PAPER

Inherited thrombophilia and stratification of ischaemic stroke risk among users of oral contraceptives

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Background: Whether use of oral contraceptives is a risk factor for arterial ischaemic stroke is controversial. In particular, few data are available on what criteria should be adopted to establish an individual profile of risk before the start of oral contraceptives.

Patients and methods: The effects of oral contraceptives and their interaction with the G1691A polymorphisms of the factor V gene, the G20210A polymorphisms of the prothrombin gene and the C677T polymorphisms of the *MTHFR* gene on the risk of cerebral ischaemia were determined in a series of 108 consecutive women aged <45 years with ischaemic stroke and 216 controls, in a hospital-based case–control study design.

Results: Use of oral contraceptives was associated with an increased risk of cerebral ischaemia (odds ratio (OR) 3.95; 95% confidence interval (CI) 2.29 to 6.78). ORs for stroke were 2.25 (95% CI 1.15 to 4.40), 3.94 (95% CI 2.28 to 6.81) and 8.87 (95% CI 3.72 to 21.1) for non-oral contraceptive users with the *TT MTHFR* genotype, oral contraceptive users without the *TT MTHFR* genotype and oral contraceptive users with the *TT MTHFR* genotype, respectively, when compared with non-oral contraceptive users without the *TT MTHFR* genotype, with a multiplicative independent effect. Compared with non-oral contraceptive users, ORs for stroke were 2.65 (95% CI 1.46 to 4.81) for oral contraceptive users with none of the studied polymorphisms and 22.8 (95% CI 4.46 to 116.00) for oral contraceptive users with at least one of the studied polymorphisms, with a synergistic effect.

Conclusions: Exposure to the effects of oral contraceptives may increase the risk of ischaemic stroke in women with an inherited prothrombotic background. Testing for these genetic variants may allow more accurate stratification of the population at risk before long-term use of oral contraceptives is prescribed.

The cardiovascular safety of widely used oral contraceptives is still debated. The risks of venous thromboembolism have been well established,[1 2] but there is controversy over whether oral contraceptives may also increase the risk of vascular arterial events. In particular, many studies have attempted to assess the risk of stroke related to use of oral contraceptives, but findings have been somewhat contradictory. Methodological differences among studies, the potential influence of confounding factors, the use of different oral contraceptive formulations and their changes over time mean the results of these analyses are not strictly comparable, making it difficult to draw definitive conclusions.[3–5] Despite these differences, evidence suggests that a relationship between oral contraceptives and stroke exists. However, although this association is fairly robust across studies and meta-analyses,[6] the crude risk seems low, with only an additional 4.1 ischaemic strokes per 100 000 non-smoking, normotensive women using low-dose oestrogen oral contraceptives.[4] Epidemiological studies have shown that the risk of cardiovascular events associated with oral contraceptives increases considerably in women with major vascular risk factors such as arterial hypertension or smoking. Accordingly, these conditions are now considered contraindications to the use of oral contraceptives, particularly after age 35 years.[7 8] However, an issue that has received little attention is whether other conditions that are not currently taken into consideration by doctors when prescribing oral contraceptives might contribute to further increase the risk of arterial ischaemic diseases, because of an independent (additive or multiplicative) or an interacting (partial or complete) effect with these drugs.

Recently, in line with the results of previous reports showing the effect of a synergistic interaction between inherited thrombophilic defects and oral contraceptives on the risk of venous thrombosis,[9] three independent studies suggested that the risk of arterial ischaemic stroke related to oral contraceptives might be higher in women with such a predisposing genetic background,[10–12] prompting speculation that even a small degree of increased risk for stroke associated with oral contraceptives in the general population might be greatly amplified in such women and be clinically relevant. When confirmed, this might allow a more accurate stratification of the population at risk before oral contraceptives are prescribed.

Owing to the potential implications of screening for these genetic markers, we undertook the current case–control study of young women aged 18–45 years to investigate the contribution of oral contraceptive use and inherited thrombophilic defects to the occurrence of ischaemic stroke, by analysing their independent and interactive effects.

SUBJECTS AND METHODS

Data were obtained in the setting of a single-centre, hospital-based study, designed at the Department of Neurology, University of Brescia, Brescia, Italy, for the evaluation of gene–environment interactions in the development of ischaemic cerebrovascular disease. A detailed description of the methods and the standardised protocol for patients' documentation has been given elsewhere.[13 14] For the purpose of this study, unselected, unrelated women from a series of consecutive patients admitted to our department with first-ever acute ischaemic stroke occurring under the age of 45 years were enrolled. After completion of the diagnostic work-up, cases

Abbreviations: AIC, Akaike's information criterion; BIC, Bayesian information criterion

Table 1 Demographic and clinical characteristics of patients with stroke and controls

	Patients with stroke (n = 108)	Controls (n = 216)		
Characteristic	**n (%)**	**n (%)**	**Crude OR**	**95% CI**
Current smokers	44 (40.8)	27 (12.5)	4.89	2.79 to 8.54
Hypertension	13 (12.0)	9 (4.2)	3.14	1.30 to 7.62
Diabetes mellitus	0 (0)	5 (2.3)	0.18	0.01 to 3.23
Hypercholesterolaemia	27 (25.0)	23 (10.7)	2.99	1.61 to 5.54
Use of oral contraceptives	43 (39.8)	31 (14.4)	3.95	2.29 to 6.78
	Mean ± SD	Mean ± SD	Crude MR	
Age (years)	33.9 (7.4)	34.6 (6.6)	−0.70	–

were subtyped into four major aetiological categories, according to a classification based on the Trial of ORG 10172 in Acute Stroke Treatment criteria, accommodated and validated for the aetiology of stroke in the young[15]:

1. Atherosclerotic vasculopathy: cerebral infarction caused by large-vessel atherosclerotic vasculopathy or small-vessel disease
2. Non-atherosclerotic vasculopathy
3. Cardiac/transcardiac embolism: also including cases with a cardioembolic source combined with a proven thrombophilic disorder
4. Other: cerebral infarction that did not meet the criteria for one of the categories outlined above.

Women from the staff of our hospital with no known history of vascular disease and matched to the cases by age in 3-year bands served as controls.[13] Both cases and controls were white and were from the same geographical area.

Assuming a gene–environment interaction model with a dominant mode of inheritance, significance level equal to 0.05 (two-sided), disease prevalence equal to the prevalence of stroke in the Italian population aged 0–44 years (65/100 000),[16] frequency of oral contraceptive use equal to 0.174,[17] and prevalence of the studied prothrombotic variants equal to 0.262 in a representative population of northern Italy,[18] we estimated that a sample of approximately 100 cases and 200 controls was necessary to meet the commonly accepted standard value of 80% for sufficient power, when the ORs of the genotype–environment interaction effect of genetic modelling are >5.[19]

Demographic data and history of hypertension, diabetes mellitus, cigarette smoking and hypercholesterolaemia were obtained from each subject. Hypertension was considered present if systolic blood pressure was >160 mm Hg and diastolic pressure >95 mm Hg in two separate measurements after the acute phase, or if the subject was under treatment with antihypertensive drugs before recruitment. The diagnosis of diabetes mellitus was established according to WHO criteria.[20] Cigarette smokers were categorised as current smokers or non-smokers (non-smokers included former smokers who had quit smoking for at least 6 months before the study). Hypercholesterolaemia was considered present if cholesterol serum concentrations were >220 mg/dl or if the subject was under treatment with cholesterol-lowering drugs. A detailed history of oral contraceptive use was recorded and the subjects categorised as users or non-users (non-users included former users who had stopped taking these drugs at least 1 month before the index event).

The study was designed and carried out in accordance with the ethical principles established by the local institutional guidelines on clinical investigation. Written, informed consent was provided by all study participants.

Genetic analyses

Venous blood sampling for biochemical determinations was performed in the early morning (before 07.00 h) after overnight fasting in all subjects. In patients, blood samples were obtained 7–10 days after the acute event. Genomic DNA was isolated in all subjects from −20°C frozen samples of EDTA anticoagulated whole blood using standard DNA extraction. The G1691A mutation in the factor V gene (factor V Leiden) and the G20210A mutation in the prothrombin gene were determined according to a standardised multiplex polymerase chain reaction method.[21] The C677T *MTHFR* genotypes were determined according to the method of Frosst *et al*,[22] using polymerase chain reaction amplification and restriction digestion with *Hinf*I to distinguish mutant from wild-type allele.

Statistical analysis

Bivariate mean differences (MD), odds ratio (OR) and 95% confidence intervals (CI) were estimated for conventional risk factors and polymorphisms. A binary variable was determined for each polymorphism (''yes'' or ''no'' based on the status of ''carrier'' or ''non-carrier'' of the FV 1691A allele, the PT 20210A allele and the *TT*677 *MTHFR* genotype). On the basis of this definition, the entire study group was stratified into two subcategories corresponding to subjects carrying at least one such genetic marker (prothrombotic, P) and subjects carrying none (non-prothrombotic, non-P), respectively.

The 4×2 table approach[23] was used to estimate the effect of interaction between genetic thrombophilic background and the use of oral contraceptives on the risk of ischaemic stroke. In particular, the interactive effect of the *TT*677 *MTHFR* genotype with oral contraceptives, and that of the inherited prothrombotic background (defined as the presence of at least one genetic thrombophilic defect) with oral contraceptives were tested.

Five genotype–environment interaction models were fitted using logistic regression: (1) genotype–environment complete interaction; (2) genotype as moderator of the environment effect; (3) environment as moderator of the genotype effect; (4) genotype–environment partial interaction; and (5) genotype and environment independent effect.[24] The referent categories were the following: (a) non-*TT MTHFR* genotype and non-oral contraceptive users for models 1, 4 and 5; (b) overall non-oral contraceptive users for model 2; and (c) overall non-*TT MTHFR* genotype for model 3. The modelling strategies included assessment of interaction models without and with adjustment for covariates (age, smoking habit, hypertension and hypercholesterolaemia). Diabetes mellitus was not entered into the final analysis as covariate owing to the low frequency of this

Table 2 Characteristics of the study group according to genotype distribution

Genotype distribution	Patients with stroke (n = 108)	Controls (n = 216)	Crude OR	95% CI
Methylenetetrahydrofolate reductase *C677T*				
CC	25 (23.1)	84 (38.9)	1	
CT	61 (56.5)	110 (50.9)	1.86	1.00 to 3.21
TT	22 (20.4)	22 (10.2)	3.36	1.60 to 7.05
FV G1691A				
GG	103 (95.3)	210 (97.2)	1	
GA	5 (4.7)	6 (2.8)	1.70	0.51 to 5.70
AA	0 (0.0)	0 (0.0)	–	
PT G20210A				
GG	98 (90.7)	213 (98.6)	1	
GA	9 (8.4)	3 (1.4)	6.52	1.73 to 24.6
AA	1 (0.9)	0 (0.0)	–	
FV 1691A or PT 20210A or *TT MTHFR*				
No	75 (69.4)	185 (85.6)	1	
Yes	33 (30.6)	31 (14.4)	2.63	1.50 to 4.59

condition in the present series. To compare such competing models, Akaike's information criterion (AIC = −2×model log-likelihood+2×number of model parameters) and the Bayesian information criterion (BIC = −2×model log-likelihood+log(n)×number of model parameters) were computed. The selected model was the one minimising either AIC or BIC.[25] Because assessment of interaction by logistic regression models was performed assuming multiplicativity of the effects, Rothman's synergy (S) measure[26] was also computed using the maximum likelihood estimates ORs of the selected model. The S index is the ratio of the observed effect with joint exposure to the effect predicted for joint exposure assuming additivity of the effects. No interaction corresponds to S = 1, whereas S>1 (S<1) can be interpreted as measure of relative increase (decrease) in the additive effect among those exposed to both factors.

Pooled ORs (95% CIs) from our data and those tabulated by Slooter *et al*[11] (for the *TT*677 *MTHFR* genotype–oral contraceptive use interaction) and by Aznar *et al*[10] (for the FV 1691A allele/PT 20210A allele–oral contraceptive use interaction) were also calculated using the same modelling approach. As frequencies of oral contraceptive users who were carriers of prothrombotic genotypes/alleles were not reported by Martinelli *et al*,[12] data from this study were not included in the pooled analysis. Data analyses were conducted with SPSS V.11.1 software.

RESULTS

Characteristics of the study group

As DNA could not be amplified in one subject, data from 108 cases were entered into the final analysis. Thus, 216 controls were recruited for a 1:2 case–control design. Table 1 presents the demographic characteristics and prevalence of selected risk factors of the participants. A cardioembolic aetiology was presumed in 36 (33.3%) patients, non-atherosclerotic vasculopathy in 20 (18.5%), atherosclerotic vasculopathy in 11 (10.2%), and 41 patients (38.0%) met the criteria for other aetiologic categories. As expected, patients with stroke were more often smokers, those with hypertension and those with hypercholesterolaemia as compared with controls. The use of oral contraceptives was associated with a more than threefold increased risk of cerebral ischaemia (OR 3.95; 95% CI 2.29 to 6.78).

Distribution of the prothrombotic genotypes

The observed distribution of the genotypes closely resembled those reported previously in other series from northern Italy.[18 27] An increase in stroke risk was associated with the *TT*677 *MTHFR* genotype when compared with the *CC* genotype (OR 3.36; 95% CI 1.60 to 7.05) and with the PT 20210A gene variant when compared with the PT 20210G gene variant (OR 6.52; 95% CI 1.73 to 24.6). By contrast, the frequency of the FV 1691A gene variant was not different in the two groups (table 2). The prevalence of subjects carrying at least one procoagulant genotype was higher in the group of patients than in the group of controls (30.6% *v* 14.4%; OR 2.63; 95% CI 1.50 to 4.59).

Interaction analysis

Tables 3 and 4 summarise the results of the analyses undertaken to find the effect of potential interactions of both the *TT*677 *MTHFR* genotype and the overall inherited prothrombotic background with the use of oral contraceptives on the risk of ischaemic stroke. Based on the AIC or BIC indices (not shown), the following two models were selected: the ''*TT MTHFR* genotype and oral contraceptive use independent effect'' model (table 3) and the ''inherited prothrombotic background as moderator of oral contraceptive use effect'' model (table 4).

Non-oral contraceptive users carrying the *TT MTHFR* genotype had an increased risk of stroke in comparison with non-oral contraceptive users without this genotype (OR 2.25; 95% CI 1.15 to 4.40). A similar effect was determined by oral contraceptive use alone (OR 3.94; 95% CI 2.28 to 6.81), whereas the combination of the *TT MTHFR* genotype and oral contraceptive use was associated with about a ninefold greater odds of ischaemic stroke (OR 8.87; 95% CI 3.72 to 21.1), as predicted by the multiplicativity of the genotype–environment effect (2.25×3.94 = 8.87). The combined effect of the *TT MTHFR* genotype and oral contraceptive use on stroke risk was about 90% greater than that predicted by assuming additivity of effects (S = 1.88). Adjustment for other covariates did not significantly change the results (table 3).

A synergistic effect was observed when the presence/absence of an inherited prothrombotic background (P/non-P) was considered (table 4). Compared with the ''corner-point'' reference category of

Table 3 4×2 table of *TT MTHFR* × oral contraceptive use × disease status, and maximum likelihood estimate odds ratios (95% confidence interval) of the model ''*TT MTHFR* and oral contraceptives use independent effects on disease status''

	Patients with stroke (n, %)	Controls (n, %)	Unadjusted OR	Unadjusted 95% CI	Unadjusted p Value	Adjusted* OR	Adjusted* 95% CI	Adjusted* p Value
Non-oral contraceptive users and non-TT	53 (24.3)	165 (75.7)	1	—		1		
Non-oral contraceptive users and TT	12 (37.5)	20 (62.5)	2.25	1.15 to 4.40	0.018	3.02	1.43 to 6.41	0.004
Oral contraceptive users and non-TT	33 (53.2)	29 (46.8)	3.94	2.28 to 6.81	<0.001	3.89	2.12 to 7.15	<0.001
Oral contraceptive users and TT	10 (83.3)	2 (16.7)	8.87	3.72 to 21.1	<0.001	11.8	4.46 to 31.0	<0.001

Synergy index = (8.87−1)/((2.25−1)+(3.94−1)) = 1.88.
Adjusted synergy index = (11.8−1)/((3.02−1)+(3.89−1)) = 2.20.
p Values of the square root of Wald test with df = 1 (two-tailed).
*Covariates: age, smoking, hypertension and hypercholesterolaemia.

Table 4 4×2 table of genetic background x oral contraceptive use × disease status, and maximum likelihood estimate odds ratios (95% confidence interval) of the model ''genetic thrombophilic background as moderator of oral contraceptives use effect on disease status''

	Patients with stroke (n, %)	Controls (n, %)	Unadjusted			Adjusted*		
			OR	95% CI	p Value	OR	95% CI	p Value
Non-oral contraceptive users and non-P	48 (23.5)	156 (76.5)	1	–		1		
Non-oral contraceptive users and P	17 (37.0)	29 (63.0)	1			1		
Oral contraceptive users and non-P	27 (48.2)	29 (51.8)	2.65	1.46 to 4.81	0.001	2.57	1.33 to 4.98	0.005
Oral contraceptive users and P	16 (88.9)	2 (11.1)	22.8	4.46 to 116	0.007	23	4.19 to 125	0.008

P, prothrombotic.
Synergy index = (22.8−1)/(2.65−1) = 13.2.
Adjusted synergy index = (23.0−1)/(2.57−1) = 14.0.
p Values of the square root of Wald test with df = 1 (two-tailed).
*Covariates: age, smoking, hypertension and hypercholesterolaemia.

non-oral contraceptive users (both with or without an inherited prothrombotic background), current use of oral contraceptives resulted in about threefold increased risk of stroke in subjects with no predisposing thrombophilic background (OR 2.65; 95% CI 1.46 to 4.81), and in about 23-fold increase in the disease risk (OR 22.8; 95% CI 4.46 to 116) when in combination with at least one thrombophilic genotype (table 4). Such a combined effect was about 13-fold greater than that predicted by assuming additivity of effects (S = 13.2). Results remained substantially unchanged after adjustment for covariates.

Pooled data analysis

The model selection based on the AIC/BIC indices of the pooled data from our study and that of Slooter *et al*[11] confirmed the multiplicative independent effect of the *TT*677 *MTHFR* genotype and oral contraceptive use in increasing the risk of stroke (OR 2.67×1.65 = 4.41; 95% CI 2.85 to 6.84; S = 1.47). The same effect resulted from the combination of the FV 1691A allele/PT 20210A allele and oral contraceptive use according to the pooled data from the present study and that of Aznar *et al*[10] (OR 3.49×3.81 = 13.3; 95% CI 5.17 to 34.2; S = 2.32).

DISCUSSION

Although oral contraceptive use is generally perceived as a predisposing condition for stroke in young women, the absolute risk seems to be small and even less relevant when the non-negligible rate of maternal mortality during pregnancy is taken into account.[28] As a general rule, premenopausal women can

Table 5 Comparison of study design and characteristics of the four studies investigating the thrombophilia–oral contraceptives–ischaemic stroke hypothesis

	Aznar *et al*[10]	Slooter *et al*[11]	Martinelli *et al*[12]	This study
Study design	Case–control analysis	Case–control analysis	Case–control analysis	Case–control analysis
No of patients/controls	34/68	193/767	105/293	108/216
Patients				
Age, years, mean (SD)	18–50	18–49, 38.6 (8.0)	Fertile age, 34.7 (9.1)	18–45, 33.9 (7.4)
Diagnosis	Ischaemic stroke	First ever ischemic stroke	First ever ischaemic stroke	First ever ischaemic stroke
Inclusion criteria	Consecutive patients with cryptogenic stroke	Consecutive patients with acute stroke	Consecutive patients with history of stroke	Consecutive patients with acute stroke
Exclusion criteria	Atherosclerosis, heart disease, foramen ovale, occlusive vessel disease	TIA, cerebral venous sinus thrombosis, carotid artery dissection, history of cardiovascular or cerebrovascular disease, terminal illness, aphasia or cognitive impairment interfering with the questionnaire, not speaking Dutch		
Controls				
Age, years, mean (SD)	<50	18–49, 39.7 (7.7)	Fertile age, 34.9 (8.6)	34.6 (6.6)
Selection criteria	Not defined	From the general population by random digit dialling	From partners and friends who accompanied patients to the centre	From the hospital staff
Inclusion criteria	Age-matched healthy subjects	Age-matched subjects, no history of coronary heart disease, cerebrovascular event or peripheral vascular disease	Healthy subjects of fertile age	Age-matched subjects (±3 years), no known history of vascular disease
Setting	Unit of thrombophilia	Stroke centre	Thrombosis centre	Stroke centre
OC users	Not defined	Within 1 month before the acute event	Within the 2 weeks before referral	Within 1 month before the acute event
Thrombophilic tests	FV G1691A PT G20210A	FV G1691A PT G20210A C677T MTHFR	FV G1691A PT G20210A C677T MTHFR Hyperhomocysteinaemia AT, PC, PS deficiency	FV G1691A PT G20210A C677T MTHFR

AT, anti-thrombin; OC, oral contraceptive; PC, protein C; PS, protein S; TIA, transient ischaemic attack.

safely be prescribed oral contraceptives, provided they have no increased cardiovascular risk at baseline. However, apart from major cardiovascular risk factors such as hypertension and cigarette smoking, which are now established contraindications to the use of oral contraceptives,[7 8] few data are available on the influence that other conditions might have in increasing the risk of stroke when combined with these drugs. The identification of these conditions has implications for public health, given the large number of women currently using oral contraceptives worldwide, and, at an individual level, would allow ''at-risk'' subjects to be stratified and an individual profile of stroke risk to be established before any decision to start long-term use of oral contraceptives. The results of our study emphasise the possibility of better defining such an individualised risk. Although an overall increased risk of disease was associated with the use of these drugs, women carrying inherited thrombophilic defects seem to be exposed to a greater risk of stroke when under treatment with oral contraceptives in comparison to those without such a genetic predisposing background.

These findings are in line with previous observations made by Aznar *et al*[10] in a small subgroup of women with cryptogenic stroke, by Slooter *et al*[11] in a separate analysis of the Risk of Arterial Thrombosis in Relation to Oral Contraceptives study, and by Martinelli *et al*[12] in a recent case–control study, and further strengthen the assumption that a pre-existing prothrombotic state should be considered an additional risk factor when oral contraceptives are prescribed. In particular, according to our results, an interactive effect with genotype seems to be operant when prothrombotic variants are analysed in combination. Methodological differences in the study design of these reports are noteworthy (table 5). Firstly, a population-based setting, such as that of Slooter *et al*, has the theoretical advantage of reducing the risk of selection bias in comparison to a hospital-based setting, such as that of Aznar *et al*, of Martinelli *et al* and ours. The observation that the summary risk estimates are generally lower in studies using population-based controls than in studies using hospital-based controls also supports this assumption.[29] As Slooter pointed out, the fact that only a limited number of the eligible patients was included in their final analysis cannot exclude a survival bias. By contrast, as patients were both included and sampled during the acute phase of stroke, the possibility of biased observations due to a survival effect is unlikely in our study. Secondly, to obtain a homogeneous group in terms of clinical phenotype, Aznar *et al* focused on patients with stroke with no recognised cause of infarct, whereas Slooter *et al* applied specific predefined criteria for patient selection and Martinelli *et al* investigated a highly selected population of stroke survivors referred to their centre for thrombophilia screening. Unlike these authors, we decided to include in our analysis all women with cerebral infarct of arterial origin irrespective of stroke subtype, as we believe this may increase the external validity of our results. Finally, the sample size of the four studies is not strictly comparable. One rule in genetic epidemiology is that small initial studies (such as that of Aznar *et al*) are likely to overestimate the true effect size[30] and their findings would need to be replicated in other, ideally larger, samples. In this regard, Slooter's, Martinelli's and our own analyses served as confirmatory.

Overall, despite differences in the study design, there is substantial consistency among the results of these four reports, which supports the assumption that a combined effect of inherited thrombophilia and oral contraceptives on the risk of cerebral ischaemia is likely to exist.

In conclusion, if oral contraceptives are prescribed to women with an increased absolute risk at baseline, or for reasons other than contraception, the benefits might not justify the risks. Our findings suggest that the existence of an inherited prothrombotic state may increase such a baseline risk and may be associated with a higher rate of cerebral ischaemic events in women exposed to the effects of these drugs. The fact that a synergistic interaction with oral contraceptive use was found only when the thrombophilic genotypes were analysed in combination, as opposed to the independent multiplicative effect observed from the analysis of single polymorphisms in the present study, clearly indicates that studying an individual polymorphism in isolation would not uncover the overall predisposing influence of a hypercoagulable state. Although this study seems to be sufficiently powered in case of ORs of the gene–environment interaction effect >5, we do not dispute the fact that the large confidence intervals make the results of our interaction analysis statistically unstable. This puts further emphasis on the need for large national and international networks to overcome sampling limitations and come to conclusive evidence. A larger sample would also allow a more accurate characterisation of the study group with regard to other relevant variables such as the effect of the dose of oestrogen and the chemical composition of progestogen, or the effect of progestogen-only compounds on stroke risk.

When confirmed, our observations may provide the opportunity to identify subgroups of individuals in whom the exposure to oral contraceptives might significantly increase the incidence of ischaemic outcomes and prompt consideration of alternative therapeutic or contraceptive approaches.

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