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## Subclinical neurological involvement does not develop if Wilson's disease is treated early

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### ABSTRACT

**Background & aims:** Wilson's disease (WD) is a genetic disorder of copper metabolism causing dysfunctions of various organs, mostly the liver and brain. If untreated, WD is fatal, but early treatment results in a good prognosis, although the long-term neurological outcome has not yet been clarified. To address this issue, we evaluated the neurological status of early-treated WD patients without overt nervous system impairment using neurophysiological, neuropsychological and neuroimaging procedures at least 10 years after treatment onset.

**Methods:** Thirty-eight WD patients (18 females, aged  $24.47 \pm 7.50$  years), who received an early diagnosis (in presymptomatic or mild/moderate liver disease stages without neurological involvement) and prompt treatment, were clinically evaluated with the Global Assessment Scale. Presentation was hepatic in 36 subjects (95%), while 2 patients (5%) were presymptomatic. A neurophysiological study was performed to explore the central motor conduction time of the upper and lower limbs, and motor cortex excitability using single pulses and paired-pulse transcranial magnetic stimulation. Neuroimages were obtained with brain magnetic resonance scans. Cognitive abilities, and psychiatric and behavioral disturbances were evaluated with neuropsychological tests.

**Results:** Patients were undergoing treatment with penicillamine (7 patients) or zinc salts (31 patients) with good adherence. They did not present any neurological signs at clinical evaluation or at specific scale of impairment, the mean Global Assessment Scale score was  $0.3 \pm 0.7$ . Magnetic resonance imaging, transcranial magnetic stimulation studies and neuropsychological/neuropsychiatric assessment ruled out subclinical involvement.

**Conclusions:** This study suggests that early diagnosis and treatment of WD may prevent the onset of neurologic damage, even at subclinical level.

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**Abbreviations in order of appearance:** Wilson's Disease, WD; Central Nervous System, CNS; Global Assessment Scale, GAS; D-Penicillamine, DPA; Magnetic Resonance Imaging, MRI; Resting Motor Threshold, RMT; Active Motor Threshold, AMT; Short Interval Intracortical Inhibition, SICI; Intracortical Facilitation, ICF; Cortical Silent Period, CSP; Mini-Mental State Examination, MMSE; Frontal Assessment Battery, FAB; Raven's 47 Colored Progressive Matrices, RCPM; Beck Depression Inventory Scale, BDI; Neuropsychiatric Inventory, NPI; Barrott Impulsiveness Scale Vision 11, BIS 11.

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### 1. Introduction

Wilson's disease (WD) is a widely known condition in many medical fields as it can present with different clinical features depending on the site of excessive copper accumulation and subsequent copper toxicity [1]. It was first described in detail in 1912 and defined as "progressive lenticular degeneration causing a familial nervous disease associated with cirrhosis of the liver" [2]. At that time, neurological dysfunctions and advanced liver impairment were considered an inevitable part of the disease. With the advent of effective drugs, prolonged survival has become the norm

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for WD patients.

In general, prognosis depends on the severity of liver and neurological disease and adherence to treatment. Early treatment with chelators or zinc salts can control symptoms related to liver or brain damage [3,4], therefore the prognosis of WD is strictly related to the time of diagnosis. Unfortunately, because of the wide clinical spectrum of WD and its variable onset time, the condition is usually diagnosed, and hence appropriately treated, almost one year after the onset of clinical manifestations. Although the efficacy of therapy with chelators or zinc is well documented, there is a subset of WD patients in whom the liver disease persists despite treatment [4–8]. Accordingly although the central nervous system (CNS) seems to be protected from copper accumulation in appropriately treated patients [7,8], it remains to be established whether minimal copper deposition can cause subclinical neurologic impairment. In an attempt to shed light on this issue, we used clinical, radiological, neurophysiological and neuropsychological tools to evaluate the CNS of a large group of WD patients who were diagnosed and treated in the early stage of the disease.

## 2. Patients and methods

We recruited 38 patients (18 females and 20 males) with clinical, biochemical and genetic or histological evidence of WD. Their mean age was  $24.47 \pm 7.50$  years with a mean disease duration (time from clinical onset) of  $19.24 \pm 6.50$  years, a mean treatment duration of  $17.54 \pm 6.46$  years and  $13.5 \pm 3.9$  years of schooling. Patients were selected from among those observed at Pediatric Liver Unit of University of Naples Federico II in the period 1984–2013, if the following inclusion criteria were met: (1) diagnosis of WD confirmed by molecular analysis of the ATP7B gene (standard methods, Regional Hospital for Microcytemia, Cagliari, Italy) or by liver copper content of dry tissue higher than  $250 \mu\text{g/g}$  or both; (2) available data of clinical-laboratory features, copper metabolism and treatment (dosage and compliance) at the time of diagnosis and throughout the observation period; (3) diagnosis of WD made during the presymptomatic or mild or moderate liver disease stages followed by prompt treatment, i.e., “early treated” patients; and (4) absence of overt neurological involvement. Presentation was hepatic in 36 subjects (95%), while 2 patients (5%) referred for family screening were diagnosed in a presymptomatic stage. Mutation analysis of the ATP7B coding region was performed in 36/38 patients. Disease-causing mutations were detected in 30 (83%) patients on both chromosomes (12 homozygotes and 18 compound heterozygotes) and in 4 (11%) subjects only on one chromosome. No mutations were detected in 2 (6%) patients (a couple of siblings) in whom WD diagnosis was confirmed by other parameters such as ceruloplasmin serum levels, urinary copper and liver copper content (Ferenci score [9] 4 and 5, respectively). Overall, 34 patients of 29 unrelated families carried 25 different mutations. The enrolled patients were prospectively examined at the Department of Neurosciences between 2011 and 2013.

Clinical features of nervous system impairment were evaluated in each patient by a neurologist using a standard neurological examination and the neurological assessment section (tier 2) of the Global Assessment Scale (GAS) for WD [10]. To assess subclinical CNS involvement, patients underwent brain magnetic resonance imaging (MRI), transcranial magnetic stimulation (TMS) studies and neuropsychological/neuropsychiatric evaluation. TMS studies were also performed in a separate group of 15 WD patients with neurologic signs and Kayser–Fleischer rings (8 males and seven females; mean age  $28.2 \pm 12.1$  years, mean treatment duration  $15.8 \pm 9.14$  years). Fifteen age-, education-, and sex-matched healthy subjects, not affected by any neurological, psychiatric or other relevant clinical conditions (10 females and five males; mean

age  $26.7 \pm 9.1$  years; years of schooling  $13.2 \pm 2.4$ ) served as the control group for clinical, neurophysiological, neuropsychological and neuropsychiatric evaluation.

Written consent to participate in the study was obtained from all subjects. The protocol was approved by the local ethics committee, and the research was conducted in accordance with the 1964 Declaration of Helsinki.

### 2.1. Treatment

As recommended by EASL and AASLD [11,12], D-penicillamine (DPA) was used as initial and maintenance therapy for symptomatic patients, zinc salts were used as initial and maintenance therapy for presymptomatic patients, and as maintenance therapy after a first phase with DPA for symptomatic patients. Starting from 1995, zinc was also used as first-line therapy in WD patients with a mild liver disease. Adherence to treatment was assessed based on treatment schedule data (prescribed dose, number of daily doses, and adequate interval between medicine and meals), and on levels of urinary copper excretion ( $<75 \mu\text{g}/24 \text{ h}$ ), urinary zinc levels ( $>2000 \mu\text{g}/24 \text{ h}$ ) and serum zinc levels ( $>150 \text{ mg/dl}$ ) for patients treated with zinc, and urinary copper levels (values between 200 and  $500 \mu\text{g}/24 \text{ h}$  after a year of treatment) for patients treated with DPA [11,12]. Treatment efficacy was evaluated based on the absence of manifestations of WD other than liver disease, on the maintenance of liver enzymes (aspartate aminotransferase, alanine aminotransferase,  $\gamma$ -glutamyltransferase) within normal range and on a basal urine copper level lower than  $75 \mu\text{g}/24 \text{ h}$  for patients treated with zinc, and between 200 and  $500 \mu\text{g}/24 \text{ h}$  for patients on DPA. A standard ultrasonography of liver was performed in all patients.

### 2.2. Neuroradiological evaluation

All Magnetic Resonance Imaging (MRI) studies were carried out with three Tesla on the same MRI scanner (Trio, Siemens Medical Systems, Erlangen, Germany). The brain MRI studies included T2-weighted turbo spinecho (TR/TE 4400/100 ms) and FLAIR (TR/TE/TI 8000/100/2200 ms) sequences, T1-weighted conventional SE (TR/TE 580/15 ms). During the MRI study, subjects laid supine with the head lightly fixed by straps and foam pads to minimize head movement, and were asked to relax with eyes closed.

### 2.3. Neurophysiological assessment

Neurophysiological study was performed to explore the central motor conduction time of the upper and lower limbs, and motor cortex excitability using single pulses and paired-pulse TMS. We evaluated the following TMS parameters of the stimulated motor cortex: (1) resting motor threshold (RMT) and active motor threshold (AMT); (2) short interval intracortical inhibition (SICI) and intracortical facilitation (ICF); (3) cortical silent period (CSP) [13]. Details regarding the transcranial stimulation procedures we used are reported in Supplementary Material.

### 2.4. Neuropsychological and neuropsychiatric assessment

The Mini-Mental State Examination (MMSE) was used to assess general cognitive abilities, and the Frontal Assessment Battery (FAB) to screen frontal functions [14]. In addition, we used the following neuropsychological tests to evaluate selected cognitive domains: (1) Corsi's block-tapping test and verbal span for words to assess short-term memory; (2) Rey's immediate and delayed recall of 15 words and of a short passage to evaluate long-term memory and learning; (3) attentional matrices and the shortened form of

the Stroop Color-Word Test to assess focused and selective attention; (4) Raven's 47 Colored Progressive Matrices (RCPM) to evaluate nonverbal intelligence; (5) semantic and phonological fluency tasks to assess cognitive flexibility; and (6) a copying test for geometrical figures to assess spatial organization and visuomotor skills [14]. All patients also underwent a psychiatric assessment using the following clinical scales: (1) the Beck Depression Inventory Scale (BDI) [15] to identify clinically relevant depression and to measure the severity of depressive symptoms; (2) the Neuropsychiatric Inventory (NPI) to evaluate 12 kinds of behavioral disturbances [16] recording their presence, severity (rated 1–3) and frequency (rated 1–4); and (3) the Barratt Impulsiveness Scale Version 11 (BIS-11) to evaluate impulsiveness [17].

### 2.5. Statistical analysis

Differences in the distribution of categorical variables among groups were assessed by the chi-square test. Demographic, clinical and cognitive variables were compared using the Mann–Whitney test (two-group comparison) or Kruskal–Wallis test (three-group comparison). Normal distribution of neurophysiological parameters was verified by means of Kolmogorov and Smirnov test. The ANOVA test for neurophysiological data was used to compare data among groups. Bonferroni's post hoc test or unpaired t-test was used for further analysis. Correlations of neuropsychological and neuropsychiatric results with patients' demographic (i.e. age, onset and disease duration) and clinical (GAS score) features were assessed by means of Spearman's rho. The significance level was set at  $p < 0.05$ . Statistical analysis was performed with STATA 12.1 for Windows (StataCorp LP, USA).

## 3. Results

Upon diagnosis, the 38 WD patients without neurological signs started treatment with DPA ( $n = 27$ ) or zinc ( $n = 11$ ). Of the 27 patients first treated with DPA 20 shifted to zinc monotherapy (2 for severe DPA-related side effects, 7 for treatment failure and 11 for transition to maintenance therapy). Of the 11 patients initially treated with zinc, all continued zinc monotherapy. The main DPA-related side effects were the following: maculo-papular rash, elastosis perforans serpiginosa, skin pigmentation, alopecia, proteinuria, leucopenia. No patient on a zinc regime needed to change treatment; the only adverse effect associated with zinc was mild gastric pain and mild elevation of serum lipase and amylase without the symptoms of pancreatitis. At the time of the study, 31 patients were receiving zinc and 7 DPA. These patients complied with treatment on the basis of the criteria reported under "Treatment" above. No patient had worsening of liver function, died or needed liver transplantation. Liver blood tests were normal in 31 patients from treatment onset. At the time of the study, hypertransaminasemia was detected in 7 patients, and values were two-fold higher than the upper normal limit in 3 of them. Ultrasound revealed steatosis in 30/38 patients (79%) and signs of portal hypertension in 3/38. At the time of the study, no patient presented neurological signs at clinical evaluation or at specific scales of impairment. The mean GAS score was  $0.3 \pm 0.7$ . All 38 patients did not have Kayser–Fleischer rings when evaluated by slit lamp.

### 3.1. Neuroradiological findings

Brain MRI was normal in all 38 WD patients without neurological signs. In particular, there was no evidence of hyperintensity in T2 weighted images in basal ganglia, brain stem and white matter.

### 3.2. Neurophysiological findings

RMT, AMT, CSP and SICI 2–3 differed significantly among the 38 WD patients without neurological signs, the 15 WD patients with neurological signs and the 15 healthy controls (RMT:  $F = (2, 44) = 42.638$ ,  $p < 0.001$ ; AMT:  $F = (2, 44) = 12.899$ ,  $p < 0.001$ ; CSP:  $F = (2, 44) = 32.213$ ,  $p < 0.001$ ; SICI2:  $F = (2, 44) = 31.844$ ,  $p < 0.001$ ; SICI3:  $F = (2, 44) = 32.033$ ,  $p < 0.001$ ). Neither ICF nor upper and lower limb central motor conduction time differed among the three groups (ANOVA, ICF:  $F = (2, 44) = 0.412$ ,  $p = 0.665$ ; upper central motor conduction time:  $F = (2, 44) = 0.720$ ,  $p = 0.493$ ; lower central motor conduction time:  $F = (2, 44) = 0.552$ ,  $p = 0.580$ ). Post-hoc comparison revealed that WD patients with neurological signs had higher RMT, AMT and SICI 2–3 and shorter CSP than WD patients without neurological signs and control subjects, while there were no statistically significant differences between WD patients without neurological signs and controls (Table 1). There was no correlation between the neurophysiological results and patients' demographic (age, disease onset and disease duration) or clinical (GAS score) features in either WD patients without neurological signs or WD patients with neurological signs.

### 3.3. Neuropsychological and neuropsychiatric findings

Neuropsychological and neuropsychiatric evaluations were carried out in the 38 WD patients without neurological signs and in the 15 healthy controls. The two-group comparison did not show any significant difference in age [ $t(28) = 0.821$ ,  $p = 0.418$ ] or educational level [ $t(28) = -0.280$ ,  $p = 0.782$ ]. No significant difference was found for any neuropsychological or neuropsychiatric measure (Table 2). The BDI did not reveal more depressive symptoms in WD patients than in controls ( $p = 0.102$ , Mann–Whitney test). There was no correlation between the neuropsychiatric measures (BDI and BIS-11 scores) and neuropsychological results. Likewise demographic features (age, disease onset and disease duration) did not correlate with neuropsychological and neuropsychiatric measures. Lastly, the NPI did not reveal any psychiatric or behavioral disturbance in WD patients.

## 4. Discussion

Although the effectiveness of pharmacological therapy in WD is well established, little is known about the impact of therapy on specific parameters of liver and/or neurological disease in homogeneous categories of WD patients. To our knowledge, this is the first study to use a multimodal strategy to evaluate, over a long period, the neurological status of early-treated WD patients with hepatic onset.

The pathophysiology of nervous system impairment in WD is still not fully understood. Copper deposition in the brain seems to be the main process causing neurologic disorders while hepatic encephalopathy occurs only in cases of severe liver dysfunction. It is conceivable that even when copper metabolism is well controlled with medical treatment, a slow progressive pathological process can lead to subclinical impairment.

None of our homogeneous group of early-treated WD patients with hepatic onset developed any clinical feature of neurological disorder. Brain scans were normal. Similarly, TMS studies exploring intracortical circuiting and central motor pathways were unremarkable, and neuropsychological testing revealed no signs of cognitive or psychiatric disturbance. Therefore, early-treated WD patients without clinical evidence of CNS impairment, on long-term treatment and with adequate adherence to therapy do not develop either clinical or subclinical nervous system alterations.

**Table 1**  
Neurophysiological data of Wilson's disease patients and controls.

	Neurological WD patients (n = 15)	Non neurological WD patients (n = 38)	Controls (n = 15)	p (N vs nN)	p (N vs controls)	p (nN vs controls)
DD (years)	20 ± 8.5	19.8 ± 5.4	NA	NS	NA	NA
GAS	13.4 ± 5.5	0.3 ± 0.7	NA	<0.001	NA	NA
UL CMCT (ms)	6.3 ± 0.7	6.5 ± 1.1	6.7 ± 0.8	NS	NS	NS
LL CMCT (ms)	16.0 ± 0.8	16.1 ± 1.0	15.8 ± 1.0	NS	NS	NS
RMT (%)	71.0 ± 6.3	47.6 ± 10.6	48.7 ± 5.5	<0.001	<0.001	NS
AMT (%)	46.6 ± 5.2	32.4 ± 6.1	35.4 ± 4.1	<0.001	<0.001	NS
CSP (ms)	74.0 ± 7.2	130.2 ± 27.1	125.3 ± 23.8	<0.001	<0.001	NS
SICI2 (%)	82.7 ± 13.5	44.3 ± 24.6	38.6 ± 5.2	<0.001	<0.001	NS
SICI3 (%)	77.7 ± 13.6	36.4 ± 24.5	32.8 ± 9.3	<0.001	<0.001	NS
ICF15 (%)	160.6 ± 50.9	153.4 ± 40.8	147.2 ± 25.5	NS	NS	NS

NS, not significant; NA, not applicable; N, neurological WD patients; nN, non neurological WD patients; DD, disease duration; GAS, Global Assessment Scale; CMCT, central motor conduction time; UL, upper limb; LL, lower limb; RMT, resting motor threshold; AMT, active motor threshold; CSP, cortical silent period; SICI2, short interval intracortical inhibition 2 ms; SICI3, short interval intracortical inhibition 3 ms; ICF15, intracortical facilitation 15 ms. Significance level was set at  $p < 0.05$ . Values are expressed as mean ± standard deviation.

**Table 2**  
Neuropsychological and neuropsychiatric data of non neurological WD patients and controls.

	Patients (n = 38)	Controls (n = 15)	p
<b>Neuropsychological measures</b>			
<b>Screening tests</b>			
Mini-Mental State Examination	27.7 ± 0.8	27.6 ± 0.7	NS
FAB	15.6 ± 0.6	15.5 ± 1.3	NS
<b>Spatial and verbal working memories</b>			
Corsi's test	4.7 ± 0.7	4.8 ± 0.9	NS
Verbal span	4.0 ± 0.7	4.0 ± 0.5	NS
<b>Long-term memory</b>			
15-word Immediate recall	41.0 ± 7.1	40.6 ± 5.6	NS
15-word Delayed recall	10.3 ± 1.9	10.0 ± 2.1	NS
Story recall test	13.2 ± 0.9	12.6 ± 1.8	NS
<b>Focused and selective attention</b>			
Attentive matrices	47.2 ± 3.4	46.8 ± 6.8	NS
Stroop task	9.7 ± 1.9	11.8 ± 5.1	NS
<b>Non verbal intelligence</b>			
RCPM	29.3 ± 1.90	28.3 ± 3.0	NS
<b>Cognitive flexibility</b>			
Phonological fluency	32.9 ± 6.8	32.2 ± 7.1	NS
Semantic fluency	10.9 ± 2.4	13.0 ± 5.9	NS
<b>Visuospatial skill</b>			
Copying task	12.5 ± 0.2	12.3 ± 0.9	NS
<b>Neuropsychiatric measures</b>			
BDI scale	4.1 ± 4.6	5.7 ± 3.3	NS
<b>BIS-11</b>			
Attentional score	15.1 ± 2.3	14.3 ± 2.9	NS
Motor score	17.7 ± 2.4	17.6 ± 2.2	NS
Non planning score	24.8 ± 3.7	23.6 ± 6.8	NS
Total score	57.6 ± 6.7	55.5 ± 11.3	NS

NS, not significant; FAB, Frontal Assessment Battery; RCPM, Raven's 47 Colored Progressive Matrices; BDI, Beck Depression Inventory; BIS-11, Barratt Impulsiveness Scale; Significance level was set at  $p < 0.05$ . Values are expressed as mean ± standard deviation.

The integrity of the CNS was confirmed also in patients who, despite a favorable response to therapy, presented signs of mild liver disease.

Electrophysiological findings of WD patients with neurological involvement showed moderate abnormalities related to intracortical circuit function while the cortical spinal system conductivity was normal. Previous TMS studies in WD patients with neurological impairment revealed more severe findings than ours [18,19]; this difference is probably related to a longer duration of treatment in our patients. Moreover, in our neurological WD patients, the reduction of CSP and the increase of SICI and AMT suggest GABA and glutamate neuronal circuitry dysfunctions. Recent electrophysiological studies conducted with animal models

demonstrated that copper inhibits GABA transmission by modulating neuronal firing by the interaction with GABA-A receptor [20], which in turn suggests that a high copper level may affect GABA transmission in WD. This pathophysiological process is consistent with the alterations of our TMS studies exploring GABA circuitry and in particular with SICI reduction that is mainly related to GABA-A transmission. Moreover, a high level of copper reserve may impair cortical glutamate circuitry [21], which is in line with the increase of AMT in our WD patients with neurological signs. Consequently, the neurophysiological findings of this group of patients suggest that CNS impairment could be due in part to a metabolic defect of specific neuronal circuits as well as to the toxicity of copper deposits in the brain. It would be interesting to test this hypothesis in a prospective study on a larger sample of patients. This study suggests that patients with WD may not present any neurological alteration or progression when copper metabolism is well controlled by prompt therapy. Most of our patients were under zinc salts although some of them were first treated with DPA and shifted to zinc later during the disease course. Although the aim of the present study was not to compare the efficacy between the two drugs, our results suggest that zinc can protect the CNS against copper accumulation and can control overall the disease. Numerous studies have demonstrated that anti-copper therapy can reverse the neurologic clinical impairment in WD patients [1,6,11]. Our study gives further proof of benefits of treatment revealing the absence of subclinical neurologic alterations in early treated patients.

### Conflict of interest

We declare that we have no conflicts of interest.

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None to declare.

### Authors' contributions

GR, RD, ME were the main authors and did the literature search; GR conceived and designed the study, and was involved in data acquisition. RD analyzed and interpreted the data. RI and FM were the co-senior authors and coordinated the work, and drafted the manuscript. FDD, AT, MM, LS reviewed and revised the work for important intellectual content. MQ was the radiologist and reviewed the imaging section. All authors were directly involved in care of patients; RI and FM were the physicians principally responsible for care of patients.



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## Appendix A. Supplementary data

Supplementary data related to this article can be found at <http://dx.doi.org/10.1016/j.parkreldis.2016.01.024>.

## References

- [1] A. Ala, A.P. Walker, K. Ashkan, J.S. Dooley, M.L. Schilsky, Wilson's disease, *Lancet* 369 (2007) 397–408.
- [2] S.A. Kinnier Wilson, Progressive lenticular degeneration: a familial nervous disease associated with cirrhosis of the liver, *Brain* 34 (1912) 295–509.
- [3] G.J. Brewer, R.D. Dick, V.D. Johnson, J.A. Brunberg, K.J. Kluin, J.K. Fink, Treatment of Wilson's disease with zinc: XV long-term follow-up studies, *J. Lab. Clin. Med.* 132 (1998) 264–278.
- [4] R. Iorio, M. D'Ambrosi, M. Marcellini, C. Barbera, G. Maggiore, L. Zancan, et al., Serum transaminases in children with Wilson's disease, *J. Pediatr. Gastroenterol. Nutr.* 39 (2004) 331–336.
- [5] K. Dzieżyc, M. Karliński, T. Litwin, A. Członkowska, Compliant treatment with anti-copper agents prevents clinically overt Wilson's disease in pre-symptomatic patients, *Eur. J. Neurol.* 21 (2014) 332–337.
- [6] M.L. Schilsky, Long-term outcome for Wilson disease: 85% good, *Clin. Gastroenterol. Hepatol.* 12 (2014) 690–691.
- [7] R. Bruha, Z. Marecek, L. Pospisilova, S. Nevsimalova, L. Vitek, P. Martasek, et al., Long-term follow-up of Wilson disease: natural history, treatment, mutations analysis and phenotypic correlation, *Liver Int.* 31 (2011) 83–91.
- [8] G. Ranucci, F. Di Dato, M.I. Spagnuolo, P. Vajro, R. Iorio, Zinc monotherapy is effective in Wilson's disease patients with mild liver disease diagnosed in childhood: a retrospective study, *Orphanet J. Rare Dis.* 9 (2014) 41.
- [9] E. Nicastro, G. Ranucci, P. Vajro, A. Vegnente, R. Iorio, Re-evaluation of the diagnostic criteria for Wilson disease in children with mild liver disease, *Hepatology* 52 (2010) 1948–1956.
- [10] A. Aggarwal, N. Aggarwal, A. Nagral, G. Jankharia, M. Bhatt, A novel Global Assessment Scale for Wilson's Disease (GAS for WD), *Mov. Disord.* 24 (2009) 509–518.
- [11] European Association for the Study of the Liver, EASL clinical practice guidelines: Wilson's disease, *J. Hepatol.* 56 (2012) 671–685.
- [12] E.A. Roberts, M.L. Schilsky, American Association for Study of Liver Diseases (AASLD). Diagnosis and treatment of Wilson disease: an update, *Hepatology* 47 (2008) 2089–2111.
- [13] V. Di Lazzaro, F. Manganelli, M. Dileone, F. Notturmo, M. Esposito, M. Capasso, et al., The effects of prolonged cathodal direct current stimulation on the excitatory and inhibitory circuits of the ipsilateral and contralateral motor cortex, *J. Neural Transm.* 119 (2012) 1499–1506.
- [14] R. Dubbioso, P. Moretta, F. Manganelli, C. Fiorillo, R. Iodice, L. Trojano, et al., Executive functions are impaired in heterozygote patients with oculopharyngeal muscular dystrophy, *J. Neurol.* 259 (2012) 833–837.
- [15] A.T. Beck, C.H. Ward, M. Mendelson, J. Mock, J. Erbaugh, An inventory for measuring depression, *Arch. Gen. Psychiatry* 4 (1961) 561–571.
- [16] J.L. Cummings, M. Mega, K. Gray, S. Rosenberg-Thompson, D.A. Carusi, J. Gornbein, The Neuropsychiatric Inventory: comprehensive assessment of psychopathology in dementia, *Neurology* 44 (1994) 2308–2314.
- [17] J.H. Patton, M.S. Stanford, E.S. Barratt, Factor structure of the Barratt impulsiveness scale, *J. Clin. Psychol.* 51 (1995) 768–774.
- [18] A. Perretti, M.T. Pellecchia, B. Lanzillo, G. Campanella, L. Santoro, Excitatory and inhibitory mechanisms in Wilson's disease: investigation with magnetic motor cortex stimulation, *J. Neurol. Sci.* 192 (2001) 35–40.
- [19] K. Jhunjhunwala, D.K. Prashanth, M. Netravathi, B.C. Nagaraju, P.K. Pal, Alterations of cortical excitability and central motor conduction time in Wilson's disease, *Neurosci. Lett.* 553 (2013) 90–94.
- [20] T.P. McGee, C.M. Houston, S.G. Brickley, Copper block of extrasynaptic GABAA receptors in the mature cerebellum and striatum, *J. Neurosci.* 33 (2013) 13431–13435.
- [21] F. Tecchio, G. Assenza, F. Zappasodi, S. Mariani, C. Salustri, R. Squitti, Glutamate-mediated primary somatosensory cortex excitability correlated with circulating copper and ceruloplasmin, *Int. J. Alzheimers Dis.* 2011 (2011) 292593.