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Levels of Apolipoproteina-1, Apolipoprotein B, and Glucose of the Elderly, Exercising, and Non-Exercising Individuals in Nnewi, Nigeria

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ABSTRACT

Background: Cardiovascular disease causes death and disability. **Objectives:** To evaluate the serum levels of Apo A-1, Apo B, and glucose in the elderly, actively exercising and non-actively exercising individuals in Nnewi. **Materials and methods:** Ninety subjects recruited for the study were grouped into; Group A- Non-exercising, Group B Exercising, and Group C- Elderly Individuals aged 55 years and above. Blood sample was collected from the participants after 10-12 hours overnight fasting. Apolipoprotein A-1, Apolipoprotein B, and Glucose levels were determined spectrophotometrically. Their systemic (SBP), and diastolic blood pressures (DBP) Heights, and weights were measured. The questionnaire obtained their socio-demographic information. Data were analyzed using ANOVA, Bonferroni Post Hoc Test, and Pearson r correlation. **Result:** A significantly higher difference existed in the mean serum value of glucose in the Elderly (6.60 ± 1.50) and exercising (5.89 ± 0.82) compared with the Non-exercising (5.40 ± 0.48)($p<0.05$). A significantly higher difference was seen in the mean SBP, DBP, and BMI levels of the Elderly compared with the non-exercising ($p \leq 0.05$). However, the exercising had significantly lower mean SBP, DBP, and BMI values than the Non-exercising group ($p \leq 0.05$). A strong positive correlation was observed between Apo B vs Apo A-1 in the exercising, and Non-exercising groups, while a weak negative correlation existed between BMI and Apo A-1 in the actively exercising. **Conclusion**: The significant differences observed in the parameters measured were not clinically significant.

Keywords: Apolipoprotein A-1, Apolipoprotein B, Glucose, cardiovascular diseases

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INTRODUCTION

In this 21st century, we have observed a shift in the disease pattern. Non-communicable diseases (NCDs) are now a massive challenge for medical science. Mortality and Morbidity rates are very high in cases of Cardiovascular diseases (CVDs). CVDs affect almost the entire world population and can be defined also as a worldwide epidemic. [1] Most of the CVDs are having chronic pathological processes, we know many risk factors causing and indicating future events of CVDs. [2]

A Sedentary lifestyle is one of the five major risk factors (along with High blood pressure, abnormal values of blood lipids, smoking, and obesity) for cardiovascular disease as outlined by American Health Association (AHA). The burden of cardiovascular diseases in developing countries such as Nigeria is worsened by the increasing prevalence of cardiovascular risk factors. [3] It has been estimated that 9.5% of 57 million deaths in the world a decade ago could be attributed to physical inactivity which equals more than 5 million deaths worldwide. [4] The population of the elderly is on the rise as life expectancy increases with implications for dependency and the need for formal and informal care. One of the challenges of increasing expectations of life in a population of people is the risk of developing component-defining criteria for cardiovascular (CV) diseases which is related to the process of aging. [5]In Nigeria, CVD accounted for 11% of all deaths in 2018. [6] The need for a newer approach was necessary as many reported cases who had already achieved the recommended level of conventional lipid profile have developed cardiac events mainly in high-risk patients. These newer markers answer many of the questions by providing a wide range of specificity over traditional markers. Some of the numerous risk scores which have been developed to predict cardiovascular risk include Apolipoprotein, Creatine kinase (Ck-MB), Cardiac Troponin, etc. [7] Apolipoproteins are structural and functional proteins of lipoprotein particles that perform important functions for lipoprotein metabolism as carriers of these hydrophobic molecules in the plasma aqueous medium, binding to the specific receptors on the cell surface to conduct the lipids correctly to the target organs and tissues of the organism. [8] Apolipoproteins are also known to determine the structural stabilities and metabolic directions of lipoproteins. Of the Apolipoproteins, Apolipoprotein B (Apo B) has been identified in VLDL-C (very high-density lipoprotein cholesterol), LDL-C (Low-density lipoprotein), IDL (intermediate density lipoprotein). Hyperglycemia in diabetic mellitus causes the formation of Advanced Glycation end products (AGEs), glycated proteins are one of the AGEs. The AGEs cause the formation of pro-inflammatory products by stimulating various receptors on various cells related to the formation of atherosclerosis. Apo B undergoes this glycosylation process and plays a vital role in the formation of plaque. This process is not limited to LDL-C alone; It affects all the lipoproteins having Apo B as Apolipoprotein. [9] So it is logical to evaluate Apo B in the case of diabetic mellitus. While Apo A-1 has been identified in HDL-C. [10] Apolipoprotein A-1 is an indispensable component and major structural protein of high-density lipoprotein cholesterol (HDL-C), which plays a vital role in reverse cholesterol transport and cellular cholesterol homeostasis. Since its identification, its multifunctional role in immunity, Inflammation, apoptosis, viral, and bacterial infection, etc, has crossed the boundary of its potential of protecting the cardiovascular system and lowering cardiovascular disease risk, attributing HDL-C to be known as a protective fat removal particle. So, it gives a fair share to check the balance between the level of "Good cholesterol" and "Bad cholesterol". Notably, CVDs are precipitated by some risk factors classified as modifiable and non-modifiable risk factors such as unhealthy diet, physical inactivity, harmful use of alcohol, and smoking which lead to the effects such as increased blood pressure, increased blood sugar levels, dyslipidemia, overweight and obesity. [6] Aging and gender strongly modulate the risk to develop cardiovascular diseases but very few studies have investigated the impact of gender on cardiovascular diseases in the elderly, which represents a growing population. Recent studies showed that patients with high plasma Apo-B levels were more predisposed to coronary diseases than patients with low plasma Apo-B levels because Apo B is essential for the binding of LDL particles to the LDL-C receptor,

allowing cells to internalize LDL and thus absorb cholesterol, Apo B is also involved in plaque vulnerability. Apo A-1 also acts as a cofactor for lecithin cholesterol acyltransferase (LCAT), [11] which is important in removing excess cholesterol from tissue and incorporating it into HDL for reverse transport to the liver thereby reducing the risk of developing coronary heart disease. Thus, evaluation of the levels of Apo A-1 and Apo B of the elderly, exercising, and non-exercising individuals may provide information on the possible risks of atherosclerosis associated with cardiovascular disease. Thus, this study evaluation of apolipoprotein A-1, apolipoprotein B, and glucose levels of the elderly, actively exercising and non-exercising individuals in Nnewi may also highlight the usefulness of these parameters (Apo A-1 and Apo B) as a detection or onset of CVD in the management of individuals with cardiovascular disease.

MATERIALS AND METHODS

The reagents and kits for the biochemical analysis were commercially obtained and the manufacturer's standard operating procedures were strictly observed. This cross sectional study was conducted in Nnewi North, Anambra state, south East of Nigeria.

Study participants

The sample size was calculated using G* Power software version 3. 0. 10 (Universitat Dusseldorf, Germany). Power to determine a sufficient sample size using an alpha of 0.05, a power of 0.80, and a medium effect size of 0.4, based on these, the calculated total sample size of 66 has 80% power to detect a difference of 0.4 at a significance level of 0.05. To take care of possible attrition, a total sample size of 90 was used for this study. Ninety (90) Participants in Nnewi (Okofia Otolo) were recruited by random sampling for this study aged between18-70years comprising of 30 Elderly (Group C(65-70years), 30 Actively Exercising (GroupB(18-30years) and 30 Non-exercising subjects (Group A(18-30years). Fasting blood samples were collected, the immunoturbidometric technique measured Apolipoprotein A-1, and Apolipoprotein B, while the glucose was analyzed immediately using an accu-answer glucometer. The questionnaire obtained socio-demographic information such as age, sex, and demographic factors, while participants' heights were obtained in meters by height scale , and weights measured by a digital body weighing scale(canny, USA), recorded, and used to calculate their body mass index. Their blood pressure readings were obtained using sphygmomanometer (Walgreens digital blood pressure, China). The study was approved by the Ethics Committee of the Faculty of Health Sciences and Technology, Nnamdi Azikiwe University, Nnewi, Anambra State, Nigeria FHST/NAU/2021/. Healthy informed, and consented participants between 18-65 years were recruited whereas; individuals that were diabetics, hypertensive, alcoholic, smokers, pregnant women, and lactating mothers were excluded.

Collection of Samples

About five milliliters (5ml) of venous fasting blood sample was collected aseptically after 10-12hours by venipuncture method from each subject. Two microlitre (2µl) of the fasting blood sample was used for glucose estimation. The remaining blood sample was dispensed into the plain sample bottle, allowed to clot, retract, and centrifuged at 4000rpm for 5minutes and the serum was then extracted into well labeled plain tube, and stored in aliquots of three at 4˚C until when required for the determination of Apolipoprotein A-1 and Apolipoprotein B.

Laboratory methods

The determination of Apo A-1, and B, were done by sandwich enzyme immunoassay

technique as described by Brodsky, and Edward. [12]

Statistical analysis

The IBM Statistical Package for the Social Sciences (SPSS) version 23.0 was used for the analysis of the results. Data was presented as mean ± standard deviation (SD) and analyzed statistically using ANOVA Bonferroni Post Hoc, the correlation of the parameters was determined using Pearson's correlation coefficient. The level of significance was set at $p<0.05$.

RESULTS

As shown in Table 1, the mean serum concentration of Apolipoprotein A-1, B, and the mean plasma concentration of glucose of the non- actively exercising (A), actively exercising (B) and elderly individuals (C). There was no significant difference in the mean value of serum Apo A-1 in elderly (224.10 ± 32.39) and actively exercising (221.70 ± 27.21) compared with the Non-exercising individuals (234.25 ± 28.44)(p>0.05). And no significant difference existed in the mean value of Apo A-1 in actively exercising Individuals (221.70 ± 27.21) compared with the elderly (224.10 ± 32.39)(p> 0.05). Also, no significant difference was seen in the mean serum value of Apo B in elderly (113.73 ± 12.00) and in the actively exercising (112.85 ± 15.19) compared with the Non-exercising individuals (113.55 ± 14.48) (p>0.05). More so, no significant difference existed in the mean serum value of Apo A-1 in actively exercising Individuals (112.85 ± 15.19) compared with the elderly (113.73 ± 12.00) (p> 0.05)**.** However, a significant higher difference was observed in the mean blood value of glucose in the Elderly (118.83 ± 27.69) and actively exercising (5.89±0.82) compared with the Non-exercising individuals (5.40 ± 0.48) (p<0.05).The mean blood glucose level in the Non-exercising individuals (5.40±0.48) was significantly lower compared with the elderly (6.60±1.54) (p<0.05).

Table 2: The systolic blood pressure (SBP), diastolic blood pressure (DBP) and body mass index (BMI) of the non-exercising (a), exercising (b), elderly individuals(c).(Mean±SD)

A significant higher mean systolic, diastolic and BMI levels existed in the elderly participants (C) (140.63 ± 26.24, 84.77 ±12.17, 27.19 ± 5.90) compared with the Non-exercising (A)(111.66 ± 9.86, 76.33 ± 8.90, 22.19 ± 2.63) and exercising (B)(109.67 ± 9.64, 77.33 ± 9.44, 23.67 ± 5.91) respectively(p<0.05).

Table 3: Correlation of the level of Glucose, BMI with Apo A-1 and Apo B, ratio of Apo B/Apo A-1 of the Non-exercising, Exercising and Elderly individuals.

For non-exercising individuals, no correlation was observed between Glucose and Apo A-1 (r= -0.07) (p=0.72),and between Glucose and Apo B (r=0.24)(p=0.21). No association was seen between BMI and Apo A-1(r=0.28)(p=0.14), and BMI and Apo B(r=0.20)(p=0.30). There is a strong positive correlation between Apo B and Apo A-1(r=0.59)(p=0.01).

Table 1: The mean serum concentration of Apolipoprotein A-1, B, and the mean plasma concentration of glucose of the non exercising (a), actively exercising (b) and elderly individuals (c). (mean±SD)

Groups	Apo A-1(mg/dl)	Apo B(mg/dl)	Glucose(mmol/l)
Non-exercising (A) (n=30)	234.25 ± 28.44	113.55 ± 14.48	5.40±0.48
Exercising (B) (n=30)	221.70 ± 27.21	112.85 ± 15.19	5.89 ± 0.82
Elderly (C) (n=30)	224.10 ± 32.39	113.73 ± 12.00	6.60± 1.54
F-value	1.54	0.22	4.61
P-value	0.22	0.97	0.01*
A vs B (p-value)	0.31	1.00	0.21
Avs C (p-value)	0.56	1.00	0.01*
B vs C (p-value)	1.00	1.00	0.72

** Statistically significant at p< 0.05*

Table 2: The systolic blood pressure (SBP), diastolic blood pressure (DBP) and body mass index (BMI) of the non-exercising (a), exercising (b), elderly individuals(c).(Mean±SD)

Groups	Systolic (mmHg)	Diastolic (mmHg)	BMI(kg/m2)
Non-exercising (A) (n=30)	111.66 ± 9.86	76.33 ± 8.90	22.19 ± 2.63
Exercising (B) (n=30)	109.67 ± 9.64	77.33 ± 9.44	23.67 ± 5.91
Elderly (C) (n=30)	140.63 ± 26.24	84.77 ±12.17	27.19 ± 5.90
F-value	30.76	5.94	11.98
P-value	0.00*	0.00*	0.00*
A vs B (p-value)	1.00	1.00	0.47
A vs C (p-value)	0.00*	0.01*	0.00*
B vs C (p-value)	0.00*	0.02*	0.00*

** Statistically significant at p< 0.05*

For actively exercising individuals, no associations existed between Glucose and Apo A-1 (r= -0.23) (p=0.23), BMI and Apo B (r= -0.13) (p=0.48), and between between Glucose and Apo B (r=0.23) (p=0.23). There is a weak positive correlation between Apo B and Apo A-1(r=0.40) (p=0.03), and a weak negative correlation exists between BMI and Apo A-1(r= -0.39) (p=0.03).

For the Elderly, no association was observed between Glucose and Apo A-1 (r= -0.04) (p=0.84), between Glucose and Apo B (r=0.07) (p=0.73), BMI and Apo A-1(r= 0.26) (p=0.17), BMI and Apo B (r= 0.09) (p=0.65) and Apo B and Apo A-1(r=0.14) (p=0.46).

Table 3: Correlation of the level of Glucose, BMI with Apo A-1 and Apo B, ratio of Apo B/Apo A-1 of the Non-exercising, Exercising and Elderly individuals.

	Parameters	r-value	P-value
	Glucose vs Apo A-1	-0.07	0.72
Non-Exercising	Glucose vs Apo B	0.24	0.21
	BMI vs Apo A-1	0.28	0.14
	BMI vs Apo B	0.20	0.30
	Apo B vs Apo A-1	0.59	0.01*
Exercising	Glucose vs Apo A-1	-0.23	0.23
	Glucose vs Apo B	0.23	0.23
	BMI vs Apo A-1	-0.39	0.03*
	BMI vs Apo B	-0.13	0.48
	Apo B vs Apo A-1	0.40	0.03*
Elderly	Glucose vs Apo A-1	-0.04	0.84
	Glucose vs Apo B	0.07	0.73
	BMI vs Apo A-1	0.26	0.17
	BMI vs Apo B	0.09	0.65
	Apo B vs Apo A-1	0.14	0.46

** Statistically significant at p< 0.05*

DISCUSSION

Regular exercise and physical activity considerably reduce the risk of cardiovascular diseases (CVDs) and are associated with impressive, widespread health benefits.Chronic obstructive pulmonary disease (COPD) and CVD have been associated with a sedentary lifestyle, characterized by consistently low levels of physical activity, and is widely acknowledged as one of the major risk factors that predisposes to the development, and progression of CVDs. [13]

The structural and functional elements of the lipoprotein particles that act as cholesterol transporters are known as apo A-1 and apo B. The levels of Glucose, Apo A-1, and Apo B in elderly, exercising, and non-exercising individuals were evaluated. In this study, there was no discernible difference between the elderly and those who were actively exercising and those who were not (p>0.05).

This research differs from that of, [13] who investigated how aerobic exercise affected the levels of apolipoproteins A-1 and B in the serum of patient with chronic obstructive pulmonary disease(COPD). It was a prospective trial, and the aerobic activity was carried out three times a week for two months. The findings demonstrated that after the exercise, the mean levels of apo A-1 and apo B were considerably greater than they were before the exercise. The variations in the study's design and exercise duration account for the discrepancy in their findings. The current cross sectional study, revealed that the Exercising participants had the lowest mean Apolipoprotein B values whereas the Elderly participants had the highest mean Apolipoprotein B values. It has been shown that apolipoprotein B lipoprotein particles play a significant role in determining cardiovascular risk and the risk of cardiovascular disease rises with age. [14] Lipoproteins carrying apolipoprotein B are able to transport cholesterol into the arterial wall and, if they are present in higher concentrations, may be the primary beginning component in atherosclerosis, which may predispose a person to cardiovascular disease. [14,15]

The elderly participants' mean plasma glucose levels were significantly greater than those of the non-exercising participants (p <0.05). The study also revealed that the non-exercising participants had the lowest mean plasma glucose levels, while the elderly participants had the highest mean plasma glucose levels. Given that it is well known that plasma glucose levels increase with aging, it is possible that the individuals' varying ages (young and old) had an effect on this. [16]

After exercise, glucose absorption is still high and the route mediated by contraction is still active for several hours. Increased muscle glucose absorption is matched by an equal increase in

hepatic glucose production during a brief, moderate-intensity exercise (VO2 max 60%) in people without diabetes, and blood glucose levels stay the same. [16] As insulin levels drop, the liver becomes more receptive to glucagon release, which boosts glucose synthesis. Additionally, following, [17] elderly people have higher glucose levels than younger people because they are more likely to be undernourished, have several chronic diseases, or take multiple drugs. This was in contrast to a similar study that showed exercise causes a considerable drop in
mean fasting blood glucose compared to the levels seen in sedentary individuals, [18] Their differing conclusions could be attributed to epigenetic modifications, various study
Designs, and various study subjects. The elderly participants in this study had significantly higher mean systolic, diastolic, and

BMI values than the non-exercising and exercising groups, respectively ($p < 0.05$), whereas the exercising group had the lowest systolic value. This is because as we become older, the vascular system—the body's network of blood vessels—changes. [19] Blood pressure rises as a result of stiffer arteries. Regular aerobic activity lowers systolic and diastolic blood pressure by 11 and 8 mmHg, respectively. This result is consistent with a study by, [20] whose participants engaged in aerobic exercise. After three months of three days a week of aerobic activity, the intervention group's mean systolic and diastolic blood pressure decreased by 3.2 and 1.2 mmHg, respectively, while the control group's mean blood pressure showed no significant change. [20] In a different study, [21] arterial compliance demonstrated resistance to a brief aerobic exercise program, and no decrease in the patient's blood pressure was discovered. [21] Exercise has been linked to immediate, considerable drops in systolic blood pressure, according to another study. [22] This initial drop in blood pressure following exercise, known as post-exercise hypotension, can last for almost 24 hours and has the strongest impact in people with greater baseline blood pressure. The observation that older people had higher diastolic and BMI values demonstrate that sedentary lifestyles favor an increase in BMI, which is exacerbated by aging as adiposity tends to increase with age. [23] This study supports, [24] which claimed that diastolic blood pressure tends to rise until around age 50 and that the rise is caused by an increase in artery resistance. Exercise-induced changes in diastolic blood pressure were closely correlated with changes in serum total cholesterol (as well as LDL cholesterol), with diastolic blood pressure rising in persons with the highest cholesterol levels and falling in those with the lowest levels. [24] Studies have shown that similar to BMI, increasing physical activity tends to reduce the incidence of cognitive problems and enhances general health in humans. [25]

High levels of physical activity have been demonstrated to improve life's physical, social, emotional, and health-related aspects. [26] Age and weight are two factors that are thought to affect physical activity. Research shows that this activity tends to decrease with age and stabilize in middle age.
The levels of glucose with Apo A-1 in the elderly, exercising and non-exercising, and the levels of glucose with Apo B in the elderly, exercising and non-exercising, did not significantly correlate negatively or positively. This might be because while Apo B is a single molecule that is found in low-, intermediate-, and very low-density lipoproteins, ApoA-1 is the major lipoprotein linked to high-density lipoprotein cholesterol (HDL-C). [27] ApoA-1 and ApoB regulated fasting blood glucose (FBG) levels by raising insulin levels, according to in vitro and animal studies. [29,5] According to research, the relationship between fasting blood glucose levels and apo A-1, apo B, and the apo B/a-1 ratio is inverse or null for apo A-1 [30,31], but positive for apo B and apo B/a-1 ratio. [32] The populations with greater BMI, larger waist-hip ratios, higher mean values of SBP and DBP, as well as higher levels of serum triglycerides and cholesterol, were also more likely to experience a cardiovascular incident. [33]
The cornerstone of cholesterol treatment continues to be LDL-C, which is closely connected to CV disease and death. [33] Exercise was also found to lower LDL-C levels in obese men and women with type 2 diabetes. [34] Therefore, it would make sense to assume that exercise training reduces the atherogenicity of the lipoproteins, as shown in the current study by the reduction in the apoB and apoB/apoA-I ratio.

The fact that this study was only a month long and was not a large-scale randomised controlled trial is one of its limitations. Large-scale, randomised controlled trials that will last six months or more may produce better results that are clinically significant.

CONCLUSION

Although it was not statistically significant, this study demonstrated that older participants and non-exercising participants had greater values of apo A-1 and apo B than exercising people. Additionally, among the participants, the elderly participants' mean blood glucose levels were the highest. The statistically significant variations in the variables that were measured were not clinically meaningful. To further validate and bolster the findings of this investigation, other studies with larger sample sizes and longer study duration should be conducted.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

Contributors

ACI, CIO, and SCM conceived and designed the research proposal. AFE, PCO, and ACI performed sample collection, experiments and data analysis. CIO, ACI, EIN, and SCM contributed to the final version of the manuscript. All authors have read and approved the final manuscript.

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Data availability

The data used to support the findings of this study are available from the corresponding author upon reasonable request.

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Conflict of interest:

None declared.

Ethical approval:

The study sought and obtained ethical approval on 18th January, 2023 from the Faculty of Health Sciences and Technology Ethics Committee of Nnamdi Azikiwe University with reference no.NAU/FHST/2021/MLS25

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