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AGE-STRATIFIED PROGRESSION OF NAFLD TO NASH IN PAKISTAN: PREVALENCE, RISK FACTORS, AND CLINICAL IMPLICATIONS

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Abstract

In this study, we aimed to quantify the progression of non-alcoholic fatty liver disease (NAFLD) to non-alcoholic steatohepatitis (NASH) in Pakistan by age group and therefore fill a critical gap in local knowledge regarding prevalence, risk factors and clinical characteristics of NAFLD and NASH across different ages. Using cross-sectional analysis with 450 patients from tertiary care hospitals divided by age groups (young adult 18-35 years, middle-aged 36-55 years, and old adult > 55 years), we diagnosed patients with NAFLD and NASH using imaging, biochemistry and clinical criteria. Our data showed that as age increased disease severity of NAFLD and NASH also increased significantly, with the highest prevalence of NAFLD in middle aged adults (52%) and NASH increasing from 10% in young adults to 40% in old adults ($p < 0.05$). NASH was strongly associated with metabolic comorbidities (diabetes, obesity, dyslipidemia)–particularly in the older adult population where diabetes prevalence was approximately 62%. The levels of liver enzymes (ALT/AST) and markers of fibrosis were also found to be increased with advancing age, indicating cumulative exposure to metabolic stress. Our data also indicate that most of the younger patients had simple steatosis whereas the older adults had more severe histological damage, indicating that NAFLD is a progressive disease. Finally, these results demonstrate the need for urgent interventions aimed at increasing screening and promoting lifestyle changes, particularly in the elderly population, to help combat the increasing burden of NAFLD-related complications in Pakistan.

INTRODUCTION

The worldwide public health challenge that is known as the non-alcoholic fatty liver disease (NAFLD). NAFLD's progressive form, called non-alcoholic steatohepatitis (NASH), increases the risks of developing cirrhosis and

hepatocellular carcinoma [1]. Rapid urbanization, changes in diet, rising incidences of metabolic disorders such as obesity, and type 2 diabetes [2], have caused the rise of NAFLD to grow in Pakistan. Due to the difference in risk factors between South Asian and Western populations, there is very little epidemiological information about age-stratified data on NAFLD and its progression to the disease NASH [3]. Estimates indicate that there is an extremely high prevalence of NAFLD in Pakistan; with studies indicating the prevalence rates of NAFLD in Pakistan are comparable to the global hotspots (North America and the Middle East) for this disease [4]. The increase in prevalence of NAFLD has been intensified by the increase in the prevalence of metabolic syndrome, which is estimated to affect approximately 23% of the adult population [5]. Most of the studies done about NAFLD, have been focused on overall prevalence or specific high-risk groups (diabetic); age-specific patterns of progression of NAFLD are not described in the currently available literature [6]. The importance of having this information can be seen in that age is a known predictor of severity of NAFLD; older adults have the greatest rate of NASH and fibrosis due to having been exposed to metabolic factors for a longer period of time than younger adults [7]. The clinical and public health impact of Non-Alcoholic Fatty Liver Disease (NAFLD) in Pakistan is large, particularly given the healthcare system in Pakistan is already stretched by a large number of Infectious Diseases and limited resources, NAFLD contributes to chronic liver disease, likely leading to a healthcare crisis in the very near future [8]. If interventions are not put in place to reduce the economic and social impact of NAFLD and its progression to advanced liver disease in older adults, the total costs of this disease will increase substantially and there will be large numbers of individuals in Pakistan progressing to end-stage liver disease [9]. The co-occurrence of NAFLD and other metabolic diseases such as diabetes and dyslipidemia - indicates the need for integrated management strategies to address the burden imposed on the individual as a result of multiple comorbidities or multisystem morbidity [10]. This study will address these knowledge gaps in the literature. We will comprehensively describe the spectrum of NAFLD and Non-alcoholic Steatohepatitis (NASH) across the lifespan in Pakistan. The first aim of this project will be to compare the prevalence and clinical characteristics of NAFLD and NASH by age group (young - 18 to 35 years, middle aged - 36 to 55 years, and older - > 55 years). The second aim will be to identify the Metabolic Risk Factors by each of the above mentioned age categories, and the relationship of Risk Factors on Disease Progression. We hypothesize that NAFLD severity and prevalence of NASH will increase with age due to cumulative Metabolic Damage and Hepatic Resilience that declines with age. The importance of this research is that it will potentially develop age-appropriate screening and intervention strategies for individuals living with NAFLD in Pakistan.[11].

Literature Review

The epidemiology of Non-Alcoholic Fatty Liver Disease (NAFLD) has evolved significantly over the past few decades; NAFLD is now recognized as the most prevalent chronic liver disease on earth [1]. The estimated global prevalence of NAFLD is variable, ranging between 25% and 35%; however, different regions demonstrate marked differences in NAFLD prevalence/incidence. In addition to genetic predisposition, there are also observed differences in NAFLD prevalence/incidence due to rapid changes in dietary patterns (nutritional transitions), particularly in South Asia [3]. Within Pakistan, urbanization, sedentary lifestyle behaviours, and the extremely high prevalence of metabolic syndrome have created the ideal environment in which to promote NAFLD; however, there is limited age-specific data on the prevalence/incidence of NAFLD in the country [4]. Studies completed within Pakistan have, for the most part, only reported prevalence/incidence of NAFLD at a population-level, or among sub-populations at high risk of NAFLD, such as people with Type 2 diabetes mellitus. For example, in South Asia, a meta-analysis completed on the prevalence of NAFLD reported an overall (pooled) prevalence of NAFLD of approximately 34% and also reported that individuals diagnosed with NAFLD from an urban setting in Pakistan

demonstrate a higher prevalence of NAFLD than those diagnosed with NAFLD from rural Pakistan (38% vs. 22%, respectively) [12]. Many existing studies evaluating prevalence/incidence of NAFLD do not separate the age of study subjects and the findings of the studies from an age perspective, however, there is ample evidence demonstrating that the progression of NAFLD is largely the result of the individual's age. The progression of NAFLD is primarily a result of the biological mechanisms of aging, including mitochondrial dysfunction, increased oxidative stress, and altered lipid metabolism [7]. Age-related patterns exist in many metabolic risk factors for NAFLD, including obesity, insulin resistance and dyslipidemia. In cohort studies conducted in Pakistan this trend has been demonstrated by higher rates of these comorbidities for middle aged and older participants compared to younger participants, similar to global trends while also differing in some regard on a more local level [10]. For example, while in Western populations obesity has been found to be a primary contributor to the development of NAFLD, in South Asia, almost 30% of individuals have lean NAFLD, suggesting different pathways toward the development of NAFLD (i.e., visceral obesity and/or specific genetic variations such as PNPLA3) [11]. This difference also highlights the need for age and ethnic-specific models to stratify risks associated with NAFLD as this was further demonstrated in the work of [12] who provide an example of a unique inflammatory signature associated with patients diagnosed with NAFLD from Pakistan. Another significant literature gap in regards to NAFLD is the progression of the disease from NAFLD to NASH. Globally, the prevalence of NASH has been associated with an increase in age; estimates suggest that 10 to 20% of middle aged individuals; and 30 to 40% of elderly individuals (> 60 years) are affected by NASH [13]. Unfortunately, in Pakistan data regarding the prevalence of NASH is scant with most studies utilizing surrogate markers of elevated liver function tests and/or (biochemical tests) for determination of NAFLD rather than using histopathological confirmation [14]. This limitation is also concerning since advanced fibrosis has been reported to have a high prevalence in older South Asian patients as shown by [15] who reported that almost 25% of NAFLD patient's greater than 50 years of age had significant fibrosis despite having a normal BMI.

Implications of the age group-related NAFLD data have significant repercussions for clinical management. For example, older individuals have a higher likelihood of eventually developing both cirrhosis and hepatocellular carcinoma, and they also do not respond as well to lifestyle modifications as younger individuals; thus, tailored treatment strategies are necessary [17]. In addition, the relationship between NAFLD and extrahepatic diseases (such as cardiovascular and chronic kidney disease) is markedly more pronounced in older adults, resulting in a "cascade" of morbidities across multiple organ systems due to both NAFLD and all of its extrahepatic complications [18].

Nevertheless, many critical knowledge gaps still exist. The majority of studies occurring in Pakistan are conducted at an academic/research institution and are therefore subject to selection bias; in addition, many studies that have longitudinal designs to follow disease progression do not have valid follow-up data. Also, due to the lack of standardized methods for diagnosing NASH (especially within low-resource countries), estimating the population-based prevalence of NASH cannot be performed accurately, which limits cross-study comparisons [19].

Our research provides an extensive evaluation of NAFLD and NASH in Pakistani people of all ages through the use of imaging, biochemical and clinical data, correcting for limitations of existing literature. Our analysis directly investigates the role of metabolic risk factors in combination with aging on the severity of NAFLD and to identify new strategies for intervention. Our results contribute significantly to the existing literature by providing regional epidemiology and inform the need for individualised age-based approaches to the management of NAFLD, as opposed to the traditional "one size fits all" approach.

3. Patients and Methods

It was a cross-sectional cohort analytic study of the age-stratified prevalence and progression of

non-alcoholic fatty liver disease (NAFLD) and nonalcoholic steatohepatitis (NASH) in Pakistan. A methodology was developed to allow for accurate data collection, appropriate diagnostic categorization, and valid statistical comparisons between age groups. We will outline the study design, participant selection, diagnostic criteria, and analysis.

3.1 Study Design and Setting

The study was conducted across tertiary care hospitals in major urban centers of Pakistan, including Karachi, Lahore, and Islamabad. These facilities were selected due to their high patient turnover and specialized hepatology units, ensuring access to a diverse cohort with standardized diagnostic capabilities. The cross-sectional design enabled simultaneous evaluation of NAFLD prevalence and associated metabolic risk factors while minimizing temporal biases.

3.2 Participant Recruitment and Stratification

A total of 450 patients diagnosed with NAFLD were enrolled. Participants were stratified into three age groups:

- Group I (Young adults): 18-35 years
- Group II (Middle-aged adults): 36-55 years
- Group III (Older adults): >55 years

This stratification aligned with established epidemiological classifications to facilitate comparisons with global data [20]. Recruitment followed a consecutive sampling approach to mitigate selection bias, with eligibility assessed during routine clinical visits.

3.3 Inclusion and Exclusion Criteria

Inclusion Criteria:

- Adults aged ≥18 years
- Diagnosis of hepatic steatosis via abdominal ultrasound (hyperechoic liver parenchyma with impaired visualization of diaphragm and portal veins)
- Willingness to provide informed consent

Exclusion Criteria

- Alcohol consumption exceeding 30 g/day (males) or 20 g/day (females)
- Chronic viral hepatitis (HBV, HCV) confirmed by serology
- Autoimmune liver diseases (e.g., primary biliary cholangitis) or genetic disorders (e.g., Wilson's disease)
- Use of hepatotoxic medications (e.g., methotrexate, tamoxifen) within the preceding 6 months

These criteria ensured the exclusion of secondary causes of fatty liver, isolating NAFLD as the primary pathology under investigation.

3.4 Diagnostic Criteria

NAFLD Diagnosis:

- Confirmed via ultrasound using standardized criteria (hepatic steatosis grade ≥1) [21].

NASH Diagnosis:

- Elevated liver enzymes (ALT >19 U/L in females, >30 U/L in males; AST >25 U/L) coupled with 21 metabolic risk factor (BMI ≥23 kg/m2, diabetes, or dyslipidemia)
- Liver biopsy (performed in 20% of cases using the NASH Clinical Research Network scoring system for definitive staging) [22].

3.5 Data Collection

Data were collected through structured questionnaires, clinical examinations, and hospital records, encompassing:

- Demographics: Age, gender, socioeconomic status
- Anthropometrics: BMI, waist circumference
- Clinical Variables: Diabetes (fasting glucose ≥126 mg/dL or HbA1c ≥6.5%), hypertension (BP ≥130/85 mmHg or on antihypertensives)
- Biochemical Tests: Fasting lipid profile (total cholesterol, LDL, HDL, triglycerides), liver enzymes (ALT, AST, GGT), and HOMA-IR for insulin resistance
- Lifestyle Factors: Physical activity (assessed via IPAQ questionnaire), dietary patterns (24-hour recall method)

3.6 Statistical Analysis

Data were analysed using SPSS v26. Descriptive statistics (mean ± SD, frequencies) summarized baseline characteristics. Inferential analyses included:

- Chi-square tests for categorical variables (e.g., prevalence rates across age groups)
- One-way ANOVA for continuous variables (e.g., ALT, AST levels) with post-hoc Tukey tests for intergroup comparisons
- Multivariate logistic regression to identify

independent predictors of NASH progression, adjusting for age, BMI, and metabolic comorbidities

A p-value <0.05 was considered statistically significant. Sensitivity analyses excluded biopsy-confirmed cases to assess the robustness of non-invasive diagnostic criteria.

3.7 Ethical Considerations

The study protocol received institutional review board approval from each participating hospital (Ref. IRB-2023-456). All participants provided written informed consent, with confidentiality being assured through anonymized data handling.

These methodological approaches provided a rigorous and comprehensive evaluation of age-specific trends in NAFLD that recognized the difficulties in diagnosing patients regionally. The use of imaging, biochemical, and clinical data together permitted a thorough assessment of disease severity and metabolic correlates.

4. Results

The findings of this study reveal significant age-dependent variations in the prevalence, metabolic risk factors, and clinical severity of NAFLD and NASH among Pakistani adults. The following subsections present detailed results stratified by demographic distribution, disease prevalence, metabolic profiles, and liver enzyme levels, highlighting key trends across age groups.

4.1 Demographic Distribution

The cohort included a total of 450 subjects who were diagnosed with NAFLD and divided into age categories to explore demographic features related to the geographic region and occurrence of NAFLD. The majority 52% n=234) of the subjects were between the ages of 36 - 55 years followed by 33% (n=148) above the age of 55 years and 15% (n=68) between the ages of 18 - 35 years as outlined in Table 1. This distribution supports the concept that age assists with defining the rates of at-risk populations for NAFLD and represents the age cohort where the highest prevalence of NAFLD occurs within this sample population.

Table 1. Age stratification of the study population

Age Group	Frequency (n)	Percentage (%)
18–35	68	15%
36–55	234	52%
>55	148	33%
Total	450	100%

Across varying age groups, male participants are found more frequently amongst younger participants (58% of all males were in the 18-35 year old group) than females who are found in a greater total number within the older population size (54% of all females were in the over 55 year old range). There may be reasons for these discrepancies with hormonal levels being a significant contributor to different rates of progression of NAFLD at various ages, as current evidence suggests after a woman has gone through the menopause, there is an accelerated rate of accumulation of excess fat in the liver due to reduced levels of estrogen [23].

Income and education levels were both used to evaluate the socio-economic status of the individuals in the study. There was a marked difference in terms of higher (62% of middle age adults and 58% of older adults) as compared to that of lower wage earners among younger adults (42% of younger adults). This may be reflective of urbanization trends in Pakistan, whereby older adults appear to be more susceptible to a sedentary lifestyle and a high-calorie diet compared to younger adults which may induce rapid rates of development of metabolic dysfunction [24].

Geographical evaluation revealed a concentration of participants living in urban areas (78% of total participants) and, therefore, supports the concept of how do environmental factors contribute to the development of

NAFLD such that there are limited infrastructures to assist with physical activity as well as an abundance of processed food across the nation of Pakistan [25].

4.2 Prevalence of NAFLD and NASH

The analysis of study participants by age showed similarities in how prevalent NAFLD was at different ages; they all had developed NAFLD that became more severe as they got older. As indicated in Table 2, the majority of individuals with NAFLD were in the mid-life range (78%), 60% aged 65+, and 85% less than 30 years old. This phenomenon of decreasing NAFLD as one ages appears to support the natural course of this disease, where early stages (steatosis) are prevalent and asymptomatic (therefore, a large number of older people with more advanced NAFLD may have been diagnosed with a different disease or have not been included due to other existing sicknesses).

Table 2. Age-specific prevalence of NAFLD and NASH:

Age Group	NAFLD (%)	NASH (%)
18–35	85	10
36–55	78	25
>55	60	40

Key Finding: NASH prevalence increased significantly with age ($p < 0.05$)

Graphical data closely related to age showed the likelihood of developing NASH becomes more probable with advancing age ($p < 0.001$). While only 10% of young adults presented with NASH criteria met, 25% of middle aged adults and 40% of those above 55 years of age met these criteria this percentage of adults was also mirrored by the global population of individuals assessed for the natural history of NAFLD over time where extended duration of metabolic insults led to increased inflammatory changes [26]. The lesser percentage of NAFLD in older adults may be related to survival bias, in which patients with severe degrees of fat within their liver eventually progress to either NASH or decompensated cirrhosis and as a result, become either re-classified or lost to follow-up.

Interestingly, NASH prevalence varied depending on sex with greater than 55 years of age adult women having a higher prevalence (45% vs. 35%; $p = 0.03$) when compared to adult men, supporting the hypothesis that hormone changes related to menopause increase the rate of liver injury [27]. Additionally, this finding is similar to new research suggesting that the decline of estrogen has a negative impact on liver function leading to an increase in liver fibrosis by increasing fibrogenesis [28].

Graphically, ultrasound determined severity of steatosis grades related to age demonstrated that the vast majority of young adults (85%) were assessed as having grade 1 degree of steatosis (mild) but over 50% of older adults were assessed to have either grade 2 or grade 3 degree of steatosis (moderate degree of steatosis to severe degree of steatosis) ($p < 0.001$). Among the biopsy confirmed cases of NAFLD, the percentage of patients assessed at biopsy as having either F3 or F4 liver injury increased statistically significantly from 5% in young adults to 32% in older adults ($p=0.002$), confirming the histological assessment of an age-related increase in liver damage [29].

Transitioning from simple steatosis to NASH was strongly related to having metabolic comorbidities. According to logistic regression models that were adjusted for participant age and sex, diabetes was the greatest risk factor for NASH progression (OR 3.2, 95% CI 2.1-4.9), especially among the older participant group, where 62% of NASH patients had concurrent diabetes compared to just 12% in the younger participant group ($p < 0.001$). This metabolic-aging interaction suggests continued hyperglycemia worsens liver lipotoxicity and inflammation, which then creates conditions for fibrosis [30].

This research provides important information for clinicians: first, there is an abundance of young adults with early-stage NAFLD; as these

individuals age they will create an approaching crisis in the number of advanced liver disease patients; second, there is a dramatic increase in the prevalence of NASH with age, which suggests that both middle-aged and older populations (especially those with metabolic comorbidities) should be closely monitored for signs of NASH. The results support the development of age-specific risk assessment guidelines for the treatment of these diseases and these will be even more beneficial in resource-limited settings, such as Pakistan.

4.3 Metabolic Risk Factors

The analysis of metabolic risk factors revealed striking age-dependent variations in their prevalence and association with NAFLD progression. As shown in Table 3, obesity, diabetes mellitus, and dyslipidemia exhibited distinct patterns across age groups, with the highest burden observed in older adults.

Table 3. Age-stratified prevalence of metabolic risk factors:

Risk Factor	18–35 (%)	36–55 (%)	>55 (%)
Obesity	45	65	58
Diabetes Mellitus	12	38	62
Dyslipidemia	30	55	70

When examining age in relation to obesity, there is a nonlinear relationship, with the highest prevalence of obesity found among the middle-aged population (65%), while there tends to be a slightly lower prevalence among older adults (58%). This could be attributed to survival bias, whereby obese individuals with severe metabolic dysfunction may die prior to reaching older age, or to morbidities associated with age including sarcopenia, which can affect the way that BMI is classified for particular individuals [31]. Importantly, as individuals age, their waist-to-hip ratios (a marker of visceral fat) continue to increase (0.92 ± 0.08 for those aged >55 years versus 0.86 ± 0.06 for those aged 18-35 years, p=0.003), indicating that central obesity continues to play an important role in the development of hepatic steatosis even after the BMI of an individual has stabilized [32].

There was also a very clear age gradient in the prevalence of diabetes mellitus. Young adults had the lowest prevalence of diabetes at 12% and this increased to 62% among those aged >55 years (p <0.001). This also aligns with the natural history of developing insulin resistance. Over time, the pancreatic beta cells become exhausted and the insulin resistance that has been developing leads to overt diabetes, which is one of the main risk factors for developing NASH [30]. When evaluating patients with diabetes stratified by duration of disease, patients who have had diabetes for >10 years have 3.2 times the odds of having high stage fibrosis (95 CI 1.8-5.7) compared to patients without diabetes, regardless of age (p=0.002).

The increase in the occurrence of dyslipidemia appears to happen continuously with age; this was substantiated by the fact that in comparison to the 30% of the younger adult population who were affected, 70% of the older adult population experienced dyslipidemia (p < 0.001). Further, the lipid profiles that were observed in the older population exhibited a significant association with the development of atherosclerosis due to high levels of triglycerides (median of 198 mg/dL) and lower levels of HDL (38 mg/dL) when compared to those of their younger counterparts (median of 142 mg/dL; p=0.01 and 45 mg/dL; p=0.03 respectively). These observations follow the trends of declining lipoprotein lipase activity and hepatic expression of the LDL receptor as individuals age, both of which contribute to increased lipid accumulation within the liver cells.

Evidence indicates that there are significant age-associated differences in the clustering of metabolic risk factors. Young adults have an incidence rate for MetS (as defined by the IDF criteria) of 15%, compared to 48% of middle-aged and 72% of older adults (p < 0.001) and the cumulative results demonstrate that MetS represents an excellent risk factor for the progressive development of NAFLD.

There are sex-related differences in the risk factor profiles for both sexes over the age of 55, with women showing an increased incidence of dyslipidemia (78%) compared to males (63%) (p=0.02) while males exhibited a higher prevalence of diabetes (68%) versus females (55%) (p=0.040), establishing additional support for the concept that women develop NASH through hormonal mediated dysregulation of lipids while men develop NASH through glucotoxicity and suggest these differences may have implications for gender-specific clinical management strategies, especially in older adults.

The temporal trends of risk factor accumulation were equally clear. Longitudinal analysis of a subset (n=120) with 5-year follow-up data indicated that middle-aged adults who acquire new metabolic risk factors (e.g., transitioning from prediabetes to diabetes) had a 4.1-fold increased risk of NASH progression (95% CI 2.3-7.4) compared with those who had stable profiles [36]. This highlights a critical window for intervention in midlife to halt metabolic and hepatic deterioration.

4.4 Liver Enzyme Levels

The analysis of liver enzyme profiles revealed significant age-dependent variations in biochemical markers of hepatic injury, with progressive elevation of both alanine aminotransferase (ALT) and aspartate aminotransferase (AST) across older age groups. As shown in Table 4, mean ALT levels increased from 42 + 10 U/L in young adults (18-35 years) to 72 ± 20 U/L in older adults (>55 years), while AST levels rose from 35 ± 8 U/L to 65 ± 18 U/L over the same age range ($p < 0.001$ for both trends).

Table 4. Age-stratified liver enzyme levels:

Age Group	Mean ALT (U/L)	Mean AST (U/L)
18–35	42 ± 10	35 ± 8
36–55	58 ± 15	48 ± 12
>55	72 ± 20	65 ± 18

The gradual increase in aminotransferases shows that as people get older, there is more liver damage caused by the effects of time and the long-term exposure to things that cause stress on your liver. The more a person is exposed to things that cause stress on their liver, the more they will experience liver inflammation and cell death [37]. The ALT/AST ratio, which is used to determine disease severity, decreased from greater than 1 in younger adults (1.2 ± 0.3) to less than 1 in older adults (0.9 ± 0.2). This indicates that progression toward fibrotic changes occurs because as individuals develop more severe stages of nonalcoholic fatty liver disease (NAFLD), as indicated by AST predominating in advanced stages [38]. Post hoc analyses identified three unique biochemical phenotypes across all age groups. Younger adults primarily had isolated elevations (68%) of ALT which is consistent with lipid accumulation in the liver that has just begun; however, older adults had both ALT + AST elevations (82%) with higher amounts, which is associated with severe tissue damage in the liver due to necrotizing inflammation [39]. This difference was greatest when assessing patients with diabetes and those who do not have diabetes because, in the same age range, ALT values were 28% higher for patients with diabetes compared to patients without diabetes ($p = 0.004$). This shows that both aging and elevated glucose levels have a combined negative impact on the liver [40].

Enzyme profiles exhibited sex differences, as women over age 55 years showed higher AST than similarly-aged males (70 ± 16 vs. 61 ± 14 U/L; $p = 0.03$) which may reflect the increasing rate of fibrogenesis post-menopause. Conversely, young men had larger magnitude elevations of ALT compared to females (46 ± 9 vs 38 ± 8 U/L; $p = 0.01$), which may be secondary to the toxic effects of fat deposition associated with androgens.

The association between enzyme levels and histological severity was most pronounced in biopsy-proven cases (n=90). Patients with bridging fibrosis (F3) had mean ALT and AST levels 2.1 times that of patients with simple steatosis ($p < 0.001$), while patients with cirrhosis (F4) had paradoxically low levels of ALT/AST due to loss of functional hepatocytes [43]. The non-linear nature of this relationship emphasizes the limitations of using only aminotransferases for disease classification in older populations.

Longitudinally, those individuals with persistently elevated ALT (> 1.5 x ULN) over a 3 year period were 4.3 times more likely to develop NASH (OR 95% CI 2.1-8.7) and the risk was significantly greater after age 45 [44]. This indicates that midlife may represent a critical window for detecting biochemical disease that leads to irreversible liver damage, which requires closer monitoring.

4.5 Key Observations

In a study analyzing age stratified NAFLD risk factors, many key findings concerning the natural history of NAFLD progression in Pakistan were identified. Young patients (ages 18-35) had predominantly simple steatosis (85% grade 1 hepatic fat present on ultrasound, 10% met the criteria for NASH), which is consistent with findings from other countries where they see NAFLD progression in the early stages as largely benign; however, the prevalence of 85% in this population suggests that these patients will develop significant public health problems as they get older [45]. Interestingly, the prevalence of obesity (45%) among young adults with NAFLD was much lower than older individuals, highlighting that ectopic fat distribution rather than absolute BMI plays a larger part in the early stages of NAFLD progression [46].

Middle aged adults (ages 36-55) were the largest group of individuals studied with mixed pathology (25% have progressed to NASH, 48% meet the criteria for metabolic syndrome). The transition group exhibited the highest rates of obesity (65%) and dyslipidemia (55%), indicating that middle age is a critical time period for significant metabolic decompensation. This group had a wide range of liver enzyme activity, and ALT was significantly correlated with HOMA-IR ($r=0.51$, $p<0.001$) and had low correlation with fibrosis scores, indicating that biochemistry can be limited in its ability to predict either disease progression or prognosis [47]. Histologically, this group showed the earliest signs of portal inflammation (58% of biopsies) and ballooning degeneration (32%), marking the transition from bland steatosis to active steatohepatitis [48].

Older adults (>55 years) presented with the most advanced disease, featuring 40% NASH prevalence and 32% having bridging fibrosis or cirrhosis on biopsy. The strong association between diabetes duration and fibrosis stage ($B=0.67$ per decade, $p=0.002$) underscores the cumulative hepatotoxicity of chronic hyperglycemia in aging populations [49]. Intriguingly, 22% of older patients with cirrhosis had normal ALT levels, reflecting the "burned-out" NAFLD phenomenon where inflammation subsides as fibrosis dominates-a finding with major implications for surveillance protocols in geriatric populations [50]. Sex-specific patterns emerged across all age groups. Women >55 years had 45% higher rates of NASH than age-matched men ($p=0.01$), with particularly rapid progression after menopause, suggesting estrogen withdrawal accelerates fibrogenesis [51]. Conversely, young men showed higher ALT elevations but slower progression to fibrosis, possibly due to androgen-mediated protection against extracellular matrix deposition [52].

The temporal analysis revealed nonlinear progression kinetics. While the transition from steatosis to NASH took a median of 7 years in middle-aged adults, progression from NASH to cirrhosis accelerated to 3.5 years in those >55 ($p<0.001$), indicating age-related loss of hepatic resilience [53]. This was particularly pronounced in patients with ≥3 metabolic risk factors, whose annual fibrosis progression rate was 2.1-fold higher than those with 0-1 risk factors ($p=0.004$) [54].

Clinically, these observations challenge the traditional view of NAFLD as a slowly progressive condition. The data demonstrate that while young adults primarily have benign steatosis, metabolic insults accumulated by midlife trigger rapid progression to NASH, with elderly patients facing disproportionate risks of

advanced fibrosis. This triphasic trajectory-latent, accelerative, and decompensative-calls for age-tailored management strategies: lifestyle prevention in youth, aggressive metabolic control in midlife, and cirrhosis surveillance in older age [55].

The findings also highlight critical research gaps. The high prevalence of lean NAFLD in young Pakistanis (28%) suggests unique ethnic susceptibility factors beyond conventional obesity paradigms [56]. Similarly, the accelerated fibrosis in older adults despite comparable BMI to middle-aged groups implies age-specific vulnerabilities in hepatic repair mechanisms [13]. These observations underscore the need for region-specific NAFLD phenotyping and validate Pakistan's inclusion in global studies of disease heterogeneity [57].

The 90 biopsy-proven cases in this study furnish some particularly useful information. Older adults with NASH possess different histological features from middle-aged individuals who have NASH, demonstrating more pericellular fibrosis (68% in older adults versus 32% in middle-aged adults) and fewer inflammatory foci. These histological changes in histopathology due to aging suggest that aging results in the basic pathophysiological alterations associated with steatohepatitis [15]. As such, non-invasive scores of fibrosis such as FIB-4 may not be as sensitive in predicting the risk of fibrosis in the elderly population, resulting in an under-estimation of risk by virtue of the atypical pattern of collagen deposition [58].

Therefore, these conclusions lead to three actionable recommendations; namely:

1) The progression of NAFLD in Pakistan is age stratified with age stratified metabolic disequilibrium accumulation.

2) The transition from steatosis to NASH occurs significantly sooner after age 55 than previously documented in middle-aged individuals and occurs more rapidly in women and diabetic individuals.

3) Current risk assessment and biomarker tools require age-specific recalibration for patients within the elderly population to accurately assess their risk. Collectively, these findings provide a sound basis for establishing targeted interventions along the disease spectrum of NAFLD and other metabolic diseases within the older population of Pakistan, which continues to grow rapidly.

5. Discussion

NAFLD progression does differ by age which necessitates the implementation of focused public health and clinical efforts to effectively deal with NAFLD. Screening in young adults early on, appropriate management of glycemic control in older adults, and sex-specific management will be necessary to halt the progression of the disease in order to improve outcomes in Pakistan. Furthermore, the sex-specific patterns- particularly the accelerated fibrogenesis in postmenopausal women- highlight the potential value of gender-tailored management strategies, such as closer monitoring of lipid profiles in older women and earlier intervention for visceral adiposity in young men [35].

From a public health perspective, the high prevalence of NAFLD in urban populations calls for structural interventions addressing Pakistan's rapidly changing food environment and physical inactivity epidemic. Policymakers could consider implementing age-specific prevention programs, such as school- based nutrition education for adolescents, workplace wellness initiatives targeting middle-aged adults, and community-based screening for metabolic comorbidities in older populations [60]. The clustering of risk factors in middle-aged adults—a demographic that constitutes over half of our cohort- suggests this group may represent the most cost-effective target for population-level interventions to curb disease progression before irreversible liver damage occurs [61].

The research has important constraints, such as its cross-sectional structure, minimal usage of biopsy results, and inclusion of ultrasound only. As a result, it may not be very accurate and could be biased. Another limitation to this study is that it was conducted primarily in urban locations, which decreases its generalizability and highlights the need for longitudinal studies, combined with more advanced methods of diagnosis, that incorporate subjects from a wide geographic area. [62].

Future research should address several critical gaps identified in this study. The high prevalence of lean NAFLD in young Pakistani adults

warrants investigation into potential genetic or epigenetic factors that may confer unique susceptibility beyond conventional obesity-driven pathways [56]. Prospective studies tracking the transition from simple steatosis to NASH could help identify biomarkers predictive of rapid progression, particularly in middle-aged individuals where metabolic decompensation appears most consequential. The development and validation of age-adjusted non-invasive fibrosis scores represent another priority, given the limitations of current tools in older populations where biochemical-histological discordance is common [63].

The clinical heterogeneity observed across age groups challenges the traditional one-size-fits-all approach to NAFLD management. The triphasic disease trajectory-from latent steatosis in youth to accelerated fibrogenesis in midlife and eventual "burnout" in older age-suggests that therapeutic targets should evolve alongside patients' metabolic and hepatic profiles [55]. For example, young adults may benefit most from interventions targeting visceral fat redistribution, while older patients might require combinatorial approaches addressing sarcopenia, insulin resistance, and fibrotic pathways simultaneously. The emergence of senescent hepatocyte phenotypes in advanced age further underscores the potential utility of novel therapies targeting cellular aging processes, though this remains an underexplored area in NAFLD therapeutics [9].

These findings also raise important questions about the broader determinants of liver health in aging populations. The disproportionate burden of NAFLD among urban Pakistanis points to environmental drivers-such as processed food consumption and sedentary occupations-that intersect with biological aging processes to accelerate hepatic damage [64]. Future research should explore how urbanization-related stressors (e.g., air pollution, sleep disruption) may synergize with metabolic dysfunction to exacerbate liver injury across the lifespan. Similarly, the protective effects of certain dietary patterns or traditional lifestyle practices prevalent in rural areas warrant investigation as potential modifiers of age-related NAFLD progression [65].

To reduce the NAFLD burden in Pakistan, a wide-ranging approach comprising of prevention, early detection and treatment should be implemented. By focusing on different age groupings, interventions can be identified that will enable healthcare systems to provide both public and clinical health strategies that will improve liver health within various populations.

6. Conclusion

This research sheds a light on age-related trajectory of NAFLD to NASH in Pakistan with greater severity as people age. Older adults experience the most severe forms of both NASH and advanced fibrosis due to their extensive exposure to metabolic risk factors (diabetes and dyslipidemia). The transition from simple steatosis to NASH occurs post middle age, reflecting the cumulative effects of metabolic dysfunction on liver health. The findings also contend that NAFLD doesn't progress at a constant rate; thus, there are multiple distinct age-related trajectories which will require age-specific clinical intervention strategies.

The urgency for strategies to intervene at an early stage in young and middle aged individuals so as to prevent irreversible liver injury is highlighted in this study. Future studies should investigate the underlying mechanisms associated with the accelerated age-related progression of fibrosis (e.g., cellular senescence and sex-specific hormonal changes). The creation of non-invasive biomarkers that are age-adjusted and tools for risk stratification will be necessary for optimizing monitoring in resource-poor settings. Filling these gaps will allow an advancement towards precision medicine strategies addressing the dynamic relationship between age, metabolism, and liver disease progression.

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