Quantifying P450 Eicosanoids Levels Following Subarachnoid Hemorrhage

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Introduction

Aneurysmal subarachnoid hemorrhage (aSAH) is associated with a high degree of secondary brain injury, often due to delayed ischemia. Growing evidence suggests that dysfunction of the microvasculature may be a key factor. P450 metabolites are important regulators of microvascular tone. Studies have shown that CSF levels of the vasoconstrictive metabolite, 20-hydroxyeicosatetraenoic acid (20-HETE) rise over the first two weeks after aSAH. A different group of metabolites the eicosatrienoic acids (EETs) have been shown in animal models to be vasodilatory. The purpose of this study is to determine if levels of p450 metabolites change over time in the CSF of patients with aSAH and secondarily if functional genetic polymorphisms in the gene (EPHX2) that controls the metabolic degradation of EETs correlates with clinical outcome.

Methods

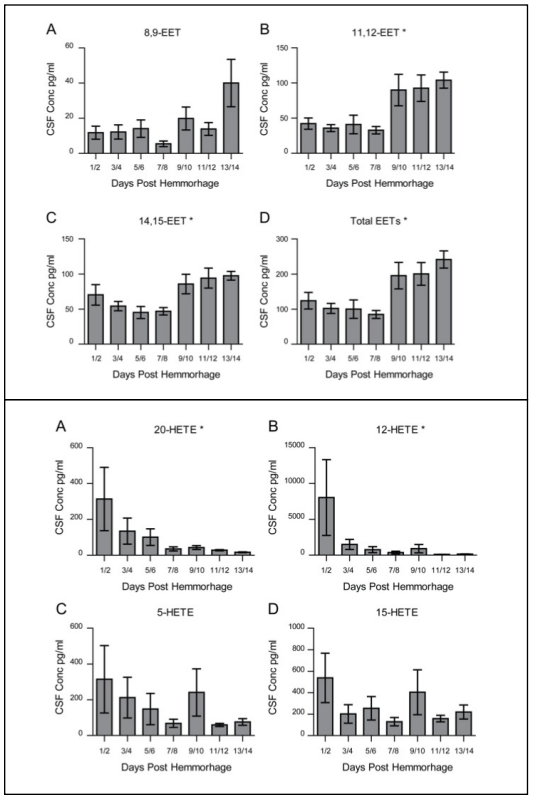
Patients 18 to 80 years old with aSAH were enrolled in the study. Clinical and radiological presentation, development of vasospasm and clinical outcomes were recorded. Serial samples of CSF were collected over 14 days in patients with ventricular access devices. Levels of P450 metabolites in the CSF were quantified using liquid chromatography tandem mass spectrometry. Genotyping was conducted on DNA extracted from whole blood samples and allelic discrimination of EPHX2 polymorphisms was performed using Taqman amplification.

Results

CSF levels of 20-HETE declined during the acute phase of hemorrhage. Inversely, CSF levels of 14,15-EET rose. EPHX2 polymorphism analysis showed a trend of poorer Glasgow Outcome Score in the K55R population at time of discharge, compared to other genotypes. No difference was noted between patients with the R287Q EPHX2 polymorphism compared to Wild Type.

Conclusions

P450 metabolites may be important in aSAH. CSF levels of the vasodilatory 14,15 EETs gradually increased over a 14 day recovery period whereas the vasoconstrictive 20-HETEs declined over this same time. Polymorphisms that speed degradation of EETs (theoretically leading to lower circulating levels) may be associated with poorer clinical outcomes suggesting an important role of p450 metabolites in regulating microvascular tone after aSAH.



Learning Objectives

Describe the potential role of the P450 metabolites in subarachnoid hemorrhage

Summary of stroke, vasospasm, delayed cerebral ischemia, and outcomes stratified by EPHX2 genotype

Characteristics*	EPHX2 Genotype Wild Type (n=46)	K55r (n=12)	R287q (n=19)	p-value
Strokes				
Stroke at admission	4 (8.9)	1 (8.3)	2 (10.5)	0.97
Stroke after intervention	20 (43.5)	3 (25)	8 (42.1)	0.50
New stroke at discharge/follow up	10 (21.7)	6 (50.0)	3 (15.8)	0.076
Any stroke at discharge/follow up	31 (72.1)	10 (90.9)	10 (52.6)	0.078
Vasospasm				
TCD	12 (26.1)	4 (33.3)	7 (36.8)	0.66
Angiographic	24 (52.2)	5 (41.7)	9 (47.4)	0.80
Angiographic intervention	19 (41.3)	2 (16.7)	6 (31.6)	0.26
Clinical deterioration d/t DCI	17 (37.0)	3 (25)	5 (26.3)	0.59
ICU discharge GCS, mean ± SD	13.3 ± 2.8	10.3 ± 5.5	12.9 ± 3.9	0.045†
Hospital discharge GOS, mean ± SD	3.2 (.95)	2.6 (1.2)	3.4 (1.2)	0.09
Discharge GOS < 3, n (%)	30 (65.2)	9 (75.0)	7 (36.8)	0.053
Hospital length of stay, mean ± SD	21 ± 12	17 ± 7	17 ± 8	0.23
Mortality at follow-up	3 (6.5)	4 (33.3)	3 (15.8)	0.044
Composite of death or new stroke	13 (28.3)	8 (66.7)	6 (31.6)	0.043
Disposition				0.040
Home / rehab	28 (60.9)	4 (33.3)	15 (78.9)	
SNF/LTAC/dead	18 (39.1)	8 (66.7)	4 (21.1)	

TCD transcranial Doppler, GCS Glasgow coma scale, GOS Glasgow outcome scale, SNF skilled nursing facility, LTAC long term acute care hospital

*Data are expressed as frequency distributions unless otherwise specified

†p=0.05 for wild type vs. k55r genotype, using Tukey-Kramer *post hoc* pairwise comparison

Unadjusted and adjusted odds ratios for primary and secondary outcome measures.

	DCI w/clinical change	Mortality	New Stroke	GOS ≤ 3
Unadjusted Model*				
k55r	0.57 (0.14-2.40)	**7.17 (1.34-38.3)**	3.6 (0.95- 13.62)	1.6 (.38-6.76)
r287q	0.61 (0.19 - 1.99)	2.69 (0.49-14.7)	0.68 (0.16 - 2.79)	**.31 (.10-.95)**
Adjusted Model*				
k55r	0.62 (0.14-2.79)	**7.62 (1.19-48.7)**	3.47 (0.89-13.57)	1.47 (0.30-7.13)
r287q	0.61 (0.17 - 2.12)	2.9 (0.47-18.12)	0.67 (0.16-2.79)	**0.23 (0.06-0.88**
age	1.0 (0.95-1.04)	1.06 (0.99-1.13)	1.01 (0.67-1.05)	1.03 (0.98-1.07)
GCS at admission	0.86 (0.76-0.96)	0.83 (0.69-0.99)	0.97 (0.86-1.11)	0.77 (0.66-0.89)

DCI delayed cerebral ischemia, GCS Glasgow coma scale, GOS Glasgow outcome scale

*Data presented as Odds ratios with 95% confidence intervals OR (95% CI).