

REVIEW ARTICLE

MEDICAL PROGRESS

The Tuberous Sclerosis Complex

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THE TUBEROUS SCLEROSIS COMPLEX (TSC), A MULTISYSTEM, AUTOSOMAL dominant disorder affecting children and adults, results from mutations in one of two genes, *TSC1* (encoding hamartin) or *TSC2* (encoding tuberlin) (see the Glossary). First described in depth by Bourneville in 1880,¹ TSC often causes disabling neurologic disorders, including epilepsy, mental retardation, and autism. Additional major features of the disease include dermatologic manifestations such as facial angiofibromas, renal angiomyolipomas, and pulmonary lymphangiomyomatosis. TSC has a wide clinical spectrum of disease, and many patients have minimal signs and symptoms with no neurologic disability. With the discovery of the two genes responsible for TSC and insights derived from observations in *Drosophila melanogaster* models, our understanding of the pathogenesis of this disorder has progressed rapidly during the past decade. Clinical trials predicated on the cellular targets of hamartin and tuberlin are under way.

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CLINICAL FEATURES AND DIAGNOSIS

The diagnostic criteria for TSC consist of a set of major and minor diagnostic features² (Table 1). Cases meeting these criteria fulfill a clinical diagnosis of TSC; the results of molecular genetic testing of the *TSC1* or *TSC2* loci are currently viewed as corroborative. Many affected persons come to medical attention because of seizures or dermatologic manifestations. However, no single feature of TSC is diagnostic; thus, an evaluation that includes consideration of all clinical features is necessary to make the diagnosis. The clinical manifestations of TSC appear at distinct developmental points (Table 1). For example, cortical tubers and cardiac rhabdomyomas form during embryogenesis and thus are typical findings in infancy. Skin lesions are detected at all ages in more than 90% of patients with TSC. Hypopigmented macules (formerly known as ash-leaf spots) are generally detected in infancy or early childhood, whereas the so-called shagreen patch is identified with increasing frequency after the age of 5 years. Ungual fibromas typically appear after puberty and may develop in adulthood. Facial angiofibromas (Fig. 1A, 1B, and 1C), formerly called adenoma sebaceum, may be detected at any age but are generally more common in late childhood or adolescence. A subependymal giant-cell tumor of the brain may develop in childhood or adolescence. Renal cysts can be detected in infancy or early childhood, whereas angiomyolipomas develop in childhood, adolescence, or adulthood. Pulmonary lymphangiomyomatosis is found in adolescent girls or women with TSC.

Clinical manifestations of TSC have variable penetrance. For example, two underrecognized groups of patients are asymptomatic adults with one or two minor features who meet the diagnostic criteria on the basis of a physical examination, radiographic findings, or both and asymptomatic women who give birth to a child with early neurologic manifestations of TSC.

Glossary

GAP: GTPase-activating protein; converts a Ras homologue protein from GTP (active state) to GDP (inactive state).
Hamartin: Protein product of the chromosome 9q34 <i>TSC1</i> gene.
Loss of heterozygosity (LOH): A DNA marker with a one-allele (homozygous) pattern in a tumor as compared with normal DNA, which has a two-allele (heterozygous) pattern.
mTOR: Mammalian target of rapamycin (now called sirolimus); kinase activated by Rheb.
p70S6K: Ribosomal protein p70 S6 kinase; target of mTOR that activates translation.
S6: Ribosomal S6 protein; regulates protein translation.
Sirolimus: FDA-approved agent that inhibits mTOR.
Rheb: Ras homologue; target of the GAP domain of tuberlin.
S6: Ribosomal protein S6, substrate of p70S6K.
Tuberlin: Protein product of the chromosome 16p13 <i>TSC2</i> gene.

RENAL LESIONS

Renal angiomyolipomas, benign tumors composed of abnormal vessels, immature smooth-muscle cells, and fat cells (Fig. 1D, 1E, and 1F), are bilateral, with multiple tumors in each kidney, in most patients with TSC. The estimated incidence of angiomyolipomas in TSC ranges from 55 to 75%.^{3,4} Angiomyolipomas may be detected by ultrasonography, computed tomography (CT), or magnetic resonance imaging (MRI). Because these tumors have abnormal vasculature and often contain aneurysms, spontaneous life-threatening bleeding is an important complication, especially when angiomyolipomas are 3 cm or greater in diameter. The rate of growth of angiomyolipomas varies among patients and lesions. For example, a longitudinal study of 25 boys and 35 girls with TSC showed that 75% had renal angiomyolipomas by the age of 10.5 years,⁴ and in 2 of the boys, the diameter of the angiomyolipomas increased by 4 cm in a year. In general, surgical resection is avoided whenever possible in order to preserve renal function; angiomyolipomas that are more than 3 to 4 cm in diameter can usually be treated successfully by embolization.^{5,6}

In addition to angiomyolipomas, epithelial renal lesions that include epithelial cysts, polycystic kidney disease, and renal-cell carcinomas may develop in patients with TSC. Epithelial cysts are generally asymptomatic and are more often associated with hypertension and renal failure than are angiomyolipomas.⁷ In addition, an estimated 2 to 3% of patients with TSC carry a contiguous

germline deletion that affects both the *TSC2* gene and one of the genes that leads to autosomal dominant polycystic kidney disease on chromosome 16p13, resulting in a polycystic kidney phenotype that is detectable in infancy or early childhood and that generally leads to renal insufficiency in the late teens to early 20s.⁷⁻⁹

The overall incidence of renal carcinoma in patients with TSC is similar to that in the general population, with a lifetime risk of 2 to 3%; however, the cancer is diagnosed at a younger age in patients with TSC. In one series of patients with TSC, renal carcinoma developed at an average age of 28 years,¹⁰ 25 years earlier than the average age at diagnosis in the general population. There are case reports of renal carcinoma in children and even in one infant with TSC.¹⁰⁻¹² An unusual feature of renal carcinoma associated with TSC is its pathological heterogeneity. Clear-cell, papillary, and chromophobe carcinoma subtypes, as well as oncocytomas, have all been reported in patients with TSC.^{11,12} In one series, four of six patients died from metastases of renal carcinoma.¹²

PULMONARY MANIFESTATIONS

Lymphangiomyomatosis, also called lymphangiomyomatosis, affects women almost exclusively and is characterized by widespread pulmonary proliferation of abnormal smooth-muscle cells and cystic changes within the lung parenchyma (Fig. 1G, 1H, and 1I).¹³ Lymphangiomyomatosis is usually diagnosed during early adulthood and is initially manifested by dyspnea or pneumothorax. Radiographic evidence indicates that the incidence of lymphangiomyomatosis among women with TSC is 26 to 39%^{14,15}; many of these women are asymptomatic. In a series of 49 TSC-related deaths reported by the Mayo Clinic, lymphangiomyomatosis was cited as the cause of 4 deaths, making it the third most frequent cause of death after renal and brain lesions.¹⁶

NEUROLOGIC MANIFESTATIONS

The neurologic manifestations of TSC, which include epilepsy,^{17,18} cognitive disability,¹⁹ and neurobehavioral abnormalities, such as autism,²⁰ appear to be intimately related to the cerebral cortical tubers (Fig. 2) that are present in over 80% of patients. Tubers are developmental abnormalities of the cerebral cortex characterized histologically by a loss of the normal six-layered structure of the cortex and by dysmorphic neurons, large astro-

cytes, and a unique type of cell known as a giant cell.^{21,22} Tubers have been identified in fetuses at a gestational age of 20 weeks.²³ The lesions persist throughout life but do not become malignant tumors. Tubers can calcify or undergo cystic degeneration.

Epilepsy may be the most prevalent and challenging clinical manifestation of TSC. Epilepsy occurs in more than 70 to 80% of patients with TSC, and virtually all subtypes of seizure (simple partial, complex partial, and generalized tonic-clonic seizures) have been reported. Seizures are often refractory to treatment, even to polytherapy with antiepileptic drugs.¹⁷ Patients with medically refractory epilepsy may require a surgical evaluation.^{24,25} In most cases, the region in which the seizure originates coincides with the location of a tuber in the brain, and it is widely believed that tubers serve as the epileptogenic focus. Thus, intractable epilepsy is often treated by resection of a tuber.^{24,25}

Infantile spasms, a devastating epileptic syndrome that is often associated with profound mental retardation and a poor neurologic prognosis, occurs in 20 to 30% of infants with TSC.¹⁸ Treatment with vigabatrin, an inhibitor of γ -aminobutyric acid transaminase, appears to be beneficial in some of these infants.¹⁸ Whether the association between infantile spasms and cognitive deficits arises from the effects of early seizures or by a distinct mechanism is unknown.²⁶ The risk and degree of intellectual impairment correlate with the time from the initiation of treatment until the cessation of the spasms and the ability to control the seizures after infantile spasms, suggesting that the seizures themselves have some role in the severity of the subsequent neurologic deficits.²⁶⁻²⁸ Clinical studies have suggested that a higher number of tubers (more than seven) in patients with TSC is associated with the development of infantile spasms and intractable epilepsy; thus, the number of tubers may also be an independent risk factor for cognitive disability.²¹

In approximately 10% of patients with TSC, the growth of subependymal giant-cell tumors (Fig. 2) can cause obstruction of cerebrospinal fluid flow, hydrocephalus, increased intracranial pressure, and even death.^{3,29} These lesions are composed of proliferative astrocytes and giant cells but do not become malignant glial tumors.²⁹ Subependymal giant-cell tumors are presumed to derive from subependymal nodules, which are

Table 1. Diagnostic Criteria for TSC.*

Criteria	Age at Onset
Major	
Facial angiofibroma	Infancy to adulthood
Ungual fibroma	Adolescence to adulthood
Shagreen patch	Childhood
Hypomelanotic macule	Infancy to childhood
Cortical tuber	Fetal life
Subependymal nodule	Childhood to adolescence
Subependymal giant-cell tumor	Childhood to adolescence
Retinal hamartoma	Infancy
Cardiac rhabdomyoma	Fetal life
Renal angiomyolipoma	Childhood to adulthood
Lymphangiomyomatosis	Adolescence to adulthood
Minor†	
Multiple pits in dental enamel	
Hamartomatous rectal polyps	
Bone cysts	
Cerebral white-matter radial migration lines	
Gingival fibromas	
Retinal achromic patch	
"Confetti" skin lesions (groups of small, lightly pigmented spots)	
Multiple renal cysts	

* Two major features or one major feature plus two minor features are required for a definite clinical diagnosis of TSC; for a probable diagnosis of TSC, one major and one minor feature are required; for a possible diagnosis of TSC, one major or two or more minor features are needed. Cerebral cortical dysplasia and cerebral white-matter radial migration lines are counted together as one feature of TSC; when both lymphangiomyomatosis and renal angiomyolipomas are present, other features of tuberous sclerosis must be present before TSC is diagnosed. Data were modified from Roach et al.²

† In addition to major diagnostic features, minor features affect the teeth (dental pits), gums (gingival fibroma), digestive tract (hamartomatous rectal polyps), blood vessels (aneurysms), and bony skeleton (bone cysts or sclerosis). Data are not available to list the typical age at onset.

asymptomatic hamartomas that protrude from the walls of the lateral and third ventricles.

CARDIAC LESIONS

Cardiac rhabdomyomas are intracavitary or intramural tumors that are present in nearly 50 to 70% of infants with TSC but cause important clinical problems in only a very small fraction of those patients. Rhabdomyomas may be detected on fetal ultrasonography and are the most common cardiac tumor diagnosed in utero. The detection of a rhabdomyoma may be useful for making a pre-

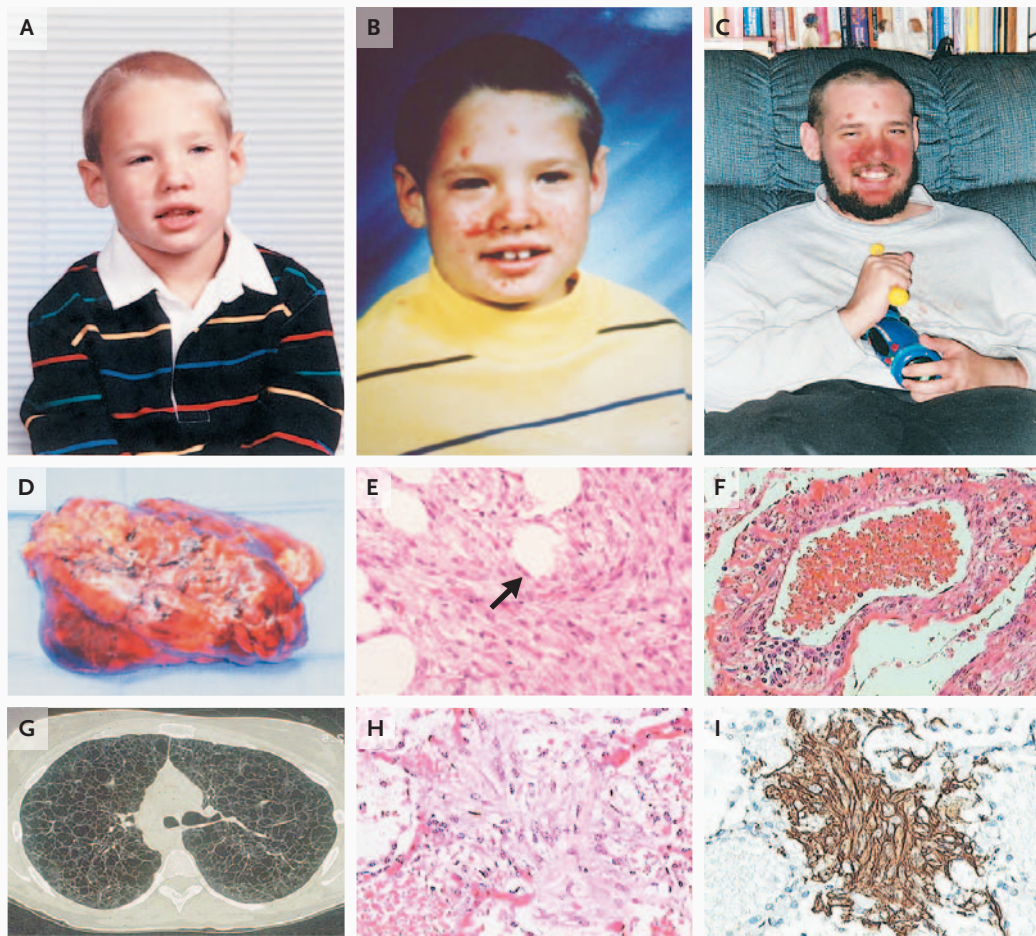


Figure 1. Dermatologic, Renal, and Pulmonary Manifestations of TSC.

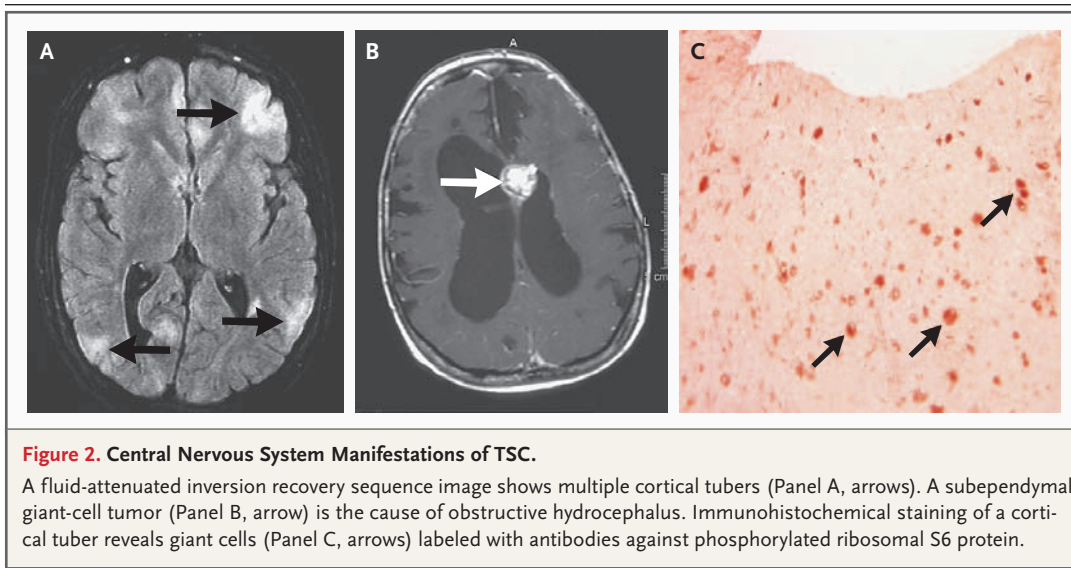
A patient with facial angiofibromas around the nose and chin is shown at 4 years of age (Panel A), 8 years of age (Panel B), and 21 years of age (Panel C). The progression of the lesions over time is evident. Panel D shows a resected kidney distorted by numerous angiomyolipomas. Sections stained with hematoxylin and eosin show fat (arrow) and smooth muscle (Panel E) and aberrant vessels (Panel F). Computed tomography (Panel G) of the lungs shows the radiographic appearance of lymphangiomyomatosis. Smooth-muscle proliferation in lymphangiomyomatosis is shown after staining with hematoxylin and eosin (Panel H) and immunohistochemical labeling with muscle-specific actin (Panel I).

natal diagnosis of TSC. In one series of cases, rhabdomyoma was identified in 19 fetuses; the diagnostic criteria for TSC were met after birth in 15 (79%).³⁰ In all 15 of those fetuses, multiple rhabdomyomas were detected, whereas the 2 fetuses with a single rhabdomyoma had no other diagnostic features of TSC. Rhabdomyoma may be associated with cardiac failure in infancy,³⁰ and 47% of patients with rhabdomyoma have also had associated cardiac dysrhythmias, including atrial tachycardia, ventricular tachycardia, complete heart block, and the Wolff–Parkinson–White syndrome.

Unlike other lesions seen in TSC, cardiac rhabdomyomas often disappear spontaneously in later life; in one series, for example, 20 of 24 patients with TSC had complete regression of the rhabdomyoma.³¹

MOLECULAR GENETICS

Linkage analysis in multigenerational families and positional cloning were used to map both the *TSC1* and *TSC2* genes (Fig. 3).^{32–34} The *TSC2* gene, which is located on chromosome 16p13, encodes a tran-



script of 5.5 kb containing 41 exons and encompassing 40 kb of genomic DNA; there are several alternatively spliced versions.³⁵ The *TSC1* gene, which is located on chromosome 9q34, encodes a transcript of 8.6 kb, containing 23 exons and encompassing 55 kb of genomic DNA.³⁴

Extensive studies of the *TSC1* and *TSC2* genes in patients with TSC have revealed a wide spectrum of mutations.^{33,34} Indeed, more than 200 *TSC1* and nearly 700 *TSC2* unique allelic variants have been reported (http://chromium.liacs.nl/lovd/index.php?select_db=TSC1_or_db=TSC2).³⁶⁻⁴⁰ There are no particular regions within the *TSC1* or *TSC2* gene in which mutations occur at a high rate, although missense mutations at Arg611 (exon 16), Pro1675Leu (exon 38), and an 18-bp in-frame deletion in exon 40 have been observed in *TSC2* in multiple patients.^{3,36,37} Missense mutations and large genomic deletions are much more frequent in *TSC2* than in *TSC1*. Missense mutations in *TSC2* tend to cluster in the GTPase-activating protein (GAP) binding domain (exons 35 through 39).⁴¹ A subgroup of large genomic deletions and rearrangements in *TSC2* also affect the adjacent *PKD2* gene, causing early-onset polycystic kidney disease.^{8,9}

Linkage studies initially suggested that there would be equivalent numbers of families with mutations in each *TSC* gene.⁴² However, the frequency of mutations reported in *TSC2* is consistently higher than in *TSC1*; *TSC1* mutations account for only 10 to 30% of the families identified with TSC.^{36-39,41,43} In sporadic cases of TSC, there is an

even greater excess of mutations in *TSC2*. Indeed, in two large studies, mutations in *TSC1* were identified in only about 15% of patients.^{3,37} Nonetheless, identification of *TSC1* mutations appears to be twice as likely in familial cases as in sporadic cases. The disparity in mutational frequency may reflect an increased rate of germline and somatic mutations in *TSC2* as compared with *TSC1*, as well as ascertainment bias, since mutations in *TSC2* are associated with more severe disease.^{3,36,37}

Among patients meeting the clinical criteria for a diagnosis of TSC, 15 to 20% have no identifiable mutations.^{3,36} These persons generally have milder clinical disease (i.e., a lower incidence of mental retardation, seizures, and dermatologic signs) than patients with identified *TSC1* or *TSC2* mutations. Somatic mosaicism has been reported in some persons with mutations in *TSC1* or *TSC2* and is thought to account for a milder clinical phenotype. This is also a credible explanation for the failure to detect a mutation.⁴⁴

In agreement with Knudson's two-hit tumor-suppressor gene model,⁴⁵ inactivation of both alleles of either *TSC1* or *TSC2* appears to be required for lesion formation in TSC. Most second-hit mutations are large deletions involving the loss of surrounding loci. These mutations are referred to as loss of heterozygosity, since they affect neighboring heterozygous polymorphic markers. Loss of heterozygosity in *TSC1* or *TSC2* has been consistently observed in the majority of TSC-associated angiomyolipomas, cardiac rhabdomyomas, subependymal giant-cell tumors, and lymphangio-

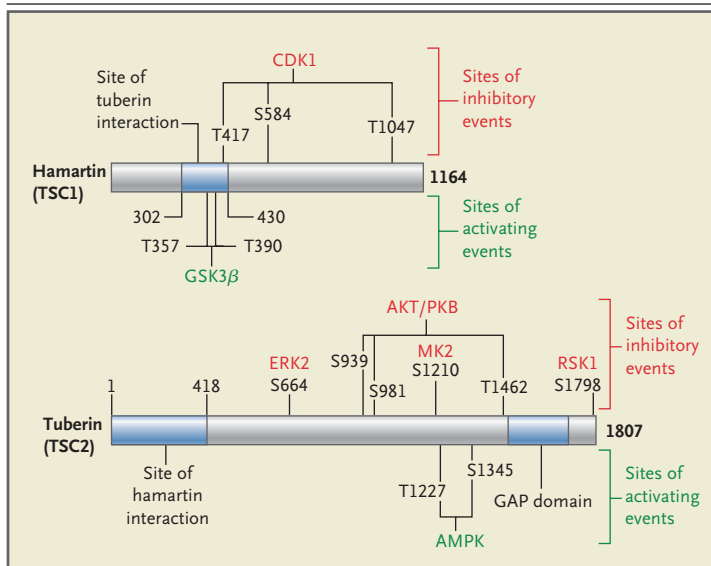


Figure 3. Structure of Hamartin (TSC1) and Tuberin (TSC2).

TSC1 is composed of 1164 amino acids and interacts with tuberlin in the region of amino acids 302 through 430. TSC2 is composed of 1807 amino acids and interacts with hamartin in the region of amino acids 1 through 418. The activity of TSC1 and TSC2 is regulated by both inhibitory and activating phosphorylation events at specific amino acid residues. CDK1 denotes cyclin-dependent kinase 1, GSK3 β glycogen synthase 3 β , ERK2 extracellular-related kinase 2, MK2 mitogen-activated protein kinase-activated protein kinase 2, AKT/PKB protein kinase B, RSK1 p90 ribosomal S6 kinase 1, AMPK AMP kinase, and GAP GTPase-activating protein.

myomatosis cells but has only rarely been found in cerebral cortical tubers.⁴⁶⁻⁴⁸ This observation may indicate that either inactivation of both alleles is not required for tuber pathogenesis or only a subgroup of cells within a tuber is affected by the second hit.

FUNCTIONS OF TSC1 AND TSC2

TSC PROTEINS AND INTERACTING FACTORS

TSC1 encodes TSC1 (hamartin), a 140-kD protein with no homology to TSC2 (Fig. 3). TSC2 encodes TSC2 (tuberlin), a 200-kD protein with a GAP domain near the carboxy terminal (Fig. 3). TSC1 and TSC2 interact physically with high affinity to form heterodimers,^{49,50} an observation that is consistent with the similar clinical features of patients with TSC1 and TSC2 mutations. TSC1 and TSC2 are coexpressed in cells within multiple organs, including the kidney, brain, lung, and pancreas.^{51,52} TSC2 has been localized to the Golgi apparatus⁵³ and the nucleus,⁵⁴ and TSC1 to the centrosome.⁵⁵

Substantial progress has been made in the past 5 years toward understanding the normal cellular functions of the TSC1–TSC2 protein complex.⁵⁶⁻⁶⁶ Much of this progress has been deduced from studies in the fruit fly, *D. melanogaster*. The TSC1–TSC2 complex interacts with several proteins (Fig. 4),⁶⁷⁻⁷⁵ although in most cases the clinical relevance of these interactions is not yet well understood. One of the first mechanistic clues to the roles that TSC1 and TSC2 have in cell function was the finding that mutations in the drosophila Tsc1 and Tsc2 homologues increased cell and organ size.⁷⁶⁻⁷⁸ Subsequent experiments demonstrated that the TSC1–TSC2 heterodimer inhibits the mammalian target of rapamycin (mTOR) cascade. In normal cells, direct phosphorylation and inactivation of TSC2 by protein kinase B (AKT) leads to mTOR activation.^{79,80} A serine–threonine kinase, mTOR has a central role in the control of cell growth and proliferation through the regulation of ribosome biosynthesis and protein translation. It functions by phosphorylating two effector molecules — p70S6 kinase and 4E-binding protein 1 (4E-BP1) — to increase cell growth and proliferation in response to growth factors, amino acids, and nutrients. By phosphorylating ribosomal protein S6, p70S6 kinase causes increased ribosome biogenesis. The phosphorylation of 4E-BP1 permits messenger RNA (mRNA) translation. In TSC-associated tumors, loss of TSC1 or TSC2 results in mTOR-dependent phosphorylation of p70S6 kinase, ribosomal protein S6, and 4E-BP1.^{81,82}

Rheb (RAS-homologue expressed in brain), a member of the RAS superfamily, is a specific GTPase downstream of TSC2 that functionally links the TSC1–TSC2 complex to the mTOR pathway.⁸³⁻⁸⁶ Rheb, like other RAS family members, cycles between an active GTP-bound state and an inactive GDP-bound state. TSC2 stimulates the conversion of Rheb–GTP to Rheb–GDP, thereby inactivating Rheb. Loss of TSC2 function leads to enhanced Rheb–GTP signaling and mTOR activation. TSC2 mutations in the GAP domain have low GAP activity with respect to Rheb,⁸⁷ suggesting that the GAP activity of TSC2 is essential for its physiologic function. Since patients with germline TSC1 mutations and those with TSC2 mutations have nearly identical phenotypes, it seems likely that TSC1 participates in the regulation of TSC2-related GAP activity with respect to Rheb, but the precise role of TSC1 is not yet clear. Given

the critical role of TSC1–TSC2 in the regulation of Rheb and mTOR activation, it is not surprising that the complex is subject to an intricate and incompletely understood series of phosphorylation events.⁶⁷⁻⁷⁴

TSC1–TSC2 SIGNALING AND CLINICAL MANIFESTATIONS OF TSC

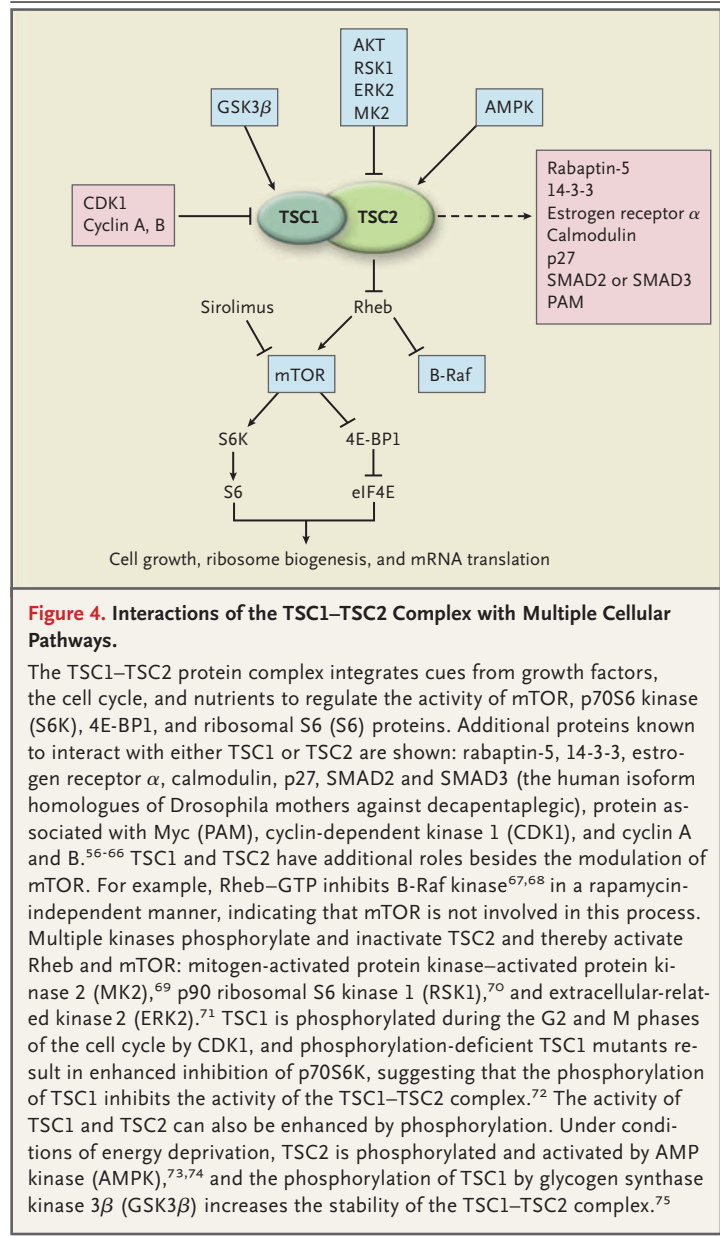
ABERRANT DIFFERENTIATION IN RENAL ANGIOMYOLIPOMAS

Loss of heterozygosity at the *TSC1* or *TSC2* locus and hyperphosphorylation of ribosomal protein S6 have been documented in each of the three components of angiomyolipomas (vessels, smooth muscle, and fat), suggesting that all three components arise from a common progenitor and that the TSC1–TSC2 complex regulates the differentiation of cells that are derived from mesenchyme. The smooth-muscle component of angiomyolipoma is histologically and immunophenotypically identical to the smooth-muscle cells of lymphangiomyomatosis, suggesting the existence of a pathogenic connection between the two disorders.

THE “BENIGN METASTASIS” HYPOTHESIS

Women with the sporadic form of lymphangiomyomatosis do not have germline *TSC1* or *TSC2* mutations.⁸⁸ Approximately 60% of such patients have renal angiomyolipomas. In patients with both sporadic lymphangiomyomatosis and angiomyolipoma, identical somatic *TSC2* mutations have been identified in the abnormal lung and kidney cells but not in the normal cells,^{89,90} suggesting that lymphangiomyomatosis and angiomyolipoma cells are genetically related and most likely arise from a common progenitor cell. These data have led to the “benign metastasis” hypothesis for the pathogenesis of lymphangiomyomatosis (Fig. 5), which proposes that histologically benign cells with mutations in *TSC1* or *TSC2* may have the ability to travel to the lungs from angiomyolipomas in the kidney.⁹⁰

Cells lacking *TSC1* or *TSC2* have an altered capacity for motility and migration. The expression of *TSC1* and *TSC2* is associated with the activation of Rho, a small GTPase^{63,91} that regulates the actin cytoskeleton and focal adhesions, and *TSC2*-deficient smooth-muscle cells exhibit increased migration in vitro.⁵⁸ These observations are consistent with a model in which *TSC2*-defi-



cient cells have increased migratory potential. The fact that pulmonary lymphangiomyomatosis occurs only in women has led to the hypothesis that estrogen regulates TSC signaling and, perhaps, also the migration of *TSC2*-deficient cells. Furthermore, the carboxy terminal of *TSC2* interacts with the estrogen receptor α and functions in vitro as a transcriptional corepressor of the estrogen receptor.⁵⁸ However, the in vivo role of estrogen in the pathogenesis of this disease is not yet understood.

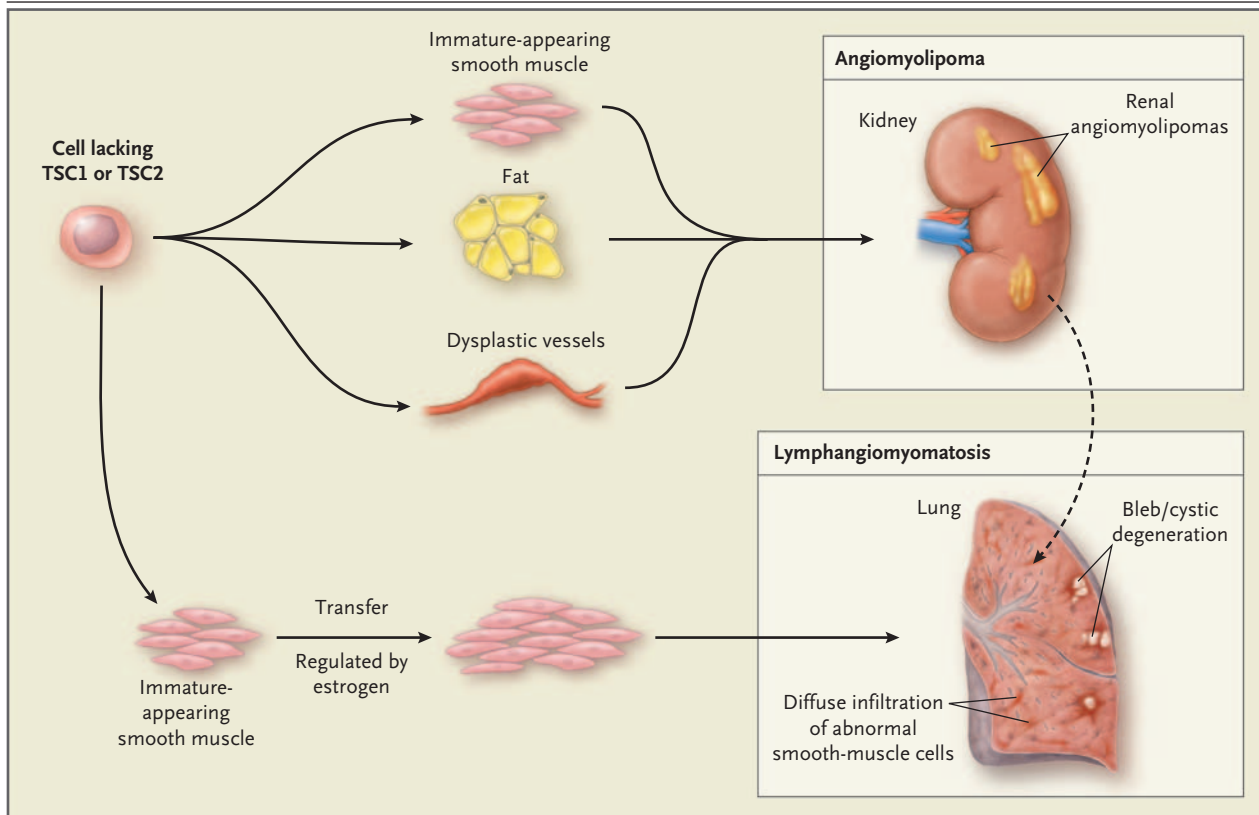


Figure 5. Model of the Pathogenesis of Angiomyolipomas and Lymphangiomyomatosis.

Molecular genetic data indicate that all three components of angiomyolipomas are derived from a common precursor cell. The pathogenic mechanism of lymphangiomyomatosis involves the aberrant migration of smooth-muscle cells harboring a somatic TSC gene mutation to the lung. Because lymphangiomyomatosis occurs almost exclusively in women, estrogen may promote this migration, although the targeting mechanism is not yet understood. It has been hypothesized that cells with *TSC1* or *TSC2* mutations may travel to the lungs from angiomyolipomas in the kidney (indicated by a dashed arrow).

CELL-SELECTIVE ACTIVATION OF mTOR IN TUBERS AND SUBEPENDYMAL GIANT-CELL TUMORS

Analysis of surgically resected tubers has revealed cell-specific activation of the mTOR cascade in giant cells, as evidenced by the expression of activated (phosphorylated) components of the mTOR cascade, including phosphorylated p70S6 kinase and phosphorylated ribosomal protein S6 (Fig. 2).^{92,93} Since mTOR is a critical regulator of cell size, it is logical to infer that the activation of mTOR is responsible for cytomegaly in tubers and subependymal giant-cell tumors. Interestingly, subependymal giant-cell tumor cells show high levels of phosphorylated p70S6 kinase, phosphorylated ribosomal S6, and phosphorylated Stat3 proteins, which are also indicative of mTOR activation.⁴⁶ The loss of heterozygosity detected in subependymal giant-cell tumors provides evidence that biallelic *TSC* gene inactivation

leads to the activation of mTOR and to cytomegaly.⁴⁶

PRACTICAL MANAGEMENT

In most patients with TSC, the first management issue is making the appropriate diagnosis by identifying major and minor diagnostic features. For initial diagnostic evaluation, careful dermatologic examination of the skin, including use of a Wood's lamp and funduscopic examination to identify retinal hamartomas, is necessary. In infants, echocardiography may reveal rhabdomyomas. MRI or CT of the brain is indicated to identify tubers and subependymal giant-cell tumors. Visualization of the kidneys by ultrasonography, CT, or MRI is warranted to identify angiomyolipomas. In women with TSC, CT of the lungs is indicated to look for subclinical lymphangiomyomatosis, and pul-

monary-function tests may provide a measure of disease progression. Currently, there are at least 15 TSC clinics across the United States that specialize in the diagnosis, care, and treatment of patients with the disorder. The clinical staff at these multidisciplinary centers includes specialists such as neurologists, dermatologists, geneticists, and pulmonologists. For the general practitioner, referring a patient to a regional TSC clinic should ensure that he or she will receive a comprehensive evaluation of the multisystem complications in TSC. If referral to a TSC clinic is not feasible, then subspecialty evaluation of the patient for individual manifestations of TSC, including lymphangio-myomatosis, angiomyolipomas, and epilepsy, is prudent. Physicians and patients can also gain useful information from the Tuberous Sclerosis Alliance Web site (www.tsalliance.org).

The second important issue in the management of TSC is long-term follow-up, including the monitoring of lesion growth. In particular, the growth of angiomyolipomas or subependymal giant-cell tumors requires continued vigilance. No conclusive guidelines for surveillance have been established for this disease, but most centers periodically image the brain and abdomen to monitor the growth of lesions in the brain and kidney.⁹⁴ (The Tuberous Sclerosis Alliance provides suggestions for surveillance on its Web site at www.tsalliance.org/pages.aspx?content=10.) For example, it is standard practice to perform brain and abdominal imaging at least every 3 years, and more often in patients with brain or renal lesions that have progressive growth. Annual MRI of the brain is suggested in patients until they are at least 21 years of age, and then MRI should be done every 2 to 3 years both to diagnose and to monitor subependymal giant-cell tumors.²⁶ In patients with multiple angiomyolipomas or a single lesion that is progressive, yearly ultrasonography, MRI, or CT is indicated.

In patients with lymphangio-myomatosis, annual pulmonary-function testing may be useful to monitor lung function, and some patients may require more frequent assessments. Although electroencephalography is not part of the diagnostic workup for TSC, it remains an important tool in patients with TSC and epilepsy to define background cerebral activity, characterize patterns such as hypsarrhythmia in infantile spasms, and identify seizure foci. Periodic dermatologic evaluation is useful, since facial angiofibromas can cause

cosmetic disfigurement and ultimately require laser therapy or surgical removal. In general, lifetime surveillance for lesion growth in patients with TSC permits early recognition of potentially life-threatening complications.

Finally, genetic counseling should be offered to patients to aid with family planning. TSC is an autosomal dominant disorder; thus, those affected should be advised that the risk of having an affected child is approximately 50%. Genetic testing for *TSC1* and *TSC2* mutations is commercially available. Prenatal or preimplantation genetic testing is becoming more widely available.

THERAPEUTIC DEVELOPMENTS

Since tumor cells from patients with TSC activate mTOR, the mTOR inhibitor sirolimus has been identified as a potential therapeutic agent. Pre-clinical studies in mice have supported the usefulness of this approach⁹⁵; clinical trials of sirolimus in patients with TSC and in those with sporadic cases of lymphangio-myomatosis are ongoing. A recent study demonstrated regression of subependymal giant-cell tumors in patients after sirolimus therapy.⁹⁶ One concern is that sirolimus treatment may restore the cell's ability to activate AKT,⁹⁷ suggesting that long-term treatment may increase the risk of malignant tumors in patients with TSC. Clearly, both the short- and long-term consequences of mTOR inhibition in such patients require further study.

CONCLUSIONS

The TSC1-TSC2 complex plays a central role in the integration of multiple cues to regulate cellular growth and differentiation, and mutations in *TSC1* or *TSC2* result in widespread, devastating consequences. Key priorities for future research include elucidating the location of and functional relationship between TSC1 and TSC2 and their pathways, determining whether Rheb is the sole downstream effector of the TSC1-TSC2 complex and whether mTOR is the only clinically relevant target of Rheb, understanding the relationship between tubers and epilepsy, and investigating the role of estrogen in the pathogenesis of lymphangio-myomatosis. The recent delineation of the TSC biochemical signaling pathway suggests strategies for developing targeted therapies including mTOR inhibition, which is being evaluated in clinical trials.

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