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Original Article

Serum and pulmonary vascular endothelial growth factor/receptors and haemodynamic measurements in cyanotic congenital heart disease with decreased pulmonary blood flow

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Abstract Tetralogy of Fallot is the most common cyanotic congenital heart disease with decreased pulmonary blood flow. Right-to-left shunt and infundibular pulmonary stenosis in this disease lead to a decrease in arterial O_2 saturation. Hypoxia is a strong stimulus for angiogenesis; however, the reason for insufficiency in the pulmonary vascular growth in patients despite chronic arterial hypoxia is still not known. This study was planned considering that the impairment in vascular endothelial growth factor-receptor relationship or the vascular endothelial growth factor-receptor deficiency in the pulmonary vascular bed during development may cause insufficiency of pulmonary vascular growth. A total of 24 patients were grouped as cyanotic – including 13 patients with tetralogy of Fallot – and acyanotic – including 11 patients with left-to-right shunt lesions. During cardiac catheterisation, vascular endothelial growth factor measurements were performed; and oxygen saturations, pressures, and haemoglobin levels were measured. Perioperative lung biopsy for vascular endothelial growth factor receptors was performed in the cyanotic group. Vascular endothelial growth factor of the aorta was higher in the acyanotic group. There was a significant negative correlation between vascular endothelial growth factor levels and aortic O_2 saturation in the cyanotic group ($p < 0.05$). Vascular endothelial growth factor tissue staining was negative in 11 out of 13 (84.6%) patients. KDR/Flk-1 receptor was positive in four out of 13 (30.7%) patients; Flt-1 receptor was positive in six out of 13 (46.1%) patients. Vascular endothelial growth factor values were found to be lower than those of the acyanotic patients in this study. Low serum vascular endothelial growth factor levels of the cyanotic group, in spite of the hypoxia, demonstrated the importance of studying vascular endothelial growth factor tissue levels and vascular endothelial growth factor receptors in these patients.

Keywords: KDR/Flk-1; Flt-1; shunt lesions

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HYPOXIA IS THE MOST POWERFUL STIMULUS FOR vascular endothelial growth factor release.[1–4] Elevated serum vascular endothelial growth factor levels were reported in cyanotic congenital heart disease compared with acyanotic and healthy groups, and secondary changes to hypoxia in the pulmonary vascular bed were shown to be related to vascular endothelial growth factor.[5–8] In our previous study, we showed that vascular endothelial growth factor was high in the shunt lesions with a pulmonary vascular resistance of greater than 2 units per squared metre, and our findings reported that pulmonary vascular changes were related to vascular endothelial growth factor.[9]

In spite of the chronic arterial hypoxia in cyanotic congenital heart disease with decreased pulmonary blood flow, the reason for inadequate pulmonary vascular growth is unknown. Tetralogy of Fallot is

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the most common cyanotic congenital heart disease with decreased pulmonary blood flow.[10] The shunt (from right to left), which is produced depending on the degree of infundibular pulmonary stenosis in these patients, leads to a decrease in arterial O_2 saturation. However, the reason for underdevelopment/maldevelopment of pulmonary vascular growth in these patients is unknown. The lack of contribution of vascular endothelial growth factor to pulmonary vascular growth, which is expected to increase because of hypoxia in cyanotic congenital heart disease with decreased pulmonary blood flow, suggests the unresponsiveness of the target organ in these patients.

The deficiency of vascular endothelial growth factor receptor in the pulmonary vascular bed or the impaired relationship between the vascular endothelial growth factor and the receptor during growth may be responsible for the impaired pulmonary vascular growth and the lack of an increase in pulmonary flow.

In this study, we assessed whether the impaired pulmonary vascular growth is due to pulmonary vascular endothelial growth factor/receptor dysfunction in spite of the chronic arterial hypoxia in cyanotic congenital heart disease.

Material and method

The study consisted of 24 patients who had been diagnosed with cyanotic and acyanotic congenital heart disease and had undergone cardiac catheterisation and angiography at the Department of Pediatric Cardiology, Gazi University Faculty of Medicine, and were operated on at the Department of Cardiovascular Surgery of the same institution. The patients were grouped as cyanotic and acyanotic groups. There were 13 patients with tetralogy of Fallot in the cyanotic group, and there were a total of 11 patients in the acyanotic group, including four patients with atrial septal defect, four patients with ventricular septal defect, and three patients with patent ductus arteriosus. Blood samples were obtained from superior caval vein, pulmonary artery, and the aorta from patients with tetralogy of Fallot during cardiac catheterisation; in addition, blood samples were obtained from the pulmonary artery and aorta from patients with acyanotic disease. Measurements of the vascular endothelial growth factor; oxygen saturations of the pulmonary artery, pulmonary vein, superior caval vein, and the aorta; and pressures and haemoglobin levels were recorded. Perioperative lung biopsy of the peripheral right lobe was performed in 13 patients with tetralogy of Fallot after thoracotomy, and prior to cardiorespiratory pump and the material was sent to the pathology laboratory in formaldehyde and kept as paraffin sections. The vascular endothelial growth factor was measured in serum samples using enzyme-linked immunosorbent assay and R&D Systems, Quantikine Human VEGR Immunoassay ELISA kit (Quantikine Human VEGR Immunoassay Lot: 224353, Catalog No. DVE00, R&D Systems, Minneapolis, MN, USA). For the immunohistochemical method, sections of 4-micron thickness were de-paraffinised in the incubator at 56°C. They were left in 3% hydrogen peroxide for 10 minutes to block endogenous peroxidase, washed with phosphate-buffered saline, and treated in the microwave within 0.01 molar sodium citrate buffer for Flt-1 and KDR/Flk-1 antibodies and within ethylenediamine tetra acetic acid for vascular endothelial growth factor antibody for a total of 20 minutes. Immunohistochemical staining was performed using the streptavidine–biotin–peroxidase method. The primary antibodies – vascular endothelial growth factor, clone VG1, ready-to-use; Flt-1, ready-to-use; KDR/Flk-1, ready-to-use (Neomarkers, Fremont, CA, USA) were applied so as to cover the sections, and kept at room temperature for 2 hours and incubated with 3-amino-9-ethylcarbazole substrate to maintain visualisation with stain. The sections were covered with Mayer's haematoxyline as the base staining. Angiosarcoma sections were used as positive tissue controls for the three antibodies. Cytoplasmic staining was accepted as positive. Negative control staining was performed, in which there was no primary antibody. The rate of stained cells to non-stained cells was calculated.

Statistics

The results were analysed using the Mann–Whitney U test and the Spearman correlation analysis. The level of significance was set as less than 0.05. The results were given as mean plus or minus standard deviation (minimum value–maximum value).

The families of patients gave informed consent and the study was approved by the Ethics Committee of the Faculty of Medicine (Date: 27.12.2004, No: B-30.2.0.01.00-0089). Vascular endothelial growth factor serum, vascular endothelial growth factor tissue, KDR/Flk-1, and Flt-1 kit were provided by the support of the Scientific Research Project of Gazi University, no: 1/2004-30.

Results

The study included 24 patients with cyanotic congenital heart disease – cyanotic group – and acyanotic congenital heart disease – acyanotic group. There were 13 patients with tetralogy of Fallot in the

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Table 1. Haemoglobin, aortic O_2 saturation, and vascular endothelial growth factor levels of the cyanotic group.

Patients	Gender	Age (months)	Haemoglobin (g/dl)	Aortic O_2 saturation (%)	Vascular endothelial growth factor-superior caval vein (pg/ml)	Vascular endothelial growth factor-aorta (pg/ml)	Vascular endothelial growth factor-pulmonary artery (pg/ml)
1. HBU	F	52	16.3	73	27	28	–
2. MG	M	30	12.3	44	42.9	34.4	39
3. KA	M	48	13.1	83	25.4	20	24
4. MFK	M	24	16.7	75	20	22.4	20
5. ST	M	48	14.2	64	138.5	143	113
6. ZO	M	36	19	16	137.5	111.4	141.5
7. MI	M	18	11.8	78	24	25.4	24
8. MA	M	36	12.0	72	22.4	24	17.8
9. SK	M	14	13.9	68	29.9	27	31.4
10. ZA	F	24	14.1	18	248.6	164.2	186.8
11. BA	F	16	16.3	56	27	28	27
12. NV	M	18	11.5	44	–	17.8	24
13. TE	F	13	16	52	106.8	127.9	90.8

cyanotic group, and 11 patients in the acyanotic group, including four patients with atrial septal defect, four patients with ventricular septal defect, and three patients with patent ductus arteriosus.

The 13 patients in the cyanotic group included four female and nine male patients, and the 11 patients in the acyanotic group included eight female and three male patients. The mean age was 29.0 ± 13.8 (13–52) months in the cyanotic group and 48.0 ± 30.7 (9–108) months in the acyanotic group, and the difference was not significant ($p > 0.05$).

Table 1 shows the haemoglobin, aortic O_2 saturation, and vascular endothelial growth factor levels of the cyanotic group.

Table 2 shows the haemodynamic measurements and vascular endothelial growth factor levels of the acyanotic group.

The haemoglobin values [14.4 ± 2.30 (11.5–19) grams per decilitre] of the cyanotic group were significantly higher than those of the acyanotic patients, and aortic O_2 saturation [57.1 ± 21.71 (16–83%)] was significantly lower than that of the acyanotic group ($p < 0.05$; Table 3).

The vascular endothelial growth factor-aorta values of the acyanotic patients were significantly higher than those of the cyanotic group ($p < 0.05$); the vascular endothelial growth factor-pulmonary artery values were also higher than those of the cyanotic group; however, it was not statistically significant ($p > 0.05$). The vascular endothelial growth factor values were compared within the groups and there was no difference between vascular endothelial growth factor-aorta values and vascular endothelial growth factor-pulmonary artery values ($p > 0.05$; Table 4).

Blood samples were obtained from the cyanotic group to measure vascular endothelial growth factor-superior caval vein. The vascular endothelial growth factor-superior caval vein level was 122 ± 147.93 (20–429) picograms per millilitre, and there was a significant negative correlation with aortic O_2 saturation ($p < 0.05$, $r = -0.848$; Fig 1); however, there was no correlation between vascular endothelial growth factor-superior caval vein and haemoglobin levels.

The vascular endothelial growth factor-aorta level of the cyanotic group was 59.5 minus or plus 54.8 (17.8–164.2) picograms per millilitre, and the vascular endothelial growth factor-pulmonary artery level was 61.5 minus or plus 57.2 (17.8–186.8) picograms per millilitre; there was no correlation with haemoglobin levels, although there was a negative correlation with aortic O_2 saturation (Fig 2).

The vascular endothelial growth factor-aorta level of the acyanotic patients was 222.9 minus or plus 219.9 (24–560.9) picograms per millilitre, and the vascular endothelial growth factor-pulmonary artery level was 128.56 minus or plus 161.96 (21.4–493) picograms per millilitre; there was no significant relationship between vascular endothelial growth factor-aorta and vascular endothelial growth factor-pulmonary artery and haemoglobin and aortic O_2 saturation.

Table 5 presents the immunohistochemical staining results of biopsy materials.

Vascular endothelial growth factor-tissue staining was negative in 11 out of 13 patients (84.6%). Figure 3 shows positive vascular endothelial growth factor tissue staining of patient number 11.

KDR/Flk-1 receptor was positive in four out of 13 patients (30.7%); there was 20% staining in

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Table 2. Haemodynamic measurements and vascular endothelial growth factor levels of the acyanotic group.

Patients (diagnoses)	Gender	Age (months)	Hb (g/dl)	Aortic O_2 saturation (%)	Pulmonary flow/systemic flow	Pulmonary resistance (U/m^2)	Pulmonary artery systolic pressure (mmHg)	Pulmonary artery mean pressure (mmHg)	Vascular endothelial growth factor-aorta (pg/ml)	Vascular endothelial growth factor-pulmonary artery (pg/ml)
1. SA ventricular septal defect	F	24	13.4	92	2.42	4.7	60	44	24	21.4
2. TT ventricular septal defect	M	18	11.7	89	4.0	1.8	46	35	78.2	88.7
3. AI ventricular septal defect	F	60	12.0	96	1.09	1.7	28	15	444.7	493
4. ST atrial septal defect	F	24	10.8	97	1.9	1.4	30	18	550.3	29.9
5. AB atrial septal defect	M	72	12.4	99	3.3	0.6	31	23	560.9	45
6. HU atrial septal defect	F	54	13.9	97	1.6	0.5	23	16	24	42
7. EC atrial septal defect	F	9	11.4	97	1.1	1.6	28	16	413	395
8. AP patent ductus arteriosus)	M	68	11.8	97	1.76	0.4	72	60	140	143
9. HH patent ductus arteriosus	F	108	13.4	97	1.1	0.2	34	25	69.1	51
10. SB patent ductus arteriosus)	F	24	10.5	98	1.9	1.2	25	7	106.2	97.8
11. BA ventricular septal defect	F	72	13.2	95	1.5	1.8	25	17	24	25.5

three patients and 10% in one patient. Figure 4 shows positive KDR/Flk-1 receptor staining (20%) in patient number 8.

Flt-1 receptor was positive in six out of 13 patients (46.1%); there was extensive receptor positivity (70%) in one patient, there was 30% staining in one patient, and 20% staining in four patients. Figure 5 shows staining results of patient number 7 whose Flt-1 receptor was positive (70%).

Discussion

Right-to-left shunt and decreased pulmonary blood flow in patients with tetralogy of Fallot are the main causes of reduced arterial O_2 saturation and systemic hypoxia.[10] In spite of hypoxia, patients with tetralogy of Fallot do not have adequate changes in the pulmonary vascular bed and do not respond to hypoxia. Previous studies in patients with cyanotic congenital heart disease have shown that the elevated levels of vascular endothelial growth factor do not contribute to pulmonary vascular growth in patients with tetralogy of Fallot, suggesting that vascular endothelial growth factor is ineffective at the tissue level in these patients, and bring into mind that there is a deficiency in tissue vascular endothelial growth factor receptors or an impaired relationship between vascular endothelial growth factor and vascular endothelial growth factor-receptors.

Our results showed a negative correlation between superior caval vein, aorta, pulmonary artery vascular endothelial growth factor levels, and aortic O_2 saturation. Vascular endothelial growth factor is affected by the severity of unsaturation. When vascular endothelial growth factor levels of the cyanotic and acyanotic groups were compared, there was no significant difference between the cyanotic and acyanotic groups in terms of vascular endothelial growth factor-pulmonary artery; on the other hand, vascular endothelial growth factor-aorta was higher in the acyanotic group. Despite the fact that aortic O_2 saturation was low in patients with tetralogy of Fallot, the expected significant elevation was not observed in vascular endothelial growth factor-aorta.

Highest vascular endothelial growth factor-aorta levels were found in the cyanotic group (range: 164.2–111.4 picograms per millilitre) in four patients with tetralogy of Fallot (patient numbers: 10, 5, 13, and 6). Of these, two patients (patient numbers: 10 and 6) had the lowest aortic O_2 saturation and they underwent emergency heart catheterisation owing to deterioration in the general status. Vascular endothelial growth factor tissue and receptor staining were negative in these patients. They (patient numbers: 10, 6, 5, and 13) also had the

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Table 3. Haemoglobin and aortic O_2 saturation values of patients (mean minus or plus standard deviation).

	Cyanotic group	Acyanotic group	p
Haemoglobin (g/dl)	14.4 ± 2.30	12.2 ± 1.12	0.015
Aortic O_2 saturation (%)	57.1 ± 21.71	95.8 ± 2.89	0.0001

Table 4. Vascular endothelial growth factor-aorta and vascular endothelial growth factor-pulmonary artery levels of patients.

	Cyanotic group (mean ± SD)	Acyanotic group (mean ± SD)	p
Vascular endothelial growth factor-aorta (pg/ml)	59.5 ± 54.8	222.9 ± 219.9	< 0.05
Vascular endothelial growth factor-pulmonary artery (pg/ml)	61.5 ± 57.2 $p > 0.05$	128.5 ± 161.9 $p > 0.05$	> 0.05

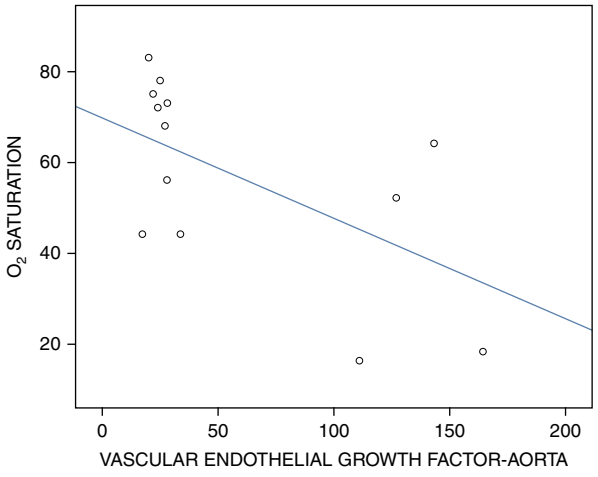

Figure 1.
Negative correlation between aortic O_2 saturation and vascular endothelial growth factor-superior caval vein levels in the cyanotic group.

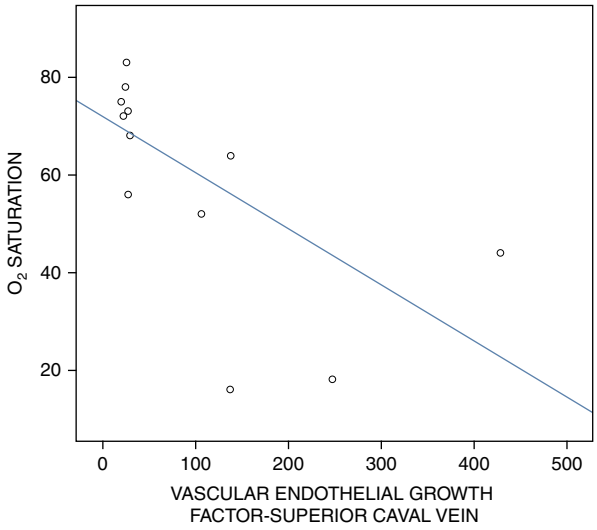

Figure 2.
*(**a**) Negative correlation between aortic O_2 saturation and vascular endothelial growth factor-aorta levels in the cyanotic group.*

highest vascular endothelial growth factor-pulmonary artery levels (range: 90.8–186.8 picograms per millilitre); however, the vascular endothelial growth factor-pulmonary artery and vascular endothelial growth factor-aorta levels were low (range: 17.8–39 picograms per millilitre) in the remaining patients.

Similar to vascular endothelial growth factor-aorta and vascular endothelial growth factor-pulmonary artery levels, the vascular endothelial growth factor-superior caval vein levels were the highest in patients with numbers 5, 6, 10, and 13. The severity of pulmonary stenosis determines pulmonary flow, the amount of the right-to-left shunt, and related decreased arterial O_2 saturation. In two patients with the lowest aortic O_2 saturation, pulmonary hypoplasia was the most severe and hypoxia was the most prominent, and vascular endothelial growth factor was high in the venous and arterial systems and in pulmonary circulation, suggesting that vascular endothelial growth factor was produced in non-pulmonary systems because of hypoxia. In spite of high vascular endothelial growth factor levels, there was no increase in pulmonary flow supporting our hypothesis that there was a problem in vascular endothelial growth factor response of the tissue.

In contrast with the results of previous studies,[5–8] the vascular endothelial growth factor levels of patients with tetralogy of Fallot were lower than those of the acyanotic patients. The expected increase in vascular endothelial growth factor level due to hypoxia could not be seen in patients with tetralogy of Fallot. Our results conflict with the

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Table 5. Immunohistochemical staining percentages of vascular endothelial growth factor-tissue and vascular endothelial growth factor receptors (KDR/Flk-1, Flt-1) in patients with tetralogy of Fallot.

Patient	Vascular endothelial growth factor-tissue (%)	KDR/Flk-1 (%)	Flt-1 (%)
1. HBU	0	10	20
2. MG	0	0	20
3. KA	0	0	0
4. MFK	0	0	0
5. ST	0	0	0
6. ZO	0	0	0
7. MI	0	20	70
8. MA	10	20	20
9. SK	0	0	0
10. ZA	0	0	0
11. BA	80	0	30
12. NV	0	20	20
13. TE	0	0	0

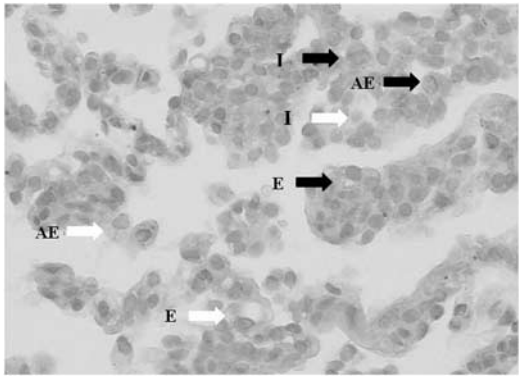

Figure 3.
Alveoli epithelial cells, interstitial cells, and endothelial cells have vascular endothelial growth factor expression (cells with red cytoplasma). The black arrow shows vascular endothelial growth factor positive cells, and white arrow shows vascular endothelial growth factor negative cells (streptavidine–biotin–peroxidase, AEC × 200). AE = alveoli epithelial cells; E = endothelial cells; I = interstitial cells.

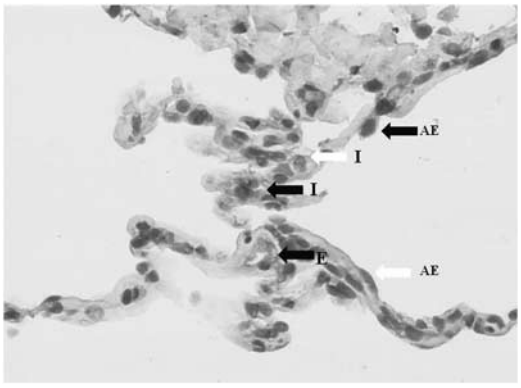

Figure 4.
The black arrow shows KDR/Flk-1 positive alveoli epithelial cells, interstitial cells, and endothelial cells; the white arrow shows KDR/Flk-1 negative cells (streptavidine–biotin–peroxidase, AEC × 200). AE = alveoli epithelial cells; E = endothelial cells; I = interstitial cells.

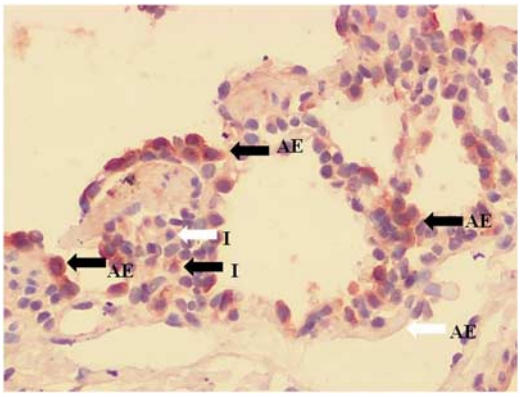

Figure 5.
The black arrow shows Flt-1 positive alveoli epithelial cells and interstitial cells; the white arrow shows the Flt-1 negative cells (streptavidine–biotin–peroxidase, AEC × 200). AE = alveoli epithelial cells; E = endothelial cells; I = interstitial cells.

results of previous studies. Himeno et al. showed that serum vascular endothelial growth factor was significantly elevated in patients with cyanotic congenital heart disease compared with healthy controls. However, the majority of cyanotic patients – 51 out of 80 patients – in this study were patients with increased pulmonary blood flow or patients with cyanotic congenital heart disease who had undergone shunt operations.[6] Starnes et al. reported that serum vascular endothelial growth factor was higher in cyanotic congenital heart disease compared with acyanotic patients and that hypoxia was the reason for the increased vascular endothelial growth factor in these patients. The majority of the patients – that is, 18 out of 22 patients – in the study by Starnes et al. were similar to those in the study by Himeno et al.[10] Ootaki et al. also reported that the vascular endothelial growth factor level was significantly elevated in the group with cyanotic heart disease. However, most patients had collaterals to increase pulmonary circulation.[7]

Our patients were a homogeneous group with cyanotic congenital heart disease with decreased pulmonary blood flow without abnormal collateral circulation. The increased pulmonary flow in patients in other studies may cause more increase in vascular endothelial growth factor levels.

The increased pulmonary blood flow in congenital heart disease with the left-to-right shunt leads to pulmonary plexogenic arteriopathy, which

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is characterised by concentric laminar intimal fibrosis, fibrinoid necrosis, and plexiform lesions.[11] In a study by Zengin and Tunaoglu, cyanotic and acyanotic patients were grouped in two groups according to pulmonary artery pressure. Vascular endothelial growth factor was found to be higher in the group with the mean pulmonary artery pressure of greater than 30 millimetres of mercury; however, this increase was not significant. The patients were grouped in two groups according to pulmonary resistance, and the vascular endothelial growth factor increase in the group with pulmonary resistance greater than 2 units per squared metre was significant.[9]

Vascular endothelial growth factor is accepted to be responsible for pulmonary vascular changes. The increased serum levels are accepted as indicators of irreversible vascular damage due to pulmonary hypertension. It is clear that vascular endothelial growth factor increases in response to increased pulmonary blood flow and is responsible for the pulmonary vascular changes.

There was no relationship between vascular endothelial growth factor-aorta and vascular endothelial growth factor-pulmonary artery levels and pulmonary flow/systemic flow, pulmonary resistance, systolic pulmonary artery pressure, mean pulmonary artery pressure in acyanotic patients. The lack of a relationship between vascular endothelial growth factor levels and pulmonary haemodynamic measures may be due to the low pulmonary artery pressure values and pulmonary resistance (less than 2 units per squared metre) in the patients. As our patients in the acyanotic group were at the beginning of the process of pulmonary hypertension according to haemodynamic measurements and as they had no persistent changes in pulmonary resistance, there was no correlation between serum vascular endothelial growth factor and haemodynamic measurements.

Pleural dissection is not performed during thoracotomy in acyanotic patients different from patients with tetralogy of Fallot; since it is not ethical to perform an additional intervention for biopsy, these patients did not undergo biopsy. Further studies are needed on tissue vascular endothelial growth factor and receptors in patients with pulmonary hypertension and with Rp greater than 2 units per squared metre and mean pulmonary artery pressure greater than 30 millimetres of mercury.

There are studies showing that one of the vascular endothelial growth factor receptors, namely KDR/Flk-1, is an initial stimulus in the differentiation of the haemangioblast.[2] Embryo mice with KDR/Flk-1 deficiency died on the eighth day and it was noticed that development of both endothelial and haematopoietic systems had fallen behind. In this study, four out of 13 patients have shown KDR/Flk-1 positivity.[4]

Flt-1 deficiency also caused death in embryo mice in the same period. Although haematopoietic precursor cells were formed in these embryos, there was no maturation in tube formation and functions, and it was found that the precursor cells were abnormally reproduced endothelial cells.[4]

The Flt-1 receptor, which was shown to be effective in the stages of maturation and preservation of vascular integrity in angiogenesis, was positive in six out of 13 patients (46.1%). In two out of six patients with Flt-1 receptor positivity, vascular endothelial growth factor tissue staining, which shows the production of vascular endothelial growth factor in the tissue, was positive and in four patients it was negative. In one (patient number 7) out of four patients with negative vascular endothelial growth factor tissue staining, there was extensive (70%) Flt-1 positivity, and the KDR/Flk-1 receptor was also positive in this patient. Kdr/Flk-1 was positive in four out of six patients with Flt-1 positivity (Table 5).

We observed that patients had variable degrees of vascular endothelial growth factor production and impaired development of tissue receptor. It is clear that vascular endothelial growth factor is needed for normal lung maturation and that serum vascular endothelial growth factor is affected by the production in the lung tissue. In our patients, the vascularity of the lung was not adequately developed because of the impairment in production and interaction, and therefore the expected increase in serum vascular endothelial growth factor was not found to be different from other studies.

In contrast with other studies, the fact that we did not find the serum vascular endothelial growth factor-aorta and vascular endothelial growth factor-pulmonary artery levels as high, and that we did not detect vascular endothelial growth factor and its receptors in tissues in 53.8% of patients may be explained by the impairment in expression of vascular endothelial growth factor and its receptors at varying degrees in our patients with tetralogy of Fallot.

The impaired production of vascular endothelial growth factor and its receptors is thought to be the cause of impairment in the growth and remodelling of the vascular bed detected in the morphometric analysis of lungs in children with CHD.

Arteries have the most rapid numeric and structural growth in infants. In spite of the growth in alveoli, the ratio of alveoli:arteries changes from a ratio of 20:1 in the neonatal period to a ratio of 8:1 in early childhood and this ratio persists. The deficiency of vascular endothelial growth factor and its receptors in infants may cause inadequate development of pulmonary vessels.

Studies on curative angiogenesis have gained speed after recognition of tumour regression and its metastasis by pharmacological inhibition of vascular endothelial growth factor.[12] In our study, patients with tetralogy of Fallot underwent the shunt operation before total correction in order to empower the vascular bed of the lungs. Identification of the role of vascular endothelial growth factor in the aetiopathogenesis of pulmonary hypoplasia may put forth the administration of vascular endothelial growth factor to patients via the pulmonary route. Identification of gene expression of the vascular endothelial growth factor/receptors, administration of vascular endothelial growth factor in infants, or provision of gene expression of vascular endothelial growth factor/receptors may spare patients from shunt operations and may reduce the post-operative mortality and morbidity after correction.

Further studies are needed on tissue vascular endothelial growth factor and receptors in patients with pulmonary hypertension and with pulmonary resistance of greater than 2 units per squared metre and mean pulmonary artery pressure of greater than 30 millimetres of mercury.

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