Pediatric Neurology 89 (2018) 3–10

Contents lists available at ScienceDirect

Pediatric Neurology

journal homepage: www.elsevier.com/locate/pnu

Review Article

Genetic Evaluation of Common Neurocutaneous Syndromes

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ARTICLE INFO

Article history:
Received 8 April 2018
Accepted 6 August 2018

Keywords:
Neurocutaneous syndromes
Neurofibromatosis
Tuberous sclerosis complex
Sturge-Weber syndrome
Genetic approach
Mosaicism

ABSTRACT

The neurocutaneous syndromes are a group of multisystem disorders that affect the skin and central nervous system. Neurofibromatosis 1, neurofibromatosis 2, tuberous sclerosis complex, and Sturge-Weber syndrome are the four major neurocutaneous disorders that mainly present in childhood. In this review, we discuss the clinical findings and genetic diagnosis, related genes/pathways and genotype–phenotype correlations of these four neurocutaneous syndromes.

Introduction

The neurocutaneous syndromes comprise a variety of disorders that include abnormalities of both the skin and the central nervous system (CNS). These disorders have also been characterized as phakomatoses, a term suggested by Dr. Jan van der Hoeve in 1923. The skin and CNS share a common ectodermal embryologic origin. Several of the neurocutaneous syndromes may be related to defective neural crest ontogeny, and these conditions are sometimes characterized as "neurocristopathies."[1-3] Neurocutaneous syndromes vary widely in their clinical manifestations as well as their genetic basis.[2,4,5] Most neurocutaneous syndromes represent single-gene disorders but exhibit various patterns of inheritance. While neurofibromatosis type 1, neurofibromatosis type 2, tuberous sclerosis complex and von Hippel-Lindau syndrome have autosomal dominant inheritance pattern, incontinentia pigmenti is X-linked and Sturge-Weber syndrome has a sporadic inheritance pattern.[6]

Here we discuss the diagnostic criteria, clinical features, genetic diagnosis, genotype–phenotype correlation as well as molecular pathways of the most common neurocutaneous syndromes.

Neurofibromatosis type 1

Neurofibromatosis type 1 (NF1), is one of the most common genetic disorders, with an incidence of approximately one in 3000 people.[7] NF1 is inherited as an autosomal dominant trait, but only half of the patients have an affected family member with NF1. Although the penetrance of NF1 approaches 100%, its expressivity is highly variable. The phenotype of NF1 can differ markedly depending on the type of the mutation, and the clinical features vary even among affected family members.[8]

NF1 is frequently diagnosed early in childhood. Diagnostic criteria have been established for NF1, but children less than two years of age are less likely to meet them.[9] Two of these clinical features are necessary for the diagnosis of NF1: (1) six or more café-au-lait macules with diameters greater than 5 mm in a prepubertal patient and greater than 15 mm in a postpubertal patient, (2) two or more neurofibromas or one plexiform neurofibroma, (3) skinfold (axillary or inguinal) freckling, (4) optic pathway tumor, (5) two or more iris hamartomas,

Conflict of interest: The authors declare no conflict of interest or financial disclosures concerning the materials or methods used in this study or the findings specified in this article.

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https://doi.org/10.1016/j.pediatrneurol.2018.08.006
0887-8994/

(6) characteristic bony lesion, (7) first-degree relative with NF1.[10,11]

Gene linkage studies primarily localized the *NF1* gene to the pericentromeric region of chromosome 17 in 1987. After that, the *NF1* gene was described by positional cloning experiments at 17q11.2.[12,13] The *NF1* gene encodes neurofibromin. It is a large multidomain protein, spanning approximately 250 kDa and containing at least 60 common and three alternatively spliced exons. Neurofibromin is a negative regulator of rat sarcoma virus (RAS) signaling and is also expressed in migrating neural crest cells during early fetal development. RAS requires guanosine triphosphate (GTP) binding to stimulate the mitogen-activated protein kinase (MAPK) pathway, which initiates cell proliferation. Neurofibromin diminishes the activity of RAS by increasing the hydrolysis of GTP to guanosine diphosphate (GDP) (Fig). Loss-of-function mutations in this gene lead to a predisposition for tumorigenesis due to improper activation of the RAS-MAPK pathway.[14,15] Additionally, due to increased RAS activity, NF1 is classified as a RASopathy and disorders other than NF1 (i.e., Noonan syndrome, cardiofaciocutaneous syndrome) are also caused by mutations in the RAS-MAPK pathway and share some clinical characteristics with NF1.[16,17] Furthermore, neurofibromin is a positive regulator of intracellular cyclic AMP production, which is responsible for maintaining neuronal viability in neurons in the setting of optic glioma.[18]

The *NF1* gene has a wide mutational spectrum (Table 1) due to the high number of coding exons. The mutations generally result in truncation and loss of function of neurofibromin and represent approximately 16% of the coding region.[19] Although some hot spots with a higher mutation rate (especially exons 4b, 7, 10b, 13, 15, 20, 29 and 37) have been described, most of the mutations are private. Truncating mutations and deletions are associated with a more severe clinical phenotype, while missense and in-frame mutations are usually associated with milder clinical manifestations. In a recent study, the authors suggested that a recurrent three base-pair deletion, c.2970-2972delAAT (p.Met992del), in exon 17 is associated with a mild phenotype. Café-au-lait macules and skinfold freckling can occur in this small in frame deletion in the absence of visible cutaneous or plexiform neurofibromas.[20] In another study, individuals with missense mutations of codon p.Arg1809 had Lisch nodules and similar clinical features as patients with three base-pair in frame deletion [c.2970-2972delAAT (p.Met992del)]. Moreover, the individuals may exhibit Noonan-like characteristics.[21,22] Five to ten percent of pathogenic *NF1* mutations include large or complete gene deletions that may be associated with intellectual disability, facial dysmorphism, cardiovascular abnormalities, higher benign tumor burden, and a higher frequency of malignant nerve sheath tumors. The size of these deletions is approximately 1.2-1.4 Mb.[23-25] Furthermore, the *NF1* deletions can be divided into four different types based on their size and breakpoint location (type 1, 2, 3 and atypical).[26] Nevertheless, it is not always possible to detect a clear genotype–phenotype correlation.

Segmental neurofibromatosis, also known as mosaic-localized neurofibromatosis (MNF1), is a rare NF1 variant that is characterized by cutaneous lesions limited to a circumscribed body segment. In the general population, the prevalence of MNF1 is approximately one in 36,000 to 40,000 individuals, but it is probably underreported.[27] Mutation usually occurs during the late embryonic development which provokes localized disease. Most patients do not have any family history of the disease.[28] MNF1 has also been divided into four groups on the basis of clinical features: (1) pigmentary anomalies only, (2) pigmentary anomalies and neurofibromas, (3) neurofibromas only, and (4) plexiform neurofibromas only.[29-30] Patients with MNF1 have milder clinical features and exhibit fewer complications compared with those with complete NF1. Individuals with gonadal mosaicism have few manifestations of NF1 but can have affected offspring with NF1.[27]

There are several disorders which are sometimes confused with NF1 (Table 2). These include a group of genetic syndromes (known as RASopathies) resulting from mutations in genes that encode the components or the regulators of the RAS/MAPK pathway. One of these syndromes is Legius syndrome and the affected patients may fulfill the diagnostic criteria for NF1. Multiple café-

TABLE 1. Molecular Genetic Testing Used in Neurofibromatosis Type 1 and Tuberous Sclerosis Complex

Gene	Test Method	Detectable Rate	
NF1	Genomic DNA sequence analysis	~60%-90%	
	Gene-targeted deletion/duplication analysis	~5%	
	Chromosomal microarray	~5%	
	Cytogenetic analysis	<1%	
TSC1	Genomic DNA sequence analysis	**Familial**	**Nonfamilial**
		~9.8%	~13.5%
	Gene-targeted deletion/duplication analysis	~0.1%	~0.5%
TSC2	Sequence analysis	13.8%	~50%
	Gene-targeted deletion/duplication analysis	~0.2%	~2%

Adapted from *Northrup H, Koenig MK, Pearson DA, et al (2015) Tuberous Sclerosis Complex. In: Adam MP, Ardinger HH, Pagon RA et al (eds) GeneReviews® [Internet]. Seattle (WA): University of Washington, Seattle; 1993-2017. *Friedman JM. Neurofibromatosis 1. 1998 Oct 2 [Updated 2018 Jan 11]. In: Adam MP, Ardinger HH, Pagon RA, et al., editors. GeneReviews® [Internet]. Seattle (WA): University of Washington, Seattle; 1993-2018.

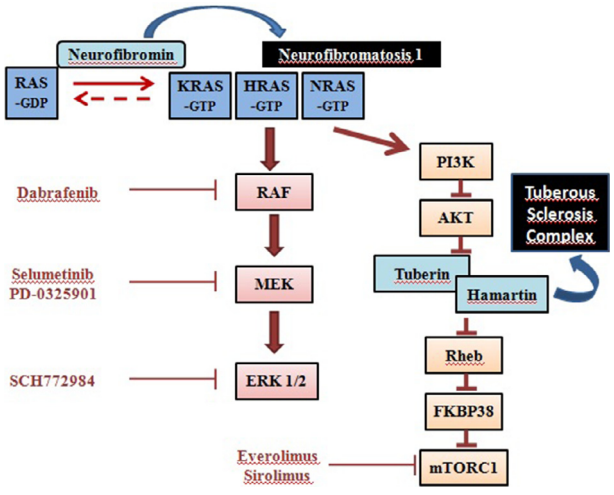

FIGURE. The scheme of the RAS/MAPK and mTOR pathways.

au-lait macules, axillary freckling, and macrocephaly can occur in individuals with Legius syndrome, but Lisch nodules, neurofibromas, and central nervous system tumors are not usually present. This syndrome results from a heterozygous mutation the sprouty related EVH1 domain containing 1 (*SPRED1*).[31] The *SPRED1* gene encodes a protein that negatively regulates RAS-MAPK signaling, and pathogenic loss-of-function variants of this gene result in attenuated inhibition of downstream RAF-MEK-ERK signaling and a consequent increase in RAS signal propagation.[32] LEOPARD syndrome is usually characterized by multiple lentigines and has clinical characteristics similar to the RASopathies. Protein tyrosine phosphatase, nonreceptor type 11 (*PTPN11*; 85%) gene is responsible for LEOPARD syndrome. Molecular diagnosis is relatively important because otherwise it is not easy to distinguish this syndrome from other RASopathies.[33] The *PTPN11* gene encodes tyrosine-protein phosphatase nonreceptor type 11, which has a role in modulating cellular proliferation, differentiation, migration, and apoptosis.[34] *PTPN11* gene is also responsible for approximately 50% of the patients with Noonan syndrome which is also a RASopathy. Neurofibromatosis-Noonan syndrome (NFNS) is a version of NF1 rather than Noonan syndrome. These patients may have the features for NF1 with café-au-lait macules and skin-fold freckling and also have some overlapping features with Noonan syndrome, including dysmorphic facial features, short stature, and congenital heart defects. The major gene involved in NFNS is *NF1*, but co-occurring *NF1* and *PTPN11* gene mutations in patients with NFNS have been reported in the literature.[35,36] It should be noted that the molecular diagnosis is important for genetic counseling, prognosis, and monitoring of potential risks.

Tumor-related phenotype of NF1 can be explained by the "second hit" theory, where the germline mutation serves as a "first hit" and another cell-specific event (the "second hit") must occur in a specific cell in order for the clinical manifestation to develop. This "second hit" contributes to the highly variable expression of the disease due to occurrence of these events in different cell numbers, types, locations, and ages in patients with NF1.[37]

Biologically targeted therapies have focused on management of the most severe NF1 complications. Signal transduction inhibitors including RTK, RAS, RAF, MEK, ERK, mTOR and PI3K inhibitors have been used for the management of NF1. MEK inhibitors target their conserved allosteric binding pocket, blocking phosphorylation, and subsequent activation. These drugs have several potential drawbacks including toxicity and acquired resistance. On the other hand, novel ERK inhibitors have a direct effect on ERK activity and inhibit MEK-mediated phosphorylation of ERK and could be considered for treatment of NF1. Furthermore, co-targeting mTOR and MEK-ERK pathways is likely to be more effective in several situations than using a single agent.[38]

NF1 is a multisystem, relatively common disorder. At each age, there are various clinical manifestations that may develop, necessitating detailed assessment of the individuals. Molecular analysis is significant for early diagnosis of NF1 and clarification of ambiguous cases. Genetic counseling should be offered to patients who are affected or at risk. Once the pathogenic variant has been identified in an affected family member, prenatal testing for a pregnancy at increased risk and preimplantation genetic diagnosis are possible.

Neurofibromatosis type 2

Neurofibromatosis type 2 (NF2), previously known as bilateral acoustic neurofibromatosis or central neurofibromatosis, is an autosomal dominant disorder with an incidence of approximately one in 35000 to 40000

TABLE 2. Disorders Most Frequently Confused With NF1

Syndrome	Most Related Gene (Proportion)	Inheritance Pattern	Clinical Features
Legius syndrome, NF1-like syndrome	*SPRED1*	AD	≥6 cafe-au-lait macules (approximately >%80 of the patients) Intertriginous freckling Lacks neurofibromas, Lisch nodules, optic gliomas and osseous lesions of NF1
LEOPARD syndrome	*PTPN11(%85)*	AD	Multiple lentigines mostly on face, neck, and upper part of the trunk Lentigines present in early infancy/childhood Similar clinical characteristics of RASopathies
Noonan syndrome	*PTPN11(%50)*	AD	Dysmorphic craniofacial features, cardiac defect, musculoskeletal abnormalities Cafe-au-lait macules (nearly 10%)
Piebaldism	*KIT*	AD	Depigmented patches of skin and hair Spots or patches of pigmented skin within or around the borders of the unpigmented areas
McCune-Albright syndrome	*GNAS1*	Sporadic	Cafe-au-lait macules which usually appear in infancy Polyostotic fibrous dysplasia Hyperfunction of endocrine glands
Mismatch repair cancer syndrome	*MSH2, MSH6, MLH1, PMS2*	AR	Cafe-au-lait macules, axillary freckling Variable neoplasms

GNAS1, guanine nucleotide binding protein (G protein), alpha stimulating activity polypeptide 1; KIT, KIT proto-oncogene receptor tyrosine kinase; MLH1, mutL homolog 1; MSH2, mutS homolog 2; MSH6, mutS homolog 6; PMS2, PMS1 homolog 2, mismatch repair system component; PTPN11, protein tyrosine phosphatase, nonreceptor type 11; SPRED1, sprouty related, EVH1 domain containing 1.

people.[39] Bilateral vestibular schwannomas (rarely unilateral), schwannomas of different parts of the neural system, meningiomas, and other low grade CNS malignancies can be observed in patients with NF2.[40] Ocular abnormalities including early-onset cataracts, optic nerve sheath meningiomas, hamartomas of the retina or retinal pigment epithelium are common. Subcutaneous nodular schwannomas and flat dermal plaques can also be seen. Furthermore, cutaneous signs in NF2 are much more subtle than in NF1 and more than six cafe-au-lait macules are rarely detected.[41]

There are generally two types of NF2 due to the clinical severity: the more severe "Wishart form" and the milder "Gardner form." The other form is congenital NF2 which is linked with bilateral vestibular schwannomas detected in the first days to months of life. In congenital NF2, the patients can be asymptomatic for one to two decades with usually unexpected and rapid progression; this form may be associated with NF2 plaques in atypical locations and other CNS tumors.[41]

The NF2 gene is located on chromosome 22q12.2 and has 16 coding exons. It encodes a 595 aminoacid protein called merlin or schwannomin. Merlin is a negative regulator of Schwann cells and the loss of function of this protein allows overproduction of Schwann cells. Merlin has been divided into three structurally different domains: (1) an amino-terminal FERM (Fourpoint-one, ezrin, radixin, and moesin) domain; (2) an α-helical coiled-coil domain; and (3) a carboxy-terminal hydrophilic tail. FERM domain, especially blue-box motif" within the subdomain F2, has a significant role in the merlin's antitumor effect. This domain is also identical to the residues 177-183 of merlin.[11,42,43]

Different types of mutations (single-base variations, insertions, and large multi-exon or whole-gene deletions) have been identified in the NF2 gene. In contrast to NF1, there is a strong genotype–phenotype correlation both with the type and the position of the NF2 mutation. Pathogenic mutations can be detected in 35% to 66% of the patients and germline mutations occur throughout the initial 15 exons. The most common mutations are nonsense mutations at CpG islands.[41] Although a truncating mutation (nonsense and frameshift) may be associated with serious features and early onset of clinical features, intracranial schwannomas and meningiomas which may cause blindness, hearing loss, paralysis, and death at age of 40 years; missense mutations may cause predominantly a mild phenotype.[39,44] Patients with mutations in exons 9-15 may also have a milder form of the disease. However, missense mutations that involve residues 177-183 in the F2 domain can be associated with a more serious phenotype. Splice site mutations may cause a variable phenotype. Besides, large scale rearrangements (especially the promoter region, exon 1, intron 1 deletions, and deletions of the whole gene) accounted for up to 15% of the patients which is associated with a milder phenotype.[43,44] *NF2* is also a tumor suppressor gene and the two alleles of the gene should be inactivated for initiation of tumorigenesis. In the beginning, all Schwann cells have a mutant allele of the gene but tumorigenesis occurs in case of a deletion or mutation in the other allele.[40]

A group of patients who are being evaluated for schwannomatosis develop multiple nonvestibular, nonintradermal cranial, spinal and peripheral schwannomas. Schwannomatosis is a different disorder from NF1 and NF2 with a 1/40.000 to 1/100.000 incidence.[45] Some patients with multiple schwannomas meet the diagnostic criteria of NF2. Furthermore, mosaic *NF2* pathogenic variants have been detected in several patients with schwannomatosis.[46] *SMARCB1* and *LZTR1* genes are responsible for schwannomatosis which are located at 22q11.23 and 22q11.21, respectively.[43] *SMARCB1* gene is a subunit of the SWI/SNF complex and exerts its tumor suppressor function by regulating the cell cycle. About 10% to 15% of the patients with schwannomatosis have a positive family member and *SMARCB1* mutation can be detected in 40% to 50% of these familial cases. The other gene *LZTR1* variants can be defined in about ~80% of the patients with no detectable mutations in *SMARCB1* gene and LZTR1 protein is involved in multiple cellular processes including regulation of chromatin conformation and the cell cycle.[45,47-49] But schwannomatosis is only partially explained by mutations in these two genes. The clinical phenotype of tumorigenesis in schwannomatosis can be explained by other complex mechanisms (the four hits-three steps model).[50]

Some of the patients with segmental or mosaic NF2 may have a unilateral eighth-nerve Schwannoma associated with ipsilateral meningiomas or schwannomas.[51] In these patients, the *NF2* gene mutation occurs after fertilization. Standard molecular techniques can detect the mutation when more than 10% of lymphocytes have the mutation. Furthermore, the ratio of affected cells can vary in different tissues.[52] As fewer cells receive a second hit in mosaic individuals, the patients may have a milder phenotype.[40]

Due to autosomal dominant inheritance, half of the offspring of individuals with NF2 are affected. Family history is present in nearly half of all NF2-affected patients. Clinical findings and phenotype tend to be similar within the families and early screening is recommended for diagnosis of tumors presymptomatically. About one third of the patients are considered to be mosaic. Moreover, molecular genetic techniques can identify the *NF2* gene mutations in about 70% to 75% of the patients. However, blood mutation analysis may not show the pathogenic mutation, therefore tumor tissue analysis is necessary in sporadic cases for identification of a mosaic mutation.[40,43]

Tuberous sclerosis complex

Tuberous sclerosis complex (TSC) is inherited as an autosomal dominant trait. TSC has an estimated incidence of one in 6000 births.[53] It's manifestations incude seizures, skin lesions, intellectual disability, and atypical tumor-like growths. TSC was initially described in 1835 by Rayer. In 1862, von Recklinghausen described a child with cardiac tumors and a large number of scleroses in the brain.[54,55] In 1880, Bourneville coined the term tuberous sclerosis, better documented the neurological

features, and described additional dermatologic abnormalities.[56]

The clinical features of TSC are variable, even among the affected family members with the same mutation. The brain and the skin are affected in more thn 90% of the patients. The major cutaneous manifestations are hypomelanotic macules ("ash leaf" lesions), facial angiofibromas, shagreen patch, and subungual fibromas. Cardiac rhabdomyomas shown by prenatal ultrasound can be the earliest signs of TSC. The presence of a single cardiac rhabdomyoma in a newborn is associated with a 25% risk of TSC, while multiple rhabdomyomas are associated with TSC over 90% of the time.[57] While multiple retinal hamartomas occur in 30% to 50% of the patients, 39% of the patients have retinal achromic patches.[58] The patients may suffer from renal disorders such as angiomyolipoma, renal cysts, and carcinoma. Benign epithelial cysts occur in 20%-50% of the patients.[59] The most common neurological features include epilepsy, autism spectrum disorders, and intellectual impairment. Seizures can be observed in most of the patients; infantile spasms are particularly seen during the first year of life which may be the first clue to diagnosis of TSC. On the other hand, some patients may not have any neurological impairment.[60] In 2012, the TSC diagnostic criteria were modified and genetic testing of the *TSC1* or *TSC2* genes was included at the International Tuberous Sclerosis Complex Consensus Conference.[61]

Mutations of the genes *TSC1* and *TSC2* have been responsible for the pathogenesis of TSC and lead to overactivation of the mTOR pathway (Fig). In normal cells, the mTOR pathway plays an essential role in growth, proliferation, and survival of the cells. *TSC1* binds to *TSC2* and this stabilizes the intracellular *TSC2* levels which serves to regulate the GTPase activating protein (GAP) activity. Therefore inactivating mutations in either gene give rise to the same clinical disorder.[62] Stimulant agents, such as growth factors, bind to tyrosine kinase receptors and cause the phosphorylation of PI3K. This activation of PI3K results in a cascade of phosphorylation events, especially leading to activation of AKT, which in turn inhibits the TSC complex. Consequently, TSC-related hamartomas (angiomyolipomas, facial angiofibromas, etc.) can be seen. Furthermore, hypoxia-inducible factor-1a, which is indirectly activated by mTORC1, also promotes tumor development.[60,63,64]

The *TSC1* gene is located on chromosome 9q34.1 and encodes hamartin (130 kD), while the *TSC2* gene is located on chromosome 16p13.3 and encodes tuberin (180 kD).[63] The incidence of mutations in *TSC2* gene is approximately five times higher than the rate of *TSC1* gene mutations, and *TSC2* mutations are associated with a more severe clinical phenotype.[65] Approximately 85% of the patients have a mutation in one of these genes and more than 1200 allelic variants have been identified (Table 1). Point mutations (missense, nonsense), splice site mutations, and small deletions are usually reported in both of these genes. Large rearrangements are responsible for 17.4% of the mutations in *TSC2*, a higher number than the frequency of large rearrangements in the *TSC1* gene.[60,62,64,66] Missense mutations of *TSC2* gene typically present with phenotypes that are as severe as *TSC2* nonsense mutations and more severe than nonsense and missense mutations of *TSC1*.[67] Nevertheless, there have been some reported associations between particular *TSC2* missense mutations and generally mild phenotypes, namely R905Q, R1200W, S1036P, R1713H, and G1579S.[68] Furthermore, the phenotype is usually milder in patients who do not have any detectable mutation.[59]

The *TSC2* gene is adjacent to the *PKD1* gene (a mutation leads to polycystic kidney disease) on chromosome 16p13. Some of the patients have a genomic deletion involving both of these continuous genes. Severe clinical phenotypes (significant renal insufficiency, need for dialysis, renal transplantation at early age) can be seen in the patients. The mechanism by which deletion of *TSC2* and *PKD1* genes generate polycystic kidney disease is not clear.[60]

TSC1 and *TSC2* are tumor suppressor genes, and two hits are required for promoting tumor formation. The first hit is the inherited germline mutation, and the second hit is somatic. While TSC lesions may develop by somatic inactivation of *TSC1/TSC2*, second hits are not always observed, especially in the brain lesions.[69] Furthermore, there are several potential mechanisms for somatic inactivation of the wild-type alleles, including loss of heterozygosity, mutation, and promoter methylation.[70]

Several drugs have been approved for the management of TSC. mTORC1 inhibitors (everolimus, sirolimus), referred to as Rapalogs, decrease the size of subependymal giant cell astrocytomas, angiofibromas, and angiomyolipomas, and delay the loss of lung function in lymphangioleiomyomatosis. mTORC1 inhibitors result in a cytostatic response rather than cytotoxic effects, with suppression while on therapy and regrowth when therapy is discontinued. Therefore continuous therapy is required. Long-term treatment can also induce several side-effects, such as interstitial pneumonia and to avoid these side-effects arising from systemic treatment, a topical sirolimus formulation has been developed for skin lesions.[60,71]

TSC usually develops as a result of *de novo* germline mutation in one of these two genes. *TSC2* gene has a larger coding region and sporadic mutations occur more frequently than *TSC1*. Nevertheless, in familial cases, the identification of *TSC1* mutations appears to be higher compared with sporadic cases.[62] But due to the incomplete investigations of the extended families, the rates of spontaneous mutations are quite variable (66%-75%). Mosaic mutations have been also identified in some patients.[57] Molecular diagnosis of TSC and determination of genotype–phenotype correlations is significant and this may help in the establishment of personalized treatment and improve quality of life among these patients. Prenatal testing for pregnancies at increased risk may be also available.[72]

Sturge-Weber syndrome

Sturge-Weber syndrome (SWS) is a vascular neurocutaneous syndrome characterized by facial/capillary nevus (port-wine birthmark), leptomeningeal vascular malformation,

and ocular abnormalities, which include choroidal vascular anomalies and glaucoma.[73] SWS, which is also defined as encephalofacial angiomatosis, is the third most common neurocutaneous disorder after NF and TSC. It occurs sporadically, with an estimated incidence in male and female newborns, in 1/20,000 to 1/50,000 live births.[74]

SWS can be clinically classified into three different types and the most common is type 1 which is the classic manifestation of port-wine nevi and intracranial leptomeningeal capillary malformation.[75] A port-wine birthmark is a common finding caused by progressive ectasia of the vascular plexus of the dermis, typically affecting the forehead and cheeks in individuals with intracranial involvement. The skin lesion may be unilateral or bilateral and is present at birth.[76] The cerebral vascular malformations are usually unilateral (on the same side as the port-wine nevus) but can also occur bilaterally, and bilateral brain lesions is associated with a worse neurological prognosis. As well as capillary-venous leptomeningeal malformations, the other brain malformations including focal cortical dysplasia or polymicrogyria can also be observed.[77] Neurodevelopmental prognosis is highly variable. In fact, patients can suffer from progressive neurological problems, such as seizures, migraines, learning difficulties, or intellectual disability. On the other hand, unlike NF and TSC, patients with SWS do not have a greatly increased risk for tumor development.[78-80] The presence of epilepsy and frequency of seizures are associated with greater risk of having intellectual, behavioral, and mood abnormalities.[81] Furthermore, localized/generalized visceral vascular malformations can be seen in the kidneys, spleen, intestine, and the other organ systems.[82]

Diagnosis of SWS is based on typical clinical symptoms, facial appearance, and brain magnetic resonance findings. The syndrome is caused by a somatic mosaic mutation of the guanine nucleotide-binding protein alpha-q gene *(GNAQ)*, which occurs post-zygotically. The *GNAQ* gene, mapped to 9q21.2, encodes the human Guanine nucleotide-binding protein G(q) subunit alpha and is important for blood vessel development. The Gαq protein is part of the heterotrimeric GTP binding proteins, which transfer extracellular stimuli to intracellular signaling cascades through the G protein coupled proteins.[81] When activated by the G protein coupled proteins ligand, Gαq binds GTP and releases GDP, dissociates from the trimeric protein complex, and activates downstream pathways. Hydrolysis of GTP to GDP and re-association of the trimeric G protein with the GPRC causes the inactivation of these pathways. A somatic mosaic mutation in *GNAQ* gene affects the autohydrolysis site of the protein. Thus the ability of Gaq to bind to GDP is impaired which results in hyperactivation of these downstream pathways including RAS-MEK-ERK, HIPPO-YAP, and indirectly, mTOR.[83-85]

Somatic mutations of the *GNAQ* gene cause persistance of the primordial embryonic venous plexus, which is normally present at five to eight weeks of the embryonic period. This leads to venous hypertension, tissue hypertrophy, and formation of developmental vascular abnormalities that grow with the body.[86] The mutations in the Q209 and R183 residues of the *GNAQ* gene, including the Q209L substitution located in the Ras-like domain, are likely to exhibit involvement in the process of tumorigenesis via the upregulation of the MAPK pathway.[87] Q209L mutation is also associated with development of uveal melanoma.[88] Additionally, *GNAQ* gene mutations are found in patients with isolated port-wine birthmarks and no brain or eye involvement.[89] Recently the researchers identified the somatic *GNAQ* mutation c.548G>A (p.R183Q) in 88% of SWS patients and 92% of patients with nonsyndromic port-wine birthmarks. This mutation is located in the GTP-binding pocket of the G-α subunit and has an important role in GTP hydrolysis. They suggested that this mutation induced moderate activation of the extracellular signal-regulated kinase pathway, which may cause pathologic growth of capillaries in the skin and brain.[83,88] Although several studies and molecular findings have dramatically improved our understanding of the pathogenesis of SWS, the management of the disorder still remains challenging.

We discussed the clinical manifestations, molecular pathways, and related genes of the most common neurocutaneous syndromes (NF1, NF2, TSC, and SWS). Neurocutaneous syndromes can affect multiple organ systems. Early diagnosis allows the clinician to anticipate possible complications, make the necessary referrals, and to offer genetic counseling to the families. The advances in molecular genetics and a better understanding of the biologic functions of the genes/gene products may guide future therapeutic developments.

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