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Neural substrates of motor and cognitive dysfunctions in SCA2 patients: A network based statistics analysis

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\textbf{ABSTRACT}

Spinocerebellar ataxia type 2 (SCA2) is an autosomal dominant neurodegenerative disease characterized by a progressive cerebellar syndrome, which can be isolated or associated with extracerebellar signs. It has been shown that patients affected by SCA2 present also cognitive impairments and psychiatric symptoms. The cerebellum is known to modulate cortical activity and to contribute to distinct functional networks related to higher-level functions beyond motor control. It is therefore conceivable that one or more networks, rather than isolated regions, may be dysfunctional in cerebellar degenerative diseases and that an abnormal connectivity within specific cerebellum-cortical regions might explain the widespread deficits typically observed in patients.

In the present study, the network-based statistics (NBS) approach was used to assess differences in functional connectivity between specific cerebellar and cerebral “nodes” in SCA2 patients. Altered inter-nodal connectivity was found between more posterior regions in the cerebellum and regions in the cerebral cortex clearly related to cognition and emotion. Furthermore, more anterior cerebellar lobules showed altered inter-nodal connectivity with motor and somato-sensory cerebral regions. The present data suggest that in SCA2 a cerebellar dysfunction affects long-distance cerebral regions and that the clinical symptoms may be specifically related with connectivity changes between motor and non-motor cerebello-cortical nodes.

\textbf{1. Introduction}

Spinocerebellar ataxia type 2 (SCA2) is an autosomal dominant neurodegenerative disease involving the cerebellum. Neuropathological studies confirmed a pattern of grey matter (GM) loss to affect the cerebellar vermis and hemispheres with sparing of the vermic lobules I, II (lingula) and X (nodulus) and of the hemispheric lobules I, II (lingula) and Crus II (Della Nave et al., 2008a) as well as a diffuse damage of the brainstem and cerebellar white matter (WM) (Durr et al., 1995, Gilman et al., 1996, Estrada et al., 1999).

In addition to typical motor deficits (Takahashi et al., 2010), the presence of cognitive impairments in subjects with degenerative ataxia has long been debated (Fehrenbach et al., 1984). Recently, the cognitive performances of SCA2 patients have been exhaustively investigated showing that the patients may present with impairment in several cognitive and emotional domains (Kline et al., 2010; Sokolovsky et al., 2010; D’Agata et al., 2011; Fancellu et al., 2013; Moriarty et al., 2016).

The evidence of motor, cognitive, and emotional impairments in the presence of cerebellar damage has been linked to alterations of cerebro-cerebellar networks (Broich et al., 1987; Clausi et al., 2009; Komaba et al., 2000; Bailleul et al., 2010).

Indeed, the cerebellum has extensive projections to and from cortical regions by means of middle and superior cerebellar peduncles, the main afferent and efferent cerebellar white matter (WM) tracts. These connections are known to be strictly controlateral and to be spatially and functionally organized in distinct parallel loops (Middleton and Strick, 1994; Ramnani, 2006), thus contributing to...
distinct functional networks (Allen et al., 2005; Habas et al., 2009; De Vivo Fallani et al., 2016) clearly related to different functional processes. Within this complex neural system the role of the cerebellum is to integrate multisensory information and then send them back to cerebral cortex (Leggio and Molinari, 2015). More specifically, it has been proposed that the cerebellum modulates the cortical activity (Di Lazzaro et al., 1994; Middleton and Strick, 2000) by detecting predictable sequences and allowing an optimized feedforward control that is necessary to accomplish the different functions successfully (Leggio et al., 2011).

Therefore, it is conceivable that an abnormal connectivity within specific cerebellum-cortical circuits might explain the widespread deficits typically observed in SCA2 patients and that one or more networks, rather than isolated regions, might be dysfunctional.

Consistent with this hypothesis, a reduction of brain size has been reported in patients with SCA2, involving not only cerebellum and brainstem, but also other cortical and subcortical areas, such as frontal regions, primary sensorimotor cortex, temporo-mesial and parahippocampal regions, substantia nigra, middle striatum, and thalamus (Estrada et al., 1999; Brenneis et al., 2003; Fanelli et al., 2013, Mercadillo et al., 2014). All these regions are known to be reciprocally connected with the cerebellum (Schmahmann, 1991; Schmahmann and Pandya, 1997; Middleton and Strick, 2001), indicating that several targets of cerebellar projections, including both motor and non-motor areas, are affected in patients with SCA2.

We hypothesize that cerebellar dysfunctions affect long-distance regions of the brain and clinical symptoms are related with changes in functional connectivity (FC) within specific cerebellum-cortical networks.

The investigation of FC may provide important information to further characterize the neural basis and examine the integrity of cerebellar and cerebral networks in SCA2 patients. Indeed FC allows the relationship between the neuronal activation patterns of anatomically separated brain regions to be described (van den Heuvel and Hulshoff Pol, 2010). Amongst the available methods to investigate the brain functional connectivity, resting-state fMRI (RS-fMRI) has been proven particularly suitable for the study of a complex structure like the cerebellum, that is necessary to accomplish the different functions successfully (Leggio et al., 2011).

Table 1

<table>
<thead>
<tr>
<th>Case code</th>
<th>Age</th>
<th>Gender</th>
<th>Years of illness</th>
<th>CAG repeats</th>
<th>ICARS total scores</th>
</tr>
</thead>
<tbody>
<tr>
<td>CA-1</td>
<td>42</td>
<td>F</td>
<td>1</td>
<td>22/39</td>
<td>47</td>
</tr>
<tr>
<td>CA-2</td>
<td>54</td>
<td>F</td>
<td>1</td>
<td>22/37</td>
<td>27</td>
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<tr>
<td>CA-3</td>
<td>65</td>
<td>M</td>
<td>3</td>
<td>22/35</td>
<td>27</td>
</tr>
<tr>
<td>CA-4</td>
<td>42</td>
<td>M</td>
<td>1</td>
<td>14/47</td>
<td>24</td>
</tr>
<tr>
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<td>42</td>
<td>F</td>
<td>1</td>
<td>22/39</td>
<td>28</td>
</tr>
<tr>
<td>CA-6</td>
<td>36</td>
<td>F</td>
<td>8</td>
<td>22/42</td>
<td>37</td>
</tr>
<tr>
<td>CA-7</td>
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<td>F</td>
<td>4</td>
<td>22/37</td>
<td>31</td>
</tr>
<tr>
<td>CA-8</td>
<td>41</td>
<td>M</td>
<td>3</td>
<td>22/38</td>
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</tr>
<tr>
<td>CA-9</td>
<td>44</td>
<td>F</td>
<td>13</td>
<td></td>
<td>28</td>
</tr>
</tbody>
</table>

ICARS = International Cooperative Ataxia Rating Scale.
was used for statistical comparison. A two-sample t-test was used to compare FC matrices between patients and controls, with 5000 permutations and setting the significance threshold at p-value at 0.05 corrected for multiple comparisons by using NBS correction (Zalensky et al., 2010).

It is important to note that all analyses were performed using Statistical Parametric Mapping (SPM) software, which is designed to analyze functional imaging data. The SPM8 (http://www.fil.ion.ucl.ac.uk/spm/) and in-house software implemented in Matlab were used for data processing. The 3D Modified Driven Equilibrium Fourier Transform (MDEFT) scan was used to obtain a connectivity matrix for each participant, and then we used the Network-based statistics (NBS) tool developed by Zalesky et al. (2010) to assess differences in functional connectivity (FC) between specific cerebellar and cerebral nodes. These results are consistent with previous findings (Zalesky et al., 2010).

### 4. Results

#### 4.1. Neuropsychological assessment

The neuropsychological assessment revealed the presence of selective and very slight impairments in some patients but did not show clear evidence of general cognitive impairment. Indeed, some patients displayed values below the cut-off in word fluency test and backward digit span (CA5), forward digit span (CA7), and Wisconsin Card Sorting Test (CA6) (see Table 2). These results are consistent with findings that patients who are affected by cerebellar damage do not present with marked cognitive deficits.

#### 4.2. Resting-state fMRI data results

No significant differences were found between total GM volumes of patients [mean ± SD = 655.7 ± 51.5] and controls [mean ± SD = 644.2 ± 49.1] as assessed by the t-test analysis (p = 0.26).

NBS analysis showed altered inter-nodal connectivity between cerebellum and several cerebral regions throughout the whole brain. Overall, 62 nodes and 110 edges showed differences in SCA2 brains compared to control ones while 57 edges and 35 nodes survived after Bonferroni correction for multiple comparisons (FWE = 0.05) (Fig. 1a).
According to the cerebellar functional topography, cerebellar nodes in the posterior cerebellum, such as Crus I and Crus II, showed reduced FC with nodes in cortical regions implicated in cognition and emotion, such as superior (SFg) and middle (MFg) frontal gyrus. Similarly, cerebellar nodes in the motor anterior cerebellum, such as lobules III, IV, V, and vermis IV–V, showed reduced FC with nodes in the cortical regions related to motor control, such as precentral (PrG) and post-central (PcG) gyrus, Rolandic Operculum (RO), supplementary motor area (SMA) (Fig. 1b).

Finally, reduced inter-nodal FC was found between cerebellar lobule VI and both cognitive and motor regions in the cerebral cortex, including supramarginal gyrus and supplementary motor area. A similar pattern was found in the vermis lobule VI, showing decreased functional connectivity with regions in the rolandic and frontal operculum as well as the supramarginal area. Detailed results of NBS analysis are reported in Table 3 that shows the cerebello-cortical edges of significant FC decrease in SCA2 patients.

5. Discussion

Despite the advancing knowledge of cerebellar functions, the specific role that the cerebellum plays in concert with other brain regions in SCA2 patients remains unclear. RS-fMRI is an ideal method for investigating functional interactions between cerebellum and cerebral cortex in the human brain and it may prove a useful tool for interpreting motor and non-motor impairment driven by the cerebellar damage.

A growing body of studies explored the use of RS-fMRI in functional disconnection in neurological and psychiatric disorders (Greicius and Menon, 2004; Greicius et al., 2007; Rombouts et al., 2005, 2009; Liu et al., 2008; Bluhm et al., 2009; Whitfield-Gabrieli et al., 2009). Disrupted functional cerebellar connectivity has been demonstrated in patients with schizophrenia (Liu et al., 2008; Collin et al., 2011), Parkinson’s disease (Liu et al., 2013), and major depressive disorder (Ma et al., 2013). Disruption of visual and motor connectivity has also been demonstrated in spinocerebellar ataxia type 7 (SCA 7) supporting the theory that neurodegenerative diseases target specific regions in large-scale networks (Seeley et al., 2007). Further, typical connectivity patterns have been characterized in patients with autosomal dominant spinocerebellar ataxia 17 (SCA17) (Reetz et al., 2012) suggesting that the broad range of symptoms observed in SCA17 patients may primarily reflect the involvement of distinct functional networks affected by the cerebellar atrophy. A disconnection syndrome has been suggested in spinocerebellar ataxia type 1 (SCA 1) by means of intrinsic functional analysis and diffusion tensor imaging (Solodkin et al., 2011).

Accordingly, RS-fMRI studies in healthy subjects provided a detailed mapping of resting state networks of the human cerebellum revealing that distinct networks are associated with each single lobule (Bernard et al., 2012), van den Heuvel and Hulshoff Pol (2010) suggested that there is a more general link between structural and functional connectivity. Indeed, it has been shown that almost all functionally linked regions of the most often reported resting-state networks are structurally interconnected by known white matter tracts (van den Heuvel et al., 2009). This suggests the existence of a general structural core of resting-state networks, supporting the notion of an overall link between structural and functional connectivity on a whole-brain scale (Damoiseaux and Greicius, 2009; Hagmann et al., 2008). These assumptions support the idea that the functional heterogeneity of the cerebellum is reflected in its connectional heterogeneity and give rise to the hypothesis that different cerebello-cortical projections and distinct functional modules can be selectively impaired by cerebellar disorders.

In the present study the pattern of FC alterations between regions in the cerebellum and cerebral cortex has been extensively characterized in SCA2 patients using the NBS approach, that is based on the whole-brain analysis allowing complex systems to be described as networks (Bullmore and Sporns, 2009; Rubinov and Sporns, 2010). We found...
nodes in the posterior cerebellum to show reduced functional connectivity with nodes in cortical regions related to cognition and emotion and nodes in the anterior cerebellum to show reduced functional connectivity with nodes in the cortical regions related to motor control. This result is, at least in part, in line with a previous FC study in SCA2 patients using a seed-based approach (Hernandez-Castillo et al., 2015a) and showing FC decrease between the right posterior cerebellum and the left superior frontal gyrus, which could impact different cognitive operations such as self-monitoring and verbal/visuospatial working memory (Cao et al., 1998; O’Reilly et al., 2010).

Additionally, we found that in both hemispheres and vermis, lobule VI shows decreased FC with regions related to motor control as well as emotion and nodes in the anterior cerebellum to show reduced functional connectivity with nodes in the posterior cerebellum. These prefrontal areas have been consistently implicated in different aspects of executive functions using both verbal and visuospatial tasks (Reverberi et al., 2005; Crescentini et al., 2011; Langdon and Warrington, 2000). In spite of this, in the present study only 3 patients presented impaired performances in executive processing and verbal working memory. This datum can be explained by the fact that most standard norms of testing do not detect cognitive impairments in cerebellar cohorts because cerebellar patients’ symptoms are present in selective domains and very often, they can be detected only when the patients are compared to matched healthy controls (Tedesco et al., 2011).

An important issue that needs to be discussed is that the pattern of decreased cerebello-cerebral functional connectivity may be at least in part explained by damage of the cortical GM in SCA2. Indeed, cerebellar atrophy has been also reported to reduce GM volume in several supratentorial areas (Brenneis et al., 2003; Della Nave et al., 2006a). Thus, even if in the present study the total GM volume was not significantly different between SCA2 patients and controls, the possibility of local GM loss cannot be ruled out. An alternative explanation for the absence of significant whole brain GM loss in patients might be that the pattern of ponto-cerebellar atrophy associated with SCA2 pathology predominantly entails a WM damage (Della Nave et al., 2006b).

Overall, the observed pattern of inter-nodal underconnectivity is consistent with previous studies using different RS-fMRI approaches (O’Reilly et al., 2010; Bernard et al., 2012) demonstrating in healthy subjects the functional segregation of the cerebellum in sensorimotor and supramodal zones, the former containing overlapping functional connectivity maps for domain-specific motor and somatosensory cortices, the latter for prefrontal and posterior-parietal cortex, and provides the latter important insight into understanding the neural circuit abnormalities in SCA2. In light of the general link between structural and functional connectivity (van den Heuvel et al., 2009) a comprehensive understanding of neural connectivity may require clear evidence as to whether structural connectivity is affected in SCA2. In SCA2 patients microstructural alterations of the cerebellar WM have been reported by Diffusion Tensor Imaging (DTI) studies, showing the prevalent involvement of the main afferent and efferent tracts (i.e. Middle and Superior Cerebellar Peduncle), connecting the cerebellum with both motor and non-motor cortical regions (Mandelli et al., 2007; Della Nave et al., 2008b; Hernandez-Castillo et al., 2015b). Although in the present study structural connectivity has not been specifically investigated, it has to be considered that microstructural abnormalities of the cerebellar WM tracts, typically reported in SCA2 patients, may underlie a deficient structural connectivity that impacts the cerebellocerebral interplay and results in a lack of functional connectivity.

Cerebellar clusters of significantly reduced functional connectivity have been recently reported by Cocozza et al. (2015) only in the default mode network, executive control network and right fronto-parietal network in patients with SCA2. However, this study used a different resting-state approach (i.e. ICA) limited to investigate connectivity within specific functional networks (Cocozza et al., 2015). In the present study, by using a whole-brain approach, we provide additional evidence that extensive and segregated functional brain changes may occur as the result of the SCA2 degenerative process.
Indeed, cerebellar-cerebral functional disconnections are observed in this patient population throughout the brain and they are consistent with the pattern of cerebellar structural alterations mainly involving vermis and cerebellar hemispheres reported by Della Nave and colleagues (2008) In particular, connectivity reduction involved segregated motor and cognitive cerebellar-cortical networks with the only exception of lobule VI involvement, not by chance a region in which both motor, cognitive, and emotional functions are localized. It has to be underlined that we also found Crus II and lobule VII to show a functional disconnection with nodes in superior and middle frontal regions. This evidence is partially consistent with previous VBM studies that have shown cerebellar grey matter reduction to spare both Crus II and lobule VII in SCA2 patients (Brenneis et al., 2003; Ying et al., 2006; Della Nave et al., 2008a,b). Nevertheless, it has to be considered that a functional coherence between the two cerebellar hemispheres has been widely demonstrated by RS-fMRI studies (Habas et al., 2009; O’Reilly et al., 2010; Buckner et al., 2011). Thus, it is reasonable to hypothesize that the cerebellar regions that are not directly affected by the degenerative process could suffer from the functional release of the affected cerebellar regions and result functionally impaired.

A limitation of this study is that, due to the small number of patients, the cognitive performance has not been directly correlated with functional connectivity alterations observed. Further investigations are needed to support our interpretation with greater patient population.

6. Conclusion

To our knowledge this is the first study using a whole-brain approach to investigate functional organization in SCA2 patients and to detect cerebellar-cerebral inter-nodal connectivity changes that can be associated with cerebellar structural abnormalities of SCA2. Allogether, the present findings show that a cerebellar dysfunction may affect long-distance regions in the cerebral cortex targeted by cerebellar projections and that specific cerebral functional alterations derive from cerebellar structural degeneration typically associated with SCA2 pathology, thus resulting into the multifariar motor, cognitive, and emotional deficits evidenced in patients.

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Conflict of interest

The authors declare that they have no conflict of interest.

References


Simani, G., Lamm, C., Ruff, C.C., Singer, T., 2013. Right supramarginal gyrus is crucial to overcome emotional egocentricity bias in social judgments. J. Neurolsci. 33 (39), 15466–15476.


Manyam, B., 1997. International Cooperative Ataxia Rating Scale for ambulatory ataxia types 1, 2, 3, 6, and 7. Orphanet J. Rare Dis. 11 (1), 82.


