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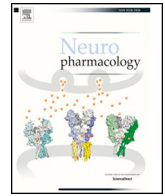
Invited review

Modifying genetic epilepsies – Results from studies on tuberous sclerosis complex

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HIGHLIGHTS

- Epileptogenesis may be modified in animal models and patients.
- Prevention of epilepsy is particularly feasible in tuberous sclerosis complex.
- Clinical trials in TSC patients suggest vigabatrin may have antiepileptogenic effects.
- TSC mouse models indicate mTOR inhibitors have strong antiepileptogenic potential.

ARTICLE INFO

ABSTRACT

Tuberous sclerosis complex (TSC) is an autosomal dominant neurocutaneous disorder affecting approximately 1 in 6,000 in general population and represents one of the most common genetic causes of epilepsy. Epilepsy affects 90% of the patients and appears in the first 2 years of life in the majority of them. Early onset of epilepsy in the first year of life is associated with high risk of cognitive decline and neuropsychiatric problems including autism.

Recently TSC has been recognized as a model of genetic epilepsies. TSC is a genetic condition with known dysregulated mTOR pathway and is increasingly viewed as a model for human epileptogenesis. Moreover, TSC is characterized by a hyperactivation of mTOR (mammalian target of rapamycin) pathway, and mTOR activation was showed to be implicated in epileptogenesis in many animal models and human epilepsies. Recently published studies documented positive effect of preventive or disease modifying treatment of epilepsy in infants with high risk of epilepsy with significantly lower incidence of epilepsy and better cognitive outcome. Further studies on preventive treatment of epilepsy in other genetic epilepsies of early childhood are considered.

This article is part of the special issue entitled 'New Epilepsy Therapies for the 21st Century – From Antiseizure Drugs to Prevention, Modification and Cure of Epilepsy'.

1. Introduction

Epilepsy is one of the few brain diseases known to man in which people at risk may be identified, but there is no prophylactic treatment to prevent the development of epilepsy in those at risk. The current medical therapy for the majority of epilepsies aims to reduce the likelihood of seizure recurrence rather than to reverse the underlying cause of the epilepsy. This approach results in seizure freedom only in about two-thirds of the patients while 30% of patients suffer from drug-resistant seizures.

Unsatisfactory results of standard antiepileptic medication have led researchers to the search for new therapeutic approaches and to novel definitions of treatments: 1) disease modifying treatment, delivered to patients after seizures, but based on the identification of the new genes

and precision medicine aimed to reduce seizure burden and its co-morbidities by reversing or slowing the progression of the underlying disease, and 2) antiepileptogenic treatment based on early identification of patients with high risk of epilepsy leading to sustained prevention of seizure development.

Epileptic encephalopathies of early childhood compose a specific group of conditions characterized mostly by genetic origin, early presentation of seizures and their strong impact on cognitive development. Early diagnosis of the underlying disease may allow therapeutic intervention before clinical seizures, ideally reducing the risk of epilepsy and avoiding unnecessary epileptic comorbidities.

In this review we will describe a current status of knowledge regarding the use of an antiepileptogenic approach in one of epileptic encephalopathies - tuberous sclerosis complex (TSC). The NIH Workshop on Antiepileptogenic therapy and Disease modification held in Washington in 2018 considered TSC as one of the most appropriate conditions for developing this kind of therapy.

2. Concept of epilepsy modification and first clinical applications

In 1881 Gowers (1881) first noted that there is often a seizure-free period lasting several months to years between brain insults and the onset of symptomatic epilepsy. This interval, between brain injury and appearance of clinically obvious seizures, called the "latent period of epileptogenesis", defines a cascade of changes that occur in the brain after an injury that lead to the first clinical seizure. Epileptogenesis itself is a process of structural and functional changes transforming the normal brain to one that can generate abnormal neuronal activity that subserves seizures. Moreover, the process of epileptogenesis does not end with the first seizure, but extends further to include progression of the disease, development of drug-resistant epilepsy, etc. (Pitkänen and Engel, 2014).

So far the "gold standard" of epilepsy treatment has been treatment *after* clinical seizures. However, animal studies indicated that pharmacological intervention during a latent period of epileptogenesis in pre-defined cohorts of animals may delay or halt the appearance of seizures (Blumenfeld et al., 2008; Yan et al., 2005; Zeng et al., 2008). The latent period may offer a window of opportunity in which an appropriate treatment may stop or modify the epileptogenic process induced by a brain insult (Stafstrom et al., 2011) (Fig. 1).

On the basis of this concept, several clinical trials have been performed to assess whether pre-seizure administration of an antiepileptic drug (AED) prevents the development of epilepsy after head trauma. These studies with the use of conventional AEDs, such as phenytoin, phenobarbital, carbamazepine, or valproate have failed to prevent epileptogenesis (Temkin, 2009, 2001), giving a rise to the concept that a true "antiepileptogenic drug" should have mechanisms of action distinct from traditional AEDs. However there are additional factors, which may be responsible for lack of preventive effect in this trials. Besides severity and location of brain injury or gap between an injury and onset of treatment, there are other factors which may contribute to the effect of therapeutic intervention, such as age of patient and brain

maturation. In recent years many studies have appeared on biomarkers of epileptogenesis, which should help in identification of proper candidates for preventive or disease modifying treatment (Auvin et al., 2017; Pitkänen et al., 2019; Wu et al., 2016).

The first studies aimed at identifying infants with high probability of epilepsy development have been reported in the latter part of the last century. Lombroso (1983), Watanabe et al. (Watanabe et al., 1987, 1983) and Walther et al. (1987) documented a prognostic utility of electroencephalography (EEG) for identification of infants with high risk of infantile spasms. In the study of Walther et al. (1987) the authors by the use of neonatal polygraphic EEG developed a compound score, which predicted development of infantile spasms or hypsarrhythmia in all 25 newborns with perinatal stress or brain malformation and positive scoring. It is important to note that they concluded: "The high validity of the risk-score based on polygraphic tracing between conceptional age 36 and 44 weeks may allow pre-onset treatment preventing secondary mental deterioration due to hypsarrhythmia and infantile spasms" (Walther et al., 1987).

However, the first use of preventive antiepileptogenic treatment should be ascribed to Ville et al. (2002). The authors compared the outcome of 21 children with Sturge-Weber syndrome (SWS) who were treated following the first seizures with the outcome of 16 patients who were treated preventively from the SWS diagnosis with phenobarbitone. Epilepsy was more frequent in those treated after seizures ($p < 0.01$). They also presented earlier onset of seizures ($p < 0.05$) and poorer cognitive outcome (Ville et al., 2002).

3. TSC as a model for epilepsy modification

Early childhood epilepsies, especially of genetic origin, seem to represent a particularly suitable group for preventive/disease modification studies. They have homogenous cause of epilepsy (one disease), early and predictable age of epilepsy presentation and lesser number of epileptogenic factors, which may contribute to the process of epileptogenesis, compared with the older population. TSC due to its epilepsy characteristics may be considered as particularly good candidate for such studies due to several reasons: 1/unlike many other conditions predisposing to epilepsy, patients can be diagnosed with TSC prior to onset of epilepsy due to non-neurological findings (cardiac rhabdomyomas, skin lesions and positive family history) allowing early intervention, 2/high risk and prevalence of epilepsy and 3/known pathophysiological mechanisms of the disease that can be targeted with preventive treatment.

3.1. Epilepsy characteristics in TSC

TSC is an autosomal dominant disease that affects approximately 1 in 6,000 individuals and is a common genetic cause of early childhood epilepsy. Patients with TSC carry loss-of-function germline mutations in either of the tumor suppressor genes *TSC1* or *TSC2*. The TSC protein complex, composed of these genes products (hamartin and tuberin respectively), inhibits the mechanistic target of rapamycin (mTOR)
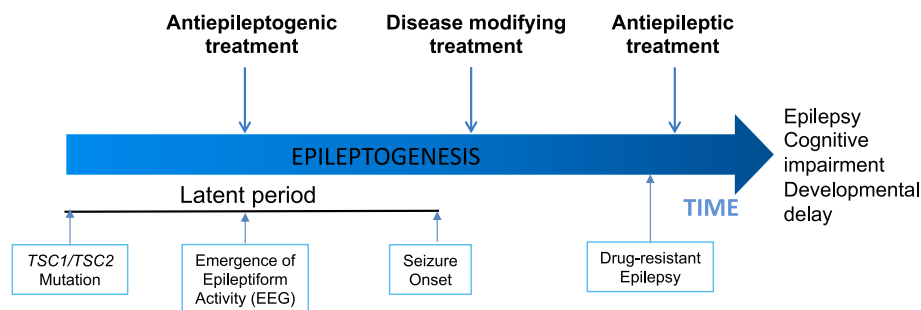

Fig. 1. Concept of epileptogenesis and different treatment approaches in TSC.

pathway (Henske et al., 2016).

Epilepsy appears in about 90% of TSC patients during their lifespan. Approximately 65% of those with epilepsy have medically refractory epilepsy, which in turn increases the likelihood of co-morbid developmental delay and autism. In a prospective study of infants followed from the neonatal period to two years of life, 71% of children developed epilepsy (Jóźwiak et al., 2011). In a large group of patients followed at The Children's Memorial Health Institute in Warsaw by video EEG (carried out every 4 weeks in the first 6 month of life) most patients had normal EEG in the first 2 months of life. In the next few months about 70% of patients developed spike and wave complexes or polyspike activity on EEG, which, if not treated, evolved to more generalized activity. First, short-lasting focal seizures could be seen in children with multifocal spikes or spike and wave complexes, mostly in the 4th or 5th month of life (Domańska-Pakieła et al., 2014). If not successfully treated, they transform to more generalized type of seizures - infantile spasms.

In about 5–6% of children with TSC, the first seizures appear in the neonatal period. In 11 out of 21 (52.4%) patients with neonatal seizures reported by Kotulska et al. (2014) brain MRI revealed large malformations of cerebral cortex, meeting the criteria for focal cortical dysplasia (FCD). Other risk factors for neonatal epilepsy included: perinatal complications and congenital SEGAs. Presence of FCD was associated with more severe epilepsy and worse neuropsychological outcome.

3.2. Role of mTOR activation in epileptogenesis in TSC and other epilepsies

Prevention or disease modification of epilepsy depends on understanding the underlying mechanisms of epileptogenesis which can then be targeted with early therapeutic intervention. Supporting TSC as a model for epilepsy modification, critical mechanisms of epileptogenesis have started to be identified in TSC, particularly related to the mechanistic target of rapamycin (mTOR) pathway (Wong, 2013). mTOR is a relatively ubiquitous protein kinase involved in a number of important molecular and cellular functions throughout the body, including cellular growth, proliferation, and protein synthesis (Lipton and Sahin, 2014; Saxton and Sabatini, 2017). The two TSC genes, *TSC1* and *TSC2*, normally inhibit mTOR and thus limit excessive cellular growth and proliferation. In TSC, mutation of either *TSC1* or *TSC2* leads to a disinhibition or hyperactivation of the mTOR pathway, which then causes excessive cellular growth and proliferation, promoting tumor formation and progression in various organs that is characteristic of TSC. Clinical trials have found that mTOR inhibitors, such as rapamycin and everolimus, represent an effective treatment for brain subependymal giant cell astrocytomas, renal angiomyolipomas, and pulmonary lymphangioleiomyomatosis (Franz, 2013; Krueger et al., 2010), leading to official approval of mTOR inhibitors for these oncological indications in TSC.

Compared with tumor growth, the role of mTOR in the brain in causing epileptogenesis is more complex, but mTOR has been implicated in a number of cellular and molecular processes in the brain that directly affect neuronal excitability and seizures (Wong, 2013). Epilepsy in TSC is generally not directly related to tumor growth per se, but is strongly linked to tubers, as surgical removal of tubers can sometimes eliminate seizures in some TSC patients (Kaczorowska et al., 2011; Ma et al., 2012; Ostrowsky-Coste et al., 2019). Patients with large tubers, as well as tubers hypointense on T1weighted, hyperintense on T2 weighted and heterogeneous on FLAIR images, were more likely reported to have severe epilepsy than patients with other types of tubers (Pascual-Castroviejo et al., 2013). Neuropathological examination of surgically removed cortical tumors from TSC patients revealed features characteristic for FCD type IIb: dysmorphic neurons and balloon cells together with disruption of cortical lamination (Grajkowska et al., 2008; Yasin et al., 2010).

There are a number of pathological, cellular, and molecular

abnormalities within tubers and the perituberal region, or potentially completely independent of tubers, that are likely mTOR-dependent and could contribute to epileptogenesis in TSC. For example, in human TSC tuber specimens or animal models, abnormalities in ion channel or neurotransmitter receptor expression, synaptic plasticity, astrocyte glutamate transport, astrogliosis, neuronal cytomegaly, hypomyelination, microglia activation, and inflammatory mechanisms have been identified (Carson et al., 2012; Goto et al., 2011; Meikle et al., 2007; Sosunov et al., 2008; Uhlmann et al., 2002; Zhang et al., 2015). Many of these abnormalities have been shown to be reversed by mTOR inhibitors, at least in animal models (see below). There may also be a link between GABAergic modulation and the mTOR pathway, as vigabatrin may also inhibit mTOR activity (Zhang et al., 2013).

In addition to TSC, there is now evidence that mTOR is dysregulated in other types or causes of epilepsy. In patients, this is most clear in related types of cortical malformations, such as focal cortical dysplasia type IIb and hemimegalencephaly, where somatic or germline mutations have been identified in upstream regulators of the mTOR pathway, such as DEPDC5, PI3K, PTEN, and mTOR itself (Iffland and Crino, 2017).

In some of these cases, there is evidence that these mutations lead to hyperactivation of the mTOR pathway, similar to TSC, suggesting that mTOR inhibitors may be rational treatments for epilepsy associated with these cortical malformations. Furthermore, mTOR activation has been shown to occur in animal models of acquired epilepsy, such as following traumatic brain injury, neonatal hypoxia, or kainate-induced status epilepticus (Guo et al., 2013; Talos et al., 2012; Zeng et al., 2009). mTOR activation leads to mossy fiber sprouting, neuronal death, and neurogenesis, which are associated with and may promote epileptogenesis in these models.

4. Clinical trials of epilepsy modification in TSC

4.1. Trials in animal models

The increase in mTOR activity and its association with epilepsy in TSC suggest that mTOR inhibitors may be a rational treatment for epilepsy modification in TSC, including possible prevention of epilepsy [1](Sadowski et al., 2015). The effects of mTOR inhibitors, typically rapamycin, on seizures have been tested in multiple animal models of TSC and related mTORopathies (Goto et al., 2011; Ljungberg et al., 2009; Meikle et al., 2008; Zeng et al., 2008). The timing of treatment can be modified to investigate potential early preventive or disease-modifying effects versus late conventional antiseizure/antiepileptic effects. In some cases, late rapamycin treatment of TSC knockout mice with established epilepsy has been shown to decrease existing seizures, consistent with a disease-modifying effect or traditional antiseizure effect. In addition, when initiated prior to the onset of epilepsy, rapamycin has also been shown to prevent the onset of epilepsy, compared with untreated or vehicle-treated mice. For example, in conditional knockout mice involving inactivation of the *TSC1* gene in astrocytes and neurons, rapamycin initiated at two weeks of age inhibited mTOR activity and completely prevented epilepsy from occurring in all treated mice, whereas 100% of vehicle-treated mice develop seizures by 4–6 weeks of age (Zeng et al., 2008). Importantly, rapamycin also prevented the underlying pathological, cellular, and molecular mechanisms that cause epileptogenesis, such as megalencephaly, astrogliosis, and impaired glutamate transporter expression, suggesting that these effects of rapamycin were consistent with true preventive or disease-modifying actions, not simply a suppression of seizures. However, it should be noted that once rapamycin was stopped, the pathological and cellular phenotype and seizures could emerge as mTOR activity rebounded, suggesting that continued treatment may be necessary to maintain a

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long-term effect. This temporal requirement could be more a function of the relatively simplistic, reductionist animal models, as human brain development may involve more intricate, complex developmental windows or critical periods, after which chronic treatment may not be necessary.

The preclinical data supporting efficacy of mTOR inhibitors in treatment of epilepsy in TSC, including prevention and disease-modification, are consistent and compelling. Animal models of TSC similarly provide the opportunity to investigate potential alternative therapies for epilepsy modification. Other therapeutic approaches could target mechanisms related to or downstream from the mTOR pathway or could be completely independent of mTOR. Vigabatrin, which is already a very effective, established treatment for infantile spasms in TSC, potentially represents a unique approach to epilepsy modification in TSC from a mechanistic standpoint. The established mechanism of action of vigabatrin involves increasing GABA levels in the brain via inhibition of its breakdown by GABA transaminase. However, other GABA potentiating drugs, such as benzodiazepines and barbituates, are not as effective as vigabatrin against infantile spasms in TSC, suggesting that vigabatrin may have another mechanism of action specific to TSC. Toward this end vigabatrin has also been found in mouse models of TSC to have some effects in inhibiting mTOR, as reflected by inhibition of phosphorylation of the downstream mTOR effector, S6 protein. (Zhang et al., 2013). These dual, combined actions might account for vigabatrin's strong efficacy for infantile spasms in TSC and support its application for potential prevention and disease-modification of epilepsy, as currently being pursued in clinical trials in TSC patients.

4.2. Trials in humans

4.2.1. Early or pre-seizure treatment with conventional drugs

Currently, treatment of epilepsy is usually introduced after the clinical diagnosis of epilepsy is set. According to the newest International League Against Epilepsy (ILAE) classification of epilepsies released in 2017, epilepsy is diagnosed when an individual had at least 2 unprovoked seizures >24 h apart, or one unprovoked seizure and at least 60% risk of having another seizure over next 10 years. All available pharmacological treatments of epilepsy are actually only seizure suppressing, or 'antiepileptic' and none are disease-modifying, or 'antiepileptogenic, when introduced after clinical seizures. Preclinical models have showed that some antiepileptic drugs, however, may prevent or alleviate epilepsy when given before seizures (Blumenfeld et al., 2008; Yan et al., 2005). In humans, the trials to prevent epileptogenesis using standard antiepileptic drugs after severe traumatic brain injury (Bakr and Belli, 2018; Mani et al., 2011; Wat et al., 2019), stroke (Angriman et al., 2019), or craniotomy (Greenhalgh et al., 2018; Lee et al., 2018) have been performed and were unsuccessful. The failure of anti-epileptogenic effects in these studies may in part result from inclusion of patients at different risk of epilepsy, highlighting the need for reliable and clinically applicable biomarkers of epileptogenesis.

Up until now, the only clinically available tool to detect epilepsy development before clinical seizures is a change in EEG pattern. An increasing body of evidence indicates that the spikes on EEG precede clinical seizures during epileptogenesis in various clinical conditions (Bernardo et al., 2018; Chang et al., 2012; Chauvière et al., 2012; van Rooij et al., 2010; Wu et al., 2016). In the study reported by van Rooij et al. [2](van Rooij et al., 2010), infants with severe hypoxic-ischemic encephalopathy were included and monitored by EEG. Antiepileptic treatment with phenobarbitone was randomly assigned in infants either before or after the onset of clinical seizures. The results suggested that epilepsy was less severe in infants treated preventively, but differences were not significant, probably due to the small study population.

In TSC, the diagnosis of the disease is being increasingly made before seizures (Słowińska et al., 2018) enabling monitoring of EEG changes and identification of patients at high risk of developing epilepsy [3](Bernardo et al., 2018; Domańska-Pakieła et al., 2014; Wu et al., 2016).

A longitudinal study was recently completed which focused on interictal scalp epileptiform discharges and was able to identify impending epilepsy in the majority (77%) of seizure-naïve TSC infants. In this longitudinal cohort TSC infant study participants were enrolled across 5 TSC centres for a total of 40 infants with a mean age of 82.4 days, and 32 completed the 24 month study. Two were lost to follow up and 6 were treated with AED either due to electrographic seizures and/or epileptiform discharges on their EEGs prior to the onset of clinical seizures. Seventeen of the 32 remaining children developed epilepsy at a mean age of 7.5 months (SD = 4.4). Generalized/focal slowing, hypsarrhythmia, and generalized/focal attenuation were not predictive for the development of clinical seizures. Presence of inter-ictal epileptiform discharges had a 77.3% positive predictive value and absence a 70% negative predictive value for developing seizures by 2 years of age. Epileptiform discharges preceded clinical seizure onset by 3.6 months (mean). The results of this study supports the use of a 1 h awake and asleep EEG as a biomarker for ongoing epileptogenesis in most but not all infants with TSC(Wu et al., 2019, 2016).

Our data suggest there is a specific association between severity of epilepsy and co-morbid conditions, including developmental delay, reported previously in retrospective and prospective series (Capal et al., 2017; Chu-Shore et al., 2010; Davis et al., 2017; Northrup et al., 2013). This study showed significant decline only in infants with ongoing seizures, but not in those who did not developed seizures or whose seizures came under control. There is emerging evidence that persistent seizures, but not so history of interictal epileptiform activity, nor history of well controlled seizures, correlated with low scores on the Vineland and Mullen tests at 2 years of age (Wu et al., 2016).

The results of these observational studies allowed studies with active preventive use of vigabatrin in TSC infants. An open-label study of preventive treatment in infants with TSC (Jóźwiak et al., 2011) demonstrated that in patients with paroxysmal discharges on EEG it is possible to modify the outcome of the disease by antiepileptic treatment before the onset of clinical seizures with vigabatrin, a drug standardly used to treat infantile spasms in TSC. Preventive vigabatrin treatment used in a group of 11 TSC infants with EEG changes improved the outcome when compared to a historical control group of 31 infants in whom neither EEG monitoring, nor prevention were introduced. Infants were followed up to the age of 24 months. Preventive treatment increased the proportion of seizure free patients (93% versus 35% in a standard treatment group), lowered the risk of drug resistant epilepsy (7% versus 42%) and the risk of intellectual disability (48% vs 14%; p = 0.031; mean IQ score 68.7 vs 92.3; p < 0.05) (Jóźwiak et al., 2011)[4]. On longer follow-up, at median age of 8.8 years of age, only 5 of 14 (35.7%) required antiepileptic treatment and in 6 subjects we were able to withdraw the treatment completely. Patients with the preventive treatment had much better neurocognitive outcome (IQ score of 81.6 in comparison to 52.8 in an age-matched group receiving standard treatment (p < 0.03 (Jóźwiak et al., 2019; Jóźwiak and Kotulska, 2014). The most important caveats about this study were the small number of the patient and open-label, single arm design with a historical control group; thus larger, prospective controlled randomized trials have been recommended.

The EPISTOP project (Clinical- Trials.gov Identifier NCT02098759), a large collaborative study funded by European Union within 7th Frame Programme to examine the risk factors and biomarkers of epilepsy has addressed this issue. In the clinical part of the project, a clinical trial

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comparing preventive and standard antiepileptic treatments has been carried out. Ninety-seven infants with definite TSC were included and followed from birth up to the age of 24 months with serial EEG, imaging, and laboratory tests. In the randomized part of the study, children were randomly assigned to either preventive or standard antiepileptic treatment with vigabatrin. Preventive treatment was introduced after the onset of spikes on EEG but prior to seizures. The results of the study are currently in preparation.

Similar randomized controlled double blind clinical trial in TSC infants, has been recently launched also in the US as the PREVeNT trial (PReventing Epilepsy Using Vigabatrin In Infants with Tuberous Sclerosis Complex; (Clinical- Trials.gov Identifier NCT02849457). The study is currently underway in the United States. The results of this study will provide important information which will determine if early interventions prior to the development of epilepsy with antiseizure medications such as vigabatrin will be useful to prevent epilepsy, the development of refractory epilepsy and cognitive impairment in children with epileptiform EEGs. Perhaps as or more important than epilepsy outcomes in the TSC patient population, is their developmental outcome and risk for autism. The PREVeNT study begins to answer the role of preventive antiepileptic therapy in infants with TSC and how targeting epilepsy prevention impacts developmental outcomes for these patients.

4.2.2. Trials with mTOR inhibitors

While controlled preventive clinical trials of vigabatrin for epilepsy in TSC are ongoing, comparable preventive clinical trials of mTOR inhibitors are still under development. However, the mTOR inhibitor, everolimus, has been tested rigorously in clinical trials for conventional antiseizure effects in TSC patients with drug-resistant epilepsy. Initial suggestion for a potential effect of everolimus against seizures in TSC patients was derived from the clinical trials of everolimus for tumors in patients who had concurrent epilepsy (Kotulska et al., 2013; Krueger et al., 2010). Open-label drug studies further supported that everolimus decreased seizure frequency in TSC patients, independent of treatment of tumors (Krueger et al., 2013; Sadowski et al., 2016). Most definitively, a recent double-blind, randomized, placebo-controlled trial (EXIST-3) showed that everolimus was significantly more effective than placebo in reducing focal seizures in TSC patients with drug-resistant epilepsy (French et al., 2016). A high exposure everolimus group had a median reduction in seizure frequency of 40% compared with 15% in the placebo group. Longer term follow-up of this study indicates that everolimus has a sustained effect on seizure frequency (Franz et al., 2018).

Given the precedent of preventive clinical trials with vigabatrin for epilepsy in TSC, similar preventive trials with mTOR inhibitors are in the planning stages, but have not yet been conducted. One barrier to progress has been the concern for potential adverse effects of mTOR inhibitors in young infants, given the role of the mTOR pathway in normal growth and development. In a large multicenter survey of 45 children below 2 years of age severe adverse events (Grade 3) were present in 7 (16%) subjects and no life-threatening (Grade 4) or death/disability (Grade 5) were reported. Treatment was discontinued due to adverse events in 9 of 45 (20%) (Krueger et al., 2018). Nevertheless, adverse events were mild to moderate in the majority of cases and overall mTOR inhibitors were well-tolerated in this patient population, suggesting that a preventive clinical trial of mTOR inhibitors for epilepsy is feasible and safe.

5. Current recommendations and clinical practice

Current treatment practices for epilepsy in TSC include medical treatment of seizures, ACTH, epilepsy surgery, ketogenic diet and its variants, vagus nerve stimulation, and, recently, preventive strategies in infants. Given that epilepsy in TSC is frequently resistant to medical treatment (Chu-Shore et al., 2010; Nabbout et al., 2019; Song et al.,

2018), a broad range of antiepileptic drugs is used in attempts to achieve seizure control, including: vigabatrin, valproic acid, levetiracetam, carbamazepine, oxcarbazepine, topiramate, phenytoin, mTOR inhibitors, and others (Overwater et al., 2015). According to he data from the TSC Natural History Database which included 1328 TSC individuals from the U.S. and Europe, 83.6% of patients with TSC have epilepsy (Song et al., 2018). Nearly all patients with TSC and epilepsy receive medical treatment, and in 64.5% of them 3 or more antiepileptic drugs are required. Other treatment options for epilepsy included epilepsy surgery in 25.3%, dietary treatment in 7.9%, and mTOR inhibitors in 1% of the patients (Song et al., 2018). It should be noted that the data in the database were captured between 2006 and 2014 and currently the use of mTOR inhibitors for epilepsy associated with TSC is likely more common.

In TSC, epilepsy usually presents within the first months of life. Another recent study based on a large multicenter registry of over 2.000 patients showed that nearly 80% of children with TSC developed epilepsy before the age of 2 years (Nabbout et al., 2019). It is established that timing of treatment initiation is crucial for epilepsy outcome in TSC (Canevini et al., 2018). In a study of Cusmai et al. (2011),[5] all children with TSC who received antiepileptic treatment later than one week after the first clinical seizure had intellectual disability and more than 60% of them had drug-resistant seizures. When antiepileptic treatment was introduced within first week after the onset of clinical seizures, the risk of intellectual disability and drug-resistant seizures decreased by about 30%–60% (Cusmai et al., 2011; Jóźwiak et al., 2011).

In infants, focal seizures are frequently followed by infantile spasms. Their incidence was reported to be as high as 70% in some older studies and decreased significantly with an introduction of EEG surveillance (Chu-Shore et al., 2010; Curatolo et al., 2001; Overwater et al., 2015). Vigabatrin is widely used for infantile spasms and focal seizures in infants (Curatolo et al., 2018; Overwater et al., 2015). In recent years, an increasing body of evidence indicates that vigabatrin might be also effective in epileptic spasms and tonic seizures in older patients with TSC (van der Poest Clement et al., 2018). It has been also shown that high dose of vigabatrin prevents the relapse of infantile spasms (Hussain et al., 2018), suggesting that it may change the natural course of epilepsy in TSC. Vigabatrin has been used in three epilepsy ion studies in infants with TSC: the open-label study published by Jozwiak et al., in 2011, as well as two randomized trials: EPISTOP and PREVeNT studies (Table 1).

Both timing of treatment initiation and the use of appropriate medication are addressed in current recommendations for the management of epilepsy in TSC. New European recommendations have been published in 2018 by the group of experts in TSC (Curatolo et al., 2018). The need for early diagnosis and presymptomatic assessment for epilepsy with EEG surveillance in newborns and infants with TSC has been highlighted. Video EEG recording is recommended in every newly diagnosed newborn and infant and follow-up video EEG recording should be performed every 4 weeks until the age of 6 months and every 6–8 weeks thereafter in infants unless seizures are disclosed. Treatment with vigabatrin should be initiated immediately after electrographic or clinical seizures, whichever first, are recognized (Curatolo et al., 2018). Vigabatrin is the first choice treatment for focal seizures [6]and infantile spasms in TSC. The recommended dose is 100–150 mg/kg as lower dosage is associated with higher rate of seizure recurrences (Hussain et al., 2018).

In non-responders, second antiepileptic drug should be selected considering the age of the patient and seizure type. Epilepsy surgery is recommended to be considered early, after the failure of second antiepileptic drug. In patients with few tubers and good anatomic-electro-

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[6] Should be: Vigabatrin is the first choice treatment for infantile spasms and focal seizures in the first year of life in TSC.

Table 1

Comparison of EPISTOP and PREVeNT trials with preventive epilepsy treatment in TSC. EEG = electroencephalography; EOS = end of study; WES = whole genome sequencing.

	EPISTOP	PREVeNT
Trial type	IIb/III	IIb
Enrolment goal	N = 100 (101 enrolled)	N = 80
Randomization	Based on presence of epileptiform activity; blinded; 1:1; Vigabatrin vs no treatment	Based on presence of epileptiform activity; blinded; 1:1; Vigabatrin vs placebo
Intervals between EEGs	4 weeks in the first 6 mo, 6 weeks in 7–12 mo,8 in 13–24 mo	6 weeks
Study design	24 mo follow up, Study end points: time to first seizure, risk of drug resistant epilepsy, proportion of patients with seizures	Time to first seizure; drug-resistant epilepsy at EOS; neuropsychological outcome; proportions of seizure-free patients at EOS
Blood biomarkers	Serial blood collection (at enrolment, at abnormal EEG, at seizures, at EOS), Biomarkers analysed in the project	Serial samples stored at TSC biorepository
Genetic testing	WGS within the project	Samples stored in TSC repository
Developmental testing	6, 12, 18, 24 mo	6, 12, 24, 36mo
Autism evaluation	6, 12, 24 mo	24 and 36 mo

clinical correlations the surgical treatment results in 50–60% chance of long-term seizure freedom (Ostrowsky-Coste et al., 2019). In patients with a significant "tuber burden" long-term monitoring and multiple neuroimaging and electrographic source localization procedures must be applied (Jansen et al., 2007). Restricted availability of highly specialized epilepsy surgery centres for such patients may be a limitation of this method.

Ketogenic diet is an important treatment option in cases of medically intractable epilepsy related to TSC especially given the mechanism of action of ketogenic diet in regulating epigenetic mechanisms and the mTOR pathway (Boison, 2017; McDaniel et al., 2011). According to Park et al. (2017) up to 80% of TSC patients with drug-resistant epilepsy may experience at least 50% reduction of seizures number.

There is an increasing interest in the use of mTOR inhibitors as a first antiepileptogenic treatment in TSC infants. Currently licensed mTOR inhibitor for epilepsy in TSC is registered after 12 months of age while such preventive treatment should be applied in the first months of life. However there is increasing number of reports on the use of mTOR inhibitors in young infants documenting that such treatment may be safe and effective (Curatolo et al., 2016a, 2016b; Curatolo and Maria, 2013; Jóźwiak et al., 2016; Kotulska et al., 2013; Krueger et al., 2018). As TSC represents a genetic encephalopathy, where encephalopathy is caused not only by epilepsy per se, but also by underlying genetic mutation and its molecular consequences, there are increasing expectations that the treatment with mTOR inhibitors may be more efficient and holistic in infants with TSC.

The U.S. recommendations of the 2012 International Tuberous Sclerosis Complex Consensus Conference highlighted the need for education of the parents and caregivers to recognize focal seizures as well as infantile spasms (Krueger et al., 2013). All children who received TSC diagnosis should undergo a baseline EEG. If no clinical seizures were recognized or reported and baseline EEG is abnormal, it should be followed with 24-hour video EEG in order to disclose electrographic or subtle clinical seizures ([7]Krueger et al., 2013).

6. Conclusions and perspectives

Prevention of epilepsy or modification of the disease outcome is one of the major Epilepsy Research benchmarks of the United States the National Institute of Neurological Disorders and Stroke/National Institutes of Health Epilepsy Research and a research priority of the European scientific community.

The recent "practical clinical definition of epilepsy" allowed the diagnosis of epilepsy after a single seizure when a probability of another seizure exceeds 60% (Fisher et al., 2014). Such conditions as TSC, Sturge-Weber syndrome, Angelman syndrome, and Rett syndrome with

an incidence of epilepsy over 60% in the first year of life, well recognized natural course of the disease, and established severe epilepsy comorbidities should be considered as a potential candidates for preventive treatment (Jóźwiak and Kotulska, 2014). In these conditions early therapeutic intervention may prevent, delay or ameliorate the development of epilepsy, drug-resistant epilepsy and intellectual disability and significantly improve the quality of life of young patients and their families.

There is an increasing number of papers demonstrating the utility of pre-seizure EEG surveillance in early diagnosis of TSC as a first step to preventive treatment (Benova et al., 2018; Chung et al., 2017; Whitney et al., 2017). Studies in SWS are in the pipeline (Pinto et al., 2016). The recent clinical trials in TSC indicate that a preventive treatment for epilepsy in TSC is feasible and could become standard of care in the near future.

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