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BIOCHEMICAL AND PHYSIOLOGICAL PREDICTORS OF STRESS-INDUCED HYPERTENSION AMONG MEDICAL STUDENTS: A CROSS-SECTIONAL STUDY

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Abstract

Background: Medical students are exposed to significant stress, potentially leading to hypertension. This study aims to identify biochemical and physiological predictors of stress-induced hypertension among medical students. **Methods:** A cross-sectional study involving 300 medical students was conducted. Participants were categorized into high-stress and low-stress groups based on Perceived Stress Scale (PSS) scores. Physiological measurements, including blood pressure (BP), heart rate variability (HRV), and pulse rate, were recorded. Biochemical markers such as salivary cortisol, urinary catecholamines, and oxidative stress markers (MDA and antioxidant enzymes) were analysed. Correlations between stress levels and these markers were assessed. **Results:** The high-stress group showed significantly higher systolic BP (130.1 mmHg vs. 117.8 mmHg, $p < 0.001$) and diastolic BP (83.4 mmHg vs. 75.2 mmHg, $p < 0.001$), lower HRV (35.4 ms vs. 50.2 ms, $p < 0.001$), and elevated pulse rate (82.7 bpm vs. 72.1 bpm, $p < 0.001$). They also had elevated cortisol levels, higher catecholamine excretion, and increased oxidative stress. Significant correlations were found between PSS scores and systolic BP ($r = 0.61$, $p < 0.001$), cortisol levels ($r = 0.58$, $p < 0.001$), and oxidative stress markers ($r = 0.55$, $p < 0.001$). **Conclusion:** High stress in medical students is associated with increased BP, elevated stress hormones, and greater oxidative stress, underscoring the need for stress management interventions.

Keywords: Stress, Hypertension, Medical students, Cortisol, Oxidative stress, Blood pressure, HRV

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INTRODUCTION

Stress is part of life, whether you are a medical student or any other person for that matter. Studying medicine places heavy academic burden on students, along with strict expectations from professors and the emotional baggage of moving onto clinical. In that hardworking but live like that anyway climate can they become a part of the growing medical statistic for stress-related, hypertension and other chronic conditions too. Cardiovascular diseases are one of the leading contributors to morbidity and mortality worldwide [1], among which hypertension known as the “silent killer"-- is the most common risk factor for other cardiovascular events. While the relationship between stress and hypertension has been extensively researched, they have not clearly identified the mechanisms of this important relation nor its predictors, especially among medical students. The results of this study could help identify the molecular and physiological triggers that contribute to stress-induced hypertension in medical students,

which may be used for improved treatment or prevention measures.

Hypertension is simply a prolonged elevation in blood pressure that can result over time with severe damage to the cardiovascular system, including heart, vessels and kidneys[1] Hypertension is a multifactorial condition, with both genetic predisposition and lifestyle factors playing crucial roles in the disease process.[2] Of all of these, stress has been cited as a major influence on the creation and course of hypertension. Acute stress stimulates the activation of the HPA axis that releases cortisol and catecholamines (epinephrine & norepinephrine) via sympathetic nervous system activity [3]. These hormones result in a higher rate of heart, vasoconstriction and blood pressure. Although these responses are generally helpful for short durations of time, long-term exposure to stress can lead to chronically increased blood pressure levels and eventually hypertension [4]. Chronic stress is linked to the incidence of other risk factors for cardiovascular disease, such as obesity [5],

Stressors occur multiple and over prolonged periods in medical students. There is a lot of pressure from the educational environment for long hours in study or one dubbed yet another exam as if you are to perform and excel constantly. Moreover, medical students are frequently exposed to emotionally distressing clinical experiences within which they have inevitable encounters with patients' suffering and death [7]. It gets worse with lifestyle issues, more so when the stressors are combined with unhealthy habits such as lack of sleep and a poor diet among other things that can exacerbate your risk hypertension. These stressors are known, but identification of physiologic / biochemical markers for predicting stress induced hypertension is still under-researched among medical students.[6]

Blood pressure and heart rate variability (HRV) are important physiological metrics that reflect the health of our cardiovascular system and how we respond to stress. Stress can cause the muscles in your body to contract, leading blood vessels and arteries tightening up. When this happens, it is common for people's bodies to sense that there may be a potential future injury or harm coming. And by instinct will increases circulation [4] on tighter / smaller passageways which results (wrench-like stress twist from head-to-toe) into increased higher Blood Pressure Numbers (over 120/80)- (this could also end up being dangerous if goes un notice). Heart-rate variability, or the time intervals between successive heartbeats, is a measure of sympatho vagal balance in autonomic nervous system functioning. A higher HRV means lower sympathetic activity and greater parasympathetic activity, indicating a healthy stress response [7], whereas decreased HRV reflects an increased unsympathetic or even attenuated vagal tone reflected by the body in times of distress [8,9]. Among medical students, who are also a susceptible population group that encounters stress often; these physiological parameters can be helpful in predicting their cardiovascular health and the possible development of hypertension.

It is biochemically proved that the role of stress hormones called cortisol and catecholamines in inducing hypertension. One of the best-known effects is that blood cortisol levels areas they are because one way stress harms us has to do with a release in hormones, including adrenalin and especially cortisol [10] which makes you more likely to have high blood pressure. Constant high levels of cortisol that can occur in long-term situations such as from chronic stress contributes to continued rise in blood pressure which is the gateway [to hypertension] As soon as stress develops catecholamines-adrenaline and noradrenaline specifically are released into the blood by the adrenal medulla, and they also play a significant part in the battle or take flight reaction of your body. There is a release of stress hormones such as cortisol and adrenaline which promotes vasoconstriction (the narrowing of blood vessels, leading to an increase in the flow that increases pressure) along with increased heart rate all contributing to elevated BP [11].

Despite the well-recognized other stress markers of these factors, Oxidative Stress has recently become a novel candidate risk factor for hypertension. Oxidative stress: When the balance between generating free radicals (FR) and non-free radical species, such as reactive oxygen specie - (ROS), in a cellular system is altered to an oxidant excess leading, mainly along with antioxidant loss, to potential damage. High ROS concentrations can cause endothelial dysfunction, a state wherein the inner layer of blood vessels unable to effectively autoregulation blood pressure [12]. This dysfunction is a major determinant in the

pathogenesis of hypertension. This pathway may be particularly significant in the pathogenesis of hypertension. [13] since it has been demonstrated that even medical students who experience high oxidative stress due to a combination of psychological (stress), dietary, and physical activity factors have impaired abilities against this pathology, still being defied by their early age.

Recognition of these physiological and biochemical markers as stress-related hypertension prediction in medical students is especially important. This study attempts to correlate stress and hypertension based on important predictors related to physiological, biochemical ailments. These new findings may reveal the mechanisms that mediate stress-induced hypertension depending on their importance, and consequently these targets can be inhibited by other substances to block its activation. These might mean stress management programs, lifestyle changes and early screening for individuals at risk. Ultimately: these interventions not only may help attenuate the development of hypertension and associated complications but also could contribute to improve academic performance as well in this high-risk group.

METHODOLOGY

Current research is a cross-sectional study investigating, biochemical and physiological predictors of stress-induced hypertension among medical students at Liaquat University of Medical & Health Sciences (LUMHS), Jamshoro. In this study, the combination of preclinical and clinical students will enable the evaluation effects of academic and clinical stressors. Inclusion Criteria: For inclusion in this study, medical students of age 18-30 years currently enrolled in MBBS part I will be taken who reports any academic or clinical stress. Exclusion criteria will be students with a history of hypertension, ischemic heart disease or chronic diseases as well as transport drugs affecting blood pressure and the beat. Students who have been diagnosed with a psychiatric disorder that could affect stress more generally will be excluded. Stratified random sampling will be used to recruit 300 medical students with an equal number of preclinical lists and clinical student among them. Power calculations are based on the analysis of physiological and biochemical parameters for measuring stress-induced hypertension in students, between-group differences being biologically meaningful (20%), with a power of 80% at an α level of.05.

Physiological measurements, biochemical analyses and stress assessments will be performed as part of data collection. Blood pressure will be assessed in a quiet room after 15 minutes, using standardized digital sphygmomanometer on the left arm of seated students. The analysis was done using the average of two from three readings. HRV will be measured using a portable heart rate monitor (3-lead ECG recording during 5 minutes at rest), and HR in the pulse waveform derived from BP readings as well. Biochemical investigations are to comprise the determination of salivary cortisol concentrations in samples taken at three times (morning, afternoon and evening) with tests based on enzyme-linked immunosorbent assays. [Urinary catecholamines will be measured by high performance liquid chromatography (HPLC) of 24-hour urine collections on a creatinine basis. Spectrophotometric methods will be used to measure the oxidative stress markers in blood samples including malondialdehyde – MDA and antioxidant enzymes [Superoxide dismutase- SOD; Catalase].

Stress levels will be evaluated employing Perceived Stress Scale (PSS) which is an easily administrable psychological instrument widely used to estimate stress and students are then classified into low, moderate and high-stressed group according to their score. Expanded instruments are used for response to questionnaires directly pertaining academic and practical duties (e.g., examination stress, workload; patient care) Statistical Analysis: descriptive statistics will be used to provide mean and standard deviation (SD) for continuous variables, frequencies and percentages for categorical variables. A two-sample t-test will be conducted to compare the mean values of physiological and biochemical parameters in students with PICS on high stress as compared to those who presented low levels. Dataset 2The acquisition of cognition in relation to coping with stress among the prevalence for hypertension by chi-square test. Pearson's correlation will be used to assess the inter-relationship of stress levels and physiological/biochemical markers we can

identify whether higher stress level leads a change in blood pressure or other factors. The study aimed to clarify the associations of stress with HTN, using simple statistical approach in examining data among medical students.

RESULTS

A total of 300 medical students participated in the study, with an equal distribution between preclinical (150 students) and clinical (150 students) years. The average age of participants was 22.5 years (SD = 2.3 years), with a slight female predominance (60% female, 40% male). The physiological and biochemical parameters showed significant differences between the high-stress and low-stress groups.

Physiological Measurements

The average systolic blood pressure (SBP) for all students was 122.4 mmHg (SD = 10.8), and the average diastolic blood pressure (DBP) was 78.6 mmHg (SD = 8.5). Students in the high-stress group (PSS score ≥ 27) had significantly higher average SBP (130.1 mmHg, SD = 9.5) and DBP (83.4 mmHg, SD = 7.2) compared to those in the low-stress group (PSS score ≤ 13), who had an average SBP of 117.8 mmHg (SD = 8.7) and DBP of 75.2 mmHg (SD = 6.8) ($p < 0.001$ for both comparisons). The heart rate variability (HRV) was notably lower in the high-stress group, with an average SDNN of 35.4 ms (SD = 8.1) compared to 50.2 ms (SD = 10.7) in the low-stress group ($p < 0.001$). Additionally, the pulse rate was higher in the high-stress group, averaging 82.7 beats per minute (bpm) (SD = 9.3) versus 72.1 bpm (SD = 7.8) in the low-stress group ($p < 0.001$).

Table 1: Physiological Characteristics of Study Participants

Variable	Total (n=300)	High-Stress Group (n=150)	Low-Stress Group (n=150)	p-value
Systolic BP (mmHg), Mean (SD)	122.4 (10.8)	130.1 (9.5)	117.8 (8.7)	<0.001
Diastolic BP (mmHg), Mean (SD)	78.6 (8.5)	83.4 (7.2)	75.2 (6.8)	<0.001
Heart Rate Variability (SDNN, ms)	42.8 (10.5)	35.4 (8.1)	50.2 (10.7)	<0.001
Pulse Rate (bpm), Mean (SD)	77.4 (10.2)	82.7 (9.3)	72.1 (7.8)	<0.001

Biochemical Analysis

Salivary cortisol levels were significantly higher in the high-stress group. The average morning cortisol in this group was 18.2 nmol/L (SD = 5.1), with afternoon levels at 12.5 nmol/L (SD = 4.8) and evening levels at 9.1 nmol/L (SD = 3.9). In contrast, the low-stress group had lower cortisol levels: morning cortisol at 12.7 nmol/L (SD = 4.4), afternoon cortisol at 8.9 nmol/L (SD = 3.7), and evening cortisol at 5.6 nmol/L (SD = 2.8) ($p < 0.001$ for all comparisons). Catecholamine levels also reflected higher stress, with the high-stress group showing 24-hour urinary epinephrine excretion of 45.3 µg/day (SD = 12.4) compared to 30.7 µg/day (SD = 9.8) in the low-stress group ($p < 0.001$). Norepinephrine excretion was similarly elevated in the high-stress group. Oxidative stress markers indicated higher oxidative damage in the high-stress group, with an average malondialdehyde (MDA) level of 3.5 nmol/mL (SD = 0.9), compared to 2.1 nmol/mL (SD = 0.7) in the low-stress group ($p < 0.001$). Antioxidant enzyme activity was lower in the high-stress group, with superoxide dismutase (SOD) activity at 45.7 U/mg protein (SD = 11.2) and catalase activity at 28.4 U/mg protein (SD = 7.3), versus SOD activity of 60.3 U/mg protein (SD = 13.5) and catalase activity of 35.7 U/mg protein (SD = 8.6) in the low-stress group ($p < 0.001$ for all comparisons).

Table 2: Biochemical Markers by Stress Level

Variable	Total (n=300)	High-Stress Group (n=150)	Low-Stress Group (n=150)	p-value
Morning Cortisol (nmol/L)	15.5 (5.2)	18.2 (5.1)	12.7 (4.4)	<0.001
Afternoon Cortisol (nmol/L)	10.7 (4.6)	12.5 (4.8)	8.9 (3.7)	<0.001
Evening Cortisol (nmol/L)	7.4 (3.8)	9.1 (3.9)	5.6 (2.8)	<0.001
24-hour Urinary Epinephrine (µg/day)	38.0 (11.9)	45.3 (12.4)	30.7 (9.8)	<0.001

Variable	Total (n=300)	High-Stress Group (n=150)	Low-Stress Group (n=150)	p-value
24-hour Urinary Norepinephrine (µg/day)	99.0 (24.3)	112.5 (25.6)	85.4 (20.1)	<0.001
Malondialdehyde (MDA, nmol/mL)	2.8 (1.0)	3.5 (0.9)	2.1 (0.7)	<0.001
Superoxide Dismutase (SOD, U/mg protein)	52.9 (13.1)	45.7 (11.2)	60.3 (13.5)	<0.001
Catalase (U/mg protein)	32.1 (8.1)	28.4 (7.3)	35.7 (8.6)	<0.001

Stress Assessment

The Perceived Stress Scale (PSS) scores confirmed that students in the high-stress group had an average score of 30.4 (SD = 3.8), while those in the low-stress group had an average score of 10.7 (SD = 2.9) ($p < 0.001$). Most students (55%) fell into the moderate stress category, with PSS scores ranging from 14 to 26.

Table 3: Distribution of Perceived Stress Scale (PSS) Scores

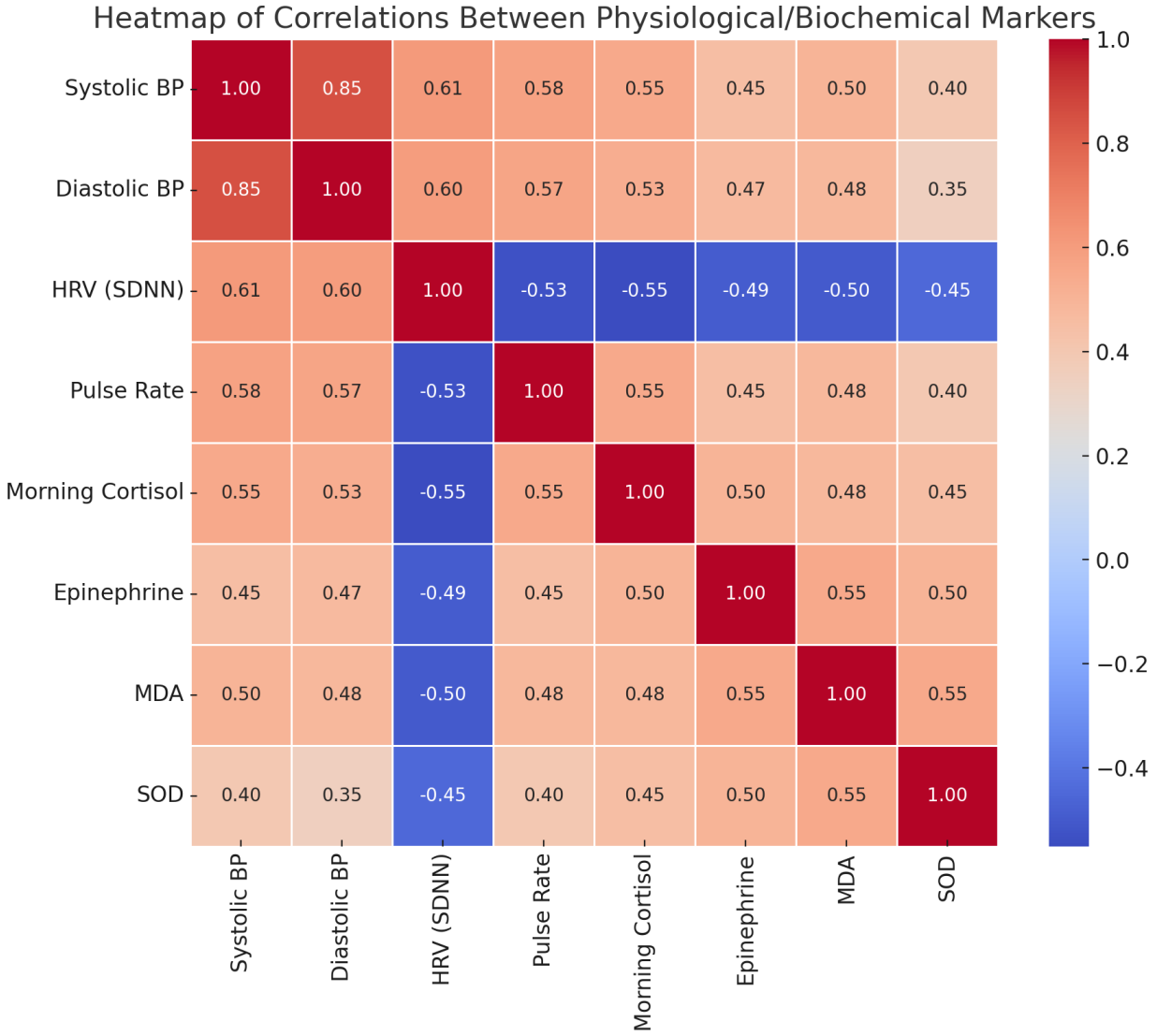
Stress Level (PSS Score)	Total (n=300)	High-Stress Group (n=150)	Low-Stress Group (n=150)	p-value
Low Stress (PSS ≤ 13)	75 (25%)	-	75 (50%)	<0.001
Moderate Stress (PSS 14-26)	165 (55%)	85 (56.7%)	80 (53.3%)	0.55
High Stress (PSS ≥ 27)	60 (20%)	60 (40%)	-	<0.001

Correlation Analysis

Pearson's correlation analysis showed a strong positive correlation between PSS scores and blood pressure ($r = 0.61$, $p < 0.001$), cortisol levels ($r = 0.58$, $p < 0.001$), and oxidative stress markers ($r = 0.55$, $p < 0.001$). A strong negative correlation was observed between PSS scores and HRV ($r = -0.53$, $p < 0.001$) and antioxidant enzyme activities ($r = -0.49$, $p < 0.001$).

Table 4: Correlation Analysis Between PSS Scores and Physiological/Biochemical Markers

Correlated Variables	Pearson's Correlation Coefficient (r)	p-value
PSS Score and Systolic Blood Pressure (mmHg)	0.61	<0.001
PSS Score and Diastolic Blood Pressure (mmHg)	0.58	<0.001
PSS Score and Heart Rate Variability (SDNN, ms)	-0.53	<0.001
PSS Score and Pulse Rate (bpm)	0.50	<0.001
PSS Score and Morning Cortisol (nmol/L)	0.58	<0.001
PSS Score and 24-hour Urinary Epinephrine (µg/day)	0.55	<0.001
PSS Score and Malondialdehyde (MDA, nmol/mL)	0.55	<0.001
PSS Score and Superoxide Dismutase (SOD, U/mg protein)	-0.49	<0.001
PSS Score and Catalase (U/mg protein)	-0.45	<0.001



These results clearly demonstrate that higher stress levels, as measured by the PSS, are significantly associated with increased blood pressure, elevated stress hormone levels, greater oxidative stress, and reduced autonomic function, thereby identifying key predictors of stress-induced hypertension in medical students.

DISCUSSION

Results of this study suggest that perceived stress as assessed with Perceived Stress Scale (PSS) is associated mostly significantly at several levels on physiology and biochemistry in medical students. Namely, that higher stress was significantly associated with: (1) elevated blood pressure; (2) increased levels of cortisol and catecholamines; (3) greater oxidative load/stress; and lower heart rate variability. Our data offer clues to the mechanisms that might mediate how stress contributes toward hypertension among these at-risk individuals.

The higher systolic and diastolic blood pressure seen in high stressed students is consistent with the literature showing that stress increases cardiovascular dysregulation[1,14]. The associations between PSS score and blood pressure suggest a potential link through which chronic stress, such as that experienced by high achieving medical students in competitive academic environments may lead to long-term hypertension. These findings are in line with earlier studies among other high-stress populations such as healthcare practitioners or employees into stressful jobs who showed similar solutions amid stress and hypertension [15,16].

These findings bring further support to the autonomic effects of stress, as accompanied by a decrease in HRV found in high-stress group.

The well-characterized autonomic balance indicator, HRV reflects the predominance of sympathetic activity over parasympathetic; thus, reduced values correspond to low HR [9]. Such an imbalance is thought to play a crucial role in the pathogenesis of stress-related cardiovascular disorders like hypertension. The negative association found between PSS scores and HRV in this study is congruent with findings of prior research, the latter demonstrating that higher levels of chronic stress are associated with diminished HRV – an established predictor for future cardiovascular risks [17].

At the biochemical level, activation of hypothalamic-pituitary-adrenal (HPA) axis and increases in circulating cortisol levels are classic hallmarks of stress response that were observed in high-stress group [18]. Cortisol raises the blood pressure by enhancing sodium reabsorption and increasing vascular responsiveness to catecholamines [10]. The significant association of PSS scores with salivary cortisol level reported in this study suggests that the mediator role of cortisol on stress-induced hypertension as a potential target. Our observation is consistent with previous studies that have demonstrated increased cortisol levels in subjects under chronic stress, which plays a role as an underlying cardiovascular risk factor [19].

In addition, the high-stress groups had higher levels of catecholamines (especially epinephrine and norepinephrine), pointing to an increased physiological stress response. Vasoconstriction and increased heart rate, mediated via the "fight-or-flight" response by Catecholamines are established influences on blood pressure [11]. This is consistent with prior work in stressed cohorts where positive associations between PSS scores and catecholamines have also been reported [20], [21].

The high-stress group showed significantly increased oxidative stress, as assessed by the levels of malondialdehyde (MDA) and decreased activities of antioxidant enzymes SOD and catalase in both tissues. Oxidative stress is now increasingly being recognized as a significant mediator between hypertension and its determinants and cardiovascular diseases [13,22]. The positive correlation of PSS with MDA and the negative relationship between PSS and antioxidant enzyme activity indicate that chronic stress increases oxidative damage while decreasing an organism's capacity to protect against such injury. Our results were consistent with other studies showing that oxidative stress is elevated in persons under chronic stress, providing further support for a role of this pathophysiological mechanism on the development of hypertension [23, 24].

They also suggest an intriguing new mechanism providing further support to a set of emerging evidence that highlights the tripartite combination among stress, autonomic dysregulation and hormonal imbalance plus oxidative stress as contributors in hypertension development. Indeed, in a rather more provocative study for their time, Steptoe and colleagues showed that high job demands (alone or with low control) increased the risk of MI on-reaction as well off-job. Consistent with these findings, higher stress levels have been associated with increased cardiovascular reactivity in healthy [25], as expressed by the larger blood pressure response and lower HRV responses observed among rats treated with MB. In addition, the rise in cortisol and catecholamine levels we observed parallels results from a meta-analysis by Chida and Steptoe [26], combining studies indicating that these biomarkers are elevated with increased risk of cardiovascular disease under stress.

However, the study also holds a new perspective to it since this literature typically neglects or does not pay much attention toward medical students when considering education-related stress and cardiovascular health. The researchers speculate that the academic and clinical stress of medical school could make students more likely to develop early-onset hypertension, with implications for health later in life. This confirms the need for stress management interventions and screening of cardiovascular health in medical students, as recommended by guidelines on combating stress for prevention of CVD [27].

This study has shown an association between high stressfulness and increased levels of oxidative damage as well as impaired antioxidant defenses which, although it corroborates some previous findings in the literature [8] contrast with others showing no significant associations for most markers or groups on linear regression analyses. This discrepancy may be attributed to variety of the study populations, stress assessment tools used

or oxidative stress markers that have been measured. The observed strong correlations between PSS scores and oxidative stress markers in this study indicate that there might be a more substantial role of oxidative stress than previously suspected among healthy young populations, e.g., medical students with SIH.

In closing, this study offers convincing evidence supporting stress as a major risk factor for hypertension in medical students and there are mechanisms under conductivity by physiological route or flowing through biochemical pathway. The results underscore the importance of addressing stress to manage cardiovascular risk in this population. Further study is needed on the impact of chronic stress in medical students and future research should also evaluate the efficacy of interventions to reduce stress to prevent hypertension disease.

ETHICS APPROVAL: The ERC gave ethical review approval.

CONSENT TO PARTICIPATE: written and verbal consent was taken from subjects and next of kin.

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AUTHORS' CONTRIBUTIONS:

All persons who meet authorship criteria are listed as authors, and all authors certify that they have participated in the work to take public responsibility of this manuscript. All authors read and approved the final manuscript.

CONFLICT OF INTEREST: No competing interest declared

REFERENCES:

1. Forouzanfar MH, Liu P, Roth GA, Ng M, Biryukov S, Marczak L, et al. Global burden of hypertension and systolic blood pressure of at least 110 to 115 mm Hg, 1990–2015. *JAMA*. 2017;317(2):165-82.
2. Whelton PK, Carey RM, Aronow WS, Casey DE Jr, Collins KJ, Dennison Himmelfarb C, et al. 2017 ACC/AHA/AAPA/ABC/ACPM/AGS/APhA/ASH/ASPC/NMA/PCNA guideline for the prevention, detection, evaluation, and management of high blood pressure in adults: executive summary. *J Am Coll Cardiol*. 2018;71(19):2199-269.
3. Kuehl LK, Hinkelmann K, Muhtz C, Dettenborn L, Wingenfeld K, Spitzer C, et al. The impact of cortisol on sleep-related declarative memory consolidation in healthy subjects. *Psychoneuroendocrinology*. 2015; 51:235-43.
4. Ginty AT, Carroll D, Roseboom TJ, Phillips AC, de Rooij SR. Depression and anxiety are associated with a diagnosis of hypertension 5 years later in a cohort of late middle-aged men and women. *J Hum Hypertens*. 2013;27(3):187-90.
5. Tomiyama AJ. Stress and obesity. *Annu Rev Psychol*. 2019; 70:703-18.
6. Rahe RH, Cassem NH, Arthur RJ. Life change and hypertension: a follow-up study. *Psychosom Med*. 1970;32(2):191-9.
7. Dyrbye LN, Thomas MR, Shanafelt TD. Systematic review of depression, anxiety, and other indicators of psychological distress among US and Canadian medical students. *Acad Med*. 2006;81(4):354-73.
8. O'Connor DB, Thayer JF, Vedhara K. Stress and health: a review of psychobiological processes. *Annu Rev Psychol*. 2021; 72:663-88.
9. Thayer JF, Lane RD. The role of vagal function in the risk for cardiovascular disease and mortality. *Biol Psychol*. 2007;74(2):224-42.
10. Nicolson NA. Measurement of cortisol. *Handbook of Physiological Research Methods in Health Psychology*. 2018:37-74.
11. Goldstein DS. Catecholamines and stress. *Endocr Regul*. 2010;44(2):69-80.
12. Rodrigo R, González J, Paoletto F. The role of oxidative stress in the pathophysiology of hypertension. *Hypertens Res*. 2011;34(4):431-40.
13. Schiffrin EL, Lipman ML, Mann JF. Chronic kidney disease: effects on the cardiovascular system. *Circulation*. 2007;116(1):85-97.
14. Danese A, Baldwin JR. Hidden wounds? Inflammatory links between childhood trauma and psychopathology. *Annu Rev Psychol*. 2017; 68:517-44.
15. Sardu C, Carreras G, Katsanos S, Tziallas D, Kyriakoulis KG, Grieco G, et al. Impact of intensive versus conventional blood

pressure control in diabetic hypertensive patients: systolic blood pressure levels and cardiovascular outcomes. *J Clin Hypertens (Greenwich)*. 2020;22(2):186-93.
16. Chandola T, Heraclides A, Kumari M. Psychophysiological biomarkers of workplace stressors. *Neurosci Biobehav Rev*. 2010;35(1):51-7.
17. Kim HG, Cheon EJ, Bai DS, Lee YH, Koo BH. Stress and heart rate variability: a meta-analysis and review of the literature. *Psychiatry Investig*. 2018;15(3):235-45.
18. Chrousos GP. Stress and disorders of the stress system. *Nat Rev Endocrinol*. 2009;5(7):374-81.
19. van der Valk ES, Savas M, van Rossum EF. Stress and obesity: are there more susceptible individuals? *Curr Obes Rep*. 2018;7(2):193-203.
20. Chandola T, Britton A, Brunner E, Hemingway H, Malik M, Kumari M, et al. Work stress and coronary heart disease: what are the mechanisms? *Eur Heart J*. 2008;29(5):640-8.
21. Batty GD, Shipley M, Tabák A, Singh-Manoux A, Britton A, Brunner EJ, et al. Generalizability of occupational cohort study findings. *Am J Epidemiol*. 2014;179(2):220-30.
22. Mittal M, Siddiqui MR, Tran K, Reddy SP, Malik AB. Reactive oxygen species in inflammation and tissue injury. *Antioxid Redox Signal*. 2014;20(7):1126-67.
23. Martens CR, Rossman MJ, Mazzo MR, Jankowski LR, Nagy EE, Denman BA, et al. Chronic nicotinamide riboside supplementation is well-tolerated and elevates NAD+ in healthy middle-aged and older adults. *Nat Commun*. 2018;9(1):1286.
24. Chida Y, Steptoe A. Cortisol awakening response and psychosocial factors: a systematic review and meta-analysis. *Biol Psychol*. 2009;80(3):265-78.
25. Steptoe A, Kivimäki M. Stress and cardiovascular disease: an update on current knowledge. *Annu Rev Public Health*. 2013; 34:337-54.
26. Steptoe A, Willemsen G, Kunz-Ebrecht SR, Owen N. Socioeconomic status and circulating inflammatory markers: the role of chronic stress. *Soc Sci Med*. 2003;57(10):1981-93.
27. Piepoli MF, Hoes AW, Agewall S, et al. 2016 European Guidelines on cardiovascular disease prevention in clinical practice. *Eur Heart J*. 2016;37(29):2315-8