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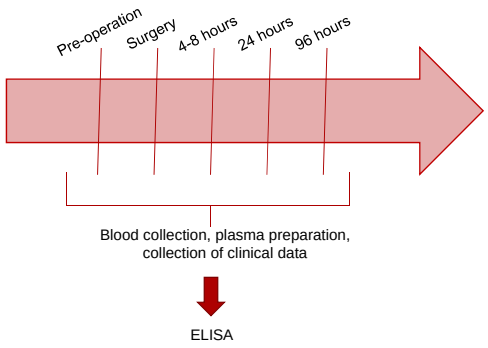
Introduction

- Coronary artery disease (CAD) causes local hypoxia due to reduced blood flow
- Hypoxic conditions are known to induce vascular endothelial growth factor (VEGF) production, a key contributor to angiogenesis
- The purpose of this study was to determine the potential of VEGF as a marker of myocardial stress in subjects with CAD undergoing coronary artery bypass grafting (CABG) surgery

Methods

- Research was performed in accordance with study protocols approved by Maine Medical Center Institutional Review Board
- The study cohort consisted of plasma samples from 73 patients undergoing CABG surgery at Maine Medical Center (MMC) in Portland, ME
- Plasma samples were collected prior to operation (pre-op), during surgery, and 4-8, 24 and 96 hours following surgery
- VEGF concentration was determined using a DuoSet enzyme-linked immunosorbent assay (ELISA) kit (R&D Systems, sensitivity range 31.3-2000 pg/mL)
- Undetectable levels of VEGF (<31.3 pg/mL) were assigned a concentration equal to one-half of the lowest calibration point (15.6 pg/mL)
- All statistical analyses were performed in GraphPad Prism and a p-value <0.05 was considered statistically significant

Study Timeline



Hypothesis

Due to local hypoxic conditions during surgery, we hypothesize that circulating VEGF levels increase immediately following CABG surgery. However, at time points following surgery, we expect a decrease in VEGF, indicating successful revascularization.

Results

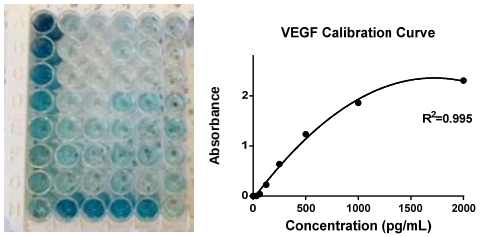


Figure 1. Determining concentration of VEGF in plasma samples

Left Representative VEGF ELISA plate. Lane 1 contains standard concentrations, and lanes 2 through 6 contain subject plasma samples

Right Standards were plotted in GraphPad Prism and plasma VEGF concentrations were back-calculated based on the calibration curve

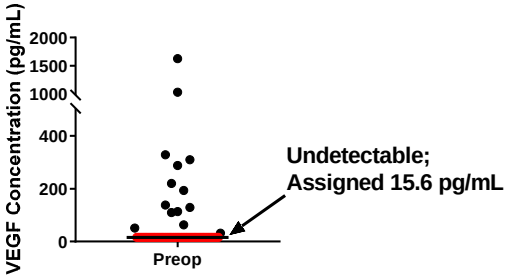


Figure 2. Subject expression of VEGF at pre-operative time point

The majority of patients (69.7%) did not have detectable levels of VEGF at any time point (red). VEGF protein was characterized by interindividual variability (CQD=34%). Therefore, subjects with detectable levels of VEGF at the pre-operative time point (black) were considered VEGF expressors and subjected to further statistical analysis.

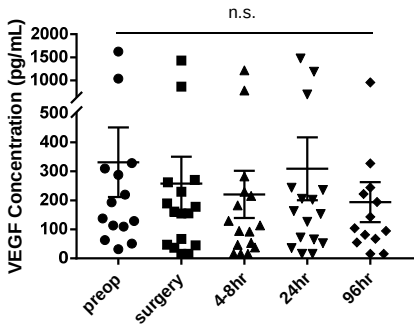


Figure 3. VEGF levels do not differ between time points analyzed

VEGF expression for the 16 subjects with detectable VEGF levels was subjected to Friedman's test and Dunn's multiple comparisons test to determine statistical significance. Analysis incorporated 11 subjects due to missing data at one or more time points for n=5 subjects. We did not observe any significant changes in VEGF expression across the five time points tested.

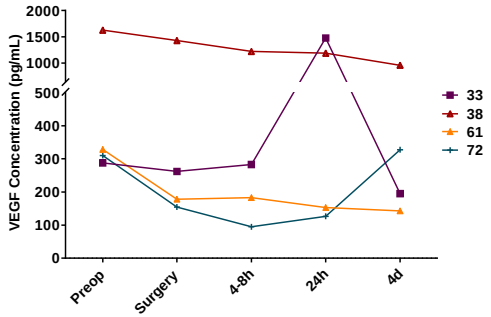


Figure 4. Representative data demonstrating different changes in VEGF expression following surgery

VEGF expression in response to CABG surgery differed within the study cohort. Although some individuals showed no changes following surgery, others were characterized by dynamic changes in VEGF expression. These changes included a decrease, an increase, or an increase followed by a decrease in VEGF expression.

Table 1. Demographic data of VEGF expressors is not significantly different from those with undetectable levels

Study Subject Demographics	VEGF Expressors	Undetectable	p-value
Number of subjects	16	50	
Age*	66 ± 10	63 ± 11	0.47‡
Female†	5 (31%)	20 (40%)	0.57§
Male†	11 (69%)	30 (60%)	
BMI*	32.3 ± 7.5	29.6 ± 5.8	0.18‡
Aortic cross clamp time (min)*	85 ± 28	82 ± 30	0.55‡
CPB time (min)*	100 ± 34	105 ± 35	0.60‡
Smoking history†	8 (50%)	36 (72%)	0.13§
HbA1c %*	7.0 ± 1.9	6.7 ± 1.8	0.67‡
EF<50%†	2 (13%)	12 (24%)	0.49§

*mean ± SD, †number of research subjects (%), BMI=body mass index, CPB=cardiopulmonary bypass, HbA1c=hemoglobin A1c, EF=ejection fraction
‡Mann-Whitney t-test, §Fisher exact test

Conclusions

- Plasma levels of VEGF are characterized by interindividual variability
- Individual VEGF expression appears to vary in response to CABG surgery
- CABG surgery did not induce changes in the level of circulating VEGF, limiting its potential use as a biomarker of cardiometabolic stress in CABG patients

Future Directions

- Determine Hypoxic Inducible Factor 1α (a transcription factor for VEGF) expression by ELISA
- Investigate the biological activity of VEGF in CAD patients
- Examine potential relationships with cytokine, and clinical data and outcomes
- Understand if patient subpopulations (diabetes, heart failure) have varying VEGF levels and if they are impacted clinically

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