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ABSTRACT
Objective The second National Diabetes Survey of Pakistan (second NDSP) was planned to ascertain the updated prevalence of diabetes, pre-diabetes and associated risk factors at the national and provincial levels.

Research design and methods The survey was conducted by using multistage clustering technique in all four provinces of Pakistan from February 2016 to August 2017. An estimated sample size of 10,800 was calculated using probability sampling and multistage stratified sampling technique.Twenty-seven clusters were selected out of total 213 clusters from all four provinces (strata) of Pakistan. A total of 46 subclusters were selected by using the ‘Rule of thumb’. Out of 12,486 targeted individuals, 10,834 study subjects finally participated in the study (87% response rate). Seventeen trained teams collected data using the structured questionnaire. The clinical and anthropometric measurements included height, weight, blood pressure, waist circumference and waist-to-hip ratio while the blood tests included Oral Glucose Tolerance Test (OGTT), haemoglobin A1c and fasting lipid profiles. WHO criteria were used for the diagnosis of diabetes and pre-diabetes.

Results Overall weighted prevalence of diabetes was 26.3%, of which 19.2% had known diabetes, and 7.1% were newly diagnosed people with diabetes. Prevalence of diabetes in urban and rural areas was 28.3% and 25.3%, respectively. Prevalence of pre-diabetes was 14.4% (15.5% in urban areas and 13.9% in rural areas). Age greater than or equal to 43 years, family history of diabetes, hypertension, obesity and dyslipidaemia were significant associated risk factors for diabetes.

Conclusion The findings of the 2nd NDSP imply that diabetes has reached epidemic proportion and urgently need national strategies for early diagnosis and effective management as well as cost-effective diabetes primary prevention programme in Pakistan.

INTRODUCTION
The epidemic of diabetes is one of the most alarming public health issues of the 21st century, especially for lower middle-income countries.1 It was predicted that from 2010 to 2030, there will be a 67% increase in the prevalence of diabetes in these countries.2 Diabetes-related complications are the major cause of premature deaths in the world,3 4 with a death occurring in every 6 s due to the consequences of diabetes.5 6 In 2015, approximately five million diabetes-related deaths were reported in low-income and middle-income countries.7

The first National Diabetes Survey of Pakistan (1st NDSP) was done by the Diabetic Association of Pakistan (DAP) in collaboration with WHO between 1994 and 1998.7–11 Overall, glycaemic dysregulation (diabetes plus pre-diabetes) was reported to be 22.04% and 17.15% in urban and rural areas, respectively, by the 1st NDSP.7 The prevalence of diabetes as assessed by fasting plasma glucose had doubled from 7.2% in 2002 to 14.2% in 2009 in the rural areas of Baluchistan.12 Similarly, the prevalence of impaired fasting glucose (IFG) has risen from 6.5% to 11%.12 Furthermore, studies conducted in school-age children had also warned of a rise in childhood obesity.13 Pakistan Health Research Council (PHRC) conducted a
questionnaire-based survey using STEPS, a ‘WHO STEPwise approach to surveillance’, in the provinces of Punjab and Sindh. This survey along with other studies reported the prevalence of diabetes between 13.1% and 26.9%. Hence the epidemic of diabetes was predicted nearly a decade ago. Most of these studies had been smaller scale, focusing on specific towns or villages and because of diversified ethnic groups within Pakistani population, could not accurately reflect the prevalence of diabetes in Pakistan. Therefore, there was a need for a repeat survey, that is, second NDSP.

This led to a joint collaboration of Ministry of National Health Services, Regulation and Coordination, PHRC, DAP and Baqai Institute of Diabetology and Endocrinology, Baqai Medical University, to conduct the second NDSP. The aim of this survey is to ascertain prevalence of diabetes, pre-diabetes and associated risk factors at the national and provincial levels. The results are expected to explore ethnic and geographical variation in diabetes and pre-diabetes phenotypes.

RESEARCH DESIGN AND METHODS

The second NDSP was conducted from February 2016 to August 2017 in all four provinces of Pakistan, that is, Punjab, Sindh, Khyber Pakhtunkhwa and Baluchistan. Pakistani nationals aged 20 years or more were included in the survey, whereas pregnant women and those not residents of the selected households were excluded. An estimated sample size of 10,800 was calculated using probability sampling and multistage stratified sampling techniques. Sample size was calculated based on an expected prevalence of 18% (based on previous surveys), level of significance 97%, margin of error 1% with a design effect of 2, considering complex multistage design.

Stratification of population was done on the basis of urban and rural domains for all four provinces as defined in the latest available census. Each province was considered as a stratum and the districts (geographical subdivision of provinces legally described by government) considered as clusters were selected from each stratum. Tehsils or towns (further bifurcation of districts legally described by government) considered as subclusters were selected from each identified cluster for the survey.

Clusters and subclusters were randomly selected using probability proportional to size technique, and number of clusters were selected from each province using the ‘rule of thumb’ number of clusters (k)=(sample size of stratum/2)^0.5. Twenty-seven clusters were selected out of a total 215 clusters from all over Pakistan. A total of 46 subclusters (21 from urban and 25 from rural) were selected (figure 1).

Seventeen teaching hospitals and/or diabetes centres participated in the second NDSP. The training sessions of these 17 teams were conducted from February 2016 to July 2016. The teams were trained to identify households, to fill the questionnaire, to take anthropometric and clinical measurements and to collect blood samples. The questionnaire was adopted from the WHO Questionnaire used in the 1st NDSP. Each team was led by a physician as provincial coordinator of that cluster and each team comprised laboratory technicians, paramedical staff and survey officers.

Door-to-door assessment was done following systematic sampling technique. The first household in the lane was selected randomly and afterwards every 10th house was identified. In case residents of the identified household were not present or if they refused to participate, the next consecutive household was taken. Teams marked the houses and informed the adult residents. The selected household members were requested to come after an overnight fast (at least 8 hours) to the camp on the specific day. Two hundred and fourteen camps were conducted to recruit the required number of study subjects. Each participant was expected to stay within the screening facility for at least 2 hours, that is, for the post 75 g anhydrous glucose load. Meanwhile, the anthropometric and clinical data were collected by the trained paramedic staff under the supervision of the provincial coordinator.

Behavioural and social assessment

For each participant, we administered a detailed behavioural and social assessment with the help of a predesigned questionnaire. All information was gathered through one-to-one interviews by a trained survey officer. Tobacco users were defined as self-reported smoking and chewing of tobacco products daily irrespective of quantity consumed and duration.

Anthropometric and clinical assessment

Height, bodyweight, waist circumference and blood pressure were measured using standardised techniques. Weight was taken to the nearest of 0.1 kg with participants in light clothes and without shoes. Height was recorded to the nearest of 0.1 cm, while subjects standing in erect posture vertically touching the occiput, back, hip and heels on the wall. Body mass index (BMI) was calculated as weight in kilograms divided by height in metre squared (kg/m²). Waist circumference and hip circumference were measured between the centre point of the lower margin of the ribs and iliac crest the mid-point. Blood pressure was measured with mercury sphygmomanometer. Individuals were requested to take 10 min rest in a sitting position before measurement of blood pressure to reduce variation. Mean of two readings was taken.

BIOCHEMICAL ASSESSMENT

Blood samples were collected by using sterilised disposable vacutainer tubes containing sodium fluoride (for glucose), EDTA K2 (for haemoglobin A1c; HbA1c) and gel (for lipids). Within 1 hour of blood collection, the samples were centrifuged, separated and sent to the laboratory. Plasma glucose was measured using the glucose oxidase peroxidase method, total cholesterol...
by cholesterol oxidase phenol 4-aminoantipyrine peroxidase (CHOD-PAP) method, triglycerides by glycerol phosphate oxidase-p-aminophenazine (GPO-PAP) method, high-density lipoprotein cholesterol (HDL-C) by homogeneous enzymatic calorimetric method, low-density lipoprotein cholesterol (LDL-C) by CHOD-PAP method and HbA1c by high-performance liquid chromatography method. Plasma glucose was performed both fasting and 2 hours post 75g glucose load (2-hour PGL) at the designated laboratory close to the survey site with the specified methodology. Samples for HbA1c and lipid profile were transported as per the standardised protocol to PHRC, Jinnah Postgraduate Medical Centre, Karachi, for analysis. Equipment with same specifications was used throughout the study for standardisation and as a measure of quality assurance. Calibration and controls were run as per the specified guidelines. Interassay and intra-assay coefficients of variability for these biochemical parameters were within the acceptable ranges.

**Diagnostic criterion**

WHO definition was used to diagnose diabetes and pre-diabetes (intermediate hyperglycaemia). Results of plasma glucose testing were categorised as follows: isolated IFG was defined as fasting plasma glucose level between 110 mg/dL and 125 mg/dL with 2-hour PGL ≤140 mg/dL. Isolated impaired glucose tolerance was defined as fasting glucose level <110 mg/dL and 2-hour PGL between 141 mg/dL and 199 mg/dL. Newly diagnosed diabetes was defined as fasting plasma glucose level ≥126 mg/dL or 2-hour PGL ≥200 mg/dL or both. Known diabetes was considered if the subject had been diagnosed as diabetic by a physician.
For the diagnosis of diabetes using HbA1c as diagnostic tool, the American Diabetes Association (ADA) standards of care were used. HbA1c ≥6.5% (48 mmol/mol) was diagnosed as diabetic while HbA1c between 5.7% and 6.4% (39 and 46 mmol/mol) was considered as pre-diabetes.24 People were considered hypertensive if they were already diagnosed by a physician or if they were taking any antihypertensive medication or if the systolic blood pressure was ≥140 mm Hg and/or diastolic blood pressure ≥90 mm Hg.8 9 As per WHO Asia Pacific Guidelines, obesity was defined as a BMI of 25 kg/m² or higher for both males and females with or without abdominal obesity.25 Central obesity was defined as waist circumference ≥90 cm and ≥80 cm in males and females, respectively.8 9

Using the Adult Treatment Panel III guidelines, dyslipidaemia was classified as one or more of the following conditions in fasting state: serum cholesterol >200 mg/dL, serum LDL-C >130 mg/dL, serum HDL-C <40 mg/dL and <50 mg/dL for male and female, respectively, and serum triglycerides >150 (mg/dL).26 People were also considered as dyslipidaemic if they were taking any lipid-lowering medications.

**PATIENT AND PUBLIC INVOLVEMENT**

It is a community-based epidemiological survey conducted to ascertain the prevalence of people with type 2 diabetes in Pakistan. The results of this survey will help the National and International stakeholders to take appropriate measures for prevention of diabetes at all levels. With the informed consent, 10 834 individuals from all four provinces of Pakistan were involved in the survey. The participation of the study subjects was limited to the collection of study data approved by the ethical review committee while the whole survey was performed by the survey team members. The tests involved in the survey were conducted free of cost and the results were communicated to study participants as printed medical reports through local NDSP team members. Complimentary medical consultation was provided in case of any abnormal finding. Subjects with newly diagnosed diabetes and impaired glucose tolerance were referred to the nearest centre for registration and treatment.

**STATISTICAL ANALYSIS**

Data analysis was conducted on SPSS V.20. Descriptive analysis included the estimation of mean values and SDs for continuous variables. Categorical variables and prevalence values were presented in the form of frequency and percentage. For calculating the prevalence of diabetes, the following formula was used27:

\[
\hat{p} = \left(\frac{\text{Sample size of district}}{\text{Sample size of province}}\right) \times \left(\frac{\text{Number of diabetics from that district}}{\text{Number of individuals sampled from that district}}\right)
\]

| Table 1 Baseline characteristics of the study participants, by province urban and rural areas of Pakistan |
|-------------------------------------------------|------------------|---------------------|------------------------|------------------|
| **Number of participants** | 6221 | 2531 | 1544 | 538 |
| **Age (years)** | 43.5±14.1 | 45.5±14.2 | 40.3±12.9 | 48.4±12.81 | 43.8±14.0 |
| **Gender** | 2457 (39.5) | 1192 (47.1) | 835 (54.1) | 257 (47.8) | 4756 (43.9) |
| **Female** | 3764 (60.5) | 1339 (52.9) | 709 (45.9) | 281 (52.2) | 6078 (56.1) |
| **Primary education or more** | 2675 (49.6) | 1353 (61.3) | 759 (55.6) | 278 (54.9) | 5065 (53.5) |
| **Tobacco addiction** | 614 (11.2) | 493 (22.2) | 117 (8.9) | 152 (29.1) | 1376 (14.5) |
| **Positive family history of diabetes** | 1509 (27.8) | 760 (37) | 240 (22.2) | 236 (43.9) | 2745 (30.2) |
| **Body mass index (kg/m²)** | 27.5±6.1 | 26.5±5.6 | 27.2±6.1 | 26.7±5.0 | 27.2±6.0 |
| **Waist circumference (cm)** | 27.0 (23.3–31.2) | 25.9 (22.6–29.6) | 26.4 (23.4–30.1) | 25.6 (23.5–29.2) | 26.6 (23.1–30.5) |
| **Blood pressure (mm Hg)** | | | | | |
| <140/90 | 2482 (45%) | 958 (44.9%) | 922 (65.5%) | 187 (34.9%) | 4549 (47.4%) |
| ≥140/90 | 3032 (55%) | 1178 (55.1%) | 486 (34.5%) | 349 (65.1%) | 5045 (52.6%) |

Data are presented as mean±SD or n (%).

*Median (IQR).*
Associated risk factors of diabetes, pre-diabetes, hyper-tension, obesity and dyslipidaemia were investigated using multivariable logistic regression.

Univariate logistic regression was carried out to select potential predictors \( (p<0.25) \). Multivariate logistic regression analyses were undertaken to estimate the
independent effect of predictors on the prevalence of diabetes and pre-diabetes. Models were built and compared by stepwise forward selection method and likelihood ratio test.

Multicollinearity for variables was checked using variance inflation factor (VIF) with a cut-off point mean VIF >10. Goodness of fit for the final fitted model was checked using the Hosmer and Lemeshow test. Association between predictors and occurrence of diabetes and pre-diabetes was summarised using adjusted OR and statistical significances were tested at \( p<0.05 \). Final model equation was written as:

\[
g_0(t, X_1, X_2, ..., X_k) = g_0(t) \exp \left( \sum_{i=1}^{k} \beta_i X_i \right)
\]

For all estimates, the study population was weighted to the latest available demographic information at Pakistan Bureau of Statistics.18

RESULTS

In this survey, 12486 individuals were approached, out of which 10834 individuals finally participated in the study (87% response rate). Basic characteristics of the study population are presented in **Table 1**. A total of 10834 subjects were screened for diabetes; 43.9% were males and 56.1% were females. More than half of all the participants (53.5%) had at least primary level education. Around one-third (30%) of the population had positive family history of diabetes, and 14.5% were tobacco users. Mean BMI of participants was almost similar in all provinces of Pakistan with an overall mean of 27.23±6.0 kg/m² (mean±SD).

According to OGTT criteria, overall age-adjusted weighted prevalence of diabetes was 26.3%, of which 19.2% had known diabetes and 7.1% were newly diagnosed people with diabetes. Prevalence of diabetes in urban and rural areas was 28.3% and 25.3%, respectively. Highest prevalence of diabetes was observed in Sindh followed by Punjab. Prevalence of pre-diabetes was 14.4%, urban and rural distribution was 15.5% and 13.9%, respectively. Overall glycaemic dysregulation (diabetes, plus pre-diabetes) was 43.8% and 39.2% in urban and rural areas, respectively. Prevalence of pre-diabetes and
newly diagnosed diabetes was higher in Baluchistan compared with other provinces (table 2). According to the ADA standards of care, on the basis of HbA1c criteria, prevalence of diabetes and pre-diabetes was 30.1% and 5.9%, respectively. Urban women showed significantly higher prevalence of diabetes than rural women above the age of 40 years while in men this trend was seen in the age group of 60 years and above (p<0.05). On the other hand, urban men in the age group 30–39 years showed significantly (p<0.05) lower prevalence of diabetes than rural men (figure 2).

Rural men showed significantly higher prevalence of pre-diabetes than urban men for the age group of 40–49 years while for women significant difference was seen in urban compared with rural population for the age group of 30–39 years (figure 3).

Table 3 shows the multivariable logistic regression for identifying the associated risk factors for diabetes, pre-diabetes, hypertension, obesity and dyslipidaemia. Significant association was found between diabetes, hypertension, obesity and dyslipidaemia (p≤0.05). Table 3 also shows the associated risk factors for diabetes, pre-diabetes, hypertension, obesity and dyslipidaemia. Age (≥43 years), family history of diabetes, hypertension, obesity and dyslipidaemia were significant risk factors for diabetes (p≤0.0001). Similar were the risk factors for pre-diabetes including education with the exception of obesity (p≤0.05). Significant association was found between diabetes, hypertension, obesity and dyslipidaemia (p≤0.05).

We estimated the overall age-adjusted weighted prevalence of diabetes and pre-diabetes as 26.3% and 14.4%, respectively. Overall, glycemic dysregulation, that is, diabetes plus and pre-diabetes was 43.8% and 39.3% in urban and rural areas, respectively. This suggests doubling of glycemic dysregulation compared with the 1st NDSP done during 1994–1998.7

Our study has several strengths. To our knowledge, this is the largest survey on diabetes in Pakistan to include all the four provinces and both urban and rural populations. Proper epidemiological methods with multistage stratified sampling technique were used. Oral GTT and HbA1c were done for the diagnosis of diabetes for a sample of over 10,000 people. Another strength is that the whole study was completed within 1 year with the help of 17 teams across the country. There are also some limitations in our study. According to WHO, ideally blood tests shall be repeated for the diagnosis of diabetes, but we were not able to conduct the repeat specimen collection because of resource constraints. We also excluded individuals less than 20 years of age as the survey mainly was designed for the diagnosis of type 2 diabetes. Furthermore, we would not be able to differentiate between type 1 and type 2 diabetes on the basis of our survey. This is because diagnostic approaches for type 1 and type 2 diabetes are different.

We estimated the overall average age-adjusted prevalence of diabetes as 26.3% and 14.4%, respectively. Overall, glycemic dysregulation, that is, diabetes plus and pre-diabetes was 43.8% and 39.3% in urban and rural areas, respectively. This suggests doubling of glycemic dysregulation compared with the 1st NDSP done during 1994–1998. Our study has several strengths. To our knowledge, this is the largest survey on diabetes in Pakistan to include all the four provinces and both urban and rural populations. Proper epidemiological methods with multistage stratified sampling technique were used. Oral GTT and HbA1c were done for the diagnosis of diabetes for a sample of over 10,000 people. Another strength is that the whole study was completed within 1 year with the help of 17 teams across the country. There are also some limitations in our study. According to WHO, ideally blood tests shall be repeated for the diagnosis of diabetes, but we were not able to conduct the repeat specimen collection because of resource constraints. We also excluded individuals less than 20 years of age as the survey mainly was designed for the diagnosis of type 2 diabetes. Furthermore, we would not be able to differentiate between type 1 and type 2 diabetes on the basis of our survey. This is because diagnostic approaches for type 1 and type 2 diabetes are different.
Various regional surveys have shown higher prevalence of diabetes among Indo-Asians compared with other ethnic groups which support our findings. The prevalence of diabetes in Malaysia was 22.9% but among Indians residing in Malaysia, its prevalence was 37.9%. In addition, studies from Bangladesh and Turkey had also shown similar results. A recent study from China showed that 11.6% adults (218 years) had diabetes and almost 50% of the population had pre-diabetes. Similarly, a recent study from 15 states of India showed that 7.3% had diabetes and 10.3% of the population had pre-diabetes. Also worth noting is the prevalence of diabetes in the UK which had doubled from 2.39% in 2000 to 5.32% in 2013. Moreover, the prevalence of pre-diabetes had increased from 11.6% in 2003 to 35.3% in 2011.

In our study, it was noted that in three out of four provinces, the prevalence of diabetes was higher than the prevalence of pre-diabetes. The age and gender-weighted prevalence also suggests that in early years pre-diabetes is higher but diabetes prevalence rises steeply after the age of 30. The possible explanation is the rapid transition to diabetes from pre-diabetes in these provinces. Whereas, in the province of Baluchistan, diabetes to pre-diabetes ratio is almost 1:2, suggesting that a large number of individuals are at risk of developing type 2 diabetes.

Prevalence of diabetes on the basis of HbA1c was slightly more than on the basis of OGTT. However, prevalence of pre-diabetes was much lower comparatively, that is, 5.9% by HbA1c criteria compared with 14.4% OGTT. This pattern has also been found in other studies. Considering limitations of resources and HbA1c variation with anaemia and haemoglobinopathies, we would still take OGTT as a gold standard diagnostic tool in an epidemiological setting.

One of the unanswered questions from the study was much lower prevalence of diabetes and pre-diabetes in the province of Khyber Pakhtunkhwa. On the other hand, diabetes and pre-diabetes were much higher in the province of Baluchistan. A number of smaller studies had already warned of higher prevalence of risk factors leading to diabetes in Baluchistan in the last 20 years, but still this high proportion needs to be evaluated. Further researches are required to study genetic and environmental influences among various ethnicities.

Also addressed in this survey were parameters like hypertension, obesity and dyslipidaemia in Pakistan. These numbers are also correspondingly high and strongly suggestive of the rise of risk factors leading to diabetes. Hypertension nationally was surveyed by the Pakistan Health and Research Council (previously called Pakistan Medical Research Council) in 1998, and the prevalence of hypertension was reported as 33%. In our survey, hypertension in the community is 52.6%. Similar are the results for overweight and obesity. For overweight and obesity, we have chosen the WHO cut-offs, and the prevalence is 62.1% and 47.5%, respectively, while from Asian cut-offs, these figures are 76.2% and 62.1%.

This study has tremendous future implications. Diabetes has now become a major public health challenge in Pakistan. If appropriate actions are not taken, the burden of disability and deaths from diabetes will be enormous. The existing infrastructure of healthcare services for managing diabetes and its complications is suboptimal. Poverty both as a cause and consequence of diabetes and its complications is a major threat to health, and economic and social development.

It requires multiple stakeholders including policy-makers to integrate and execute their actions to save millions of people from premature morbidity and mortality. Urgent strategies need to be developed for nationwide network of diabetes care and management. Also, primary prevention ought to be addressed at all levels. More importantly, healthy lifestyle changes must be educated and encouraged at school level. Maternal and child health must be given top priority to prevent transgenerational obesity and diabetes.

CONCLUSION
The findings of the second NDSP imply that diabetes has reached epidemic proportion and urgently needs national strategies for early diagnosis and effective management, as well as cost-effective diabetes primary prevention programme in Pakistan.

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Contributors AB: concept, design, interpretation of data, wrote and approved the final submitted version. AF: concept, design, literature search, designing quality assurance measures, research data, wrote and approved the final submitted version. NDSP: involved in the quality control, edited and approved the final submitted version. ASS: concept, design and involved in the quality control. NDSP members were responsible for the supervision of the survey, concept, design, involved in the quality control and data management in their respective areas. All members approved the final submitted version. AB and AF are the guarantors and undertake the full responsibility for the contents of the article submitted for publication.

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Patient consent Obtained.

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REFERENCES


The previous version of this manuscript contains incorrect versions of figures 2 and 3 in the published article. Figures 2 and 3 should appear as:

Figure 2: Age-stratified prevalence of diabetes among men and women with urban and rural distribution

![Figure 2](image1)

Figure 3: Age-stratified prevalence of pre-diabetes among men and women with urban and rural distribution

![Figure 3](image2)

The following related text should read as:

Urban women showed significantly higher prevalence of diabetes than rural women above the age of 40 years while in men this trend was seen in the age group of 60 years and above (p<0.05) (figure 2).
Relating to figure 3, the following text should read as:

Urban men showed significantly higher prevalence of pre-diabetes than rural men for the age group of 60 years and above while for women significant difference was seen in urban compared with rural population for the age groups of 40-49 and 60 years and above (figure 3).

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