Generalized arterial calcifications in a twin recipient treated with etidronate: description of a case report within a narrative review

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Generalized arterial calcification of infancy (GACI) was initially identified by Bryant and White in 1901. The incidence is unknown, but around 1000 cases have been documented, mostly diagnosed post-mortem [1]. However, GACI has also been detected antenatally through fetal echocardiography [2]. Hajdu *et al.* [3] observed hyperechogenic and hypokinetic walls in both atria and ventricles, along with enlarged great vessels (the fetus died in utero). Samon *et al.* [4] diagnosed GACI antenatally by noting hyperechogenicity in the proximal aorta and central pulmonary vessels, with hypertrophic cardiomyopathy. In two other fetuses with GACI, the cardiac chambers were dilated, and severe myocardial dysfunction with atrioventricular valve regurgitation was evident. Additionally, hyperechogenic foci were found in the wall of both atria and great arteries. Dystrophic calcification in GACI leads to reduced vessel compliance and can cause systemic and/or pulmonary hypertension, resulting in ventricular hypertrophy or dilation, potentially leading to cardiac failure [5, 6]. Furthermore, GACI can cause fetal heart failure with polyhydramnios, fetal hydrops, and, ultimately, fetal demise during the early part of the last trimester of pregnancy [7, 8]. Postnatally, GACI presents with severe congestive heart failure and refractory systemic hypertension [9–11]. Medical treatment of cardiovascular effects is generally unsuccessful.

Therapeutic approaches

Relative to pharmacological interventions, some infants treated with bisphosphonate (an inhibitor of calcium salt precipitation) [12, 13] or a combination of corticosteroids, estrogens, and thyroid hormones have survived. Additionally, spontaneous remission of the disorder has been reported [14]. Etidronate, one of the first-generation bisphosphonates, has a profile favoring anti-mineralization over anti-osteoclastic activity. It has been shown to prevent ectopic mineralization in soft connective tissues [15]. At the same time, etidronate increases bone mineral density and corrects the mineralization defect noted in GACI, at least in animal

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models [15]. In patients with pseudoxanthoma elasticum, etidronate reduces arterial calcification and subretinal neovascularization events [16]. Therefore, etidronate may offer dual benefits by preventing ectopic mineralization and reducing bone loss over the long term through its anti-osteoclastic effect [17]. In adult patients with arterial calcification, etidronate can slow the progression of vascular disease and is both safe and well-tolerated [18].

GACI in twin-twin transfusion syndrome

Concerning the topic of our study, GACI has been reported in two sets of monochorionic and dichorionic twins [19–21]. In these cases, both twins were affected and tested positive for ENPP1 (ecto-nucleotide pyrophosphatase/phosphodiesterase). One case of GACI was also described in the recipient twin of a twin-twin transfusion syndrome (TTTS) [4]. This latter is a condition where blood flow is uneven between twins sharing a placenta (monochorionic twins). TTTS affects about 4% to 26% of monochorionic, diamniotic twins. In TTTS, there is an unequal sharing of blood that passes between the twins through blood vessel connections on the surface of the placenta. One

twin (the donor) pumps blood to the other twin (the recipient). The recipient twin receives an excessive amount of blood, leading to polyhydramnios, increased pressure, volume overload, and the release of mediators associated with hypertension. Conversely, the donor twin receives a low amount of blood, leading to oligohydramnios. The right ventricle of the recipient twin initially undergoes compensatory hypertrophy, followed by dilation and heart failure with tricuspid regurgitation and pulmonary stenosis and/or regurgitation, potentially progressing from functional to organic, with pulmonary atresia. Fetal hydrops due to heart failure can also occur. Some case reports describe the calcifications at both the pulmonary valve and trunk (supravalvular) levels in recipient twins [19–21]. The authors speculate that pulmonary artery thickening, and calcification may result from vascular injury caused by excessive volume load, as there is no pressure gradient between the aorta and pulmonary artery during fetal life.

GACI has been described in only two sets of monochorionic and dichorionic twins [22, 23]. In these cases, both twins were affected, with positive genetics for ENPP1 (ecto-nucleotide pyrophosphatase/phosphodiesterase).
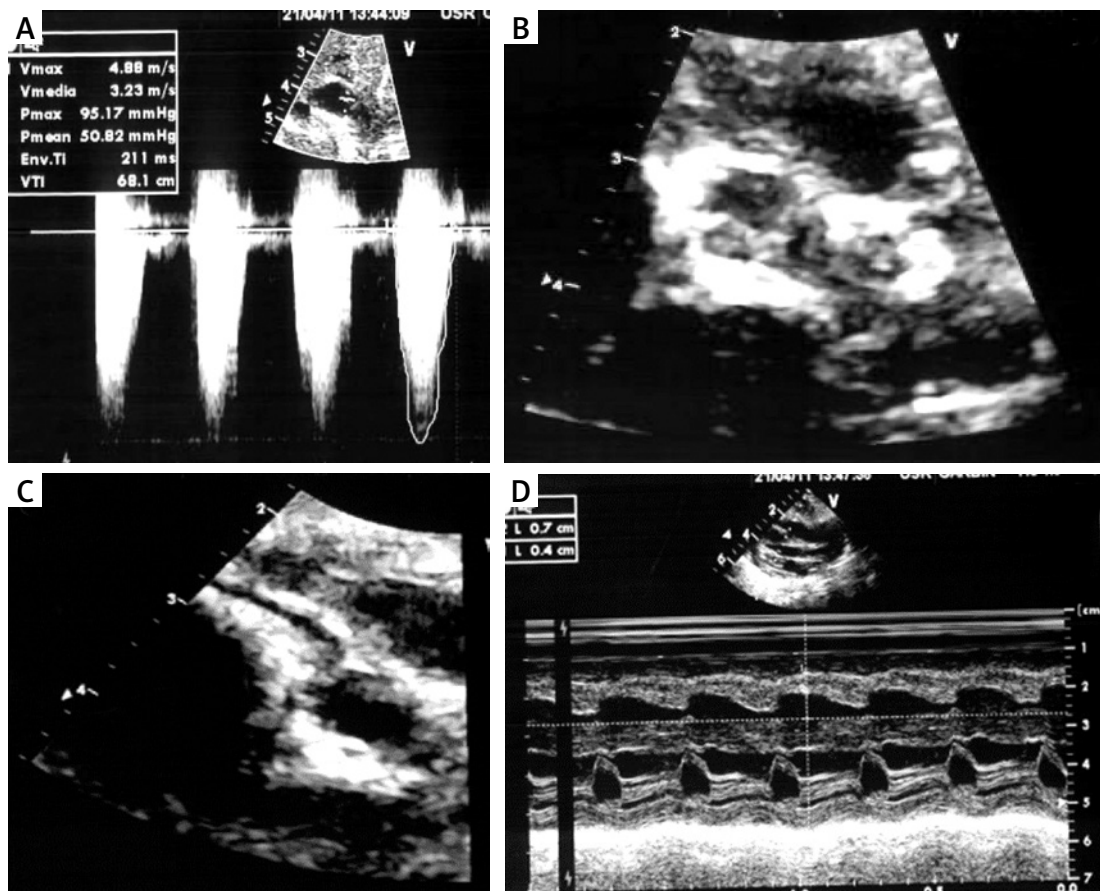

Figure 1. Echocardiogram. **A** – Severe pulmonary supravalvular stenosis, **B** – diffuse pulmonary supravalvular calcifications, **C** – coronary nodular calcifications, **D** – left and right ventricle hypertrophy

Case report

Here we present our experience with a recipient twin from a monochorionic diamniotic twin pregnancy complicated by stage three TTTS, treated with fetoscopic selective laser ablation of placental anastomoses according to Solomon technique [24]. An echocardiogram 1 week after laser surgery showed moderate biventricular hypertrophy and dilatation, with a reduced caliper of the ascending aorta and pulmonary trunk. At birth, the echocardiogram revealed mild supravalvular pulmonary stenosis with mild stenosis of pulmonary branches, moderate supravalvular aortic stenosis, and dilatation of coronary arteries with small diffuse calcific nodules. Virological, metabolic, hormonal and autoantibody investigations were negative.

The degree of supravalvular aortic stenosis, characterized by a tubular appearance, hyperechogenicity, and thickening of the aortic wall, rapidly progressed during the first month. Additionally, significant calcification of the pulmonary trunk with severe pulmonary supravalvular stenosis, diffuse calcifications at the bifurcation and pulmonary branches, and diffuse nodular calcifications at the left coronary artery were also demonstrated (Figure 1). Computed tomography (CT) total body CT angiography [25] confirmed the echocardiographic supravalvular calcifications and excluded other vascular calcifications in the head, neck, thorax and abdomen vessels (Figure 2). The CT also revealed thickening of the wall of the abdominal aorta and renal arteries (Figure 2). The search for ENPP1 gene mutation (6q22) was negative. The mutation in ABCC6 (ATP Binding Cassette Subfamily C Member 6) gene was not tested because its link to GACI was unknown at the time of birth.

Initially, β-blocker therapy was administered, but it resulted in only minimal reduction of supravalvular aortic and pulmonary gradients. Due to the severity of the stenoses, we, in agreement with the parents, initiated etidronate treatment (18 mg/kg/day). The drug was given after obtaining informed consent from parents, in galenic formulation provided by our hospital pharmacy. Monthly radiographic checks of the radius and ulna were performed to monitor for possible bone mass reduction. After 6 months of treatment, we observed mild residual supravalvular aortic stenosis and mild-to-moderate supravalvular pulmonary stenosis, both of which showed continuous improvement. An echocardiographic examination after 3 years showed minimal supravalvular aortic gradients with wall calcification and post-stenotic dilation. Additionally, there was mild supravalvular pulmonary stenosis with calcification of pulmonary branches, and the previously visible coronary artery calcific nodules were no longer present (Figure 3). Etidronate therapy was then discontinued with any side effects or reduction in bone mass. At 10-year follow-up, the echocardiographic findings remain unchanged.

Discussion

Stenosis of the pulmonary valve, up to atresia, has been demonstrated in several recipient twins in TTTS-complicated pregnancies. The recognized pathophysiology involves volume and pressure overload, predominantly affecting the right ventricle of the recipient twin. This overload occurs due to passage of hypertension mediators from the donor to the recipient, including renin, angiotensin and endothelin 1. The initial compensatory response of the right ventricle, characterized by progressive hypertrophy, itself contributes to reduced flow through the pulmonary valve, leading to functional stenosis or atresia. This functional form may progress to an organic one.
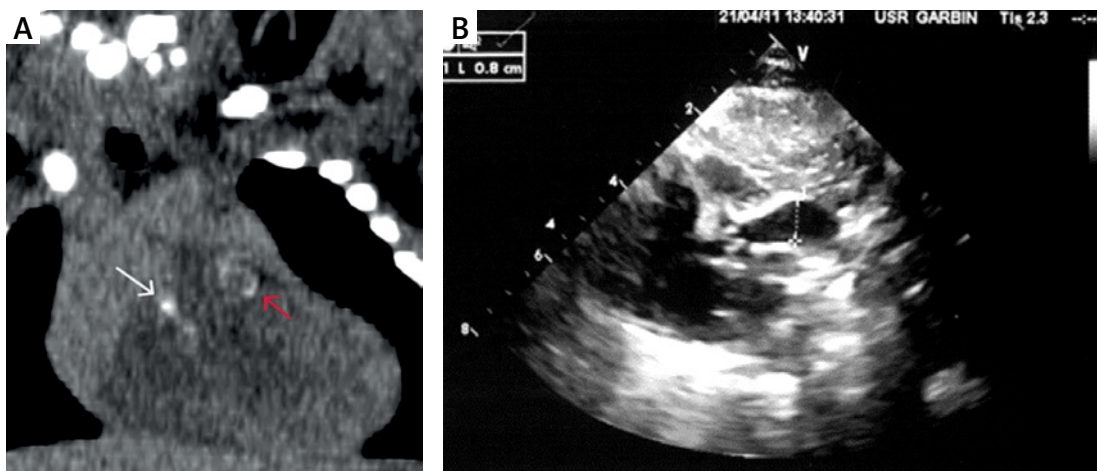
Figure 2. Computed tomography (CT) total body CT angiography. **A** – Coronal reformation from unenhanced CT. White and red arrows show supravalvular pulmonary and aortic calcifications. **B** – Aortic supravalvular tubular calcifications
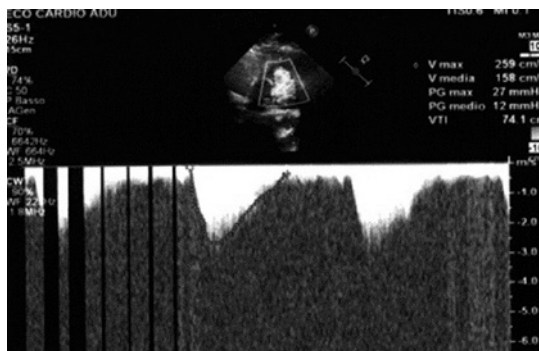
Carla Giuseppina Corti, Luisa Federica Nespoli, Alessandro Campari, Eleonora Gnan, Massimiliano Ruscica, Stefano Carugo

Figure 3. Echocardiogram. Mild residual pulmonary supravalvular stenosis

The vasoconstrictor and mitogenic effects of endothelin 1 may partly explain the increased thickness of the tunica intima of both the aorta and pulmonary artery, as observed in some recipient twins.

Literature reports on supravalvular pulmonary and aortic calcification, along with diffuse calcification of the pulmonary branches and aortic wall, consistently show negative genetics for both *ENPP1* and *ABCC6* mutations. These cases typically follow a benign course, with gradual resolution of the cardiovascular involvement in the absence of etidronate therapy.

However, in 1 case, aortic stenosis progression was observed despite therapy, leading to surgical intervention at the age of 4 months. Interestingly, both twins in that case showed the *ABCC6* gene mutation [15]. This observation prompt us to hypothesize how epigenetic factors may influence the phenotype and the development (or lack) of calcifications.

Conversely, the renin-angiotensin-aldosterone system (RAAS) is involved in all stages of cardiovascular homeostasis, as demonstrated by Mahieu-Caputo *et al.* [26]. The renal expression of renin was up-regulated in donors and down-regulated in recipient twins, potentially serving as the link between abnormalities in fetal blood volume and vascular disturbances in TTTS. This initial positive adaptation then becomes deleterious, particularly in the recipient twin, who may be exposed to the transfer of various molecules produced by the donor via intertwin vascular anastomoses, including vasoactive RAAS hormones [27].

Some experimental studies have shown that chronic infusion of angiotensin II (AII) in rats increased the left ventricular mass, even when the pressor activity of AII was blocked [28]. In a rat model of "pressure-overload" cardiac hypertrophy (abdominal aortic constriction), treatment with an ACE inhibitor completely prevented the increase in the left ventricular mass, without effecting cardiac afterload [29]. These observations suggest that AII has a direct hypertrophic action on heart tissue, in addition to indirect effects mediated through increases in blood pressure and vascular resistance. All could be one of the most important epigenetic factors leading to the cardiovascular changes in the recipient twin, as observed in our patient. Conversely, studies performed on animal models of aortic valve stenosis (AS) show that the administration of angiotensin-receptor-blockers (ARBs) slows the progression of the disease compared to ACE inhibitors.

This difference might be explained by the presence of elevated levels of chymase in calcific aortic valve tissue. Similar to ACE, chymase converts AI to AII. Therefore, the inhibition of ACE activity may be bypassed by the action of chymase, resulting in the production of AII, while the effects of AII can be blocked by ARBs.

Several studies have been conducted to elucidate the processes involved in the progression of aortic valve stenosis, allowing us to hypothesize another epigenetic mechanism related to the described cases of diffuse aortic and pulmonary stenosis in TTTS twin recipients. Hong *et al.* [30] demonstrated how increasing concentrations of oxidized low-density lipoproteins (oxLDLs) lead to the uncoupling of endothelial nitric oxide synthase. This uncoupling results in a switch from nitric oxide (a protective antioxidant) production to the generation of reactive oxidative species (ROS) superoxide. Further lipid peroxidation and endothelial injury contribute to the inflammatory and calcification processes within the valve. The pro-inflammatory cytokines, coupled with reduced availability of nitric oxide, disrupt the balance between extracellular matrix production and breakdown in the valve [31, 32], ultimately leading to dystrophic microcalcification [33].

In conclusion, to the best of our knowledge, this is the first report of long-term follow-up of progressive arterial calcifications in a recipient twin from a TTTS-complicated pregnancy, without a positive genetic mutation. The patient was successfully treated with etidronate, thus avoiding the need for surgical intervention.

The mechanisms underlying the development of diffuse arterial calcifications in our patient extend beyond the known pressure-volume imbalance and in vasoactive RAAS hormones in the recipient twin. Significant epigenetic modifications affecting antioxidation processes may play a significant role.

Our experience, supported by clinical and laboratory data, demonstrates that etidronate effectively induces regression of arterial calcifications and is essentially free of major side effects.

Acknowledgments

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Funding

Ethical approval

Ethical approval was not required for the study involving human samples in accordance with the local legislation and institutional requirements. Written informed consent was obtained from the minor's parents for the publication of any potentially identifiable images or data included in this article.

Conflict of interest

The authors declare no conflict of interest.

References

1. Van Reempts PJ, Boven KJ, Spitaels SE, Roodhooft AM, Vercruyssen ELJ, Van Acker KJ. Idiopathic arterial calcification of infancy. Calcif Tissue Int 1991; 48: 1-6.
2. Nasrallah FK, Baho H, Sallout A, Qurashi M. Prenatal diagnosis of idiopathic infantile arterial calcification with hydrops fetalis. Ultrasound Obstet Gynecol 2009; 34: 601-4.
3. Hajdu J, Marton T, Papp C, Hruby E, Papp Z. Calcification of the fetal heart--four case reports and a literature review. Prenat Diagn 1998; 18: 1186-90.
4. Samon LM, Ash KM, Murdison KA. Aorto-pulmonary calcification: an unusual manifestation of idiopathic calcification of infancy evident antenatally. Obstet Gynecol 1995; 85: 863-5.
5. Corbacioglu Esmer A, Kalelioglu I, Omeroglu RE, et al. Prenatal ultrasonographic diagnosis of generalized arterial calcification of infancy. J Clin Ultrasound 2015; 43: 50-4.
6. Pala HG, Bilgili G, Artunc Ulkumen B, Alkan F, Coskun S. A case of antenatal diagnosis and postnatal characteristics of ıdiopathic ınfantile arterial calcification (IIAC and prenatal diagnosis). J Obstet Gynaecol 2016; 36: 665-7.
7. Jones DE, Pritchard KI, Gioannini CA, Moore DT, Bradford WD. Hydrops fetalis associated with idiopathic arterial calcification. Obstet Gynecol 1972; 39: 435-40.
8. Pu L, Dai X, Liu H, Li L, Zhao F, Chen J. Prenatal diagnosis of generalized arterial calcification of infancy in the second trimester. Prenat Diagn 2022; 42: 1538-44.
9. Făgărăşan A, Gozar L, Ghiragosian SR, et al. Severe early-onset manifestations of generalized arterial calcification of infancy (mimicking severe coarctation of the aorta) with ABCC6 gene variant - Case report and literature review. Front Cardiovasc Med 2022; 9: 1032519.
10. Carles D, Serville F, Dubecq JP, Alberti EM, Horowitz J, Weichhold W. Idiopathic arterial calcification in a stillborn complicated by pleural hemorrhage and hydrops fetalis. Arch Pathol Lab Med 1992; 116: 293-5.
11. Ferreira CR, Kintzinger K, Hackbarth ME, et al. Ectopic calcification and hypophosphatemic rickets: natural history of ENPP1 and ABCC6 deficiencies. J Bone Miner Res 2021; 36: 2193-202.
12. Akhtar Ali S, Ng C, Votava-Smith JK, Randolph LM, Pitukcheewanont P. Bisphosphonate therapy in an infant with generalized arterial calcification with an ABCC6 mutation. Osteoporos Int 2018; 29: 2575-9.
13. Boyce AM, Gafni RI, Ferreira CR. Generalized arterial calcification of infancy: new insights, controversies, and approach to management. Curr Osteoporos Rep 2020; 18: 232-41.
14. Ferreira CR, Hackbarth ME, Ziegler SG, et al. Prospective phenotyping of long-term survivors of generalized arterial calcification of infancy (GACI). Genet Med 2021; 23: 396-407.
15. Li Q, Sundberg JP, Levine MA, Terry SF, Uitto J. The effects of bisphosphonates on ectopic soft tissue mineralization caused by mutations in the ABCC6 gene. Cell Cycle 2015; 14: 1082-9.
16. Kranenburg G, de Jong PA, Bartstra JW, et al. Etidronate for prevention of ectopic mineralization in patients with pseudoxanthoma elasticum. J Am Coll Cardiol 2018; 71: 1117-26.
17. Kauffenstein G, Chappard D, Leftheriotis G, Martin L. ABCC6 deficiency and bone loss: a double benefit of etidronate for patient presenting with pseudoxanthoma elasticum? Exp Dermatol 2022; 31: 1635-7.
18. Ferrante EA, Cudrici CD, Rashidi M, et al. Pilot study to evaluate the safety and effectiveness of etidronate treatment for arterial calcification due to deficiency of CD73 (ACDC). Vasc Med 2024; 29: 245-55.
19. Saxena A, Soni NR. Pulmonary artery calcification in recipient twins of twin-to-twin transfusion syndrome: a report of three cases. Pediatr Cardiol 2003; 24: 80-3.
20. Popek EJ, Strain JD, Neumann A, Wilson H. In utero development of pulmonary artery calcification in monochorionic twins: a report of three cases and discussion of the possible etiology. Pediatr Pathol 1993; 13: 597-611.
21. Eronen M, Pohjavuori M, Heikkilä P. Fatal outcome of two siblings with idiopathic arterial calcification of infancy diagnosed in utero. Pediatr Cardiol 2001; 22: 167-9.
22. Votava-Smith JK, Pitukcheewanont P, Randolph LM, Chmait RH. Generalized arterial calcification in a recipient twin: discordant fetal hemodynamics result in differing phenotypes in monozygotic twins with an ABCC6 mutation. Fetal Diagn Ther 2017; 41: 234-6.
23. Chen H, Fowler M, Yu CW. Generalized arterial calcification of infancy in twins. Birth Defects Orig Artic Ser 1982; 18: 67-80.
24. Shamshirsaz AA, Chmait RH, Stirnemann J, et al. Solomon versus selective fetoscopic laser photocoagulation for twin-twin transfusion syndrome: a systematic review and meta-analysis. Prenat Diagn 2023; 43: 72-83.
25. Santangelo G, Gherbesi E, Donisi L, et al. Imaging approaches in risk stratification of patients with coronary artery disease: a narrative review. Arch Med Sci 2024. doi:10.5114/aoms/188808.

Carla Giuseppina Corti, Luisa Federica Nespoli, Alessandro Campari, Eleonora Gnan, Massimiliano Ruscica, Stefano Carugo

26. Mahieu-Caputo D, Dommergues M, Delezoide AL, et al. Twin-to-twin transfusion syndrome. Role of the fetal renin-angiotensin system. Am J Pathol 2000; 156: 629-36.
27. Khairallah PA, Kanabus J. Angiotensin and myocardial protein synthesis. In: Perspectives in Cardiovascular Research. Tarazi RD, Dunbar JB (ed.). Raven, New York 1983; 8: 337-47.
28. Baker KM, Dostal DE, Chemin MI, Wealand AL, Conrad KM. Angiotensin II-mediated cardiac hypertrophy in adult rats. J Cell Biochem 1991; 15 C Suppl.: 167.
29. Baker KM, Chemin MI, Wixson SK, Aceto JF. Renin angiotensin system involvement in pressure overload cardiac hypertrophy in rats. Am J Physiol 1990; 259: H324-32.
30. Hong FF, Liang XY, Liu W, et al. Roles of eNOS in atherosclerosis treatment. Inflamm Res 2019; 68: 429-41.
31. Rajamannan NM. Bicuspid aortic valve disease: the role of oxidative stress in Lrp5 bone formation. Cardiovasc Pathol 2011; 20: 168-76.
32. Hutson HN, Marohl T, Anderson M, Eliceiri K, Campagnola P, Masters KS. Calcific aortic valve disease is associated with layer-specific alterations in collagen architecture. PLoS One 2016; 11: e0163858.
33. Akahori H, Tsujino T, Masuyama T, Ishihara M. Mechanisms of aortic stenosis. J Cardiol 2018; 71: 215-20.