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Biochemical Consequences of Cigarette Smoking in the Blood Profile of Adult Males in Peshawar, Pakistan

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ABSTRACT

Cigarette remains one of the most preventable and ongoing public health dangers, especially in developing countries such as Pakistan, where poverty, culture, and regulation obstruct effective tobacco control. The current research aims to investigate the early stage of biochemical effects cigarette smoking has on liver function and glucose handling in young adult males living in the district Peshawar rural area. A cross sectional comparative study is performed on 100 medically healthy male participants between 20-35 years of age. The participants are split into two groups, smokers ($n = 50$) who are smokers for over five years, and non-smokers ($n = 50$). Venous blood is drawn from each subject and sent to the laboratory for analysis with a semi-automated biochemical analyzer assessing the individuals Aspartate Transaminase (AST), Alkaline Phosphate (ALP), and Lactate Dehydrogenase (LDH) levels; capillary blood glucose levels (BGL) were drawn using a hand held glucometer. The study's statistics are running using SPSS version 26 and statistical analysis used independent sample t-tests to determine significance for intra group differences ($p < 0.05$). The findings of the data analysis showed statistically significantly increased ALP level ($p = 0.047$); BGL level ($p = 0.012$) in the smokers group than the non-smokers rather indicating the combined effects of early hepatic stress and inappropriate glucose disposition. Although AST and LDH levels are marginally higher in smokers, these differences are not statistically significant ($p > 0.05$), possibly due to the light to moderate smoking intensity among participants. These findings indicate that even low daily cigarette consumption can initiate subtle biochemical disturbances, particularly affecting liver enzyme activity and carbohydrate metabolism, which may predispose individuals to long term conditions such as liver disease and type 2 diabetes. This research contributes valuable data to the limited body of literature on smoking related biochemical alterations in rural Pakistani populations. It underscores the need for early biochemical screening and region specific public health interventions. Additionally, the study emphasizes the utility of liver enzyme profiling and glucose monitoring as non-invasive, cost effective tools for early detection of sub clinical damage in high risk groups. The findings advocate for the implementation of culturally tailored smoking cessation programs and preventive healthcare policies targeting young adults in under researched communities.

INTRODUCTION

Cigarette smoking is identified as a major cause of death all over the world accounting for about 63% of deaths on the planet (Hameed & Malik, 2021). In Pakistan, it is still one of the leading risk factors for a variety of health problems and diseases (Alam et al., 2008). It is noted that about 36% of men and about 9% of women smoke tobacco in Pakistan according to some national surveys (Jamshed et al., 2017; Nawaz & Naqvi, 2008). Smoking of tobacco is increasing in a rapid way although it is an important issue for public health. There is no doubt the control of tobacco consumption faces a number of challenges, especially when smoking has such an important economic impact. The tobacco industry adds significant revenue to the state and makes it more difficult for policy makers to take action to inhibit) tobacco consumption for public health (Mushtaq et al., 2011). Cigarette smoke contains more than 4,000 chemical compounds, at least 400 of which are known carcinogens. When a cigarette is burned, its temperature reaches about 316°C to about 482°C, and a large variety of substances are formed and they are in turn inhaled.

Cigarette smoke can be categorized into two types: mainstream smoke and side stream smoke. Mainstream smoke is the type directly inhaled through the filtered end of a cigarette, and it is typically denser, hotter, and contains a higher concentration of harmful chemicals. In contrast, side stream smoke is emitted from the burning tip of a cigarette and, although less concentrated, still poses health risks to passive smokers. Both forms are hazardous, but mainstream smoke is considered more dangerous due to its deeper penetration into the respiratory system. Once inhaled, the particles in cigarette smoke rapidly absorb moisture in the upper respiratory tract, increasing in size. Mainstream smoke travels into the lungs and deposits approximately 50% to 95% of its chemical load in the bronchi, bronchioles, and alveoli (Stratton et al., 2001).

Nicotine, the main addictive substance found in cigarettes, succumbs quickly after the oral mucosa absorbs it. It takes just seven seconds for nicotine to reach the brain, which causes a dopamine release and a brief feeling of relaxation (Juranić et al., 2018). Leaving smoking is difficult. Smokers may try many times and different options to quit smoking. An effective therapeutic option includes five forms of nicotine replacement therapy (NRT) (patch, nasal spray, inhaler, lozenge, and gum) and two non-nicotine medications, bupropion SR and varenicline. Other agents like naltrexone and cytosine have also been shown to assist in quitting smoking (Walker et al., 2014). Cigarette smoking effectively exposes the smoker to a wide variety of harmful chemicals, many of which negatively affect cells via oxidative stress and injury (Yao et al., 2008). Cigarettes also increase the risk of developing lung cancer by 60% in heavy smokers (Siddiqui et al., 2010), and increase the risk of developing type 2 diabetes depending on the number of cigarettes smoked daily (Afridi et al., 2022). Although this literature on smoking is evident, the prevalence of studies discussing cigarette smoking on biochemical variables in young adult men recruited from Pakistan is scarce.

BACKGROUND

Tobacco use, through multiple means but primarily through smoking cigarettes, has developed into one of the main threats to global health, and the preeminent preventable threat to global health. Tobacco is Longley associated with negative outcomes such as respiratory disease and cancer, but recent advances in the area of biochemistry have shown that tobacco is far more systemic in its effects than previously appreciated, especially in the context of metabolic and hepatic function. Cigarette smoke is no longer simply a pulmonary hazard, it is now a known biochemical disruptor of the essential processes of the human body (U.S. Department of Health and Human Services, 2020).

In developing nations like Pakistan, smoking prevalence for adult males remains extremely high. The Global Adult Tobacco Survey Pakistan 2014 indicated nearly one in four adults in the country are tobacco users with smoking cigarettes being the most common form of tobacco use. Much has been done from an awareness standpoint, however, tobacco control efforts are challenging given prevailing cultural norms, economic pressures from tobacco farming, and insufficient enforcement of tobacco control policies (WHO 2021).

Emerging studies in biomedical sciences have shifted the focus from long term morbidity to early, subclinical markers of disease. Liver function markers such as Aspartate Transaminase (AST), Alanine Transaminase (ALT), Alkaline Phosphatase (ALP), and Lactate Dehydrogenase (LDH), as well as blood glucose levels, have recognized as sensitive indicators of physiological disruption due to oxidative stress induced by smoking (Pérez-Rial et al., 2019). Reactive oxygen species (ROS) generated from tobacco smoke play a pivotal role in damaging cellular membranes, DNA, and proteins, leading to metabolic dysregulation and the activation of inflammatory pathways (Valavanidis et al., 2013).

The specific risks of cigarette smoking apply to young adults, especially men between Tracy and 35 years Gill neutral). When they are constantly exposed to tobacco, these individuals do not usually know the early internal damage happening to their body during their tobacco exposure, as basic physical symptoms which are easily diagnosed are not visible. This justifies utilizing detailed biochemical profiling as a proactive diagnostic strategy to monitor the latent effects of tobacco on their health. Furthermore, the country effect of smoking in conjunction with glucose homeostasis is being recognized for its link to insulin resistance and potential link to type 2 diabetes, particularly in habitual light to moderate smokers (Pan et al. 2015).

As discussed earlier there is very little data concerning rural Pakistani groups, and habitual cigarette smoking and how that relates to biomarkers. There are no studies completed regarding even the normal patterns of biochemical changes exhibited by young male cigarette smokers in less urbanized areas Badge paste314ha, but this level of understanding about the early biochemical changes is something that public health professionals would need to educate themselves on to assist in developing area specific health initiatives, enhancing a aspects of screening events for screening,, and developing cessation programs to continue to provide support to populations at risk.

SIGNIFICANCE OF THE STUDY

This research has a considerable value to both science and public health, which is particularly relevant to Pakistan, where cigarette smoking is most prevalent among young adult males. The dangers of tobacco have been well documented, however much of the research has focused on severe distal clinical outcomes from cigarette addiction, such as cancer, cardiovascular disease, and chronic respiratory disease. This study contributes a new dimension to the existing literature by examining liver function markers and blood glucose levels in a medically healthy young population; which reflects the biochemical effects of smoking in real time, as opposed to longitudinally examining the (often severe) clinical consequences to health. These biochemical markers (AST, ALP, LDH, and BGL) of the liver and glucose metabolism are early biochemical flags of physiological potential disruption of tissue function long before any clinical manifestations of disease or detrimental behaviors occur.

Additionally, another important contribution of the study lies in the fact that the study cohort is a semi-urban under researched population in District Peshawar. Localized biomedical data on the subclinical physiological and pathological impact of tobacco consumption and the harmful effects on specific rural populations in Pakistan is an uncelebrated gap within the literature. Filling this gap is significant in terms of future research and informing on the importance of regionally specific health assessments. The use of structured methods, including blood serum analysis and statistical comparisons between smokers and non-smokers, provides credible evidence of smoking-induced changes in metabolic and hepatic functions. This evidence may prove essential in improving local public health awareness and preventive healthcare efforts. The findings of elevated ALP and blood glucose levels among smokers are particularly significant. These changes, although subtle, suggest early hepatic stress and impaired glucose regulation, both of which are risk factors for more serious conditions such as liver disease and type 2 diabetes. By identifying these biomarker changes early enough we can improve the quality and power of prevention services through their uptake and opportunity for lifestyle changes before there are permanent effects. This study is invaluable for the field of early detection and risk mitigation especially for identifying maritime illicit tobacco users so that health policy makers can identify the population of interest.

In addition, this research has particular relevance for the consideration of culturally safe public health campaigns and cessation programs for young adults that recognize and represent, and incorporate key messaging that is appropriate to their rural context, specifically where use and/or normalization of tobacco use occurs in various social situations. This research also clearly demonstrates a means to transfer evidence into practice which integrates biochemistry, botany and narcotic science, and contributes to the evidence base for interdisciplinary research as a rationale for substantial progress for public health and the shifting nature of complex health issues.

addition, the connection between using naturally extracted products (tobacco) with undesirable biochemical effects suggests a further degree of understanding lifestyle constructs, and the paradigm around how these types of actions may work in a biological system in our body. In addition to the comments about health, the methods of the study present promising avenues for future research. The methods that embraced semi-automated analyzers, intentional sampling of participants, and analytical measures like an independent sample t-test outline an investigative process that can be a procedural framework for future research conducted by others that can even be used in larger cohorts, longer durations of tobacco product usage, and even biochemical measures that could capture broader spectra of health risk factors relating smoking. The results can address the surveillance of non-communicable diseases (NCD) in Pakistan, with regard to identifying subclinical measures that can impact screening and monitoring practices (in terms of tobacco use) within primary health settings.

METHODOLOGY

PARTICIPANT SELECTION AND GROUPING

This research was completed from January to March 2021 in different locations of Peshawar District. For this study, 100 adult males were randomly chosen and randomized into a test and control group. The test group was made up of 50 smokers. The control group consisted of 50 non-smokers. All subjects in the smokers group had a history of smoking cigarettes for a minimum of 5 years, smoked 10 or less cigarettes, and were medically stable. Sociodemographic Information like age, number of cigarettes smoked per day, and relevant medical history were determined through a structured interview administered to the subjects. The smokers group had a mean age of 25.96 ± 3.886 years, and the non-smokers group had a mean age of 28.96 ± 3.987 years. The overall mean age for both groups combined was 27.45 ± 4.193 years.

BLOOD SAMPLE COLLECTION

Up to 5 ml of venous blood is collected from subjects using sterile and disposable 5 ml syringes. The blood is placed in labelled gel tubes immediately to avoid contamination. The gel tubes are labelled with the subject's name and their age; when collecting blood, some blood samples are coagulated, and the rest were refrigerated in gel tubes, labels on each gel tube were placed upright, identification tag is removed from the tube and placed on a master document where the blood collection is identified and all related subjects' biochemical tests arelisted.

The blood samples are centrifuged at 3000 rpm for 10 minutes at room temperature, when ready to be analyzed, the above was followed for each blood sample.

LIVER FUNCTION TEST EVALUATION

Assessment of liver function is undertaken by analyzing serum samples using the Micro lab biochemical analyzer. The biochemical analyzer uses a photometric approach by measuring the analytic concentration based on ultraviolet light absorption driven by Beer Lambert's Law (Maire, 1990). The resulting intensity of the light passing through the solution created an electrical signal to the intensity of the analytic concentration. Salt is measured for concentration using the calibration procedures to ensure accuracy of measures by comparing the light absorbance with analytic concentrations known (Mäntele & Deniz, 2017). Diagnostic kits produced by Spinreact (Spain) and Diatech (Switzerland) for Aspartate Transaminase, Alkaline Phosphatase, and Lactate Dehydrogenase are used. Results, documented and results tabulated ready for future investigation.

BLOOD GLUCOSE

Blood glucose levels are taken using a portable glucometer. The portable glucometer contains a test strip that is inserted. A small drop of capillary blood is acquired through the pricked index finger with sterile single use, spring loaded lancet. A drop of blood is placed on the test strip and 5 minutes later a reading was obtained. This was conducted for each participant to ensure reliability and all readings were carefully recorded.

STATISTICAL ANALYSIS

Data gathered are analyzed, using SPSS version 26. All measured parameters are presented means ± standard error (SE). An independent sample t-test includes a comparative analysis between group's smoker and non-smoker, examining statistical differences in the biochemical parameters for the test and control group and checking the significance of the variance. A p-value of less than 0.05 is considered statistically significant.

RESULTS

Visual Analysis of Smoking Effects on Biochemical Parameters

- **Figure 1**: Cigarette Consumption Distribution
- **Figure 2:** Biochemical Parameters Comparison (Bar Chart)
- **Figure 3:** Separate Pie Charts for Biochemical Parameters
- **Figure 4:** Trend Line of Biochemical Parameters

DISTRIBUTION OF CIGARETTE CONSUMPTION AMONG SMOKERS

Table 1 provides the distribution of the frequency of daily cigarette use of participants in the experimental group (smokers) using the following intervals:

- 6 respondents using 1 to 2 cigarettes in one day
- 8 respondents using 2 to 3 cigarettes in one day
- 11respondents using 3 to 4 cigarettes in one day
- 6 respondents using 4 to 5 cigarettes in one day
- 7 respondents using 5 to 6 cigarettes in one day
- 2 respondents using 6 to 7 cigarettes in one day
- 3 respondents using 7 to 8 cigarettes in one day
- 2 respondents using 8 to 9 cigarettes in one day
- 1 respondents using 9 to 10 cigarettes in one day

- 2respondents using 10 to 11 cigarettes in one day
- 2 respondents using 11 to 12 cigarettes in one day

TABLE 1. *DAILY CIGARETTE CONSUMPTION AMONG SMOKERS (N = 50)*

Number of Cigarettes\Day	Frequency	Percentage
1 to 2	6	12
2 to 3	8	16
3 to 4	11	22
4 to 5	6	12
5 to 6	7	14
6 to 7	2	4
7 to 8	3	6
8 to 9	2	4
9 to 10	1	2
10 to 11	2	4
11 to 12	2	4

COMPARATIVE STUDY OF BIOCHEMICAL PARAMETERS IN SMOKERS AND NON-SMOKERS

The comparison of blood biochemical parameters in smokers and non-smokers is Summarized in Table 2. The study found that:

- Aspartate Transaminase (AST) levels are increased in smokers compared to non-smokers by 17% but not statistically significant.
- Alkaline Phosphatase (ALP) levels are higher with smokers, and by 12%, and statistically significant.
- Lactate Dehydrogenase (LDH) is increased in smokers by 4% but not significant.
- Blood Glucose Level (BGL) is a 7% increase in smokers compared to non-smokers, and statistically significant.

TABLE 2. MEAN ± STANDARD DEVIATION AND P-VALUES OF BLOOD BIOCHEMICAL PARAMETERS IN SMOKERS AND NON-SMOKERS (N = 50 PER GROUP).

PARAMETER (Units)	SMOKERS(Mean ± SD)	NON- SMOKERS GROUP(Mean ± SD)	P - VALUE	SIGNIFICANCE
Aspartate Transaminase (IU\L)	28.71 ± 1.854	24.51 ± 1.667	0.094	Not Significant
Alkaline Phosphatase (IU\L)	224.77 ± 7.197	200.31 ± 9.817	0.047	Significant
Lactate Dehydrogenase (IU\L)	250.79 ± 10.454	241.01 ± 8.728	0.491	Not Significant
Blood Glucose Level (IU\L)	95.81 ± 2.465	89.15 ± 0.808	0.013	Significant

Note: Data represents mean ± standard deviation. Values were calculated using an independent sample *t*-test via SPSS (version 26). A P-value < 0.05 is considered statistically significant.

DISCUSSION

Liver enzyme abnormalities are widely recognized as early indicators of liver dysfunction. One of the primary functions of the liver is detoxification, and cigarette smoking exerts considerable strain on this process. This burden can trigger inflammation and fat accumulation in liver tissues, eventually contributing to the onset of liver-related diseases. While several risk factors for altered liver enzymes are known, there remains ongoing scientific debate about the extent to which smoking directly influences these changes. Liver function can vary significantly under physiological and environmental conditions such as hypoxia, exposure to pathogens, certain medications, hormonal fluctuations (e.g., menstruation), and rapid musculoskeletal development.

Cigarette smoking is also associated with insulin resistance. In male smokers, the body's ability to utilize glucose in response to insulin is reduced by 10% to 40% compared to non-smokers (Chang, 2012). Moreover, recent studies have highlighted that active smokers exhibit diminished pancreatic beta cell activity, a critical factor in insulin production and glucose regulation (Morimoto et al., 2013). This study was designed to explore the association between cigarette smoking, liver enzyme activity, and blood glucose levels in adult males.

Of all the serum biomarkers investigated, Alkaline Phosphatase (ALP) is indeed a rather intriguing marker. ALP elevation may have clinical relevance because it could indicate liver damage, bone disease, or other disease processes. A surge in ALP indicates hepatic injury to hepatocytes resulting in release of ALP to circulation. Although normal values for ALP vary between labs, gender and age; the general reference range is between 98 - 279 IU/L in the adult (Jatoi et al., 2007). We demonstrate markedly greater levels of ALP in smokers when compared to non-smokers which are consistent with findings from Salihu (2019), Atta et al. (2019), Modawe et al., (2019) and Hamza and Naji (2020) which indicate also higher serum levels of ALP in smokers. The elevations are likely due to the harmful effect of nicotine and other chemicals in tobacco smoke which create an environment of nitrosative stress which overwhelms nitrogen based reactive species' ability to be removed from the system. Moreover, this is likely to interfere with the restoration of enzymes being released from the liver (El-Zayadi, 2006).

Aspartate Aminotransferase (AST) is another enzyme of significant clinical importance which occurs in liver cells, cardiac tissues, muscles, red blood cells, kidneys, and pancreas. AST is generally located in serum concentrations of 5-40 IU/L. AST can increase significantly with tissue damage to as much as 10-20-fold normal concentrations which makes it a suitable indicator of tissue injury (Hafkenscheid & Dijt, 1979). Lipid peroxidation would damage the plasma membranes of liver cells due to the cigarette smoke; therefore, a serum aminotransferase level would indicate typing of liver damage (Rochling, 2001). However, in this study, a comparison of smokers to non-smokers did not show AST significantly different. In fact, findings of Ruchir et al. (2017) determined that while there was a trend of smokers with increased AST, it was most truly observed in heavy smokers, while Hamad et al. (2020) and Turki (2021) found increased AST in other smokers, Atta et al. (2019) found a decrease in AST. One factor as a reason for the variability in AST may be smoking exposure dose and/or duration.

Lactate Dehydrogenase (LDH) is an enzyme involved in anaerobic metabolism found in almost all body cells and tissues, especially in cells associated with carbohydrate metabolism. LDH is higher in the liver, heart, skeletal muscles, red blood cells, kidneys, and lower in the brain, lungs, and smooth muscles. Because LDH is present in many tissues, it can be elevated after a number of physiological and pathological states (Klein et al., 2020). The reference range for LDH is approximately 140–280 IU/L (Dmour et al., 2020). In this study, levels of LDH did not differ between smokers and non-smokers, supporting earlier findings by Wannamethee and Shaper (2010), Turki (2021), Abdul-Razaq and Ahmed (2013) and Atta et al. (2009) showing changes in liver enzymes due to smoking, which was a dose-dependent response to exposure. Other studies however have reported elevated LDH levels in smokers (e.g., Modawe et al., 2019; Jaafar, 2020; Hamza and Naji, 2020), suggesting LDH variation can be influenced by multiple variables, including an individual's smoking history, general health and metabolism.

One of the most significant findings of this study is the increasing blood glucose levels in smokers compared to non-smokers. This finding is in support of an earlier study that found elevated blood glucose levels in smokers, with the explicit mechanism being oxidative stress from cigarette smoking producing excessive oxidative species (ROS) in the body, leading to reactive oxygen species stress (Bhattacharjee et al., 2015) . It has been established that reactive oxygen species (ROS) can affect insulin signaling and impair glucose tolerance, which can lead to elevated blood glucose, although the exact biochemical mechanism is still unknown. Smoking may be involved in releasing stress hormones such as epinephrine and norepinephrine that appear to lead to gluconeogenesis and glycogenolysis, and both of these processes cause elevated glucose levels (Vu et al., 2014). This was consistent with the findings of Lakshmi (2018), Sahab (2019), Bassey et al. (2020), and Hmood et al. (2020) all of whom found that smokers were more likely to be hyperglycemic.

In summary, this study suggests that smoking cigarettes I.T. could be associated with a statistically significant increase in some biochemical parameters. To clarify, liver function tests such as ALP are statistically significant suggesting some hepatic stress; however, the mild smokers in this study only had a minimal increase in AST and LDH; conversely, the blood glucose did yield a statistically significant increase in blood glucose in the smoking group.

CONCLUSION

The findings of this study demonstrated that the effect of cigarette smoking on the biochemical indicators of health is very prevalent on factors including both dosing and the frequency at which producers use cigarettes on a daily basis. The study indicated that smoking to a small extent may affect liver health as suggested with the significant finding of higher mean Alkaline Phosphatase (ALP) concentrations in smokers; however, there are no significant differences when it came to the other liver indicators, Aspartate Aminotransferase (AST) and Lactate Dehydrogenase (LDH), as related to hepatic liver health, which means that liver dysfunction may not be of concern to people smoking at low doses or low frequencies.

An additional significant finding that is identified is that blood glucose levels are found to be higher in cigarette smokers when compared to non-smokers, as smokers can be reported to be skewed in glucose regulation, which may place smokers at higher risk for type II diabetes later on.

Since the research only investigated a small number of liver enzymes, I would recommend future studies which include additional indicators of liver health to fully measure the impacts of smoking related health. Moreover, as the biochemical analyses are conducted using a semi-automated Micro lab system susceptible to human handling errors future studies should employ fully automated, high precision laboratory equipment to ensure greater accuracy and reliability of results.

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