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Blood–brain barrier leakage years after pre-eclampsia: dynamic contrast-enhanced 7-Tesla MRI study

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KEYWORDS: 7 Tesla; blood–brain barrier; cerebrovascular disorder; dynamic contrast-enhanced MRI; pre-eclampsia

CONTRIBUTION

What are the novel findings of this work?
Dynamic contrast-enhanced MRI at 7 Tesla revealed subtle blood–brain barrier (BBB) leakage several years after a pre-eclamptic pregnancy. The BBB leakage rate and fractional leakage volume were globally higher in formerly pre-eclamptic women than in control women, indicating diffusively spread endothelial dysfunction as a pathophysiological mechanism.

What are the clinical implications of this work?
Pre-eclampsia is associated with an increased cerebrovascular risk. BBB leakage often precedes cerebrovascular disorders. This study demonstrates global BBB disruption years after pre-eclampsia. The BBB could be a novel target for treatment and prevention strategies aimed at reducing the cerebrovascular risk after pre-eclampsia.

ABSTRACT

Objective *Pre-eclampsia is a hypertensive complication of pregnancy that is associated with an increased risk of long-term cardiovascular and cerebrovascular disorders. Although the underlying mechanism of persistent susceptibility to cerebral complications after pre-eclampsia remains largely unclear, impaired blood–brain barrier (BBB) integrity has been suggested to precede several cerebrovascular diseases. In this study, we aimed to investigate the integrity of the BBB years after pre-eclampsia.*

Methods *This was an observational study of premenopausal formerly pre-eclamptic women and controls with a history of normotensive pregnancy who underwent cerebral magnetic resonance imaging (MRI) at ultra-high field (7 Tesla) to assess the integrity of the BBB. Permeability of the BBB was determined by assessing leakage rate and fractional leakage volume of the contrast agent gadobutrol using dynamic contrast-enhanced MRI. BBB leakage measures were determined for the whole brain and lobar white and gray matter. Multivariable analyses were performed, and odds ratios were calculated to compare women with and those without a history of pre-eclampsia, adjusting for potential confounding effects of age, hypertension status at MRI and Fazekas score.*

Results *Twenty-two formerly pre-eclamptic women (mean age, 37.8 ± 5.4 years) and 13 control women with a history of normotensive pregnancy (mean age, 40.8 ± 5.5 years) were included in the study. The time since the index pregnancy was 6.6 ± 3.2 years in the pre-eclamptic group and 9.0 ± 3.7 years in controls. The leakage rate and fractional leakage volume were*

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significantly higher in formerly pre-eclamptic women than in controls in the global white (P = 0.001) and gray (P = 0.02) matter. Regionally, the frontal (P = 0.04) and parietal (P = 0.009) cortical gray matter, and the frontal (P = 0.001), temporal (P < 0.05) and occipital (P = 0.007) white matter showed higher leakage rates in formerly pre-eclamptic women. The odds of a high leakage rate after pre-eclampsia were generally higher in white-matter regions than in gray-matter regions.

Conclusion *This observational study demonstrates global impairment of the BBB years after a pre-eclamptic pregnancy, which could be an early marker of long-term cerebrovascular disorders.* © *2022 The Authors. Ultrasound in Obstetrics & Gynecology published by John Wiley & Sons Ltd on behalf of International Society of Ultrasound in Obstetrics and Gynecology.*

INTRODUCTION

Pre-eclampsia is a hypertensive complication of pregnancy that affects multiple organ systems[1]. A history of pre-eclampsia raises the long-term risk of cerebrovascular disorders[2,3]. Impaired cerebrovascular function during a pre-eclamptic pregnancy is thought to originate from decreased cerebrovascular resistance as a consequence of the loss of cerebral blood flow autoregulation, which is accompanied by hyperperfusion, blood–brain barrier (BBB) disruption and vasogenic edema[4–9]. An increase in BBB permeability has been demonstrated in animal models exposed to blood plasma from women with pre-eclampsia[10–14]. However, studies assessing BBB permeability in pre-eclamptic women without overt neurological disturbances are lacking, and potential long-term effects of pre-eclampsia on the BBB have not been assessed.

The BBB plays an important role in maintaining the homeostasis of the brain environment, which is essential for brain health[15]. BBB dysfunction may create an unfavorable microenvironment with eventual neuronal dysfunction and has been associated with a decrease in cognitive functioning[15,16]. Given that formerly pre-eclamptic women report subjective cognitive and psychological problems years after a pregnancy complicated by pre-eclampsia[17,18], the long-term condition of the BBB may provide new insights into the underlying pathophysiological mechanism of persistent susceptibility to cerebral disorders in this population.

Localized BBB dysfunction can be investigated using dynamic contrast-enhanced magnetic resonance imaging (DCE-MRI)[19] by measuring the leakage of paramagnetic contrast agent into the brain tissue[20,21]. While some pathologies, such as high-grade tumors[22] and multiple sclerosis[23], show relatively high contrast agent leakage into a lesion, other disorders, such as cerebral small vessel disease (cSVD), have shown subtler contrast agent leakage[24]. Considering the cognitive[18] and psychological[17] problems and white-matter (WM) abnormalities[25–27] observed in patients following pre-eclampsia, which are similar to the symptoms observed in early cSVD, subtle contrast agent leakage from the cerebral microvasculature into the brain tissue may also be present in formerly pre-eclamptic women. Ultra-high field (7-Tesla) MRI, instead of clinically used 1.5- or 3-Tesla MRI, can be employed to aid in the detection of subtle BBB leakage[28,29]. In this study, we used a dedicated 7-Tesla DCE-MRI protocol to investigate differences in BBB permeability between formerly pre-eclamptic women and controls with a history of normotensive pregnancy.

METHODS

Participants and procedure

This was an observational study of premenopausal formerly pre-eclamptic women and premenopausal control women with a history of normotensive pregnancy, which was carried out at the Maastricht University Medical Center in The Netherlands and was approved by the local medical ethics committee. Women who participated in a large ongoing investigation into cardiovascular problems related to pregnancy (the Queen of Hearts study (ClinicalTrials.gov identifier: NCT02347540)) were invited to undergo additional testing. Additionally, women were recruited by their obstetric caregiver and through advertisements. All women provided written informed consent prior to inclusion.

Premenopausal women over 18 years old were considered eligible for the study. Pre-eclampsia was defined as new-onset hypertension (systolic blood pressure $\geq$ 140 mmHg and/or diastolic blood pressure $\geq$ 90 mmHg) in combination with the development of proteinuria ($\geq$ 300 mg/24 h or a protein-to-creatinine ratio of $\geq$ 0.3) and/or significant end-organ dysfunction after 20 weeks of pregnancy. The control group had a history of normotensive pregnancy not complicated by pre-eclampsia, hypertension, intrauterine growth restriction and/or placental abruption. In the control group, the first pregnancy was considered to be the index pregnancy. In the pre-eclampsia group, the first pre-eclamptic pregnancy was considered to be the index pregnancy. In both groups, women could participate if they had preterm delivery. Women could not participate if they had delivered within the past 6 months or more than 30 years ago, were diagnosed with chronic hypertension, renal disease or autoimmune disease prior to the index pregnancy, or if they had a contraindication to MRI, such as permanent facial tattoos or an intrauterine device.

Using a standardized format, trained interviewers recorded information about obstetric and medical history, age, educational level and lifestyle habits, such as smoking and alcohol use. Smokers were defined as women who were current smokers or had smoked in the past, while alcohol use was defined as consumption of at least three units of alcohol a week. Height, weight, blood pressure and heart rate were measured on site. Women were diagnosed with hypertension if they had systolic blood pressure $\geq$ 140 mmHg and/or diastolic blood pressure $\geq$ 90 mmHg, or used antihypertensive drugs. Body mass index was calculated by dividing body weight (in kg)

by the square of height (in m). Body surface area was calculated using the formula of Du Bois and Du Bois[30]. Serum creatinine was determined to assess the glomerular filtration rate, used to define kidney health, to inspect for potential risk related to administration of contrast agent. Blood pressure was measured in a sitting position using a semiautomatic oscillometric device (Dinamap Vital Signs Monitor 1846; Critikon, Tampa, FL, USA) at 3-min intervals. The median of 11 measurements was used to represent blood pressure. Educational level was recorded using a scale from 1 (primary school) to 8 (academic level). Educational level was categorized further into three subgroups: low (1–3), average (4–6) and high (7–8) following the International Standard Classification of Education.

MRI

Images were obtained using a 7-Tesla MRI system (Magnetom; Siemens Healthineers, Erlangen, Germany) with a 32-channel phased-array head coil (Nova Medical Inc., Wilmington, MA, USA). To improve the image (i.e. transmit field) homogeneity, dielectric pads were placed proximal to the temporal lobe[31].

To detect BBB leakage, a DCE-MRI imaging protocol with dual-time resolution was used, which consisted of two nested T_1-weighted slow and fast sequences[29,32]. Before the administration of contrast agent (gadobutrol), precontrast scans of the slow and fast sequences were acquired. Subsequently, contrast agent was injected into the antecubital vein using a power injector (1.0 M gadobutrol, 3 mL; flow rate of 0.3 mL/s), followed by a 20-mL saline flush.

To visualize the anatomy and to enable conversion of signal intensity changes to gadobutrol concentration, quantitative T_1-mapping was performed prior to the administration of contrast agent. Additionally, whole brain transmit field (B_1^+) mapping and a fluid-attenuated inversion recovery (FLAIR) sequence were performed to improve segmentation. Details of the imaging sequences used are provided in Appendix S1 and Table S1.

An expert radiologist (S.C.G.), blinded to group status, assessed all MRI scans for incidental findings. Additionally, the extent of WM hyperintensities (WMH), defined as WM abnormalities that appear as areas of hyperintense signal on FLAIR images, was investigated using the Fazekas scale[33]. WMH are considered to be of cerebral microvascular origin and are related to hypertension and cerebrovascular risk factors. The Fazekas score provides an overall impression of the presence of WMH in the entire brain.

Image processing

Preprocessing included transmit field correction[34], bias field correction, skull stripping and gradient distortion correction. The preprocessed images were segmented automatically into subject-specific gray matter (GM) and WM regions (FreeSurfer software, version 6.0[35]; Laboratory for Computational Neuroimaging, Athinoula A. Martinos Center for Biomedical Imaging, Massachusetts General Hospital, Charlestown, MA, USA). The segmentation was inspected visually and corrected manually when necessary.

To correct for head displacement, all dynamic images were registered to an averaged precontrast image (FLIRT, FMRIB version 6.0.1; Functional MRI of the Brain Group, Oxford, UK). A participant-specific signal time course of the blood was obtained in the superior sagittal sinus through manual delineation.

BBB leakage quantification

While the data obtained using the fast DCE-MRI scan were used only to assess the concentration of gadobutrol in the blood, BBB leakage in the tissue was determined using the slow scan data[29,32]. Voxel-wise pharmacokinetic modeling was applied using the graphical Patlak approach to calculate the BBB leakage rate from the blood plasma to the brain tissue (slope, K_i) and the fractional blood plasma volume in the capillaries (intercept, v_p). Owing to the expected low leakage, the spatial distribution of K_i was noisy. Therefore, a histogram approach was used to obtain the corrected K_i (magnitude of the K_i) and the fractional leakage volume (v_L, the spatial extent of detectable leakage) as the area under the histogram curve after the subtraction of noise[32]. This histogram noise correction approach is explained in Appendix S2 and visualized in Figure S1. The mean BBB measures were calculated for the global GM and WM, deep GM, whole cortical GM and the GM and WM of the frontal, temporal, occipital and parietal lobes. The analysis was performed in MATLAB (R2018b; MathWorks Inc., Natick, MA, USA).

Statistical analysis

Demographic and clinical characteristics were compared between the pre-eclampsia and control groups using an independent-sample, two-sided Student *t*-test for continuous variables and the Pearson χ^2 test for categorical variables.

BBB leakage measures of the two groups, including K_i, v_p and v_L, are presented as mean ± SD. Frequency distribution, normal probability plot and the Shapiro–Wilk test were used to check for normality, and Levene's test was used to test for homogeneity of variances. The K_i, v_L and v_p values of the global GM and WM, deep GM, whole cortical GM and the GM and WM of individual brain lobes were assessed between the pre-eclampsia and control groups using the Mann–Whitney *U*-test.

Additionally, odds ratios were calculated to measure the association between higher BBB leakage measures and a history of pre-eclampsia. BBB leakage was considered high if the values were greater than the mean + 2 SD of those of the control women. Finally, to check whether the observed differences were significant after adjusting for *a priori* defined potential covariates, including age, hypertension status at MRI and Fazekas score, multivariable linear regression analysis was used to obtain corrected *P*-values, and multiple logistic regression was used to estimate adjusted odds ratios; $P < 0.05$ was considered to indicate

statistical significance. Statistical analysis was performed using statistical software SPSS version 23.0 (IBM Corp., Armonk, NY, USA).

RESULTS

Twenty-two formerly pre-eclamptic women and 13 parous controls participated in the study (Figure 1). Table 1 presents the demographic and clinical characteristics of the groups. In addition to hypertension, all formerly pre-eclamptic women developed proteinuria after 20 weeks of the index pregnancy. The average time since the index pregnancy was 6.6 years (range, 1–13 years) in formerly pre-eclamptic women and 9.0 years (range, 4–15 years) in control women. Fewer women with a history of pre-eclamptic pregnancy had a subsequent pregnancy. None of the women in the pre-eclampsia or control group suffered from cardiovascular comorbidities, such as heart attack, stroke, diabetes mellitus or deep vein thrombosis. Formerly pre-eclamptic women had significantly higher systolic and diastolic blood pressure than did controls. Demographic and clinical characteristics of the formerly pre-eclamptic women participating in this study were not significantly different from those of all formerly pre-eclamptic women participating in the larger ongoing Queen of Hearts study (Table S2).

Examples of leakage maps in a formerly pre-eclamptic woman and a control woman are presented in Figure 2. The results of the BBB leakage assessment in various brain regions are presented in Table 2. K_i in the global WM and GM was significantly higher in formerly pre-eclamptic women than in controls ($P = 0.001$ and $P = 0.02$, respectively). The odds of a high K_i were 37 times higher for the global WM and four times higher

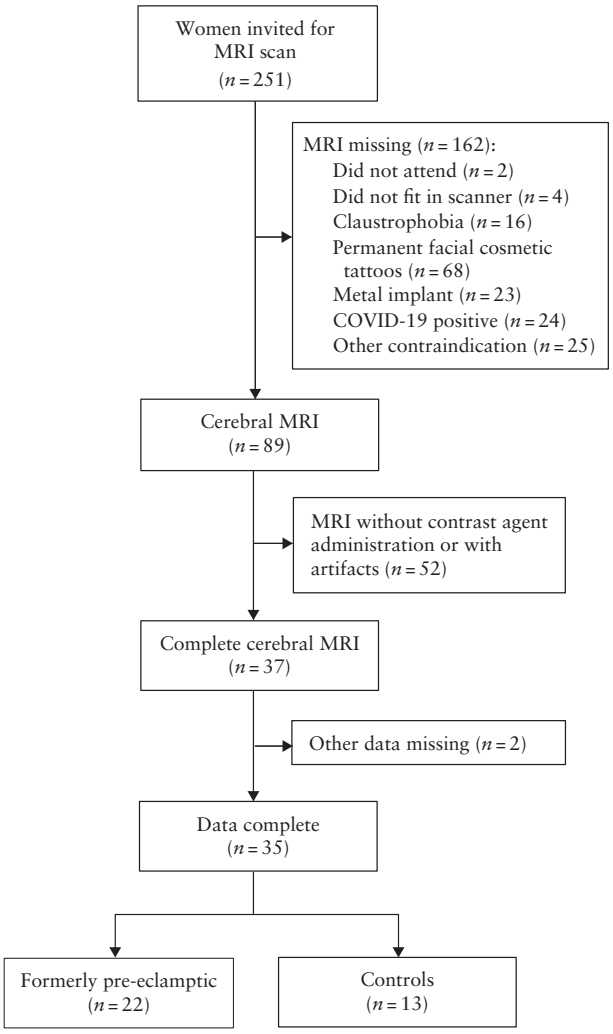

Figure 1 Flowchart showing inclusion in study population of formerly pre-eclamptic women and controls with a history of hypertensive pregnancy. COVID-19, coronavirus disease 2019; MRI, magnetic resonance imaging.

Table 1 Demographic and clinical characteristics of women who formerly had pre-eclampsia (PE) and women with a history of normotensive pregnancy (controls)

Characteristic	*Formerly pre-eclamptic* (n = *22*)	*Controls* (n = *13*)	P
Demographic			
Age (years)	37.8 ± 5.4	40.8 ± 5.5	0.13
Educational level			0.56
Low	2 (9)	0 (0)	
Average	8 (36)	6 (46)	
High	12 (55)	7 (54)	
Index pregnancy			
Early-onset PE (< 34 weeks)	13 (59)	NA	
HELLP syndrome	18 (82)	NA	
Eclampsia	2 (9)	NA	
Preterm delivery	17 (77)	0 (0)	0.001
Time since index pregnancy (years)	6.6 ± 3.2	9.0 ± 3.7	0.05
Subsequent pregnancy	9 (41)	10 (77)	0.02
Early-onset PE (< 34 weeks)	0 (0)	NA	
HELLP syndrome	1 (5)	NA	
Eclampsia	0 (0)	NA	
Lifestyle			
Smoker	9 (41)	5 (38)	0.75
Alcohol consumer	14 (64)	9 (69)	0.28
Vascular risk profile			
Weight (kg)	72 ± 11	64 ± 8	0.03
Height (cm)	169 ± 5	167 ± 6	0.24
BMI (kg/m^2)	25.1 ± 4.0	22.9 ± 2.8	0.08
BSA (m^2)	1.81 ± 0.1	1.71 ± 0.1	0.03
Hypertension	6 (27)	1 (8)	0.19
SBP (mmHg)	118 ± 12	108 ± 8	0.01
DBP (mmHg)	74 ± 9	68 ± 5	0.01
MAP (mmHg)	91 ± 10	83 ± 5	0.005
Heart rate (bpm)	66 ± 10	67 ± 11	0.96
Total cholesterol (mmol/L)	4.5 ± 1.0	4.3 ± 0.7	0.42
Glomerular filtration rate (mmol/L/1.73 m^2)	86 ± 12	82 ± 11	0.33
Fazekas score			0.60
0	14 (64)	10 (77)	
1	8 (36)	3 (23)	
2	0 (0)	0 (0)	
3	0 (0)	0 (0)	

Data are presented as mean ± SD or *n* (%). Index pregnancy defined as first pregnancy in controls and first PE pregnancy in PE group. BMI, body mass index; BSA, body surface area; DBP, diastolic blood pressure; MAP, mean arterial pressure; NA, not applicable; SBP, systolic blood pressure.

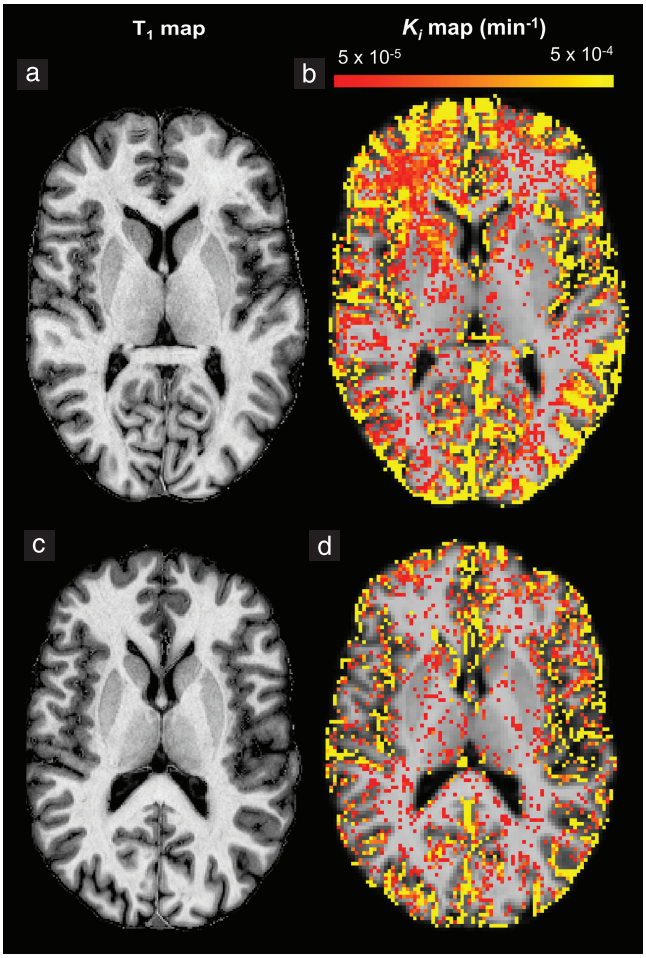

Figure 2 T1-weighted images (a,c) and leakage rate (K_i) maps (b,d) in a 39-year-old formerly pre-eclamptic woman (a,b) and a 39-year-old control woman (c,d). Note the higher K_i values (yellow) and higher number of pixels showing any blood–brain barrier leakage, particularly in the frontal lobes, in the formerly pre-eclamptic woman.

for the global GM in formerly pre-eclamptic women compared with controls. While a significantly higher K_i was observed in the cortex of formerly pre-eclamptic women ($P = 0.02$), K_i was not significantly different in the deep GM. Additionally, formerly pre-eclamptic women had a significantly higher K_i in the frontal ($P = 0.04$) and parietal ($P = 0.009$) GM and in the frontal ($P = 0.001$), temporal ($P < 0.05$) and occipital ($P = 0.007$) WM (Figure 3). The odds of a high K_i in formerly pre-eclamptic women, when compared with control women, were generally higher in the WM *vs* GM of different brain lobes. In the same regions, formerly pre-eclamptic women also had a significantly higher v_L than did the control group (Table S3). No significant differences between groups were found in v_p (Table S4). A similar trend in BBB leakage differences was found after adjusting for age, hypertension at MRI and Fazekas score (Tables 2, S3 and S4).

DISCUSSION

BBB leakage was more pronounced throughout the whole cerebrum in formerly pre-eclamptic women than in parous control women years after the index pregnancy. Higher K_i and v_L were especially prominent in the frontal and parietal GM, and in the frontal, temporal and occipital WM.

Although in one study BBB permeability was shown to be increased in pregnant and non-pregnant rats in response to acute hypertension, this difference was not statistically significant[6]. Other studies using pre-eclampsia models showed increased BBB permeability to small solutes, such as sodium fluorescein (0.47 kDa) and Evans blue (0.96 kDa)[8,12]. Increased BBB permeability during gestation was also observed in humans, but the results varied depending on the method of assessment employed[36–38]. BBB leakage of smaller molecules, such

Table 2 Blood–brain barrier leakage rates in women who formerly had pre-eclampsia (PE) and in women with a history of normotensive pregnancy (controls)

	Leakage rate ($\times 10^{-7}$/min)					
Brain region	*Women with PE*	*Controls*	*Difference*	P	*OR (95% CI)*	*aOR (95% CI)*
Whole cerebrum						
White matter	3.6 ± 3.4	0.5 ± 0.9	3.1	0.001	37.4 (3.8–364.6)	48.3 (3.5–651.2)
Gray matter	13.6 ± 2.7	6.0 ± 5.1	7.6	0.02	3.8 (1.1–35.9)	3.5 (1.1–30.8)
Gray matter						
Deep	3.9 ± 3.7	2.4 ± 3.6	1.5	0.26	1.1 (0.1–13.5)	0.7 (0.1–11.0)
Cortex	14.8 ± 14.2	7.1 ± 5.2	7.7	0.02	4.2 (1.5–12.4)	3.9 (1.1–13.2)
Lobar gray matter						
Frontal	18.1 ± 16.5	7.7 ± 8.2	10.4	0.04	3.4 (1.2–32.6)	2.8 (1.0–28.7)
Temporal	10.4 ± 1.4	5.2 ± 7.0	5.2	0.06	1.6 (0.1–17.2)	1.0 (0.1–13.1)
Parietal	19.9 ± 17.4	6.8 ± 6.5	13.1	0.009	6.9 (1.7–64.0)	6.6 (1.1–64.4)
Occipital	14.7 ± 13.5	7.9 ± 7.8	6.8	0.12	0.8 (0.1–22.1)	0.9 (0.0–11.6)
Lobar white matter						
Frontal	5.6 ± 5.8	0.9 ± 1.2	4.7	0.001	10.0 (1.1–92.0)	10.3 (1.2–102.3)
Temporal	4.3 ± 7.2	0.8 ± 1.2	3.5	< 0.05	5.1 (1.1–48.0)	4.1 (1.4–41.0)
Parietal	3.3 ± 4.1	1.5 ± 1.9	1.8	0.09	2.4 (0.2–24.8)	1.8 (0.1–22.1)
Occipital	3.4 ± 4.2	0.5 ± 0.6	2.9	0.007	25 (1.2–95.0)	25.8 (1.2–98.5)

Data are given as mean ± SD, unless indicated otherwise. Adjusted odds ratios (aOR) were obtained by correcting for age, hypertension status at magnetic resonance imaging and Fazekas score.

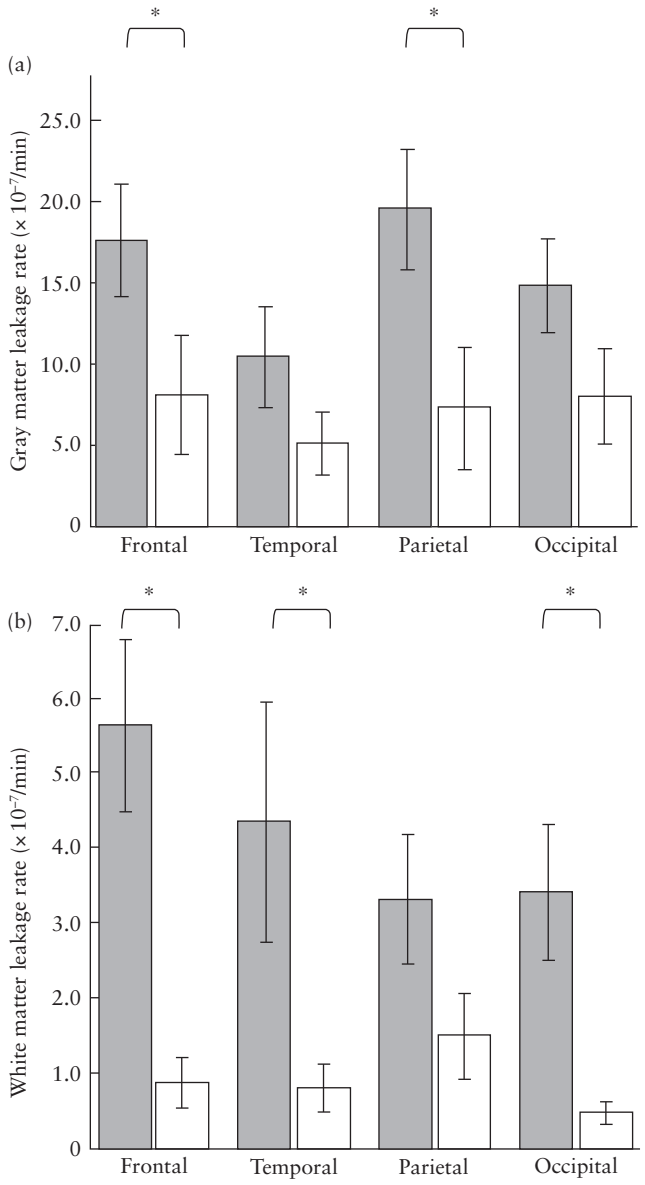

Figure 3 Average leakage rate in gray matter (a) and white matter (b), according to brain region, in formerly pre-eclamptic women (■) and in controls with a history of normotensive pregnancy (□). Error bars indicate SD. *Regions with a significantly different leakage rate between groups.

as gadobutrol (0.60 kDa), may be more likely when compared with that of larger biomolecules, such as albumin (67 kDa)[39,40]. A case report of a 27-year-old woman with pre-eclampsia and posterior reversible encephalopathy syndrome (PRES), described in the case series by Schwartz *et al.*[41,42], demonstrated BBB breakdown in the occipital lobe and parieto-occipital junction using a contrast agent. In the present study, we show greater long-term BBB leakage in women with a history of pre-eclampsia than that observed in controls with a history of normotensive pregnancy. However, our findings did not match fully the features of PRES, which are often present in the parieto-occipital region[43], that were seen in the pregnancy complicated by pre-eclampsia in studies of Schwartz *et al.*[41,42]. This could be because we investigated the long-term outcome of pre-eclampsia instead of the changes in the acute phase. The relationship between the acute and long-term cerebrovascular outcomes of pre-eclampsia needs to be investigated in the future.

The increased BBB permeability following pre-eclampsia could result from temporary elevated blood pressure, as has been suggested by the increase in circulating cells and activation of the endothelium in hypertensive patients[44]. The hypertensive state during a pre-eclamptic pregnancy could be the cause of BBB leakage. The increased BBB permeability could be related to the breakdown of tight junctions, but circulating factors may also be involved. Although the effect of chronic hypertension on the condition of the BBB is currently unknown, accumulating evidence from animal models suggests that chronic hypertension is accompanied by BBB dysfunction[45–47]. Even though the correction for hypertension status at MRI in our statistical analysis did not significantly alter the results, because of the small number of women with hypertension at the time of MRI ($n = 6$), we cannot exclude the possibility that long-term hypertension may contribute to BBB impairment.

WMH have been reported in formerly pre-eclamptic women up to 5 years after the problematic pregnancy[25–27]. While some studies have suggested a significant difference in terms of WMH between formerly pre-eclamptic and control women[26,27], other research did not[25]. Wiegman *et al.*[25] observed WMH in 36.5% of formerly pre-eclamptic women 5 years after the pregnancy and in 21.3% of control women. Although WMH was more common in pre-eclamptic women, the difference was not statistically significant. We observed mild WMH (Fazekas score 1) in 36% of cases 6.6 years after the index pregnancy and in 23% of control women 9.0 years after the index pregnancy, similar to the results of Wiegman *et al.*[25].

Similarly to our study, Siepmann *et al.*[48] did not observe a significant difference in the Fazekas score but observed an increase in WMH volume in women with a history of pre-eclampsia compared with controls. However, the volume of these WMH was very low (~0.11 cm^3) when compared with the hundred times larger volume observed in patients with cSVD[49]. Regardless of pregnancy history, WMH have been linked to BBB disruption and diffuse cerebrovascular endothelial failure[50]. Although we did not observe significantly more WMH in our study, we found an association between greater BBB leakage and pre-eclampsia, suggesting widespread endothelial dysfunction. The 10 times higher non-adjusted odds ratio for a high BBB leakage after pre-eclampsia in the global WM compared with the global GM indicates vulnerability of the generally less vascularized WM[51,52]. BBB leakage in WM could be an early marker of WMH and may indicate an increased risk of cerebrovascular disorders, as Hase *et al.*[53] found evidence of widespread microvascular pathology in neurodegenerative disorders and dementia caused by vascular disease. They observed increased capillary width in the deep WM, suggesting that chronic hypoperfusion induces

microvascular modification in the deep WM, which may affect perfusion and result in greater BBB leakage.

A limitation of this study is the relatively small sample size of the formerly pre-eclamptic group ($n = 22$) compared with the full study sample ($n = 1065$). The study characteristics of the formerly pre-eclamptic women with and those without a MRI scan were not significantly different, which indicates that this sample is representative of the population.

Whether the higher BBB leakage observed is causally related to the impact of pre-eclampsia on the brain or to the risk profile in which pre-eclampsia is a vascular stress test cannot be concluded, as other characteristics, such as hypertension and other cardiovascular risk factors, may underlie BBB disruption and pre-eclampsia. Nevertheless, it is likely that BBB impairment may be involved in (initiating) the pathophysiology and affect brain health years after pre-eclampsia, as correction for age, hypertension at the time of MRI and Fazekas score did not have a vast impact on the results, indicating that these factors do not underlie the observed differences.

Pre-eclampsia with severe features, such as eclampsia and HELLP syndrome, has been associated with poorer cognitive performance[54]. These severe features may be associated with greater BBB leakage. However, this should be investigated in the future, as this study aimed to assess the presence and extent of BBB leakage, and its statistical power was too low to assess the impact of pregnancy characteristics.

In conclusion, this study has shown greater BBB leakage in a sample of patients with a history of pre-eclampsia years after the index pregnancy compared with controls. The increased BBB permeability is subtle but may indicate diffusively spread endothelial dysfunction as a pathophysiological mechanism underlying cerebral injury associated with pre-eclampsia. Women with a history of pre-eclampsia may benefit from treatment and prevention strategies aimed at reducing the risk of cerebrovascular disease, such as prolonged neuroprotective treatment, antihypertensive treatment after delivery or lifestyle modifications, such as a healthier diet and physical exercise. Future studies should determine the optimal management strategy for this population.

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SUPPORTING INFORMATION ON THE INTERNET

The following supporting information may be found in the online version of this article:

Appendix S1 Details of magnetic resonance imaging

Appendix S2 Histogram approach for noise correction

Table S1 Pulse sequence parameters

Table S2 Demographic and clinical characteristics of formerly pre-eclamptic women with and without a cerebral MRI scan participating in the ongoing Queen of Hearts study

Table S3 Fractional leakage volume (v_L) in women with a history of pre-eclampsia and control women with a history of normal pregnancy

Table S4 Fractional blood plasma volume (v_p) in women with a history of pre-eclampsia and control women with a history of normal pregnancy

Figure S1 Averaged histograms of the blood–brain barrier leakage rate (K_i) values in the gray and white matter (A1 and B1, respectively) and the histograms corrected for noise by subtraction of the mirrored negative values in the gray and white matter (A2 and B2, respectively). Note the differences in the axis scales in fraction of leaking tissue. The histograms for the formerly pre-eclamptic women show slightly higher bins for higher K_i values (A). After noise correction (B), a larger fraction of voxels with detectable leakage is seen in formerly pre-eclamptic women.