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The value of cerebral Doppler venous haemodynamics in the assessment of multiple sclerosis

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ABSTRACT

Iron stores in the white and deep grey matter in course of multiple sclerosis (MS) have never been explained and could be related to abnormalities in venous drainage, but this possibility has never before been investigated.

From an initial cohort of 320 subjects, after application of exclusion criteria, we selected 109 patients affected by MS, and 177 controls respectively composed by age- and sex-matched, healthy aged, and patients affected by other neurological diseases. They blindly underwent transcranial and extracranial Color–Doppler sonographic examination (TCCS-ECD), aimed at investigating five parameters related to normal cerebral venous outflow haemodynamics.

Overall we analyzed 1430 TCSS-ECD parameters. In controls we found 861 normal parameters of cerebral venous return vs. 24 anomalous, whereas in MS 288 parameters were normal and 257 anomalous, respectively. Consequently, each of the considered Doppler haemodynamic parameters, when compared to revised McDonald criteria as a gold standard of MS diagnosis, showed separately a highly significant sensitivity and a noteworthy specificity. However, the detection ≥ 2 parameters in the same subject, never observed in controls, perfectly overlapped the diagnosis of MS (value, 95%CI: sensitivity 100%, 97–100; specificity 100%, 98–100; positive predictive value 100%, 97–100, negative predictive value 100%, 98–100; $p<0.0001$). Moreover, this study demonstrates a significant impairment of cerebral venous drainage in patients affected by MS, a mechanism potentially related to increased iron stores.

Since the time of Charcot's first description, we have known that plaques in multiple sclerosis (MS) are venocentric [1,2]. It has been described that these veins can also be dilated and split the MS lesions longitudinally, as demonstrated by post-mortem studies and MR venography [3–5]. Histological examination of the involved veins sometimes reveals the presence of aspects particular to any status of chronic venous insufficiency, such as fibrin cuffs and perivenous iron deposits, in form of extracellular haemosiderin and iron-laden macrophages [6–9]; Finally, brain atrophy in MS has also been related in cross-sectional and longitudinal studies to T2-hypointense lesions in deep grey matter, suggesting a link between tissue iron deposition and atrophy [10].

All of these elements convinced us to investigate the cerebral venous drainage as a possible mechanism related to increased iron deposition in the MS plaques.

In normal subjects reference parameters of cerebral venous return have been previously studied by the means of echo-color Doppler (ECD) [11–18] and transcranial color-coded Doppler sonography (TCCS) [18–21]. In contrast to MR limitations, these techniques of vascular-imaging are capable of evaluating the cerebral venous haemodynamics pre and post changes of posture and activation of the respiratory thoracic pump, both mechanisms impacting the cerebral venous flow pattern [11–18] (Table 1).

For instance, by means of a transcranial color-coded Doppler sonography (TCCS) examination, we have recently demonstrated that in the cerebral veins of MS patients the hemodynamic parameters are consistently altered, with a high frequency of inversion of the physiological flow direction, suggesting possible venous outflow abnormalities also at an extracranial level [21].

The purpose of this study is to investigate the cerebrovenous haemodynamics by combining TCCS with extracranial Color–Doppler sonographic examination (ECD) in MS patients and in controls.

1. Methods

1.1. Patients and controls

We admitted to the first part of the study 120 patients affected by clinically defined MS (CDMS), diagnosed according to the recommended

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Table 1
Venous glossary and abbreviations used in the article.

Mechanism of the thoracic pump	Increased negative pressure achieved by inspiration increases in turn aspiration of venous blood toward the chest
Postural control of cerebral venous outflow	The internal jugular veins are the predominant outflow route in supine posture. Conversely, the vertebral veins are predominant in upright position. The cross sectional area is modified in accordance with the posture and the drained volume of blood.
ECD	EchoColor–Doppler
TCCS	Transcranial color-coded Doppler sonography
IJV	Internal Jugular Vein
VV	Vertebral Vein
DCVs	Deep cerebral veins, including internal cerebral vein, basal vein, and Galen vein
HM-C; HA-C; OND;	Healthy controls matched for age and gender with MS patients; healthy aged controls; controls affected by other neurological diseases
CSA	Cross sectional area of the IJV
ΔCSA	Difference in CSA assessed in supine and sitting posture in the IJV
Aim of physiologic venous haemodynamics	To permit, by a mono-directional flow, a drainage of a volume of blood per unit of time adequate to a certain tissue. This assumption has to be valid in all conditions.
Reflux	Venous flow reversal to the physiologic direction for a duration >0.88 s in the extracranial pathways, and >0.50 s in the DCVs. In the present study reflux was assessed in different body postures and never under Valsalva manoeuvre.

criteria [22]. We also recruited 200 controls subdivided into three groups: i) 60 healthy subjects homogeneous for age and gender with MS patients (HM-C); ii) 80 healthy subjects older than the median age of the European MS population (HA-C) [23]; and iii) 60 patients affected by other neurological diseases (OND).

1.1.1. Exclusion criteria

We excluded from the study those subjects having, or showing the potential for developing, a nervous system pathology of a venous refluxive and/or obstructive nature, including:

1. Chronic venous insufficiency of the lower limbs
2. History of venous thrombosis and\or post-thrombotic syndrome
3. Genetic thrombophilia
4. Congenital angiodysplasias
5. Congenital vascular malformations
6. Budd–Chiari syndrome
7. Behcet disease
8. Other Vasculitis

By applying the above stated exclusion criteria 109/120 CDMS patients and 177/200 controls entered the study; CDMS were subdivided into 69 with a relapsing-remitting (RR) clinical course, 31 secondary progressive (SP), and 9 primary progressive (PP) [22–24], attributing to each group a relative expanded disability disease score (EDSS) [25]. The control groups included 60 HM-C, 72 HA-C, and 45 OND patients (Tables 2a and 2b).

Table 2b
Demographics of other neurological diseases control population (OND).

	Group OND	Subgroup Neurodegenerative Disease (ALS; Parkinson's)	Subgroup OIND (Myasthenia; MMN)	Subgroup Cerebro-vascular Disease (Stroke; TIA)
	($n=45$)	($n=19$)	($n=7$)	($n=19$)
Age				
median	60	64	50	69
(interquartile range)	(26)	(24)	(12)	(25)
Sex %M	55.5%	47%	57%	58%
M/F	25/20	9/10	4/3	11/8

Patients and controls underwent a noninvasive study of cerebral venous return at the Vascular Lab; the ultrasound technicians and the physicians interpreting the data were blinded to the patient diagnostic category. This study was approved by the Ethical Committee of Ferrara University Hospital.

1.2. Study of cerebral venous return

Cerebral venous return was examined with the subjects positioned on a tilt bed by combining the TCCS methodology for studying the deep cerebral veins (DCVs) [18–21] with that of extracranial EchoColor–Doppler (ECD) for insonating the internal jugular veins (IJVs) and vertebral veins (VVs) [11–18] We focused in particular on the detection of five parameters previously described and related to normal cerebral venous drainage [18]. In the Appendix A is given a detailed technical description of the proposed investigation, performed by using the ultrasonographic instrument Esaote–Biosound My Lab 25, equipped with 2.5 and 7.5–10 Mhz transducers (Genoa, Italy).

1.2.1. Physiologic flow direction in the IJVs and\or VVs with the head in any position

The physiologic direction of venous flow was assessed during a short period of apnea following a normal exhalation, as previously reported, with the head positioned at 0°, +15°, +30°, +45°, +90° in the four extracranial venous drainage pathways [11]. We assess the eventual presence of reflux in the IJVs and VVs in the same experimental condition and never in a forced condition as Valsalva manouevre [15,16]. According to a recent study on reflux time cut-off values, we considered reflux a flow directed toward the brain for a duration >0.88 s [15].

1.2.2. Physiologic flow direction in the DCVs

In human physiology the venous flow in the cerebral veins is monodirectional [19,20]. TCCS investigation allows to assess, through

Table 2a
Demographics of patient and healthy control populations.

	All MS Patients	Group HM-C	Group HA-C	Group MS-RR	Group MS-SP	Group: MS-PP
	($n=109$)	($n=60$)	($n=72$)	($n=69$)	($n=31$)	($n=9$)
Age						
Median	40	37	58	38	44	57
(interquartile range)	(12)	(25)	(21)	(13)	(12)	(14)
Sex %M	41%	46%	40%	40.5%	42%	44%
M/F	45/64	28/32	29/43	28/41	13/18	4/5
EDSS						
Median	2			1.5	5.5	5
(interquartile range)	(3)			(1)	(3)	(3)
Disease duration(years)						
Median	6			4	13	9
(interquartile range)	(10)			(5)	(13)	(12)

Table 3
ECD-TCCS findings of abnormal cerebral venous outflow.

ECD-TCCS finding	MS	Control populations		Sensitivity Specificity PPV-NPV	p
	(N; %)	(N; %)		(95% CI)	
1. Reflux constantly present in an outflow pathway (IJV and\or VV) with the head in any position	76|109 70%	0/177	0%	100% (95–100) 84% (79–89) 70% (60–78) 100% (98–100)	<0.0001
2. Reflux propagated upward to the DCVs	55/109 50%	0/177	0%	100% (93–100) 77% (71–82) 50% (41–60) 100% (98–100)	<0.0001
3. High resolution B-mode evidence of proximal IJV stenoses	30/109 28%	1/177	0.6%	97% (83–99) 69% (63–75) 28% (19–37) 99% (97–100)	<0.0001
4. Flow not Doppler detectable in the IJVs and/or VVs despite numerous deep inspirations	35/109 32%	1/177	0.6%	97% (85–99) 70% (64–76) 32% (23–42) 99% (97–100)	<0.0001
5. Negative ΔCSA in the IJV	61/109 58%	21/177	12%	74% (63–83) 76% (70–82) 56% (46–65) 88% (82–92)	<0.0001
Conclusive analysis: ≥2 positive criteria	109/109 100%	0/177	0%	100% (97–100) 100% (98–100) 100% (97–100) 100% (98–100)	<0.0001

Sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) were tested for significance by the two-sided Fisher exact test, by comparing the gold standard represented by clinical and MRI revised McDonald criteria for diagnosis of MS with the proposed ECD-TCCS protocol.

the transtemporal window, the flow direction and the eventual presence of reflux at least one of the DCVs, including internal cerebral vein, basal vein of Rosenthal, great vein of Galen. Participants were examined in both sitting and supine positions, and the venous flow was elicited by inviting the subject to breathe and setting the reflux time to a value >0.5 s, as previously reported [21].

1.2.3. Absence of high resolution B-mode proximal IJV stenoses

We assessed the absence of stenosing venous imaging reducing the CSA ≥50%, by means of a complete ECD high resolution B-mode exploration of the cervical vessels, and measurement of the CSA of the IJV.

1.2.4. Flow not Doppler detectable in the IJVs and/or VVs

We assessed the lack of a Doppler detectable venous flow in the IJVs and/or VVs despite numerous deep inspirations. In normal subjects this finding was never observed with the head in any position [11], but was reported in supine position in 6% of cases [14].

1.2.5. Postural control of the main cerebral venous outflow pathway

In human physiology ECD clarified that the IJV is the predominant outflow pathway in the supine position, confirmed by an increased cross sectional area (CSA) related to increased blood volume in that posture; redirection of venous flow to the VVs occurs in the upright position, with compliant reduction of the CSA of the IJV [11,12]. Consequently we measured:

- The CSA of both IJVs, in supine and sitting postures.
- The difference in CSA (ΔCSA) obtained by subtracting the CSA measured in the supine from that in the erect position [18].

1.3. Statistical analysis

Clinical and demographic characteristics are expressed as median and 25th–75th percentile. CSA in sitting and supine postures, ΔCSA, are expressed as mean ± SD. Differences among groups were tested for significance with the one-way ANOVA analysis of variance.

The two-sided Fisher's exact test was used for determining sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV) of the proposed ECD-TCCS technique respect to the gold standard represented by the revised Mc Donald criteria, and by comparing the whole MS group with a group including all controls.

P-values up to 0.05 were considered statistically significant.

2. Results

2.1. Patients and controls

Tables 2a and 2b show clinical and demographic characteristics for the entire group of MS patients, as well as for the subgroups. Significant differences were found, as expected, in the followings:

- Age: SP vs RR, $p<0.01$. PP vs RR, $p<0.01$ (ANOVA).
- EDSS: SP vs RR, $p<0.01$. PP vs RR, $p<0.01$ (ANOVA).
- Disease duration: SP vs RR, $p<0.01$ (ANOVA).

2.2. Study of cerebral venous return

2.2.1. Physiologic flow direction in the IJVs and\|or VVs with the head in any position

Venous flow direction was consistently directed toward the heart with the head in any position, and increased by inspiration in all controls. Bidirectional flow limitedly to one posture was sometimes observed in controls. By contrast, the persisting presence of reflux with the head positioned at 0° and +90° (supine and sitting posture), as well as at +15°, +30°, +45°, in at least one IJV and/or VV was never observed in any subject among the three control populations. This finding was observed in 77/109 MS patients (70%), particularly in those with RR and SP courses, in 46/69 (66%) and in 28/31 (90%), respectively (Table 3), (Fig. 1).

2.2.2. Physiologic flow direction in the DCVs

In controls, flow was always monodirectional in the DCVs, including OND patients, and reflux was detected at the activation of the thoracic pump exclusively in half of the MS group (Table 3). In the PP course, reflux in the DCVs was observed more frequently as

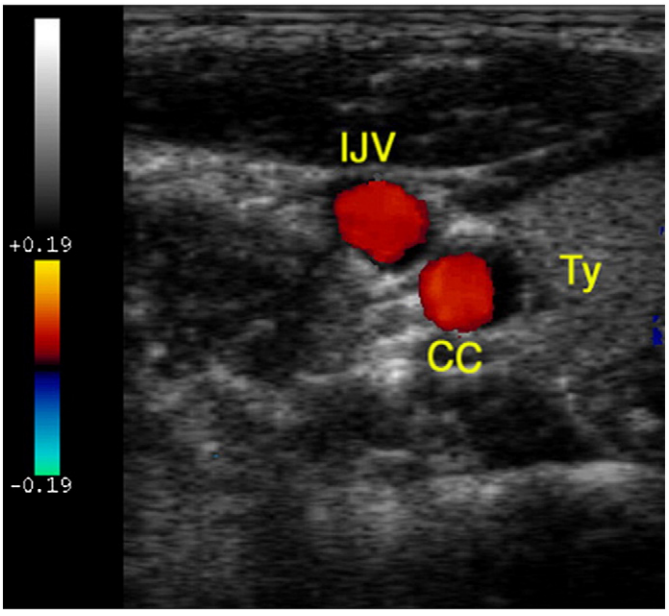

Fig. 1. *Reflux detection in the IJVs.* ECD cervical transversal access, at the level of the thyroid (Ty), showing IJV and carotid artery (CC) with the same direction of flow toward the brain assessed by the red colour code (reflux).

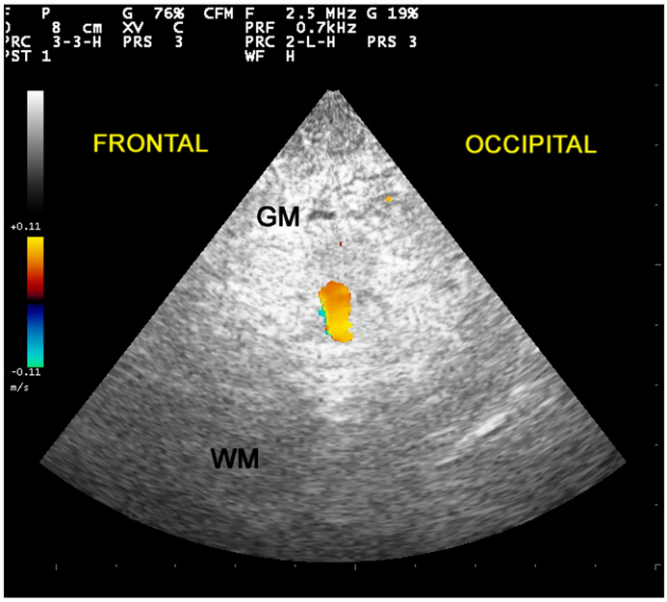

Fig. 2. *Reflux is documented (red colour) in veins of the subcortical gray matter coming from veins of the white matter.* TCSS up to the ventricular plane of the trans-temporal window, axial section: left) Normal emptying (red colour) assessed in veins of the subcortical grey matter toward the DCVs during inspiration, in a case of PP male patient aged 60; right) In contrast, reflux is documented blue colour during expiration.

compared to RR-SP (78% vs. 41%, respectively) and never in controls. Interestingly, reflux in progressive cases was also observed in veins going up-ward the cortical or subcortical grey matter starting from the ventricular plane (Fig. 2).

2.2.3. Absence of high resolution B-mode proximal IJV stenoses

B-mode analysis at high resolution of the cervical veins allowed for the direct observation, in 30 MS patients vs. 1 control, of the presence of closed stenosis in the proximal segment of an IJV, almost always the left (Table 3, Fig. 3).

2.2.4. Flow not Doppler detectable in the IJVs and/or VVs

The lack of flow in one of the extra-cranial venous routes, limitedly to the supine posture, was observed once in controls. A further finding never seen in controls but recorded in 33% of RR-SP and in 35% of PP cases, respectively, was the lack of Doppler detectable flow velocity in the IJVs and/or VVs despite numerous deep inspirations (Table 3), in any position of the body.

2.2.5. Postural control of the main cerebral venous outflow pathway

In Fig. 4, the physiologic postural control of cerebral venous outflow in the IJVs route in both healthy control populations (HM-C, HA-C) is very apparent. CSA values in the sitting position are consistently lower than those assessed in the supine position, resulting in a wider ΔCSA [18], also obvious in the figure. The same correct physiologic response to a change in hydrostatic pressure condition was also demonstrated in the OND group, with no significant differences from the CSA assessed in the HM-C and HA-C groups. In contrast, Fig. 4 shows an overturning of this physiologic mechanism of postural regulation in the entire MS population. Redistribution of blood in the supine posture, in accordance with the principle of communicating vessels, seemed to be impeded in MS patients, and CSA at 0° was significantly lower in the MS patients than in the healthy controls, and even in the OND patients. Consequently, the ΔCSA levels were significantly reduced in MS as compared to the three control groups, as shown in Fig. 4. Finally, ΔCSA was negative in 58% of MS cases vs. 12% of the three control groups, as shown in Table 3.

2.3. Sensitivity, specificity, PPV, NPV

Overall we analyzed 1430 TCCS-ECD parameters of cerebral venous haemodynamics. In controls we found 24 positive vs. 861 negative parameters of abnormal cerebrovenous return, whereas in MS 257 parameters were positive and 288 negative, respectively. Sensitivity, specificity, PPV, and NPV were calculated for each parameter, and are given in Table 3. Each of the considered Doppler haemodynamic parameters, when compared to revised McDonald criteria as a gold standard of MS diagnosis, showed separately a highly significant sensitivity and a noteworthy specificity. However, the detection ≥ 2 parameters in the same subject, never observed in controls, perfectly overlapped the diagnosis of CDMS [22] (value, 95%CI: sensitivity 100%, 97–100; specificity 100%, 98–100; positive predictive value 100%, 97–100, negative predictive value 100%, 98–100; $p<0.0001$).

3. Discussion

The principal result of our study is the original description of the overturning of the physiologic regulation of venous return assessed in our CDMS population, with parameters that are significantly different from those of all control groups (Table 3, Figs. 1–4). The latter include also older subject (Tables 2a) and OND patients (Tables 2b). Had Doppler hemodynamic anomalies been present in the healthy aged control group, we would not have been able to maintain that they have a role in MS, since these subjects are older than the median age of onset of MS [23]. Therefore, the absence of Doppler venous outflow abnormalities in the OND patients would indicate that in no other disease of the nervous system can this mechanism play a role, neither in other pathologies that, like MS, present neurodegenerative, neuroimmunitary, and neurovascular aspects, nor iron deposition as epiphenomenon [26–31].

The diagnostic accuracy of the detection of ≥ 2 anomalous parameters of cerebral Doppler venous return in recognizing CDMS in comparison to the gold standard of the revised Mc Donald criteria, clearly indicate the high potentiality of the experimented cerebral Doppler venous investigation. It could be in future proposed as a novel, non-invasive, cost-effective tool in the assessment of MS. However, further studies are necessary to achieve this goal, in order to evaluate the reproducibility, the inter and intra observer variability, as well as the need of specific training of the operators.

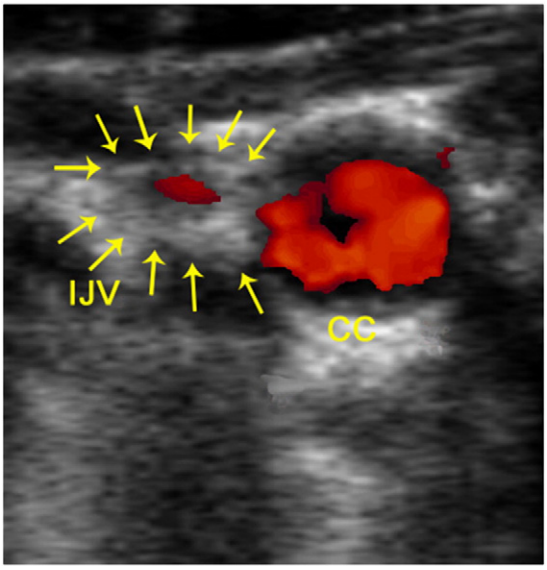

Fig. 3. *B-mode detection of IJV stenoses.* ECD cervical transversal access: stenoses of the left internal jugular vein (IJVl) yellow arrows with severe reduction of the lumen. Reflux flow is documented by the same red colour inside the lumen of the common carotid artery (CC), indicating a flow direction upward the brain.

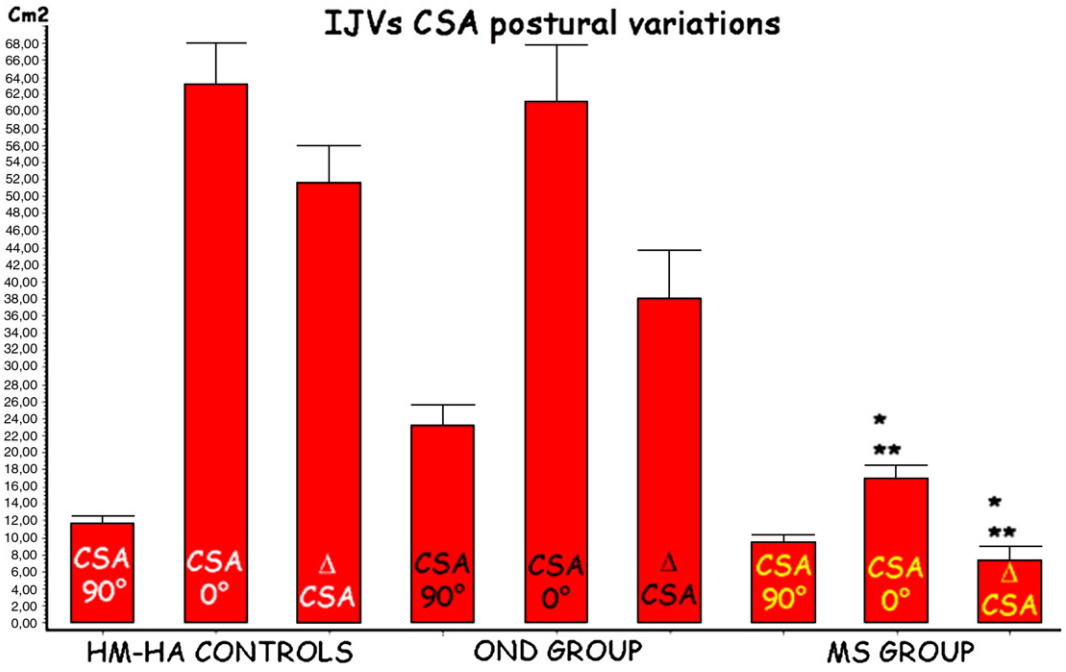

Fig. 4. *CSA and ΔCSA in MS and in controls.* CSA in sitting and supine postures, and ΔCSA measured in the HM-C/HA-C healthy control groups, and in the OND control group, respectively. The increasing of CSA in the IJVs in the supine posture is quite apparent. In contrast, this mechanism is lost in the MS population, and the differences are significant as compared to the healthy controls (*$p<0.001$), and to OND (**$p<0.001$), respectively (ANOVA).

The physiologic mechanism of postural control of cerebral venous outflow was found significantly compromised in MS. Our study demonstrates the paradoxical behaviour of the IJVs in MS patients during change in hydrostatic pressure, leading to significant reduction of ΔCSA, which finally represents a rough estimation of the compliance of the jugular system (Table 3, Fig. 4) [18].

Among the other anomalies discovered, certainly the persistence of the reflux in the IJVs–VVs in any position of the head is the criterion most significantly associated with CDMS. This reflux has a mechanism that differs from that caused by incompetence of the jugular valve, yet. In the latter case, valvular insufficiency tested with Valsalva can be related to a picture of transient global amnesia [16]. In our study extracranial venous reflux occurred naturally in any body position without the need to elicit it by a forced movement, becoming a long-lasting reverse flow (Fig. 1). Therefore, approximately in half of cases, extracranial reflux seems to be propagated up-ward to the DCVs; in progressive cases we also observed reflux directed toward the subcortical grey matter (Fig. 2)[21]. It should be interesting to assess in the deep grey matter the amount of iron stores by means of advanced MRI, in relation to such a TCCS finding [10,31].

What did ECD-TCCS tell us about the origin of this reflux? The detection of reflux would be interpreted together either the consistent presence of blocked outflow in the main outflow pathways (32%, Table 3), or with the significant reduction of venous compliance, well apparent in Fig. 4. From a haemodynamic point of view, all these findings point up the suspected presence of venous obstruction distally to the point of measurement, which cannot be crossed through the physiologic postural and respiratory mechanisms. Venous stenoses was definitely observed at the cervical level in 28% of CDMS (Fig. 3), leading us to stronghly suspect the presence of venous obstruction even in the chest. ECD is of course limited in investigating the venous outflow route at the thoracic level; the hypothesis of venous stenosis should require further investigation of the main venous pathways at the thoracic level, even by selective venography, expecially when the Doppler non invasive parameter are consistently altered. Giving the results of the present study, venography could be proposed when the cut-off of ≥ 2 positive parameters are detected by the ECD-TCCS investigation [32].

Alternatively, it could be hypothesized that venous intracranial reflux results not only from a intra-thoracic outflow block, but also from an awry intracerebral distribution of blood flow, or from improper relationship between pressure in intracranial veins and pressure of cerebrospinal fluid; relevant data, however, are missing, and more investigations and experiments, undoubtedly, should be done.

Unfortunately, the impact of the cerebrovenous system on the different physiological parameters of normal brain function has not been fully estimated to date, but our preliminary data suggest that it may be greater than previously assumed. It is accepted that a chronic insufficiency of venous drainage leads to increased iron stores in the affected tissue [9,26]. Several studies investigated the relationship between iron overload and neurodegenerative disease such as Alzheimer disease, Parkinson disease, and other disorders [27–30]. However, differently from the above reported diseases, in which the increased iron stores are not related to impaired venous function, our study demonstrates a strong association between MS and anomalies of cerebral venous drainage. The hypothesis that iron plays a role in MS pathogenesis is strongly supported by a recent study of induced iron deficiency in mice with experimental autoimmune encephalomyelitis (EAE), a useful animal model of the disease [33]. EAE did not develop in iron-deficient mice; on the contrary, the incidence of EAE was 71% in mice with normal iron levels, and 62% in iron-overloaded mice.

In conclusion, our findings support the concept of impairment in MS of the physiologic extracranial venous outflow mechanisms, with obvious reflection on cerebral tissue drainage, in turn potentially linked to the inflammatory aspect of MS [34]. If confirmed, these findings could lead to important advancements in the diagnosis and in the understanding of MS.

4. Competing financial interests

None.

5. Funding

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Appendix A. Technical methodology for ECD-TCCS investigation

Patient position

The posture of the subject under examination obtains a crucial part determining the main route of cerebral outflow. For this reason the subject should be investigated at least in both supine and sitting position (0°and 90°). This objective can be realized with the subjects being positioned on a tilt bed/chair.

Equipment and transducers

The investigation of the cerebral venous haemodynamics can envisage the contemporaneous analysis of both the intracranial and extracranial pathways, by combining with the same ultrasound machine respectively the examination of the DCVs with that of the IJVs–VVs by means of the proper ultrasonographic probe. The transducer at the intracranial level is at low frequency, usually 2.5 Mhz, whereas at the cervical level is at high frequency, 7.5–10 Mhz or more, according to the different depth of the veins respective to the body surface where the transducer is placed.

DCVs flow direction assessment

The transducer is placed at the level of the trans-temporal bone window, and the depth of the insonation is adjusted to 10 cm. At an insonation depth of about 7 cm it is possible to consistently identify the echo-lucent third ventricle, limited by two echogenic bright margins, as well as the two comma-shaped frontal horns of the lateral ventricles. Subjects can be examined in both sitting and supine positions and the venous flow is enhanced by inviting the subject to breathe, and setting the PRF of the instrument to lower values, ranging between 0.5 and 1.4. Using the trans-temporal acoustic bone window, it is possible to insonate around the third ventricle at least one of the DCVs, or all three branches, including basal veins of Rosenthal (flow expected up-ward and to-ward the Galen vein), great vein of Galen (flow expected to-ward the occipital side into the straight sinus), and internal cerebral veins (flow expected down-ward and to-ward the Galen vein) (Fig. 5). Reflux is a reverse flow for a duration >0.5 s.

IJVs–VVs flow direction assessment

Examination is performed with high frequency transducers as above reported. Subjects should be examined at least in sitting and supine positions. Flow recording begins two minutes after the change in posture and after several deep breaths in order to permit blood redistribution in the venous system. For each assessment the direction of flow is analyzed either with the pulsed wave mode and the sample placed in the vessel, at a 60° angle, or with the colour coded mode, by comparing the colour of the flow in the IJV/VV with that of the satellite carotid and/or vertebral artery, respectively. Either the IJVs or the VVs can be examined by using both the transversal and/or the longitudinal cervical access. The operator uses minimal pressure over the skin in order to prevent compressing the vein and thereby affecting the measurement. Flow direction is normally directed to-ward the heart. Reflux is a reverse flow assessed in the respiratory pause for a duration >0.88 s.

Assesment of CSA

The level of IJV insonation corresponds to the thyroid gland, and the point of measurement is outlined over the skin with waterproof pen. Patients and controls were examined in sitting and supine positions using the transversal access (by the longitudinal scan the operator is unaware if the CSA is elliptical or circular, so affecting the measurement).

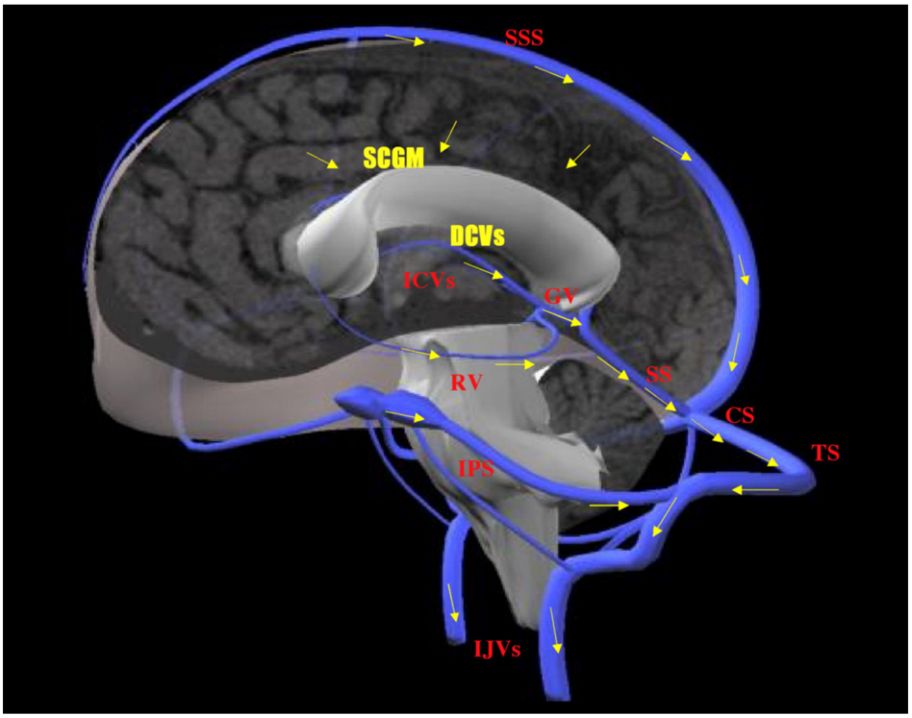

Fig. 5. *Appendix: flow direction in the cerebral venous system.* Flow in the cerebral veins is monodirectional (yellow arrows). Any reverted flow for a duration >0.5 s is considered reflux: the veins of the subcortical grey matter (SCGM) drain into the deep cerebral veins DCVs (ICVs internal cerebral veins, RV Rosenthal vein, GV Galen vein). Legenda: SSS superior saggital sinus, IPS inferior petrosus sinus, SS straight sinus, CS confluence sinus, TS transverse sinus, IJVs internal jugular veins.

The operator uses minimal pressure over the skin in order to prevent compressing the vein and thereby affecting the measurement. CSA is measured in the supine and sitting positions by means of the software for elliptical or circular shapes included in the ECD instrumentation, separately in the right and left IJV. Alternatively, it is possible to measure the diameter by means of the software and to calculate the CSA according to geometric formulas respectively for circle and ellipse. In case of spontaneous fluctuations in CSA due to activation of the thoracic pump, CSA can be measured during a short period of apnea following a normal exhalation. ΔCSA is calculated by subtracting the CSA measured in the supine position from that in the erect position, separately in the right and left IJV.

References

[1] Charcot JM. Histology of 'sclerose en plaque' [in French]. Gazette Hosp (Paris) 1868;41:554–66.
[2] Barnett MH, Sutton I. The pathology of multiple sclerosis: a paradigm shift. Curr Opin Neurol 2006;19:242–7.
[3] Fog T. The topography of plaques in multiple sclerosis with special reference to cerebral plaques. Acta Neurol Scand Suppl 1965;15:1–161.
[4] Schelling F. Damaging venous reflux into the skull or spine: relevance to multiple sclerosis. Med Hypotheses 1986;21:141–8.
[5] Tan IL, van Schijndel RA, Pouwels PJ. MR venography of multiple sclerosis. Am J Neuroradiol 2000;21:1039–42.
[6] Adams CW, Poston RN, Buk SJ. Pathology, histochemistry and immunocytochemistry of lesions in acute multiple sclerosis. J Neurol Sci 1989;92:291–306.
[7] Adams CW. Vascular aspects of multiple sclerosis. A colour atlas of multiple sclerosis and other myelin disorders. London: Wolfe Medical Publication; 1989. p. 184–7.
[8] Adams CW. Perivascular iron deposition and other vascular damage in multiple sclerosis. J Neurol Neurosurg Psychiatry 1988;51:260–5.
[9] Zamboni P. Iron-dependent inflammation in venous disease and proposed parallels in multiple sclerosis. J R Soc Med 2006;99:589–93.
[10] Zivadinov R, Bakshi R. Role of MRI in multiple sclerosis II: brain and spinal cord atrophy. Front Biosci Jan 1 2004;9:647–64.
[11] Valdueza JM, von Munster T, Hoffman O, Schreiber S, Einhaupl KM. Postural dependency of the cerebral venous outflow. Lancet 2000;355:200–1.
[12] Gisolf J, van Lieshout JJ, van Heusden K, Pott F, Stok WJ, Karemaker JM. Human cerebral venous outflow pathway depends on posture and central venous pressure. J Physiol 2004;560:317–27.
[13] Schreiber SJ, Lurtzing F, Gotze R, Doepp F, Klingebiel R, Valdueza JM. Extrajugular pathways of human cerebral venous blood drainage assessed by duplex ultrasound. J Appl Physiol 2003;94:1802–5.
[14] Doepp F, Schreiber SJ, von Münster T, Rademacher J, Klingebiel R, Valdueza JM. How does the blood leave the brain? A systematic ultrasound analysis of cerebral venous drainage patterns. Neuroradiology 2004;46:565–70.
[15] Nedelmann M, Eicke BM, Dieterich M. Functional and morphological criteria of internal jugular valve insufficiency as assessed by ultrasound. J Neuroimaging 2005;15:70–5.
[16] Sander K, Sander D. New insights into transient global amnesia: recent imaging and clinical findings. Lancet Neurol 2005;4:437–44.
[17] Lichtenstein D, Saïfi R, Augarde R, Prin S, Schmitt JM, Page B, et al. The Internal jugular veins are asymmetric. Usefulness of ultrasound before catheterization. Intensive Care Med 2001;27:301–5.
[18] Menegatti E, Zamboni P. Doppler haemodynamics of cerebral venous return. Curr Neurovasc Res 2008;5:259–64.
[19] Baumgartner RW, Nirkko AC, Müri RM, Gönner F. Transoccipital power-based color-coded duplex sonography of cerebral sinuses and veins. Stroke 1997;28:1319–23.
[20] Stolz DE, Kaps M, Kern A, Babacan SS. Reference data from 130 volunteers transcranial color-coded duplex sonography of intracranial veins and sinuses. Stroke 1999;30:1070–5.
[21] Zamboni P, Menegatti E, Bartolomei I, Galeotti R, Malagoni AM, Tacconi G, et al. Intracranial venous haemodynamics in multiple sclerosis. Curr Neurovasc Res 2007;4:252–8.
[22] Polman CH, Reingold SC, Edan G, Filippi M, Hartung HP. Diagnostic criteria for multiple sclerosis: 2005 revisions to the "McDonald Criteria". Ann Neurol 2005;58:840–6.
[23] Confavreux C, Vukusic S. Natural history of multiple sclerosis: a unifying concept. Brain 2006;129:606–16.
[24] Lublin DF, Reingold SC. Defining the clinical course of multiple sclerosis: results of an international survey. Neurology 1996;46:907–11.
[25] Kurtzke JF. Rating neurological impairment in multiple sclerosis: an expanded disability scale (EDSS). Neurology 1983;33:1444–52.
[26] Bergan JJ, Schmid-Schonbein GW, Smith PD, Nicolaides AN, Boisseau MR, Eklof B. Chronic venous disease. N Engl J Med 2006;355:488–98.
[27] Sipe JC, Lee P, Beutler E. Brain iron metabolism and neurodegenerative disorders. Dev Neurosci 2002;24:188–96.
[28] Dekker MC, Giesbergen PC, Njajou OT, wan Swieten JC, Hofman A, Breteler MM, et al. Mutation in the hemochromatosis gana (HFE), Parkinson's disease and parkinsonism. Neurosci Lett 2003;348:117–9.
[29] Lehmann DJ, Worwood M, Ellis R, Wimhurst VL, Merryweather-Clarke AT, Warden DR, et al. Iron genes, iron load and risk of Alzheimer's disease. J Med Genet 2006;43:e52.
[30] Sullivan JL. Is stored iron safe? J Lab Clin Med 2004;144:280–4.
[31] Haacke EM, Ayaz M, Khan A, Manova ES, Krishnamurthy B, Gollapalli L, et al. Establishing a baseline phase behavior in magnetic resonance imaging to determine normal vs. abnormal iron content in the brain. J Magn Reson Imaging 2007;26:256–64.
[32] Zamboni P, Galeotti R, Menegatti E, Malagoni AM, Tacconi G, Dall'ara S, et al. Chronic Cerebrospinal venous insufficiency in patients with multiple sclerosis. J Neurol Neurosurg Psychiatry December 2008 on-line first.
[33] Grant SM, Wiesinger JA, Beard JL, Cantorna MT. Iron-deficient mice fail to develop autoimmune encephalomyelitis. J Nutr 2003;13:2635–8.
[34] Zamboni P, Lanzara S, Mascoli F, Caggiati A, Liboni A. Inflammation in venous disease. Int Angiol 2008;27:361–9.