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Paradoxical Increase in Mortality and Rupture of Intracranial Aneurysms in mPGES-1 Deficient Mice: Attenuation by Aspirin

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Abstract

Background—Inflammation plays an important role in formation and rupture of intracranial aneurysms. Expression of microsomal prostaglandin E₂ (PGE₂) synthase type 1 (mPGES-1) is increased in the wall of intracranial aneurysms in humans. PGE₂, a by-product of mPGES-1, is associated with inflammation and cerebrovascular dysfunction.

Objective—To test the hypothesis that deletion of mPGES-1 decreases the formation and rupture of intracranial aneurysms in a murine model.

Methods—Intracranial aneurysms were induced in wild type (WT) and mPGES-1 deficient (mPGES-1 KO) mice using a combination of deoxycorticosterone acetate (DOCA)-salt-induced hypertension and intracranial injection of elastase in the basal cistern. Prevalence of aneurysms, subarachnoid hemorrhage (SAH), and mortality were assessed. We also tested effects of administration of aspirin (6mg/kg/d) by gavage and PGE₂ (1mg/kg/d) by subcutaneous infusion.

Results—Systolic blood pressure (SBP) and prevalence of aneurysm were similar in WT and mPGES-1 KO mice. However, mortality and prevalence of SAH were markedly increased in mPGES-1 KO mice ($p < 0.05$). Bone marrow reconstitution studies suggest that mPGES-1 derived from leukocytes does not appear to increase rupture of intracranial aneurysms. Aspirin, but not PGE₂, attenuated the increased mortality in mPGES-1 KO mice ($p < 0.05$).

Conclusion—Vascular mPGES-1 plays a protective role in blood vessels and attenuates rupture of cerebral aneurysms. In contrast to effects on abdominal aneurysms, mPGES-1 deficiency is associated with an increase in rupture of cerebral aneurysms and mortality, which are attenuated by low-dose aspirin.

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Keywords

Prostaglandin E₂; intracranial aneurysm; subarachnoid hemorrhage; mPGES-1; aspirin

INTRODUCTION

Prostaglandin E₂ (PGE₂) is associated with human cardiovascular^{1, 2} and cerebrovascular disease³. PGE₂ also is involved in the pathogenesis of several animal models of cardiovascular⁴⁻⁶ and cerebrovascular disease^{7, 8}. PGE₂ is synthesized from arachidonic acid by the sequential activity of cyclooxygenase (Cox) 1 and 2, and microsomal PGE₂ synthase type 1 (mPGES-1)^{9, 10}. Genetic deletion of mPGES-1 decreases atherogenesis and formation of aneurysms of the abdominal aorta in mice^{4, 5}. Because PGE₂ contributes to inflammation and vascular damage, inhibition of PGE₂ synthesis is an attractive target for development of therapeutics in inflammation, pain, and cardiovascular disease.¹¹

We have found that levels of Cox2 and mPGES-1 are increased in human intracranial aneurysms³. mPGES-1 is also increased in cerebral arteries collected from mice with experimental intracranial aneurysms¹². The role of mPGES-1, however, in formation and rupture of intracranial aneurysms is not clear. In this study, we explored the hypothesis that mPGES-1 deficiency decreases the risk of intracranial aneurysm rupture in mice. Surprisingly, we found that mPGES-1 deficiency increases mortality and rupture of intracranial aneurysms in mice. In this study, we explored mechanisms by which mPGES-1 may protect against aneurysm rupture, and found that aspirin, but not reconstitution of PGE₂, attenuates mortality in mPGES-1 deficient mice.

METHODS

Experimental animals

Studies were performed in 157 adult wild type (WT) (CD45.2 and CD45.1 for bone marrow studies) and mPGES-1 knockout (mPGES-1- KO) mice. The mPGES-1 KO mice were bred on the C57BL/6 background^{6, 13}. WT mice (B6-LY5.1 and B6LY5.2 mice) were obtained from the National Cancer Institute. These mice are phenotypically normal; however, the variant in the CD45 marker (LY5.1 or Ly5.2) can be used to trace the hematopoietic cell engraftment after bone marrow reconstitution¹⁴. All experimental protocols and procedures conform to the National Institute of Health guidelines and were approved by the Institutional Animal Care and Use Committee of the University of Iowa.

Induction of Aneurysms

Intracranial aneurysms were induced as described previously^{15, 16}. Following anesthesia (ketamine-xylazine i.p.) and analgesia (buprenorphine 0.2mg/kg i.p.), a right nephrectomy was performed. One week later, under anesthesia, an injection of bovine elastase (35 mU in 2.5 µl) was made under stereotactic guidance using the following coordinates: 2.7 mm posterior to the bregma, 1 mm to the right of the midline, depth of 6.2 mm from the skull. A deoxycorticosterone acetate (DOCA) pellet (66mg/28 days) was then implanted subcutaneously in the back. Systolic blood pressure was measured using the tail cuff method

between 9–11 am by the same operator. Systolic blood pressure data are presented in Supplemental Digital Content 1. Mice were given food and water (1% NaCl) *ad libitum*. Some groups received a low dose of aspirin (6 mg/kg/d)¹⁷ by gavage from day 2, or PGE₂ (1mg/kg/d)¹⁸ by subcutaneous infusion from day 0.

Mice that recovered completely after surgery (87%, 137/157) were included in the studies. Mice were euthanized when signs of neurological deficit suggested subarachnoid hemorrhage (SAH), weight loss was >20% baseline, or after 3 weeks. A survival curve was made, based on animals that were found dead (12%, 17/137) or euthanized because of neurological deficit or weight loss and evidence of SAH.

Aneurysm analysis

Immediately after euthanasia, ice-cold physiologic saline containing papaverine (100 µM) was perfused transcardially. Then, a mix of 2 mg/ml of bromophenol blue dye in 8% gelatin/saline was infused to facilitate visualization of arteries and aneurysms. The brain was removed and assessed for intracranial aneurysms and/or SAH. Intracranial aneurysms were operationally defined as any dilatation greater than 1.5x the diameter of the parent artery. Both saccular and fusiform dilatations were included, because either may rupture and cause SAH¹⁹. The distribution of findings in brains of WT and MPGES-1 KO mice is presented in Supplemental Digital Content 2. The brains of mice that were found dead were photographed without perfusion with the gelatin-dye mix, and assessed for SAH and aneurysm.

Bone Marrow Reconstitution Studies

Young CD45.2 WT or mPGES-1 KO female mice were used as donors. Bone marrow was harvested under aseptic conditions from the femur, tibia, and iliac of donor mice. Erythrocytes were lysed and the remaining cells were used for bone marrow transplant. Recipient mice were lethally irradiated using 2 doses of 5 Gy on the same day. After the last irradiation, the mice received 5 million donor cells via tail vein. Mice received ampicillin (2 g/L in drinking water) for 6 weeks, and bone marrow reconstitution was assessed after 2 months using the expression of CD45.2 marker by flow cytometry. The following antibodies were used: CD45.1 clone A20 from BD biosciences and CD45.2 clone 104 from Biolegend. Samples were run on a BD LSR II flow cytometer and analyzed with Flowjo (Treestar) software.

Drugs

All reagents were obtained from Sigma (St Louis, MO) except for DOCA (Innovative Research of America, Sarasota, FL) and PGE₂ (Cayman Chemical, Ann arbor, MI).

Statistical analysis

Analysis was performed using Prism 6 (Graphpad, La Jolla, CA). Categorical data (prevalence of aneurysms and SAH) were compared using one-tailed Chi square test. Survival was analyzed with Gehan-Beslow-Wilcoxon test. A P value less than 0.05 was considered significant.

RESULTS

Effect of mPGES-1 deficiency on formation and rupture of intracranial aneurysms

Compared to control mice (Figure 1A), about half of mPGES-1 KO mice died during follow-up ($P<0.05$). Prevalence of intracranial aneurysms (Figure 2) did not differ in both groups [73% (8/11) in WT, 81% (13/16) in mPGES-1 KO mice] (Figure 1B and Supplemental Digital Content 2). Incidence of SAH was significantly higher in mPGES-1 KO [56% (9/16)] mice than in WT controls [0% (0/11)] ($P<0.05$).

Contribution of vascular vs leukocyte mPGES-1 in formation and rupture of intracranial aneurysms

In bone marrow reconstitution studies (see Figure A, Supplemental Digital Content 3), reconstitution with donor cells was optimal (above 90%) (see Figure B–C, Supplemental Digital Content 3). Mortality was similar in WT mice that received WT or mPGES-1 KO bone marrow (Figure 3A). Similarly, no difference in mortality was observed in mPGES-1 KO that received WT or KO bone marrow (Figure 3A). Mortality in transplanted mPGES-1 KO mice was higher than in transplanted WT mice ($P<0.05$). Prevalence of aneurysms was similar in all groups [90% (9/10) WT to WT, 91% (10/11) KO to WT, 91% (10/11) WT to KO, 94% (15/16) KO to KO] (Figure 3B). Prevalence of SAH tended to be increased in transplanted mPGES-1 KO mice than in transplanted WT mice [50% (5/10) WT to WT, 27% (3/11) KO to WT, 73% (8/11) WT to KO, 88% (14/16) KO to KO]. (Figure 3C).

Effect of PGE₂ reconstitution in mPGES-1 KO mice

Infusion of PGE₂ in mPGES-1 KO mice did not decrease mortality ($P>0.05$) (Figure 4A). Similarly, PGE₂ infusion in mPGES-1 KO mice did not reduce prevalence [81% (13/16) with PGE₂ vs 86% (12/14) with vehicle] or rupture [75% (12/16) vs 71% (10/14)] of intracranial aneurysms in PGE₂ vs vehicle treated mice, respectively ($P>0.05$).

Effect of aspirin in mPGES-1 KO mice

Survival was significantly higher in mPGES-1 KO mice treated with aspirin than in vehicle (water) treated mice ($P<0.05$) (Fig 5). Aspirin did not reduce prevalence of aneurysms [85% (11/13) vs 100% (12/12)] ($P>0.05$) but tended to reduce incidence of SAH [46% (6/13) vs 75% (9/12)] ($P=0.07$) in mPGES1 KO mice treated with aspirin vs vehicle, respectively. Expression of markers of inflammation (TNF- α , MCP-1, IL-6, CD-68, Cox-2) and the expression of NOX-2 (a subunit of the NADPH oxidase) was examined using RT-PCR. Expression of these genes was not different in mPGES-1 KO mice treated with or without aspirin (data not shown).

Discussion

Because PGE₂ is associated with vascular disease and cerebrovascular dysfunction, we hypothesized that decreased formation of PGE₂ in mPGES-1 KO mice would protect against intracranial aneurysm rupture. Surprisingly, mPGES-1 appears to have a protective role in cerebral arteries, because its genetic deletion increases rupture of intracranial aneurysms in mice. Bone marrow reconstitution studies suggest that leukocyte-derived mPGES-1 does not

increase aneurysm rupture. Increased mortality in mPGES-1 KO mice was decreased by aspirin, but not by infusion of PGE₂.

PGE₂ is associated with cardiovascular disease and cerebrovascular disease in humans and experimental models of cardiovascular disease⁴⁻⁸. Expression of Cox-2 and mPGES-1 is increased in intracranial aneurysms in humans³. PGE₂ augments endothelial dysfunction induced by angiotensin II in systemic^{20, 21} and cerebral arteries^{7, 8, 22}. PGE₂ and mPGES-1 also augment neointimal growth after vascular injury^{6, 23}. Genetic deletion of mPGES-1 decreases formation of aneurysms of the aorta after infusion of angiotensin II⁶, and attenuates atherogenesis in low density lipoprotein receptor deficient mice⁴. Thus, several lines of evidence suggest that PGE₂ levels or synthesis are increased and may augment progression of cardiovascular disease in several experimental models.

It was surprising that mPGES-1 deficiency is associated with increased rupture of intracranial aneurysm in mice. A possible explanation for the finding is that mPGES-1 deficiency is associated with impaired natriuresis and increased sensitivity to salt loading and DOCA²⁴. We did not find a significant increase in blood pressure mPGES-1 deficient mice vs wild type mice. We acknowledge that we measured blood pressure during the day, and it is known that the DOCA model of hypertension displays a circadian variation²⁵. This circadian oscillation of blood pressure may limit the recording of consistently high blood pressures during the day. Moreover, because mice in this model of intracranial aneurysms start to die a couple of days after surgery, some of them may have intracranial hemorrhages and become sick. These conditions can also increase the variability of blood pressure data.

It could be speculated that mPGES-1 deficiency may disturb formation of elastin in cerebral arteries and aggravate the effects of elastase in this intracranial aneurysm model. Our data, however, suggest that mPGES-1 deficiency did not aggravate aneurysm formation, because both WT and mPGES-1 deficient mice had similar incidence of aneurysms. Additionally, recent studies revealed that PGE₂ disrupts elastogenesis in mice²⁶. Therefore, it would be expected that lack of PGE₂ in mPGES-1 deficient mice would enhance the integrity of the elastic lamina.

In a model of injury to the femoral artery produced by a wire, vascular and leukocyte mPGES-1 play different roles in vascular remodeling²⁷. Deletion of mPGES-1 in either smooth muscle or endothelium resulted in increased neointimal formation and damage after wire injury⁵, which suggests that vascular mPGES-1 is protective. In contrast, deletion of mPGES-1 in macrophages resulted in reduction of neointimal formation and vascular damage after wire injury, which suggests that macrophage-derived mPGES-1 is deleterious in vascular disease²⁷. Our results after bone marrow transplantation, however, do not suggest a contribution of hematopoietic mPGES-1 to rupture of intracranial aneurysms, because survival was similar in mPGES-1 KO recipient or WT recipient mice, independently of whether WT or mPGES-1 KO bone marrow was transplanted. Importantly, mortality of mPGES-1 KO recipients was higher than mortality in WT recipients. It is interesting that mortality was higher in irradiated- recipient WT mice (Fig 3) versus non-irradiated mice (Fig 1). We speculate that irradiation sensitizes cerebral arteries to effects of elastase and hypertension; hence, aneurysm rupture and mortality are increased in irradiated mice.

Effects of irradiation are not clear in humans, but some studies suggest that irradiation may increase the risk of rupture of intracranial aneurysms in patients²⁸.

Because mPGES-1 is protective in our model, we speculate that increased aneurysm rupture in mPGES-1 KO mice could be explained by two mechanisms. First, PGE₂ may protect against aneurysm rupture. It was shown that infusion of PGE₂ decreased inflammation and fibrosis in a model of pulmonary disease¹⁸. We therefore infused PGE₂ in mPGES-1 KO mice to determine whether PGE₂ increases survival and decreases aneurysm rupture. PGE₂ infusion, however, did not affect survival, prevalence of aneurysms formation, or rupture in mPGES-1 KO mice. Second, in the absence of mPGES-1, PGH₂, the product of cyclooxygenase activity¹⁰, cannot be converted to PGE₂ and could be converted to a harmful prostaglandin. Thus, blocking prostaglandin synthesis using a cyclooxygenase inhibitor, such as aspirin, may attenuate aneurysm rupture. We have reported that acetylsalicylic acid (aspirin) reduces the risk of SAH in patients with intracranial aneurysms²⁹. This observation was confirmed in a different retrospective study, in which chronic low-dose aspirin decreased the risk of SAH in patients³⁰. Aspirin inhibits both Cox-1 and Cox-2⁹. Low doses of aspirin decrease inflammatory responses in humans³¹. Aspirin also appears to decrease inflammation in intracranial aneurysms³². After induction of intracranial aneurysms, aspirin increased survival in mPGES-1 KO mice. Aspirin did not attenuate formation of aneurysms, but tended to decrease aneurysm rupture. We speculate that a substrate of mPGES-1 accumulated in mPGES-1 KO mice and aspirin blocked this pathway, and thereby decreased mortality (see Figure, Supplemental Digital Content 4).

Limitations

A limitation of this study is that, because it is difficult to collect enough tissue for biochemical assays from mouse intracranial aneurysms, we could not identify a prostaglandin that is associated with increases in mortality in mPGES-1 KO mice.

We studied the role of MPGES-1 in intracranial aneurysm rupture using exclusively a genetic model of MPGES-1 deficiency. There is little information about potential compensatory changes in mPGES-1 biology after genetic knockout that could explain the increased mortality in mPGES-1 KO mice, after induction of intracranial aneurysms. We considered inhibiting MPGES-1 in WT mice using a pharmacological approach. This approach is not feasible in mice, however, because available inhibitors of human mPGES-1 may not be completely specific³³. There are also structural differences between human and mouse mPGES-1³⁴ that limit the access of available inhibitors to the active site of mouse mPGES-1³⁵. Finally, compensatory responses are not unique to genetic models. Chronic treatment with pharmacological agents or inhibitors can also induce compensatory responses.

Conclusion

Inflammation is associated with rupture of intracranial aneurysms in humans. Because PGE₂ is associated with inflammatory responses, vascular disease, and cerebrovascular dysfunction, we initially hypothesized that decreased formation of PGE₂ in mPGES-1 deficient mice would protect against rupture of intracranial aneurysms. Instead, our findings

indicate that mPGES-1 plays a protective role in cerebrovascular disease. We suggest that cerebrovascular outcomes should be addressed in studies of development of mPGES-1 inhibitors.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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References

1. Holmes DR, Wester W, Thompson RW, Reilly JM. Prostaglandin E₂ synthesis and cyclooxygenase expression in abdominal aortic aneurysms. *J Vasc Surg.* May; 1997 25(5):810–815. [PubMed: 9152308]
2. Camacho M, Dilme J, Sola-Villa D, et al. Microvascular COX-2/mPGES-1/EP-4 axis in human abdominal aortic aneurysm. *J Lipid Res.* Dec; 2013 54(12):3506–3515. [PubMed: 24133193]
3. Hasan D, Hashimoto T, Kung D, Macdonald RL, Winn HR, Heistad D. Upregulation of cyclooxygenase-2 (COX-2) and microsomal prostaglandin E₂ synthase-1 (mPGES-1) in wall of ruptured human cerebral aneurysms: preliminary results. *Stroke.* Jul; 2012 43(7):1964–1967. [PubMed: 22588264]
4. Wang M, Zukas AM, Hui Y, Ricciotti E, Pure E, Fitzgerald GA. Deletion of microsomal prostaglandin E synthase-1 augments prostacyclin and retards atherogenesis. *Proc Natl Acad Sci USA.* Sep 26; 2006 103(39):14507–14512. [PubMed: 16973753]
5. Wang M, Lee E, Song W, et al. Microsomal prostaglandin E synthase-1 deletion suppresses oxidative stress and angiotensin II-induced abdominal aortic aneurysm formation. *Circulation.* Mar 11; 2008 117(10):1302–1309. [PubMed: 18285567]
6. Wang M, Ihida-Stansbury K, Kothapalli D, et al. Microsomal prostaglandin E₂ synthase-1 modulates the response to vascular injury. *Circulation.* Feb 15; 2011 123(6):631–639. [PubMed: 21282500]
7. Capone C, Faraco G, Anrather J, Zhou P, Iadecola C. Cyclooxygenase 1-derived prostaglandin E₂ and EP1 receptors are required for the cerebrovascular dysfunction induced by angiotensin II. *Hypertension.* Apr; 2010 55(4):911–917. [PubMed: 20194308]
8. Cao X, Peterson JR, Wang G, et al. Angiotensin II-dependent hypertension requires cyclooxygenase 1-derived prostaglandin E₂ and EP1 receptor signaling in the subfornical organ of the brain. *Hypertension.* Apr; 2012 59(4):869–876. [PubMed: 22371360]
9. Wang M, Fitzgerald GA. Cardiovascular biology of microsomal prostaglandin E synthase-1. *Trends Cardiovasc Med.* Aug; 2010 20(6):189–195. [PubMed: 22137640]
10. Ricciotti E, Fitzgerald GA. Prostaglandins and inflammation. *Arterioscler Thromb Vac Biol.* May; 2011 31(5):986–1000.
11. Chang HH, Meuillet EJ. Identification and development of mPGES-1 inhibitors: where we are at? *Future medicinal chemistry.* Nov; 2011 3(15):1909–1934. [PubMed: 22023034]
12. Peña Silva RA, Kung DK, Mitchell IJ, et al. Angiotensin 1–7 Reduces Mortality and Rupture of Intracranial Aneurysms in Mice. *Hypertension.* May 5; 2014 64(2):362–368. [PubMed: 24799613]
13. Trebino CE, Stock JL, Gibbons CP, et al. Impaired inflammatory and pain responses in mice lacking an inducible prostaglandin E synthase. *Proc Natl Acad Sci USA.* Jul 22; 2003 100(15):9044–9049. [PubMed: 12835414]

14. Martin MD, Condotta SA, Harty JT, Badovinac VP. Population dynamics of naive and memory CD8 T cell responses after antigen stimulations in vivo. *J Immunol.* Feb 1; 2012 188(3):1255–1265. [PubMed: 22205031]
15. Nuki Y, Tsou TL, Kurihara C, Kanematsu M, Kanematsu Y, Hashimoto T. Elastase-induced intracranial aneurysms in hypertensive mice. *Hypertension.* Dec; 2009 54(6):1337–1344. [PubMed: 19884566]
16. Tada Y, Wada K, Shimada K, et al. Roles of hypertension in the rupture of intracranial aneurysms. *Stroke.* Feb; 2014 45(2):579–586. [PubMed: 24370755]
17. Tancevski I, Wehinger A, Schgoer W, et al. Aspirin regulates expression and function of scavenger receptor-BI in macrophages: studies in primary human macrophages and in mice. *FASEB J.* Jul; 2006 20(9):1328–1335. [PubMed: 16816107]
18. Dackor RT, Cheng J, Voltz JW, et al. Prostaglandin E₂ protects murine lungs from bleomycin-induced pulmonary fibrosis and lung dysfunction. *Am J Physiol Lung Cell Mol Physiol.* Nov; 2011 301(5):L645–655. [PubMed: 21856819]
19. Findlay JM, Hao C, Emery D. Non-atherosclerotic fusiform cerebral aneurysms. *Can J Neurol Sci.* Feb; 2002 29(1):41–48. [PubMed: 11858533]
20. Francois H, Athirakul K, Mao L, Rockman H, Coffman TM. Role for thromboxane receptors in angiotensin-II-induced hypertension. *Hypertension.* Feb; 2004 43(2):364–369. [PubMed: 14718360]
21. Guan Y, Zhang Y, Wu J, et al. Antihypertensive effects of selective prostaglandin E₂ receptor subtype 1 targeting. *J Clin Invest.* Sep; 2007 117(9):2496–2505. [PubMed: 17710229]
22. Kawano T, Anrather J, Zhou P, et al. Prostaglandin E₂ EP1 receptors: downstream effectors of COX-2 neurotoxicity. *Nat Med.* Feb; 2006 12(2):225–229. [PubMed: 16432513]
23. Zhang J, Zou F, Tang J, et al. Cyclooxygenase-2-derived prostaglandin E₂ promotes injury-induced vascular neointimal hyperplasia through the E-prostanoid 3 receptor. *Circ Res.* Jul 5; 2013 113(2):104–114. [PubMed: 23595951]
24. Jia Z, Zhang A, Zhang H, Dong Z, Yang T. Deletion of microsomal prostaglandin E synthase-1 increases sensitivity to salt loading and angiotensin II infusion. *Circ Res.* Nov 24; 2006 99(11):1243–1251. [PubMed: 17095726]
25. Hilzendeger AM, Cassell MD, Davis DR, et al. Angiotensin type 1a receptors in the subfornical organ are required for deoxycorticosterone acetate-salt hypertension. *Hypertension.* Mar; 2013 61(3):716–722. [PubMed: 23266541]
26. Yokoyama U, Minamisawa S, Shioda A, et al. Prostaglandin E₂ inhibits elastogenesis in the ductus arteriosus via EP4 signaling. *Circulation.* Jan 28; 2014 129(4):487–496. [PubMed: 24146253]
27. Chen L, Yang G, Xu X, et al. Cell selective cardiovascular biology of microsomal prostaglandin E synthase-1. *Circulation.* Jan 15; 2013 127(2):233–243. [PubMed: 23204105]
28. Nanney AD 3rd, El Tecle NE, El Ahmadi TY, et al. Intracranial aneurysms in previously irradiated fields: Literature review and case illustration. *World neurosurgery.* Oct 19.2013
29. Hasan DM, Mahaney KB, Brown RD Jr, et al. Aspirin as a promising agent for decreasing incidence of cerebral aneurysm rupture. *Stroke.* Nov; 2011 42(11):3156–3162. [PubMed: 21980208]
30. Garcia-Rodriguez LA, Gaist D, Morton J, Cookson C, Gonzalez-Perez A. Antithrombotic drugs and risk of hemorrhagic stroke in the general population. *Neurology.* Aug 6; 2013 81(6):566–574. [PubMed: 23843464]
31. Morris T, Stables M, Hobbs A, et al. Effects of low-dose aspirin on acute inflammatory responses in humans. *J Immunol.* Aug 1; 2009 183(3):2089–2096. [PubMed: 19597002]
32. Hasan DM, Chalouhi N, Jabbour P, et al. Evidence that acetylsalicylic acid attenuates inflammation in the walls of human cerebral aneurysms: preliminary results. *Journal of the American Heart Association.* Feb.2013 2(1):e000019. [PubMed: 23525414]
33. Koeberle A, Werz O. Inhibitors of the microsomal prostaglandin E(2) synthase-1 as alternative to non steroidal anti-inflammatory drugs (NSAIDs)—a critical review. *Curr Med Chem.* 2009; 16(32):4274–4296. [PubMed: 19754418]

34. Pawelzik SC, Uda NR, Spahiu L, et al. Identification of key residues determining species differences in inhibitor binding of microsomal prostaglandin E synthase-1. *J Biol Chem.* Sep 17; 2010 285(38):29254–29261. [PubMed: 20605783]
35. Mbalaviele G, Pauley AM, Shaffer AF, et al. Distinction of microsomal prostaglandin E synthase-1 (mPGES-1) inhibition from cyclooxygenase-2 inhibition in cells using a novel, selective mPGES-1 inhibitor. *Biochem Pharmacol.* May 15; 2010 79(10):1445–1454. [PubMed: 20067770]

Figure 1.

Effect of mPGES-1 deficiency in intracranial aneurysm rupture. A: Survival of WT and mPGES-1 KO mice after induction of aneurysms. B: Formation of aneurysms in WT and mPGES-1 KO mice. C: Prevalence of subarachnoid hemorrhage was markedly higher in mPGES-1 KO mice than in WT mice.

Figure 2. Intracranial aneurysms in mice. Example of unruptured aneurysm in a WT mouse (left) and a ruptured aneurysm in a mPGES-1 KO mouse (right). Scale bar=1mm.

Figure 3.

Effect of leukocyte-derived mPGES-1 on intracranial aneurysm formation and rupture. A: Survival of WT and mPGES-1 KO recipient mice that received WT or KO bone marrow. B: Formation of aneurysms in WT and mPGES-1 KO transplanted mice. C: Prevalence of subarachnoid hemorrhage in WT and mPGES-1 KO transplanted mice

Figure 4.

Effect of PGE₂ (1mg/kg/d) infusion in mPGES-1 KO mice. A: Survival of mPGES-1 KO mice after infusion of PGE₂ or vehicle (10% ethanol). B: Formation of aneurysms in mPGES-1 KO mice infused with vehicle or PGE₂. C: Prevalence of subarachnoid hemorrhage in mPGES-1 KO mice infused with vehicle or PGE₂.

Figure 5.

Effect of aspirin (6mg/kg/d) treatment in mPGES-1 KO mice. Mice received aspirin from day 2 after surgery until death. A: Survival was increased in mPGES-1 KO mice treated with aspirin compared to mPGES-1 KO mice treated with vehicle (water) by gavage. B: Similar prevalence of aneurysms in mPGES-1 KO mice treated with aspirin or vehicle. C: Incidence of subarachnoid hemorrhage in mPGES-1 KO mice treated with aspirin or vehicle.