Ca2+ reduces inward current through Alpha-Amino-3-hyroxy-5-methyl-4-isoxazolepropionic Acid (AMPA) channels and depresses the release of excitatory Neurotransmitters (NT).
- It can affect potassium channels and potentiate GABA sensitivity.

These all responses are mediated by either calcium influx or some calcium sensing receptors.

Thus, reduced calcium levels can also exacerbate seizure episodes and thereby may worsen the condition. This develops into a vicious circle, leading on to prescription of higher doses of drug.

Bone mineral density (BMD)

The diagnosis of osteoporosis or AED-induced osteopenia is based on measurement of bone mineral density measured by Dual-Energy X-ray Absorptiometry (DEXA), single energy X-ray absorpiometry, quantitatative CT and an ultrasound. The gold standard for BMD is DEXA scan with 99% accuracy. It can be measured at any site like calcaneum, forearm, fingers but the most common sites are lumbar spine and hip. Here, two X-ray energies are used to estimate the area of mineralized tissue, and the mineral content is divided by the area, which partially corrects for body size. It is a two dimensional technique so can not estimate the depth or posteroanterior length of the bone [11]. Results are expressed in the form of T-score and Z-score.

CT scan can also be used to assess BMD but this technique is more expensive, involves greater radiation exposure and is less reproducible than DEXA. Ultrasound and high resolution CT scan can also be considered for assessing BMD. The hip is the most preferred site in most individuals as it predicts the risk of hip fracture which is the most common complication of osteoporosis. But in young individuals, spine measurement may be the most sensitive indicator of bone loss.

Status of vitamin D and bone health in epilepsy

There are certain factors which contribute to vitamin D deficiency and bone depletion in patients with epilepsy other than antiepileptic drugs. It has been suggested that having epilepsy promotes sedentary lifestyles, low self-esteem due to social stigma and hence isolation. Due to frequent episodes of seizures and a myth that physical exercise may provoke seizures, patients may not go outside and have inadequate physical activity. Hence, these patients tend to lack sun exposure which immensely affects their vitamin D synthesis.

Patients with epilepsy perse and due to other side effects of the drug have already an increased propensity to fractures due to ataxia, coexisting neurological deficits and seizure related falls.

AEDs effects on vitamin D, calcium and bone markers

- Bone loss as ranges from the ability of enzyme inducers to increase vitamin D catabolism and impairment of calcium absorption from gastrointestinal tract to phenytoin’s direct effect on bone-forming osteoblasts via a biphasic mechanism, as low doses seem to stimulate and high therapeutic doses to inhibit osteoblast proliferation and differentiation.BMD decreases after around 1 year of phenytoin therapy but it has no significant correlation with serum phenytoin levels or calcium levels.

- Changes in serum total ALP and its isoenzymes, calcium, inorganic phosphorus, vitamin D, magnesium, total proteins, albumin, urine calcium and phosphorus levels occur in ambulatory patients having adequate sun exposure with new onset epilepsy on phenytoin or valproic acid monotherapy. Calcium metabolism is deranged involving the bone as early as 2 months after starting AED monotherapy, indicating predisposition to development of osteomalacia in these patients. Urinary calcium, tartarate resistant acid phosphatase and urinary calcium/kg/BW also got reduced after 6 months.

- The exact mechanism by which AEDs induce bone loss is not fully understood. Phenytoin and carbamazepine may inhibit calcium transport from apical to basolateral side of Caco 2 cells under physiologic calcium conditions in a dose related manner. This effect is supported by effect of AEDs on vitamin D levels. There is decrease of 40%-50% in intestinal permeability to calcium with therapeutically relevant levels of phenytoin and carbamazepine. Decreased
serum calcium leads to a cascade of events to move calcium from bone into plasma. Caco-2 cells are cultured human colon adenocarcinoma cells which can be used as a model to study the transport of calcium, including effect of vitamin D on calcium absorption, and also quantitation of drug transport and calculation of drug permeability.

➢ In patients with epilepsy receiving chronic anticonvulsant therapy, urinary deoxypyridinoline levels of male patients can be increased significantly along with reduction in vitamin D levels which may be due to the fact that only resorption phase of bone turnover is affected due to chronic therapy with AEDs.

➢ Carbamazepine, which is also an enzyme inducer, causes a significant decrease in bone mineral density along with decreased vitamin D levels while valproic acid and lamotrigine increases osteocalcin, a bone formation marker.

➢ Cytochrome P450 enzyme inducing AEDs are most commonly associated with a negative impact on bone. They up-regulate the enzymes responsible for vitamin D metabolism, thus, resulting in a decrease in vitamin D levels leading to reduced calcium absorption, with consecutive secondary hyperparathyroidism, increased bone resorption and accelerated bone loss [13].

➢ Carbamazepine, phenytoin, phenobarbitone, primidone activate nuclear receptor pregnane X, which in turn, leads to up regulation of cytochrome P (CYP24) gene expression, further leading to raised degradation of 25 hydroxycalciferol and 1,25 dihydroxycholecalciferol and thus, low vitamin D levels which, in turn, may lead to calcium deficiency.

➢ Serum concentrations of γ-glutamyl transpeptidase, 5-nucleotidase, and leucine aminopeptidase can also be raised in children, suggesting that, this might be caused by drug induction of membrane bound hepatic enzymes.

References