Smoking Causes Fatal Subarachnoid Hemorrhage A Case-Control Study of Finnish Twins

Rautalin, Ilari

2020-10

Rautalin , I , Korja , M & Kaprio , J 2020 , ' Smoking Causes Fatal Subarachnoid Hemorrhage A Case-Control Study of Finnish Twins ' , Stroke , vol. 51 , no. 10 , pp. 3018-3022 . https://doi.org/10.1161/STROKEAHA.120.031231

http://hdl.handle.net/10138/328099
https://doi.org/10.1161/STROKEAHA.120.031231

acceptedVersion

Smoking causes fatal subarachnoid hemorrhage – a case-control study of Finnish twins

Ilari Rautalin, BM[1]; Miikka Korja, MD, PhD[1]; Jaakko Kaprio, MD, PhD[2];

[1]Department of Neurosurgery, University of Helsinki and Helsinki University Hospital, P.O. Box 266, FI-00029 Helsinki, Finland

[2]Institute for Molecular Medicine Finland FIMM, University of Helsinki, P.O. Box 20, FI-00014 Helsinki, Finland

Correspondence to Ilari Rautalin, Department of Neurosurgery, University of Helsinki, P.O. Box 266, FI-00029 Helsinki, Finland; E-mail address: ilari.rautalin@helsinki.fi; Telephone: +358 947187604; Fax: +358 947187616; ORCID 0000-0002-6283-0398

Title character count: 84

Tables: 2

Figures: 0

Number of references: 12

Word count manuscript: 3182

Word count abstract: 293

Cover title: Smoking, twins and fatal subarachnoid hemorrhage

Keywords: subarachnoid hemorrhage, familial, matched pair design, twin, smoking

Subject terms: Risk Factors; Cerebrovascular Disease/Stroke; Cerebral Aneurysm

Abstract

Background and Purpose. One of the largest twin studies to date suggested that subarachnoid haemorrhage (SAH) is mainly of non-genetic origin, but the causal effect of environmental factors on SAH is yet unknown. We hypothesized that if only one of the twins suffer from fatal SAH, they do not share the most important environmental risk factor for SAH, namely smoking. If true, such finding would suggest that smoking causes SAH.

Methods. Through the nationwide cause-of-death register, we followed 16,282 same-sex twin pairs of Finnish origin from the older Finnish Twin Cohort between 1976 and 2018, and identified all participants that died from SAH. For the baseline, we collected risk factor information about smoking, hypertension, physical activity, body mass index, alcohol consumption and education. We classified the pairs as monozygotic, dizygotic or of unknown zygosity. We examined the within-pair risk factor differences in the pairs discordant for SAH – that is, where one twin died from SAH and the other did not. We computed both individual (whole cohort) and pairwise (discordant pair) hazard ratios (HRs) and 95% confidence intervals (CIs).

Results. During the 869,469 person-years follow-up, we identified 116 discordant and 2 concordant (both died from SAH) twin pairs for fatal SAH. Overall, 25 of the discordant twin pairs were monozygotic. For the whole cohort, smoking (occasional/current) was associated with increased risk of SAH death (HR=3.33, CI 2.24–4.95) as compared to non-smokers (never/former). In the pairwise analyses for discordant twin pairs, we found that the twin who smoked had an increased risk of fatal SAH (HR=6.33, CI 1.87–21.4) as compared to the non-smoking twin. The association remained consistent regardless of the twin pairs' zygosity or sex.

Conclusions. Our results provide strong evidence for a causal – rather than associative – role of smoking in SAH.

Non-standard Abbreviations and Acronyms.

CI = Confidence interval

HR = Hazard ratio

SAH = Subarachnoid hemorrhage

Introduction

Twin cohorts are unparalleled in estimating the relative roles of genetic and environmental influence on a given phenotype. Therefore, large twin cohorts are of great value in studying familial clustering of any disease. In addition to genetic origin, familial clustering may also arise from environmental factors aggregating in families. In 2010, the results of a population-based Nordic Twin Cohort of 79,664 twin pairs from Denmark, Finland and Sweden suggested that the role of genetic factors is at most modest, indicating that familial clustering of subarachnoid haemorrhage (SAH) risk factors contributes more to the incidence of familial SAHs than susceptibility genes[1]. Although the Nordic Twin Study indicated that most twin pairs were discordant for SAH, i.e. only one twin died from SAH, the role of risk factors in explaining this discordance was not studied. To clarify why most often only one of the twins suffer from fatal SAH, we studied a large twin cohort consisting of 32,564 twin siblings. We hypothesized that smoking – the most important environmental risk factor for SAH[2, 3] – may associate with such discordance. If true, such finding would suggest that smoking causes SAH.

Materials and Methods

Data availability. The Finnish Twin Cohort data is not publicly available due to the restrictions of informed consent. However, the data is available through the Institute for Molecular Medicine Finland Data Access Committee (fimm-dac@helsinki.fi) for authorized researchers who have institutional review board/ethics approval and an institutionally approved study plan. To ensure the protection of privacy and compliance with national data protection legislation, a data use/transfer agreement is needed, the content and specific clauses of which will depend on the nature of the requested data.

Ethical consideration. The study cohort was established with approval from the National Board of Health; register linkages for our study received ethical approval from the ethical committee of the Department of Public Health, University of Helsinki. All participants provided their informed consent for participation. In addition, the study was conducted in line with the Declaration of Helsinki[4].

Study Subjects and Data Collection. Details of the study protocol are described elsewhere[5, 6]. Briefly, the older Finnish Twin Cohort was established in 1974, and it consisted of 16,282 same-sex twin pairs (total 32,564 individuals) alive in 1974 and born before 1958. Using the baseline surveys mailed for cohorts enrolled in 1975 and 1981, we collected data on smoking (never, occasional, former and current), hypertension (hypertension diagnosis or use of antihypertensive medications), leisure-time physical activity (metabolic equivalent of task scores divided into quintiles), body mass index (weight in kilograms divided by the square of height in meters, based on the World Health Organization's categories[7]), schooling (years of education divided into primary, secondary and tertiary educational attainment) and alcohol use (daily consumption categorized per ten grams of alcohol). We dichotomized the participants

into smokers (occasional and current) and non-smokers (never and former), and classified current smokers at baseline according to the number of cigarettes per day: light (<10), moderate (10–19) or heavy smokers (20 or more).

Follow-up and Case Identification. We obtained the follow-up mortality data through linking to the computerized nationwide cause-of-death registry, which is managed by Statistics Finland. Fatal SAHs were recorded during the follow-up time, which started in May 1976 and ended with death, migration out of Finland, or on December 31, 2018. In order to capture fatal SAHs, we used the International Classification of Diseases codes 430 (8^{th} and 9^{th} versions) and I60.0-I60.9 (10^{th} version) as the underlying cause of death. Data on non-fatal SAH cases were not available for this cohort. Six deaths due to SAH were excluded as they occurred prior to the start of the follow-up in 1974 and 1975.

Determination of Zygosity and Discordance. We determined zygosity by standardized and validated questionnaire methods known to classify more than 95% of twin pairs as monozygotic or dizygotic correctly[8]. If twin pairs did not reply or replied conflictingly to the questionnaires, or the zygosity of the pair was unknown, we classified the zygosity as uncertain. Twin pairs were classified as discordant for fatal SAH if only one twin died from SAH during the follow-up, regardless of whether the co-twin had died from any another cause or was still alive at the end of the follow-up. Twin pairs were concordant if both twins died from SAH.

Data Analysis. We estimated hazard ratios (HRs) with 95% confidence intervals (CIs) for fatal SAH by the studied risk factors in the entire cohort. For this analysis, we used the Cox proportional hazards model adjusted to the sampling of twins within pairs by using the cluster option in Stata. In addition, we adjusted the hazard models to sex. We then conducted pairwise

analyses for the variables that were significantly ($p<0.05$) associated with fatal SAH risk, in order to evaluate whether the risk factors differentiate within the pairs discordant for fatal SAH. All analyses were performed using the Stata statistical package (version 15).

Results

During the follow-up of 869,469 person-years, we identified 120 fatal SAH events (71 women; median age at death 61.4 years). Of these 120 fatal SAHs, four cases (two twin pairs, zygosity uncertain) occurred in concordant twins whereas the remaining 116 cases were discordant (i.e. the co-twin did not die from SAH). After a fatal SAH in one twin, the median follow-up time for the co-twin was 6.6 years overall. For the 73 pairs where the co-twin lived longer, the follow-up time was 15.1 years; for the 42 pairs where the SAH twin lived longer, the co-twin died on average 8.1 years earlier. In one pair, we ended the follow-up at the time when the co-twin migrated. Of the discordant pairs, 25 were monozygotic, 72 dizygotic and 19 were of uncertain zygosity.

Risk factors for fatal SAH. Table 1 presents the sex-adjusted HRs for fatal SAH by risk factors measured at baseline. Smoking was associated with increased risk of fatal SAH (Table 1). Both heavy (HR 3.01, CI 1.58–5.73) and moderate smokers (HR 3.98, CI 2.44–6.49) had a slightly higher risk than light smokers (HR 2.83, CI 1.64–4.9), but with overlapping confidence intervals. In addition, we found significant association between strong alcohol consumption and the risk of fatal SAH, but this association was no longer apparent after adjusting for smoking. Although the previously reported SAH risk factors, namely female sex[2], hypertension[9] and low leisure time physical activity[10] showed trends towards increased risk of fatal SAH, the findings were non-significant (Table 1).

Risk factor differences within twin pairs. Of the risk factors that were significant in the cohort-wise analysis (alcohol and smoking), smoking was associated with the increased risk of fatal SAH also in pairwise analyses for discordant twin pairs when comparing to non-smokers. The association remained when adjusting for covariates (Table 2). Moreover, the association

remained consistent with sex and zygosity (Table 2), even though the association was statistically non-significant in men and monozygotic twins, most likely due to low sample sizes. On the contrary, we found an elevated but non-significant association between strong alcohol consumption (over 21 grams of alcohol per day) and the risk of fatal SAH (HR=3.17, 0.43–23.3). After adjustment for smoking, the HR decreased to 2.5 (CI 0.29–21.5).

Discussion

Using the twin cohort of 16,282 same-sex Finnish twin pairs, we found that the participants who died from SAH were more commonly smokers at baseline. More interestingly, based on the pairwise analyses, smoking predicted SAH death within discordant sibling pairs, i.e. where only one of the twins died from SAH. Impressively, of the 24 twin pairs also discordant for smoking, the smoking twin died from SAH in 20 pairs; the non-smoking twin died from SAH in the remaining four pairs. Moreover, smoking increased the fatal SAH risk regardless of the twin pairs' sex or zygosity. Our findings suggest that smoking may contribute to the familial manifestation of SAH, and is consistent with strong evidence for a causal effect. This finding is in line with the previously reported strong relationship between smoking and this life-threatening stroke type[2, 3, 9].

According to one of the largest population-based twin cohort studies to date[1], environmental risk factors influence the occurrence of SAH more than inheritance. In addition to smoking, high blood pressure values have been associated with the increased risk of the deadliest type of SAH, i.e. outside-hospital sudden-death SAH[9] whereas female sex[2] has been associated with a slightly increased SAH risk. In our analyses, hypertension as well as female sex had an increasing trend towards a higher risk for fatal SAH, but none of these associations were as evident as the relationship between smoking and SAH. This may at least partly relate to the

effect size, i.e. to the fact that the number of SAH events was insufficient to distinguish relatively weak SAH risk factors. In addition, contrary to hypertension and smoking[9], the impact of female sex on the SAH fatality is unknown. In terms of hypertension, we had no information on measured blood pressure values, which have been shown to be more reliable variables in risk factor assessments than diagnosed hypertension or used medication[11]. Regarding the increased risk of fatal SAH among people with a heavy alcohol consumption (over 21 grams of alcohol per day), our findings are also in line with the previous population-based study showing that the increased risk is attributed to the high smoking rate in heavy alcohol users, rather than to alcohol use per se[12].

Strengthening previous findings reporting that the contribution of genetic factors in the origin of SAH appears to be slight[1], we identified only four fatal SAHs in two concordant twin pairs (3% of all SAHs) and none of them occurred before the age of 43 years. Unfortunately, they did not take part in our surveys, so we have no risk factor information on them. As a major limitation, we did not have data on non-fatal SAH events, which means that we may have missed concordant events in cases where both twins or the co-twin of the discordant pair have suffered a non-fatal SAH. On the other hand, the previous Nordic Twin Study[1], which included both non-fatal and fatal SAHs of the very same Finnish Twin Cohort, found no concordant twin pairs among the Finnish study participants (the two concordant twin pairs included in our study were excluded due to uncertain zygosity) and only 6 (2% of all SAH events) concordant pairs for SAH of all 79,664 Nordic twin pairs. As smoking also increases the risk for non-fatal SAHs[9], we believe it is extremely unlikely that including non-fatal SAH events would substantially impact the study findings and conclusions. Given that only six former smokers died from SAH during the follow-up, we were not able to estimate the impact of former smoking on the fatal SAH risk. Therefore, we ended up combining former and never-smokers

into the non-smoking category. As the risk of SAH among former smokers decreases rapidly after quitting and reaches the risk level of never-smokers within 5 years[2], combining former and never-smokers may perhaps be somewhat justified. Lastly, we were not able to confirm the aneurysmal origin of SAHs (no patient data available) and thus, we may have included a few non-aneurysmal SAH events. Given that the overall number and mortality rate of non-aneurysmal SAHs is relatively small, we believe that this possible bias does not strengthen the reported associations. In fact, since the majority of non-aneurysmal SAHs are not fatal, the inclusion of non-fatal SAHs could have reduced the quality of the results and conclusions.

Conclusions

Smoking increased the risk of fatal SAH, also within twin pairs, and may contribute to a familial manifestation of SAH. Most strikingly, these results suggest that smoking causes fatal SAH.

Acknowledgements

We would like to thank Jacquelin De Faveri for language revision. All authors contributed to design, writing and critical revisions of the presented manuscript. JK performed the statistical analyses of the study but all authors participated in interpreting the results. All authors approved the final version of the manuscript and its submission.

Sources of Funding

IR would like to thank Aarne and Aili Turunen, Paulo and Aarne Koskelo Foundations for research grants. The Finnish Twin Cohort was supported by a grant (# 312073) from the

Academy of Finland Centre of Excellence in Complex Disease Genetics to JK. The funders had no role in the design and conduct of the study; in collection, management, analysis, and interpretation of the data; or in preparation, review, or approval of the manuscript.

Disclosures

None

References

1. Korja M, Silventoinen K, McCarron P, Zdravkovic S, Skytthe A, Haapanen A, de Faire U, Pedersen NL, Christensen K, Koskenvuo M, et al. Genetic epidemiology of spontaneous subarachnoid hemorrhage: Nordic twin study. *Stroke*. 2010;41:2458-2462
2. Lindbohm JV, Kaprio J, Jousilahti P, Salomaa V, Korja M. Sex, smoking, and risk for subarachnoid hemorrhage. *Stroke*. 2016;47:1975-1981
3. Feigin V, Parag V, Lawes CM, Rodgers A, Suh I, Woodward M, Jamrozik K, Ueshima H, Asia Pacific Cohort Studies C. Smoking and elevated blood pressure are the most important risk factors for subarachnoid hemorrhage in the asia-pacific region: An overview of 26 cohorts involving 306,620 participants. *Stroke*. 2005;36:1360-1365
4. General Assembly of the World Medical A. World medical association declaration of helsinki: Ethical principles for medical research involving human subjects. *J Am Coll Dent*. 2014;81:14-18
5. Kaprio J, Bollepalli S, Buchwald J, Iso-Markku P, Korhonen T, Kovanen V, Kujala U, Laakkonen EK, Latvala A, Leskinen T, et al. The older finnish twin cohort - 45 years of follow-up. *Twin Res Hum Genet*. 2019;22:240-254
6. Kaprio J, Koskenvuo M. Genetic and environmental factors in complex diseases: The older finnish twin cohort. *Twin Res*. 2002;5:358-365
7. WHO. Obesity: Preventing and managing the global epidemic. *2000*
8. Sarna S, Kaprio J, Sistonen P, Koskenvuo M. Diagnosis of twin zygosity by mailed questionnaire. *Hum Hered*. 1978;28:241-254
9. Lindbohm JV, Kaprio J, Jousilahti P, Salomaa V, Korja M. Risk factors of sudden death from subarachnoid hemorrhage. *Stroke*. 2017;48:2399-2404
10. Lindbohm JV, Rautalin I, Jousilahti P, Salomaa V, Kaprio J, Korja M. Physical activity associates with subarachnoid hemorrhage risk- a population-based long-term cohort study. *Sci Rep*. 2019;9:9219
11. Lindbohm JV, Kaprio J, Korja M. Survival bias explains improved survival in smokers and hypertensive individuals after asah. *Neurology*. 2019;93:e2105-e2109
12. Korja M, Silventoinen K, Laatikainen T, Jousilahti P, Salomaa V, Hernesniemi J, Kaprio J. Risk factors and their combined effects on the incidence rate of subarachnoid hemorrhage--a population-based cohort study. *PLoS One*. 2013;8:e73760

Tables

Table 1. Baseline characteristics by fatal subarachnoid hemorrhage status.

Variable	Fatal SAH, n (%)		HR (95% CI)
	No	Yes	
Number of cases	31 058	120	-
Age at baseline (mean, SD)	36.9 (14.9)	40.1 (14.9)	-
Sex			
• Men	15 717 (50.6)	53 (44.2)	(Reference)
• Women	15 341 (49.4)	67 (55.8)	1.12 (0.77–1.62)
Hypertension			
• No	22 202 (71.5)	80 (66.7)	(Reference)
• Yes	3 792 (12.2)	18 (15.0)	1.05 (0.61–1.83)
• Missing	5 064 (16.3)	22 (18.3)	
Smoking			
• No (never/former)	17 647 (56.8)	46 (38.3)	(Reference)
• Yes (occasional/current)	9 991 (32.2)	60 (50.0)	3.33 (2.24–4.95)
• Missing	3 420 (11.0)	14 (11.7)	
LTPA (MET quintiles)			
• Very low	5 345 (17.2)	26 (21.7)	(Reference)
• Low	5 378 (17.3)	24 (20.0)	0.95 (0.55–1.67)
• Moderate	5 319 (17.1)	16 (13.3)	0.66 (0.36–1.24)
• High	5 233 (16.8)	17 (14.2)	0.70 (0.38–1.30)
• Very high	5 262 (16.9)	18 (15.0)	0.77 (0.42–1.41)
• Missing	4 521 (14.6)	19 (15.8)	
Education, (years)			
• Low (<3)	12 403 (40.0)	50 (41.7)	(Reference)

• Moderate (3–6) • High (≥7) • Missing	11 491 (37.0) 2 037 (6.6) 5 127 (16.5)	41 (34.2) 6 (5.0) 23 (19.2)	1.17 (0.77–1.79) 0.91 (0.39–2.13)
Alcohol, grams per day • 0 • 1–10 • 11–20 • 21+ • Missing	 6 595 (21.2) 14 376 (46.3) 2 479 (8.0) 2483 (8.0) 5 125 (16.5)	 29 (24.2) 42 (35.0) 11 (9.2) 15 (12.5) 23 (19.2)	 (Reference) 0.90 (0.56–1.46) 1.62 (0.74–3.53) 2.44 (1.22–4.88)
Obesity, (BMI) • Underweight (<18.5) • Normal weight (18.5–24.9) • Overweight (25.0–29.9) • Obese (≥30) • Missing	 888 (2.9) 18 527 (59.7) 7 024 (22.6) 1 193 (3.8) 3 426 (11.0)	 1 (0.8) 72 (60.0) 25 (20.8) 8 (6.7) 14 (11.7)	 0.40 (0.06–2.87) (Reference) 0.67 (0.42–1.09) 1.20 (0.57–2.53)

BMI=body mass index; CI=confidence interval; HR= hazard ratio; IQR=interquartile range; LTPA=leisure time physical activity; MET=metabolic equivalent of task; SAH=subarachnoid hemorrhage

Table 2. Within-pair hazard ratios (HRs) for fatal SAH between non-smoking (never/former) and smoking (occasional/current) discordant twins.

	N of discordant twin pairs	HR (95% CI)
Overall		
• Partly adjusted*	106	6.33 (1.87–21.4)
• Fully adjusted†	94	7.61 (1.68–34.4)
By sex		
• Men	48	7.00 (0.86–56.9)
• Women	58	6.00 (1.34–26.8)
By zygosity		
• DZ	68	5.50 (1.22–24.8)
• MZ	25	4.00 (0.45–35.8)

*partly adjusted model=adjusted for sex, age and zygosity

†fully adjusted model=adjusted for sex, age, zygosity, body mass index, alcohol and hypertension

CI=confidence interval; DZ=dizygotic; MZ=monozygotic; HR=hazard ratio