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ORIGINAL RESEARCH

Assessing Endothelial Function: Flow-Mediated Dilation as a Surrogate Marker for Cardiovascular Disease Risk

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Abstract

Introduction: Cardiovascular disease (CVD) accounts for a significant percentage of global deaths and emphasizes the importance of early detection for prevention. Endothelial dysfunction serves as a reversible precursor of atherosclerosis and plays a crucial role in the development of complications such as heart attacks and strokes. Assessment of endothelial function using flow-mediated dilation (FMD) in the brachial artery utilizing noninvasive ultrasound imaging provides promising opportunities for early detection and intervention strategies in cardiovascular health.

Material and Method: The study employs meticulous data collection procedures through ultrasonographic scanning of the brachial artery to ensure precise measurements of parameters such as mean velocity, diameter, flow, shear stress, and percentage flow-mediated dilation.

Result: Significantly, the study results revealed variations in blood sugar levels and % FMD between the control and case groups, highlighting potential differences in endothelial function and vascular health. Additionally, the study conducted a comparative analysis of data from different studies, providing valuable insights into the impact of shear stress on endothelial homeostasis and cardiovascular health.

Conclusion: The comprehensive assessment of endothelial function and its correlation with cardiovascular disease risk, through advanced methodologies and meticulous data analysis, empowers researchers and healthcare practitioners with crucial insights into refining risk assessments and personalized treatment strategies in cardiovascular health.

Keywords: Brachial artery, Endothelial dysfunction, Flow mediated Dilation, CVD, Hemodynamics.

Introduction

Cardiovascular disease (CVD), accounting for 31% of global deaths according to the World Health Organization's (WHO) statistical survey in (2017), underscores the significance of prevention and early detection over subsequent treatment[1]. Atherosclerosis serves as the underlying process of CVD, characterized by arterial wall thickening due to inflammatory responses and the accumulation of substances like cholesterol, culminating in plaque formation[2].

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Endothelial dysfunction stands as a reversible precursor of atherosclerosis and plays a vital role in the development of complications like heart attacks and strokes[3]. The World Health Organization (WHO) has incorporated assessment of endothelial dysfunction into various diseases screening criteria[4], recognizing its importance in identifying individuals at moderate and high risk for early interventions to enhance vascular function.

The endothelium serves a critical role in maintaining vascular homeostasis by facilitating the conversion of mechanical signals to chemical signals within blood vessels[5]. It regulates vascular tone through the synthesis and release of vasoactive substances. When endothelial cells are exposed to blood flow, they produce physiologically active substances, including both vasodilators (e.g., nitric oxide, prostacyclin, adenosine, and hydrogen peroxide) and vasoconstrictors (e.g., endothelin-1, angiotensin II, and thromboxane A2). In addition to regulating vascular tone, endothelial cells inhibit platelet aggregation, white blood cell adhesion, and vascular smooth muscle cell proliferation[6]. Moreover, they produce vascular endothelial growth factor (VEGF), influencing the growth and metabolism of surrounding tissue.

Flow-mediated dilation, a physiological test used to assess endothelial function, presents an opportunity for screening programs aimed at improving vascular health[7-8]. By evaluating endothelial-derived flow-mediated dilation in the brachial artery using non-invasive ultrasound imaging, this study aims to investigate the utility of this assessment as a surrogate marker for cardiovascular disease risk[9]. The accessibility and affordability of this assessment tool make it a promising avenue for early detection and intervention strategies in cardiovascular health.

Methodology

The data collection procedure for ultrasonographic scanning of the brachial artery involved meticulous steps to ensure accurate and comprehensive data acquisition. Prior to the examination, detailed subject preparation was conducted, including obtaining informed consent and noting relevant information such as age, sex, cardiovascular history, medication use, the presence of risk factors and any acute illnesses. The procedure took place in a quiet, temperature-controlled environment ranging from 22-26^0 C to optimize conditions for examination.

The baseline measurements phase began with the subject lying supine on an exam table, with a 3-lead EKG attached in a standard position. After a 5-minute rest period to stabilize cardiovascular parameters, blood pressure was measured using an automated device. A tourniquet cuff was applied proximally on the upper arm, and a cross-sectional scan of the brachial artery was performed using colour flow imaging to identify the artery and locate reference points. Various adjustments were made to the probe angle and position to ensure optimal visualization, including marking the subject's skin and adjusting the Doppler gate for velocity measurement.

During the occlusion phase, the cuff was inflated above systolic blood pressure, and the artery was monitored using 2D-Doppler imaging (**Figure 1)**. The release of the cuff was recorded to capture time-to-peak diameter accurately. Subsequently, measurements were taken during hyperaemia, with careful adjustments made for arterial shifts post-cuff release. The probe position was modified to optimize image quality and intima-media thickness visualization. Diameter measurements were recorded meticulously for a specified duration to capture relevant data accurately.

Figure 1: Measurments in Doppler Scan (double lines of pignoli)

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Overall, the data collection process involved systematic and detailed steps to ensure precise measurement of parameters such as mean velocity, diameter, flow, shear stress, and percentage flow-mediated dilation (% FMD) in the brachial artery. These steps are crucial for assessing vascular health, endothelial function, and cardiovascular status accurately.

By ultrasonographic scanning of brachial artery following data was collected:

- Mean Velocity (cm/sec): The mean arterial velocity of blood in the middle 50% of the lumen during one cardiac cycle estimated from Doppler spectral waveforms, proportional to blood flow and inversely proportional to cross-sectional area.
- Diameter (mm): The intima-intima distance (double lines of pignoli) as measured from a longitudinal view along the vessel axis (**Figure 1**). This is measured at Baseline and during reactive hyperaemia.
- Flow (ml/min): The bulk flow of fluid in the circulation, derived mathematically from mean velocity and diameter (**Equation 2**).
- Shear Stress (dynes/cm^2): The frictional force exerted by circulating blood on the intima surface, proportional to velocity and inversely proportional to diameter, derived from mean velocity and diameter (**Equation 3**).
- % FMD: The change in arterial diameter after occlusion in response to hyperemia, over the baseline diameter (**Equation 1**).

Equation 1. % FMD is defined as:

$$\frac{HyperemiaDiameter - BaselineDiameter}{BaselineDiameter} * 100$$

Equation 2. Mean Flow in ml/min is defined as:

$$Q = Velocity * \pi \left(\frac{Diameter}{2}\right)^2$$

Equation 3. Shear Stress is defined as:

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$$T_w = \frac{4\mu Q}{\pi\left(\frac{diameter}{2}\right)^3}$$

Where T_w is shear stress in dynes/cm^2, Q is mean volumetric flow, and μ, the viscosity of blood, is assumed to be 0.035 poise.

Result

In the form of a master chart, the collected data was entered and compiled in a Microsoft Excel 2010 worksheet. Quantitative data was expressed as mean and standard deviation, while qualitative data was presented as percentage and proportion. To infer the significance of the difference in means between the groups, an unpaired 't' test was utilized. Similarly, the significance of the difference in proportion was assessed using a chi-square test, with a p-value less than 0.05 considered as statistically significant.

The study results revealed significant findings in various parameters between the Control and Case groups (Table 1):

Table 1: Parameters between the Control and Case groups

Parameters (present study)	Control (mean ± SD) n=70	Case(mean ± SD) n=70	P value
Blood sugar mg/dl	89.56 + 5.19	114.17 + 10.74	< .001
Mean Baseline Artery Diameter (mm)	3.27+ 0.54	3.44+0.67	> .05
Mean Velocity Baseline (cm/s)	49.68 + 9.94	49.46 + 16.10	> .05
FlowBaseline(cm3/s)	5.45 + 1.46	4.45 + 1.55	< .001
Shear stress at wall (dynes/cm2) Baseline	38.11 ± 10.20	43.14 ± 20.40	> .05
Mean Hyperemi**a** Artery Diameter (mm)	3.42± 0.71	3.59±0.69	> .05
Mean Velocity Hyperemia (cm/s)	56.88 + 12.46	48.94 + 14.78	< .001
Flow Hyperemia(cm3/s)	7.89 + 2.13	4.88 + 1.53	< .001
Shear stress at wall (dynes/cm2) Hyperemia	38.69 ± 10.60	40.34 ± 17.98	> .05
% FMD	12.3 + 1.69	5.50 + 2.30	< .001

The Case group exhibited substantially higher mean Blood Sugar levels compared to the Control group, with a significant difference indicated by a p-value of less than .001. No significant difference was observed in the Baseline Artery Diameter between the Control and Case groups, with a p-value greater than 0.05. The Mean Velocity was comparable between the two groups, with a p-value exceeding 0.05, signifying no statistically significant variation. A stark difference was noted in the % FMD values between the Control and Case groups, with the Case group displaying significantly lower % FMD compared to the Control group, denoted by a p-value of less than .001. The shear stress at the wall during hyperaemia phase did not exhibit a statistically significant difference between the Control and Case groups, with a p-value exceeding 0.05.

Overall, these results highlight variations in Blood Sugar levels and % FMD between the groups, suggesting potential differences in endothelial function and vascular health, while other parameters like Artery Diameter and Velocity did not show significant disparities.

Discussion

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Table 2: Comparison of different Study Data

Characteristics	Tatsua Maruhashi et al.pdf	Result Present Study	Verma I et al.pdf[11]	Alley H et al (2014)	Bellien et al. (2010)[10]
Subjects (cases)	5735 (risk cases for CVD)	70 (CVD)	35 (RA)	25 (PAD)	24 (diabetic)
Subjects (control)	1542	70	25	25	24
Age (case)	53.3+10.3	57.83 + 6.95	46.0 ± 9.1	68 ± 6	34±3
Age (control)	44.5+9.8	39.00 + 12.28	42.8 ± 7.6	68 ± 11	37±3
BMI (case)	24.0 3.3	23.69 + 2.45	24.1 ± 3.9	29 ± 7	24.5±0.8
BMI (control)	21.8 2.8	24.36 + 2.40	23.2 ± 2.6	30 ± 4	23.7±0.8
Sys BP (case)	130	130	125	139	118
Sys BP (control)	117.1	114.2	118.8	134	118

Table 3: Cut-off Values of Flow-Mediated Dilation (FMD) in Case and Control Groups among different studies

	Number of Cases	Number of Control	FMD% of cases	FMD% of control
Bellien et al (2010)[10]	24	24	9.2±0.8*	10.5±0.6*
Alley H et al (2014)	25	25	6.8 ± 3.5	9.1 ± 3.6
Tatsua Maruhashi et al (2020)	5735	1542	5.9 ± 2.9	7.5 ± 3.3
Holder et al (2021)[12]	1920	821	5.45±2.72	6.66±3.24
Present study (2024)	70	70	5.50 + 2.30	12.3 + 1.69

*haemodynamic measured on radial artery.

Discussion on Endothelial Function and Cardiovascular Disease Risk

Neunteufl T et al[13], in 1997, conducted a study to test the hypothesis that an impairment of FMD in the brachial artery is related to the presence and/or extent and severity of coronary artery disease (CAD)[14]. The investigation into endothelial function and its association with cardiovascular disease risk presents compelling insights drawn from a comparative analysis of multiple studies. Notably, the study delves into the impact of shear stress on endothelial homeostasis and the subsequent implications for cardiovascular health. The meticulous assessment of endothelial function using Shear Stress has unraveled significant disparities in vascular function between distinct patient cohorts, shedding light on crucial cardiovascular risk factors.

Study Comparisons and Findings

Tatsua Maruhashi et al. and Alley H et al. exhibited varying gender-specific insights, emphasizing optimal FMD cutoff values differently for both genders. Verma S et al.[15] and Tatsua Maruhashi et al.[6] provided valuable analyses on optimal FMD cutoff values across different age brackets and age decades. The distribution of subjects, especially the predominance of over-50-year-olds, showcases demographic nuances influencing cardiovascular health.

Present study was conducted on brachial artery like others[16] while many studies were conducted on radial artery[17]. Brachial artery FMD was found more approaching in a roll of a screening tool. Lars Lind, in 2014, conducted a study onflow-mediated vasodilation over five years in the general elderly population and evaluate its relation to cardiovascular risk factors[18].

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Significance of Findings

The observed significant differences in Blood Sugar Levels and % FMD values between the study groups emphasize key variations in endothelial functionality and cardiovascular health profiles. The non-significant variances in Artery Diameter and Velocity provide additional context to the complex interplay of factors impacting vascular health in distinct patient cohorts. These discrepancies underscore the need for tailored interventions and further explorations in endothelial function assessment.

Conclusion

This discussion encapsulates the pivotal role of studying endothelial function and its correlation with cardiovascular disease risk through advanced methodologies and meticulous data analysis. The study arms researchers and healthcare practitioners with vital insights into refining risk assessments and personalized treatment strategies in the realm of cardiovascular health.

References

1. World Health Organization. (n.d.). Cardiovascular diseases (CVDs). Retrieved from WHO Fact Cardiovascular Diseases 2014.
2. Cahill PA, Redmond EM. Vascular endothelium - Gatekeeper of vessel health. Atherosclerosis. 2016 May;248:97-109. doi:10.1016/j.atherosclerosis.2016.03.007. Epub 2016 Mar 9. PMID: 26994427; PMCID: PMC6478391.
3. Chhabra N (2009). Endothelial dysfunction – A predictor of atherosclerosis. Internet J Med Update 4, 33–41.
4. Wilson JMG &Jungner G (1968). Principles and practice of screening for disease. Geneva, Switzerland: World Health Organization.
5. Alley H et al. Ultrasound Assessment of Endothelial-Dependent Flow-Mediated Vasodilation of the Brachial Artery in Clinical Research. J. Vis. Exp. (92), e52070,doi:10.3791/52070 (2014).
6. Maruhashi, Tatsuya et al. “Diagnostic Criteria of Flow-Mediated Vasodilation for Normal Endothelial Function and Nitroglycerin-Induced Vasodilation for Normal Vascular Smooth Muscle Function of the Brachial Artery.” Journal of the American Heart Association vol. 9,2 (2020): e013915. doi:10.1161/JAHA.119.013915
7. Thijssen, D.H.J., Bruno, R.M., Van Mil, A., Holder, S.M., Faita, F., Greyling, A., Zock, P., Taddei, S., Deanfield, J.E., Luscher, T., Green, D.J., & Ghiadoni, L. (2019). Expert consensus and evidence-based guidelines for the assessment of flow-mediated dilation (FMD) in humans. European Heart Journal, 40(30), 2534-2547. doi: 10.1093/eurheartj/ehz350.
8. Atkinson, G., Batterham, A. M., Thijssen, D. H. & Green, D. J. (2013). A new approach to improve the specificity of flow-mediated dilation for indicating endothelial function in cardiovascular research. Journal of Hypertension, 31, 287-91.
9. Celermajer DS, et al. Non-invasive detection of endothelial dysfunction in children and adults at risk of atherosclerosis. The Lancet. 1992;340:1111–1115. [PubMed]
10. Bellien et al. Earlystage detection of conduit artery endothelial dysfunction in patients with type 1 diabetes. Diabetes & Vascular Disease Research. 2010; 7(2) 158–166.
11. Verma I et al. Predictors of endothelial dysfunction and atherosclerosis in rheumatoid arthritis in Indian population. Indian Heart Journal. Volume 69, Issue 2, 2017, Pages 200-206, ISSN 0019-4832, https://doi.org/10.1016/j.ihj.2016.10.013.
12. Holder, S.M., Dawson, E.A., Brislane, Á., Hisdal, J., Green, D.J., & Thijssen, D.H.J. (2019). Fluctuation in shear rate, with unaltered mean shear rate, improves brachial artery flow-mediated dilation in healthy, young men. Journal of Applied Physiology, 126(6), 1687-1693. doi: 10.1152/japplphysiol.00009.2019.
13. Neunteufl T, et al, Systemic endothelial dysfunction is related to the extent and severity of coronary artery disease. Atherosclerosis. 1997 Feb 28;129(1):111-8.

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14. Healy B, Endothelial cell dysfunction: an emerging endocrinopathy linked to coronary disease. J Am Coll Cardiol. 1990 Aug;16(2):357-8.
15. Verma S et al. Cross-sectional evaluation of brachial artery flow-mediated vasodilation and C-reactive protein in healthy individuals. Eur Heart J. 2004 Oct;25(19):1754-60. doi: 10.1016/j.ehj.2004.06.039. PMID: 15451155. [PubMed]
16. Thijssen D. H. et al. Assessment of flow-mediated dilation in humans: a methodological and physiological guideline. Am J Physiol Heart Circ Physiol. 2011 Jan; 300(1): H2–H12.
17. Doshi SN, et al. Clinical science. Vol. 101. London, England: 2001. Flow-mediated dilatation following wrist and upper arm occlusion inhumans: the contribution of nitric oxide; pp. 629–635.[PubMed]
18. Lars Lind. Flow-mediated vasodilation over five years in the general elderly population and its relation to cardiovascular risk factors. Atherosclerosis. Volume 237, Issue 2, December 2014, Pages 666-670