

## Article

# Effect of Superficial Anastomoses on Circulatory Dynamics in Twin-Twin Transfusion Syndrome

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## Abstract

The role of superficial anastomoses in the survival of fetuses with twin-twin transfusion syndrome after fetoscopic laser photocoagulation is unknown. This study aimed to evaluate how superficial anastomoses affect the circulatory dynamics of both fetuses with twin-twin transfusion syndrome using ductus venous Doppler waveforms. We included all twin-twin transfusion syndrome (TTTS) patients who underwent fetoscopic laser photocoagulation in our institution from 2006 to 2019; fetal demise cases after fetoscopic laser photocoagulation were excluded. We recorded ductus venous Doppler waveforms on the same day or one day before fetoscopic laser photocoagulation and one day after fetoscopic laser photocoagulation and measured the ductus venous pulsatility index and velocity ratios. We compared these z-scores of donor and recipient twins between a group without superficial anastomoses and the groups with arterio-arterial or veno-venous anastomoses. A total of 115 surviving TTTS placentas after fetoscopic laser photocoagulation were analyzed. The ductus venous pulsatility index and all ratios were better in recipient twins with arterio-arterial anastomoses than in those without. The a-wave-related ratios were better in recipient twins with veno-venous anastomoses than in those without. Superficial anastomoses reduced the blood volume and arterio-arterial anastomoses protected the diastolic cardiac function in recipient twin-twin transfusion syndrome twins before fetoscopic laser photocoagulation. Superficial anastomoses in TTTS equilibrate blood pressure between donor and recipient twins.

**Keywords:** arterio-arterial anastomoses; veno-venous anastomoses; twin-twin transfusion syndrome; fetoscopic laser photocoagulation; ductus venous Doppler

(Received 8 September 2022; revise received 16 November 2022; accepted 21 November 2022; First Published online 6 January 2023)

Twin-twin transfusion syndrome (TTTS) is caused by the uneven transfer of blood between fetuses via placental vascular anastomoses in monochorionic-diamniotic twin pregnancies. In terms of hemodynamics, some studies have indicated that the recipient twin is typically hypervolemic; in contrast, the donor twin is hypovolemic (Wohlmuth et al., 2016).

Arterio-arterial (AA) and veno-venous (VV) anastomoses are superficial anastomoses that can facilitate bidirectional blood flow via their arteriovenous (AV) branches. AA anastomoses can protect against TTTS, although they may sometimes lead to poor prognoses (Kontopoulos et al., 2007; Murakoshi et al., 2008; Murakoshi et al., 2003). Similarly, VV anastomoses may play a role in the development of TTTS; however, they can also help protect against TTTS (de Villiers et al., 2015; Diehl et al., 2001; Zhao et al., 2015; Zhao et al., 2014). Superficial anastomoses have been labeled ‘unstable’ because they have several roles that are poorly understood.

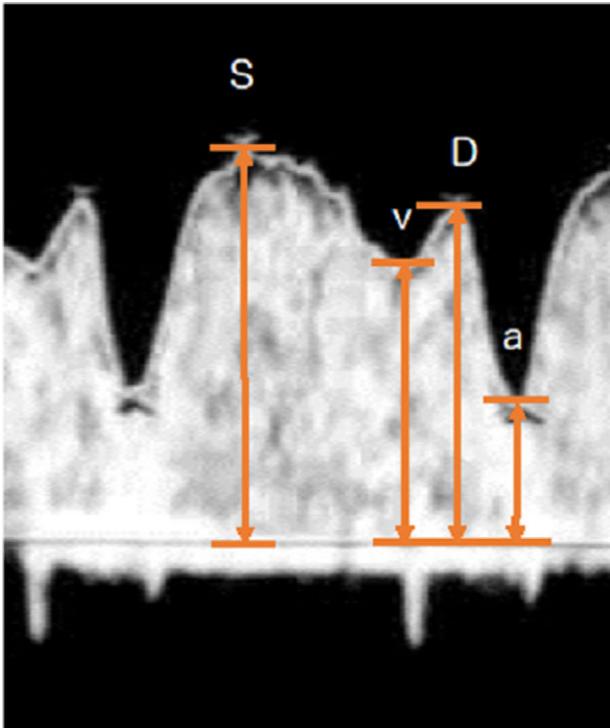
We previously reported that the presence of AA anastomoses is a risk factor for the demise of the donor twin after fetoscopic laser photocoagulation (FLP) because the AA anastomoses protect the

donor twin from hypovolemia before FLP (Konno et al., 2019). Furthermore, demise of the recipient twin or both fetuses occurs significantly more often after FLP with VV than after FLP without VV, because VV anastomoses protect the circulation of the recipient twin or both fetuses before FLP (Konno et al., 2019). Superficial anastomoses might facilitate hemodynamic changes in both twins because they are anastomosed directly without the cotyledon of the placenta. However, the role of such superficial anastomoses in the survival of TTTS cases after FLP is unknown.

The ductus venous (DV) acts as a pressure transducer and reflects atrial filling pressure and afterload from the ventricular end-diastolic pressure. In TTTS, diastolic dysfunction is often pronounced in the recipient because of increased ventricular pressure, which creates abnormal DV Doppler waveforms that reflect abnormal forward flow during passive diastolic filling (Turan, Turan, Sanapo, Rosenbloom et al., 2014). In contrast, hypovolemia in the donor twin causes the DV inlet to dilate (Wohlmuth et al., 2016), prolonging diastolic time intervals and causing abnormal flow during end-systolic relaxation in DV Doppler waveforms (Turan, Turan, Sanapo, Rosenbloom et al., 2014; Wohlmuth et al., 2016). DV Doppler waveforms reflect the cardiac function or the blood volume of fetuses; these waveforms change given that blood is carried through the anastomoses (Tachibana et al., 2015; Turan, Turan, Sanapo, Rosenbloom et al., 2014; Turan, Turan, Sanapo, Wilruth et al., 2014). In particular, the recipient twins with an

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**Cite this article:** Konno H and Murakoshi T. (2022) Effect of Superficial Anastomoses on Circulatory Dynamics in Twin-Twin Transfusion Syndrome. *Twin Research and Human Genetics* 25: 245–250. <https://doi.org/10.1017/thg.2022.38>



**Fig. 1.** Components of the ductus venosus Doppler waveform. The peak velocities were measured; during ventricular systole (S), end-systolic ventricular relaxation (v), early diastole (D), atrial systole (a)

increased pulsatility index of DV (DV-PI) and/or absent or reverse flow during atrial contraction of DV are expected to have increased their cardiac load (Takano et al., 2022).

This study aimed to evaluate how superficial anastomoses affect the circulatory dynamics of twin fetuses with TTTS in survival cases after FLP using DV Doppler waveforms.

## Materials and Methods

All TTTS patients who underwent FLP between 2006 and 2019 at our institution were included in this retrospective cohort study. We excluded cases of fetal demise after FLP.

DV flow velocity waveforms were recorded on the day of or one day before FLP and one day after FLP. The DV was visualized in either a mid-sagittal or an oblique transverse plane and recorded in the absence of fetal breathing movement. The examinations were performed using Voluson 730 Expert, E8 or E10 (GE Healthcare, WI, USA) by fetal medicine specialists. The sample volume conformed to the vessel diameter. We measured the DV-PI and peak velocities during ventricular systole (S), end-systolic ventricular relaxation (v), early diastole (D), and atrial systole (a); see Figure 1. We calculated velocity ratios, including the S/D, S/v, S/a, v/a, D/a, and v/D ratios. Based on a previous study, an absent a-wave was expressed as 1 cm/s and reversed a-waves were calculated as a ratio by adding the absolute a-wave value to the S, v, and D values (Turan, Turan, Sanapo, Rosenbloom et al., 2014). The *z* scores of the DV-PI and each ratio were converted according to the previously established reference ratios to eliminate the influence of gestational age (Turan, Turan, Sanapo, Wilruth et al., 2014). We defined that low *z* score of DV-PI and high *z* scores of all ratios were more desirable in past studies (Turan, Turan, Sanapo, Rosenbloom et al., 2014).

We identified and recorded the type of anastomosis as either AA, VV or AV, and recorded the number of anastomoses and direction of the AV anastomoses during FLP. AA and VV anastomoses are considered superficial anastomoses. We established a control group, with no superficial anastomoses, for comparison with the groups identified with AA anastomoses or VV anastomoses. Following delivery, we checked for occlusion of the intertwined vascular anastomoses by injecting colored dye or air into the umbilical vessels. None of the patients had residual anastomoses.

The chi-square test was used to analyze categorical variables, and a *t* test or Mann–Whitney test was used to analyze continuous variables, as required. Statistical significance was defined as  $p < .05$ . Statistical analyses were performed using SPSS Statistics version 16.0 J (SPSS Inc., Shibuya-Ku, Japan).

This study was approved by our institutional research review board, and informed consent was obtained from participating mothers.

## Results

In total, 115 TTTS placentas were analyzed. There were 26 and 11 cases of AA and VV anastomoses respectively. There were 84 cases in the control group, which had no superficial anastomoses. Six cases had both AA and VV anastomoses and were included in both groups. We compared the data of the examined groups with the corresponding data of the control group. These characteristics are shown in Table 1. There were significant differences in the body weight discordance before FLP between the AA or VV groups and the control group ( $p = .048$  and  $p = .047$  respectively). There was a significant difference in the birth weights of ex-donor infants in the AA group and infants in the control group ( $p = .045$ ). However, there were no statistically significant differences in other characteristics between the groups.

The presence of each vascular anastomosis according to TTTS stage is shown in Table 2. The presence of AA anastomoses was higher in TTTS stage I than in any other stage. The presence of other anastomoses was not significantly different according to the TTTS stage.

The *z* score of the DV-PI and *z* scores of the S/D, S/v, S/a, v/a, D/a, and v/D ratios before FLP are shown in Table 3. The *z* scores of the DV-PI of recipient twins were lower (better) in each group with AA and VV anastomoses than in the control group. The *z* scores of all ratios except for the S/D ratio (S/v, S/a, v/a, D/a and v/D) were higher (better) in recipient twins with AA anastomoses than in recipient twins without AA anastomoses ( $p = .006$ ,  $.000$ ,  $.001$ ,  $.000$  and  $.041$  respectively). The *z* scores of the S/a, v/a and D/a ratios were higher (better) in recipient twins with VV anastomoses than in recipient twins without VV anastomoses ( $p = .004$ ,  $.005$ , and  $.019$  respectively). There were no significant differences in all ratios for donor twins, regardless of the presence of superficial anastomoses.

The *z* scores of the DV-PI and *z* scores of the S/D, S/v, S/a, v/a, D/a and v/D ratios after FLP are shown in Table 4. The *z* scores of the DV-PI after FLP of recipient twins were lower (better) in the group with AA anastomoses than in the control group. The *z* scores of all ratios (S/D, S/v, S/a, v/a, D/a and v/D) were higher (better) in recipient twins with AA anastomoses than in those without AA anastomoses ( $p = .033$ ,  $.002$ ,  $.001$ ,  $.003$ ,  $.000$  and  $.036$  respectively). The *z* scores of the S/a and v/a ratios were higher (better) in recipient twins with VV anastomoses than in those without VV anastomoses ( $p = .023$  and  $.019$  respectively). There were no

**Table 1.** Patients' characteristics

	Control <i>n</i> = 84	With AA <i>n</i> = 26	<i>p</i> -value	With VV <i>n</i> = 11	<i>p</i> -value
Maternal age (years)	31.0 ± 5.6	30.0 ± 5.6	.313	30.6 ± 4.6	.817
Maternal height (cm)	158.4 ± 5.6	158.2 ± 4.2	.864	156.5 ± 4.4	.293
Maternal weight (kg)	54.6 ± 7.3	53.1 ± 5.8	.342	52.1 ± 5.5	.283
Nulliparity	40 (48%)	14 (54%)	.579	4 (36%)	.481
Onset of TTTS (weeks)	19.9 ± 2.5	20.6 ± 2.4	0.252	21.1 ± 2.1	0.148
GW at FLP (weeks)	20.5 ± 2.3	21.1 ± 2.5	0.314	21.7 ± 2.0	0.109
Body weight discordance (>25%) before FLP	49 (58%)	20 (77%)	0.048*	10 (91%)	0.047*
GW at delivery (weeks)	32.7 ± 2.7	31.5 ± 4.6	0.280	32.0 ± 4.0	0.652
Birth weight of ex-donor (g)	1563.8 ± 665.9	1256.7 ± 647.1	0.045*	1318.5 ± 526.4	0.245
Birth weight of ex-recipient (g)	1830.5 ± 700.4	1743 ± 751.3	.594	1753 ± 617.6	.729
FLP-delivery (weeks)	12.1 ± 5.2	10.5 ± 6.0	.174	10.3 ± 5.4	.274
DR > RD	25 (30%)	10 (38%)	.405	3 (27%)	1.000
AA	0	–	–	6 (55%)	.000*
VV	0	6 (23%)	.000*	–	–

Note: Data are presented as means ± standard deviations (SD) or numbers (%). VV, veno-venous anastomoses; AA, arterio-arterial anastomoses; DR, arteriovenous anastomoses from donor to recipient; RD, arteriovenous anastomoses from recipient to donor; DR > RD, a majority of DR compared to RD; TTTS, twin-twin transfusion syndrome; GW, gestational week; FLP, fetoscopic laser photocoagulation; FLP-delivery, time to delivery from FLP.

\**p* < .05.

**Table 2.** Presence of vascular anastomoses according to stages of TTTS

	I <i>n</i> = 23	II <i>n</i> = 29	III		IV <i>n</i> = 8	Total <i>n</i> = 115	<i>p</i> -value
			c <i>n</i> = 44	a <i>n</i> = 11			
VV	3 (13%)	2 (7%)	5 (11%)	1 (9%)	0 (0%)	11 (10%)	.813
AA	9 <sup>†</sup> (39%)	4 (14%)	7 (16%)	5 <sup>‡</sup> (45%)	1 (13%)	26 (23%)	.045*
AA + VV	3 (13%)	1 (7%)	1 (2%)	1 (9%)	0 (0%)	6 (5%)	.333
DR > RD	9 (39%)	8 (28%)	9 (20%)	6 (55%)	2 (25%)	34 (30%)	.185

Note: Data are presented as numbers (%). c, Stage III classical donor with nonvisible bladder and abnormal Doppler waveform; a, stage III atypical donor with visible bladder and abnormal Doppler waveform; TTTS, twin-twin transfusion syndrome; AA, arterio-arterial anastomoses; VV, veno-venous anastomoses; AA + VV, cases that have both AA and VV; DR, arteriovenous anastomoses to recipient from donor; RD, arteriovenous AV anastomoses to donor from recipient; DR > RD, a majority of DR compared to RD.

\**p* < .05; <sup>†</sup>Adjusted residual=2.0; <sup>‡</sup>Adjusted residual=1.8.

significant differences in all ratios for donor twins, regardless of the presence of superficial anastomoses.

## Discussion

In surviving TTTS cases after FLP, the DV-PI and all ratios were better in recipient twins with AA anastomoses than in those without. The a-wave-related ratios were better in recipient twins with VV anastomoses than in those without. Therefore, superficial anastomoses can reduce the venous return blood volume of the recipient twin, and AA anastomoses might not worsen the diastolic function in recipient twins before FLP.

In TTTS, recipient twins are more likely to present with abnormal forward flow during passive diastolic filling in DV Doppler waveforms (Turan, Turan, Sanapo, Rosenbloom et al., 2014). In this study, the S/D ratio was better after FLP in recipient

twins with AA anastomoses than in those without. The S/D ratio quantifies ventricular systolic to early passive diastolic filling and may better reflect diastolic filling abnormalities (Smrcek et al., 2005). Our results indicate that recipient twins with AA anastomoses are more likely to have improved diastolic filling after FLP than those without. AA anastomoses could help increase passive diastolic filling relative to ventricular systole and keep diastolic function from worsening in recipient twins.

All other ratios were better in recipient twins with AA anastomoses than in those without, both before and after FLP. In a previous study, v-wave abnormalities were more specific for myocardial relaxation and compliance issues, while D-wave abnormalities reflected global diastolic venous dysfunction (Turan, Turan, Sanapo, Rosenbloom et al., 2014). The S/v ratio is an indicator of forward flow during ventricular relaxation immediately following ventricular systole, and the v/D ratio represents the

**Table 3.** Z score of the DV-PI and each ratio before FLP

	Control before	AA before	p-value	VV before	p-value
<b>Recipient</b>					
DV-PI	1.92 (-7.90 to 24.58)	-0.34 (-4.90 to 11.81)	.000*	-1.72 (-4.54 to 4.10)	.014*
S/D	-1.39 (-3.30 to 2.97)	-1.34 (-2.79 to 0.47)	.191	-1.31 (-2.68 to 0.47)	.500
S/v	-1.35 (-4.33 to 0.46)	-0.63 (-2.61 to 0.90)	.006*	-0.74 (-2.58 to 0.23)	.071
S/a	-1.82 (-3.85 to 1.38)	-0.62 (-3.58 to 2.56)	.000*	0.17 (-2.15 to 0.81)	.004*
v/a	-1.51 (-3.37 to 9.51)	-0.47 (-3.13 to 2.16)	.001*	0.29 (-1.63 to 1.43)	.005*
D/a	-1.38 (-3.23 to 6.10)	0.33 (-3.00 to 6.55)	.000*	0.62 (-1.76 to 1.28)	.019*
v/D	-0.52 (-4.58 to 2.73)	0.48 (-2.03 to 1.21)	.041*	0.59 (-2.22 to 1.04)	.114
<b>Donor</b>					
DV-PI	-0.92 (-5.75 to 23.88)	-0.52 (-7.21 to 6.61)	.547	0.90 (-7.21 to 4.49)	.811
S/D	-0.43 (-2.78 to 1.95)	-0.29 (-2.41 to 1.95)	.437	-0.51 (-2.41 to 1.34)	.617
S/v	-0.74 (-3.99 to 1.57)	-1.15 (-2.82 to 1.27)	.382	-1.09 (-2.82 to 1.02)	.332
S/a	-0.29 (-3.62 to 2.82)	-0.35 (-4.29 to 2.31)	.612	-1.37 (-3.80 to 0.88)	.116
v/a	-0.18 (-3.18 to 2.56)	-0.22 (-4.02 to 1.77)	.749	-1.21 (-3.35 to 1.11)	.077
D/a	-0.25 (-3.03 to 7.54)	0.08 (-3.21 to 6.20)	.741	-1.26 (-3.21 to 0.68)	.049
v/D	-0.29 (-3.48 to 3.20)	-0.95 (-2.25 to 1.12)	.163	-0.73 (-2.36 to 0.53)	.146

Note: Data are presented as medians (ranges). AA, arterio-arterial anastomoses; VV, veno-venous anastomoses; DV-PI, ductus venous pulsatility index; FLP, fetoscopic laser photocoagulation; S, ventricular systole; D, early diastole; v, end-systolic ventricular relaxation; a, atrial systole.  
\* $p < .05$ .

subsequent passive diastolic forward flow (Turan, Turan, Sanapo, Rosenbloom et al., 2014). These abnormalities are mainly observed in fetuses with hydrops or right-sided heart lesions. Right ventricular strain is sometimes present in recipient twins in TTTS, and v-wave abnormalities are also observed. AA anastomoses may be beneficial in right ventricular strain in the recipient twin.

On the other hand, a-wave-related ratios (S/a, v/a, and D/a) were better in recipient twins with superficial anastomoses than in those without, both before and after FLP. These ratios were also better in recipient twins with VV anastomoses than in those without, both before and after FLP. The S/a ratio quantifies ventricular systolic to active diastolic filling, the v/a ratio quantifies late systolic to late diastolic filling (Kanzaki & Chiba, 1990), and the D/a ratio is a diastolic parameter related to the magnitude of forward flow during diastolic filling (Turan, Turan, Sanapo, Rosenbloom et al., 2014). Abnormalities in a-wave-related ratios were not predominant in any fetal condition (Turan, Turan, Sanapo, Rosenbloom et al., 2014). In the recipient twin, abnormal a-wave-related ratios might represent worse cardiac function or increased venous volume. Only a-wave-related ratios show abnormalities, indicating that late diastolic events play a role in abnormal DV-PI (Turan, Turan, Sanapo, Rosenbloom et al., 2014). Furthermore, the recipient twins with an increased DV-PI, absent

or reverse flow of a-wave are expected to have increased their cardiac load (Takano et al., 2022). Considering this, together with other ratios, recipient twins with AA anastomoses may have better cardiac function than those without anastomoses, and those with VV anastomoses may not have as much venous volume as those without VV anastomoses.

It is important to note that some ratios changed in the surviving recipient twins with superficial anastomoses. Superficial anastomoses could protect cardiac function and circulation in surviving recipient twins. Previous studies have indicated that superficial anastomoses have unstable characteristics. However, we believe that the role of superficial anastomoses is not unstable; they simply equalize the blood pressure between one twin and the other. Superficial anastomoses always work as AV anastomoses with blood pressure gradients when there are differences in blood pressure between twin fetuses. Superficial anastomoses can reduce the venous return blood volume of the recipient twin, and AA anastomoses might not worsen the diastolic function in recipient twins before FLP.

The limitations of this study include the small sample size and its retrospective, single-center design. This precluded further assessment and some bias inherent to this study design was unavoidable. Nonetheless, variability between examiners was

**Table 4.** Z-score of the DV-PI and each ratio after FLP

	Control Day 1	AA Day 1	p-value	VV day 1	p-value
Recipient					
DV-PI	2.51 (-7.61 to 24.08)	-0.13 (-7.47 to 4.70)	0.000*	0.59 (-4.95 to 3.91)	0.078
S/D	-1.09 (-3.28 to 4.48)	-0.78 (-2.35 to 1.95)	0.033*	-0.95 (-2.32 to 115.7)	0.524
S/v	-2.02 (-3.95 to 0.45)	-0.92 (-2.91 to 1.34)	0.002*	-1.43 (-3.17 to 6.76)	0.258
S/a	-2.24 (-3.79 to 0.69)	-0.92 (-3.38 to 1.59)	0.001*	-1.16 (-2.54 to 2.74)	0.023*
v/a	-1.76 (-3.34 to 1.65)	-0.67 (-2.79 to 0.95)	0.003*	-0.66 (-2.27 to 0.74)	0.019*
D/a	-1.84 (-3.18 to 7.12)	-0.30 (-2.44 to 5.46)	0.000*	-1.00 (-2.19 to 0.09)	0.088
v/D	-1.31 (-3.69 to 3.35)	-0.44 (-2.56 to 2.68)	0.036*	-0.83 (-3.00 to -0.23)	0.775
Donor					
DV-PI	1.98 (-7.611 to 20.70)	0.55 (-7.47 to 19.63)	0.070	1.34 (-7.21 to 6.03)	0.577
S/D	-0.58 (-2.58 to 4.40)	-0.42 (-1.92 to 1.95)	0.611	-0.15 (-1.92 to 2.45)	0.503
S/v	-1.86 (-3.92 to 1.04)	-1.88 (-4.28 to -0.06)	0.868	-1.87 (-3.96 to -0.17)	0.455
S/a	-1.82 (-3.48 to 2.35)	-1.47 (-6.40 to 1.50)	0.217	-1.65 (-2.84 to -0.13)	0.715
v/a	-1.22 (-3.02 to 1.85)	-0.80 (-6.50 to 9.24)	0.109	-0.59 (-2.43 to 9.20)	0.385
D/a	-1.72 (-2.96 to 6.88)	-1.19 (-2.86 to 6.03)	0.078	-1.38 (-2.77 to -0.02)	0.927
v/D	-1.53 (-4.18 to 0.87)	-1.56 (-4.51 to 0.30)	0.779	-1.81 (-4.19 to -0.08)	0.361

Note: Data are presented as medians (ranges).

AA, arterio-arterial anastomoses; VV, veno-venous anastomoses; DV-PI, ductus venosus pulsatility index; FLP, fetoscopic laser photocoagulation; S, ventricular systole; D, early diastole; v, end-systolic ventricular relaxation; a, atrial systole.

\* $p < .05$ .

minimized. Additionally, some cases had both AA and VV anastomoses. It remains unclear how AA and VV anastomoses interact for the circulation of TTTS. Furthermore, the extent to which each parameter of DV Doppler waveform accurately reflects fetal cardiac function is still debatable. These points should be examined in future work.

In conclusion, superficial anastomoses reduced the blood volume of recipient twins and AA anastomoses protected the diastolic cardiac function in recipient TTTS twins before FLP. Thus, the role of superficial anastomoses in TTTS is to balance the blood pressure between the donor and recipient twins.

**Data Availability Statement.** The data that support the findings of this study are available from the corresponding author upon reasonable request.

**Financial Support.** This research received no specific grant from any funding agency, commercial or not-for-profit sectors.

**Conflicts of Interest.** None.

**Ethical Standards.** The authors assert that all procedures contributing to this work comply with the ethical standards of the relevant national and institutional committees on human experimentation and with the Helsinki Declaration of 1975, as revised in 2008.

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