WWW.IJRPR.COM

International Journal of Research Publication and Reviews

Journal homepage: www.ijrpr.com ISSN 2582-7421

MALAT1 Gene Rs3200401 polymorphism in Patients with arterial Hypertension

Omar Mohamed Saleh[1] *, Yassmin Hassan Mohamed Elsayed [2], Loaa Abd-Allah Tag Eldeen[2], Moushira Abd El Wahab Mahmoud [2]

[1] Cardiology Department, Suez Canal University, Faculty of Medicine, Egypt
[2] Medical Biochemistry and Molecular Biology Department, Suez Canal University, Faculty of Medicine, Egypt
* E-Mail: omar_hassan@med.suez.edu.eg
ORCID ID: 0000-0001-7467-9210
DOI: https://doi.org/10.55248/gengpi.4.723.48907

ABSTRACT

Background: Systemic arterial hypertension (HTN) is a common and important preventable risk factor for morbidity and mortality of cardiovascular disease (CVD) worldwide. Metastasis associated lung adenocarcinoma transcript 1 (MALAT1), a long non-coding RNA, plays a role in endothelial cell function and facilitates the inflammatory cascade providing the key events in vascular remodeling. Previous studies have shown that single nucleotide polymorphisms (SNPs) in MALAT1 contribute to the risk of CVD attracting attention to its possible risk role.

Aim: To investigate the association between MALAT1 rs3200401 polymorphism with hypertension susceptibility, as well as its relation to lipid profile and the risk of left ventricular hypertrophy (LVH) in Egyptians.

Patients and Methods: In this pilot study a total of 55 patients with Hypertension and 50 -age and sex matched- healthy control subjects were recruited from Suez Canal University teaching hospitals. Genotyping assay was done by real time PCR using two TaqMan® MGB probes. Lipid profile and echocardiography were also done for all study population.

Results: The CC genotype was significantly more frequent in hypertensive patients' group, while CT+ TT genotypes were more frequent in control group after adjusting for age and gender (OR = 2.56, 95% CI = 1.06-6.18; P-value = 0.034). Allele C frequency is significantly higher in patients' group (71%) versus control group (57%). (OR= 0.54, CI= 0.31 – 0.97, P-value= 0.03). MALAT1 SNP (rs3200401) is associated with dyslipidemia, as HDL levels in total population were lower in TT genotype than other genotypes. There's no association between MALAT1 SNP (rs3200401) with LVH.

Multivariate regression analysis demonstrated that the rs3200401 polymorphism of MALAT1 was independent risk factor for HTN in Egyptian patients (OR = 0.151, 95% CI = 0.036 - 0.265, P = 0.01).

Conclusion: These results provide that MALAT1 SNP (rs3200401) is a risk factor for HTN and presence of T allele may be protective against HTN development.

Key words: Hypertension, Polymorphism, MALAT1, rs3200401

INTRODUCTION

The most prevalent global preventable risk factor for disability and early mortality is systemic arterial hypertension (HTN) [1]. A national survey conducted in Egypt estimated the prevalence of hypertension is 29.5% [2]. Hypertension is a complex pathology with genetic basis and environmental impacts can transmit across generations [3].

Long non-coding RNAs (lncRNAs) are more than 200 nucleotides of transcribed genetic material. These lncRNAs can alter gene expression via various mechanisms. In mammals' lncRNAs, Metastasis Associated Lung Adenocarcinoma Transcript 1 (MALAT1) is substantially conserved. Its length is longer than 8000 nucleotides, and it is located at chromosome 11(11q13.1) long arm [4].

MALAT1 plays important roles in cardiovascular diseases. It affects endothelial cells function and angiogenesis by direct control of vascular endothelial growth factor receptor 2 [5]. It can promote the inflammatory cascade by increasing the expression of tumour necrosis factor-α (TNF-α) as well as interleukin-6 (IL-6), which can then sustain an endothelial inflammatory response [6]. The key events in the onset and development of hypertension are likely endothelial cell dysfunction and inflammation of the arterial wall [7, 8].

Spontaneously hypertensive rats **(**SHRs) had highly expressed MALAT1 in their thoracic aorta and myocardium and were presented with signs of left ventricular hypertrophy (LVH) [8].

Single nucleotide polymorphisms (SNPs) in MALAT1 have been linked to increased inflammation [9], dyslipidemia and the risk of coronary artery disease (CAD), according to earlier research [4, 10].

On the MALAT1 gene, there are 16 SNPs with a minimal allele frequency (MAF) > 0.01, however only rs3200401 has a MAF > 0.10 over the entire genome [11].

There were many studies investigated (rs3200401) MALAT1 polymorphism in cardiovascular diseases, however, none investigated its relation with hypertension [12, 13].

As there is currently no preventative or curative treatment for hypertension, the major management goal is to improve quality of life of patients, decrease disability as well as uncovering early biomarkers that can either predict the occurrence of HTN, its risk factors, or consequences.

Therefore, the purpose of this research is to look at the association between Egyptian patients' hypertension and the MALAT1 rs3200401 polymorphism.

PATIENTS AND METHODS:

This research was conducted in the outpatient clinics of Suez Canal University Teaching Hospital as an observational cross-sectional study. It included 55 patients previously diagnosed with primary hypertension and 50 non-hypertensives, apparently-healthy subjects which were age and gender-matched. Patients suffering from secondary hypertension, any structural heart disease, malignant diseases or autoimmune diseases were excluded. Measurement of blood pressure, calculation of BMI (dividing the kilogrammes of body weight by the square of the meters of height (kg/m^2)) were done.

Echocardiographic assessment: The American Society of Echocardiography's recommended procedures 2016 were followed for performing the echocardiographic measures, using a commercially available system (Philips Ultrasound field Service Company, EPIQ 7 Q lab version 10.8.5 machine). To determine the left ventricular mass index (LVMI), left ventricular mass (LVM) was computed and indexed to body surface area.

Lipid profile measurement: Measurements were made of triglycerides (TG), total cholesterol (TC) as well as high-density lipoprotein cholesterol (HDL-C) by colorimetric method using a semi-automatic, single beam filter photometer (Photometer 5010 v5+)**.** Using the Friedewald equation, low density lipoprotein cholesterol (LDL-C) was calculated.

Genotyping of MALAT1 rs3200401 polymorphism:

Using the Spin-column technique, genomic DNA was extracted from a 200 µL peripheral whole blood sample (QIAamp® DNA Blood Mini Kit; Cat No. 51104). Then the DNA yield with assessment of the purity using the NanoDrop® (ND)-1000 v3.8 spectrophotometer known as (NanoDrop Technologies, Inc. Wilmington, USA).

TaqMan® SNP Genotyping Assays were used for SNP genotyping in accordance with Life Technologies' manufacturer's instructions. (Thermo Fisher Scientific Inc. USA). *Cat No 4351379*

Ethical approval: Every case-sharing participant in the study provided his or her consent. Any patient who was mulling over taking part in this research was given a thorough explanation of all the steps, presented in a manner they could fully comprehend. The Ethical Committee Council of Suez Canal University approved the study. The study was done in compliance with the Declaration of Helsinki, which is the World Medical Association's code of ethics for studies for humans.

Statistical analysis: Gathered information were processed with SPSS version 21 (SPSS Inc., Chicago, IL, USA).

RESULTS:

A total of 55 patients with HTN (28 male and 27 female) and 50 matched for age and gender, supposedly healthy control individuals (28 females and 22 males) participated in this study. The patients mean age of was 56.5±8.6 years; while that of the control subjects was 53.4±7.3 (p >0.05).

No significant difference was found between the patients and controls as regard smoking (25% versus 24%), BMI (26.7 ± 1.7 versus 27.3 ±1.4 kg/m^2) or pulse (76 ± 11 versus 77 ± 9 beat/minute)

As expected, significantly increased systolic and diastolic blood pressure in hypertensive group (144 ± 23/90 ± 9 versus 113 ± 10/76 ± 5 mmHg) (P=0.000). Hypertension duration in the patients' group was 8±6 years.

The lipid profile was investigated in the study population. It was found that values of HDL-C are significantly lower in hypertensive than in control group (34±7.5 versus 44±11 mg/dl, P = 0.000), while LDL-C levels are significantly higher in patients (114±37 versus 94±21 mg/dl), P = 0.001). However, the two groups did not differ significantly from one another. as regard TC or TG levels (P-value = 0.351 and 0.2, respectively). Hypertensive patients shows significantly higher LVMI than the controls with values (130±54 versus 85±18 gm/m^2, P=0.000).

MALAT1 rs3200401 polymorphism and risk of hypertension:

Figure (1) shows genotypes distribution in study population; there were no variations from the Hardy-Weinberg equilibrium in the cases or the controls (P-value > 0.05).

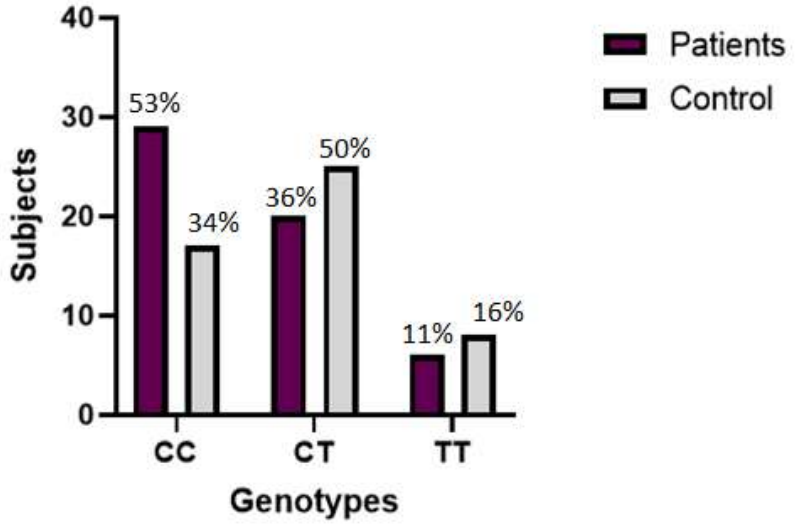

Figure 1: MALAT1 rs3200401 polymorphism genotype distribution in hypertensive patients and control group.

Table (1) showed the genotypic connection of the MALAT1 rs3200401 polymorphism with HTN in various inheritance models. Before adjustment, it was found that CC genotype was more frequent in patients' group, while CT+ TT genotypes were more frequent in control group with borderline P-value (Odds R = 0.462, 95% CI = 0.216-1.015; P = 0.052).

However, when adjusting for age and gender the CC genotype was significantly higher in patients' group while CT+TT genotypes were significantly greater in control group. (Odds R = 2.56, 95% CI (1.06-6.18); P = 0.034).

Table 1: HTN and the MALAT1 rs3200401 polymorphism association

Model	**Genotype**	**Patients (N = 55)**	**Control (N = 50)**	**OR (95% CI)**	**P-value**	***Adjusted OR (95% CI)**	***Adjusted P-value**
Codominant	**C/C**	29 (53%)	17(34%)	1.00 (Reference)		1.00 (Reference)	0.11
	C/T	20 (36%)	25(50%)	0.469 (0.198-1.048)	0.0941	0.39 (0.153– 1)	
	T/T	6 (11%)	8 (16%)	0.439 (0.134-1.384)	0.2232	0.39 (0.102-1.51)	
Dominant	**C/C**	29(53%)	17(34%)	0.462 (0.216-1.015)	0.052	2.56 (1.06-6.18)	**0.034**
	C/T-T/T	26 (47%)	33(66%)				
Recessive	**C/C-C/T**	49 (89%)	42(84%)	0.64 (0.198-1.977)	0.56	0.62 (0.46-5.51)	0.46
	T/T	6 (11%)	8 (16%)				

*CI= Confidence interval, OR= Odds ratio and *= adjusted by age and gender.*

In addition, the frequency of allele C is significantly higher in patients (71%) versus control (57%). However, compared to the control group, the T allele was considerably lower in the hypertensive patients, suggesting that allele T can be protective from HTN. (OR= 0.54, CI = 0.31 – 0.97, P = 0.03) (Table 2).

Table 2: MALAT1 rs3200401 polymorphism allele frequency in hypertensives and control

Allele type	Patients (N=55)	Control (N=50)	P-value	Odds Ratio	Confidence Interval
Allele C	71%	57%	**0.03**	0.54	0.31 – 0.97
Allele T	29%	43%			

Upon investigating the relationship between genotypes and lipid profile, table (3) shows that HDL-C, level in total population is significantly lower in TT genotype than other genotypes. However, there are no significant differences in TG, LDL-C, or TC levels between various genotypes.

There are no significant differences between genotypes and echocardiographic results across the population, as shown in Table (4).

Multivariate regression analysis revealed that HDL-C, LDL-C and rs3200401 are independent risk factor for hypertension (Odds R = 0.151, 95% Confidence I = 0.036 - 0.265, P < 0.005) (Table 5).

Table 3: Relationship between the genotypes and laboratory findings among total population

Parameter	Genotype C/C	Genotype C/T	Genotype T/T	P-value
TC (mg/dl)	169±40	177±34	156±40	0.186
HDL-C (mg/dl)	38±10	41±11	32±7	**0.028**
LDL-C (mg/dl)	104±34	107±31	99±28	0.702
TG (mg/dl)	132±50	144±46	124±67	0.369

HDL-C (High density lipoprotein cholesterol), LDL-C (Low density lipoprotein cholesterol), TC (Total Cholesterol), , and TG (Triglycerides).

Table (4) Relationship between the genotypes and Echocardiographic findings among total population

Parameter	C/C Genotype	C/T Genotype	T/T Genotype	p-value
LVMI	106±44	113±53	99±33	0.598
EF	64±8	64±7	61±9	0.439

EF (Ejection fraction), LVMI (Left ventricular mass index)

Table 5: Hypertension risk factors in the multivariate regression analysis

Variables	OR	95% CI	P-value
Age	0.005	(0.015 – 0.005)	0.349
Sex	0.116	(0.045 – 0.276)	0.156
HDL-C	0.022	(0.015 – 0.029)	**0.000**
LDL-C	0.005	(0.003 - 0.008)	**0.000**
Rs3200401	0.151	(0.036 - 0.265)	**0.010**

DISCUSSION:

The goal of the current study was to determine how the MALAT1 rs3200401 polymorphism in Egyptian patients compared to healthy control participants related to hypertension, dyslipidemia, and left ventricular hypertrophy.

A number of studies has investigated rs3200401 MALAT1 polymorphism in cardiovascular diseases. For an instant, a study looked into the connection between major adverse cardiac and cerebrovascular events and the MALAT1 gene polymorphism reported that the rs3200401 (CT and TT) genotypes were independent predictors of MACCEs in MI patients [12].

Additionally, in Egypt, according to **Fathy et al. (2021),** the rs3200401 genotypes CT and TT were distinct predictors of cerebral ischemic stroke in Egyptian individuals [11].

On the other hand, a study including CAD patients from Egypt discovered that MALAT1 rs3200401 (T/C) heterozygosity was linked to a lower Gensini score so it may operate as a protective measure for CAD [14].

Coronary atherosclerotic heart disease, onset of congenital heart disease and pulmonary arterial hypertension have all been linked to MALAT1 polymorphisms, according to previous research [4, 15 and 16].

Research studied four polymorphisms in MALAT1 including rs3200401 and their relationship with CAD in Chinese patients and age-, gender-, and ethnicity-matched control but they found no statistical significance [4].

The outcomes of SNPs in MALAT1 among hypertensives were considered in the light of aforementioned findings of earlier studies, and the SNP in MALAT1 with the highest minor allele frequency, rs3200401, was selected to be studied in this study.

When associating MALAT1 (rs3200401) polymorphism with HTN under different models of inheritance it was found that CT+ TT genotypes were more frequent in control group with borderline P-value before adjustment (Odds R = 0.462, 95% CI = 0.216-1.015; P = 0.052). However, after adjusting for age and gender the CT+TT genotypes found to be significantly higher in control than the hypertensives (Odds R = 2.56, 95% CI = 1.06-6.18; P = 0.034).

Furthermore, we discovered that the allele T was more prevalent in the control group than in the patients (Odds R= 0.54; 95% CI = 0.31 – 0.97, P = 0.03).

These results can be explained by findings by **Li *et al.* (2018)** who demonstrated that The C>T of the MALAT1 rs3200401 SNP causes a shift in the minimal free energy of 1.62 kcal/mol, which may be accompanied by the loss of binding sites on MALAT1 and result in the ineffectiveness of miRNA-lncRNA interactions [17]. This suggests that T allele can decrease MALAT1 sponging of miR-155, which allows miR-155 to demonstrate its protective effect [18]. This protective function of miR-155 emerged from its ability to recognize the 3′- untranslated region of human angiotensin II type-1 receptor (AT1R); a receptor which its overexpression can increase the potential of hypertension development. Therefore, miR-155 can lessen AT1R ability to raise HTN development by binding to it [19]. Thus, the T allele of rs3200401 can indirectly decrease HTN development.

Interestingly, using logistic regression analysis, in the population under study, MALAT1 rs3200401 SNP was discovered to be an independent risk factor for HTN (OR = 0.151, 95% CI (0.036 - 0.265), P = 0.01).

Up to our knowledge, our current research findings have revealed the first connection between MALAT1 SNP (rs3200401) & HTN. Our results point to the potential role of lncRNA MALAT1 as a fascinating target for future therapeutic intervention in hypertension management.

The present study shows that hypertensive patients have evidence of dyslipidemia in the form of significantly increased levels of LDL cholesterol with decreased HDL levels in comparison to the controls. However, there is no discernible variation in TC level.

About 50–80% of hypertensives have plasma lipid abnormalities. Endothelial dysfunction, a critical stage in the pathophysiology of atherosclerosis, thrombosis, insulin resistance, and hypertension, is influenced by dyslipidemia. Endothelial cells have been demonstrated to be toxic for TG-rich lipoproteins and LDL-C, although HDL-C is protective [20].

The current study also revealed that the levels HDL-C is significantly less in TT genotype than other genotypes. However, in 2020 a study discovered that in MI patients' rs3200401 TT genotype carriers had greater levels of total cholesterol than CC + CT genotype carriers did [21].

Data showed that TT genotype is associated with dyslipidemia. The current study showed association between TT genotype and decreased levels of HDL-C, moreover, **Li *et al.* (2020)** found association between TT genotype and increased levels of TC and both are markers of dyslipidemia [21].

Our study also revealed that LDL-Cholesterol and HDL- Cholesterol are independent risk factors for HTN. This result is in agreement with a study in Bangladesh that stated that lipid composition; LDL-Cholesterol, HDL- Cholesterol, TC and TG are independently associated with HTN [22].

Additionally, our research somewhat supports a study by Chen and Cheng (2022) who also found that LDL-C, and non-HDL were strong risk factors for HTN, but HDL-C and TG were not significantly related to incidence of HTN [23].

However, in the entire study population, there were no appreciable differences between various genotypes and LVMI, which means no association of this polymorphism and LVH.

To date, there are no other studies on the relationship between MALAT1 rs3200401 and LVMI. However, prior investigation found that the MALAT1 gene downregulates the target gene and suppresses the transcription of MyoD, leading to cardiac remodelling in hypertensive rats. They examined vascular remodeling and the rat left ventricle [24].

Their findings demonstrated that spontaneously hypertensive rats had LV weights that were considerably bigger than those of non-hypertensive rats, as well as moderate myocardial fibrosis, increased myocardial cell cross-section area and hypertrophic myocardial cells. Interestingly, SHRs had increased expression of MALAT1 in their heart tissues and aorta compared to non-hypertensive rats [24].

CONCLUSION

This study concluded that MALAT1 rs3200401 variants are independent risk factor for hypertension. The T allele was significantly lower in hypertensive patients than in control group; findings suggest that the T allele may be an HTN protecting one. Besides, MALAT1 rs3200401 is associated with

dyslipidemia, as HDL levels in total population were lower in TT genotype than other genotypes. However, there is no association between MALAT1 SNP (rs3200401) with LVMI.

REFERENCES

[1] Mills K, Stefanescu A, and He J (2020): The global epidemiology of hypertension. Nature Reviews Nephrology, 16 (4): 223–237.

[2] Reda A, Ragy H, Saeed K, and Alhussaini M (2021): A semi-systematic review on hypertension and dyslipidemia care in Egypt—highlighting evidence gaps and recommendations for better patient outcomes. J. Egypt. Public Health Assoc., 96 (1): 1–14.

[3] Dominiczak A, Delles C, Padmanabhan S (2017): Genomics and Precision Medicine for Clinicians and Scientists in Hypertension. Hypertension, 69 (4): e10–e13.

[4] Wang G, Li Y, Peng Y, J. Tang, and Li H (2018): Association of polymorphisms in MALAT1 with risk of coronary atherosclerotic heart disease in a Chinese population. Lipids Health Dis., 17 (1):1–7.

[5] Zhang X, Tang X, Hamblin M et al. (2018) :Long non-coding RNA Malat1 regulates angiogenesis in hindlimb ischemia. Int. J. Mol. Sci., 19 (6): 1723.

[6] Puthanveetil P, Chen S, Feng B et al. (2015): Long non-coding RNA MALAT1 regulates hyperglycaemia induced inflammatory process in the endothelial cells. J. Cell. Mol. Med., 19 (6):1418–1425.

[7] Rafieian-Kopaei M, Setorki M, Doudi M et al (2014): Atherosclerosis: Process, indicators, risk factors and new hopes. International Journal of Preventive Medicine, 5(8):927–946.

[8] Li D, Zhang C, Jian Li J et al.(2019): Long non-coding RNA MALAT1 promotes cardiac remodeling in hypertensive rats by inhibiting the transcription of MyoD. Aging, 11 (20):8792–8809.

[9] El-Sayed, E.H., Fathy, A., Al-Deen Younes, S.E. et al. (2023): Long Non-coding RNA Genes Polymorphisms H19 (rs2251375) and MALAT1 (rs3200401) Association with Rheumatoid Arthritis and Their Correlation with Disease Activity in a Cohort of Egyptian Patients: A Pilot Study. Biochem Genet

[10] Hu W, Ding, H, Ouyang A et al. (2019): "LncRNA MALAT1 gene polymorphisms in coronary artery disease: A case–control study in a Chinese population. Biosci. Rep., 39 (3)1–13.

[11] Wang J, Xiang J, Wu L et al. (2017): A genetic variant in long non-coding RNA MALAT1 associated with survival outcome among patients with advanced lung adenocarcinoma: A survival cohort analysis. BMC Cancer, 17 (1):1–8,

[12] Fathy N, Kortam M, Shaker O et al. (2021): Long Noncoding RNAs MALAT1 and ANRIL Gene Variants and the Risk of Cerebral Ischemic Stroke: An Association Study. ACS Chem. Neurosci., 12 (8):1351–1362.

[13] Zhang T, Luo J, Liu F et al. (2022): Long noncoding RNA MALAT1 polymorphism predicts MACCEs in patients with myocardial infarction. BMC Cardiovasc. Disord., 22 (1):1–9.

[14] Elwazir M, Hussein M, Toraih E et al. (2022): Association of Angio-LncRNAs MIAT rs1061540/MALAT1 rs3200401 Molecular Variants with Gensini Score in Coronary Artery Disease Patients Undergoing Angiography," Biomolecules, 12 (1)

[15] Zhuo Y, Zeng Q, Zhang P et al. (2017): Functional polymorphism of lncRNA MALAT1 contributes to pulmonary arterial hypertension susceptibility in Chinese people. Clin. Chem. Lab. Med., 55 (1): 38–46.

[16] Li Q, Zhu W, Zhang B et al. (2018): The MALAT1 gene polymorphism and its relationship with the onset of congenital heart disease in Chinese. Biosci. Rep., 38 (3):1-7.

[17] Li Y, Zhang, Yanxiu, Xu X et al. (2018): Clinical Chemistry and Laboratory Medicine Association of lncRNA Polymorphisms with Myocardial Infarction : A Case- Control Study and a Meta-Analysis, 1 (2): 1–8.

[18] Liang Z and Tang F (2020): The potency of lncRNA MALAT1/miR-155/CTLA4 axis in altering Th1/Th2 balance of asthma. Biosci. Rep., 40 (2).

[19] Zheng L, Xu C, Chen W et al. (2010): MicroRNA-155 regulates angiotensin II type 1 receptor expression and phenotypic differentiation in vascular adventitial fibroblasts, Biochem. Biophys. Res. Commun., 400 (4): 483–48.

[20] Ayoade O, Umoh I, Amadi C (2020): Dyslipidemia and Associated Risk Factors among Nigerians with Hypertension. Dubai Med J, 3:155–161.

[21] Li Y, Zhang D, Zhang, Yanxiu et al. (2020): Association of lncRNA polymorphisms with triglyceride and total cholesterol levels among myocardial infarction patients in Chinese population. Gene, 724: 143684.

[22] Choudhury N. K., Mainuddin A. K. M., Wahiduzzaman M. et al (2014): Serum lipid profile and its association with hypertension in Bangladesh. Vasc. Health Risk Manag., 10: 327.

[23] Chen S. and Cheng W. (2022): Relationship Between Lipid Profiles and Hypertension: A Cross-Sectional Study of 62,957 Chinese Adult Males. Front. Public Heal., 0: 1202.

[24] Li D. Zhang C., Li J et al. (2019): Long non-coding RNA MALAT1 promotes cardiac remodeling in hypertensive rats by inhibiting the transcription of MyoD, Aging (Albany. NY). 11(20): 8792–8809.