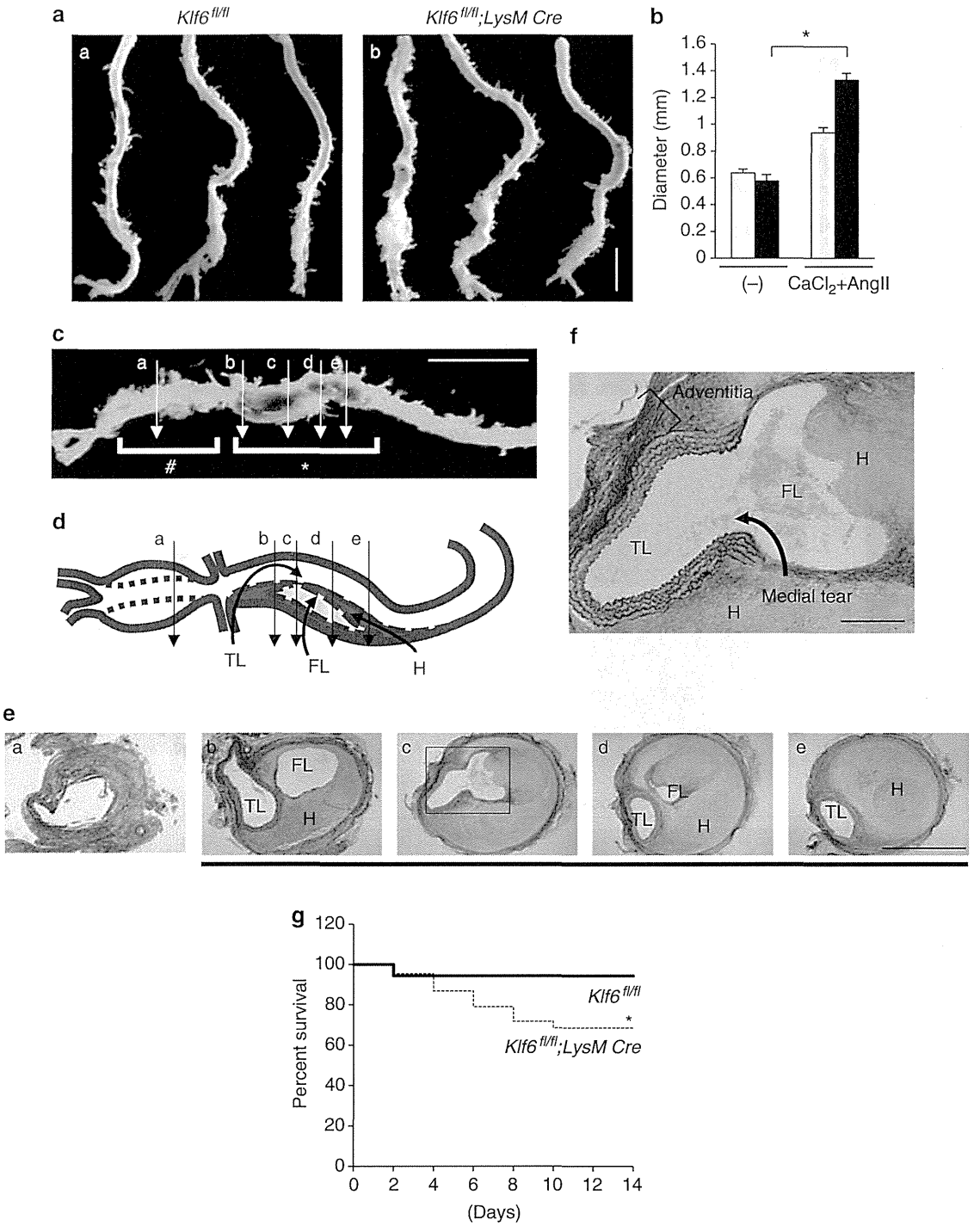



**Figure 2 | Myeloid *Klf6*-deficient mice show aortic dissection/haematoma.** (**a**) Representative aorta of *Klf6*$^{fl/fl}$ control mice (**a**) and *Klf6*$^{fl/fl}$;*LysM Cre* mice (**b**) after 2 weeks of AngII infusion with $CaCl_2$ application. Scale bar, 5 mm. (**b**) Quantification of infrarenal aortic diameters before ((−), $n=3$) and after 2 weeks of AngII infusion with $CaCl_2$ application ($CaCl_2$ + AngII, $n=5$). Results are from three independent experiments. All values are presented as means ± s.e.m. *$P<0.05$, Student's *t*-test. (**c**) Thoracic-abdominal aorta subjected to $CaCl_2$ application and AngII infusion (infrarenal aorta: hash, suprarenal aorta: asterisk). Note that intramural thrombus formation is present in the suprarenal region. Scale bar, 5 mm. (**d**) Schematic illustration of the diseased aorta (TL: true lumen, FL: false lumen, H: haematoma). (**e**) Cross-sectional histological sections stained by Elastica van Gieson. (a) Cross-section of the infrarenal abdominal aorta ($CaCl_2$ application region). (b) At the level of the renal arteries. (c) Suprarenal level where the intima-medial layer shows a tear. (d and e) Suprarenal descending thoracic aorta beyond the intima-medial tear. Scale bar, 1 mm. (**f**) High-magnification cross-section at the suprarenal level (**e**(c, boxed)). Intima-medial tear and false lumen/mural thrombus formation are present. Scale bar, 200 μm. (**g**) Survival curve between *Klf6*$^{fl/fl}$ control mice ($n=19$) and *Klf6*$^{fl/fl}$;*LysM Cre* mice ($n=22$) with $CaCl_2$ application and AngII infusion. *$P<0.05$, log-rank test.

macrophages (Fig. 5a). Macrophages obtained from the aorta of *Klf6*$^{fl/fl}$;*LysM Cre* mice showed markedly increased expression of *GM-CSF* under experimental conditions of $CaCl_2$ application and AngII infusion (Fig. 5b), and in macrophages derived from bone marrow of these mice (Supplementary Fig. 2a). Expression of *GM-CSF* in the aorta was elevated from 3 days after treatment

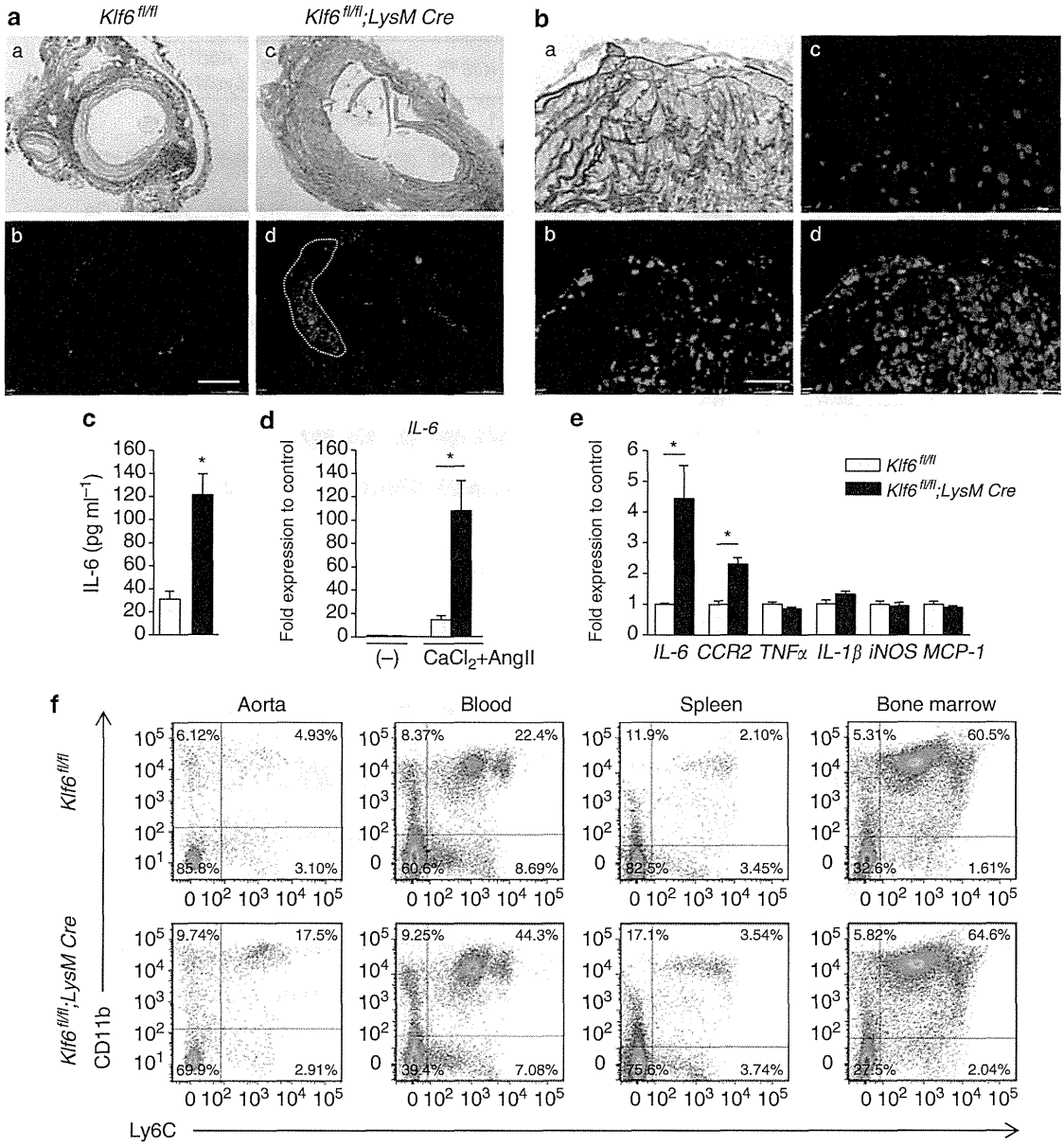



**Figure 3 | Marked inflammation in the aortic lesion of myeloid *Klf6*-deficient mice.** (**a**) Infiltrated macrophages were visualized by immunofluorescent staining (dotted line, green, Mac3) in aorta of *Klf6*$^{fl/fl}$;*LysM Cre* mice (right panels, c and d) compared with *Klf6*$^{fl/fl}$ mice (left panels, a and b). Scale bar, 100 μm. (**b**) Immunofluorescent staining for macrophages (b: green, Mac3), pSTAT3 (c: red) and nuclei (d: 4′, 6-diamidino-2-phenylindole, blue) in diseased aorta (a) of *Klf6*$^{fl/fl}$;*LysM Cre* mice. Scale bar, 20 μm. (**c**) Plasma concentration of IL-6 in *Klf6*$^{fl/fl}$ mice ($n=7$) and *Klf6*$^{fl/fl}$;*LysM Cre* mice ($n=9$) after 2 weeks of AngII infusion with $CaCl_2$ application. *$P<0.05$, Student's *t*-test. (**d**) Expression of RNA levels of *IL-6* were examined in aorta from *Klf6*$^{fl/fl}$ mice and *Klf6*$^{fl/fl}$;*LysM Cre* mice before (( − ), $n=3$) and after 2 weeks of AngII infusion with $CaCl_2$ application ($CaCl_2$ + AngII, $n=5$) using real-time PCR and normalized by *GAPDH* messenger RNA. (**e**) Expression of RNA levels of *IL-6*, *CCR2*, *TNFα*, *IL-1β*, *iNOS* and *MCP-1* were examined in bone-marrow-derived macrophages subjected to AngII stimulation (10 μM) for 3 h ($n=3$ mice per group). (**f**) Population of CD11b$^+$ Ly6C$^{hi}$ cells in aorta, peripheral blood, spleen and bone marrow in *Klf6*$^{fl/fl}$ and *Klf6*$^{fl/fl}$;*LysM Cre* mice after 2 weeks of AngII infusion with $CaCl_2$ application. Results represent three independent experiments. All values are presented as means ± s.e.m. *$P<0.05$, Mann-Whitney test (**d**,**e**).

(before onset of aortic dissection) of *Klf6*$^{fl/fl}$;*LysM Cre* mice (Fig. 5c). Whether deletion of KLF6 in macrophages affects secretion of GM-CSF and further systemic circulating levels was next asked. Macrophages and GM-CSF co-localized in the aorta of *Klf6*$^{fl/fl}$;*LysM Cre* mice, and GM-CSF was markedly produced by macrophages in response to pro-inflammatory stimuli (Fig. 5d; Supplementary Fig. 2b). Circulating levels of GM-CSF were at least 73.3-fold higher in *Klf6*-deleted mice (Fig. 5e). It therefore seems that a markedly increased response in GM-CSF is a hallmark feature of the aorta in *Klf6*$^{fl/fl}$;*LysM Cre* mice.

We next sought to understand mechanisms underlying regulation of GM-CSF expression and secretion by KLF6. Overexpression of *KLF6* significantly attenuated *GM-CSF* expression induced by pro-inflammatory stimuli in macrophages (Supplementary Fig. 2c). Transcriptionally, several KLF-binding sites were present in the promoter region of GM-CSF to which KLF6 was recruited by agonistic stimuli treatment in macrophages (Supplementary Fig. 2d). These results demonstrated that, mechanistically, GM-CSF is a direct target of KLF6 and that KLF6 represses expression of GM-CSF.

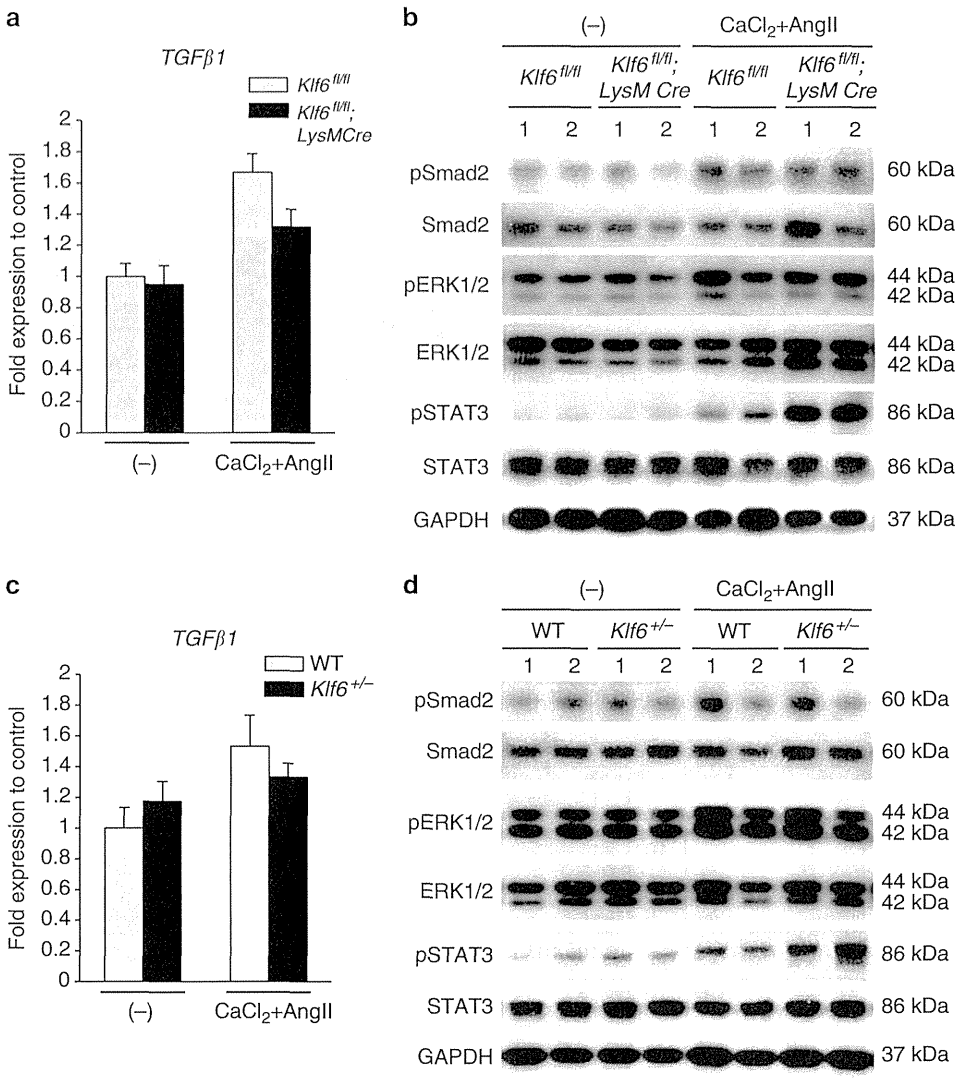



**Figure 4 | Involvement of TGFβ pathways in aortic dissection/haematoma.** Expression of messenger RNA (mRNA) levels of TGFβ1-related factors in aorta from *Klf6*$^{fl/fl}$ and *Klf6*$^{fl/fl}$*;LysM Cre* mice (**a**), and in wild-type (WT) littermates and *Klf6*$^{+/-}$ mice (**c**), using real-time PCR normalized by *GAPDH* mRNA. $n=5$ per group. All values are presented as mean ± s.e.m. Western blot analysis for pSmad2, Smad2, pERK1/2, ERK1/2, pSTAT3, STAT3 or GAPDH in aorta before (−) and after 2 weeks of AngII infusion with $CaCl_2$ application ($CaCl_2$ + AngII) in *Klf6*$^{fl/fl}$ and *Klf6*$^{fl/fl}$*;LysM Cre* mice (**b**) and WT littermates and *Klf6*$^{+/-}$ mice (**d**). Results represent three independent experiments.

**GM-CSF manipulation regulates aortic dissection/haematoma.** To next test the requirement of GM-CSF in aortic dissection in these mice, the actions of GM-CSF were blocked using a neutralizing antibody, which abrogated aortic dissection/intramural haematoma (Fig. 6a,b) as well as expression of *GM-CSF* receptor α, *MMP9*, *F4/80* and *IL-6* (Fig. 6d) in addition to serum levels of IL-6 (Fig. 6c). GM-CSF was therefore required for the aortic phenotype in *Klf6*$^{fl/fl}$*;LysM Cre* mice.

We further investigated whether GM-CSF is sufficient to induce aortopathy. Administration of GM-CSF in wild-type mice subjected to aortic inflammation ($CaCl_2$ + AngII) caused aortic dissection/intramural haematoma, confirming the generality of the role of GM-CSF in the pathogenesis of the condition. Mice died from aortic rupture due to the aortic lesion and showed pathological features of the condition (for example, fragile aorta, intimal tear with haematoma) (Fig. 6e–h,j). However, aortic dissection/intramural haematoma did not develop by administration of GM-CSF alone, even with abnormally increased circulating levels (at least 180.9-fold) of GM-CSF (Supplementary Fig. 3a,b). As AngII, $CaCl_2$ or GM-CSF alone was not sufficient to induce the condition, it seems that a combination of aortic inflammation with GM-CSF infusion is necessary for the phenotype (Supplementary Fig. 4a,b). Consistent with this, circulating levels of GM-CSF in mice were only markedly elevated when treated with the combination of measures as compared with each alone (Fig. 6i). Note that these elevated levels were comparable to those in *Klf6*$^{fl/fl}$*;LysM Cre* mice, suggesting that highly elevated levels of GM-CSF are required but not sufficient to cause aortic dissection/intramural haematoma (Fig. 5e).

Finally, whether manipulation of GM-CSF affects the number of peripheral leukocytes was examined. With GM-CSF administration, the number of circulating lymphocytes did not change in either the early phase (5 days) or developed phase (14 days) of the model (Supplementary Tables 2 and 3). With respect to neutrophils, the number in peripheral blood was markedly increased in the early phase, but no difference was observed at 14 days of GM-CSF administration. This was similarly seen in the group in which GM-CSF alone was administered, which did not result in the aortic phenotype. While these changes might be due to acute effects by exogenous GM-CSF treatment, this alone

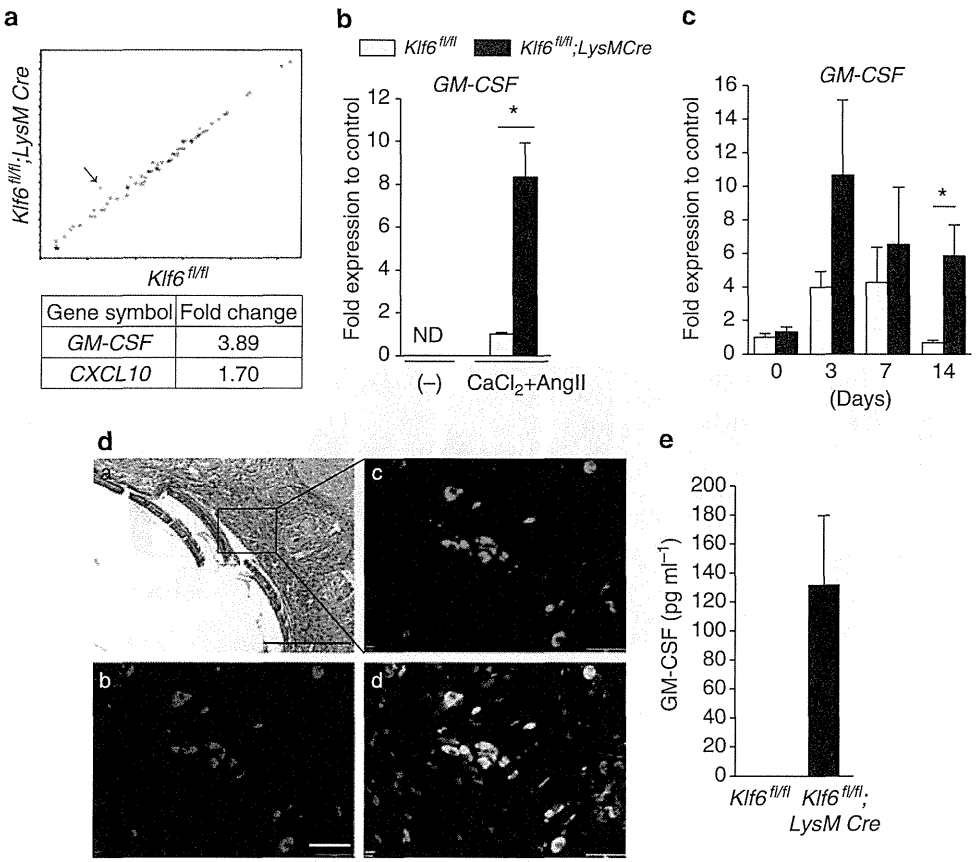



**Figure 5 | GM-CSF is a direct target of KLF6 in macrophages.** (**a**) RT2 profiler PCR array analysis of genes related to IL-6/STAT3 inflammatory pathway between bone marrow (BM)-derived macrophages from *Klf6*$^{fl/fl}$ and *Klf6*$^{fl/fl}$*;LysM Cre* mice with AngII stimulation (10 μM) for 3 h. Arrow indicates *GM-CSF*. List of genes that showed consistent changes between BM-derived macrophages from *Klf6*$^{fl/fl}$ and *Klf6*$^{fl/fl}$*;LysM Cre* mice stimulated with AngII (10 μM) for 3 h. (**b**) Messenger RNA (mRNA) expression of *GM-CSF* in aortic macrophages obtained from *Klf6*$^{fl/fl}$ (sham; (−), $n=3$; $CaCl_2$ + AngII; $n=3$) and *Klf6*$^{fl/fl}$*;LysM Cre* mice (sham; (−), $n=3$; $CaCl_2$ + AngII; $n=6$). ND indicates not detected. (**c**) mRNA expression of *GM-CSF* in aorta of *Klf6*$^{fl/fl}$ and *Klf6*$^{fl/fl}$*;LysM Cre* mice at 0 ($n=3$), 3 ($n=3$), 7 ($n=3$) and 14 ($n=4$) days. (**d**) Immunohistochemistry for macrophages (red: F4/80, b; scale bar, 30 μm), GM-CSF (green: c) and nucleus (blue: 4′, 6-diamidino-2-phenylindole, d) in aorta of *Klf6*$^{fl/fl}$*;LysM Cre* mice with EVG-stained infrarenal aorta (a, scale bar, 200 μm). (**e**) Plasma GM-CSF concentration between *Klf6*$^{fl/fl}$ ($n=8$) and *Klf6*$^{fl/fl}$*;LysM Cre* mice ($n=4$) after 2 weeks of AngII infusion with $CaCl_2$ application. Results represent three independent experiments. All values are presented as means ± s.e.m. *$P<0.05$, Mann-Whitney test.

had no bearing on the phenotype. Moreover, the number of circulating granulocytes and lymphocytes was not affected when GM-CSF was depleted by the neutralizing antibody (Supplementary Table 4). On the basis of these results, manipulation of GM-CSF did not affect the number of circulating leukocytes in the present model, at least during the observation period (14 days).

**Upregulation of GM-CSF in patients with aortic dissection.** To confirm the clinical relevance of our findings, circulating levels of GM-CSF were measured in sera of patients with acute aortic dissection, which showed marked increases, in contrast to patients with coronary artery disease, aortic aneurysm or healthy volunteers, which showed markedly lower if not negligible levels (Fig. 7a). Furthermore, inflammatory infiltration ($CD68^+$ monocytes/macrophage) and GM-CSF expression were upregulated and co-localized in dissected aorta (Fig. 7b). Thus, GM-CSF is associated with aortic acute dissection not only in mice but also in human conditions.

## Discussion

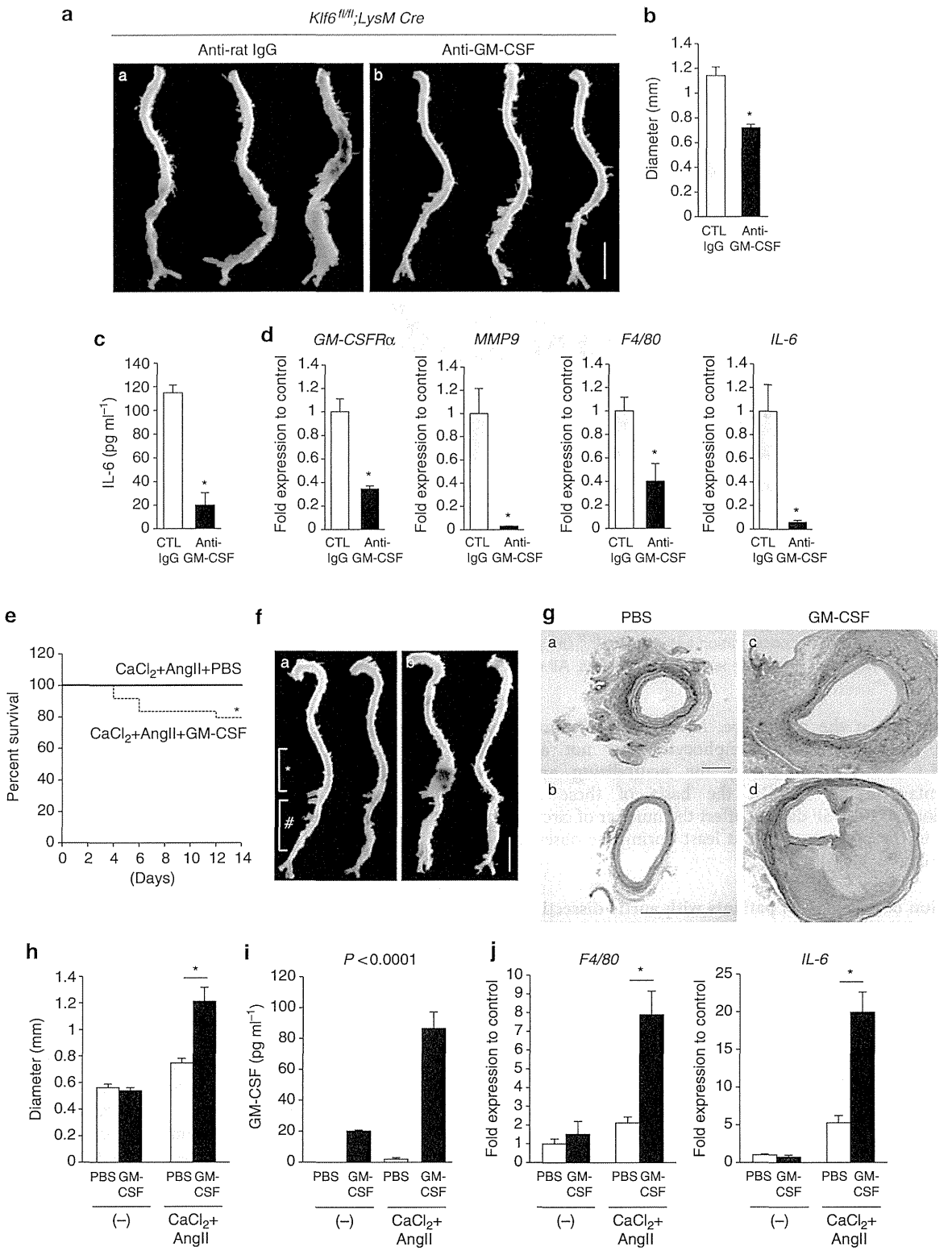
The present findings show that GM-CSF is a key regulatory molecule causative of aortic dissection/intramural haematoma in a murine model of the condition and to also be associated with the condition in humans. In mice, modulation of GM-CSF by a neutralizing antibody or exogenous administration, respectively, prevented or induced onset of this phenotype. In humans, elevated serum GM-CSF levels and expression of the cytokine in aortic tissue were seen in patients with aortic dissection.

GM-CSF was a central component of the aortic dissection/intramural haematoma phenotype in our murine model. Previous studies had suggested a limited role of GM-CSF in the pathogenesis of aortic disease[36–39]. For example, mice that lack *Smad3* manifested a phenotype of aortic aneurysm formation[39], and GM-CSF was shown to play a pivotal role in the pathogenesis; however, it was assumed that because smad3 is a downstream target of TGFβ, which is a central molecule associated in Marfan aortopathy, the pathogenic mechanism was limited to this genetic aortopathy. Our findings show that activation of the GM-CSF pathway in a manner independent of the TGFβ-SMAD pathway is sufficient to trigger this condition in a model of inflammatory and degenerative aorta (calcium chloride treatment causes stiffening of the aorta to mimic the condition as seen in atherosclerotic human aortas[40]). This model is reflective of aortic dissection/intramural haematoma seen in the elderly adult in humans and should be differentiated from the genetic aortopathy in young patients with Marfan syndrome.. GM-CSF tissue expression had also been shown to be increased in

a patient presenting with aortic dissection in Cogan's disease[41], an apparently autoimmune condition that is characterized by recurrent corneal inflammation[42], which was thought to be an isolated finding.

Effects on other non-macrophage myeloid cells were investigated, which showed that dendritic cells ($CD11c^+MHCII^+$ cells) were increased in the diseased aorta but not in the circulation under KLF6-deficient conditions, and lack of effects on neutrophils ($Ly6G^+$ cells) either in the circulation or in the aortic tissue (Supplementary Fig. 1; Supplementary Fig. 5). The contributory role of non-macrophage myeloid cells (for example, dendritic cells) needs to be further investigated.

Macrophage colony-stimulating factor (M-CSF) has been also suggested to be an important regulator of vascular remodelling[43,44]. Although the precise molecular mechanisms of the actions of M-CSF are still unclear, different actions as



compared with GM-CSF are envisioned, given different expression patterns in the vascular wall. Whereas M-CSF is constitutively expressed under physiological conditions in endothelial cells, fibroblasts, macrophages and smooth muscle cells, GM-CSF, by contrast, is expressed only in minute amounts in these cells under basal conditions, but instead is induced by inflammatory stimuli (for example, tumour necrosis factor)[45] or oxidized low-density lipoprotein cholesterol stimulation[46]. In murine and human lesions, M-CSF is detected both in healthy arteries and in atherosclerotic lesions associated with macrophage and foam cell content, and is correlated with plaque progression in the latter. By contrast, only minute levels of GM-CSF are seen in smooth muscle cells and endothelial cells of healthy human arteries, but are elevated upon atherosclerotic development and macrophage accumulation[47]. On the basis of these observations, collectively, while M-CSF is a constitutively expressed cytokine in the vasculature, GM-CSF is markedly induced in diseased vessels to regulate pathological conditions including the described aortopathy.

In the experimental model, most previous studies have used AngII infusion alone as an intervention to induce a dissection phenotype[16,48]. However, the limitation of this procedure for mechanistic investigations including inflammation was the low reproducibility (less than 30%), need for long-term infusion of AngII (more than 4 weeks) and incidence/expression of phenotype in aged mice (over 7–10 months age) with a specific genetic background (*apoE*$^{-/-}$ or *ldl receptor*$^{-/-}$ mice). Most noteworthy is that the present model could induce aortic dissection/intramural haematoma within 2 weeks with high reproducibility (at least 70%) even in young wild-type mice. Mechanistically, this model might involve hemodynamic stress on the suprarenal dissection site due to loss of Windkessel effect[49] because of increased stiffening in the infrarenal aorta (for example, downward shift of the pressure-diameter curve after $CaCl_2$ application with continuous AngII infusion)[40] that showed aneurysmal formation, which when exposed to inflammatory effects of GM-CSF triggered dissection/intramural haematoma formation in the weak and fragile suprarenal aorta. As aortic aneurysm is commonly co-present in patients with dissection[4], the described animal model and findings closely resemble the condition seen in patients.

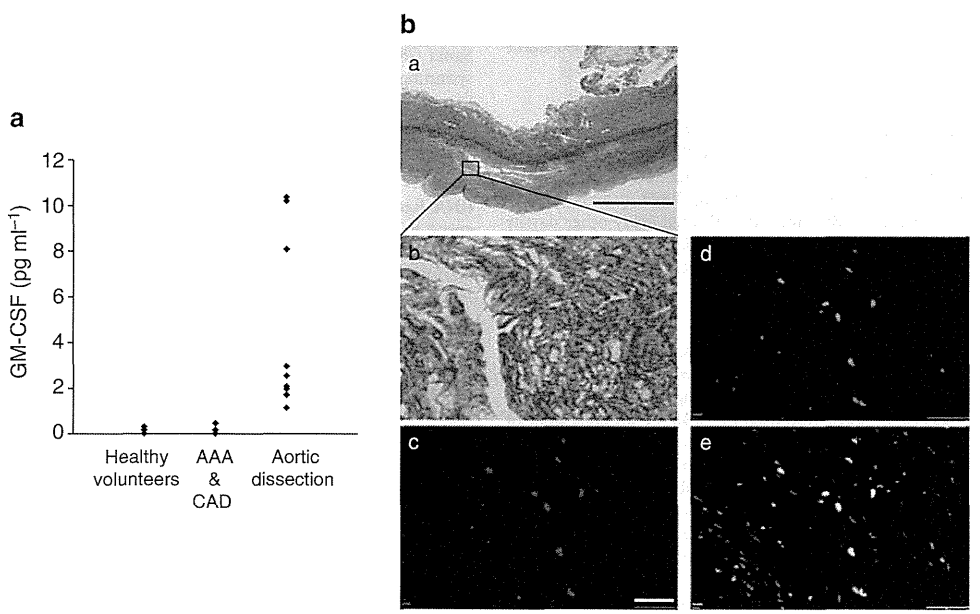



**Figure 7 | Increased GM-CSF in patients with acute aortic dissection.** (**a**) Plasma GM-CSF concentration in healthy volunteers ($n=12$) and patients with aortic aneurysm (AAA, $n=3$), coronary artery disease (CAD, $n=11$) or aortic dissection ($n=10$). (**b**) Immunofluorescent staining for CD68 (red, c, scale bar, 50 μm), GM-CSF (green, d) and 4′,6-diamidino-2-phenylindole (blue, e) in descending dissected aorta (boxed area, a, scale bar, 2 mm) with EVG staining (b). Results represent three independent experiments.

**Figure 6 | GM-CSF is required for aortic dissection/intramural haematoma.** (**a**) Representative aortas of *Klf6*$^{fl/fl}$*;LysM Cre* mice with administration of anti-GM-CSF neutralizing antibody (b, anti-GM-CSF, $n=8$) or control IgG antibody (a, $n=10$) after 2 weeks of AngII infusion with $CaCl_2$ application. Scale bar, 5 mm. Quantification of infrarenal aortic diameters (**b**, anti-GM-SCF: $n=7$; anti-control IgG: $n=9$) and plasma concentration of IL-6 (**c**, $n=5$ or 6) between anti-GM-CSF antibody-administered and anti-control IgG-administered mice. *$P<0.05$, Student's *t*-test. (**d**) Expression levels of RNA of *GM-CSFRα*, *MMP9*, *F4/80* and *IL-6* were examined in aorta of anti-GM-CSF antibody-administered mice or anti-control IgG-administered mice using real-time PCR and then normalized by *GAPDH* messenger RNA (mRNA) ($n=5$ mice per group). (**e**) Survival curve of mice with administration of recombinant GM-CSF ($n=26$) or PBS ($n=19$) with $CaCl_2$ application and AngII infusion in wild-type mice. *$P<0.05$, log-rank test. (**f**) Representative aorta of wild-type mice with administration of recombinant GM-CSF (**b**) or PBS (**a**) with $CaCl_2$ application and AngII infusion (infrarenal aorta: hash, suprarenal aorta: asterisk) for 4 weeks. Scale bar, 5 mm. (**g**) Histopathological analysis of infrarenal aorta (upper panels: a and c, scale bar, 200 μm) and suprarenal aorta (lower panels: b and d, scale bar, 1 mm) by EVG staining. (**h**) Quantification of infrarenal aortic diameters between recombinant GM-CSF-administered mice or PBS-administered mice (sham; (−), $n=3$, $CaCl_2$ + AngII; $n=5$). (**i**) Plasma GM-CSF concentration after 2 weeks infusion of recombinant GM-CSF or PBS with or without $CaCl_2$ application and AngII infusion ($n=3\sim5$ mice per group). (**j**) Expression levels of RNA of *F4/80* and *IL-6* were examined in aorta from mice administered recombinant GM-CSF or PBS using real-time PCR and then normalized with *GAPDH* mRNA (sham; (−), $n=3$, $CaCl_2$ + AngII; $n=5$). Results are from three independent experiments. All values are presented as means ± s.e.m. *$P<0.05$, Mann–Whitney test (**d**,**h**,**j**) and one-way analysis of variance with Dunn's post-test (**i**).

Taken together, our findings suggest that GM-CSF is a central regulator of aortic dissection/intramural haematoma in the atherosclerotic and inflammatory aorta, which is typically seen in the elderly patient with this condition, and may serve as a potential target for diagnostic and therapeutic exploitation (for example, aortic stabilization using GM-CSF antagonists), as well as a diagnostic biomarker.

## Methods

**Mice.** Heterozygous *Klf6*$^{+/-}$ mice (C57BL/6) were originally generated by Tarocchi *et al.*[50] To generate macrophage-specific *Klf6*-knockout mice, *Klf6*$^{fl/fl}$ mice (C57BL/6;129Sv) were cross-bred with *LysM Cre* mice (C57BL/6, Jackson laboratory)[51]. Only male mice, 10- to 13-weeks of age, and C57BL/6 as wild-type mouse (CLEA Japan) were used. All experimental protocols were approved by the Ethics Committee for Animal Experimentation at the Graduate School of Medicine, the University of Tokyo, and conducted in accordance with the Guidelines for the Care and Use of Laboratory Animals of the Department of Medicine, the University of Tokyo.

**Murine aortic dissection/intramural haematoma model.** To induce aortic dissection/intramural haematoma, peri-aortic application of $CaCl_2$ was performed on the abdominal aorta, followed by a 2-week infusion of AngII (2,000 ng kg$^{-1}$ min$^{-1}$)[40]. In detail, mice were anaesthetized and underwent laparotomy at 10–13 weeks of age. The abdominal aorta between the renal arteries and bifurcation of the iliac arteries was isolated from the surrounding retroperitoneal structure, and 0.5 M $CaCl_2$ was applied to the external surface of the infrarenal aorta. NaCl (0.9%) was substituted for $CaCl_2$ in sham control mice. The aorta was rinsed with 0.9% sterile saline after 15 min and the incision was closed.

**Macrophage depletion and manipulation of GM-CSF.** Wild-type mice were injected intraperitoneally with 110 mg kg$^{-1}$ of clodronate liposomes or equal volume of PBS liposomes 2 days prior and 7 days after induction of aortic dissection. Neutralizing antibody against GM-CSF (300 μg, R&D systems) or control anti-rat IgG antibody (Equitech Bio) was administered every other day by intraperitoneal injection. Recombinant murine GM-CSF (10, 50 and 100 μg kg$^{-1}$ per day, PeproTech) was administered for 2 or 4 weeks after induction of aortic dissection.

**Histological analysis and immunohistochemistry.** Aortas from mice were embedded in paraffin and then 5-μm-thick serial sections were prepared for Elastic Van Gienson (EVG) and haematoxylin/eosin staining. Digital images of EVG-stained aortas with a reference scale were used for absolute measurement of diameter. Human aortic tissue was obtained from patients undergoing surgical aortic repair with informed consent under a protocol approved by the University of Tokyo Hospital Research Ethics Committee. Paraffin-embedded sections were taken from the aorta for EVG staining and immunohistochemistry. For immunohistochemistry, after deparaffinization and blocking, serial sections were incubated with the following antibodies: Mac3 (dilution 1:200; rat; BD Pharmingen) or F4/80 (1:100; rat; Serotec) for macrophages in mice and CD68 (1:50; mouse; Dako) in humans and GM-CSF (1:100, rabbit; Abcam for mouse and 1:50; rabbit; Acris for humans) or pSTAT3 (1:200; rabbit; Cell Signaling Technology), followed by biotinylated secondary antibodies (1:200; Dako). For detection, anti-streptavidin-conjugated AlexFluor 488 or AlexFluor 594 (1:200; Invitrogen) was used. The nuclei were stained with 4′, 6-diamidino-2-phenylindole (1:5,000; Sigma-Aldrich) after the final series of washes.

**Cell preparation from aorta, spleen, bone marrow and blood.** Aortas were minced into 3- to 4-mm pieces and placed in 1 ml digestion solution containing collagenase type II (1.25 mg ml$^{-1}$, Worthington) and porcine pancreatic elastase (50 μg ml$^{-1}$, Worthington) in a base solution of Accumax (Innovative Cell Technologies). Aortic tissue was digested at room temperature with agitation for 1 h. After digestion, cells were washed in FACS buffer (5% FCS in PBS) at 2,000 r.p.m. for 5 min (ref. 16). Aortic macrophages were isolated using CD11b microbeads according to the manufacturer's instructions (Miltenyi Biotec). Spleen was homogenized and passed through a cell strainer to obtain single-cell suspensions. Bone-marrow-derived cells were taken from the femur and tibia of 5–6-week-old mice. Blood was collected in heparin-coated vials and then 1.2% dextran was added for 45 min at room temperature. Counting of peripheral leukocytes was done by an automated hematology analyzer (XT-2,000i, Sysmex). Neutrophils were isolated from bone marrow using a neurophil isolation kit according to the manufacturer's instructions (Miltenyi Biotec). From single-cell suspensions of spleen, bone marrow and blood, erythrocytes were lysed using ACK lysis buffer for 5, 3 and 2 min on ice, respectively. Cells were centrifuged at 2,000 r.p.m. for 5 min to remove the ACK lysis buffer, then the single-cell suspensions were resuspended and washed in FACS buffer, followed by centrifugation at 2,000 r.p.m. for 5 min.

**Cell cultures.** Bone-marrow-derived cells were prepared from femur and tibia of *Klf6*$^{fl/fl}$ mice or *Klf6*$^{fl/fl}$;*LysM Cre* mice to assess the role of GM-CSF in macrophages. KLF6 overexpression was induced by a retrovirus construct for KLF6 (pMXs-KLF6) in the presence of RetroNectin (5 μg cm$^{-2}$, Takara Bio.).

**Flow cytometry.** Murine Fc receptors were blocked using antibodies against murine CD16/32 antigens (eBioscience) for 15 min on ice, after which cells were washed and then resuspended in 100 μl FACS buffer. Fluorochrome-conjugated antibodies (all from BioLegend) for APC-CD11b[M1/70], PerCP-Cy5.5-Ly-6c[HK1.4], APC-Cy7-Ly6G[1A8] or APC-CD11c[N418] were added for 30–45 min at room temperature. FITC-CD3e[145-2C11], FITC-Ly6G[RB6-8C5], FITC-CD11b[M1/70], FITC-CD45R/B220[RA3-6B2] and FITC-Ly76[Ter-119] (erythroid lineage marker) were used as lineage markers. Corresponding isotype control antibodies were added to samples at the same concentrations as the antibodies of interest. After incubation, samples were washed three times and analysed by FACSverse (BD Pharmingen). Compensation was done using positive samples containing single-colour-stained aortic macrophages. Debris and dead cells, as defined by low forward scatter, were excluded from analysis. Data were analysed with FlowJo (Tree Star) (ref. 16).

**Chromatin immunoprecipitation.** Chromatin immunoprecipitation analysis was performed using a Chromatin Immunoprecipitation Kit (Active Motif) according to the manufacturer's instructions. Briefly, bone-marrow-derived macrophages were stimulated with or without AngII (10 μM), TNFα (10 ng ml$^{-1}$) and IL-1β (20 ng ml$^{-1}$) for 3 h prior to crosslinking for 10 min with 1% formaldehyde. Chromatin was sheared by sonication to an average size of 200 ~ 1,000 base pairs (Covaris). Immunoprecipitation was performed using anti-KLF6 antibody (25 ng μl$^{-1}$, Santa Cruz Biotechnology) and rabbit IgG antibody (25 ng μl$^{-1}$, Santa Cruz Biotechnology). PCR amplification of the GM-CSF promoter region spanning KLF-binding elements was performed using the following primers: forward: 5′-AAGC CCTTCCAAGAACTGGC-3′ and reverse 5′-GGCCCCTCAAA AAGGAGAGG-3′. KLF6 recruitment was normalized by input DNA and compared with the control group using KLF6 antibody.

**RNA isolation and quantitative real-time PCR.** Total RNA from cultured cells, aortic macrophages, bone-marrow-derived neutrophils or murine aortic samples was extracted using either the RNeasy minikit (Qiagen) or RNA*later* (Qiagen) according to the manufacturer's instructions. In total, 0.5–1 μg RNA was reverse-transcribed using Superscript III (Invitrogen) according to the manufacturer's instructions. Real-time PCRs were performed using 2 μl of resulting cDNA per 20-μl reaction volume containing SYBR green I master (Roche). Glyceraldehyde 3-phosphate dehydrogenase was used as an internal control. Using bone-marrow-derived macrophages with AngII (10 μM, 3 h) stimulation, RT2 profiler PCR array (Qiagen) was performed with 84 related genes for the IL-6/STAT inflammatory pathway. PCR was performed on a LightCycler 480 Real-time PCR system (Roche) in accordance with the manufacturer's recommended procedure. Real-time PCR primers are shown in Supplementary Table 5.

**Western blot analysis.** Mouse aortic specimens were homogenized with lysis buffer (T-PER, Thermo Scientific) containing the protease inhibitor complex (Roche) and phosphatase inhibitors (Roche). Protein concentration was assayed using the BCA protein assay kit (Pierce), and 5 μg of the protein were resolved by 10% NuPAGE (Invitrogen) and then transferred to polyvinylidene difluoride membrane. The blot was probed with primary antibodies: pSmad2 (dilution 1:400), pERK1/2 (1:3,000), pSTAT3 (1:3,000), Smad2 (1:1,000), ERK1/2 (1:3,000) or STAT3 (1:3,000) (all rabbit antibodies obtained from Cell Signaling Technology) and anti-GAPDH antibody (1:1,000, Ambion). Membranes were washed and incubated with the corresponding horseradish peroxidase-conjugated secondary antibody (Cell Signaling Technology). Protein bands were detected by ECLplus (Thermo scientific) and GAPDH served as an internal control for protein loading. The original blots for the representative images are displayed in Supplementary Fig. 6.

**Enzyme-linked immunosorbent assay.** Plasma levels of IL-6, MCP-1 and GM-CSF in mice or in humans with or without aortic dissection/intramural haematoma were assayed with commercially available quantikine ELISA kits (R&D systems) according to the manufacturer's instructions. Sera of healthy volunteers and of patients with aortic aneurysm, coronary artery disease or with aortic dissection were obtained with informed consent under a protocol approved by the University of Tokyo Hospital Research Ethics Committee. Baseline characteristics of human subjects are shown in Supplementary Table 6.

**Statistical analyses.** All data are presented as means ± s.e.m. Statistical difference between two groups was determined using the Student's *t*-test (two-tailed) for parametric data or the Mann–Whitney test for non-parametric data after testing for normality by F-test analysis. For data containing multiple time points, two group comparisons at the same time point were done. When comparing multiple

groups, data were analysed by the Kruskal–Wallis non-parametric one-way analysis of variance with Dunn's post-test. Survival curves were created using the Kaplan–Meier method and compared by a log-rank test. Statistical power for mouse experiments was calculated using Biomath (biomath.info/power). All samples sizes were equal to or greater than the recommended minimum group size. All data were analysed using Prism 6.0 (GraphPad Software). A $P$ value of less than 0.05 was considered significant.

## References


1. Nienaber, C. A. & Powell, J. T. Management of acute aortic syndromes. *Eur. Heart J.* **33,** 26–35b (2012).
2. Tsai, T. T., Nienaber, C. A. & Eagle, K. A. Acute aortic syndromes. *Circulation* **112,** 3802–3813 (2005).
3. Hiratzka, L. F. *et al.* 2010 ACCF/AHA/AATS/ACR/ASA/SCA/SCAI/SIR/STS/SVM guidelines for the diagnosis and management of patients with thoracic aortic disease. A Report of the American College of Cardiology Foundation/American Heart Association Task Force on Practice Guidelines, American Association for Thoracic Surgery, American College of Radiology, American Stroke Association, Society of Cardiovascular Anesthesiologists, Society for Cardiovascular Angiography and Interventions, Society of Interventional Radiology, Society of Thoracic Surgeons, and Society for Vascular Medicine. *J. Am. Coll. Cardiol.* **55,** e27–e129 (2010).
4. Hagan, P. G. *et al.* The International Registry of Acute Aortic Dissection (IRAD): new insights into an old disease. *JAMA* **283,** 897–903 (2000).
5. Suzuki, T. *et al.* Biomarkers of aortic diseases. *Am. Heart J.* **165,** 15–25 (2013).
6. Suzuki, T., Distante, A. & Eagle, K. Biomarker-assisted diagnosis of acute aortic dissection: how far we have come and what to expect. *Curr. Opin. Cardiol.* **25,** 541–545 (2010).
7. Clough, R. E. & Nienaber, C. A. Management of acute aortic syndrome. *Nat. Rev. Cardiol.* **12,** 103–114 (2014).
8. Song, J. K. Aortic intramural hematoma: aspects of pathogenesis 2011. *Herz* **36,** 488–497 (2011).
9. von Kodolitsch, Y. *et al.* Intramural hematoma of the aorta: predictors of progression to dissection and rupture. *Circulation* **107,** 1158–1163 (2003).
10. Milewicz, D. M., Regalado, E. S. & Guo, D. C. Treatment guidelines for thoracic aortic aneurysms and dissections based on the underlying causative gene. *J. Thorac Cardiovasc. Surg.* **140,** S2–S4 discussion S45–51 (2010).
11. Milewicz, D. M. *et al.* Genetic basis of thoracic aortic aneurysms and dissections: focus on smooth muscle cell contractile dysfunction. *Annu. Rev. Genomics Hum. Genet.* **9,** 283–302 (2008).
12. Holm, T. M. *et al.* Noncanonical TGFbeta signaling contributes to aortic aneurysm progression in Marfan syndrome mice. *Science* **332,** 358–361 (2011).
13. Li, W. *et al.* Tgfbr2 disruption in postnatal smooth muscle impairs aortic wall homeostasis. *J. Clin. Invest.* **124,** 755–767 (2014).
14. Dietz, H. C. TGF-beta in the pathogenesis and prevention of disease: a matter of aneurysmic proportions. *J. Clin. Invest.* **120,** 403–407 (2010).
15. Ju, X. *et al.* Interleukin-6-signal transducer and activator of transcription-3 signaling mediates aortic dissections induced by angiotensin II via the T-helper lymphocyte 17-interleukin 17 axis in C57BL/6 mice. *Arterioscler. Thromb. Vasc. Biol.* **33,** 1612–1621 (2013).
16. Tieu, B. C. *et al.* An adventitial IL-6/MCP1 amplification loop accelerates macrophage-mediated vascular inflammation leading to aortic dissection in mice. *J. Clin. Invest.* **119,** 3637–3651 (2009).
17. Date, D. *et al.* Kruppel-like transcription factor 6 regulates inflammatory macrophage polarization. *J. Biol. Chem.* **289,** 10318–10329 (2014).
18. Starkel, P. *et al.* Oxidative stress, KLF6 and transforming growth factor-beta up-regulation differentiate non-alcoholic steatohepatitis progressing to fibrosis from uncomplicated steatosis in rats. *J. Hepatol.* **39,** 538–546 (2003).
19. Holian, J. *et al.* Role of Kruppel-like factor 6 in transforming growth factor-beta1-induced epithelial-mesenchymal transition of proximal tubule cells. *Am. J. Physiol. Renal Physiol.* **295,** F1388–F1396 (2008).
20. Hamilton, J. A. & Anderson, G. P. GM-CSF biology. *Growth Factors* **22,** 225–231 (2004).
21. Daugherty, A., Manning, M. W. & Cassis, L. A. Angiotensin II promotes atherosclerotic lesions and aneurysms in apolipoprotein E-deficient mice. *J. Clin. Invest.* **105,** 1605–1612 (2000).
22. Johnston, K. W. *et al.* Suggested standards for reporting on arterial aneurysms. Subcommittee on Reporting Standards for Arterial Aneurysms, Ad Hoc Committee on Reporting Standards, Society for Vascular Surgery and North American Chapter, International Society for Cardiovascular Surgery. *J. Vasc. Surg.* **13,** 452–458 (1991).
23. Nagasawa, A. *et al.* Important role of the angiotensin II pathway in producing matrix metalloproteinase-9 in human thoracic aortic aneurysms. *J. Surg. Res.* **183,** 472–477 (2013).
24. Ezekowitz, R. A. & Gordon, S. Down-regulation of mannosyl receptor-mediated endocytosis and antigen F4/80 in bacillus Calmette-Guerin-activated mouse macrophages. Role of T lymphocytes and lymphokines. *J. Exp. Med.* **155,** 1623–1637 (1982).
25. Ezekowitz, R. A., Austyn, J., Stahl, P. D. & Gordon, S. Surface properties of bacillus Calmette-Guerin-activated mouse macrophages. Reduced expression of mannose-specific endocytosis, Fc receptors, and antigen F4/80 accompanies induction of Ia. *J. Exp. Med.* **154,** 60–76 (1981).
26. Atreya, R. *et al.* Blockade of interleukin 6 trans signaling suppresses T-cell resistance against apoptosis in chronic intestinal inflammation: evidence in crohn disease and experimental colitis in vivo. *Nat. Med.* **6,** 583–588 (2000).
27. Cheuk, B. L. & Cheng, S. W. Can local secretion of prostaglandin E2, thromboxane B2, and interleukin-6 play a role in ruptured abdominal aortic aneurysm? *World J. Surg.* **32,** 55–61 (2008).
28. Dawson, J. *et al.* Aortic aneurysms secrete interleukin-6 into the circulation. *J. Vasc. Surg.* **45,** 350–356 (2007).
29. Dawson, J., Cockerill, G., Choke, E., Loftus, I. & Thompson, M. M. Aortic aneurysms as a source of circulating interleukin-6. *Ann. N. Y. Acad. Sci.* **1085,** 320–323 (2006).
30. Treska, V., Topolcan, O. & Pecen, L. Cytokines as plasma markers of abdominal aortic aneurysm. *Clin. Chem. Lab. Med.* **38,** 1161–1164 (2000).
31. Wright, H. L., Moots, R. J., Bucknall, R. C. & Edwards, S. W. Neutrophil function in inflammation and inflammatory diseases. *Rheumatology* **49,** 1618–1631 (2010).
32. Habashi, J. P. *et al.* Angiotensin II type 2 receptor signaling attenuates aortic aneurysm in mice through ERK antagonism. *Science* **332,** 361–365 (2011).
33. Lindsay, M. E. *et al.* Loss-of-function mutations in TGFB2 cause a syndromic presentation of thoracic aortic aneurysm. *Nat. Genet.* **44,** 922–927 (2012).
34. Sawaki, D. & Suzuki, T. Targeting transforming growth factor-beta signaling in aortopathies in Marfan syndrome. *Circ. J.* **77,** 898–899 (2013).
35. Feng, X. H. & Derynck, R. Specificity and versatility in tgf-beta signaling through Smads. *Annu. Rev. Cell Dev. Biol.* **21,** 659–693 (2005).
36. Shindo, J. *et al.* Granulocyte-macrophage colony-stimulating factor prevents the progression of atherosclerosis via changes in the cellular and extracellular composition of atherosclerotic lesions in watanabe heritable hyperlipidemic rabbits. *Circulation* **99,** 2150–2156 (1999).
37. Zhu, S. N., Chen, M., Jongstra-Bilen, J. & Cybulsky, M. I. GM-CSF regulates intimal cell proliferation in nascent atherosclerotic lesions. *J. Exp. Med.* **206,** 2141–2149 (2009).
38. Haghighat, A., Weiss, D., Whalin, M. K., Cowan, D. P. & Taylor, W. R. Granulocyte colony-stimulating factor and granulocyte macrophage colony-stimulating factor exacerbate atherosclerosis in apolipoprotein E-deficient mice. *Circulation* **115,** 2049–2054 (2007).
39. Ye, P. *et al.* GM-CSF contributes to aortic aneurysms resulting from SMAD3 deficiency. *J. Clin. Invest.* **123,** 2317–2331 (2013).
40. Kimura, T. *et al.* Tenascin C protects aorta from acute dissection in mice. *Sci. Rep.* **4,** 4051 (2014).
41. Weissen-Plenz, G. *et al.* Aortic dissection associated with Cogans's syndrome: deleterious loss of vascular structural integrity is associated with GM-CSF overstimulation in macrophages and smooth muscle cells. *J. Cardiothorac. Surg.* **5,** 66 (2010).
42. Kessel, A., Vadasz, Z. & Toubi, E. Cogan syndrome—Pathogenesis, clinical variants and treatment approaches. *Autoimmun. Rev.* **13,** 351–354 (2014).
43. Babamusta, F. *et al.* Angiotensin II infusion induces site-specific intra-laminar hemorrhage in macrophage colony-stimulating factor-deficient mice. *Atherosclerosis* **186,** 282–290 (2006).
44. Brocheriou, I. *et al.* Antagonistic regulation of macrophage phenotype by M-CSF and GM-CSF: implication in atherosclerosis. *Atherosclerosis* **214,** 316–324 (2011).
45. Filonzi, E. L., Zoellner, H., Stanton, H. & Hamilton, J. A. Cytokine regulation of granulocyte-macrophage colony stimulating factor and macrophage colony-stimulating factor production in human arterial smooth muscle cells. *Atherosclerosis* **99,** 241–252 (1993).
46. Sakai, M. *et al.* Glucocorticoid inhibits oxidized LDL-induced macrophage growth by suppressing the expression of granulocyte/macrophage colony-stimulating factor. *Arterioscler. Thromb. Vasc. Biol.* **19,** 1726–1733 (1999).
47. Plenz, G., Koenig, C., Severs, N. J. & Robenek, H. Smooth muscle cells express granulocyte-macrophage colony-stimulating factor in the undiseased and atherosclerotic human coronary artery. *Arterioscler. Thromb. Vasc. Biol.* **17,** 2489–2499 (1997).
48. Saraff, K., Babamusta, F., Cassis, L. A. & Daugherty, A. Aortic dissection precedes formation of aneurysms and atherosclerosis in angiotensin II-infused, apolipoprotein E-deficient mice. *Arterioscler. Thromb. Vasc. Biol.* **23,** 1621–1626 (2003).
49. Cavalcante, J. L., Lima, J. A., Redheuil, A. & Al-Mallah, M. H. Aortic stiffness: current understanding and future directions. *J. Am. Coll. Cardiol.* **57,** 1511–1522 (2011).
50. Tarocchi, M. *et al.* Carcinogen-induced hepatic tumors in KLF6 + / − mice recapitulate aggressive human hepatocellular carcinoma associated with p53 pathway deregulation. *Hepatology* **54,** 522–531 (2011).

51. Clausen, B. E., Burkhardt, C., Reith, W., Renkawitz, R. & Forster, I. Conditional gene targeting in macrophages and granulocytes using LysMcre mice. *Transgenic Res.* **8**, 265–277 (1999).

## Acknowledgements

*Klf6* floxed mice were provided by Genentech. We thank Yasushi Imai for human samples and Naoko Sato for technical assistance. This research was funded in part by the Ministry of Health, Labour and Welfare of Japan; Grants-in Aid from the Ministry of Education, Culture, Sports, Science and Technology of Japan and the Japan Society for the Promotion of Science through its Funding Program for World-Leading Innovative R&D on Science and Technology (FIRST Program). This study was also supported by the National Institutes of Health National Cancer Institute (DK37340).

## Author contributions

B.-K.S. conducted experiments and partially wrote the manuscript. D.S. discussed on results. S.T. conducted experiments. D.F. provided human aortic samples and performed analysis. K.A., H.A., M.A., I.M., S.L.F. and I.K. consulted on the project. R.N. and T.S. planned and supervised the project. T.S. designed the study and wrote the manuscript. All authors discussed results and commented on the manuscript.

## Additional information

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**Competing financial interests:** The authors declare no competing financial interests.

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**How to cite this article:** Son, B.-K. *et al.* Granulocyte macrophage colony-stimulating factor is required for aortic dissection/intramural haematoma. *Nat. Commun.* 6:6994 doi: 10.1038/ncomms7994 (2015).

Perspective

# Medical management in type B aortic dissection

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Medical management is generally the preferred treatment for uncomplicated type B acute aortic dissection cases. It is often centered on the use of antihypertensive agents, which alleviates hemodynamic stress on the damaged aortic wall. Methods of medical management and drug selection are still based mainly on personal experience, expert opinion and historical observational studies as randomized controlled studies are lacking. Guidelines from European (ESC), American (ACC/AHA) and Asian (Japan) societies in the last decade have made recommendations on use of medications, but also reaffirmed the lack of evidence for therapeutic approaches and targeted medical management. More recent evidence suggests that there may be type-selective benefits for antihypertensive medications. Here, we will discuss the present understanding of medical management of acute aortic dissection.

**Keywords:** Aortic dissection; medication

Submitted Jun 16, 2014. Accepted for publication Jun 20, 2014.
doi: 10.3978/j.issn.2225-319X.2014.07.01
**View this article at:** http://dx.doi.org/10.3978/j.issn.2225-319X.2014.07.01

## Introduction

The aim of treatment in aortic dissection is to limit propagation of the false lumen and its negative consequences on end-organ perfusion by reducing and stabilizing hemodynamic stress on the aortic wall (1-8). As the majority of these type B dissection patients are hypertensive, medical therapy is centered on the use of anti-hypertensive agents. Medical therapy also aims to maintain hemodynamic stability in the chronic phase to promote aortic stability and to prevent aortic expansion, which might cause possible rupture and/or recurrent dissection.

Medical management of aortic dissection is still based mainly on personal experience, expert opinion and historical observational studies as there is a paucity of randomized controlled studies (1-8). As a result, in the last decade, efforts have been made to better understand the medical management of the disease. These efforts have ranged from proposal of guidelines from European, American and Asian societies, to the analysis of the International Registry of Acute Aortic Dissection, which is presently the largest and most comprehensive global registry database for this disease (9).

## Guideline-based medical management

### European guidelines

The European Society of Cardiology (ESC) was the first society to publish guidelines on aortic dissection in 2001 (2). In the initial assessment of the disease, immediate management of pain and blood pressure, with the target of lowering systolic blood pressure to 100-120 mmHg was recommended. For this, morphine sulfate is typically used for pain control and beta-blockers are most favored to reduce the force of left ventricular ejection (dP/dt), which will otherwise continue to weaken the arterial wall. Detailed recommendations were also presented on the use of beta-blockers, especially on intravenous use (e.g., loading and maximal dose) for agents such as propranolol, esmolol, metoprolol, atenolol, and labetalol. In patients who do not

tolerate beta-blockers well, such as patients with bronchial asthma, bradycardia or signs of heart failure, esmolol was stated to be a reasonable choice to test the patients' reaction to beta-blockers given its short half-life compared to metoprolol. The guidelines note that there is no data supporting the use of calcium antagonists (e.g., verapamil, diltiazem, or nifedipine) but also suggest that these drugs may be necessary to reduce blood pressure, particularly in patients with bronchial asthma. In cases where beta-blockade alone is insufficient to control hypertension, vasodilators were recommended as an ideal additional agent to control blood pressure; however, they should always be combined with beta-blockers because they can increase the force of left ventricular ejection. While beta-blocking agents are usually adequate in patients with slightly elevated blood pressure, it was noted that combined use of beta-blockers with intravenous sodium nitroprusside might also be required for more severe hypertension. Finally, the guidelines recommended that lowering of systolic blood pressure needs to be modified if oliguria or neurological symptoms develop.

It must be noted however, that the ESC guidelines did not make recommendations on long-term management but focused specifically on management in the acute phase with the inference to continue this as definitive treatment as necessary. Furthermore, these guidelines did not make recommendations on heart rate as mentioned in the American guidelines. Given that other guidelines have become more recently available, these items may be discussed in an upcoming update in the near future.

### Asian (Japanese) guidelines

The next available guidelines emerged from Japan in 2006 and were updated in 2011 (3). Medical management is classified according to acute (immediate) and chronic phases. Aims in the acute phase are to control blood pressure (100-120 mmHg), heart rate and pain. While this blood pressure target is generally accepted, there is a lack of robust evidence supporting this. Furthermore, it is thought that pain reflects the extension of the dissection, and alleviation of pain through blood pressure management will be beneficial.

In regards to specific anti-hypertensive agents, intravenous use of nicardipine, nitroglycerin and diltiazem in combination with beta-blockers was recommended. Intravenous use in the immediate phase is preferred given the ease of titration with transition to oral agents. It was noted that there is little evidence on oral agents. The use of beta-blockers was recommended to control heart rate to preferably less than 50 bpm in the immediate phase and to reduce dissection-related events in the chronic phase. Morphine or buprenorphine were also recommended to control persistent pain. After the immediate phase, blood pressure control should aim between 100-120 mmHg with some flexibility. In the chronic phase, it was emphasized that blood pressure control is important as favorable blood pressure control can reduce re-dissection by two-thirds. Only beta-blockers have evidence stating that they can reduce dissection-related events and inhibit aortic dilatation. There are reports stating that systolic blood pressure targets at 130 mmHg or less than 135/80 mmHg are appropriate, but again there is a lack of clear evidence. In cases with visceral ischemia, targets may need to be lowered. During rehabilitation, it was noted that systolic blood pressure preload should be kept below 130 mmHg and afterload to less than 150 mmHg.

Furthermore, uncomplicated type B dissections have a 30-day mortality rate of less than 10%, a rate which is comparable to surgical outcomes, and therefore medical treatment is appropriate in the acute phase. However, surgery should be considered for complicated cases, and that thoracic endovascular aortic repair (TEVAR) has shown promising results as a new and alternative option. Notably, patients resistant to anti-hypertensive therapy are not necessarily indicated for surgical management as they previously were, given that recent reports have suggested that elevated blood pressure is not necessarily a cause of increased risk for rupture. In the chronic phase (after two weeks), medical treatment should be continued for stable patients as they generally have a favorable prognosis.

### American guidelines

The American Heart Association/American College of Cardiology (AHA/ACC) guidelines were published in 2010 and classify aortic dissection among the acute aortic syndromes. These guidelines note that 71% of patients that sustain type B aortic dissections have a systolic blood pressure greater than 150 mmHg at presentation. Initial management of thoracic aortic dissection was recommended to decrease aortic wall stress by controlling heart rate and blood pressure.

Initial medical stabilization using beta-blockers was recommended to control aortic wall stress that is affected by the velocity of ventricular contraction,

 

the rate of ventricular contraction, and blood pressure parameters. Initial targets of heart rate less than 60 bpm and systolic blood pressure between 100-120 mmHg were recommended in order to maintain adequate end-organ perfusion. It should be noted that unlike the European guidelines, the American guidelines emphasize heart rate control. Intravenous propranolol, metoprolol, labetalol, or esmolol are suggested as excellent choices for initial treatment. In patients who are unable to tolerate beta-blockade, non-dihydropyridine calcium channel antagonists (verapamil, diltiazem) were suggested to offer acceptable, although less-established, alternatives. The use of beta-blockers, verapamil or diltiazem for rate control in patients with significant aortic regurgitation was noted to be potentially problematic due to deleterious effects on reflex tachycardia. In cases where vasodilators may be required to control blood pressure in addition to beta-blockade, intravenous sodium nitroprusside is the most established agent and offers the advantage of being rapidly titratable. It is important to consider, however, that vasodilator therapy without prior beta-blockade may cause reflex tachycardia and increased force of ventricular contraction leading to greater aortic wall stress and potentially cause false lumen propagation. Nicardipine, nitroglycerin and fenoldopam were also listed as being appropriate. Following initial stabilization with intravenous antihypertensives, most patients will require long-term antihypertensive treatment including the use of a beta-blocker plus additional classes of agents. It was noted that angiotensin-converting enzyme inhibitors or angiotensin receptor blockers may also retard aortic dilatation and that their use may be indicated.

## Findings from the International Registry of Acute Aortic Dissection

The International Registry of Acute Aortic Dissection (IRAD) is the world's largest multi-center registry-based study focused on aortic dissection to understand the clinical profiles, diagnosis, treatment and outcomes of the disease. Medical management of aortic dissection was addressed by analyzing the IRAD global registry database (579 type B cases). Data regarding medication prescription to patients with type B dissections at discharge was analyzed to investigate the association of medications and mortality. Initial univariate analysis showed that use of beta-blockers was associated with improved survival in all patients (P=0.03), and that use of calcium channel blockers was associated with improved survival in type B patients receiving medical management (P=0.03). Multivariate models further showed that use of calcium channel blockers was associated with improved survival in type B medically-treated patients (OR 0.55; 95% CI, 0.35-0.88, P=0.01). The findings of the IRAD analysis collectively demonstrated that while use of beta-blockers was associated with improved outcome in all patients with dissection, the use of calcium channel blockers was associated with improved survival selectively in type B dissections. Use of ACE inhibitors did not improve survival. Consequently, the IRAD analysis suggests the possibility of type-selective benefits of medications in acute aortic dissection.

## Conclusions

Recent society guidelines and findings from global registry databases (e.g., IRAD) have made significant contributions to our approach to medical management of acute type B aortic dissection. Close examination of guidelines show that each emphasizes control of different parameters (see *Table 1*). Looking to the future, these guidelines will serve as a working model to shape our medical management of dissection and the question of preferred treatment should only be revisited after they have been thoroughly implemented. If a randomized controlled trial ever becomes possible for dissection given ethical concerns, a definitive answer on the optimal treatment of the condition may become clearer.

**Table 1** Guideline-based medical management of aortic dissection

| | Class | Evidence |
|---|---|---|
| European Society of Cardiology (2001) | | |
| Immediate treatment | | |
| Pain relief (morphine sulphate) | I | C |
| Reduction of systolic blood pressure using beta-blockers (i.v. propranolol, metoprolol, esmolol or labetalol) | I | C |
| Additional vasodilator for severe hypertension (i.v. sodium nitroprusside to titrate BP to 100-120 mmHg) | I | C |
| For obstructive pulmonary disease, blood pressure lowering with calcium channel blockers | II | C |
| Japanese Circulation Society (2006, revised 2011) | | |
| Immediate treatment | | |
| Medical treatment for uncomplicated type B cases (patent/thrombosed false lumen, ulcer-like projection) | I | C |
| Medical treatment for cases resistant to anti-hypertensive treatment | IIa | C |
| Chronic treatment | | |
| Medical treatment if aortic diameter less than 50 mm and absence of rapid dilatation | I | C |
| Exercise (e.g., cycling, running) should be kept to a blood pressure of less than 180 mmHg | IIa | C |
| Beta-blockers should mainly be used for blood pressure treatment | IIb | C |
| Target for systolic blood pressure is 130-135 mmHg | IIb | C |
| American (AHA/ACC) guidelines 2010 | | |
| Initial management of thoracic aortic dissection | | |
| Intravenous beta-blockade should be initiated and titrated to a target heart rate of <60 bpm | I | C |
| Non-dihydropyridine calcium channel blocking agents as an alternative for rate control | I | C |
| If systolic BP remains >120 mmHg after adequate heart rate control, angiotensin-converting enzyme inhibitors and/or other vasodilators should be administered to further reduce BP that maintains adequate end-organ perfusion | I | C |
| Beta-blockers should be used cautiously with aortic regurgitation because they will block compensatory tachycardia | I | C |
| Vasodilator therapy should not be initiated prior to rate control to avoid associated reflex tachycardia | III | C |

AHA, American Heart Association; ACC, American College of Cardiology; BP, blood pressure.

## Acknowledgements

*Disclosure:* The authors declare no conflict of interest.

## References

1. Hiratzka LF, Bakris GL, Beckman JA, et al. 2010 ACCF/AHA/AATS/ACR/ASA/SCA/SCAI/SIR/STS/SVM guidelines for the diagnosis and management of patients with Thoracic Aortic Disease: a report of the American College of Cardiology Foundation/American Heart Association Task Force on Practice Guidelines, American Association for Thoracic Surgery, American College of Radiology, American Stroke Association, Society of Cardiovascular Anesthesiologists, Society for Cardiovascular Angiography and Interventions, Society of Interventional Radiology, Society of Thoracic Surgeons, and Society for Vascular Medicine. Circulation 2010;121:e266-369.
2. Erbel R, Alfonso F, Boileau C, et al. Diagnosis and management of aortic dissection. Eur Heart J 2001;22:1642-81.
3. Guidelines for diagnosis and treatment of aortic aneurysm and aortic dissection (JCS 2011): digest version. Circ J 2013;77:789-828.
4. Tsai TT, Nienaber CA, Eagle KA. Acute aortic syndromes. Circulation 2005;112:3802-13.
5. Golledge J, Eagle KA. Acute aortic dissection. Lancet 2008;372:55-66.
6. Nienaber CA, Eagle KA. Aortic dissection: new frontiers in diagnosis and management: Part I: from etiology to

 

diagnostic strategies. Circulation 2003;108:628-35.
7. Nienaber CA, Eagle KA. Aortic dissection: new frontiers in diagnosis and management: Part II: therapeutic management and follow-up. Circulation 2003;108:772-8.
8. Hagan PG, Nienaber CA, Isselbacher EM, et al. The International Registry of Acute Aortic Dissection (IRAD): new insights into an old disease. JAMA 2000;283:897-903.
9. Suzuki T, Isselbacher EM, Nienaber CA, et al. Type-Selective Benefits of Medications in Treatment of Acute Aortic Dissection (from the International Registry of Acute Aortic Dissection [IRAD]). Am J Cardiol 2012;109:122-7.

**Cite this article as:** Suzuki T, Eagle KA, Bossone E, Ballotta A, Froehlich JB, Isselbacher EM. Medical management in type B aortic dissection. Ann Cardiothorac Surg 2014;3(4):413-417. doi: 10.3978/j.issn.2225-319X.2014.07.01

 

CLINICAL RESEARCH STUDY

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# Cocaine-related Aortic Dissection: Lessons from the International Registry of Acute Aortic Dissection

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## ABSTRACT

**BACKGROUND:** Acute aortic dissection associated with cocaine use is rare and has been reported predominantly as single cases or in small patient cohorts.

**METHODS:** Our study analyzed 3584 patients enrolled in the International Registry of Acute Aortic Dissection from 1996 to 2012. We divided the population on the basis of documented cocaine use (C+) versus noncocaine use (C-) and further stratified the cohorts into type A (33 C+/2332, 1.4%) and type B (30 C+/1252, 2.4%) dissection.

**RESULTS:** C+ patients presented at a younger age and were more likely to be male and black. Type B dissections were more common among C+ patients than in C- patients. Cocaine-related acute aortic dissection was reported more often at US sites than at European sites (86.4%, 51/63 vs 13.6%, 8/63; $P < .001$). Tobacco use was more prevalent in the C+ cohort. No differences were seen in history of hypertension, known atherosclerosis, or time from symptom onset to presentation. Type B C+ patients were more likely to be hypertensive at presentation. C+ patients had significantly smaller ascending aortic diameters at presentation. Acute renal failure was more common in type A C+ patients; however, mortality was significantly lower in type A C+ patients.

**CONCLUSIONS:** Cocaine use is implicated in 1.8% of patients with acute aortic dissection. The typical patient is relatively young and has the additional risk factors of hypertension and tobacco use. In-hospital mortality for those with cocaine-related type A dissection is lower than for those with noncocaine-related dissection, likely due to the younger age at presentation.

 • *The American Journal of Medicine (2014) 127, 878-885*

**KEYWORDS:** Acute aortic dissection; Cocaine; Outcomes

Acute aortic dissection is a lethal cardiovascular disease with many associated comorbidities and has a high mortality rate even when properly treated. Cocaine use is a known risk factor for an acute aortic dissection. Such patients may present to unsuspecting physicians or emergency departments, particularly in urban areas where cocaine use is more common.[1] However, if not properly identified, patients with acute aortic dissection associated with cocaine use may

**Funding:** See last page of article.

**Conflict of Interest:** See last page of article.

**Authorship:** See last page of article.

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0002-9343/$ -see front matter 
http://dx.doi.org/10.1016/j.amjmed.2014.05.005

have worse short- and long-term outcomes, particularly in the setting of recurrent cocaine use after a dissection, which increases the risk of subsequent aortic rupture and re-dissection.[2] Little is known about the implications of cocaine on aortic dissection, because prior studies are predominately limited to small single-center case reports or case series.

In 2002, the International Registry of Acute Aortic Dissection (IRAD) published a brief report on aortic dissection associated with cocaine use. The IRAD identified 5 patients (0.5%)[3] with aortic dissection with documented cocaine use who were enrolled in the registry at that time. The IRAD's small percentage of patients with a history of cocaine use contrasted that from a study performed by Hsue et al,[4] in which 14 (37%) of 38 patients with dissection chronically used cocaine and presented to a single urban US public hospital.[2,4] We attempt to more precisely define the clinical features, imaging, diagnosis, and outcomes among 63 consecutive cocaine-using patients enrolled in the IRAD.

**CLINICAL SIGNIFICANCE**

- Some 1.8% of patients with an acute aortic dissection who were enrolled in the International Registry of Acute Aortic Dissection had a history of cocaine use.
- Patients with cocaine-related aortic dissection were more likely to be younger and to use tobacco. Their cocaine use, hypertensive state, and tobacco use collectively induce a dissection at an age that an acute aortic dissection would be less common.
- Patients with long-term, cocaine-related aortic dissection were more likely to experience rehospitalizations.

## METHODS

### Study Population

This analysis examines patients enrolled in the IRAD, for which the rationale and methodology have been published.[5] Thirty IRAD data-collection centers, representing 11 countries, contributed patients for this study. Acute type A aortic dissection was defined on the basis of Stanford classification as any dissection involving the ascending aorta, and acute type B aortic dissection was defined as any dissection not involving the ascending aorta, each presenting within 14 days of symptom onset. Consecutive patients at each IRAD center were identified prospectively at presentation or retrospectively by searching hospital discharge diagnosis records or surgical, pathology, and echocardiology databases. A cocaine-using patient was defined as a patient who uses cocaine to the detriment of his/her health and social functioning.

For this study, the IRAD patient population was divided into type A and B dissection cohorts. Within these cohorts, patients were further stratified by the presence or absence of cocaine use as defined in the IRAD lexicon and specified earlier (**Figure 1**).

### Data Collection

Data on 290 variables were recorded on a standardized form that included information on patient demographics, history, clinical presentation, physical findings, imaging study results, details of medical and surgical treatment, and patient outcomes. Data forms were reviewed internally for completeness and face validity and entered into an online database.

Yearly follow-up data were obtained up to 5 years after discharge using standardized forms. Collected data included variables on clinical, imaging, and vital status. At each enrolling hospital, study investigators obtained approval from their ethics or institutional review board to participate in the IRAD and its follow-up study.

### Statistical Analysis

This study compared patients with documented cocaine use with those without known cocaine use. Data are shown as frequencies and percentages, and as mean ± standard deviation or median and interquartile range. Missing data were not defaulted to negative, and denominators reflect only reported cases. Categoric variables were compared using a Pearson's chi-square test or Fisher exact test where appropriate. Continuous variables with normally distributed data were compared using the Student *t* test. Variables with a skewed distribution were compared using the Mann-Whitney *U* test. Kaplan-Meier analysis was performed to analyze long-term survival and freedom from aortic-related rehospitalization. *P* values $\leq .05$ were deemed as significant. SPSS Version 20.0 (IBM Inc, New York, NY) was used for all analyses.

## RESULTS

### Patient Demographics and History

This study investigated 3584 patients enrolled in the IRAD from 1996 to 2012. Within this group, 63 (1.8%) were

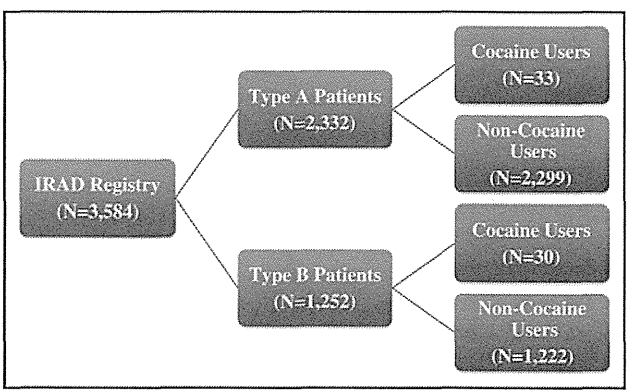


**Figure 1** Patient population. IRAD = International Registry of Acute Aortic Dissection.

**Table 1** Dissection Type per Cohort

| | Cocaine Users | Noncocaine Users | *P* Value |
|---|---|---|---|
| Type A | 33 (52.4%) | 2299 (65.3%) | .033 |
| Type B | 30 (47.6) | 1222 (34.7%) | |

documented as having used cocaine. Among these 63 patients, 33 (52.4%) had type A aortic dissection and 30 (47.6%) had type B aortic dissection (**Figure 1**). The frequency of type B aortic dissection was significantly higher in the cocaine-using cohort compared with the noncocaine-using cohort (47.6% vs 34.7%, $P = .033$) (**Table 1**).

Age at dissection was significantly younger for the cocaine-using cohort compared with others for both type A and B aortic dissections. Black race, male gender, and history of tobacco use were significantly more prevalent among the cocaine-using cohort. Patients with cocaine-related acute aortic dissection presented more often at US IRAD centers compared with European centers (86.4%, 51/63 vs 13.6%, 8/63; $P < .001$) (**Figure 2**). Body mass index was significantly higher among cocaine-using patients with dissection compared with noncocaine-using patients within the type B aortic dissection cohort (34.95 ± 7.71 kg/m$^2$ vs 28.71 ± 5.59 kg/m$^2$; $P = .001$) (**Table 2**).

There were few differences in comorbidity between cocaine users and their counterparts. Both hypertension and atherosclerosis did not differ significantly between the cocaine- and noncocaine-using cohorts. Prior cardiac surgery was lower in cocaine-using patients compared with noncocaine-using patients with type B aortic dissection (3.3% vs 19.1%; $P = .03$). There were also no significant differences seen in the number of hours between symptom onset and presentation to the initial hospital between each patient cohort (**Table 2**). No significant difference was seen in the number of hours from presentation to the initial hospital and diagnosis of acute aortic dissection between cocaine- and noncocaine-using patients (23.0 vs 18.5; $P = .161$).

## Presenting Symptoms

There were few differences seen in presenting symptoms among the cocaine users and noncocaine users. There were no significant differences in pain severity and location of pain between the 2 groups. Most cocaine- and noncocaine-using patients in both type A and B aortic dissection cohorts categorized their pain as severe and abrupt in onset (**Table 3**).

Ascending aortic measurements were significantly smaller for cocaine-using patients with both type A and B aortic dissections compared with their noncocaine-using counterparts. No significant differences were seen in measurements at other recorded aortic locations (**Table 3**).

Hypertension at presentation was nearly ubiquitous for cocaine-using patients compared with noncocaine-using patients with type B aortic dissection (92.6% vs 65.9%; $P = .004$). There was no significant difference in presenting hemodynamics when comparing the type A aortic dissection patient groups (**Table 3**). When comparing the overall cocaine user cohort with the noncocaine user cohort, hypertension at presentation was significantly higher for cocaine users (67.2% vs 42.3%; $P < .001$).

## Management

There were no differences among cocaine users and noncocaine users in the type of management they received,

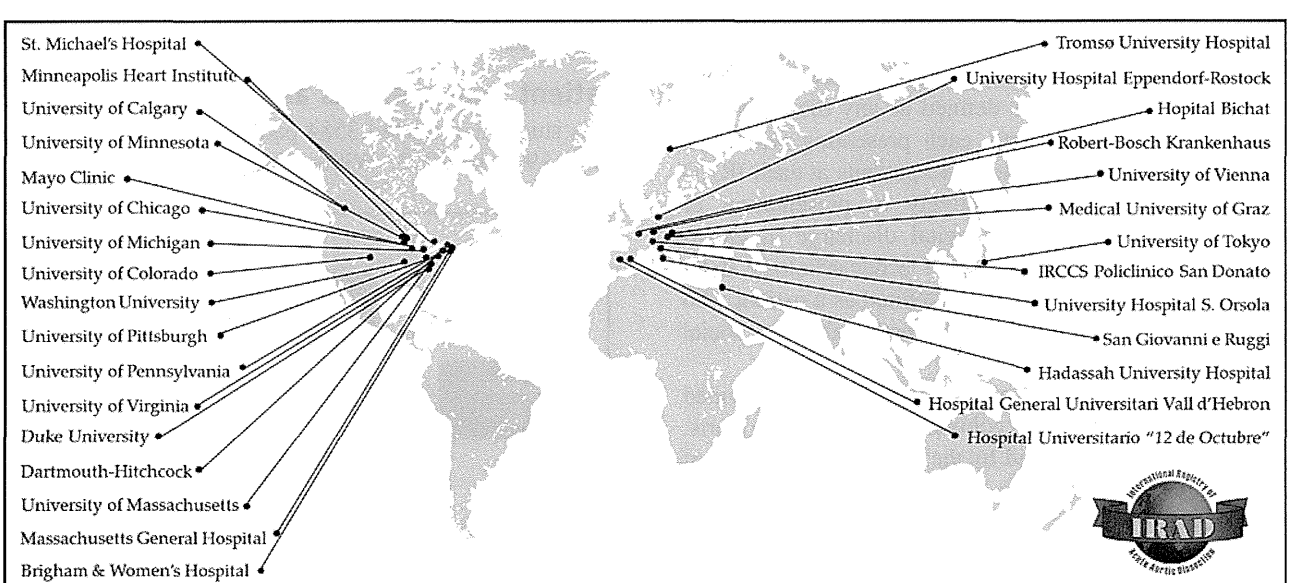


**Figure 2** IRAD collection centers. IRAD = International Registry of Acute Aortic Dissection; IRCCS = Instituto di Ricovero e Cura a Carattere Scientifico (Scientific Institutes for Research, Hospitalization and Health Care).

**Table 2** Patient Demographics and History

| | Type A Cocaine | Type A Noncocaine | *P* Value | Type B Cocaine | Type B Noncocaine | *P* Value |
|---|---|---|---|---|---|---|
| N | 33 (52.4%) | 2299 (65.1%) | .033 | 30 (47.6%) | 1222 (34.9%) | .033 |
| Age, y | 47.670 ± 10.9083 | 61.982 ± 14.5760 | <.001 | 47.594 ± 7.8155 | 64.004 ± 13.9865 | <.001 |
| Race, black | 13 (39.4%) | 103 (4.8%) | <.001 | 13 (44.8%) | 100 (8.6%) | <.001 |
| Gender, male | 28 (84.8%) | 1537 (66.9%) | .029 | 27 (90.2%) | 799 (65.4) | .005 |
| BMI, kg/m$^2$ | 30.885 ± 9.8589 | 27.942 ± 5.5892 | .211 | 34.950 ± 7.7133 | 28.710 ± 5.5892 | .001 |
| Any history tobacco use | 18 (90.0%) | 319 (51.1%) | .001 | 21 (95.5%) | 249 (60.1%) | .01 |
| Comorbidity history | | | | | | |
| Aortic dissection | 2 (6.2%) | 87 (3.8%) | .352 | 3 (10.0%) | 110 (9.1%) | .750 |
| Hypertension | 27 (81.8%) | 1658 (72.8%) | .245 | 25 (83.3%) | 977 (80.2%) | .671 |
| Atherosclerosis | 7 (21.2%) | 496 (22.0%) | .918 | 5 (16.7%) | 379 (31.5%) | .083 |
| Marfan syndrome | 3 (9.4%) | 93 (4.1%) | .137 | 0 (0%) | 47 (3.9%) | .625 |
| Aneurysm | 3 (9.4%) | 281 (12.4%) | .790 | 3 (10.0%) | 251 (20.7%) | .175 |
| Prior cardiac surgery | 3 (9.4%) | 303 (13.7%) | .611 | 1 (3.3%) | 228 (19.1%) | .03 |
| Renal insufficiency | 2 (9.1%) | 48 (5.7%) | .364 | 0 (0%) | 43 (9.5%) | .242 |
| Time (h) between symptom and presentation to initial hospital | 2.22 (1.33-10.92) | 2.00 (1.00-10.36) | .685 | 6.00 (1.50-41.00) | 3.00 (1.08-16.89) | .348 |

BMI = body mass index.

whether it be medical, endovascular, or surgical. With regard to in-hospital and postdischarge medications given to patients, no differences were seen when comparing patients with type B aortic dissection. Cocaine-using patients with type A aortic dissection were significantly more likely to initially be taking a beta-blocker (77.4% vs 53.2%; $P$ = .010), calcium channel blocker (26.7% vs 11.9%; $P$ = .023), and a diuretic (25% vs 8.5%; $P$ = .026). Cocaine-using patients with type A aortic dissection initially were less likely to be taking a vasopressor (0% vs 14.7%; $P$ = .028). From the available patients postdischarge, cocaine-using patients with type A aortic dissection were more likely to remain on a beta-blocker (100% vs 83.5%; $P$ = .024) (**Table 4**).

## In-Hospital Outcomes

There were no significant differences among the cohorts for in-hospital complications of cerebrovascular accident, coma, and myocardial infarction. However, cocaine-using patients with type A aortic dissection were significantly more likely to experience acute renal failure (42.4% vs 24.8%; $P$ = .02).

**Table 3** Presenting Symptoms

| | Type A Cocaine | Type A Noncocaine | *P* Value | Type B Cocaine | Type B Noncocaine | *P* Value |
|---|---|---|---|---|---|---|
| Chest pain | 28 (84.8%) | 1803 (81.7%) | .642 | 23 (79.3%) | 822 (69.3%) | .245 |
| Pain severity | | | | | | |
| Mild | 2 (8.0%) | 138 (7.6%) | .714 | 1 (3.8%) | 66 (6.3%) | 1.000 |
| Severe | 16 (64.0%) | 1375 (75.5%) | .185 | 22 (84.6%) | 782 (75.0%) | .359 |
| Worst | 7 (28.0%) | 308 (16.9%) | .143 | 3 (11.5%) | 194 (18.6%) | .425 |
| Pain in head or neck | 4 (14.3%) | 520 (25.5%) | .197 | 1 (3.7%) | 68 (5.9%) | 1.000 |
| Back pain | 16 (55.2%) | 895 (42.9) | .185 | 18 (62.1%) | 834 (70.7%) | .312 |
| Abdominal pain | 11 (37.9%) | 511 (24.7%) | .102 | 13 (44.8%) | 475 (40.9%) | .675 |
| Leg pain | 6 (20.7%) | 265 (13.0%) | .220 | 4 (14.3%) | 135 (11.8%) | .566 |
| Abrupt onset of pain | 26 (86.7%) | 1769 (83.1%) | .603 | 25 (86.2%) | 999 (85.8%) | .954 |
| Hemodynamics at presentation: | | | | | | |
| Hypertension | 14 (45.2%) | 631 (29.5%) | .059 | 25 (92.6%) | 765 (65.9%) | .004 |
| Hypotension | 5 (16.1%) | 344 (16.1%) | .996 | 0 (0%) | 39 (3.4%) | 1.000 |
| Shock | 1 (3.2%) | 209 (9.8%) | .357 | 0 (0%) | 14 (1.2%) | 1.000 |
| Aortic measurements | | | | | | |
| Annulus | 2.40 (2.375-2.80) | 2.50 (2.30-2.80) | .435 | 2.60 (1.55-3.05) | 2.50 (2.30-3.00) | .854 |
| Root | 4.10 (3.80-4.30) | 4.20 (3.70-5.00) | .265 | 3.50 (3.20-3.80) | 3.6 (3.30-3.80) | .611 |
| Sinotubular junction | 3.70 (3.10-4.20) | 3.90 (3.30-4.60) | .573 | 3.55 (2.80-3.60) | 3.30 (3.00-3.80) | .715 |
| Ascending aorta | 4.20 (3.90-5.00) | 5.00 (4.50-5.80) | .044 | 3.435 (3.40-3.725) | 3.80 (3.50-4.20) | .002 |
| arch | 3.85 (3.225-4.30) | 3.70 (3.20-4.10) | .633 | 3.30 (2.90-3.65) | 3.50 (3.00-4.00) | .198 |
| Descending aorta | 3.30 (2.80-3.90) | 3.30 (3.00-3.80) | .978 | 4.00 (3.30-4.20) | 4.00 (3.50-5.00) | .188 |

**Table 4** Management

| | Type A Cocaine | Type A Noncocaine | *P* Value | Type B Cocaine | Type B Noncocaine | *P* Value |
|---|---|---|---|---|---|---|
| Medical | 4 (12.1%) | 273 (11.9%) | 1.000 | 17 (56.7) | 783 (64.1) | .404 |
| Endovascular | 0 (0%) | 26 (1.1%) | 1.000 | 9 (30%) | 264 (21.6%) | .271 |
| Surgical | 27 (81.8%) | 1973 (85.9%) | .505 | 4 (13.3%) | 165 (13.5%) | 1.000 |
| Medications (initial) | | | | | | |
| ACE | 3 (15%) | 69 (10.4%) | .712 | 1 (4.8%) | 76 (18.5%) | .145 |
| ARBs | 0 (0%) | 25 (3.8%) | .632 | 0 (0%) | 28 (6.9%) | .387 |
| Beta-blocker | 24 (77.4%) | 1043 (53.2%) | .010 | 26 (89.7%) | 934 (82.1%) | .340 |
| Ca blocker | 8 (26.7%) | 226 (11.9%) | .023 | 8 (29.6%) | 299 (27.9%) | 1.000 |
| Diuretic | 5 (25%) | 56 (8.5%) | .026 | 1 (5%) | 64 (15.8%) | .228 |
| Nitroprusside | 8 (26.7%) | 446 (23.4%) | .829 | 14 (51.9%) | 419 (38.7%) | .230 |
| Statin | 0 (0%) | 37 (6.5%) | .421 | 0 (0%) | 34 (9.8%) | .379 |
| Vasodilator | 3 (10.3%) | 517 (27.1%) | .055 | 8 (27.6%) | 435 (40.4%) | .184 |
| Vasopressor | 0 (0%) | 279 (14.7%) | .028 | 0 (0%) | 34 (3.3%) | .626 |
| Medications (chronic at discharge) | | | | | | |
| ACE | 10 (40%) | 625 (40.8%) | 1.000 | 10 (38.5%) | 534 (52.9%) | .167 |
| ARBs | 2 (11.8%) | 76 (13%) | 1.000 | 0 (0%) | 59 (15.7%) | .097 |
| Beta-blocker | 25 (100%) | 1322 (83.5%) | .024 | 24 (92.3%) | 953 (91.1%) | 1.000 |
| Ca blocker | 10 (40%) | 550 (36%) | .834 | 15 (62.5%) | 631 (62.1%) | 1.000 |
| Diuretic | 6 (37.5%) | 255 (42.9%) | .800 | 11 (55%) | 147 (38%) | .159 |
| Statin | 1 (9.1%) | 143 (28.1%) | .197 | 4 (28.6%) | 107 (32.3%) | .768 |
| Vasodilator | 2 (12.5%) | 194 (14.1%) | 1.000 | 6 (30%) | 267 (29.2%) | 1.000 |

ACE = angiotensin-converting enzyme inhibitor; ARB = angiotensin receptor blocker; Ca = calcium channel.

In-hospital mortality was significantly lower for cocaine-using patients compared with noncocaine-using patients with type A aortic dissection (6.1% vs 25.5%; $P = .012$). No significant differences were observed in the length of hospital stay (**Table 5**).

## Long-Term Outcomes

The cocaine-using cohort was significantly more difficult to follow in the 5 years after dissection compared with the noncocaine-using cohort (follow-up rate 34.5% vs 51.7%; $P = .010$). Among the patients with available follow-up, no significant differences were noted for overall mortality between the 2 cohorts (**Figures 3** and **4**), and freedom from aortic rehospitalization at the 5-year follow-up was significantly lower in the group using cocaine (59.5% vs 81.5%; $P = .001$) (**Figure 5**).

## DISCUSSION

Cocaine use is a rare predisposing factor for patients with aortic dissection.[3] Our study reveals the rarity of the relationship between dissection and cocaine use, with only 63 of 3854 (1.8%) of the patients in the IRAD having a history of cocaine use. This rare coincidence may explain how a patient with a cocaine-induced acute dissection could be falsely diagnosed as having symptoms for a more common cocaine-related cardiovascular complication, such as a myocardial infarction or ischemic heart disease.[6] Furthermore, urban patient cohort studies indicate that cocaine is a more prevalent factor in predisposing a patient to an aortic dissection than in the general population.[2,7] Misdiagnosing an aortic dissection is potentially catastrophic and frequently contributes to an increased mortality rate.[5]

Cocaine-related aortic dissection predominates in young, hypertensive, obese, black men who use tobacco. Cocaine users in both type A and B aortic dissection patient cohorts were significantly younger compared with noncocaine users. On average, the cocaine-using cohort was more like to experience a dissection almost 15 years before the noncocaine-using cohort. The cocaine-using cohort was represented by a higher proportion of black patients compared with the noncocaine-using cohort. Hypertension is more frequent and more likely to be uncontrolled within black populations.[7] Furthermore, hypertensive effects of cocaine are more drastic in individuals who have a history of uncontrolled hypertension independently of drug use.[8] Such hypertensive effects may explain why cocaine users with type A aortic dissection were more likely to receive more antihypertensive medications in-hospital. Patients with cocaine-related aortic dissection are more likely to be male, which may be due in part to the increased likelihood of illicit drug use in men.[9,10]

Type B aortic dissections occurred more frequently when cocaine was related to aortic dissection. Chronic cocaine use has been associated with the development of premature atherosclerosis.[3] Atherosclerosis affects the distal descending aorta more often than other locations of the thoracic aorta.[11] Furthermore, individuals with chronic hypertension are more likely to develop type B aortic dissection than type A aortic dissection.[5] Therefore, cocaine's association with the development of both atherosclerosis and hypertension

**Table 5** In-Hospital Outcomes

| | Type A Cocaine | Type A Noncocaine | *P* Value | Type B Cocaine | Type B Noncocaine | *P* Value |
|---|---|---|---|---|---|---|
| CVA | 2 (6.5%) | 186 (9.0%) | 1.000 | 0 (0%) | 28 (2.5%) | 1.000 |
| Coma | 0 (0%) | 47 (2.3%) | 1.000 | 0 (0%) | 9 (0.8%) | 1.000 |
| MI | 2 (6.1%) | 156 (7.2%) | 1.000 | 1 (3.4%) | 29 (2.5%) | .529 |
| Acute renal failure | 14 (42.4%) | 539 (24.8%) | .02 | 9 (31.0%) | 202 (17.4%) | .058 |
| Extension of dissection | 1 (3.0%) | 179 (8.3%) | .517 | 3 (10.7%) | 101 (8.7%) | .730 |
| Hypotension | 7 (21.2%) | 656 (30.2%) | .265 | 1 (3.4%) | 122 (10.5%) | .352 |
| Limb ischemia | 6 (18.2%) | 272 (12.6%) | .336 | 3 (10.3%) | 109 (9.4%) | .750 |
| Length of hospital stay | 13.6042 (8.4819-18.8507) | 11.6875 (6.7023-20.0078) | .453 | 8.9281 (6.9531-23.7073) | 11.4708 (7.000-20.0406) | .582 |
| Death | 2 (6.1%) | 574 (25.5%) | .012 | 3 (10.0%) | 127 (10.4%) | 1.000 |

CVA = cerebrovascular accident; MI = myocardial infarction.

might be the underlying factor for the increase in type B aortic dissections among cocaine users.[3]

Our study reveals that short-term outcomes are similar between noncocaine and cocaine users. Both patient cohorts experienced a similar frequency in the number of in-hospital adverse events. Differences were seen in renal failure in cocaine users with type A aortic dissection, who were more likely to experience such an event in-hospital. This might have arisen because cocaine users with type A aortic dissection were more likely to be given beta-blocker antihypertensive medications in-hospital. The combination of cocaine and beta-blockers can result in unopposed alpha-receptor vasoconstriction, leading to severe hypertension and vasoconstriction that could contribute to the increased incidence of renal failure seen in this cohort.[1,12] Furthermore, in-hospital mortality was lower for cocaine users with type A aortic dissection, which may be due to the younger age of cocaine users. Age is an independent factor for in-hospital mortality.[13,14]

Despite the cocaine users' young age, these patients had similar long-term all-cause mortality compared with noncocaine users. This is noteworthy because older patients with aortic dissection are generally more likely to experience mortality.[14] Furthermore, cocaine users were less likely to return for follow-up after their aortic dissections. This may be related to poor compliance and medication use after discharge.[2] This might be a contributing factor to why cocaine users experience more rehospitalizations related to aortic complications after their initial discharge.

Our findings are consistent with those from important prior studies. Eagle et al[3] and Daniel et al[9] revealed that cocaine use is rare within their patient population with general dissection. In the 2002 IRAD study, cocaine-using patients were more likely to be young, black, and hypertensive.[3] The report by Hsue et al[4] revealed an increased prevalence in young, male, black and uncontrolled hypertensive patients when cocaine was involved. Furthermore, all of their cocaine users also used tobacco.[4] Singh et al[2] studied cocaine-user patient demographics and revealed cocaine-using patients with dissection to be young, hypertensive, black, and tobacco users. Daniel et al[9] confirmed that young and male patients comprise most cocaine-using patients. All of their patients used tobacco, and the majority were hypertensive.[9] For long-term outcomes, Singh et al[2] reported that cocaine users had a higher mortality rate than noncocaine users.[2] Recurrent dissection was a large contributor to the cocaine user cohort's high mortality,[2] possibly related to continuing cocaine use or medical noncompliance.

Findings from our study include the observation that although type B aortic dissections were more prevalent when cocaine was involved, type A aortic dissections still comprised a majority of our cocaine patient cohort. A similar trend was revealed for black race, which was more prevalent when cocaine was involved, but black patients did not make up a majority of the patient cohort. It was discovered that in both type A and B aortic dissection