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Spinocerebellar ataxia type 6 and episodic ataxia type 2: differences and similarities between two allelic disorders

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Abstract. Spinocerebellar ataxia type 6 (SCA6) is one of three allelic disorders caused by mutations of CACNA1A gene, coding for the pore-forming subunit of calcium channel type P/Q. SCA6 is associated with small expansions of a CAG repeat at the 3′ end of the gene, while point mutations are responsible for its two allelic disorders (Episodic Ataxia type 2 and Familial Hemiplegic Migraine). Genetic, clinical, pathological and pathophysiological data of SCA6 patients are reviewed and compared to those of other SCAs with expanded CAG repeats as well as to those of its allelic channelopathies, with particular reference to Episodic Ataxia type 2. Overall SCA6 appears to share features with both types of disorders, and the question as to whether it belongs to polyglutamine disorders or to channelopathies remains unanswered at present.

Spinocerebellar ataxia type 6 (SCA6) belongs to the group of autosomal dominant cerebellar ataxias (ADCAs), which have an estimated prevalence around 3/100,000 inhabitants (Skre, 1974; van de Warrenburg et al., 2002). SCA6 accounts for 1–11% of ADCA families collected in European centres (Leggo et al., 1997; Riess et al., 1997; Stevanin et al., 1997; Takano et al., 1998; Pujana et al., 1999; Sinke et al., 2001; Silveira et al., 2002), 6–31% in Japanese (Matsuyama et al., 1997; Takano et al., 1998; Watanabe et al., 1998; Maruyama et al., 2002) and 0–11% in Chinese series (Tang et al., 2000; Soong et al., 2001a). As most ADCAs, SCA6 is due to expansions of a CAG repeat stretch, which is embedded in the 3′ coding region of a calcium channel gene, CACNA1A, on chromosome 19p13. The gene codes for the α_{1A} pore-forming subunit of voltage-gated calcium channels type P/Q, expressed predominantly in cerebellar Purkinje and granule cells. Point mutations at the CACNA1A gene are known to cause Episodic Ataxia type 2 (EA2) and Familial Hemiplegic Migraine (FHM) (Ophoff et al., 1996). SCA6 shares some features with these disorders, particularly EA2, and differs in many aspects from ADCAs with expansions of a CAG repeat.

Gene, mutation and inheritance

The CACNA1A gene maps on chromosome 19p13.2→p13.1 (Diriong et al., 1995) and covers about 300 kb with 48 exons (Ophoff et al., 1996; Trettel et al., 1998). Well conserved, the gene is expressed as a transcript of approximately 9.8 kb in the brain (Ophoff et al., 1996), more abundantly in the cerebellum than in other cerebral areas, and particularly in Purkinje cells.

The cDNA clones predict large peptides with molecular masses of 200 to 275 kDa with four homologous domains (I–IV), each containing six hydrophobic transmembrane segments, S1–S6 (Fig. 1A). The short N-terminal and the long C-terminal tails of the protein are located in the cytoplasm. In some isoforms a polymorphic polyglutamine sequence, coded by the CAG_n repeat expanded in SCA6, is present in the C-terminal tail.

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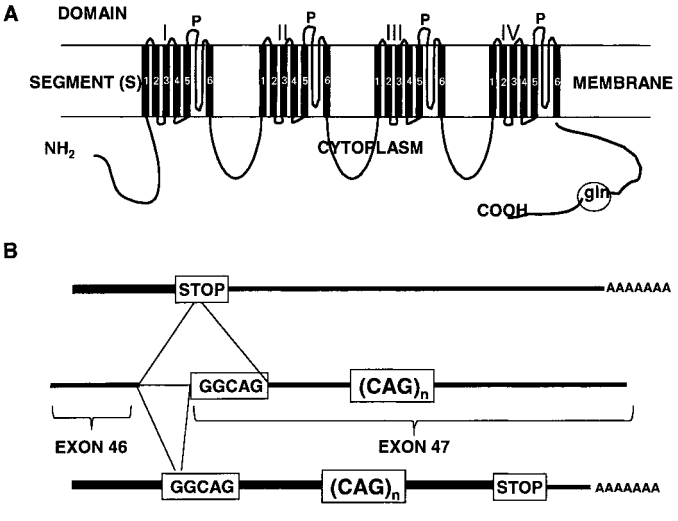

Fig. 1. (**A**) Schematic structure of the α_{1A} subunit of the voltage-gated calcium channel type P/Q. Each of the four domains (I–IV) has six α-helix transmembrane segments (S1–S6). The carboxyl terminus of the protein contains a stretch of polyglutamines (gln) in some of the isoforms (see text). (**B**) Alternative splicing between exons 46 and 47. If exon 46 is joined to exon 47 excluding the GGCAG segment, a TGA stop codon is encountered and the poly-CAG stretch is not expressed. Whereas, if exon 46 is joined to exon 47 including the GGCAG segment, the translation continues beyond the poly-CAG, until a TAA stop codon is encountered, thus producing a protein containing a polyglutamine stretch.

The transcript undergoes considerable alternative splicing, producing several isoforms (Mori et al., 1991; Zhuchenko et al., 1997). In particular, the presence or absence of a 5-nucleotide stretch, GGCAG, at the 3′ end of the gene (between exons 46 and 47) is critical for the expression of the CAG repeat stretch (Fig. 1B): when the 5 nucleotides are left in place, the CAG repeat is translated and expressed as a polyglutamine sequence at the protein level; when they are spliced out, a stop codon is encountered upstream the CAG_n stretch, and the polyglutamine repeat is not included in the resultant protein. Isoforms both with or without the GGCAG insertion, i.e. expressing or not expressing the polyglutamine tract, have been found in the cerebellar cortex (Ishikawa et al., 1999a). Whether these isoforms have distinct functional properties, what is their pattern of expression and how this is regulated in the human brain is largely unknown.

The normal allele size of the polymorphic CAG repeat ranges from 4–18 units (Zhuchenko et al., 1997; Shizuka et al., 1998a, b), while that of expanded alleles is from 20–30 repeats (Jodice et al., 1997; Matsuyama et al., 1997; Shizuka, et al., 1998a, b; Katayama et al., 2000). Data on the intermediate allele with 19 repeats are contradictory. Katayama et al. (2000) report an ataxic patient with a 19/7 genotype and lacunar lesions in the pons and basal ganglia. In other studies heterozygotes for this allele were normal even at old ages (Ishikawa et al., 1997; Mariotti et al., 2001).

When compared to other CAG_n expansion disorders the size of SCA6 expanded alleles appears much smaller, overlapping their wildtype allele distribution (Margolis, 2002). The size of SCA6 expanded alleles is more stable, as expected on the basis of their relatively low number of repeats. No variation is usually observed in families over successive generations, and no mosaicism is apparent in cells from different parts of the brain (Ishikawa et al., 1999b) or in sperm (Shizuka et al., 1998b). However, some degree of meiotic instability should be assumed since in three families an intergenerational jump of the expanded allele size has been reported (Jodice et al., 1997; Matsuyama et al., 1997; Mariotti et al., 2001).

SCA6 has a dominant pattern of inheritance. Homozygous patients for a repeat expansion, however, are slightly differing from heterozygous ones, showing an earlier onset and a more rapid course (Geschwind et al., 1997; Matsumura et al., 1997; Takiyama et al., 1998; Fukutake et al., 2002). With intermediate size (19 repeat) alleles, however a recessive rather than a dominant pattern has been suggested, based on the observation of an ataxic proband carrying a 19/19 genotype, and of a normal phenotype in its heterozygous relatives (Mariotti et al., 2001)

SCA6 expansions have been found in a number of sporadic ataxia cases (Ikeuchi et al., 1997; Matsumura et al., 1997; Riess et al., 1997; Zhuchenko et al., 1997; Shizuka et al., 1998b), but a new mutation has been documented in one patient only (Shizuka et al., 1998a). Should all the reported sporadic cases be new mutations, the mutability of normal alleles would be very high. If a mutation/selection equilibrium is assumed, a high frequency of SCA6 *de novo* mutations, would be in disagreement with the, presumably, small or absent selection against a disease, such as this one, with a late onset and a long life span. In addition, linkage disequilibrium among SCA6 families was also found (Dichgans et al., 1999; Soong et al., 2001a) which, again, seems in contrast with a high gene mutability. Incomplete penetrance or presence of neglected mild expression of the disorder in probands' relatives appear to be more likely explanations for the high number of sporadic cases.

Clinical features

The SCA6 clinical picture was initially described (Zhuchenko et al., 1997) as characterized by a late onset slowly progressive cerebellar ataxia with nystagmus and dysarthria, brainstem involvement (dysphagia), vibratory and proprioceptive sensory loss, often preceeded by episodes of "wooziness" and imbalance. This phenotype, similar to that reported by some later studies (Geschwind et al., 1997; Stevanin et al., 1997; Filla et al., 1999), matches that of ADCA type 1, according to the classification of Harding (1982), i.e. a cerebellar deficit with the involvement of other nervous functional systems. According to other reports, instead, SCA6 has the features typical of a "pure" cerebellar ataxia with no extracerebellar involvement (ADCA type III according to Harding, 1982) (Ikeuchi et al., 1997; Ishikawa et al., 1997; Matsumura et al., 1997; Stevanin et al., 1997; Nagai et al., 1998; Watanabe et al., 1998; Garcia-Planells et al., 1999). Still other studies, however, show a more complex clinical picture. Patients may initially experience episodes of ataxia and dysarthria, and/or of vertigo and nausea, accompanied by visual disturbance (such as diplopia or blurred vision), and tinnitus, lasting from minutes to days and triggered by head move-

ments and physical or emotional stress (Calandriello et al., 1996; Geschwindt et al., 1997; Jodice et al., 1997; Koh et al., 2001; Sinke et al., 2001). Episodes have a variable frequency (from yearly to daily), and duration (from seconds to days). Interictally, patients may be neurologically normal or show mild cerebellar signs such as gaze-evoked nystagmus and/or saccadic pursuit, mild dysarthria and dysmetria, and mild gait imbalance (Calandriello et al., 1996; Geschwindt et al., 1997; Jodice et al., 1997; Koh et al., 2001; Sinke et al., 2001). This early phase can have a variable duration. It commonly lasts a few years before the onset of a permanent and progressive ataxia, usually of a "pure" cerebellar type. The neurological examination shows trunk and limb ataxia, dysmetria, dysarthria, hypotonia, gaze evoked nystagmus with or without a downbeat component, dysmetric saccades, saccadic pursuit and hyperactive vestibulo-ocular reflex (Gomez et al., 1997). The presence of extra-cerebellar signs, such as dysphagia, opthalmoplegia, hyperreflexia, sensory loss and/or dementia is sometimes reported (Geschwind et al., 1997; Zhuchenko et al., 1997) particularly in older subjects (Ikeuchi et al., 1997; Ishikawa et al., 1997; Stevanin et al., 1997) or in patients with other coexisting disorders (Takiyama et al., 1998). Vertigo episodes may continue during this second progressive phase (Gomez et al., 1997), and periodical exacerbations of the cerebellar signs can be present (Yabe et al., 1998). In some cases the disease may not progress towards a full-blown disorder and maintains its episodic character with mild non-progressive interictal cerebellar signs (Calandriello et al., 1996; Jodice et al., 1997; Takiyama et al., 1998; Koh et al., 2001). More frequently the disease very slowly progresses towards inability; autonomous walking is maintained even after 18–20 years from the onset (Geschwind et al., 1997). However a very rapid course has also been reported (Watanabe et al., 1998). The life span appears to be normal, although no statistical analysis is available.

The above picture of SCA6 phenotype strikingly overlaps that of the allelic disorder EA2, due to point mutations of the same gene. EA2 has been described as an early onset disorder characterised by episodes of vertigo/ataxia with dysartria, visual disturbances and variable interictal cerebellar signs, from nystagmus only to severe progressive cerebellar ataxia (Farmer and Mustian, 1967; Moon and Koller, 1991; Baloh et al., 1997). Acetazolamide, a carbonic anhydrase inhibitor, is a well-known treatment preventing EA2 episodes. Differences and similarities between the two disorders emerged by comparing the features of 315 SCA6 and 44 EA2 patients, reported in the literature, and carrying, respectively, a CAG expansion or a point mutation of CACNA1A gene (Frontali, 2001). Both can have an episodic and/or a progressive ataxia phase, but they differ in the proportion of patients with progressive cerebellar involvement, which is larger in SCA6, and of those with vertigo and/or ataxia episodes, which is larger in the EA2 group. As in EA2, SCA6 episodes respond to acetazolamide treatment (Calandriello et al., 1996; Jen et al., 1998; Koh et al., 2001), although no control for a placebo effect has ever been performed. Additional extracerebellar signs, such as decrease in vibration sense, hyperreflexia with or without Babinski sign, dysphagia and/or ophtalmoplegia, brainstem atrophy, appear to affect only a minority of SCA6 patients.

Overall, the main features distinguishing SCA6 from EA2 appear to be a higher frequency of progressive ataxia occasionally associated with extracerebellar signs. It should be noted, however, that the two groups have different ages at examination, SCA6 patients being older than EA2 patients (mean age 58.5 vs. 38.0 years). It is tempting to speculate that the phenotypic differences in SCA6 patients are the consequence either of other neurological disorders intervening with age, or to a longer disease duration, with a greater likelihood of progression to permanent ataxia. This would suggest the possibility that the two conditions might be expressions of the same highly variable disorder. A strong support for such a hypothesis comes from families carrying an SCA6 expansion mutation in which different members show either the typical features of EA2 with a prominent paroxystic ataxia or those of SCA6 with a progressive and permanent ataxia (Jodice et al., 1997; Koh et al., 2001).

Another possible discrepancy between SCA6 and EA2 concerns the age at onset. In most studies SCA6 age at onset refers to the beginning of the progressive ataxia phase and is reported around 50 years of age, with a range from 16–73 (Gomez et al., 1997; Matsumura et al., 1997; Watanabe et al., 1998; van de Warrenburg et al., 2002). In EA2, instead, the age at onset, referred to the beginning of vertigo/ataxia episodes, is usually in the first or second decade of life (Yue et al., 1997; Jen et al., 1998, 1999, 2001; Denier et al., 1999; Guida et al., 2001). It should be noted, however, that this discrepancy is probably in part, if not completely, due to an ascertainment bias: EA2 is more easily diagnosed in infancy or adolescence when prominent vertigo/ataxia episodes have fewer alternative causes, than in older ages, when such symptoms are more frequently (but inappropriately) ascribed to low blood pressure, cervical arthrosis, or other common disorders. On the other hand SCA6 evokes, by analogy with other SCAs, a permanent and progressive cerebellar ataxia and a possible early phase of ataxic/vertigo spells is overlooked both by patients and physicians, as a minor and non pertinent disturbance.

As in other CAG repeat expansion disorders, a significant inverse correlation between age at onset and size of expanded alleles has been reported in several studies (e.g. Ikeuchi et al., 1997; Ishikawa et al., 1997; Riess et al., 1997; Zhuchenko et al., 1997; Maruyama et al., 2002) as well as an anticipation of age at onset over successive generations (e.g. Ikeuci et al., 1997; Matsumura et al., 1997; Matsuyama et al., 1997; Watanabe et al., 1998; Sinke et al., 2001). The latter observation is rather surprising considering that no intergenerational variation of the expanded allele size was present. The phenomenon, hence, should be ascribed, again, to an ascertainment bias, being more likely to observe a parent-child pair with the offspring affected earlier than the parent, rather than vice versa (Penrose, 1948). Alternative explanations can rely on the exposure to different environmental factors affecting age at onset in different generations, or on the difficulty of assessing age at onset in older patients.

Neuropathology

Neuroimaging in SCA6 patients reveals a cerebellar vermis atrophy predominating in the anterior portion, which might later extend to cerebellar hemispheres, usually with preservation of brainstem (Calandriello et al., 1996; Gomez et al., 1997; Jodice et al., 1997; Nagai et al., 1998; Satoh et al., 1998; Shizuka et al., 1998a, b; Takiyama et al., 1998). An early involvement of the cerebellar vermis appears also to characterize EA2 (e.g. Denier et al., 1999, Guida et al., 2001). Occasionally, at MRI, a size reduction of pons as well as of red nucleus and middle cerebellar peduncle has been reported (Murata et al., 1998; Arpa et al., 1999; Nakagawa et al., 1999). A widespread reduction of glucose metabolism at PET scan, with particularly low values in the brainstem and cerebellar hemispheres, was observed in a group of SCA6 patients as compared to normal controls (Soong et al., 2001b). Unfortunately the controls were younger than patients, requiring a further confirmation of data.

Consistently with most neuroimaging studies, the brains of autopsied SCA6 patients showed a marked atrophy of the cerebellar vermis and, to a lesser extent, of the hemispheres. Histologically, the cerebellar cortex is characterised by a remarkable loss of Purkinje cells. Granule cells are also affected, although less severely (Subramony et al., 1996; Gomez et al., 1997; Sasaki et al., 1998; Ishikawa et al., 1999b; Tashiro et al., 1999). Loss of neurons in the dentate and inferior olivary nuclei has also been found in one study (Subramony et al., 1996). Atrophy of brainstem has been occasionally described (Zhuchenko et al., 1997).

By virtue of the analogy with other $(CAG)_n$ expansion disorders, ubiquitin and polyglutamine immunoreactive nuclear inclusions, have been looked for in SCA6 brains and in cultured cells transfected with the α_{1A} subunit cDNA coding for an expanded polyglutamine repeat. Non ubiquitinated cytoplasmic protein aggregates were detected by anti-α_{1A} antibodies (Ishikawa et al., 1999a, 2001) both in transfected cells and in patients' cerebella. In addition, the cytoplasm and nucleus of Purkinje cells showed small aggregates immunoreactive with 1C2, i.e. an antibody detecting polyglutamine sequences larger than 40 repeats. However, aggregates immunoreactive with the α_{1A} subunit were not reactive with 1C2. The interpretation of this finding is not straightforward. It should be noted, in fact, that protein aggregates in other polyglutamine disorders are most likely due to the tendency of polyglutamine stretches to form β-sheets by linking to one another through hydrogen bonds between their main chain and the side chain amides (Perutz, 1994). This process is strongly dependent on the number of repeat units: no aggregate formation is observed with polyglutamine stretches below 27 repeats (Scherzinger et al., 1999). The structural transition permitting aggregation begins, instead, in an expansion range between 32 and 37 glutamines, corresponding to the lower limit of the expansion range seen in patients affected with non-SCA6 polyglutamine expansion disorders. In light of these data the significance of aggregates in SCA6 brains remains obscure. Are the anti-polyglutamine reactive aggregates due to the SCA6 expansion inducing an aggregation of other polyglutamine proteins with longer numbers of units? And if so, what is the chemical basis for such an aggregation? Or, are aggregates aspecific by-products of Purkinje cell neurodegenerative process caused by a dysfunction of calcium channels? On the other hand the anti-α_{1A} reactive aggregates are hardly comparable with those found in other SCAs, being non-ubiquitinated and located in the cytosol only. It should be reminded that the α_{1A} subunit has many isoforms, only some of which contain the polyglutamine stretch (see below). Is the expression pattern of isoforms, or their likelihood of being included in the membrane or their turnover, different when the polyglutamine stretch is expanded compared to when it is not? If yes, then these aggregates could be due to the degradation of unused or obsolete isoforms. An increased expression of the α_{1A} subunit with a polyglutamine expansion, as compared to wildtype, has indeed been reported (Ishikawa et al., 1999, 2001; Piedras-Renteria et al., 2001). This would imply that SCA6 is completely different from other polyglutamine disorders for which a comparable expression level is reported for wildtype and mutated genes (see e.g. Persichetti et al., 1995; Servadio et al., 1995).

Pathogenesis

Several studies addressed the question whether the small polyglutamine expansion in SCA6 patients exerts its pathological effect by altering the calcium channel function or, as in other SCAs, by acquiring a new toxic function, independent from the channel kinetics. In other words the question to be answered is: does SCA6 belong to the group of channelopathies (Ptacek, 1997), thus influencing the level (decrease or increase) of calcium influx into the cell, or does it belong to the group of polyglutamine expansion disorders together with other SCAs?

So far experimental evidence (obtained by transfecting α_{1A} subunit cDNA with different number of CAG units in non neuronal cells, usually renal embryonic cells HEK293) is highly contradictory on this point. Two studies report that the α_{1A} protein with expanded polyglutamines induces a hyperpolarizing shift in the voltage dependence of channel inactivation (Matsuyama et al., 1999; Toru et al., 2000). The change is predicted to exert a considerable decrement on channel availability at resting potentials, approximately halving the Ca^{2+} influx, which may in turn lead, directly or indirectly, to neuronal cell death. This effect appears to be dependent on the type α_{1A} subunit isoform expressed and on the number of repeat units (Toru et al., 2000). It is notable that in these experiments the current density in transfected cells is not reduced, implying that the mutated protein is normally transported to the membrane and is not sequestered into aggregates (Matsuyama et al., 1999). Opposite results were reported by Piedras-Rentería et al. (2001). They observed a sharp increase of current density in cells expressing the mutated, as compared to wildtype, protein. Consistently, a higher expression of the mutated subunit was observed both in the membrane and in the cytoplasm. The effect, however, was not dependent on the number of polyglutamines. No significant alteration of channel function was observed either in the activation or in the inactivation kinetics. Although a confirmation of the functional analysis through sin-

Table 1. SCA6 as compared with EA2 and other SCAs with CAG expansions

Features	SCA 1, 2, 3, 7	SCA 6	EA2
Mutation	CAG expansion (large) Range 32–100	CAG expansion (small) Range 20–30	Point mutation
Meiotic/mitotic instability of mutation	High	Very low / absent	Absent
Frequency among sporadic ataxia cases	Low or absent	High	Not reported
Ataxia/vertigo episodes	Absent	Frequent	Very frequent
Response to acetazolamide	Not assessed	Positive	Positive
Progressive ataxia	Invariably present	Very frequent	Frequent
Extracerebellar deficits	Invariably present	Low frequency	Extremely rare
Neuropathology	Generalized cerebellar atrophy + extra-cerebellar involvement	Atrophy of cerebellar vermis + hemispheres later on + (occasionally) extra-cerebellar structures	Atrophy of cerebellar vermis + hemispheres later on + very rarely extracerebellar structures
Age at onset (range)	10–65 (progressive ataxia)	16–73 (progressive ataxia)	I–II decade (episodes)
Inverse correlation age at onset/allele size	yes	yes	–
Age at onset anticipation	Yes (intergenerational jump of allele size)	Yes (no intergenerational jump of allele size)	No
Nuclear and cytoplasmic aggregates	Yes (reactive to anti-ubiquitin, -polyglutamine, -protein)	Yes (non ubiquitinated, dissociated reaction to polyglutamine and protein antibodies)	Never assessed
Electrophysiological characteristics of mutated protein	–	Decrease of calcium influx, in some studies only	Decrease of calcium influx in all studies
Increased expression of mutated protein	No	Yes in some studies	No

gle channel recording would have been appropriate, these data would imply that SCA6 is induced by an abnormal expression or turnover of the mutated protein rather than by a channelopathy as previously suggested. Still distinct results were obtained by Restituito et al. (2000) in a different expression system (Xenopus oocytes). These authors observed an abnormal channel function, but in a direction opposite to that reported by Matsuyama et al. (1999) and Toru et al. (2000): an increased influx of Ca^{2+}, in fact, was predicted as a consequence of a hyperpolarizing shift in the voltage dependent channel activation and a slowed rate of inactivation. This effect was not obtained when the mutated protein had less than 30 glutamine units and an auxiliary β subunit different from β4 was coexpressed.

The above discrepancies could be ascribed to a number of variables in the experimental setting, namely to cDNAs used for transfecting cells (rabbit, human or chimeric rabbit/human cDNA), expression of different alternative exons (e.g. exon 37a or 37b), or coexpression of different auxiliary β units isoforms. Should these differences really account for the discrepant results obtained, this would indicate that the SCA6 mutations are interacting with the isoforms of the channel proteins. A more reliable picture of SCA6 pathogenesis, therefore, would require a detailed knowledge of functional characteristics and expression regulation of the different wildtype protein isoforms, in order to understand their interactions with the gene mutations.

Conclusions

SCA6 is one of three disorders due to mutations of the pore-forming subunit of calcium channels type P/Q and is due to small expansions of a polyglutamine sequence at the COOH terminal of the protein. Its clinical phenotype shows a wide overlap with its allelic disorder EA2, associated with point (missense or protein truncating) mutations. The two disorders share, among other things (see Table 1): a) a similar, highly variable phenotype ranging from vertigo/ataxia episodes with interictal nystagmus and, possibly, other mild cerebellar signs with no overt ataxia (Denier et al., 1999; Guida et al., 2001) to a severe progressive pure cerebellar ataxia preceded or not by episodes (Yue et al., 1997; Guida et al., 2001); b) a predominant atrophy of the cerebellar vermis (Denier et al., 1999; Guida et al., 2001); and c) a sensitivity of episodes to acetazolamide treatment (Calandriello et al., 1996; Jen et al., 1998; Koh et al., 2001). The issue of the phenotype similarities between SCA6 and EA2 has implications for the functional significance of the protein. In fact, when different mutations in the same gene are causing the same phenotype it is far more likely that they both lead to a loss rather than to a gain of protein function. Electrophysiological experiments have, indeed, assessed that CACNA1A point mutations leading to EA2 are associated with a loss of channel function (Guida et al., 2001; Jen et al., 2001; Wappl et al., 2002). Moreover, FHM point mutations show a loss of channel function when the phenotype includes a cerebellar deficit (Hans et al., 1999), while a gain of function can be present when the phenotype is purely migraineous. Overall the above evidence would suggest that a loss of channel function is underlying the cerebellar involvement in all the three CACNA1A allelic disorders. Electrophysiological evidence on SCA6 mutation, however, is far from clear on this point and no conclusions can be drawn.

With other SCAs (1, 2, 3, 7, 17) due to polyglutamine expansion, SCA6 shares the type of mutation and the protein aggregates (Table 1). However major differences are present: a) the range of repeat units in SCA6 expanded alleles falls within the distribution of other SCA normal alleles and, what is more, below the threshold for polyglutamine aggregation; b) the gene product is a membrane rather than a nuclear or cytoplasmic protein; c) the intracellular aggregates are not ubiquitinated and their relationship with the mutated protein are far from clear.

In this situation the pathogenic mechanism underlying SCA6 remains unanswered: i.e. whether a gain of a new toxic activity of the polyglutamine expansion per se should be envisaged or there exists an altered (possibly decreased) channel function, as in EA2. The presence of both mechanisms has been suggested in order to reconcile conflicting evidence (Gomez, 2002). However when no sufficient support is available for each separate hypothesis, the same holds true for their combination.

A solution for the above question will hopefully arrive when more information about the physiological properties of the channel protein isoforms and their interactions with different mutations will be attained through cell and animal models.

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