STUDY OF IMPAIRED FASTING GLUCOSE IN PATIENTS WITH ONE OR MORE RISK FACTORS FOR DIABETES MELLITUS

Submitted By

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Under the guidance of

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2011

DECLARATION BY THE CANDIDATE

I hereby declare that this dissertation entitled **"STUDY OF IMPAIRED FASTING GLUCOSE IN PATIENTS WITH ONE OR MORE RISK FACTORS FOR DIABETES MELLITUS "** is a bonafide and genuine research work carried out by me under the guidance of **DR.S.S.DEVARMANI** $_{\text{M.D.}}$ Professor Dept Of Medicine.

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LIST OF ABBREVIATIONS

AICAR	-	Aminoimidazole carboxamide ribonucleotide.
BMI	-	Body Mass Index.
CAD	-	Coronary Artery Disease
CVD	-	Cardiovascular Disease.
DM	-	Diabetes mellitus.
GDM	-	Gestational Diabetes Mellitus.
GLUT 4	-	Glucose tansporter 4.
FFA	-	Free fatty acids.
FATP	-	Fatty acid transport protein.
FABP	-	Fatty acid binding protein.
FPI	-	Fasting plasma insulin
GIP	-	Glucose dependent Insulinotrophic peptide.
GLP	-	Glucagon like peptide-1.
HDL	-	High Density Lipoprotein.
HTN	-	Hypertension.
HSL	-	Hormone sensitive lipase.
IGF-1	-	Insulin like growth factor - 1.
IFG	-	Impaired Fasting Glucose.
IGT	-	Impaired Glucose Tolerance.
IRR	-	Insulin receptor related peptide.
IRS	-	Insulin receptor substrate.
LDL	-	Low Density Lipoprotein.
METS	-	Metabolic syndrome

NGT	-	Normal Glucose Tolerance.
PPAR-γ	-	Peroxisome proliferator-activator receptor-γ.
SD	-	Standard deviation.
TNF- α	-	Tumour necrosis factor alpha.
USFDA	-	United States Food And Drug Administration
VLDL	-	Very Low Density Lipoprotein.
WHO	-	World Health Organisation.

ABSTRACT

Background and objective: Impaired fasting glucose is a forerunner of diabetes mellitus. In individuals with risk factors for diabetes mellitus,it is important to screen for Impaired fasting glucose to prevent future occurrence of diabetes mellitus and cardiovascular mortality and morbidity. Hence we aimed to study Impaired fasting glucose in patients with one or more risk factors for diabetes mellitus.

Material and methods: The study was carried out on patients coming to B.L.D.E.U's Shri B.M. Patil Medical College Hospital and Research Center, Bijapur. Detailed history was taken with special regard to the risk factors for diabetes mellitus, and general and systemic examination was carried out and they were also subjected to certain investigations to determine risk factors like Dyslipidemia, Ischemic heart disease etc. The investigations done were

a. Fasting blood sugar and Post prandial blood sugar

b. Lipid profile.

C. ECG

Results: In this study 8.46% individuals had impaired fasting glucose,the results correlating with other studies.

Comparing the physical activity, out of 8.46% individuals with Impaired fasting glucose,8.41% were moderate workers,14.29% were in heavy workers.

Comparing individuals with history of hypertensive heart disease and ischemic heart disease,13.79% individuals had history of hypertensive heart disease and ischemic heart disease and 4.17% did not have any history.

Out of 130 individuals,105 individuals were overweight and 14 individuals were obese,11 individuals were with normal BMI.

Also in this study out of 8.46% individuals, 54.55% individuals had positive family history of first degree relatives with diabetes and 45.45% did not have positive family history.

This study shows that individuals with one or more risk factors for diabetes are at increased risk of impaired fasting glucose and future risk of diabetes.

Conclusion: In our study we concluded that Individuals with risk factors for diabetes are associated with having impaired fasting glucose also known as prediabetes.

Hence, identifying individuals with impaired fasting glucose will lead to preventing future occurrence of diabetes mellitus and other cardiovascular risk factors,by life style modification and if needed pharmacological therapy.

Keywords: Risk factors for diabetes mellitus, Impaired fasting glucose.

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INTRODUCTION

Diabetes mellitus is the single most important metabolic disease recognised worldwide as one of leading cause of death and disability. The problem has reached pandemic population.

Type 2 diabetes is the commonest form of disease constituting all most 90% of the diabetic population. Prevalence of diabetes was estimated to be 4% in 1995 and expected to be 5.4% by year 2030[1]and its incidence being highest in developing countries than developed countries.

Today India is the leading country in the world with largest number of diabetic subjects as compared with any given country, currently estimated diabetic population of India is 31.7 million individuals and its number is expected to reach,79.4 million by year 2030[1],and WHO has already declared India as global capital of diabetes.

In light of the dramatic epidemic of type 2 diabetes ,there is great interest in identifying and implementing interventions to prevent or delay its onset.Early detection and prompt treatment may reduce the burden of disease and its complications. There is also good evidence that screening tests can detect diabetes during early asymtomatic phase. Targeted screening among individuals with one or more high risk factors is more cost effective than universal screening at all ages. The American Diabetes Association has proposed the major risk factors for type 2 diabetes mellitus and has created guidelines for early screening in individuals with one or more risk factors, so as to reduce the burden of disease and its complications.

AIMS AND OBJECTIVES

To study Impaired fasting glucose in patients with one or more risk factors for diabetes mellitus.

REVIEW OF LITERATURE

Diabetes mellitus in all forms is one of the most important chronic disease of the developed and developing world. The number of individuals developing type 2 diabetes mellitus is increasing worldwide and is imposing growing burden on health services. Diabetes mellitus has both acute and chronic complications which include diabetic retinopathy, nephropathy, neuropathy, peripheral vascular diseases. Individuals with diabetes are more prone for cardiovascular diseases like ischemic heart disease and cerebrovascular accident.So to reduce the burden of disease we tend to screen individuals with risk factors for diabetes.

Screening for type 2 diabetes in asymtomatic individual has been under consideration for many years[2].There is also general evidence that universal screening is not recommendable.The largest study of this kind tested over 60,000 people in cleveland,USA and concluded that indiscriminate mass screening for diabetes was of questionable value and such programme should be directed to individuals with high risk group[3].

Hence screening was done for individuals with high risk factors for diabetes mellitus as recommended by American Diabetes Association.

The United states preventive task force recommended in 2004[4],screening for Impaired fasting glucose in patients with risk factors of hypertension or hyprelipidemias and when these patients were specifically targeted for screening, the number needed to screen to prevent a cardiovascular event was much lower than screening general population.

In year 2003 American Diabetes Association[5] adviced study of Impaired fasting glucose among persons with high risk factors for diabetes mellitus

Also study conducted by Simmi dube,Vk sharma,tn dubey,ankit mehta,d gour[6] in 1008 population divided into two groups based on presence or absence of risk factors, showed that Impaired fasting glucose was prevalent in 4.61% in first group and 1.41 in% second group.Also their study concluded that Impaired fasting glucose in asymtomatic individuals warrants to prevent or delay their progression to diabetes and its related complications.

In 2007 study of David et al[7] showed that Impaired fasting glucose was seen in people with risk factors for diabetes mellitus and the progression to diabetes was more common in subjects having Impaired fasting glucose with one or more risk factors.

Also study by Jacqeline james in 2005[8],advised screening of high risk population.According to them certain genetic,environmental and biochemical variablities have been identified which increases risk of developing type 2 diabetes mellitus.

Study by Denice et al in 2005[8] together with canadian task force showed that there is strong evidence to recommend adults with Impaired fasting glucose together with high risk factors like hypertension,hyperlipidemia and BMI > 25.

Harris mi et al studied[9] that in the third national health and nutrition examination survey in 1988-1994,using American Diabetes Association criteria,showed that prevalence of Impaired fasting glucose was 6.95%.

There is a steady increase in prevalence of type 2 diabetes mellitus and Impaired fasting glucose ,both globally and in India.The prevalence of Impaired fasting glucose in age group between 40-60 years will be around 75-80% and number of Impaired fasting

glucose individuals will be 308 and 418 million in 2007 and 2025 respectively[10].In India the expected growth in diabetes mellitus prevalence is from 6 to 74% in 2025 and it is mainly due to increase in life expectancy and increase in urbanization(31-43%). The socioeconomic growth will increase the prevalence of Impaired fasting glucose even in rural areas.Also the prevalence of Impaired fasting glucose is high in India and south east asian countries,which is expected to increase from 85.6 million to 132 million by 2025.[11] The concept of prediabetes has been given to either Impaired fasting glucose or Impaired glucose tolerance either isolated or in combination.The transition from Impaired fasting glucose to frank diabetes takes several years.Current estimates are that upto 70% of people with Impaired fasting glucose eventually develop diabetes[12].

Prediabetes –The terminology:

The British diabetes association recommended that "PREDIABETES" should only be used retrospectively to describe life of a diabetic person before diagnosis of diabetes mellitus.WHO replaced term Prediabetes with statistical risk classes.It was only later in 1997 that ADA and WHO introduced the term Impaired fasting glucose[13].Prediabetes in its current concept,owes its rebirth to Mr.Tommy. G.Thompson, U.S secretary of state of health in 2002.It was used to describe people with either Impaired fasting glucose or Impaired glucose tolerance in an attempt to warn Americans of higher future risk of development of diabetes.

Also it is observed that at time of diagnosis,good number of new diabetic patients already had long term complications[14].These observations suggest that they were having diabetes from a much early period and remained aymptomatic and so undiagnosed and long term

complications may start earlier in a subject who do not satisfy the criteria for diabetes,but are having blood sugar higher than the normal range.This range of blood sugar in between 100-125mg/dl, called Impaired fasting glucose and 2 hour post 75 gram glucose value between 140-199mg/dl known as Impaired glucose tolerance.These two together are known as "Prediabetes."The patient should have been fasting for atleast 8 -14 hours(water is allowed).Worldwide the number of diabetic people are estimated to be 314 million and is projected to be 418 million in 2025[15] .Diabetes prevention programme has observed that 7.9% of Prediabetes population have retinopathy[16].

Even with the present diagnostic criteria of diagnosis of diabetes mellitus,long term complications are found at diagnosis means that they remain undiagnosed before or long term complications may start before the blood sugar remains at the Prediabetes range.So the Impaired fasting state of sugar is not benign and some degree of increased microvascular and macrovascular complications of diabetes has been described and this constitutes risks for diabetes[17].

The risk of progression to diabetes is more if subject has both Impaired fasting glucose and Impaired glucose tolerance.The risk of coronary artery disease maintains a linear correlation with glycemia well below the present diagnostic criteria for diabetes.

Significance of Impaired fasting glucose in Indians:

One of the earliest study in India[18] in 1986-87,in an urban population in a township in south India, showed 5% prevalence of diabetes and 14 patients had Impaired glucose tolerance. A family history of diabetes was present in 16 of 34 diabetic subjects and 9 of 15 with Impaired Glucose tolerance.India has the highest number of IGT.

Prevalence of IGT and IFG are also high in India and South east asia in general[19,20],which is expected to increase from 85.6 million(2003) to 132 million by 2025.

DECODE data show that of all European people with IFG defined by a fasting plasma glucose of 6.1–6.9mmol/l alone, 64.8% had isolated IFG, 28.6% had IGT and 6.6% have diabetes. Similarly, DECODE data shows that Asian people with IFG defined by a fasting plasma glucose of 6.1–6.9mmol/l alone, 45.9% had isolated IFG, 35.2% had IGT,18.9% had diabetes.The prevalence of IFG varies between populations and across different age groups within population. Overall prevalence rates in the order of 5% or more are common. IFG is typically more common in men than in women. The DECODE study showed an increase in prevalence of isolated IFG from 5.2% in 30–39yr old men to 10.1% in 50–59yr old men and then a decrease to 3.2% in 80–89yr old men, whereas in women prevalence increased from 2.6% in 30–39yr old to 5.9% in 70–79yr old.[21]

In Asian populations prevalence of isolated IFG generally increases with age, except in the Indian population where prevalence does not change much with age.[22]

Data from Mauritius[23] indicate that in people with IFG at baseline, 40% reverted to normal, 15% remained as IFG, 20% changed to IGT and 25% developed diabetes over a 11 yr follow up period. Two studies which assessed the reproducibility of IFG with retesting within 6 weeks showed that the proportion of people classified as IFG on the first test and on retesting was 64% and 51% respectively with the majority being reclassified as normal and less than 10% as having diabetes on repeat testing[24].

The annualized relative risk of people with isolated IFG progressing to diabetes compared with people with normal glucose tolerance showed a 4.7 fold increase in the three studies included in the review by the McMaster group[21].IFG was associated with increased risk of adverse outcomes with a relative risk ranging from 1.19–1.28 for non-fatal myocardial infarction, non-fatal cardiovascular disease, cardiovascular mortality and all-cause mortality[24].

Also it is seen that in overweight latino adolescents with family history of type 2 diabetes mellitus, IFG was associated with impaired beta cell function and therefore may identify children likely to be at risk of progression to type 2 diabetes mellitus.The actual risk of IFG to type 2 diabetes mellitus remained to be determined by prospective longitudinal studies.

Natural history and pathogenesis of Impaired fasting glucose

The pathogenesis of Impaired fasting glucose is linked to relative insulin deficiency and tissue insulin resistance.The pathophysiology of Impaired fasting glucose is different. Hepatic insulin resistance with normal muscle insulin sensitivity is the basis of Impaired fasting glucose,but in isolated Impaired glucose tolerance ,moderate to severe muscle insulin resistance is the predominanat feature.Insulin resistance at both these sites co exists when there is a combination of IFG and IGT.Impaired fasting glucose has a defect in early phase insulin secretion.

Determinants and risk correlates of pre-diabetes:

1.Age more than 45 years.

2.Family history of diabetes.

3.Physical inactivity.

4.Obesity and body fat patterning.

5.Insulin resistance and the metabolic syndrome.

6.Gestational diabetes.

7.Maternal undernutrition,low birth weight and childhood catch up obesity.

8.Genetic predisposition.

9.Race and ethnicity:

a.American Indian.

b.African Indian.

c.Hispanic.

d.Asian/pacific islander.

e.Asian Indian.

Risk factors for developing diabetes:

1.Body fat distribution:Individuals with central obesity(android type) and high BMI are high likely to develop diabetes. Waist hip ratio gives a good indicator of central obesity and has been found to correlate well with development of type 2 diabetes mellitus.Insulin resistance is probably the most important link between obesity and diabetes.Hyperinsulinemia is correlated well with BMI and central obesity.

2.Fat intake:There is evidence that dietary fat is the most likely component to have etilogical relationship to development of diabetes.In rats fed with high fat diets,insulin resistance tends to develop and it is possible that a similar phenomenon may occur in humans especially, with a high intake of saturated fats.

3.Lack of physical activity: Higher levels of plasma insulin are associated with lower level of physical activity and physical training can decrease insulin resistance. In one study the prevalence of diabetes was three times higher in individuals in light physical activity compared to those involved in heavy work.

4.Smoking:Smoking has been found to be an independent risk factor for development of type 2 diabetes mellitus. Data from Osaka health survey has shown that risk for development of type 2 diabetes in current smokers is higher than in nonsmokers and that the number of cigarettes smoked and the number of pack years are positively related to the development of type 2 diabetes in a dose dependent manner.

5.Role of stress:During periods of stress,the body responds by secreting excess of catecholamine,cortisol,growth hormone and glucagon.These hormones increase insulin resistance and over period of time,the increased stress may promote the development of type 2 diabetes.

6.Genetic background:There is a strong genetic element in the development of type 2 diabetes mellitus and a genetic susceptibility probably predisposes individuals with change in their lifestyle to develop diabetes.The same is true for asian Indians like us.An individual's risk for diabetes is doubled if one member of family already has the disease and the risk is quadrupled if there are two family members with diabetes mellitus.

7. Previous Gestational diabetes:Development of gestational diabetes during pregnancy indicates a significantly higher risk of developing diabetes in future.In fact the progression to type 2 diabetes mellitus is 5% per year with almost 50% of these women with gestational diabetes developing diabetes over a 10 year follow up.

Early detection of gestational diabetes mellitus by universal screening and appropriate management can not only prevent perinatal mortality and morbidity,but also prevents birth of either low birth weight baby or over weight baby,both of which are associated with higher risk of development of diabetes mellitus.Exposure of fetus to higher glycemic levels in mother has also been associated with greater risk of development of type 2 diabetes mellitus.Thus measures directed towards early detection and management of GDM can therefore ensure longterm benefit over three generations and contribute to prevention of diabetes.

Metabolic characters of Impaired fasting glucose:

A variety of methods have been utilized to evaluate the contributions of impaired insulin sensitivity and decreased insulin secretion to the genesis of IFG. Insulin secretion is markedly influenced by the route of glucose administration. When glucose is administered via gastrointestinal tract, a much greater stimulation of insulin secretion is observed compared with similar hyperglycemia created with intravenous glucose. The difference in insulin secretion between intravenous versus oral glucose administration is referred to as the incretin effect[25] and is mediated by glucagon like peptide-1(GLP-1) and glucose-dependent insulinotrophic peptide (GIP)[26] . Reduced glucose-stimulated GLP-1 secretion consistently has been observed in type 2 diabetes[27].In contrast to GLP-1,

GIP levels are elevated in type 2 diabetes and resistance to GIP is a characteristic feature of the diabetic state[28]. GLP-1 and GIP secretion in individuals with IGT and IFG has been less well characterized.[29,30.] .Insulin secretion in response to intravenous glucose also differs from oral glucose in its temporal pattern. Following glucose ingestion there is a gradual rise in plasma glucose concentration (reflecting the slow rate of glucose absorption) and this is accompanied by gradual increase in plasma insulin. The abrupt rise in plasma glucose following intravenous glucose causes a rapid and transient increase in plasma insulin concentration (first-phase insulin secretion), which lasts for 10 minutes. This is followed by a slower, sustained rise in plasma insulin (second phase insulin secretion), which persists as long as plasma glucose remains elevated[31].

In IFG patients,following glucose ingestion,plasma glucose rises rapidly at 30 minutes,continues to rise after 60 minutes and progressively declines at 120 minute.Plasma glucose concentration at 30 and 60 min increases to levels greater than in normal glucose tolerance tests.So subjects with isolated IFG have elevated FPG,exaggerated early rise in plasma glucose concentration following ingestion and plasma glucose concentration similar to normal glucose tolerance at 120 minutes.

Insulin resistance in IFG:

IFG individuals are primarily characterized by hepatic insulin resistance with normal muscle insulin sensitivity.In isolated IFG,the hepatic insulin resistance index(basal hepatic glucose production×FPI(Fasting plasma insulin) is markedly increased.Subjects with IFG have impaired first phase insulin secretion,which explains their high risk for conversion to type 2 diabetes mellitus.In IFG, patients have decrease in

first phase insulin secretion in response to intravenous glucose and early phase insulin response to oral glucose.So subjects with IFG start with high fasting plasma glucose due to hepatic insulin resistance but incremental rise in plasma glucose concentration at 30 and 60 minute is slightly greater than in normal glucose tolerance test and at 120 minute,plasma glucose concentration are similar to that of normal glucose tolerance test values.

Basal insulin release is also impaired in IFG.Also in IFG,reduced hepatorenal insulin sensitivity combined with impairment in basal insulin secretion and first phase insulin secretion causing fasting hyperglycemia.Normal or near normal second phase insulin response and muscle insulin sensitivity may prevent IFG individuals from having post prandial hyperglycemia.

Early recognition and its clinical significance and risks:

Impaired fasting glucose is not only an early forerunner of diabetes but also carries high risk of cardiovascular disease.Indians have high insulin resistance on this background in adding together to presence of all other cardiovascular risk factors. IFG occurs for a much younger age in Indians and they are predisposed to get diabetes more or less a decade prior to as compared to rest of high risk population worldwide.

Early recognition is very important in initiating early intervention to stop onset of diabetes related complications.IFG is diagnosed by studying fasting glucose measurement.

ADA recommendations for diagnosis of IFG:

1.Male and female individuals,specially over 45,those with overweight to be screened for IFG.

2.Screening also to be considered for age <45 years if they are overweight and have one or more additional risk factors.

3.If test is positive for IFG,then repeat tests to be performed on subsequently to confirm diagnosis.

4.People with IFG should be monitored with regular retesting over one or two years to monitor for type 2 diabetes mellitus.Individuals with normal screening result can be retested every 3 years.

Overall prediabetes confers a six fold increase in risk of diabetes compared to normal glucose tolerance.In most populations studied,the rate of conversion of IFG to diabetes mellitus are similar.But IGT has high sensitivity but less specificity than IFG in predicting diabetes risk.[32]

In a 11 year follow up study among young adults with IFG in Mauritius,38% developed DM,17% remained unchanged in category,17% developed IGT,and glucose level normalized in 38%.After a protracted follow up, it was seen that 50% people with IGT/IFG developed diabetes mellitus.

Patients with IFG are now referred as Prediabetes.IFG is not a clinical entity,but rather a risk factor for future diabetes mellitus as well as cardiovascular disease.IFG is associated with metabolic syndrome which includes obesity(abdominal or visceral),dyslipidemia with high triglyceride and low HDL cholesterol type and hypertension.Medical nutritional therapy,exercise and pharmacological agents have invariably demonstrated to prevent or delay development of DM.Individuals with IFG may have normal or near normal HbA1C levels.

Cardiovascular system risks in people with IFG:

1.People with IFG have increased risk of cardiovascular disease and cardiovascular and all cause mortality.

2.There is two-three fold increased risk of CVS events which is more marked in younger adults with prediabetes.

3.Also data suggests that people with IGT are at a higher risk compared to people with IFG.

Association with metabolic syndrome:

The metabolic syndrome refers to clustering in individual of CVD risk factors and diabetes susceptibility.People with metabolic syndrome have about two fold increased risk of developing diabetes and cardiovascular disease,compared with those without the syndrome.Most adults who have prediabetes will also have metabolic syndrome.The data demonstrated by Diamantopoulos et al[33]showed that METS and prediabetes have an overlapping pattern.METS appear to have a more pronounced effect on early renal dysfunction and increased inflammatory activation,while prediabetes tends to be associated with early carotid structural changes.

Interventions to prevent Prediabetes:

There are different ways by which natural history of IFG can be altered.The progression to diabetes is a time dependent phenomenon;one possible alteration is to simply reset the clock without changing the rate of deterioration.It is possible that some interventions will lower glycemia initially but do nothing to change the subsequent rate

of rise of glycemia.This mechanism will delay crossing the glycemic threshold that defined diabetes.Prediabetes is a condition that doesnot fall squarely into primary or secondary prevention group and hence is inadequately addressed by interventions in either health promotion or disease management.There is substantial evidence that even at these blood glucose levels,significant risks exist for both microvascular and macrovascular complications.Biuso et al[34] introduced a conceptual framework of care for prediabetes that includes both screening and provision of up to date clinical therapies in conjugation with an evidence based health coaching intervention.In combination these modalities represent the most effective means for delaying or even preventing the onset of diabetes in a prediabetes population. Research studies have found that lifestyle changes can prevent or delay the onset of type 2 diabetes among high risk adults. The three components of lifestyle modification are Diet, Exercise, Behavioural therapy. Several reviews have found that standard lifestyle modification programme conducted in academic medical centre induce a mean weight reduction of approximately 8-10% of initial weight in 16-26 weeks of treatment[35]. These studies included people with IFG and other high risk characteristics for developing diabetes. Lifestyle interventions included diet and moderate intense physical activity such as walking for atleast 2.5 hours in a week In Diabetes Prevention Programme[36] , a large prevention study of people at high risk for diabetes, the development of diabete was reduced by 58% over 3 years.

Principles and Practices of Management:

1.Community awareness , sensitization and education aimed at behavioral modification constitutes the triology underlying principles of management. Early intervention is the

key to successful outcome of lifestyle modification. Nevertheless, with epidemiological and demographic transition,the world now has more fat people than hungry ones, with more than a billion overweight people compared to 800 million who are undernourished.

2. In a meta analysis, lifestyle interventions reduced diabetes by approximately one-half and pharmacological interventions by approximately one third.
3. Prevention of complications rather than prevention of worsening of glycemia should be the goal of treatment-major drawback being cost effectiveness of lifestyle modifications in preventing diabetes.

Management:

Management approach involves a set of measures designed to address the abnormalities and cardiometabolic risk factors.As the Prediabetes progresses, drug therapy directed towards hyperglycemia and the individual coronary artery disease risk factors may be required.Strict control of all known risk factors of CAD and microvascular complications in prediabetes by aggressive management of hypertension,dyslipidemia and glycemia has proven beneficial.Glucose directed therapies alone are not sufficient for prediabetes.

Lifestyle modifications:

1. This is the cornerstone of management of IFG people .It should be reinforced in every visit of patient to the clinic. Lifestyle interventions is the fundamental management approach that can effectively prevent or delay progression of prediabetes to diabetes,as well as reduce both microvascular and macrovascular disease risks.Life style

intervention also improves component of metabolic syndrome(ex: obesity, hypertension, dyslipidemia and hyperglycemia)

2. Emphasis should be given to weight loss in overweight pre-diabetic subjects.Diabetes Prevention Programme findings indicate that 5 to 10% weight loss will be beneficial.Importance should be given to long term maintainence after reducing the body weight.Modest degree of weight loss, results in decreased fat mass,lowers blood pressure,glucose,LDL cholesterol and triglycerides level.These benefits can also translate into longterm outcome,especially when they are maintained.

Nutritional factors:

Nutritional factors leading to over weight and excess body fat,example,high total calorie,high glycemic load,high saturated fat always should be avoided in daily meal plan for prediabetes.

Pharmacology in Prediabetes:

1. Currently there is wide variation in opinion regarding pharmacotherapy in this period of prediabetes.[37,38.].Even after diagnosis of diabetes mellitus,a trial of diet and exercise for 6 weeks to 3 months is given by most of physicians.
2. Unlike diabetes where Metformin is recommended,in prediabetes no pharmacotherapy is approved by authorities like USFDA.Thus any decision to start pharmacotherapy is off label and requires careful judgement regarding risks and benefits of each specific agent in each individual patient.
3. Several trials have shown beneficial effect may be due to some extent to prevent subjects of prediabetes to go to the diabetic state.

Metformin, alphaglucosidase inhibitors and glitazones all have shown promising results[39,40].American Diabetes Association, Australian Diabetes Association,Indian Health Service guidelines for cases of Prediabetes or Metabolic syndrome recommended metformin in high risk individuals where there is clear evidence of glycemic deterioration or progression of underlying disease as evidenced by increase in Hba1c,fasting plasma glucose, or 2 hour post prandial plasma glucose. The use of metformin is supported by its safety, cost effectiveness and long term data in several studies.

4. The ideal pharmacological therapy must demonstrate long term safety,health benefits(reduced incidence of occurrence of diabetes,microvascular and macrovascular complications and mortality.),cost effectiveness and the ability to halt the progression from prediabetes to diabetes.These factors will remain the subject of future research in the prevention of diabetes.

Current treatment recommendations[41]:

Population	Treatment
IFG	Life style modification.
IFG plus any of the following 1.Age > than 60 years. 2.BMI>35 kg/m^2. 3.Family history of diabetes in first degree relatives with diabetes. 4.Elevated triglycerides. 5.Reduced HDL-cholesterol 6.Hypertension. 7.Hbalc > 6.	Life style modification and oral medications.

Novel targets for Pre-diabetes treatment[42]:

1.Apsirin-reduces NF-KB induced islet cell inflammation.

2.Salisylates can prevent progression from Prediabetes to Diabetes.

3.Cholestyramine can reduce glycemia.

4.Weight control by any measures can prevent the progression from Pre-diabetic to diabetic state.

5.Newer agents like HSD-1 inhibitors to prevent cushingoid changes.

6.Statins may be helpful in reduction of vascular outcome in a Prediabetic individual.

Impact of Impaired fasting glucose:

1.Impaired fasting glucose affects multiple systems just like diabetes,but the impact is relatively less severe compared to diabetes.

2.Clinical association of Pre-diabetes:

a.Atherogenic dyslipidemia.

b.Coronary heart disease.

c.Hypertension.

d.Polycystic ovarian syndrome.

e.Non alcoholic fatty liver disease.

f.Sleep disordered breathing.

g.Microangiopathies:Microalbuminuria,Neuropathy,Retinopathy.

h.Cognitive decline.

Coronary heart disease(CHD):

1.Risk of CHD begins to increase in atleast 15 years before onset of hyperglycemia in a diabetic individual. Increased intimo medial thickness has been observed across all categories of increasing glucose tolerance. Clearly the risk of CHD is higher in pre-diabetic individual as compared to normal population. A progressive increase in cardiovascular risk from NGT to pre-diabetes and diabetes was clearly seen in Nurses Health study.

2.Non-alcoholic fatty liver disease:It is astonishing that 1/3rd of Indian population may have non alcoholic fatty liver disease(NAFLD).Hepatic triglyceride content is two fold higher in asian indians as compared to controls. Studies also found that IFG was observed in 23.1 % of cases with NAFLD compared to controls(4.9%) Fasting blood glucose is an independent risk factor for NAFLD.

3.Sleep disordered breathing:Habitual snoring is associated with abnormal fasting glucose and insulin values independent of age and BMI[43].

4.Microangiopathies:Microalbuminuria:It is often seen with insulin resistance and IGT. Micrialbuminuria is suggested as one of the diagnostic criteria for metabolic syndrome.

Subjects with microalbuminuria have increased 2 hour insulin and triglyceride concentration and higher prevalence of hypertension,and decreased level of HDL-C,thus making microalbuminuria a close congener of prediabetes.

Fuel Reserve :Hormone-Fuel Interrelationships: Fed State, Starvation, and Diabetes Mellitus

Basic principles

Humans have a constant requirement for energy but eat only intermittently. To cope with this problem, we usually ingest food in excess of the immediate caloric needs of our vital organs and store the extra calories in the form of hepatic and muscle glycogen, adipose tissue triglyceride, and to a certain extent, tissue protein. In turn, during starvation and in response to various stresses, we break down these fuel reservoirs to provide energy for organ metabolism and function.

The two principal circulating fuels in humans, glucose and free fatty acids (FFAs), are stored intracellularly as glycogen and triglycerides, respectively. The largest reservoir of glycogen (300 to 500 g) is skeletal muscle[44] .However, the principal reservoir of glycogen from which free glucose can be released into the circulation is the liver (Table.1). The major site of triglyceride storage is adipose tissue. Adipose tissue triglyceride is the most efficient form of energy storage in humans. Triglyceride contains 9.5 kcal per g and the average caloric content of an adipocyte, including its cytosol, is approximately 8 kcal per gm[45]. In contrast, glycogen contains 4 kcal / gm. Furthermore, because 3 mL of water is needed to maintain the intracellular osmolality of each gram of glycogen in vivo[46] , in reality glycogen provides only 1 kcal per g. Thus, if the 15 kg of adipose tissue triglyceride in a normal 70-kg man were replaced with an equicaloric quantity of glycogen, the individual would weigh an additional 120 kg.

TABLE 1. Fuel Reservoirs in Humans

Source	gm	kilocal
Liver glycogen	75	300
Muscle glycogen	400	1,600
Blood glucose	20	80
Adipose tissue triglyceride	15,000	141,000
Protein	6,000	24,000
Data are estimates for an overnight-fasted man weighing 70 kg.		

Body protein, although of considerable mass (Table 1), is not, strictly speaking, a fuel reservoir. Protein molecules serve specific roles in maintaining organ structure and function and are less expendable than glycogen or triglycerides. On the other hand, a portion of body protein (e.g., some of the contractile protein of muscle as well as other proteins in liver and muscle) is degraded during starvation and other periods of stress and provides amino acid substrate for gluconeogenesis.

The Brain and Other Vital Organs

The brain has a continuous need for fuel but stores almost no energy as glycogen or fat. Instead, it uses glucose derived from the liver either directly from glycogen or indirectly from other fuel reservoirs through gluconeogenesis. The brain does not use FFAs directly. During prolonged starvation, however, it is able to use energy derived from FFAs after their conversion to ketone bodies. Other vital organs, such as liver, heart,

and skeletal muscle, also have a continuous requirement for fuels (Table 2), but unlike the brain, these organs can utilize fatty acids directly to meet their energy needs .

TABLE 2. Typical Daily Fuel Requirements of Liver, Muscle, and Brain of a Physically Active, Normally Fed Human

Organ	Fuel	~kcal/d
Liver	Amino acids, fat, glucose	280
Muscle	Glucose, fat	880
Brain	Glucose	480

Hormonal Regulators of Fuel Homeostasis

Energy reservoirs in humans are built up and broken down in response to hormonal messages. The principal hormonal messenger is insulin. In the fed state, insulin levels increase, promoting glycogen synthesis in liver and muscle, lipid formation in adipocytes, and amino acid uptake and protein synthesis in most cells. In the postabsorptive state, during starvation and in response to many stresses, decreased insulin levels contribute to glycogen breakdown, lipolysis, hepatic ketogenesis, and decreased synthesis and increased degradation of protein. In the latter situations, a major role of insulin is to act as a restraint on these catabolic events.

Multiple hormones counter the effects of insulin. Glucagon stimulates glycogenolysis, gluconeogenesis, and ketogenesis in the liver [47,48,49,50]. Glucagon also can stimulate lipolysis in adipose tissue, although the physiologic relevance of this latter effect is unclear. Catecholamines have effects similar to those of glucagon on the liver

and are key regulators of lipolysis in adipose tissue and glycogenolysis in muscle and other tissues. In general, the counterinsulin hormones (also called counter regulatory hormones) liberate energy from fuel reservoirs by actions opposite to those of insulin . However, not all of the actions of these counterinsulin hormones are catabolic. For instance, growth hormone, although catabolic in the sense that it stimulates lipolysis in adipose tissue, also has significant anabolic effects and enhances cell growth[51]. Similarly, glucagon has the anabolic property of stimulating amino acid uptake by the liver. The potential roles of leptin and other hormones released by the adipocyte in regulating fuel homeostasis will be discussed in the section on adipose tissue.

Nonhormonal regulation of fuel homeostasis

Although fuel homeostasis has been classically envisaged in the context of its regulation by hormones, changes in the concentrations of the fuels themselves may also play a direct role. Thus, increase in circulating glucose levels have been shown to diminish hepatic gluconeogenesis and glycogenolysis and enhance glycogen synthesis independent of their effects on hormone secretion [52,53]. In addition, FFAs have been shown to stimulate hepatic gluconeogenesis; indeed recent studies suggest that much of the antigluconeogenic action of insulin in humans and other mammals may be secondary to its antilipolytic action on the fat cell [54].

Glucose homeostasis

A principal objective of the interplay between insulin and the counterinsulin hormones in humans is the maintenance of normoglycemia. The concentration of glucose

in the circulation is more closely controlled than that of any other fuel. Thus, plasma glucose levels are maintained between 4 and 7 mm in normal humans despite varying rates of glucose utilization (Table 3), whereas levels of FFAs and ketone bodies may range 10-fold to more than a 100-fold, respectively [55,56]. Prevention of hypoglycemia is important because central nervous system (CNS) function is impaired at low plasma glucose concentrations. Likewise, significant hyperglycemia resulting in glycosuria causes a loss of fuel and may contribute to the complications of diabetes mellitus. Whether plasma glucose levels modestly above or below the "normal" range are undesirable remains to be determined.

TABLE 3. Rates of glucose utilization in the fed and fasting state

	Glucose utilization (g/d)		
Tissue	12-h fast	8-day fast	Marathon run
Brain	120	45	120
Muscle	30	Very low	500

Insulin lowers plasma glucose levels both by stimulating glucose uptake into muscle and adipose tissue and by inhibiting hepatic glycogen breakdown and gluconeogenesis. The different counterinsulin hormones balance these effects of insulin in order to maintain normoglycemia. Thus, glucagon, epinephrine, and norepinephrine are released into the circulation in response to hypoglycemia and during stresses such as exercise, when glucose utilization is altered by other factors [57,58]. In addition to stimulating hepatic glycogenolysis and gluconeogenesis, the catecholamines inhibit

insulin-stimulated glucose utilization in muscle and promote lipolysis in adipose tissue [59], thereby providing tissues with an alternative fuel to glucose. Glucocorticoids also are released into the circulation in increased quantities in response to hypoglycemia and other stresses . Glucocorticoids appear to be necessary for the mobilization of energy stores by catecholamines and glucagon; however, their role may be permissive rather than regulatory [60].

Five phases of fuel homeostasis

Immediately after a carbohydrate or mixed meal has been ingested, the concentrations of insulin, glucose, and glucagon in plasma favor fuel storage. Once absorption of the ingested food is complete, the concentrations of these and other hormones and substrates change, causing a shift from energy storage in fuel reservoirs to energy mobilization. Further alterations in fuel homeostasis occur with more prolonged food deprivation. These changes can be broken down into five phases on the basis of the source and quantity of glucose entering the circulation. Figure 2 illustrates these changes in a hypothetical human who ingests 100 g of glucose and then begins a prolonged fast .

Fed state

During the first few hours after a carbohydrate meal, glucose absorbed from the gastrointestinal tract provides for the metabolic needs of the brain and other organs. The absorbed glucose in excess of these needs is used to rebuild fuel reservoirs in liver, muscle, fat, and presumably in other tissues . In this setting, plasma insulin levels are

high, plasma glucagon levels are low, and glycogen synthesis is stimulated in liver and muscle. Approximately 75 g of carbohydrate is stored as glycogen in liver, and 300 to 500 g is stored in muscle in a human who has fasted overnight.

As noted earlier, the major form of lipid storage in humans is triglyceride and the major site for triglyceride storage is adipose tissue . Smaller amounts of triglyceride are stored in muscle, liver, and other tissues. Triglycerides also are present in the circulation as constituents of lipoproteins. However, the major circulating lipid fuels are the FFAs. After a carbohydrate meal, high concentrations of insulin favor the use of both glucose and lipoprotein triglycerides for triglyceride synthesis in adipose tissue.

In addition to promoting the synthesis of glycogen and triglycerides in the fed state, insulin inhibits the breakdown of these fuel reservoirs[61](i.e., it is anticatabolic). The concentrations of insulin that inhibit lipolysis appear to be lower than those that stimulate glucose transport in muscle. Presumably, it is for this reason that patients with mild type 2 diabetes and glucose intolerance are hyperglycemic in the absence of significant elevations of plasma FFAs or ketone bodies.

Early Starvation

With the decrease in plasma insulin and the increase in plasma glucagon that accompany an overnight fast, fuel homeostasis shifts from energy storage to energy production . At this stage, glucose no longer enters the circulation from the gastrointestinal tract but is derived principally from the breakdown of liver glycogen and, via gluconeogenesis, from lactate, amino acids, and glycerol, a process that takes place predominantly in the liver but that also occurs in the kidney [62] and intestines [63]. In

addition, circulating FFAs, derived from the hydrolysis of adipocyte triglycerides, become a major source of fuel [64]. As will be discussed later, by using FFAs, muscle and liver decrease their oxidation of glucose as a fuel, thereby conserving it for the brain.

Mobilization Of Carbohydrate And Lipid Stores

In the earliest phase of starvation (i.e., the postabsorptive state), hepatic glycogen is a major source of the glucose entering the circulation and remains so for 12 to 24 hours [65,66]. Glucagon seems to be necessary for hepatic glycogenolysis during this period, although an increase in the level of plasma glucagon does not appear to be the primary stimulus [67,68,69]. After an overnight fast, the average rate of glucose utilization by a healthy human is approximately 7 g per hour (Table 3.) . By extrapolation, the 70 to 80 g of glycogen present in the liver can provide glucose to the brain and peripheral tissues for 12 to 16 hours. Two events allow the maintenance of blood glucose levels beyond this time: (a) Muscle and other tissues begin to oxidize lipid-derived fuels in place of glucose, and (b) hepatic gluconeogenesis, which is also stimulated by fatty acids, replaces glycogenolysis as the principal source of glucose entering the circulation . As will be discussed later, glycogen breakdown in muscle does not yield significant quantities of free glucose, and after an overnight fast, gluconeogenesis by the kidney is of minor importance.

Two factors stimulate the breakdown of adipocyte triglyceride during starvation. First, the concentration of circulating insulin diminishes and, consequently, triglyceride synthesis is decreased and lipolysis is enhanced [70]. Second, norepinephrine is released from sympathetic nerve endings and directly stimulates lipolysis by raising levels of

cyclic adenosine monophosphate (cAMP) in adipocytes [70]. Epinephrine, which is secreted from the adrenal medulla, appears to play a lesser role. The principal users of FFAs during the early phases of starvation are skeletal muscle and liver.

Gluconeogenesis

Because the brain is unable to use FFAs as a fuel, it must continue to use glucose during the early phases of starvation. Gluconeogenesis is an important source of the glucose that enters the circulation even after an overnight fast [71] and becomes the major source as hepatic glycogen stores become depleted. Gluconeogenesis is responsible for approximately 35% to 60% of the hepatic glucose output after an overnight fast (12 to 15 hours postabsorptive) and for more than 97% of the output by 60 hours of starvation [72,73]. At 60 hours, glucose production is limited not by the enzymatic capacity of the liver but by the concentration of gluconeogenic substrate in the circulation [74]. During the early phases of starvation, the two principal gluconeogenic precursors are lactate and alanine (Table 4) [75,76,77].

TABLE 4. Gluconeogenic Substrates in Humans Starved for 24 Hours

Substrate	Amount generated (g/d)
Lactate	60
Amino acids except alanine	25
Alanine only	25
Glycerol	14
Pyruvate	5
Total	129

Lactate comprises 50% of the gluconeogenic substrate of liver in a human who has fasted overnight and is the major gluconeogenic substrate throughout starvation . When glucose cannot be metabolized beyond pyruvate in peripheral tissues, much of the pyruvate is reduced to lactate, which is then released into the circulation In red blood cells and renal medulla, this reduction occurs because there are no mitochondria in which pyruvate can be oxidized. In muscle and other tissues, lactate and pyruvate are released during starvation because the activity of pyruvate dehydrogenase, the enzyme that decarboxylates pyruvate to form acetyl coenzyme A (CoA), is decreased. For the most part, lactate generated from glucose in this way is taken up by the liver and reconverted to glucose by the gluconeogenic pathway .This recycling of glucose between liver and peripheral tissues via lactate is referred to as the Cori cycle.

A second major group of gluconeogenic substrates is the amino acids. Skeletal muscle is the principal reservoir of amino acids in humans. During early starvation, however, the gut and liver also appear to be important sources of the amino acids entering

the circulation . A major stimulus to protein catabolism (both decreased synthesis and increased degradation) during starvation is the decrease in plasma insulin concentrations. Glucagon stimulates protein degradation in liver, and glucocorticoids inhibit protein synthesis in muscle and other tissues. Although increases in the plasma levels of these counterinsulin hormones almost certainly play a role in modulating protein catabolism in stressful states (e.g., diabetic ketoacidosis and trauma), their concentrations are not dramatically altered during starvation, and their role here is thought to be limited.

The principal amino acids released into the circulation from muscle are alanine and glutamine. Most of the alanine is taken up directly by the liver, whereas glutamine is metabolized in the gastrointestinal tract, which can use it for gluconeogenesis and to generate alanine, and by the kidney, where it is the principal gluconeogenic substrate as well as a major source of the ammonia used for neutralizing acid in urine. Glutamine and alanine, despite comprising only 15% to 20% of muscle protein, account for 50% of the amino acids released by muscle because these amino acids can be generated from other constituents in muscle as well as from the degradation of protein. Alanine is formed by the transamination of pyruvate by alanine aminotransferase and glutamine by the amidation of glutamate by free ammonia, a reaction catalyzed by glutamine synthetase.

The rate of release of alanine increases markedly during starvation and other states of insulin deficiency. Despite this, the concentration of alanine in plasma is usually diminished in these situations because its uptake by the liver is stimulated to an even greater extent. Since the interorgan relationships of alanine are very much like those of lactate, a “glucose-alanine cycle” similar to the Cori cycle has been proposed .Impaired release of alanine from muscle has been postulated as a contributor to impaired

gluconeogenesis and hypoglycemia in patients with uremia, maple syrup urine disease, and ketotic hypoglycemia of infancy and in starved women during pregnancy .

The other major gluconeogenic substrate is glycerol, which is derived principally from the hydrolysis of adipose tissue triglyceride. In nondiabetic subjects, the rate with which glycerol appears in the circulation correlates with adipose mass and increases during starvation. Glycerol comprises about 10% of total gluconeogenic substrate during early starvation and a much higher percentage during prolonged starvation, when gluconeogenesis from amino acids is markedly diminished.

Prolonged starvation

Ketone bodies and the brain

With the prolongation of starvation, several events occur that limit the need for gluconeogenesis and thereby conserve body protein. The first of these, as already noted, is an increase in the reliance of muscle and other peripheral tissues on lipid-derived fuels: initially FFAs and later both FFAs and the ketone bodies, acetoacetate and β-hydroxybutyrate. The second is a change in the fuels used by the brain. During early starvation, the CNS continues to use glucose as its exclusive fuel. However, as starvation is prolonged, plasma levels of the ketone bodies increase to values even greater than the level of glucose. Under these circumstances, the brain, or at least parts of it, increases its use of these lipid-derived fuels. A third factor could be a decrease in plasma leptin, which by diminishing sympathetic nervous system activity, would diminish the basal metabolic rate.Ketone bodies are produced from acetyl-CoA via the β-oxidation of fatty acids in the liver. This process, termed ketogenesis, is enhanced by glucagon and inhibited by insulin.

In contrast to long-chain FFAs, the ketone bodies are water-soluble and cross the blood-brain barrier via specific carrier proteins. Furthermore, the activity of these carriers is increased in physiologic states associated with sustained hyperketonemia such as diabetic ketoacidosis and starvation. These physiologic adaptations enhance the use of ketone bodies in place of glucose by the brain and diminish the need to degrade proteins for gluconeogenesis. It is because of these adaptations that humans of normal weight are able to survive fasts of up to 60 to 70 days.

Gluconeogenesis And Protein Catabolism

The decreased use of glucose by the brain during prolonged starvation is accompanied by a diminished rate of gluconeogenesis in the liver. The latter appears to be due to decreases in protein catabolism and secondarily to the release of gluconeogenic amino acids (mostly alanine) from muscle .Some studies suggest that these adaptations in protein metabolism are related to the increased use of lipid fuels during prolonged starvation. Whatever the mechanism, as one proceeds from early to prolonged starvation, urinary excretion of nitrogen decreases from 12 g per day to 3 to 4 g per day, indicating a decrease in protein catabolism from 75 g per day to 12 to 20 g per day. The relative contribution of the kidney to gluconeogenesis increases from 5% to 10% after an overnight fast to 50% after 3 to 4 weeks of starvation. However, in absolute amounts, renal production of glucose is still much lower than hepatic production of glucose after 1 to 2 days of fasting. The increase in renal gluconeogenesis during prolonged starvation is linked to an increase in NH_3 (ammonia) generation from glutamine.

Unlike amino acids, the relative importance of glycerol as a gluconeogenic precursor increases during prolonged starvation. This reflects the fact that the release of glycerol from fat is approximately 14 g per day and remains nearly constant during early and late starvation . After several weeks of starvation, gluconeogenesis from glycerol hypothetically provides upwards of half of the glucose oxidized by the brain.

Hormonal controls

The gradual decrease in plasma insulin modulates the orderly breakdown of fuel reservoirs during the early phases of starvation. However, during prolonged starvation, the decreases in protein degradation and in the use of glucose and ketone bodies in muscle occur in the absence of further changes in plasma insulin level. Some studies suggest that a decrease in thyroid hormone activity contributes to these adaptations. Presumably, the low levels of insulin during prolonged starvation are needed for these adaptations to occur. Thus, patients without any insulin (e.g., during diabetic ketoacidosis) have an impaired ability both to limit the breakdown of their fuel reservoirs and to use glucose and ketone bodies in peripheral tissues. The precise connection between the presence of insulin and these adaptations remains to be determined.

Recent studies suggest that another factor that plays a role in the adaptation of fuel homeostasis during starvation is leptin. As will be discussed in more detail in the next section, during periods of calorie deprivation when plasma insulin levels and adipocyte lipid stores are low, the release of leptin from adipose tissue diminishes, leading to an altered release of neuropeptide Y (NPY) and other CNS peptides and secondarily to a decrease in activity in the sympathetic nervous system. Although the

precise interrelation of this chronic regulation of fuel metabolism to that modulated by insulin and counterinsulin hormones is only partially understood, it is highly likely that leptin plays a significant role in the adaptation of mammals to prolonged starvation.

Hormone–fuel interrelations at an organ level

Adipose tissue

The adipocyte in metabolic regulation

While the adipocyte has classically been viewed as a storage depot for metabolic fuel in the form of lipid, it is now clear that the fat cell plays a central role in the endocrine regulation of energy homeostasis .Adipocytes not only respond to numerous hormones to regulate the storage and release of lipids but also secrete hormones (such as leptin,) that act to control energy balance and endocrine function throughout the rest of the body. The adipocyte also secretes a number of other protein factors, including resistin (also known as Fizz3) ,tumor necrosis factor- α (TNF- α), an inflammatory cytokine ; and Acrp30 (or adiponectin and adipoQ), that may regulate insulin sensitivity elsewhere in the body. Although it was initially suggested that resistin increases with increasing adiposity and plays a role in insulin resistance associated with obesity, a number of subsequent studies failed to support this notion, and more research is necessary to determine the physiologic function of resistin. TNF- α mediates elements of insulin resistance and type 2 diabetes syndromes in some mouse models of obesity and diabetes, although the relevance of TNF-α to obesity and insulin resistance in humans remains unclear .In contrast to resistin and TNF-α, Acrp30 appears to mediate insulin sensitization. Acrp30 is an adipocyte-derived complement-related protein that is secreted

as a high-order multimeric complex. Its production by adipocytes is decreased in obesity and other states of insulin resistance, and exogenously increased levels of this protein enhance numerous insulin actions. Although a great deal remains to be learned about Acrp30 (e.g., the identity of the functional proteolytic product, receptor identity), this molecule currently commands a great deal of attention as an insulin sensitizer.

Insulin action in adipocytes

In general, the hormones that regulate energy storage and release in adipocytes are similar to those that regulate these events throughout the body . Insulin promotes the uptake of metabolites such as glucose and lipids and their storage as triglyceride. Insulin mediates its effects in adipose and other tissues by binding a cell-surface insulin receptor, activating the tyrosine kinase in the intracellular portion of the receptor . The activated insulin receptor then recruits and phosphorylates downstream intracellular substrates, such as the insulin-receptor substrate (IRS)-proteins and Shc. These molecules in turn activate two main intracellular signaling pathways: the phosphoinositide (PI) 3–kinase regulated pathway, and the ras-mitogen-activated protein (MAP) kinase pathway. The counterregulatory hormones, such as catecholamines, oppose the effects of insulin and mediate breakdown and release of stored fats. In general, the counterregulatory hormones act via seven-transmembrane receptors coupled to heterotrimeric G proteins to stimulate adenylyl cyclase, increasing intracellular levels of cAMP and activating protein kinase A (PKA).

Insulin acts at several levels to promote energy storage. Insulin increases uptake of glucose from the extracellular space by promoting the movement of the insulin-

responsive glucose transporter (GLUT4) to the cell surface to increase the rate of glucose flux into the cell . Insulin drives the metabolism of glucose to form glycerol 3-phosphate and increases the activity of glycerol phosphate acyltransferase; coupled with the increased FFA uptake also mediated by insulin, the net result is increased triglyceride storage. Insulin increases uptake of FFA by increasing the synthesis and secretion of lipoprotein lipase (LPL) ; LPL degrades triglycerides and phospholipids in adipocyte-bound lipoproteins to FFA. FFAs are then shuttled into the adipocyte by simple diffusion and/or by specialized fatty acid-transport proteins (FATPs) and fatty acid-binding proteins (FABPs) , then coupled to CoA by acyl-CoA synthetase (ACS), and finally esterified with glycerol to form triglycerides. Insulin generally acts to increase the production of these and other proteins involved in lipid storage in adipocytes . Inhibitor studies suggest that most of these effects of insulin require the action of PI 3–kinase but not the ras→MAP kinase pathway.

Nuclear Factors In Adipocyte Function

The lipid storage function of insulin acts in concert with a number of important nuclear factors that have recently been described. Adipocyte differentiation and determination factor-1/steroid response element binding protein (ADD/SREBP) mediates transcription in response to low levels of cholesterol and other lipids in adipocytes as well as in hepatocytes. ADD/SREBP is synthesized as an integral membrane protein that is retained in the endoplasmic reticulum (ER). Low cellular levels of cholesterol and other lipids result in the movement of the membrane-bound ADD/SREBP to the Golgi, where it is proteolytically cleaved and released from the membrane. Dissociation from the

ER/Golgi allows ADD/SREBP to translocate to the nucleus and increase the transcription of a number of genes required for the synthesis of cholesterol, fatty acids, and triglycerides. The peroxisome proliferator-activator receptor-γ (PPAR-γ) is a so-called orphan nuclear receptor that promotes the differentiation of adipocytes and increases the expression of proteins involved in insulin-stimulated lipid storage, including FATPs, FABPs, ACS, and GLUT4. While the endogenous ligand for PPAR-γ remains unknown, the insulin-sensitizing antidiabetic thiazolidinedione compounds act by stimulating PPAR-γ-mediated transcription.

Control Of Hormone-Sensitive Lipase By Counterregulatory Hormones

The hormone-sensitive lipase (HSL)-mediated breakdown of lipids is one of the best-characterized pathways downstream of counterregulatory hormones such as catecholamines. Although other neutral lipid lipases exist, HSL is the only hormone-regulated neutral lipid lipase .HSL is a neutral lipid esterase that mediates the regulated step of triglyceride hydrolysis by removing the first fatty acid moiety from the triglyceride. After generating FFA from triglycerides, FABPs and FATPs transport the FFA through the cytoplasm and out of the cell . During counterregulatory hormone signaling, PKA mediates the serine phosphorylation of HSL; this phosphorylation event does little to alter the assayable activity of the enzyme, however. Instead, serine phosphorylation of HSL mediates the translocation of HSL in complex with a protein known as lipotransin from the cytosol to the lipid droplet. Access to the lipid droplet may be increased by the PKA-mediated phosphorylation of the perilipin protein that coats the droplet and by the subsequent dissociation of perilipin from the droplet . Insulin

decreases the phosphorylation of HSL, probably by decreasing intracellular levels of cAMP via increases in phosphodiesterases, but perhaps also by increasing phosphatase activity toward HSL.

Muscle

Fiber Types

Muscle comprises approximately 40% of body mass in a man of normal weight. It accounts for 20% to 30% of the body's consumption of oxygen at rest and for up to 90% during exercise . Muscle fibers in the rat are classified as slow-twitch red (type 1), fast-twitch red (type 2a), and fast-twitch white (type 2b) according to their contractile characteristics and their capacity for oxidative metabolism .The same fiber types are found in human muscle. In general, the red fibers have a high oxidative capacity and oxidize fatty acids and other fuels in addition to glucose. White fibers have a lesser ability to oxidize fuels and generate a greater portion of their adenosine triphosphate (ATP) from glycolysis. With respect to exercise, white fibers are those recruited principally during brief periods of intense exercise such as sprinting or weight lifting and red fibers are those recruited during endurance-type activities of low-to-moderate intensity such as walking and running .

All of the fiber types in muscle respond to insulin. However, red fibers have a greater number of insulin receptors and GLUT4 glucose transporters than do white fibers. In addition, muscles rich in red fibers have more capillaries per mass , which could enhance diffusion of insulin and glucose from the plasma to the muscle cell . Perhaps for all of these reasons, glucose uptake in red muscle is more sensitive to insulin than is white muscle both *in vivo* and *in vitro* . Physical training, which causes white fibers to

assume some of the characteristics of red fibers, is associated with an increase in their GLUT4 contents.

Fuel Reservoirs

Glycogen

Glycogen in muscle is synthesized from circulating glucose after meals and exercise and is broken down during exercise and starvation. Glycogenolysis in muscle does not result in the release of free glucose into the circulation, since muscle cells are deficient in glucose-6-phosphatase. As a result, the 300 to 500 g of carbohydrate stored as glycogen in muscle is used solely for its own energy needs and for generating lactate and other gluconeogenic substrates for the liver.

The importance of glycogen as a fuel in contracting muscle is underscored by the association of glycogen depletion with the phenomenon of "hitting the wall" in runners . Likewise, patients with McArdle syndrome, a hereditary deficiency of muscle phosphorylase, are unable to maintain high-energy phosphate stores during exercise . During starvation, muscle glycogen in normal individuals diminishes by approximately 33% after 2 to 3 days and then remains constant as muscle switches over more completely to a lipid fuel economy . During early starvation (phases I and II) and exercise, a considerable portion of the lactate, pyruvate (about 0.1 as much as lactate), and alanine released by muscle is presumably derived from the breakdown of glycogen . The regulation of glycogen synthesis and degradation in muscle by insulin is similar to its regulation of adipocyte triglyceride. Insulin stimulates glycogen synthesis by enhancing glucose transport and activating (dephosphorylating) a key regulatory enzyme, glycogen synthase . Likewise, insulin concurrently diminishes the breakdown of glycogen by

inhibiting the conversion (phosphorylation) of phosphorylase b to phosphorylase a . These effects of insulin are thought to be mediated by specific phosphatases that both activate glycogen synthase and eventually inhibit phosphorylase b kinase and possibly by the inhibition of a kinase (glycogen synthase kinase 3) that, when active, phosphorylates and inhibits glycogen synthase . After exercise, glycogen synthesis is also increased , although the responsible mechanism may be different. Catecholamines and exercise per se stimulate the breakdown of muscle glycogen . As in adipose tissue, catecholamines (epinephrine) act by increasing cAMP and secondarily by increasing cAMP-dependent protein kinase, which in turn activates (phosphorylates) phosphorylase b kinase.

Lipids

Red muscle fibers, in particular, store some energy as triglycerides, and triglyceride hydrolysis may provide a significant portion of their fuel needs during exercise. The question of whether the synthesis and breakdown of triglycerides are regulated in muscle by the same mechanisms as in adipose tissue has not been intensively studied. The activities of lipoprotein lipase in muscle and adipose tissue go in opposite directions during feeding and starvation, suggesting differences between the two tissues with respect to their use of circulating triglycerides . A particularly intriguing observation is the strong association of increases in intramuscular triglycerides and insulin resistance.

Protein

The synthesis and degradation of protein in muscle also are regulated by insulin. Insulin promotes protein synthesis in the fed state and probably acts as a brake on protein degradation during starvation. The mechanisms by which insulin acts on protein

metabolism are more complex than those by which it acts on carbohydrate and fat metabolism and have been reviewed elsewhere .

Fed State and Starvation

Following a carbohydrate meal or insulin administration, glucose derived from the circulation is the principal oxidative fuel of muscle . During early starvation, however, it is placed in this role by fatty acids . As noted earlier, this transition to lipid fuels conserves glucose for use by the brain and conserves protein by reducing the need for gluconeogenesis. The precise mechanism by which these events are modulated has not been resolved but is almost certainly related initially to a decrease in circulating levels of insulin to values lower than those in the fed state. Among the changes attributable to the decreased levels of insulin during starvation are (a) diminished glucose transport into muscle; (b) higher plasma levels of FFA; (c) inhibition of pyruvate dehydrogenase in muscle, resulting in a decrease in glucose oxidation and an increase in release of lactate and pyruvate and, secondarily, of alanine; and (d) decreased levels of malonyl-CoA, an inhibitor of carnitine palmitoyltransferase I (CPTI, also referred to as CAT1), which controls the oxidation of fatty acids in muscle and other tissues by regulating the transport of long-chain fatty acyl CoA into the mitochondria . Recent studies have demonstrated that the concentration of malonyl-CoA increases substantially after 1 to 2 hours of refeeding following a fast and that this correlates closely with a decrease in fatty acid oxidation.

Another group of fuels used by muscle during starvation is the ketone bodies. Acetoacetate and β-hydroxybutyrate are oxidized by muscle more or less as a function of

their concentration in plasma. In rats starved for 48 hours and in humans starved for 1 to 2 weeks , the metabolism of these ketone bodies may account for upwards of 70% of the oxygen consumed by muscle. A role for insulin in regulating the utilization of ketone bodies by muscle has been suggested by studies in rats and humans.

Exercise and Adenosine Monophosphate-Activated Protein Kinase

Exercise can increase oxygen consumption by muscle in excess of 10-fold and, depending on its intensity and duration, may increase both fatty acid and glucose oxidation. It has long been appreciated that the increase in glucose oxidation is the result of increases in glucose transport, glycogenolysis, glycolysis, activation of pyruvate dehydrogenase, and changes in intracellular Ca^{2+} and adenine nucleotides. Recent studies have linked the increase in fatty acid oxidation, at least in part, to activation of an AMP-activated protein kinase (AMPK). Studies in humans and experimental animals have shown that the activity of AMPK is increased within seconds or minutes of the onset of exercise (muscle contraction) and that the activated AMPK phosphorylates acetyl-CoA carboxylase, which it inhibits, and malonyl-CoA decarboxylase, which it activates , leading to a decrease in malonyl-CoA. Abundant evidence suggests that this results in an increase in fatty acid oxidation. It is interesting that treatment with a cell-permeable AMPK-activator, AICAR, also activates glucose transport in the muscle cell, and when administered in vivo for several days or longer, increases the expression of the GLUT4, hexokinase 1 and 2, and several mitochondrial enzymes. In other words, it mimics many (although not all) of the effects of exercise, suggesting that activation of AMPK may be an integral component of both the acute- and long-term response of the muscle cell during physical activity. Interestingly, it has recently been shown that the action of the

antidiabetic drug metformin and the adipocyte hormone adiponectin might be mediated by AMPK.

Liver

The liver is the key regulatory site of glucose homeostasis. Blood glucose levels are maintained in a narrow range in great measure because the liver is able to take up glucose in the fed state and to release it in varying amounts into the circulation during starvation, exercise, and other situations in which the ratio of insulin to counterinsulin factors is decreased . Although the liver does not play a key role in determining the rate at which FFAs enter the circulation, it does appear to play a major role in the disposition of FFAs. Thus, the liver can oxidize FFAs for its own energy needs or production of ketone bodies or it can utilize FFAs for the synthesis of triglycerides and phospholipids, which it can export as constituents of very-low-density lipoprotein (VLDL).

Fed State

High levels of circulating insulin and decreased levels of glucagon, such as occur after a carbohydrate meal, stimulate glycogen synthase and inhibit glycogen phosphorylase in the liver. These changes in insulin and glucagon also inhibit hepatic gluconeogenesis . However, gluconeogenesis does not appear to cease immediately but may continue for several hours after the termination of a fast with a meal. This persistence of gluconeogenesis after a meal may allow hepatic glycogen synthesis to continue when glucose absorption by the gut is no longer in excess of the needs of other organs. According to this glucose paradox hypothesis , dietary glucose is metabolized

initially to pyruvate or lactate in peripheral cells; the liver then takes up these gluconeogenic precursors and resynthesizes glucose-6-phosphate, which can be used to synthesize glycogen. Studies in humans suggest that glucose is incorporated into glycogen by this indirect route as well as by the classical direct pathway. Studies using ^{13}C nuclear magnetic resonance (NMR) spectroscopy suggest that glucose conversion to glycogen via the direct pathway predominates immediately after a standard meal .

Starvation

As starvation proceeds through its different phases, the liver releases fuels into the circulation by three distinct processes: glycogenolysis, gluconeogenesis, and ketone body formation. The breakdown of glycogen in the liver is essentially regulated by insulin and counterinsulin hormones in a manner analogous to that in skeletal muscle . A major difference between the two tissues, as previously stated, is that liver can generate free glucose, which is released into the circulation, because of the presence of glucose-6-phosphatase. Another difference is that a primary stimulus of hepatic glycogenolysis is glucagon, which does not act on muscle.

Binding of glucagon to its receptor activates adenylyl cyclase in liver, producing cAMP from ATP . Besides initiating glycogenolysis, an increase in liver cAMP suppresses glycogen synthesis and increases gluconeogenesis . As noted earlier, liver glycogenolysis is critical in meeting the body's energy requirements in the early stages of starvation. Between one third and two thirds of the glucose released by the liver after an overnight fast is derived from hepatic glycogen . Several inherited metabolic disorders of glycogen storage or breakdown have been described. These include von Gierke disease

(type I glycogen storage disease), in which glucose-6-phosphatase is deficient and the liver cannot release free glucose into the circulation; Hers disease (type VI glycogen storage disease), in which liver glycogen phosphorylase is absent; and Cori disease (type III glycogen storage disease), in which the debranching enzyme that hydrolyzes the 1,6 linkage of the glycogen molecule is absent

Gluconeogenesis

In humans, the maintenance of euglycemia during starvation depends on the ability of the liver to synthesize glucose from nonhexose precursors (i.e., gluconeogenesiss). The molecular mechanisms regulating gluconeogenesis and the disorders of this pathway in humans have been reviewed elsewher. Gluconeogenesis use many of the enzymes involved in glycolysis but requires unique enzymes to circumvent the reactions catalyzed by glucokinase, phosphofructokinase, and pyruvate kinase . As with glycogenolysis, glucagon is a major positive hormonal modulator of gluconeogenesis and insulin is the primary inhibitor. Catecholamines also stimulate gluconeogenesis and may be the principal positive regulator in some patients with long-standing type 1 diabetes in whom glucagon secretion is impaired . Glucocorticoids appear to play an important permissive role, since the stimulation of gluconeogenesis by glucagon and catecholamines is diminished in their absence.

Formation of ketone bodies

Synthesis of ketone bodies occurs almost exclusively within the liver. Mitochondrial acetyl-CoA produced from oxidation of fatty acids either can combine with oxaloacetate and enter the tricarboxylic acid (TCA) cycle or can be used for the

synthesis of acetoacetate and β-hydroxybutyrate within the mitochondrion . When rates of FFA-CoA uptake by mitochondria are high, much of the energy needs of the liver are generated by their β-oxidation to acetyl-CoA. Under these conditions, acetyl-CoA preferentially enters the pathway for ketone-body formation and its oxidation in the TCA cycle is diminished . The high ratio of glucagon to insulin and the increase in intrahepatic fatty acids during prolonged starvation stimulate the enzyme CPTI, which is located within the outer leaflet of the inner mitochondrial membrane and is rate-limiting for fatty acid oxidation.

Interrelations between fatty acid and glucose metabolism and insulin resistance.

Glucose in the presence of insulin inhibits the oxidation of fatty acids in muscle, liver, the pancreatic β-cell, and undoubtedly other tissues. Conversely, elevated levels of fatty acids can inhibit the oxidation of glucose. From a functional perspective, such increases in plasma levels of FFA have been linked to events such as the stimulation of hepatic gluconeogenesis and ketogenesis and the maintenance of the low but finite rate of insulin secretion during starvation. The notion that high plasma levels of FFA could also contribute to the insulin resistance (defined as a decrease in the biologic effect of the hormone) in diabetes and obesity was initially given credibility by the studies of Randle et al[78,79]. that led them to propose the existence of a glucose-fatty acid cycle. The Randle mechanism was worked out in a preparation of isolated perfused rat heart, and although it has been difficult to apply it to other tissues, recent studies have strongly suggested a link between abnormalities in fatty acid metabolism and insulin resistance. Thus, the administration of lipids to prevent decreases in plasma FFA levels during an infusion of

insulin and glucose (euglycemic-hyperinsulinemic clamp) has been shown to diminish the ability of insulin to increase glucose uptake by muscle and to diminish its production by liver in humans and experimental animals[80,81] . Likewise, a close association between insulin resistance (assessed by the clamp procedure) and intramuscular triglycerides, quantified by NMR imagery, has been reported by several groups . Concurrently, studies in rodents have found that insulin resistance in skeletal muscle in a wide variety of conditions is characteristically associated with increases in the concentrations of malonyl-CoA, long-chain fatty acyl-CoA, diacylglycerol, and triglycerides and alterations in the distribution and activation of certain protein kinase C isoforms[82]. The possible linkage of this or a similar mechanism to obesity and to the disordered function and metabolism of other tissues in individuals with type 2 diabetes and obesity has been discussed elsewhere[83]. An attractive feature of this mechanism is that it could explain why exercise, acting through AMPK, could attenuate insulin resistance and exert effects on multiple tissues.

The molecular mechanism of insulin action and the regulation of glucose and lipid metabolism

More than 18 million people in the United States have diabetes mellitus, and about 90% of these have the type 2 form of the disease. In addition, between 17 and 40 million people have insulin resistance, impaired glucose tolerance, or the cluster of abnormalities referred to variably as the metabolic syndrome, the dysmetabolic syndrome, syndrome X, or the insulin resistance syndrome . In all of these disorders, a central component of the pathophysiology is insulin resistance, i.e., reduced

responsiveness to insulin in tissues such as muscle, fat, and liver. In type 2 diabetes, the β-cell can no longer secrete sufficient insulin to compensate for insulin resistance, leading to relative insulin deficiency. Insulin resistance is also closely linked to other common health problems, including obesity, polycystic ovarian disease, hyperlipidemia, hypertension, and atherosclerosis.

Glucose homeostasis and insulin resistance

Despite periods of feeding and fasting, in healthy individuals plasma glucose remains in a narrow range between 4 and 7 mM (70 to 120 mg/dL). This tight control of glucose concentration is determined by a balance between glucose absorption from the intestine, glucose production by the liver, and glucose uptake from the plasma. In tissues such as muscle, fat, and liver, glucose uptake and/or storage is regulated by insulin, whereas insulin has no apparent role in stimulating glucose metabolism in tissues such as brain, kidney, and erythrocytes. In addition to promoting glucose utilization, insulin inhibits both basal and glucagon-stimulated hepatic glucose production, thus serving as the primary regulator of blood glucose concentration during fasting. Insulin also has a general anabolic role promoting the storage of substrates in fat, liver, and muscle by stimulating lipogenesis and glycogen and protein synthesis; inhibiting lipolysis, glycogenolysis and protein breakdown; and stimulating cell growth and differentiation. In type 1 diabetes, the autoimmune destruction of the pancreatic β-cell leads to severe insulin deficiency with unrestrained hepatic glucose output, unrestrained lipolysis, and increased ketogenesis. In type 2 diabetes, insulin resistance in muscle, adipose tissue, and liver combined with a relative failure of the β-cell leads to increased glucose levels and a

variable cluster of metabolic alterations in lipid and protein metabolism. Insulin resistance in patients with type 2 diabetes is usually defined by defects in insulin-stimulated glucose transport, glycogen synthesis, and glucose oxidation, but other pathways of metabolism are clearly altered. The most characteristic feature of the β-cell failure is a specific defect in glucose sensing characterized by loss of first-phase insulin secretion in response to a glucose stimulus, while response to other secretagogues is normal or only mildly depressed.

The control of blood glucose depends upon the balance between glucose production by the liver and glucose utilization by insulin-dependent tissues, such as muscle and fat, and insulin-independent tissues, such as the brain. In mammals, up to 75% of insulin-dependent glucose disposal occurs in skeletal musce. This preeminence of muscle, however, has recently been challenged by the finding that mice with a muscle-specific knockout of the insulin receptor exhibit minimal abnormalities in glucose tolerance . Adipose tissue accounts for only a small fraction (5% to 15%) of insulin-stimulated glucose disposal. Despite this, knockout of the insulin-sensitive glucose transporter in fat leads to impaired glucose tolerance, apparently by inducing insulin resistance in muscle and liver through a yet undetermined mechanism .Adipose tissue also plays a special additional role in glucose homeostasis through its release of free fatty acids, tumor necrosis factor-α (TNF-α) leptin, Acrp30/adiponectin, and other adipokines that have been shown to contribute to insulin action and insulin resistance . Furthermore, both obesity (increased fat mass) and lipoatrophy (decreased fat mass) cause insulin resistance and predisposition to type 2 diabetes.

The liver does not exhibit insulin-stimulated glucose uptake but plays a major role in glucose homeostasis, especially in the fasting state [84]. When insulin levels are low, the liver releases glucose into the blood as a result of glycogenolysis and gluconeogenesis, providing substrate for tissues with obligate glucose requirements. In the fed state, when insulin levels are high, glucose in the liver is converted to glycogen. Recent studies using knockout and other technologies suggest that insulin action in other tissues, including brain and β-cells, although not major sites of insulin-stimulated glucose uptake, may also play important roles in glucose homeostasis and metabolism.

Proximal Signaling Pathways

The Insulin Receptor and Its Substrates

The insulin receptor is a tetrameric protein consisting of two α-subunits and two β-subunits that belongs to a subfamily of receptor tyrosine kinases that also includes the insulin-like growth factor-1 (IGF-1) receptor and an orphan receptor called the insulin receptor-related receptor (IRR). Each of these receptors is the product of a separate gene in which the two subunits are derived from a single-chain precursor or proreceptor that is processed by a furin-like enzyme to give a single α-β subunit complex . Two of the α-β dimers then undergo disulfide linkage to form the tetramer.

The insulin receptor is widely distributed throughout the body, including in tissues classically regarded as “responsive” and “nonresponsive” to insulin. Recent studies suggest that the receptor in most of these tissues has an important functional role, but in some cases this may relate to actions other than the control of glucose or lipid homeostasis. For example, in ovarian granulosa cells, insulin signaling is coupled to

regulation of estrogen/androgen balance , whereas the role of the insulin receptor in the endothelial cell may be to promote vasodilators or transcytosis of the insulin molecule from the intravascular space to the interstitial space and its target tissue; in neural or endocrine cells, insulin may have a role regulating hormone production, secretory function, or signal sensing .

Functionally, the insulin receptor behaves as a classical allosteric enzyme in which the α-subunit inhibits the tyrosine kinase activity intrinsic to the β-subunit. Insulin binding to the α-subunit, or removal of the α-subunit by proteolysis or genetic deletion, leads to a derepression, i.e., activation, of the kinase activity in the β-subunit. Following this initial activation, there is transphosphorylation of the β-subunits, i.e., one subunit phosphorylates the other, leading to a conformational change and a further increase in activity of the kinase domain . The α-β heterodimers of the insulin, IGF-1, and the IRR receptors can form functional hybrids in which occupancy of one receptor's binding domain leads to activation of the other receptor in the heterodimer by this transphosphorylation process. Likewise, a dominant-negative form of one of these receptor subtypes can inhibit the activity of the other receptors by forming heterodimers . This may explain in part why individuals with mutations in the insulin receptor exhibit both insulin resistance and growth retardation .

The insulin/IGF-1 signaling system is evolutionarily very ancient. Homologues of the insulin/IGF-1 receptor have been identified in Drosophila, Caenorhabditis elegans, and even metazoan marine sponges of the phylum Porifera that date back over 500 million years . In the lower organisms, this system uses many of the same downstream signals used in mammalian cells, i.e., phosphatidylinositol 3-kinase (PI 3-

kinase)/ Akt/forkhead transcription factors, and may also be involved in regulation of metabolism . In Drosophila, the insulin-secreting cells are neurons. Ablation of these cells results in changes in the major circulating carbohydrate in flies, trehelose . In C. elegans, a major effect of the insulin/IGF system is on aging, such that animals with mutations of the receptor that reduce insulin action live much longer than normal animals, whereas mutations in other parts of the pathway may reverse this longevity . It is interesting that chronic food restriction and leanness, which are associated with lower circulating insulin levels, increase longevity in rodents. This raises a number of interesting questions about the association of hyperinsulinemia and insulin resistance with conditions that shorten life span in humans, such as obesity, diabetes, and accelerated atherosclerosis.

At least nine intracellular substrates of the insulin and IGF-1 receptor tyrosine kinases have been identified . Four of these belong to the family of insulin/IGF-1 receptor substrate (IRS) proteins . These IRS proteins are characterized by the presence of both pleckstrin homology (PH) and phosphotyrosine binding (PTB) domains near the N-terminus that account for the high affinity of these substrates for the insulin receptor and up to 20 potential tyrosine phosphorylation sites spread throughout the center and C-terminal region of the molecule. The molecular mass of IRS proteins ranges from 60 to 180 kDa. IRS-1 and IRS-2 are widely distributed, whereas IRS-3 and IRS-4 have more limited distributions. IRS-3 is most abundant in adipocytes, and its mRNA is also detected in liver, heart, lung, brain, and kidney. In contrast, the levels of mRNA for IRS-4 are very low, but are detectable, in fibro-blasts, embryonic tissues, skeletal muscle, liver, heart, hypothalamus, and kidney . Interestingly, in humans the IRS-3 gene appears

to be nonfunctional, leaving only IRS-1, -2, and -4 . Other direct substrates of the insulin/IGF-1 receptor kinases include Gab-1 , $p62^{dok}$, Cbl , and the various isoforms of Shc . Following insulin stimulation, the receptor directly phosphorylates most of these substrates on multiple tyrosine residues. The phosphorylated tyrosines in each of these substrates occur in specific sequence motifs and once phosphorylated serve as "docking sites" for intracellular molecules that contain SH2 (Src-homology 2) domains . Thus, the insulin-receptor substrates function as key intermediates in insulin-signal transduction.

The SH2 proteins that bind to phosphorylated IRS proteins fall into two major categories. The best studied are adapter molecules, such as the regulatory subunit of PI 3-kinase or the molecule Grb2, which associates with SOS to activate the Ras-mitogen-activated protein (MAP) kinase pathway . The other major category of proteins that bind to IRS proteins are enzymes, such as the phosphotyrosine phosphatase SHP2 and cytoplasmic tyrosine kinases, such as Fyn. A few proteins that bind to phosphotyrosines in the IRS proteins do not contain known SH2 domains; these include the calcium adenosine triphosphatases (ATPases) SERCA 1 and 2 and the SV40 large T antigen . IRS proteins also undergo serine phosphorylation in response to insulin and other stimuli. In general, serine phosphorylation appears to act as a negative regulator of insulin signaling by decreasing tyrosine phosphorylation of IRS proteins, as well as by promoting interaction with 14-3-3 proteins . A number of different intracellular enzymes have been suggested as being involved in this serine phosphorylation, including some in the insulin-signaling pathway, such as Akt , JNK kinase , and PI 3-kinase (which also has serine kinase activity) , thereby providing a form of autoinhibition of signaling, and others that mediate

the effects of some inhibitors of insulin action, such as the inhibitor kappa B kinase β (IKKβ) .

Although the IRS proteins are highly homologous and possess many similar tyrosine phosphorylation motifs, recent studies in knockout mice and knockout cell lines suggest that the various IRS proteins serve complementary rather than redundant roles in insulin and IGF-1 signaling . The IRS-1 knockout mouse exhibits IGF-1 resistance as manifested by prenatal and postnatal growth retardation, as well as insulin resistance, primarily in muscle and fat, resulting in impaired glucose tolerance. IRS-2 knockout mice also exhibit insulin resistance, but primarily in the liver, and have defects in growth in only selected tissues of the body, including certain regions of the brain, β-cells, and retinal cells. Likewise at the cellular level, IRS-1 knockout preadipocytes exhibit defects in differentiation , whereas IRS-2 knockout preadipocytes differentiate normally but fail to respond to insulin-stimulated glucose transport .

The β-cell compensatory responses of the IRS knockout mice also differ. In the IRS-1 knockout, although there is some element of β-cell dysfunction, there is sufficient islet hyperplasia such that the animals develop only mildly impaired glucose tolerance. In the IRS-2 knockout mouse, there is a decrease in islet mass due to altered β-cell development. The combination of multifactorial insulin resistance and decreased β-cell mass leads to the development of early-onset diabetes in IRS-$2^{-/-}$ mice , although the frequency of this phenotype varies considerably in different laboratories . By contrast, IRS-3 knockout mice have normal growth and metabolism, whereas IRS-4 knockout mice exhibit only minimal abnormalities in glucose tolerance . It is interesting that when IRS-1-deficient mice are crossed with IRS-3-deficient mice to produce a double

knockout, the resultant animals exhibit severe hyperglycemia and marked lipoatrophy, indicating that, at least in adipocytes, there is at least some compensation of these two substrates .

The differential roles of the IRS proteins may be due to differences in tissue distribution, subcellular localization, and intrinsic activity of the proteins. IRS-1 and IRS-2 are widely distributed, whereas IRS-3 is limited largely to the adipocyte and brain and IRS-4 is expressed primarily in embryonic tissues or cell lines . Furthermore, IRS-1 is more closely associated with low-density microsomes, whereas IRS-2 is found in low-density microsomes and in the cytosol . IRS-3 is associated more with the plasma-membrane fraction in rat adipocytes . In some studies IRS-1 and IRS-3 appear to translocate to the nucleus , and IRS-3 has been suggested to possess DNA-binding activity .

Turning off the insulin signal

Unlike the prolonged actions of steroid and thyroid hormones, insulin action on glucose homeostasis demands a rapid on-and-off response to avoid the dangers of hypoglycemia. Several different mechanisms play a role in turning off the insulin signal.First, insulin may simply dissociate from the receptor and be degraded. Following dissociation of the ligand, phosphorylation of the insulin receptor and its substrates is rapidly reversed by the action of protein tyrosine phosphatases (PTPases). Several PTPases have been identified that are capable of catalyzing dephosphorylation of the insulin receptor *in vitro* or *in vivo*, and some are even upregulated in insulin-resistant state. Most attention has focused on the cytoplasmic phosphatase PTP-1b. Disruption of the gene encoding this enzyme in mice produces increased insulin-dependent tyrosine phosphorylation of the insulin receptor and IRS proteins in muscle and leads to a state of

improved insulin sensitivity . PTP-1b knockout mice are also resistant to diet-induced obesity, suggesting an effect of PTP-1b deletion in the brain, with subsequent changes in energy uptake and expenditure. This is opposite the effect of knockout of insulin receptor in the brain .

Several other mechanisms may also be involved in turning off the insulin signal in normal or pathologic states. The insulin receptor itself may be internalized and undergo degradation . As noted above, serine phosphorylation of the insulin receptor and its substrates also inhibits insulin action . Finally, the phosphorylated receptor may interact with proteins in the cell that block insulin action. This latter mechanism has recently been observed for the SOCS (suppressors of cytokine signaling) proteins in the case of the insulin resistance associated with inflammation and obesity .

Regulation Of Glucose Transport

The classical effect of insulin on glucose homeostasis is its ability to stimulate glucose transport in fat and muscle. This occurs via a translocation of GLUT4 glucose transporters from intracellular sites to the plasma membrane . The GLUT4 protein consists of 12 transmembrane helices with a characteristic C-terminal tail containing two adjacent leucine residues commonly found in proteins that undergo regulated trafficking. In the basal state, GLUT4 continuously recycles between the cell surface and various intracellular compartments. The GLUT4 vesicle is highly specialized and appears to form from a sorting endosomal population. Insulin markedly increases the rate of GLUT4-vesicle exocytosis and slightly decreases the rate of internalization of the GLUT4 protein. Although the exact domains of the protein involved in localization and trafficking remain

controversial, the C- and N-terminal tails of the protein, both of which are oriented on the cytoplasmic side of the vesicle, appear to be required . It is likely that the GLUT4 vesicle moves along microtubule tracks to the cell surface, perhaps via kinesin motors . These vesicles then fuse with the plasma membrane, allowing for the extracellular exposure of the GLUT4 protein.

Recent evidence also suggests that the actin cytoskeleton plays a critical role in insulin-stimulated GLUT4 translocation. Insulin has been shown to cause a remodeling of actin filaments just below the plasma membrane in a variety of cellular systems, with an induction of actin polymerization and membrane ruffling. This effect on ruffling is likely to reflect polymerization and depolymerization beneath the membrane, involving lamellipodia and/or filopodia formation. Actin-depolymerizing agents, such as cytochalasin D and the actin monomer-binding toxins latrunculin A and B, inhibit insulin-stimulated GLUT4 translocation . The C-terminal tail of GLUT4 in adipocytes has been shown to indirectly interact with F-actin by binding to the glycolytic enzyme aldolase, suggesting a homeostatic mechanism in which glucose metabolism might feedback regulate GLUT4 translocation along the actin cytoskeleton .

The docking and fusion of the GLUT4 vesicle at the plasma membrane are subject to regulation by insulin. This involves a series of proteins termed the SNARE proteins. The v-SNARE protein VAMP2 is present on GLUT4-containing vesicles and appears to physically interact with its t-SNARE counterpart syntaxin 4 during GLUT4-vesicle docking and fusion with the plasma membrane , although neither SNARE protein appears to be a direct target of insulin action. However, the SNARE accessory proteins Synip and Munc18c may be involved in the control of GLUT4 docking and fusion in an

insulin-dependent, PI 3-kinase-independent manner . One interesting possibility is that the PI 3-kinase-independent arm of insulin action may be directed at the docking and fusion step of GLUT4 regulation.

Regulation of glucose and lipid synthesis, utilization, and storage

Glucose oxidation and storage

Upon entering the muscle cell, glucose is rapidly phosphorylated by hexokinase and either stored as glycogen via the activity of glycogen synthase or oxidized to generate adenosine triphosphate (ATP) synthesis via enzymes such as pyruvate kinase. In the liver and adipose tissue, glucose can also be stored as fat. Some of the enzymes involved in glycolysis, as well as in glycogen and lipid synthesis, are regulated by insulin via changes in their phosphorylation state due to a combination of protein kinase inhibition and phosphatase activation. In addition, some of these enzymes are regulated at the transcriptional level.

Insulin stimulates glycogen accumulation through a coordinated increase in glucose transport and glycogen synthesis. Activation of glycogen synthase involves the promotion of its dephosphorylation via both the inhibition of kinases that can phosphorylate glycogen synthase, such as PKA , and the activation of phosphatases that dephosphorylate glycogen synthase, such as protein phosphatase 1 . This process is downstream of PI 3-kinase and involves Akt phosphorylation of GSK-3. This inactivates GSK-3, resulting in a decrease in the phosphorylation of glycogen synthase and an increase in its activity state. However, the inhibition of GSK-3 is not sufficient for full

activation of glycogen, because GSK-3 does not phosphorylate all of the residues of glycogen synthase that are dephosphorylated in response to insulin.

Activation of PP1 correlates well with changes in glycogen synthase activitvty. However, insulin does not appear to globally activate PP1 but rather to activate specific pools of the phosphatase localized on the glycogen particle. The compartmentalized activation of PP1 by insulin is due to glycogen-targeting subunits that serve as "molecular scaffolds," bringing together the enzyme with its substrates glycogen synthase and glycogen phosphorylase in a macromolecular complex. Four different proteins (G_M, G_L, PTG, and R_6) have been reported to target PP1 to the glycogen particle. Overexpression of these scaffolding proteins in cells or in vivo by adenovirus-mediated gene transfer results in a dramatic increase in basal cellular glycogen levels . Furthermore, glycogen stores in cells overexpressing PTG are refractory to breakdown by agents that raise intracellular cyclic adenosine monophosphate (cAMP) levels, suggesting that PTG locks the cell into a glycogenic mode . The mechanism by which insulin activates glycogen-associated PP1 remains unknown. Although it had been proposed that activation of MAP kinase leads to the phosphorylation of the targeting protein G_M and the subsequent release of inhibition of the enzyme by insulin, blockade of this pathway had no effect on the activation of glycogen synthase by insulin and mutation of the identified phosphorylation sites did not impair insulin action. However, inhibitors of PI 3-kinase can block activation of PP1 by insulin, indicating that PIP_3-dependent protein kinases are involved.

Regulation of Gluconeogenesis

Insulin inhibits the production and release of glucose by the liver and, to a lesser extent, by the kidney by blockade of gluconeogenesis and glycogenolysis. Insulin achieves these effects by directly controlling the activities of a subset of metabolic enzymes via the process of phosphorylation and dephosphorylation described above, as well as by regulation of the expression of a number of genes encoding hepatic enzymes. Insulin dramatically inhibits the transcription of the gene encoding phosphoenolpyruvate carboxylase (PEPCK), the rate-limiting step in gluconeogenesis. The hormone also decreases transcription of the genes encoding fructose 1,6-bisphosphatase and glucose 6-phosphatase and increases transcription of those encoding glycolytic enzymes such as glucokinase and pyruvate kinase and lipogenic enzymes such as fatty acid synthase and acetyl CoA carboxylase.

Several transcription factors play a role in this insulin-mediated regulation .Hepatic nuclear factor-3 (HNF3) and HNF4 both appear to be involved in regulation of the PEPCK gene, which is the rate-limiting enzyme of gluconeogenesis. Sterol regulatory element-binding protein-1c (SREBP-1c) is regulated by insulin in its phosphorylation and may play a role in the effect of insulin on PEPCK gene transcription). The forkhead transcription factor FKHR (now known as FOXO1) also appears to be involved in the regulation of PEPCK and glucose 6-phosphatase, because both PEPCK and glucose 6-phosphatase contain putative FKHR binding sites in their promoter sequences, and overexpression of FKHR in hepatoma cells markedly increases the expression of the catalytic subunit of glucose 6-phosphatase). Recently, it was showed that both HNF4 and FOXO1 may be modified in their activity by a single co-activator known as PGC-1.

PGC-1 levels are increased in insulin-deficient and insulin-resistant diabetes. This creates an attractive hypothesis by bringing together multiple regulators under one common master regulator.

Although there is no doubt that insulin plays a key role in the regulation of the enzymes of gluconeogenesis, insulin can also indirectly influence glucose metabolism. This occurs via changes in the availability of substrates for gluconeogenesis that are being released from muscle and fat. Thus, when insulin levels are low, there is a breakdown of muscle protein and adipocyte triglycerides, leading to increased levels of gluconeogenic substrates such as alanine and free fatty acids. Careful physiologic experiments in the dog that included time courses and dose responses of insulin action have suggested that under some circumstances this indirect pathway may be the major pathway of insulin regulation of gluconeogenesis .However, recent experiments with mice with a genetic knockout of the insulin receptor in liver indicate that the direct pathway is more important in that species . In any case, in humans the indirect pathway may contribute to the pathogenesis of diabetes, especially in individuals with central obesity, because visceral fat is less sensitive than subcutaneous fat to insulin inhibition of lipolysis, resulting in direct flux of fatty acids derived from these fat cells through the portal vein to the liver.

Regulation of lipogenesis and lipolysis

As is the case with carbohydrate metabolism, insulin also promotes the synthesis of lipids and inhibits their degradation. Recent studies suggest that many of these changes also might require an increase in levels of the transcription factor SREBP1-c. Dominant-negative forms of SREBP1 can block expression of these gluconeogenic and lipogenic

genes, and overexpression of SREBP-lc can increase their expression . Interestingly, hepatic SREBP levels are increased in rodent models of lipodystrophy, and this is associated with coordinated increases in fatty acid synthesis and gluconeogenesis, mimicking the phenotype observed in genetic models of obesity-induced diabetes. These observations led to speculate that increased expression of SREBP-1c might lead to the mixed insulin resistance observed in the diabetic liver, with increased rates of both gluconeogenesis and lipogenesis. The pathways that account for the changes in SREBP1-c expression lie downstream of the IRS/PI 3-kinase pathway.

In adipocytes, glucose is stored primarily as lipid. This is the result of increased uptake of glucose and activation of lipid synthetic enzymes, including pyruvate dehydrogenase, fatty acid synthase, and acetyl CoA carboxylase. Insulin also profoundly inhibits lipolysis in adipocytes, primarily through inhibition of the enzyme hormone-sensitive lipase. This enzyme is acutely regulated by control of its phosphorylation state, activated by PKA-dependent phosphorylation, and inhibited owing to a combination of kinase inhibition and phosphatase activation. Insulin inhibits the activity of the lipase primarily via reductions in cAMP levels due to the activation of a cAMP-specific phosphodiesterase in fat cells .

What causes insulin resistance?

Defining Insulin Resistance and the Sites of Insulin Resistance.

Insulin resistance is said to exist any time a normal amount of insulin produces a less than normal biologic response. Insulin resistance can be further divided into states in which there is a rightward shift in the dose response to the hormone but the maximal response remains normal (decreased insulin sensitivity) or states in which the dose

response is normal but the maximal response is decreased (decreased responsiveness), or a combination of the two . Insulin resistance is extremely common, occurring both in disease states such as type 2 diabetes, obesity, hypertension, polycystic ovarian disease, and a variety of genetic syndromes and in physiologic conditions such as puberty and pregnancy. Insulin resistance also is present in many states of stress, in association with infection, and secondary to treatment with a variety of drugs, particularly glucocorticoids. From a molecular perspective, insulin resistance can occur at multiple levels and be either acquired or genetic. Prereceptor insulin resistance is rare today but formerly was exemplified by patients with high levels of circulating antibodies to insulin that blocked binding of the ligand to its receptor and by patients with what appeared to be increased subcutaneous degradation of injected insulin .Insulin resistance at the level of the receptor may be the result of genetic alterations in receptor expression or structure, secondary changes in receptor activity due to serine phosphorylation, or downregulation of receptor concentration. At the postreceptor level, insulin resistance can occur almost anywhere on one of the common or branched pathways of insulin signaling.

In the most common states of insulin resistance, there appear to be defects at multiple levels. For example, in type 2 diabetes, there are decreases in receptor concentration, in receptor kinase activity, in the concentration and phosphorylation of IRS-1 and IRS-2, in PI 3-kinase activity, and in glucose-transporter translocation and defects in activity of intracellular enzymes. Interestingly, in type 2 diabetes, there does not appear to be a reduction in insulin action on the MAP kinase pathway .This blockade of the PI 3-kinase pathway with continued MAP kinase signaling might account for some of the detrimental effects of the chronic hyperinsulinemia on the vasculature .

Genetic forms of insulin resistance

Insulin resistance due to genetic defects in insulin-receptor expression or sequence is relatively rare but represents the most severe forms of insulin resistance. In humans, these may present as several different disease syndromes, including two congenital diseases termed leprechaunism and the Rabson-Mendenhall syndrome, in which there is insulin resistance, intrauterine and postnatal growth retardation, and other developmental defects; and the type A syndrome of insulin resistance that appears in childhood, adolescence, or early adulthood. Although there is some correlation between the severity of the genetic defect in receptor function and the severity of the clinical presentation, the correlation is relatively weak, indicating that other genetic or acquired factors can modify the insulin-resistant state significantly .Interestingly, none of these diseases matches the phenotype of the insulin-receptor knockout mouse, which shows normal intrauterine growth but develops diabetic ketoacidosis in the first few days of life and dies .This difference in behavior may represent differences in the role of the insulin receptor in different species or the state of development of the human versus that of the mouse at birth. Alternatively, the mutant receptors may produce more complex disease phenotypes as a result of formation of hybrids with IGF-1 or other receptors that also interfere with their function.

Acquired forms of insulin resistance

Acquired forms of insulin resistance may occur as a result of multiple mechanisms .The first of these to be described was that of insulin-receptor downregulation .In this situation, mild hyperinsulinemia that occurs in response to tissue insulin resistance results in an increase in internalization and degradation of the insulin receptor. This occurs to some extent in the most common insulin-resistant states, i.e., obesity and type 2 diabetes. Recent studies have also shown that hyperinsulinemia can lead to downregulation of insulin-receptor substrates, producing an even greater decrease in insulin signaling .In both humans and rodents, the levels of insulin receptor and IRS-1 in some tissues can each be reduced by more than 50% in some of these insulin-resistant states. Most of the changes in the insulin receptor and its substrates are due to increased protein turnover, but there may also be an element of downregulation at the transcriptional level, especially for IRS-2.

In addition to downregulation, there may be many other factors that contribute to acquired insulin resistance. As noted above, in hyperinsulinemic and other insulin-resistant states, there is increased serine phosphorylation of the receptor and its substrates .This leads to decreased kinase activity of the receptor and decreased tyrosine phosphorylation of the receptor substrates. Several different serine kinases have been implicated in this serine phosphorylation, including Akt, various isoforms of PKC, and the stress-induced MAP kinases (p38 and JNK) and IKB kinase .The upstream stimulators of these kinases may also be multiple. For example, in obesity and type 2 diabetes, there are increased levels of circulating free fatty acids (FFAs), and in obesity, adipose tissue makes and releases a number of other factors, including TNF-α, leptin,

various complement-related peptides, and two recently discovered hormones, resistin and adiponectin (also called Acrp30 and AdipoQ). Another class of proteins that can act as inhibitors of insulin signaling are the SOCS proteins. These certainly play a role in stress-induced states, such as that created by injection of bacterial lipopolysaccharide, and perhaps in obesity-linked insulin resistance. These SOCS proteins act to inhibit insulin signaling by binding to the phosphorylated insulin receptor and inhibiting phosphorylation of the IRS proteins.

Role of free fatty acids and intracellular triglycerides in insulin resistance

Circulating FFAs are elevated in many insulin-resistant states and have been suggested to play a central role in the pathogenesis of the insulin resistance. Physiologic increases in plasma FFA levels have been shown to cause insulin resistance by several mechanisms in both diabetic subjects and obese, nondiabetic subjects. FFAs inhibit insulin-stimulated glucose uptake at the level of glucose transport and/or phosphorylation, inhibit insulin-stimulated glycogen synthesis, and inhibit insulin-stimulated glucose oxidation. As noted above, FFAs might have a special role in the insulin resistance associated with central obesity. Since central adipocytes are more resistant to insulin inhibition of lipolysis, there is an increased delivery of FFAs to the liver. This leads to increased accumulation of triglycerides that could also contribute to increased hepatic glucose output, reduced hepatic extraction of insulin, and hepatic insulin resistance. In experimental lipid-induced insulin resistance, insulin-stimulated IRS-1 phosphorylation and IRS-1-associated PI 3-kinase activity is also reduced. There is also an increase in membrane-bound, i.e., activated, PKC that may serve as a mediator of

the insulin resistance by increased serine phosphorylation of the insulin receptor and/or IRS-1 .

A common link between increased levels of FFAs and the insulin resistance in type 2 diabetes, obesity, and syndrome X could be accumulation of triglycerides in muscle. Recent studies using magnetic resonance spectroscopy have demonstrated that at least some of the lipid accumulation is inside the myocyte itself. Factors leading to the accumulation of triglycerides are not clear, but it has been speculated that the triglyceride is derived from elevated levels of both circulating FFAs and triglycerides and is also the result of reduced muscle fatty acid oxidation. Whatever the mechanism, there is a close correlation between muscle triglyceride content and whole-body insulin resistance. The notion of a glucose-fatty acid cycle (Randle cycle) has been hypothesized for 40 years as a mechanism by which glucose might autoregulate its own use. It is likely that cytosolic accumulation of the long-chain fatty acyl CoAs is involved in the altered insulin signaling. Several mechanisms have been implicated in the inhibition of insulin signaling, including increased serine phosphorylation of the insulin receptor and its substrates or direct inhibition of enzymes such as glycogen synthase. Insulin sensitizers, such as the PPARγ agonists, reduce muscle lipid accumulation and increase insulin sensitivity. Other potent systemic lipid-lowering agents, such as PPAR γ agonists (e.g., fibrates) or antilipolytic agents (e.g., nicotinic acid analogues), might also improve insulin sensitivity by this mechanism.

Transgenic mice with muscle- and liver-specific overexpression of lipoprotein lipase have recently been developed to help define the roles of muscle FFAs and triglycerides in insulin resistance. Muscle-specific lipoprotein lipase-deficient mice have

a threefold increase in muscle triglyceride content and exhibit insulin resistance due to decreases in insulin-stimulated glucose uptake in skeletal muscle and insulin activation of IRS-1-associated PI 3-kinase activity . Mice with overexpression of lipoprotein lipase in the liver have increased triglyceride content in the liver and exhibit insulin resistance due to an impaired ability of insulin to suppress endogenous glucose production, along with defects in insulin activation of IRS-2-associated PI 3-kinase activity. In both tissues, these defects in insulin action and signaling are associated with increases in intracellular fatty acid-derived metabolites, such as diacylglycerol and fatty acyl CoA.

The Fat cell as a secretory cell and insulin resistance

Over the past several years, it has become clear that the adipocyte plays a role in insulin resistance not only by storing fat but also as a secretory cell producing several cytokines and hormones, as well as releasing FFAs .The first of the cytokines to be described as being increased in fat cells of obese animals and humans was TNF-α. TNF-α could lead to insulin resistance by increasing serine phosphorylation of IRS-1 and decreasing insulin-receptor kinase activity. This mechanism is clearly important in rodents, in which anti-TNF α reagents significantly improve insulin resistance. However, the importance of this mechanism in humans is much debated, and limited studies of anti-TNF reagents have shown little or no effect on the insulin-resistant state .

Leptin is a member of the cytokine family of hormones that is produced by adipose tissue and acts on receptors in the central nervous system and other sites to inhibit food intake and promote energy expenditure. Leptin has been shown to interfere with insulin signaling systems in vitro; however, it is not clear if leptin has anti-insulin

effects in vivo. Indeed, in states of severe leptin deficiency, such as in the ob/ob mouse or several genetic models of lipoatrophic diabetes, administration of exogenous leptin improves glucose tolerance and insulin sensitivity. This appears to be primarily the result of an action of leptin at the liver to increase insulin sensitivity, an effect that might be direct or centrally mediated.

Adiponectin (also called Acrp30, adipoQ, APM-1, and GBP28) is a peptide of 247 amino acids that possesses a collagenous domain at the N-terminus and a globular domain that shares significant homology with subunits of complement factor C1q. The expression of adiponectin is highly specific to adipose tissue. Adiponectin is among the most abundant proteins in adipocytes, is secreted into the bloodstream, and is present at very high circulating concentrations. Several recent studies have pointed to a potentially important role of adiponectin in the insulin resistance of obesity. First, expression of adiponectin mRNA is decreased in obese humans and mice and in some models of lipoatrophic diabetes. Acute treatment of mice with the globular head domain of Acrp30 significantly decreased the elevated levels of plasma FFAs and caused weight loss in mice consuming a high-fat diet. Administration of adiponectin to obese mice also decreases insulin resistance and triglyceride content of muscle and liver. Moreover, insulin resistance in lipoatrophic mice is completely reversed by the combination of physiologic doses of adiponectin and leptin, but only partially by administration of either adiponectin or leptin alone. Administration of adiponectin/Acrp30 also lowers glucose levels in normal mice and mouse models of type 1 diabetes, such as NOD mice and streptozotocin-treated mice. Recent genome-wide scans have mapped a susceptibility locus for type 2 diabetes and metabolic syndrome to chromosome 3q27, a region where

the gene encoding adiponectin is located. These data suggest that decreased levels of adiponectin are important factors in the insulin resistance of obesity and lipoatrophy and that replacement of adiponectin might provide a novel treatment for some insulin-resistant states.

Resistin is the most recently discovered peptide hormone secreted by adipocytes. Resistin belongs to a family of tissue-specific secreted proteins termed resistin-like molecules (RELMs) and the FIZZ (found in inflammatory zone) family. Initial studies suggested that resistin levels were increased in both genetic and acquired obesity in mice and reduced by antidiabetic drugs of the thiazolidinedione class. Further, administration of antibody to resistin appeared to improve blood glucose levels and insulin action in mice with diet-induced obesity. Moreover, insulin-stimulated glucose uptake by adipocytes was enhanced by neutralization of resistin and reduced by resistin treatment.

Regulation of glucagon secretion

In the absence of ketosis, glucose is the obligate fuel used by the brain. Because the brain cannot synthesize glucose and only has enough glycogen stores to last a few minutes, the glucose required by the brain to maintain viability must be provided by the circulation. The importance of the maintenance of adequate blood glucose levels has resulted in the evolution of insulin counterregulatory hormones, among which glucagon is critical. Impaired formation and action of glucagon results in hypoglycemia. Insulin and glucagon are physiologic antagonists; insulin removes glucose from the circulation by stimulating uptake of glucose into liver, muscle, and fat during meals, whereas glucagon stimulates the formation and release of glucose into the circulation, particularly

by the liver. Thus, in direct contrast to insulin secretion, glucagon secretion is increased during periods of fasting and is suppressed by elevated plasma levels of glucose. The balanced counteractions of insulin and glucagon maintain blood glucose levels in a relatively narrow range (~120 mg/dL during feeding and 60 mg/dL during fasting).

The most effective regulator of glucagon secretion is glucose, which suppresses glucagon secretion. However, certain amino acids produced by the digestion of proteins, such as arginine, stimulate glucagon secretion. Notably, carnivores, such as canines, have α-cells in their stomachs, suggesting an adaptive mechanism to provide glucagon early during the digestion of a high-protein meal to defend against insulin-induced hypoglycemia. Whether orally ingested fat affects glucagon secretion is unclear, although elevated plasma levels of free fatty acids reduce plasma concentrations of glucagon and inhibit the release of glucagon from the perfused rat pancreas. Several hormones regulate α-cell functions and coordinate the secretion of glucagon during changes in nutrient availability. These include the gastrointestinal hormones GLP-1 and GIP and the pancreatic-islet hormones insulin and somatostatin. In conditions of metabolic stress, such as hypoglycemia, additional hormones are released (e.g., growth hormone, catecholamines, glucocorticoids, and endorphins) all of which augment the secretion and actions of glucagon on peripheral tissues to ensure the maintenance of adequate blood glucose levels. Glucopenic stress also activates parasympathetic autonomic neural input to the islets that enhances glucagon secretion.

The intracellular signaling mechanisms within α-cells that culminate in the secretion of glucagon are complex. In situ hybridization studies of rat α-cells have shown the presence of the mRNAs encoding the K_{ATP} channel subunits Kir6.2 and SUR1.

Electrophysiologic studies of mouse α-cells in intact islets have determined that, although the expression of ion channels on α-cells is somewhat similar to that of β-cells and δ-cells, responses to glucose differ. α-Cells are distinguished from β-cells and δ-cells by the presence of a large tetrodotoxin-sensitive Na^+ current, a triethylamine-resistant K^+ current, and two kinetically separable Ca^{2+} currents: low- (T-type) and high-threshold (L-type) Ca^{2+} channels. In contrast to β-cells, α-cells are electrically silent in the presence of insulin-releasing glucose concentrations. The action potentials generated in the absence of glucose are inhibited by tetrodotoxin, nifedipine, and tolbutamide. These findings suggest that the electrical activity in and secretion of glucagon from α-cells is dependent on the generation of Na^+-dependent action potentials. Furthermore, the K_{ATP} channel opener diazoxide inhibits the electrical activity and increases the whole-cell K^+ conductance leading to glucagon secretion. Thus, glucagon secretion depends on a low activity of K_{ATP} to maintain sufficient negativity of membrane potential to prevent voltage-dependent inactivation of voltage-gated membrane currents. It was postulated that glucose inhibits glucagon release by depolarizing the α-cell, with resultant inactivation of the ion channels that participate in the generation of action potentials.

Metabolism and Degradation of Glucagon

Glucagon is cleared relatively rapidly from the circulation, with a half-life of about 5 minutes [metabolic clearance rate (MCR) ~10 $mL \cdot kg^{-1} \cdot min^{-1}$]. Both the kidney and the liver remove glucagon from the circulation, accounting for 30% and 20% of disposal, respectively. Notably, the remaining 50% of glucagon is destroyed in the circulation by enzymes, including serine and cysteine proteases, cathepsin B, and primarily DPP-IV, which cleave glucagon into proteolytic fragments. Not all cleavages of

glucagon are in the pathway for its disposal. Cleavage after arginines 17 and 18 by an endopeptidase isolated from rat liver membranes results in the formation of glucagon 19–29, so-called miniglucagon. Remarkably, miniglucagon was found to be a highly potent activator of Ca^{2+} channels in heart and liver cells. In heart cells it stimulates the accumulation of Ca^{2+} into sarcoplasmic reticular stores, which are targets for Ca^{2+} induced Ca^{2+} release by glucagon. Unlike glucagon, miniglucagon does not activate adenylyl cyclase. The cAMP-independent actions of miniglucagon in heart cells are mediated by the release of arachidonic acid, which acts synergistically with glucagon-induced formation of cAMP to trigger inotropic responses. There is also evidence that miniglucagon can inhibit glucose-induced insulin secretion from β-cells at picomolar levels. The physiologic roles of other potentially biologically relevant truncated glucagon peptides remain to be elucidated.

Physiologic actions of Glucagon

The most important physiologic action of glucagon occurs during the postabsorptive and fasting states. Through its actions on key enzymes, glucagon induces glycogenolysis, gluconeogenesis, and ketogenesis by the liver, and, to some extent, lipolysis in adipose tissue and glycogenolysis in muscle to mobilize stored energy. In the fasting state, the liver is the essential organ that provides glucose fuel to the brain. In humans the brain requires (utilizes) ~6 g of glucose per hour, whereas all other tissues utilize ~4 g of glucose per hour in the resting state. Collectively, the liver must provide ~10 g of glucose per hour to maintain euglycemia. The actions of glucagon on the liver account for ~75% of the glucose production in the fasting state. Additional fuel is derived

from the mobilization of fatty acids from adipose tissue metabolized in the liver to ketone bodies. This process is particularly important during periods of stress or starvation, as these ketones can be used by tissues, particularly the brain, to generate ATP. Other important mediators of this response include catecholamines, growth hormone, and glucocorticoids. The central nervous system senses glucopenia and, in response, triggers neural-sympathoadrenal hormones to counteract hypoglycemia. The ventromedial hypothalamus is proposed to be an important sensor of hypoglycemia and to initiate neural afferent signals to stimulate counterregulatory responses by way of the secretion of catecholamines, growth hormone, and glucocorticoids.

Glucagon is a major contributor to the immediate responses of the "fight-or-flight" circumstance. During times of sudden and intense physical effort, an instant increase in fuel for skeletal muscle is required without a decrease in fuel delivery to the brain. The skeletal muscles contain a limited supply of glycogen and lipids that can provide energy for the muscles for a short time. Sustained muscular activity requires an increase in circulating glucose and free fatty acids. Catecholamines play a critical role in these fight-or-flight circumstances during which increased fuel in the form of glucose and free fatty acids must be delivered to the circulation. Catecholamines stimulate glucagon secretion and reduce insulin levels during periods of stress and exercise. Notably, the reduction in insulin levels does not limit glucose uptake by exercising skeletal muscle but rather curtails glucose uptake by liver and fat, thereby protecting skeletal muscle from potential deleterious effects of a lowering of blood glucose levels. The increase in glucagon levels, stimulated by increased catecholamines, drives increased glycogenolysis

and gluconeogenesis in liver and lipolysis in fat to further augment circulating levels of glucose and free fatty acids.

Mechanisms of Glucagon action

The glucagon receptor belongs to group IV of the "B" family of seven membrane-spanning G-protein-coupled receptors. This family of receptors includes those for the hormones glucagon, GLP-1, GLP-2, GIP, vasoactive intestinal peptide (VIP), pituitary adenylyl activating peptide (PACAP), secretin, calcitonin, parathyroid hormone (PTH), and growth hormone releasing-hormone (GRH). A common feature of the glucagon receptor and these structurally related receptors is their coupling to the G-protein, G_s, that activates adenylyl cyclase and the resultant production of cellular cAMP. A major component of the cAMP-dependent signaling pathway is the activation of protein kinase A (PKA), an enzyme that phosphorylates and activates the functions of many different proteins in cells. PKA can activate proteins in the mitogen-activated protein kinase pathways by phosphorylation of Rap-1 that activates B-Raf and the downstream targets MEKKs (MAPK/ERK kinases), including ERK P-42, and ERK P-44. In addition, cAMP can bind directly to and activate a group of cellular mediators known as GEFs (guanine exchange factors). These additional pathways activate phosphoinositol 3-kinase and release of intracellular stores of Ca^{2+}. Thus, the intracellular signaling pathways mediated by the actions of glucagon on its target tissues are highly complex.

The expression of the glucagon receptor has been demonstrated in a large number of different tissues, including not only the generally recognized target tissues of liver, fat, and muscle but also the kidney, heart, lung, brain, intestine, adrenal gland, spleen, ovary,

thyroid, and the pancreatic islet α-, β-, and δ-cells. On the basis of these findings, it seems certain that glucagon has wide-ranging metabolic actions on the functions of many organs in the body. Within the pancreatic islets, the glucagon receptor is expressed in most β-cells and on a substantial subpopulation of the α- and δ-cells. It has been suggested that the glucagon receptors on β-cells may stimulate small amounts of insulin during periods of fasting to maintain the β-cells in a primed state awaiting a nutrient challenge and also may provide some insulin to peripheral tissues to facilitate glucose uptake during fasting.

Notably, the actions of glucagon may be regulated at the level of the expression of the glucagon receptor. In rats, expression of glucagon receptor in brown adipose tissue is downregulated following exposure to cold under the control of the sympathetic nervous system. The hepatic expression of glucagon receptor increases progressively from the first day of life to the adult stage in rodents. However, the expression can be increased under conditions in which intrahepatic glucose metabolism is activated, such as during fasting or in diabetes. Glucagon, acting through increased cellular levels of cAMP, is capable of downregulating the expression of hepatocyte glucagon receptors. Therefore, regulation at the level of the expression of receptors appears to be another means by which glucagon actions are modulated.

Relevance of glucagon in human diseases

It is generally believed that diabetes mellitus is a bihormonal disease consisting of an absolute or relative deficiency of insulin and a relative excess of glucagon. Insulin deficiency results in impairment in the utilization of glucose, and glucagon excess causes

an overproduction of glucose. Both circumstances contribute to the hyperglycemia in diabetic individuals. Therefore, inhibition of the actions of glucagon in diabetes is a rational approach to lowering blood glucose levels in individuals with diabetes. Theoretically, inhibition of glucagon actions could be achieved either by lowering blood glucagon levels or by antagonizing the actions of glucagon on the liver to reduce hepatic output of glucose. The former might be achieved by inhibiting the production/secretion of glucagon by α-cells or by neutralizing circulating glucagon. The latter could be accomplished by administering an effective glucagon-receptor antagonist. Such an antagonist should lower both fasting and postprandial glucagon levels and could theoretically be used in combination with insulin or with agents that increase insulin secretion (e.g., sulfonylureas), inhibit glucose absorption (α-glucosidase inhibitors), or enhance insulin action (thiazolidinediones).

Glucagon antagonists consisting of peptide analogues of glucagon have been shown to lower blood glucose levels in diabetic rodents. More recently, nonpeptidyl glucagon-receptor antagonists have been developed that may be more amenable to oral delivery. However, there are yet no proof-of-concept efficacy studies of glucagon antagonists in humans.

Excessive blood levels of glucagon are seen in rare instances of the development of neoplasms of the α -cells of the pancreas (glucagonomas). Glucagonomas are believed to originate from the islet progenitor cells in the pancreatic ducts. The clinical manifestations of glucagonoma, known as the “glucagonoma syndrome,” are quite distinct, consisting of glucose intolerance, weight loss, anemia, and migratory erythrodermatitis; 60% to 80% of glucagonomas are malignant and often metastasize to

the liver. The best success for treatment is surgical removal of the tumor from the pancreas before it has metastasized. Thereafter, chemotherapy is required.

Glucagon deficiency is occasionally a consequence of extensive damage of the pancreas, including the islets, caused by inflammatory or neoplastic disease. A panhormonal deficiency may occur in these circumstances. Deficiencies of both insulin and glucagon result in a type 1 insulin-dependent diabetes with a markedly enhanced sensitivity to administered insulin. Clear examples of hereditary α -cell (glucagon) deficiencies are quite rare. Two published cases of isolated glucagon deficiency describe individuals who manifested severe, intractable hypoglycemia incompatible with life. Other reports describe recurrent hypoglycemia and no detectable plasma glucagon by radioimmunoassay. Treatment with glucagon alleviated the hypoglycemia. Glucagon continues to be used therapeutically for acute treatment of hypoglycemia due to insulin overdose, so-called insulin shock. Glucagon also is given to individuals during gastrointestinal radiologic procedures because its actions of reducing gastrointestinal motility and relaxing and dilating the proximal and distal regions of the gut enhance double-contrast imaging

Regulation of secretion of glucagon-like peptides

GLP-1 and GLP-2 are produced in and secreted from the intestinal L cells located in the distal small intestine, colon, and rectum. Before the processes of formation of hormones and peptides by cleavages from proglucagon were understood, the intestinal products derived from the expression of the proglucagon gene in the intestine were known as GLIs (glucagon-like immunoreactivities), as they were measured in blood

plasma by various radioimmunoassays. The major intestinal GLI measured at that time was glicentin, a peptide consisting of the N-terminal sequence of proglucagon known as glicentin-related pancreatic peptide, glucagon, and the intervening peptide-1. Glicentin has no clearly defined biologic activities, and with the discovery of the structure of proglucagon, is generally believed to be a leftover, discarded product of proglucagon after cleavages of GLP-1s and GLP-2s have taken place. The development of radioimmunoassays specific for the detection of the GLPs (GLP-1 and GLP-2) has allowed physiologic assessment of the dynamics and factors controlling GLP secretion.

The regulation of the secretion of GLPs from the L cells is complex and appears to involve a combination of nutrient, hormonal, and neural stimuli. Numerous *in situ* luminal perfusion studies of isolated gut in experimental animals have shown that luminal nutrients, carbohydrate, fat, and to some extent amino acids stimulate the release of GLP-1 into the circulation. However, a paradox arose early in studies of the physiology of GLP-1 secretion (i.e., GLP-1 levels rise in the circulation as early as 15 minutes after a meal or an oral glucose tolerance test, a time much too short for nutrients to reach the distal intestine and colon and mediate luminal stimulation of GLP-1 secretion). The existence of signals arising from the proximal gut to the L cells of the distal gut was proposed to account for the rapid release of GLP-1 in response to ingestion of nutrients. Indeed, the presence of nutrients in the duodenum of rats can stimulate GLP-1 release, a process that seems to involve the enteric vagal nervous system as well as the duodenal hormone GIP. Thus, L cells that release GIP sense fat and/or glucose in the duodenum, which in turn activates a vagal pathway that ultimately leads to GLP-1 release. Enteric nerves originating in the duodenum may also carry the nutrient signal to the distal L cells.

Given the important actions of GLP-1 in regulating glucose homeostasis, studies have been done to determine whether subjects with type 2 diabetes have impaired GLP-1 secretion. In patients with type 2 diabetes, insulin release is not stimulated more by an oral than by an isoglycemic intravenous glucose load, indicating a loss of incretin-mediated stimulation of insulin secretion. However, this issue remains unclear, as there are reports of both decreased and increased blood levels of GLP-1 in individuals with type 2 diabetes. relative to levels in controls. Additional studies are required to clarify these apparent discrepancies. Perhaps approaches that increase the endogenous production of GLP-1 have the therapeutic benefit of improving glucose tolerance. Thus, further investigation into the mechanisms controlling the release of GLP-1 in humans also is required.

Metabolism and degradation of glucagon-like peptides

The rate of removal of GLPs from the circulation is an important determinant of their biologic actions. GLPs are eliminated by at least three processes: renal extraction, hepatic extraction, and proteolytic inactivation within the circulation. The kidney removes GLP-1 from the circulation by a mechanism that involves glomerular filtration and tubular catabolism. Thus, circulating levels of GLP-1 are elevated in patients with renal failure and in nephrectomized rats. Hepatic extraction of GLP-1 also contributes to the clearance of GLP-1. The MCR (the least amount of plasma totally cleared of GLP-1 per unit time) in humans is ~10 mL. kg^{-1}. min^{-1}, providing a half-life of about 5 minutes. However, the half-life of biologically active GLP-1 is 1 to 2 minutes because of its inactivation by proteolytic cleavage in the circulation. The biologically active forms of

GLP-1, GLP-1 (7–36) amide and GLP-1 (7–37) are rapidly cleaved by DPP-IV (CD26) that removes the N-terminal dipeptide, His-Ala, resulting in the formation of the believed-to-be inactive GLP-1 (9–36) amide and GLP-1 (9–37) isopeptides, respectively. Likewise, GLP-2 is rapidly inactivated by DPP-IV through the conversion of GLP-2 (1–33) to GLP-2 (3–33). Therefore, DPP-IV-mediated inactivation of both GLP-1 and GLP-2 is a critical determinant of the biologic activities of these hormones.

Physiologic Actions of Glucagon-like Peptide-1(GLP-1):

GLP-1 exerts several distinct physiologic actions, reflecting the different organ systems in which the GLP-1 receptor is expressed. Receptors have been identified on the pancreatic islet cells (α, β, and δ), stomach, brain, lung, intestine, heart, kidney, and anterior pituitary. There are some findings, predominantly from in vitro studies, of GLP-1 actions on liver, fat, and muscle in which GLP-1 stimulates glycogenolysis and lipogenesis. Although GLP-1 binding sites on these tissues appear to exist, attempts to identify expression of the GLP-1 receptor mRNA in these tissues have been unsuccessful. This circumstance suggests that an additional GLP-1 receptor(s) may exist. Considerable understanding has occured about the physiologic relevance of the actions of GLP-1 on pancreatic islets, stomach, and brain. Understanding of the physiologic responses of GLP-1 actions on lung, intestine, heart, kidney, and anterior pituitary are incomplete.

The first physiologic action of GLP-1 to be elucidated, after the initial discovery that GLP-1 is a split product of proglucagon derived in the intestinal L cells, was that it is a glucose-dependent stimulator of insulin secretion. GLP-1 is a potent intestinal incretin hormone that acts in concert with GIP to augment glucose-dependent insulin secretion in response to oral nutrients. Although both GLP-1 and GIP augment glucose-mediated

insulin secretion, only GLP-1, and not GIP, appears to retain insulinotropic activity in hyperglycemic individuals with type 2 diabetes. The mechanistic basis for this difference between the dual incretin hormones GLP-1 and GIP remains unknown. Not only is the stimulation of insulin secretion by GLP-1 dependent on a threshold level of plasma glucose (~ >60 mg/dL) but the actions of glucose to stimulate insulin secretion require GLP-1 or other factors that stimulate cAMP formation in β-cells. The interdependence of the dual signaling by glucose metabolism and cAMP generation, such as provided by GLP-1, is described as the "glucose competence concept". The successful achievement of nutrient-stimulated insulin secretion by β-cells depends on the concomitant activations of cAMP and glucose metabolism-driven signaling pathways.

In addition to stimulating insulin secretion, GLP-1 stimulates the biosynthesis of proinsulin and the transcription of the proinsulin gene. In this manner, GLP-1 contributes to repleting stores of insulin lost from β-cells by secretion. The GLP-1 property of stimulating the formation of insulin distinguishes it from the sulfonylurea drugs, which stimulate the secretion but not the biosynthesis of insulin.

MATERIALS AND METHODS

Source of data

All patients coming to BLDE's Shri.B.M. Patil Medical college, Hospital and Research Center, Bijapur , between October 2008 to March 2010, with one or more risk factors for diabetes mellitus as per ADA 2008 guidelines[10] .

Sample size

As the incidence of Impaired fasting glucose is 4.61%5 and 95% confidence, level, margin of error being 4% the worked out sample size is 130.Using statical formula $(n)=4pq/L^2$

Statistical analysis

a) Diagnostic presentation

b) Mean- standard deviation

Method of collection of data

1. The study was carried out on patients coming to B.L.D.E.U's Shri B.M. Patil Medical College Hospital and Research Center, Bijapur. Detailed history was taken with special regard to the risk factors for diabetes mellitus, and general and systernic examination was carried out and they were also subjected to certain investigations to determine some risk factors like dyslipidemia, ischemic heart disease etc.

The investigations done were

a. Fasting blood sugar and post prandial blood sugar

b. Lipid profile

c. Electrocardiogram

2. Individuals selected by inclusion criteria undergwent fasting glucose analysis of venous sample and sample was tested by glucose oxidase peroxidase method.

Inclusion criteria

1. First degree relative with diabetes.

2.Overweight (BMI > 25 Kg/m^2)

3. Age >45 YEARS

4. Dyslipidemia (HDL Cholesterol < 35mg/dl or Triglyceride > 250 mg /dl or both.

5. Hypertension (140/90 mm of hg or on therapy for hypertension).

6. History of gestational diabetes mellitus or delivery of a baby over 4. l kg.

7. Race or ethnicity with high risk of diabetes.

8. Physical inactivity: World Health Organization Criteria was applied to define physical inactivity , which says:

   Light work : 75% of time spent sitting and 25% of time spent working.

   Moderate work : 40% of time spent sitting and 60% of time spent working.

   Heavy work : 25% of time spent sitting and 75% time spent working.

9. Clinical conditions associated with insulin resistance (Severe obesity, acanthosis nigricans).

10. History of cardiovascular disease.

    - History suggestive of hypertensive heart disease
    - History suggestive of ischemic heart disease

11. Women with polycystic ovarian syndrome.

Exclusion criteria

1. Patients who are already diagnosed cases of diabetes mellitus by to ADA 2008 criteria.
2. Patients who are on drugs causing hyperglycemia.

Diagnostic criteria

Diagnostic criteria for Impaired fasting glucose or Impaired glucose tolerance depends whether it is identified through the fasting plasma glucose or the oral glucose tolerance test. :

- Impaired fasting glucose = fasting plasma glucose between 100 mg/dl (5.6 mmol/1) to 125 mg/dl (6.9 mmol/1).
- Impaired glucose tolerance = two hour plasma glucose between 140 mg/dl (7.8 mmol/1) to 199 mg/dl (11 mmol/1)

Impaired fasting glucose and Impaired glucose tolerance have been officially termed as Prediabetes.

Study design

The study design consisted of 130 cases having one or more risk factors for diabetes mellitus and screened for fasting venous blood glucose and were categorised between normal and Impaired fasting glucose patients. The Impaired fasting glucose criteria was according to ADA criteria 2008.

Collection of blood sample

About 10ml of blood is collected in morning, after an over night fasting for 8-12 hrs.2ml of blood is collected into bottle in which heparin is added as an anticoagulant.The remaining 8ml is taken into another bottle and is allowed to clot.

Precautions:

Usage of sterile needles and syringes, cleaning of patients skin, blood collection in clean and dry tubes.

Serum:

Serum is supernatant fluid that can be collected after centrifuging the clotted blood. It is most frequently used.

Heparin:

Anticoagulant which inhibits the conversion of prothrombin to thrombin. The following investigations were done on the same day.

1. From the heparinised blood, the following investigation is carried out.

- Fasting blood sugar (FBS).
- Postprandial blood sugar (PPBS)

2. From the whole blood, serum is separated and the following biochemical investigations were carried out.

    - Total cholesterol.
    - HDL Cholesterol.
    - Serum Triglycerides.
    - VLDL and LDL are calculated.

Determination of blood glucose:

[Glucose Oxidase Peroxidase Method]

AIM:

To estimate the serum glucose by Glucose Oxidase Peroxidase Method.

Principle:

Glucose is oxidized to glucuronic acid and hydrogen peroxide in the presence of glucose Oxidase. Hydrogen peroxide further reacts with phenol and 4 – amino antipyrine by the catalytic action of peroxidase to form a red coloured quinoneimine dye complex. Intensity of the colour formed is directly proportional to the amount of glucose present in the sample. The assay depends on.

- Oxidation of D – glucose to D – glucoronic acid.
- Colorimetric estimation of H_2O_2 formed.

$$\text{Glucose} + O_2 + H2O \xrightarrow[\text{Oxidase}]{\text{Glucose}} \text{Gluconic acid} + H_2O_2$$

$$H_2O_2 + \text{Phenol} + \text{4 Amino antipyrine} \xrightarrow{\text{Peroxidase}} \text{Red Quinoneimine} + H_2O$$

Specimen:

Whole blood collected with heparin as an anticoagulant.

Reagents:

0.2 M phosphate buffer P^H 7.4

Buffer solution A:

0.2 M monobasic sodium dihydrogen phosphate ($NaH_2Po_42H_2O$) is prepared by dissolving 7.8 gm in 250 ml distilled water.

Buffer Solution B:

0.2 M dibasic disodium hydrogen phosphate (Na_2Hpo_4) is prepared by dissolving 8.9 gm in 250 ml of water.

The two solutions are mixed as follows and P^H is adjusted to 7.4. (36.4ml of NaH_2Po_4. $2H_2O$ + 213.6ml of $Na_2Hpo_4.2H_2O$).

Colour reagents

4 amino antipyrine (0.01% - 20mg / 200ml)

Horse raddish peroxidase (0.73 IU / ml – 1 mg/200ml).

Glucose Oxidase – (30 IU or 22 mg / 200 ml)

Dissolve the solutions in 200ml of buffer. To this add 100mg of phenol (0.05%) store the reagent in brown coloured glass bottle in refrigerator.

Glucose standards

1mg of glucose is dissolved in 100ml distilled water for storage, dissolve 200 mg of benzoic acid (0.2% benzoic acid) in 100 ml water + 1 mg of glucose.

Procedure

Addition sequence	B (ml)	S (ml)	T (ml)
Glucose reagent	1.0	1.0	1.0
Distilled water	0.01	-	-
Glucose standard	-	0.01	-
Serum sample	-	-	0.01

Mixed well and this is allowed to stand for 10 minutees incubation at 37^0C. Measure the absorbance of the Standard(S), Test (T), Blank (B) within 60 minutes.

Measurements

Colour : Red

Wavelength : 505nm

Incubation : 10 min

Sample Volume : 0.01 ml

Reagent Volume : 1 ml

Units : mg/dl

Sample required : Serum Sample

Calculations

$$\text{Total Glucose in mg/dl} = \frac{\text{Abs T}}{\text{Abs S}} \times 100$$

Normal values:

Fasting Blood Sugar = 70 – 100 mg/dl

Post Prandial (2hr) = Upto 150 mg/dl

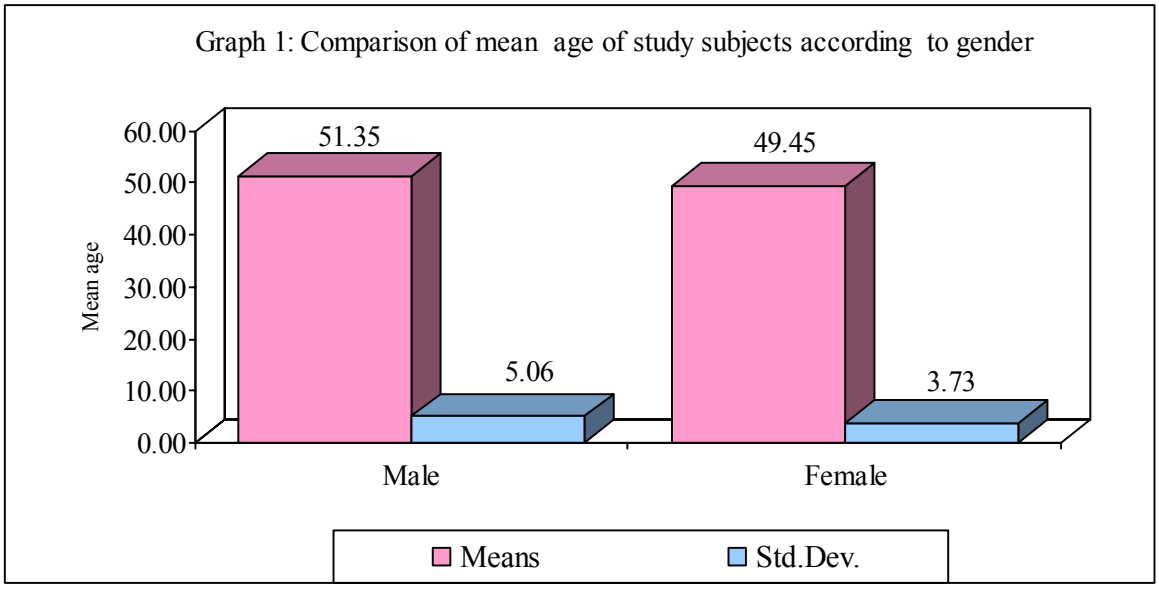
Clinical significance

Glucose is major carbohydrate present in blood. Its oxidation in the cells is the source of energy for the body. Increased levels of glucose are found in diabetes mellitus, hyperparathyroidism, pancreatitis, renal failure. Decreased levels are found in insulinoma, hypothyroidism, Hypopituitarism and extensive liver disease.

OBSERVATION AND RESULTS

Table 5: Mean and SD age of study subjects according to gender

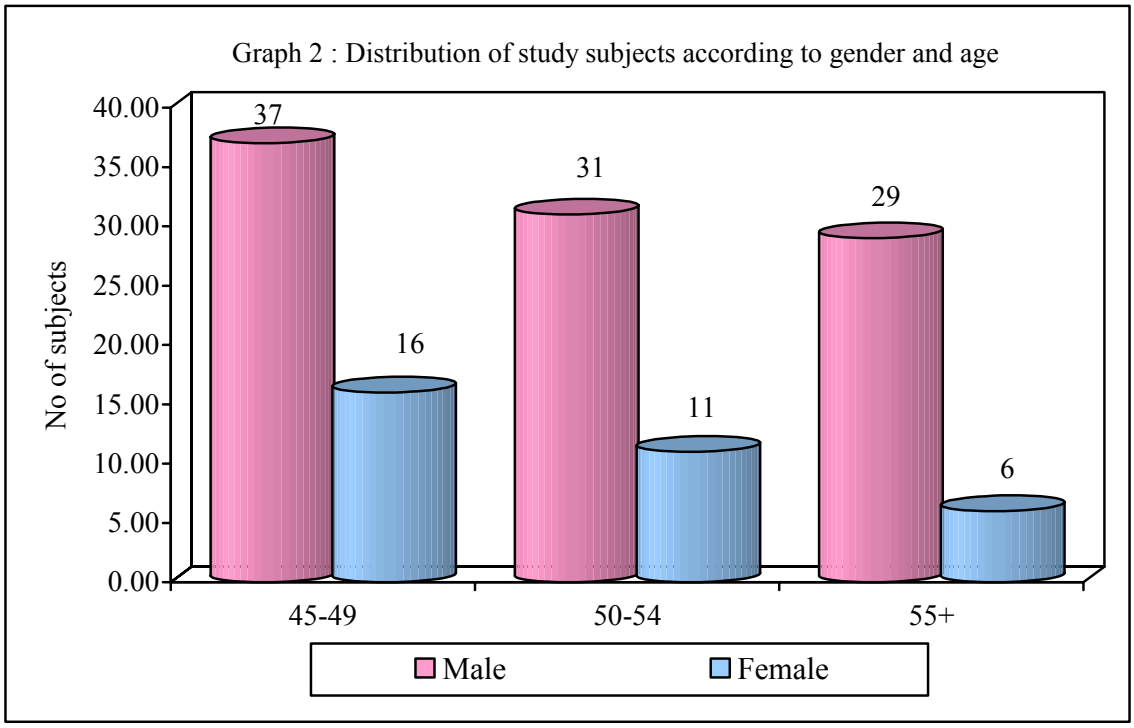
Gender	Mean	Standard deviation(SD)
Male	51.35	5.06
Female	49.45	3.73
Total	50.87	4.81



The mean and standard deviation of subjects acoording to gender,showing a mean of 51.35 in males and 49.45 in females,with standard deviation of 5.06 for males and 3.73 in females.

Table 6 : Distribution of study subjects according to gender and age.

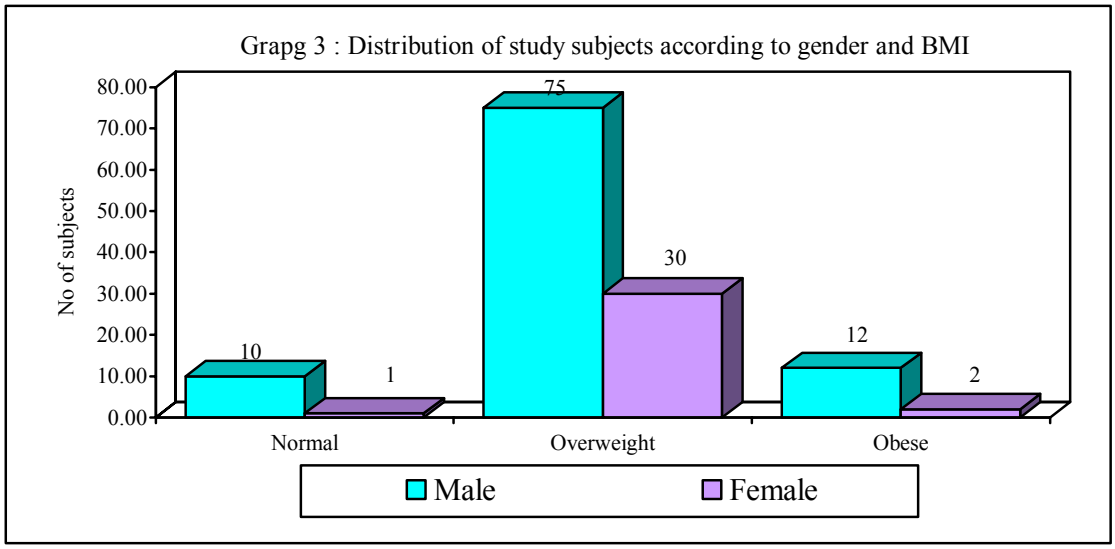
Age (in yrs)	Male	%	Female	%	Total	%
45-49	37	69.81	16	30.19	53	40.77
50-54	31	73.81	11	26.19	42	32.31
55+	29	82.86	6	17.14	35	26.92
Total	97	74.62	33	25.38	130	100.00



There were 97 males and 33 females in the study population,In males 37 were in the age group 45-49 years,31 were in the age group of 50-54 years and 29 were more than 55 years.In the female group 16 were in the age group of 45-49 years,11 were in the age group of 50-54 years and 6 were more than 55 years.

Table 7 : Distribution of study subjects according to gender and BMI

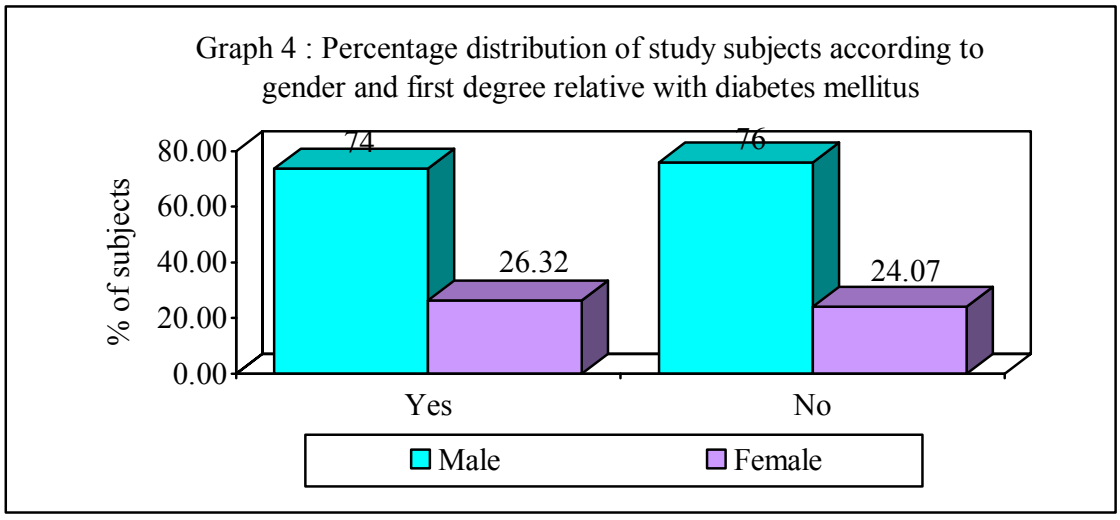
BMI	Male	%	Female	%	Total	%
Normal	10	90.91	1	9.09	11	8.46
Overweight	75	71.43	30	28.57	105	80.77
Obese	12	85.71	2	14.29	14	10.77
Total	97	74.62	33	25.38	130	100.00
Chi-square=3.0150 df=2 p=0.22144						



Out of 97 males,75 were overweight ,12 were obese and 10 had normal BMI,in females out of 33 females,30 were overweight,2 were obese and 1 female had normal BMI.

Table 8 : Distribution of study subjects according to gender and first degree relative with diabetes mellitus.

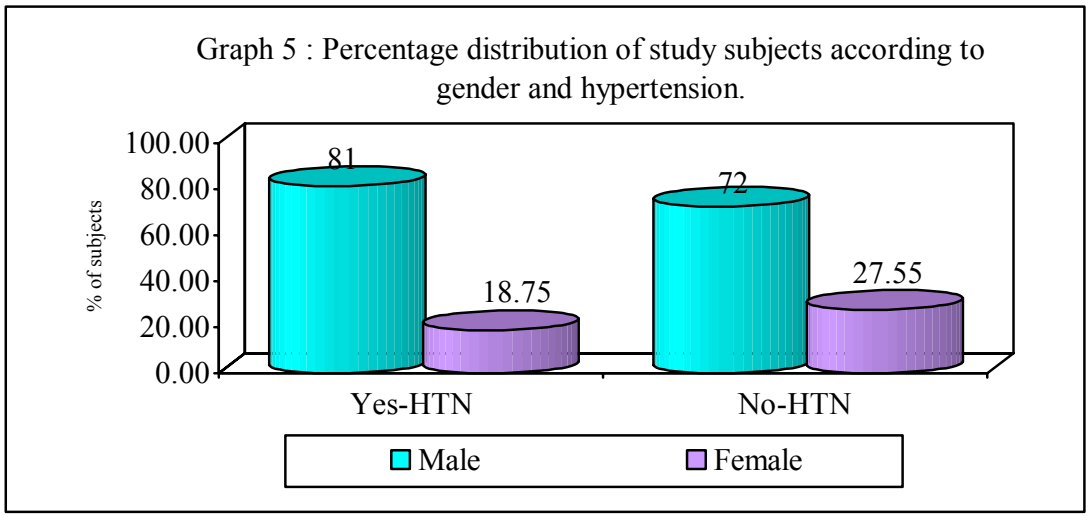
First degree relative With diabetes mellitus.	Male	%	Female	%	Total	%
Yes	56	73.68	20	26.32	76	58.46
No	41	75.93	13	24.07	54	41.54
Total	97	74.62	33	25.38	130	100.00
Chi-square=00840 df=1 p=0.77227						



Out of 97 male individuals ,56 males had positive family history of first degree relative with diabetes mellitus,and in females,20 individuals had positive family history.

Table 9 : Distribution of study subjects according to gender and Hypertension.

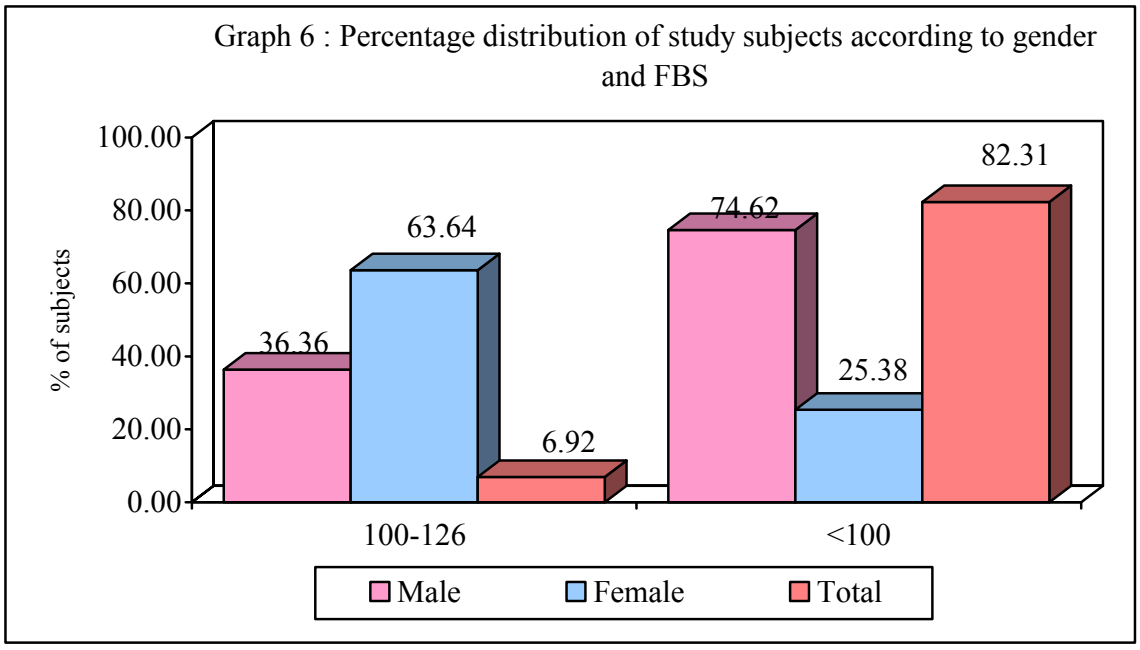
HTN	Male	%	Female	%	Total	%
Yes-HTN	71	81.25	27	18.75	32	24.62
No-HTN	26	72.45	6	27.55	98	75.38
Total	97	74.62	33	25.38	130	100.00
Chi-square=1.0330 df=1 p=0.30954						



Out of 97 male individuals ,71 were hypertensives and among 33 female individuals,27 were hypertensive individuals.

Table 10: Distribution of study subjects according to gender and FBS

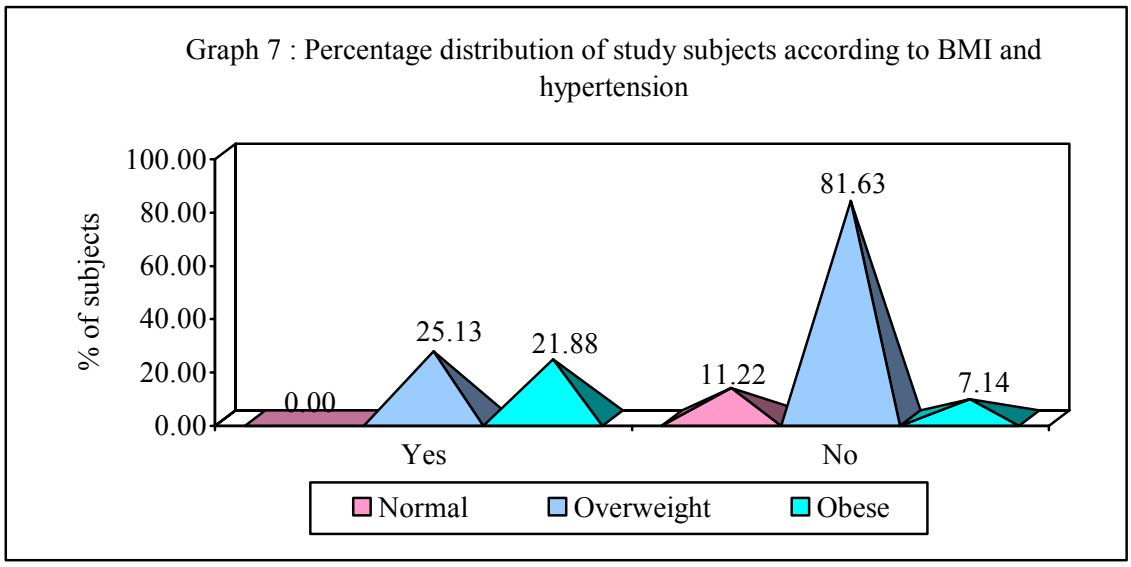
FBS	Male	%	Female	%	Total	%
100-126	4	36.36	7	63.64	11	8.46
<100	93	78.15	26	21.85	119	91.54
Total	97	74.62	33	25.38	130	100.00
Chi-square=9.2830 df=1 p=0.00231, S						



Out of 97 males and 33 females,4 males had Impaired fasting glucose and 7 females had Impaired fasting glucose.

Table 11:Relation between BMI and Hypertension of study subjects

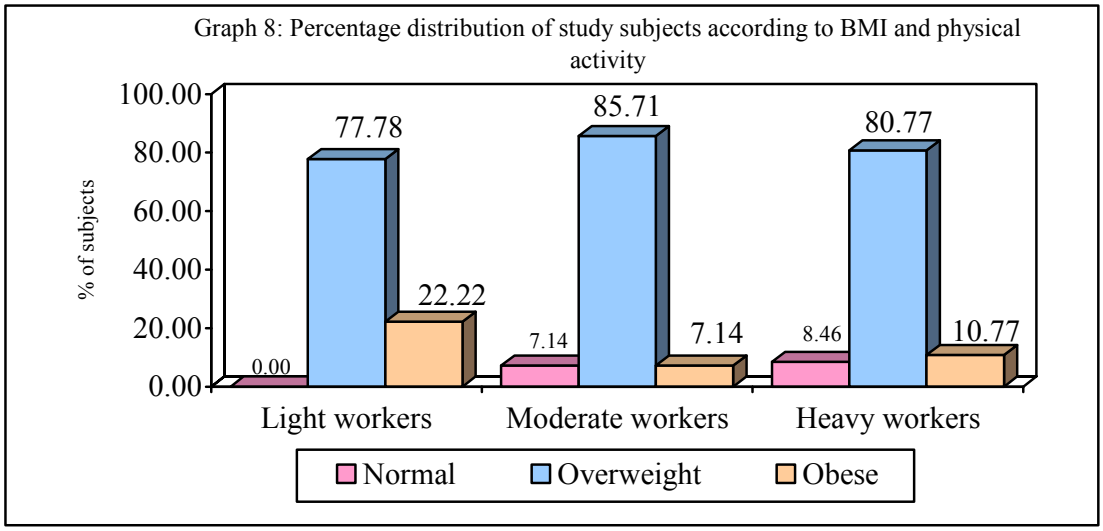
HTN	Normal	%	Overweight	%	Obese	%	Total	%
Yes	0	0.00	80	81.63	7	21.88	32	24.62
No	11	11.22	25	25.13	7	7.14	98	75.38
Total	11	8.46	105	80.77	14	10.77	130	100.00
Chi-square=8.4901 df=2 p=0.01434, S								



With normal BMI one was hypertensive,among 105 overweight individuals,25 were hypertensives and among 14 obese individuals,7 were hypertensive individuals.

Table 12 Relationship between BMI and physical activity of study subjects

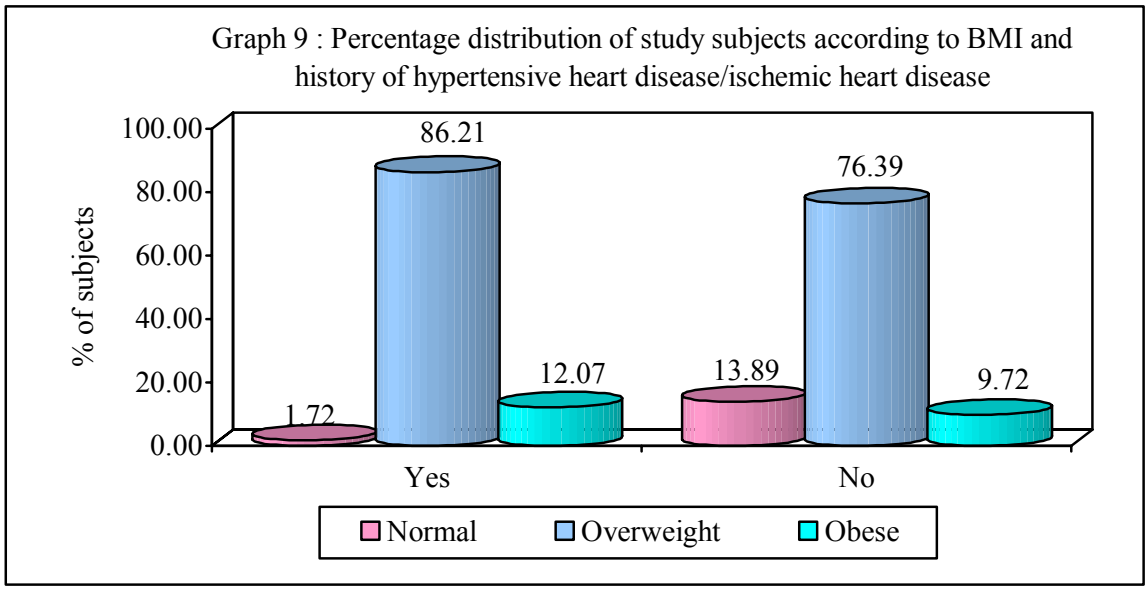
Physical activity	Normal	%	Overweight	%	Obese	%	Total	%
Light workers	0	0.00	7	77.78	2	22.22	9	6.92
Moderate workers	10	9.35	86	80.37	11	10.28	107	82.31
Heavy workers	1	7.14	12	85.71	1	7.14	14	10.77
Total	11	8.46	105	80.77	14	10.77	130	100.00
Chi-square=2.2351 df=4 p=0.69271								



Out of 11 individuals with normal BMI,10 were moderate workers and 1 was a heavy worker.With overweight individuals,7 were light workers,86 individuals were moderate workers and 12 individuals were heavy workers.Among obese individuals,1 was light worker,11 were moderate worker and 1 individual was heavy worker.

Table 13:Relationship between BMI and history of hypertensive heart disease/ischemic heart disease of study subjects.

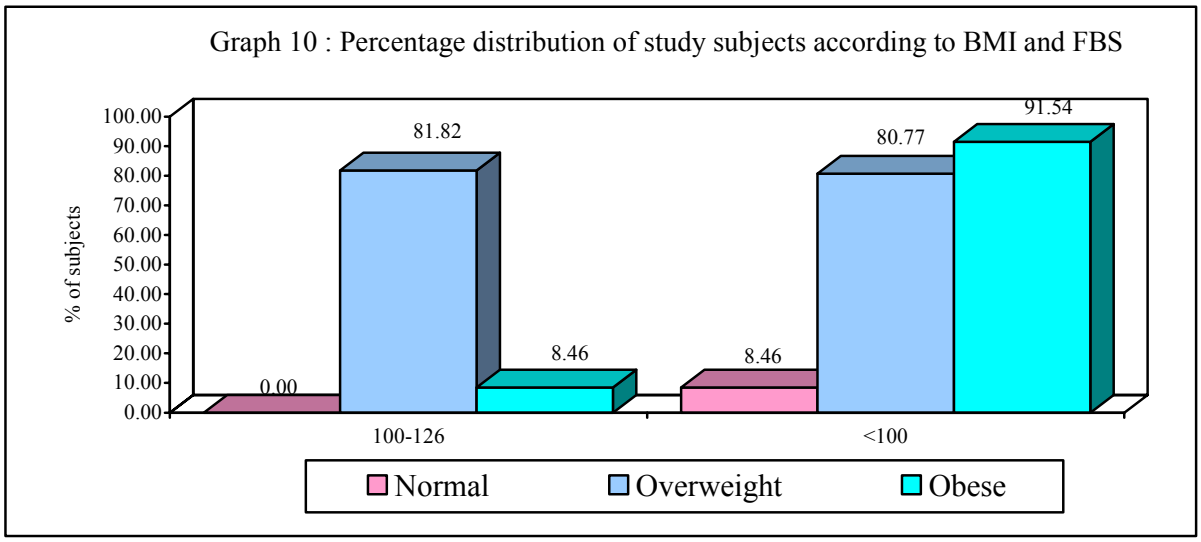
History of heart disease	Normal	%	Overweight	%	Obese	%	Total	%
Yes	1	1.72	55	86.21	7	12.07	58	44.62
No	10	13.89	50	76.39	7	9.72	72	55.38
Total	11	8.46	105	80.77	14	10.77	130	100.00
Chi-square=6.1660 df=2 p=0.04585, S								



Among individuals with normal BMI,only 1 individual had history of hypertensive/ischaemic heart disease,and in overweight and obese group,there were 55 and 7 individuals with history of hypertensive/ischaemic heart disease respectively.

Table 14:Relationship between BMI and FBS of study subjects

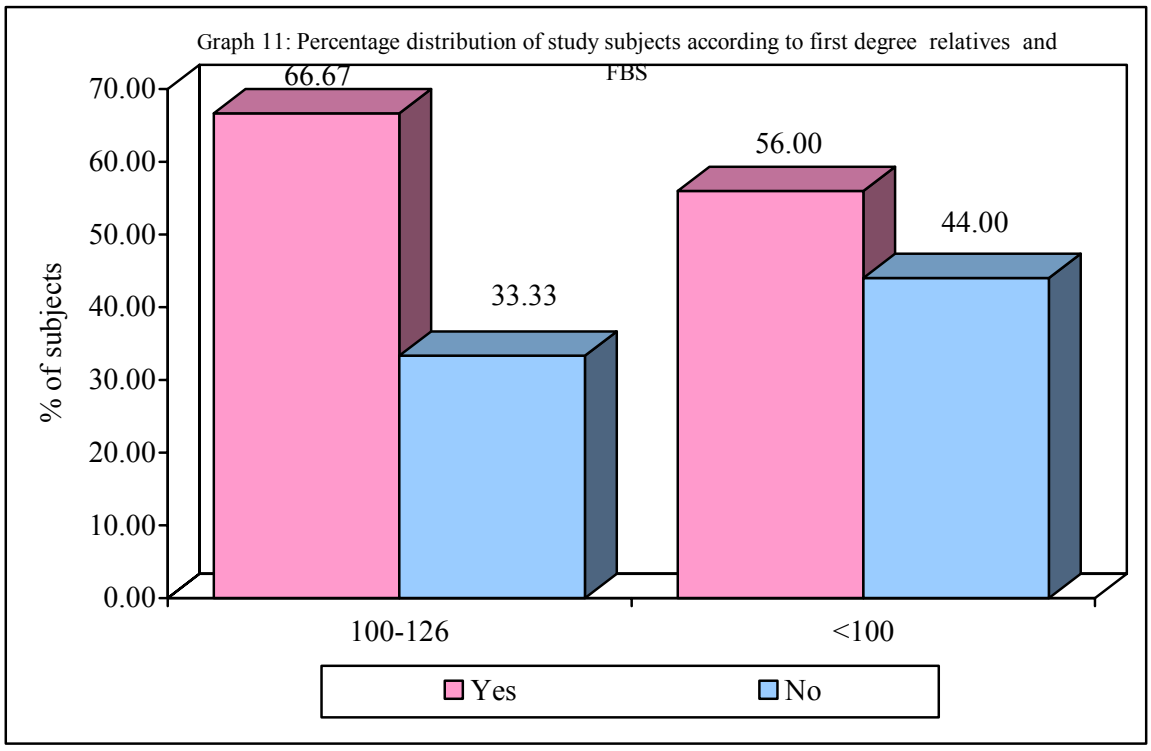
FBS	Normal	%	Overweight	%	Obese	%	Total	%
100-126	0	0.00	9	81.82	2	18.18	11	8.46
<100	11	9.24	96	80.67	12	10.08	119	91.54
Total	11	8.46	105	80.77	14	10.77	130	100.00
Chi-square=1.6320 df=2 p=0.44230								



Out of 11 individuals with Impaired fasting glucose,9 individuals were overweight and 2 individuals were obese.

Table 15:Relationship between first degree relatives with diabetes mellitus and FBS

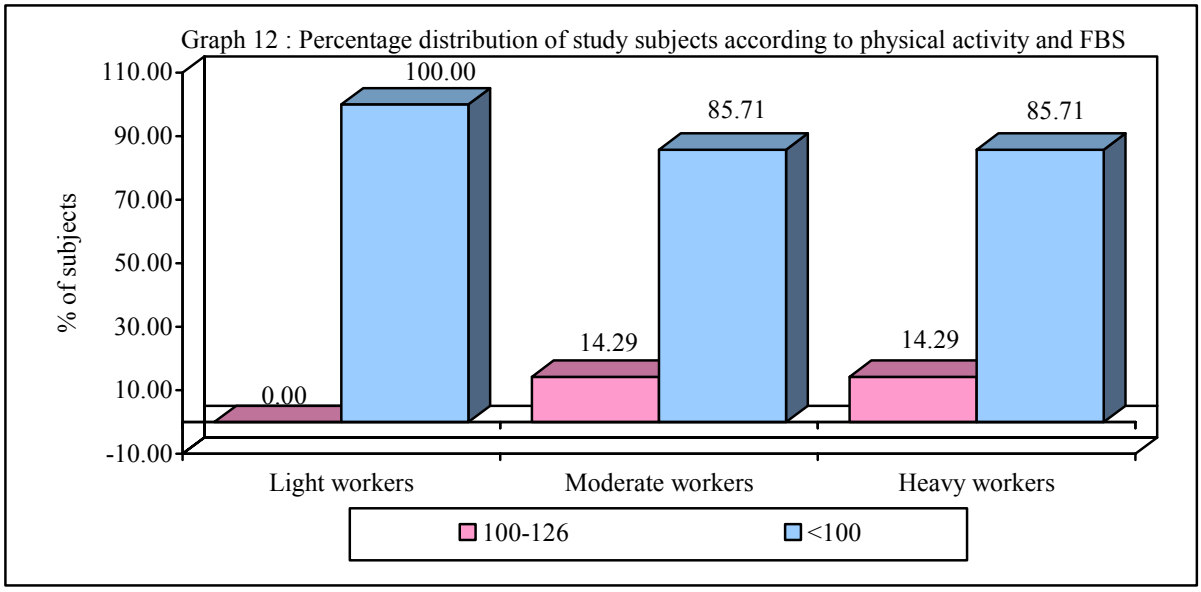
FBS	Yes	%	No	%	Total	%
100-126	6	54.55	5	45.45	11	8.46
<100	70	58.82	49	41.18	119	91.54
Total	76	58.46	54	41.54	130	100.00
Chi-square=0.0761 df=1 p=0.7829						



Out of 11 individuals with Impaired fasting glucose,6 individuals had positive family history of diabetes mellitus in immediate first degree relatives.

Table 16:Relationship between physical activity and FBS of study subjects

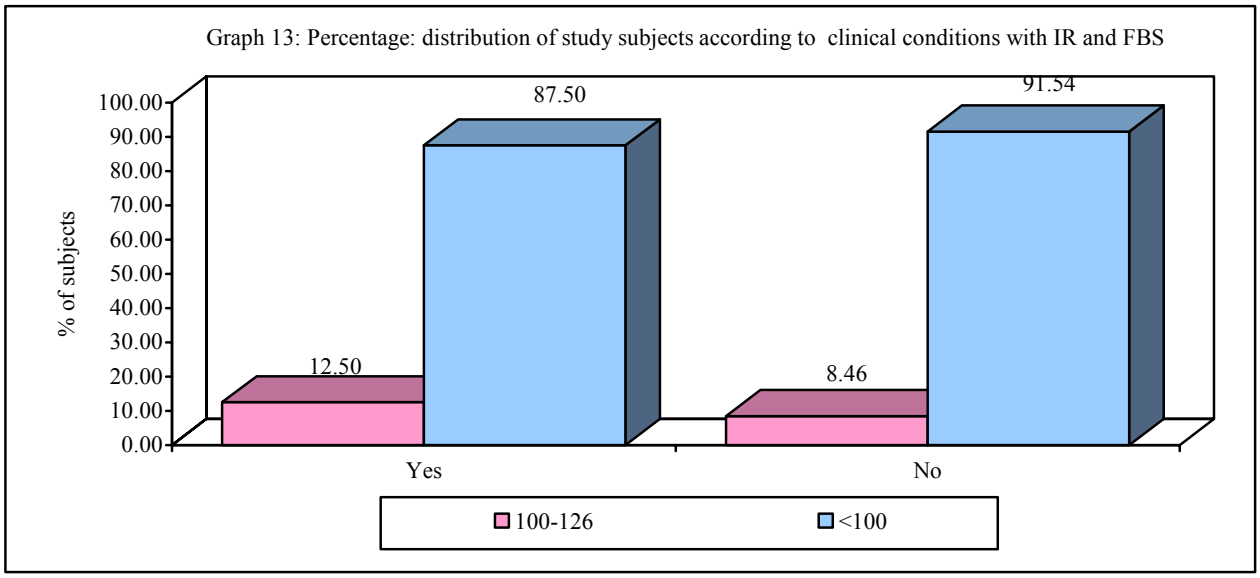
Physical activity	100-126	%	<100	%	Total	%
Light workers	0	0.00	9	100.00	9	0
Moderate workers	9	8.41	98	91.59	107	9
Heavy workers	2	14.29	12	85.71	14	2
Total	11	8.46	119	91.54	130	11
Chi-square=1.4450 df=2 p=0.48545						



Out of 11 individuals with Impaired fasting glucose,9 were moderate workers and 2 individuals were heavy workers.

Table 17:Relationship between clinical condition associated with Insulin resistance (IR) and FBS

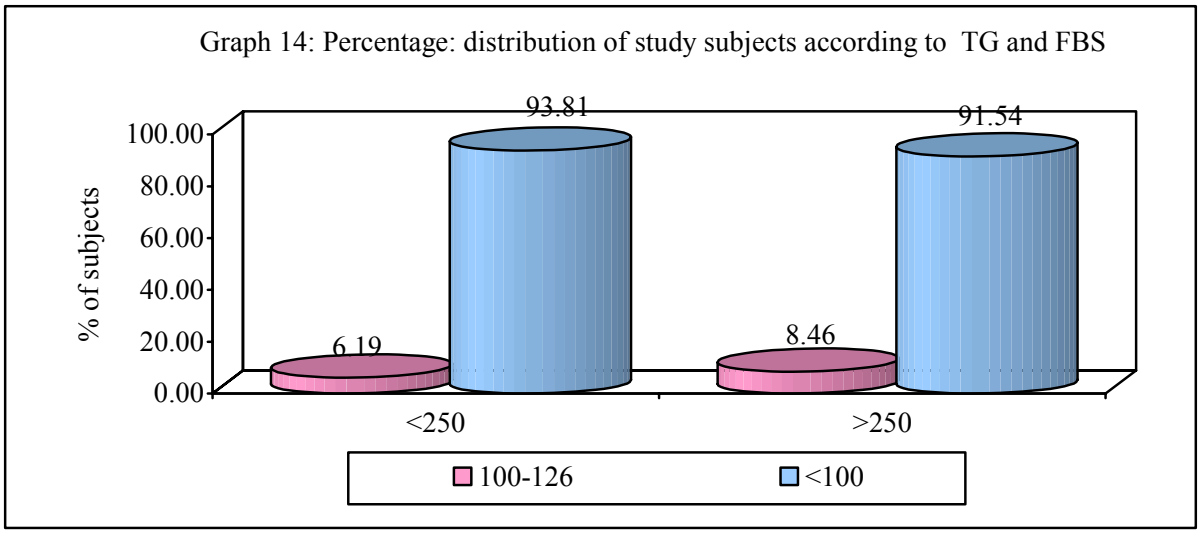
Clinical condition IR	100-126	%	<100	%	Total	%
Yes	1	12.50	7	87.50	8	6.15
No	10	8.20	112	91.80	122	93.85
Total	11	8.46	119	91.54	130	100.00
Chi-square= 0.1790 df=1 p=0.67181						



Out of 11 individuals with Impaired fasting glucose,only 1 individual had,clinical condition with insulin resistance.

Table 18:Relationship between Triglyceride (TG) and FBS:

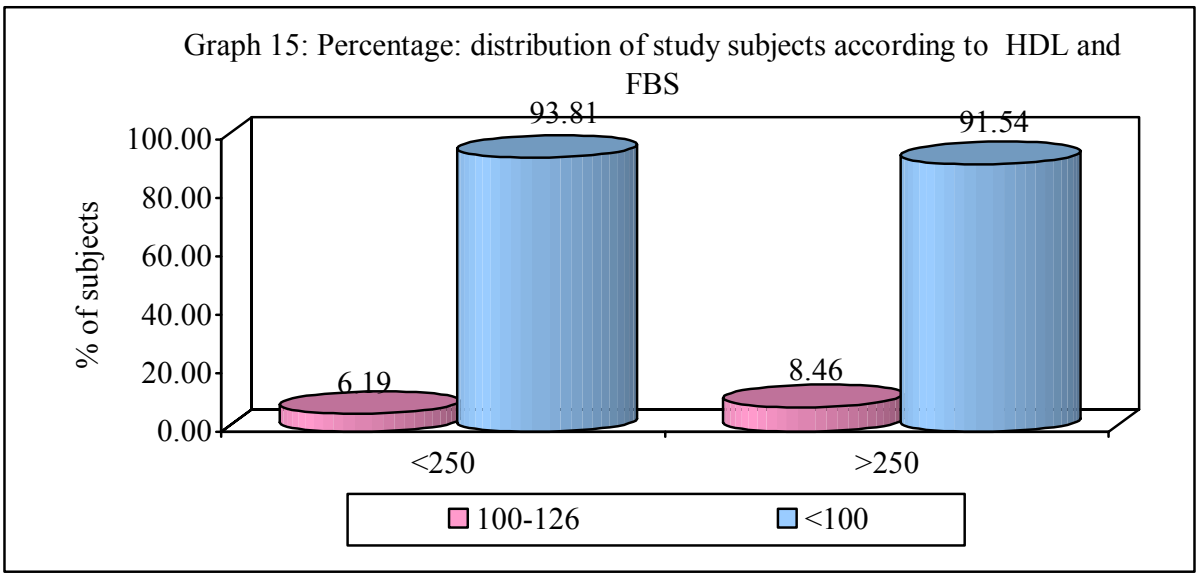
TG	100-126	%	<100	%	Total	%
<250	5	6.19	91	93.81	97	74.62
>250	6	15.15	28	84.85	33	25.38
Total	11	8.46	119	91.54	130	100.00
Chi-square=0 .1192 df=1 p=0.72993						



Out of 11 individuals with Impaired fasting glucose,6 individuals had triglyceride level >250 and 5 individuals had triglyceride level < 250 mg/dl.

Table 19:Relationship between HDL and FBS

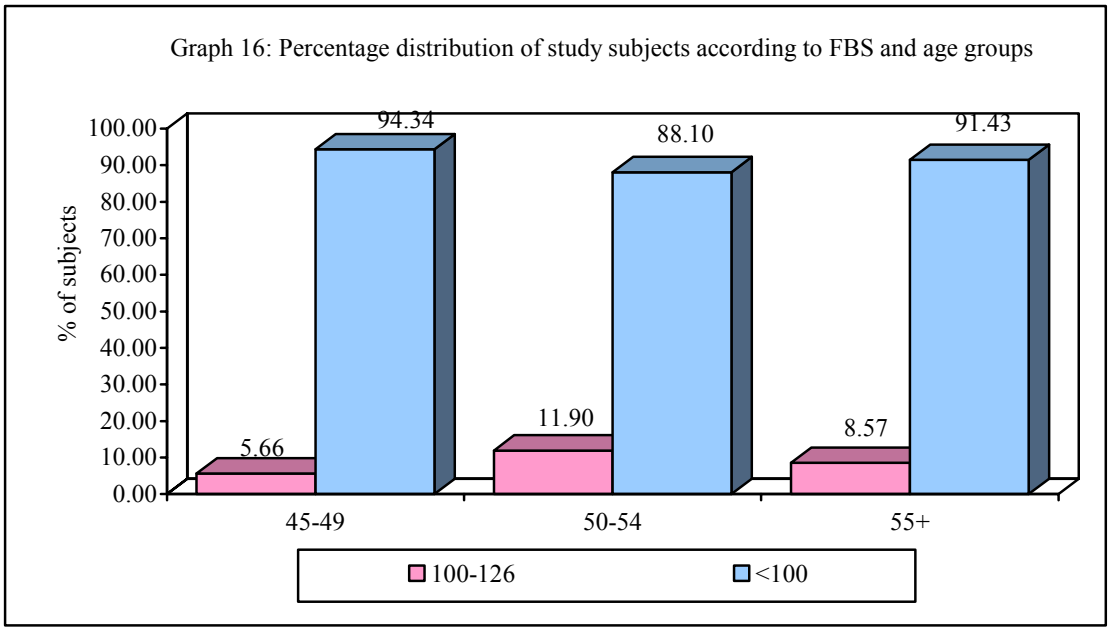
HDL	100-126	%	<100	%	Total	%
<35	7	12.07	68	94.44	72	55.38
>35	4	5.56	51	87.93	58	44.62
Total	11	8.46	119	91.54	130	100.00
Chi-square= 1.7590 df=1 p=0.18470						



Out of 11 individuals with Impaired fasting glucose,4 individuals had HDL level >35 mg/dl and 7 individuals had HDL level < 35 mg/dl.

Table 20: Table showing relationship between FBS and age group.

Age (inyrs)	100-126	%	<100	%	Total
45-49	3	5.66	50	94.34	53
50-54	5	11.90	37	88.10	42
55+	3	8.57	32	91.43	35
Total	11	8.46	119	91.54	130
Chi-square= 1.1801 df=2 p=0.55424					



Out of 11 individuals with Impaired fasting glucose,3 were in age group 45-49 years,5 individuals were in 50-54 years age group and 3 individuals were in age group > 55 years.

Table 21:Comparision of Impaired fasting glucose and normal fasting glucose with respect to various other parameters by independent t test

Variable	FBS	n	Mean	SD	t-value	p-value
BMI	100-126	11	27.9945	4.3572	1.2093	0.2288
	<100	119	27.0808	2.1509		
PPBS	100-126	11	132.1818	18.0378	-0.1084	0.9139
	<100	119	132.9664	23.3390		
TG	100-126	11	143.5455	149.2001	0.5982	0.5508
	<100	119	131.3445	51.5559		
TC	100-126	11	188.7273	34.5372	-0.1076	0.9145
	<100	119	194.0672	163.7639		
HDL	100-126	11	36.1273	8.1497	0.2663	0.7904
	<100	119	35.4118	8.5573		
LDL	100-126	11	118.8727	42.9247	0.4641	0.6433
	<100	119	113.3361	37.3902		
VLDL	100-126	11	31.3636	28.3901	0.5003	0.6178
	<100	118	28.8322	14.5178		

Table22:Correlations among risk factors for Impaired fasting glucose patients:

Risk factors	Age	BMI	First degree relative with DM	Phys1cal act1v1ty	FBS	PPBS	TG	TC	HDL	LDL	VLDL
Age	1.0000										
BMI	0.0565	1.0000									
First degree relative with DM	0.1174	-0.0666	1.0000								
Phys1cal act1v1ty	-0.1469	-0.1793*	0.0344	1.0000							
FBS	0.0961	0.1056	-0.0869	-0.2004*	1.0000						
PPBS	0.1055	0.1254	-0.0552	-0.1769*	0.5884*	1.0000					
TG	-0.1196	0.0391	-0.0003	0.0728	0.2509*	0.1750*	1.0000				
TC	-0.0482	0.0142	-0.1247	0.0330	0.1574	0.1522	0.0885	1.0000			
HDL	0.0824	0.0808	0.0064	0.0187	0.1420	-0.0051	0.0390	0.0499	1.0000		
LDL	-0.1102	0.1234	-0.2162*	0.2224*	0.1619*	0.1354	0.0296	0.2777*	0.2781*	1.0000	
VLDL	-0.1030	0.1180	-0.0021	0.1167	0.1673	0.0400	0.7857*	0.0881	0.0093	-0.0250	1.0000

*p<0.05

Table 23 :Correlation among risk factors for Impaired fasting glucose patients(Male samples)

Risk factors	Age	BMI	First degree relative with DM	Phys1cal act1v1ty	FBS	PPBS	TG	TC	HDL	LDL	VLDL
Age	1.0000										
BMI	0.0614	1.0000									
First degree relative with DM	0.0856	-0.0761	1.0000								
Phys1cal act1v1ty	-0.1606	-0.1367	-0.0171	1.0000							
FBS	0.1145	0.0734	-0.1576	-0.1682	1.0000						
PPBS	0.0775	0.1350	-0.0871	-0.1892	**0.5643***	1.0000					
TG	-0.1297	-0.0503	0.0193	0.0952	0.1204	0.1708	1.0000				
TC	-0.0407	0.0075	-0.1344	0.0264	0.1478	0.1537	0.0772	1.0000			
HDL	0.1509	0.1481	0.0452	-0.0439	**0.2062***	0.0531	0.0895	0.0309	1.0000		
LDL	-0.0593	0.1749	**-0.2691***	0.1687	0.1329	0.1506	0.0480	0.2524	**0.2659***	1.0000	
VLDL	-0.1099	0.1265	-0.0159	0.0586	0.0510	-0.0295	**0.6976***	0.0675	0.0434	-0.0728	1.0000

*p<0.05

Table 24 : Correlation among risk factors for Impaired fasting glucose patients(Female samples)

Risk factors	Age	BMI	First degree relative with DM	Phys1cal act1v1ty	FBS	PPBS	TG	TC	HDL	LDL	VLDL
Age	1.0000										
BMI	-0.0589	1.0000									
First degree relative with DM	0.2382	-0.0458	1.0000								
Phys1cal activity	0.1096	-0.2282	0.2025	1.0000							
FBS	-0.0227	0.1791	0.0861	-0.2138	1.0000						
PPBS	0.1805	0.0517	0.0505	-0.0904	**0.6786***	1.0000					
TG	-0.0737	0.2416	-0.0177	-0.0500	**0.4712***	0.2736	1.0000				
TC	-0.2236	0.0991	-0.1294	0.1902	**0.4161***	0.2228	**0.3588***	1.0000			
HDL	-0.0296	-0.0839	-0.0968	-0.0366	0.0384	-0.1660	-0.0941	**0.3457***	1.0000		
LDL	-0.2129	0.0220	-0.0751	0.2536	**0.2669***	0.1357	-0.0291	**0.8557***	**0.2586***	1.0000	
VLDL	-0.0212	0.1492	0.0391	0.1379	**0.4318***	0.2682	**0.9248***	**0.3907***	-0.1413	0.0284	1.0000

*p<0.05

DISCUSSION

The present study included 130 individuals with one or more risk factors for type 2 diabetes mellitus,their mean age was in the range of 45 to 72 years,out of which there were 97,(74.62%) males and 33(25.38%) females.

The number of risk factors were determined in each individual based on presenting history,past history,family history,physical activity and vital parameters.Fasting plasma glucose with lipid profile were determined in all these subjects.Present study showed that 8.46% individuals with one or more risk factors for diabetes mellitus had fasting blood glucose in the range of 100-125mg/dl that is in the range of Impaired fasting glucose(p value <0.0001 highly significant).Individuals with risk factors for diabetes mellitus were more likely to have blood sugar in the range of Impaired fasting glucose.

When this study parameters were compared with another study done by Simmi dube et al[6],where individuals with and without risk factors for type 2 diabetes mellitus were screened for Impaired fasting glucose-out of 1008 individuals divided ,585 were in group 1(with risk factors) and 433 were in group 2(without risk factors). IFG was seen in 4.61% and 1.41% of group 1 and group 2, respectively.This slightly lower prevalence of IFG individuals in this study was due to lesser number of each individual risk factors in group 1 individuals.

This study is also showing similar results as another study done by David m Nathan et al[7],where individuals with risk factors for diabetes were screened for Impaired fasting glucose and found out that 11.6% individuals with risk factors for diabetes were having Impaired fasting glucose.

This study is also similar to study done by G Vijayakumar, R Arun, VR Kutty[85],where individuals with one or more risk factors for diabetes were screened for Impaired fasting glucose.Cross sectional study was done in 1990 individuals (women: 1149; men: 841). The crude and age adjusted prevalence of IFG was 5.1% and 4.6 % respectively.The lower incidence of IFG individuals in this study can be attributed to more number of total subjects selected for this study and also more number of individuals in age group less than 45 years.

A similar study was done by Wan Nazaimoon W Mohamud, M Suraiami[86] at Institute for Medical Research, Jalan Pahang-50588 Kuala Lumpur . A total of 119 female OA(Orang Asli (OA) are the indigenous people of Peninsular Malaysia.), aged ≥18 years were studied. The subjects underwent physical examination, and fasting blood samples were collected for plasma glucose and lipid profile. A cohort of 76 subjects was followed up for 2 years. Results were, Prevalence of diabetes, IFG and METS was 8.4, 16.8 and 22.7%, respectively.Conclusion drawn was, Prevalence of diabetes and IFG among the OA has increased significantly over the last decade.This study is similar to current study where thers is increased prevalence of Impaired fasting glucose among people with risk factors for diabetes.

In the current study,there was increased incidence of Impaired fasting glucose with hypertension as a risk factor,this study is further supported by a similar study done by chika suematsu, Tomoshige Hayashi et al[87]at Osaka City University Medical School,this study demonstrated direct correlation between IFG and hypertension.Hence it can be concluded that hypertension is a major risk factor for diabetes mellitus.

In the present study,it is seen that individuals with cardiovascular risk factors like obesity,dyslipidemia,hypertension are associated with increased incidence of Impaired fasting glucose,this study correlates with a similar study done by C Snehalatha, A Ramachandran, K Satyavani, S Sivasankari, V Vijay[88] at Chennai where 389 non diabetic individuals with risk factors for diabetes were screened for Impaired fasting glucose, Subjects with IFG (76.9% vs. 58.1%, p = 0.02) had a significantly higher prevalence of clustering of risk variables than the normal glucose tolerance subjects (58.1%). The results of this study showed that IFG is associated with several cardiovascular risk factors. Clustering of the risk factors occurred more frequently in IFG.

A similar study was done by Yun HE,Han Ma et al at Graduate School of Chosun University, Korea[89]. This study was performed to investigate the prevalence of Impaired fasting glucose (IFG) and its related characteristics among healthy adults in some Korean rural areas. A cross-sectional study using the data from 1352 adults who were over the age of 40 and under the age of 70 and who were free of diabetes mellitus (DM). The prevalence of metabolic syndrome (METS) and METS components was higher in the subjects with IFG than in those with normal fasting glucose (NFG).Since the components of metabolic syndrome are also risk factors for Impaired fasting glucose,hence it can be concluded that there is increased prevalence of metabolic syndrome among individuals with Impaired fasting glucose.

This current study also correlated triglyceride levels with increased incidence of Impaired fasting glucose,and resulted more cases of Impaired fasting glucose had hypertriglyceridemia. This study is in accordance with Study done by Quin y et al[90]at Wuxi Center for Disease Prevention and Control, Wuxi 214023, Jiangsu, China. Fasting

plasma glucose (FPG) was positively correlated with BMI, systolic blood pressure (SBP), diastolic blood pressure (DBP), triglyceride (TG), and total cholesterol (TC). In this study, important predictors of IFG were compared. Abnormal TG as a lipid marker was more strongly associated with IFG than with TC and HDL-C. TG was an important predictor of IFG, with odds ratios of 1.76 (95%CI: 1.31-2.36) for subjects with borderline high TG level (1.70 mmol/l < or = TG < 2.26 mmol/l) and 3.13 (95% CI: 2.50-3.91) for those with higher TG level (TG > or = 2.26 mmol/l), when comparing to subjects with TG < 1.70 mmol/l.IN the same study,they also noted BMI was also more strongly correlating with Impaired fasting glucose,as is seen in this current study.

In the current study it was also seen that positive family history correlates with increased incidence of Impaired fasting glucose,this indicates the importance of positive family history as an important criteria for screening.This strongly correlates with similar study done by Tabitha A. Harrison MPH, Lucia A. Hindorff MPH et al[91] at Department of Epidemiology (Harrison, Hindorff, Kim, Edwards), School of Public Health and Community Medicine, University of Washington, Seattle, Washington, USA,where they concluded that Overall, a family history approach appears to be a promising new public health tool to fight the growing epidemic of diabetes in the United States.

In the current study it was also seen that increased physical activity was associated with decreased occurence of Impaired fasting glucose,this observation correlates very well with a similar study done by Susan P. Helmrich, , David R. Ragland[92], where they also found that increased physical activity was associated with decreased incidence of diabetes mellitus.

CAUSE OF IMPAIRED FASTING GLUCOSE IN PEOPLE WITH RISK FACTORS FOR DIABETES MELLITUS:

The natural history of IFG is, with 25% progressing to diabetes, 50% remaining in their abnormal glycemic state, and 25% reverting to NGT over an observational period of 3–5 years. Individuals who are older, overweight, and have other diabetes risk factors are more likely to progress. Moreover, low insulin secretion and severe insulin resistance identify individuals more likely to progress to diabetes. With longer observation, the majority of individuals with IFG appear to develop diabetes. IFG has a heterogeneous pathogenesis. Individuals with both IFG and IGT have approximately double the rate of developing diabetes compared with individuals with just one of them. Numerous longitudinal studies indicate that both IFG and IGT are associated with a modest increase in the hazard ratio (1.1–1.4) for CVD, with IGT being a slightly stronger risk predictor .

The majority of this risk appears to be conferred by progression to diabetes, when the risk of CVD increases two- to fourfold. Many cardiovascular risk factors (e.g., low HDL cholesterol, hypertension, and elevated triglycerides) are prevalent in IFG and IGT, but it is unclear whether they occur more frequently in one state than the other. However, after adjustment for known cardiovascular risk factors, IFG remains as independent, albeit weak, risk factors for CVD in some studies but not in others . Even so, it is unclear whether the CVD risk associated with IFG can be attributed to the development of diabetes during follow-up or whether these states per se convey such risk.

The pattern of insulin secretion also differs in IFG. People with isolated IFG have a decrease in first-phase (0–10 min) insulin secretory response to intravenous glucose and a reduced earlyphase (first 30 min) insulin response to oral glucose. However, the late-

phase (60–120 min) plasma insulin response during the OGTT is normal in isolated IFG. The combination of hepatic insulin resistance and defective insulin secretion in isolated IFG results in excessive fasting hepatic glucose production accounting for fasting hyperglycemia. The impairment in early insulin response in combination with hepatic insulin resistance results in the excessive early rise of plasma glucose in the 1st hour of the OGTT. Subjects with IFG predominantly have hepatic insulin resistance and normal muscle insulin sensitivity. The pattern of impaired insulin secretion also differs between the two groups. Subjects with isolated IFG manifest a decrease in first-phase insulin secretory response to intravenous glucose and early-phase insulin response to oral glucose. However, late-phase plasma insulin response during OGTT is less severely impaired.

SUMMARY

This study was conducted at shri B.M.Patil Medical College Hospital and research centre,Bijapur.The present study included 130 individuals with one or more risk factors for diabetes mellitus as criteria laid down by American Diabetes Association.These individuals were selected ,in detail history was taken and subjected to laboratory investigations of fasting blood glucose,lipid profile,and post prandial blood glucose.In this study there were 51.35% males and 49.45 % females.

In this study 8.46% individuals had impaired fasting glucose,the results correlating with other studies.

Comparing the physical activity,out of 8.46% individuals with Impaired fasting glucose,8.41% were moderate workers,14.29% were in heavy workers.

Comparing individuals with history of hypertensive heart disease and ischaemic heart disease, 13.79% individuals had history of hypertensive heart disease and ischaemic heart disease and 4.17% did not have any history.

Out of 130 individuals,105 individuals were overweight and 14 individuals were obese,11 individuals were with normal BMI.

Also in this study out of 8.46% individuals,54.55% individuals had positive family history of first degree relatives with diabetes and 45.45% did not have positive family history.

This study shows that individuals with one or more risk factors for diabetes are at increased risk of Impaired fasting glucose and future risk of diabetes. Because individuals with Impaired fasting glucose are associated with future risk of diabetes and cardiovascular diseases risk, it may be prudent in clinical practice to periodically monitor individuals with risk factors for diabetes to prevent future occurrence of diabetes and various complications due to diabetes mellitus.

Hence this study very well goes in accordance with various Indian studies where the incidence of impaired fasting glucose was in the range of 8-11%, in individuals with one or more risk factors for diabetes mellitus.

CONCLUSION

Individuals with risk factors for diabetes are associated with having Impaired fasting glucose also known as prediabetes. Hence,identifying individuals with Impaired fasting glucose will lead to preventing future occurrence of diabetes mellitus and other cardiovascular risk factors,by life style modification and if needed pharmacological therapy.

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BLDEA'S SHRI. B. M. PATIL MEDICAL COLLEGE HOSPITAL AND RESEARCH CENTRE, BIJAPUR

STUDY OF IMPAIRED FASTING GLUCOSE IN PATIENTS WITH ONE OR MORE RISK FACTORS FOR DIABETES MELLITUS.

Name:
Age:
Sex:
Occupation:
Religion:

IP. No:
Address:
Date of Admission:
Date of Discharge:
Status at Discharge:

History of presenting illness:
Past history:
H/o of previous symptoms suggestive of ischemic heart disease (Chest pain, giddiness, palpitation).

H/o of previous symptoms suggestive of hypertensive heart disease (Chest pain, giddiness, breathlessness, palpitation).

H/o of delivery of a baby weighing > 9 lb or were diagnosed with gestational diabetes mellitus.

FAMILY HISTORY:

Diabetes mellitus:
Father: Yes/No
Mother: Yes/No
Brother: Yes/No
Sister: Yes/No
Grand father: Yes/No
Grand mother: Yes/No

Hypertension.
Father: Yes/No
Mother: Yes/No
Brother: Yes/No
Sister: Yes/No

Personal History

General physical examination:

Pallor:

Icterus:

Cyanosis:

Clubbing:

Pedal edema:

Lymphadenopathy:

Height:_____ meters

Weight:_____ Kg

BMI: _____Kg/m^2

Sclera: Clear/Muddy/Yellow

Xanthelasma: Present /Absent.

Skin:

Neck: Thyroid gland: normal / enlarged

Vital Signs:

Pulse rate:

Blood pressure:

Temperature:

Respiratory rate:

Gastrointestinal System

Inspection:

a. Shape of the abdomen.

b. Visible peristalsis.

c. Movement with respiration.

d. Engorged veins: Direction of flow of blood.

e. Umbilicus.

f. Hernial orifices.

g. Divertification of recti

h. Skin of abdomen wall.

i. Scrotal examination

j. Signs of chronic hepatocellular failure.

Palpation:

Spleen: Palpable/Not palpable

Liver: Palpable / Not palpable

Percussion:

a. For free fluid in abdomen.

1. Fluid thrill

2. Shifting dullness

3. Horseshoe shaped dullness.

4. Puddles sign.

b. Organ percussion

1. Liver

2. Spleen

3. Other lump

Auscultation:

1. Peristaltic sound.

2. Arterial bruit.

3. Venous hum.

4. Fetal heart sounds and uterine souffle.

Cardiovascular system :

I. Arterial system

a. Pulse: 1. Rate

2. Rhythm

3. Volume

4 .Character of pulse

5. Condition of vessel wall

6. Radial to radial comparison

7. Radial to femoral comparison.

8. Other peripherial pulsations.

	Right	**Left**
Dorsalis pedals		
Posterior tibial		
Popliteal		
Femoral		
Brachial		
Radial		
Carotid		

b.Blood pressure:

II.Venous system:

a.Jugular venous pulsations:

b.Jugular venous pressure:

III. Precordial examination:

Inspection :

1. Precordial bulge

2. Apical impulse location.

3. Other pulsations

a. Epigastric

b. Left parasteral.

c. Pulmonary area

d. Suprasternal

e. Supra clavicular

Palpation

a. Apical impulse: location, character

b. Left parasternal heave

c. Epigastric pulsations

d. Diastolic shock

e. Supra clavicular pulsations

f. Thrills

g. Any other pulsations

h. Tracheal tug

Percussion:

a. Right border.

b. Left border.

c. Right second space

d. Left third space

e. Sternum : Upper and Lower

Auscultation :

a. Heart sounds

b. Murmurs

Respiratory system:

Inspection

a. Shape of chest

b. Lie of ribs

c. Intercostal spaces

d. Subcostal angle

e. Distance between costal margin and iliac crest

f. Spine

g. Supraclavicular fossa

h. Movement with respiration

i. Position of mediastinum

Palpation

a. Position of mediastinum

b. Rib and intercostal space tenderness

c. Measuresments

d. Tactile vocal fremitus

e. Friction fremitus

Percussion

a. Identical areas bilateral

b. Upper border of liver dullness

c. Tidal percussion

d. Kroing's isthmus

e. Superficial cardiac dullness

Auscultation

a. Intensity of breath sounds

b. Type of breath sounds

c. Adventitous sounds

d. Vocal resonance

CNS

Higher mental functions:

Cranial nerves:

Motor system:

Sensory system:

Reflexes:

a. Superficial

b. Deep

Cerebellar signs:

Provisional diagnosis :

Investigations

I. Biochemistry

1.	Fasting venous blood glucose	
2.	Post prandial venous blood glucose	

Lipid profile

Triglyceride	
Total cholesterol	
HDL cholesterol	
LDL cholesterol	
VLDL cholesterol	

Diagnosis

Impression:

CONCLUSION:

SIGNATURE OF GUIDE

SIIRI B.M. PATIL MEDICAL COLLEGE, HOSPITAL AND RESEARCH CENTRE, BIJAPUR - 586103.

CONSENT FORM

TITLE OF RESEARCII : STUDY OF IMPAIRED FASTING GLUCOSE IN PATIENTS WITH ONE OR MORE RISK FACTORS FOR DIABETES MELLITUS.

GUIDE : DR. SHASHIDHAR..S. DEVARMANI

P.G. STUDENT : DR. SACHIN. C. GUDAGE

PURPOSE OF RESEARCH :

I have been informed that the purpose of this study is to screen for Impaired Fasting glucose among patients with one or more risk factors of diabetes mellitus. This study will help in preventing occurrence of complications of diabetes and also early diagnosis of diabetes.

Procedure:

I understand that I will undergo detailed history and clinical examination after which blood will be collected & sent to the laboratory for investigations.

Risks and discomforts:

I understand that there is no risk involved and I may experience mild pain during the collection of blood.

Benefits:

I understand that my participation in this study will help in early detection and prompt intervention of diabetes mellitus which will reduce the burden of disease and its complications.

Confidentiality:

I understand. that the medical information produced by this study will become a part of hospital records and will be subjected to confidentiality and privacy regulations of the said hospital information of a sensitive personal nature will not be a part of medical records, but will be stored in the investigators research file and identified only by a code number. The code key connecting name to numbers will be kept in a separate secure location. If the data is used for publications, the identity of the patient will not be revealed.

Other identifiers such as photographs and audio tapes will be used only with special written permission .I understand I may see the photographs and videotapes and hear the audiotapes before giving this permission.

Request for more information:

I understand that I may ask for more information about the study at any time and concerned researcher is available to answer my questions or concerns. I understand that I will be informed of any significant new findings discovered during the course of this study, which might influence my continued participitation.

Refusal or with drawal of participation :

I understand that my participation is voluntary and I may refuse to participate or may withdraw consent and discontinue participation in the study at any time without prejudice to my present or future care at this hospital. I also understand that my researcher may terminate my participation in this study at any time after he/she has explained the reasons for doing so

and has arranged for my continued care by my own physician or physical therapist. if this is appropriate.

Injury statement:

I understand in the unlikely event of injury to me during the study will get medical treatment but no further compensation would be provided. I understand that by my agreement to participate in this study, I am not waiving any of my legal rights. I have explained to (patient / relevant / guardians' name) the purpose of the research, the procedures required and the possible risks and benefits to the best of my ability in patients own language.

(Signature of investigator/PG GUIDE) **Date :**

I confirm that _____(name of the pg guide/chief researcher),has explained to me the research, the study procedure that I will undergo and the possible risks and discomforts as well as benefits that I may experience. I have read and I understand this consent form. Therefore I agree to give my consent to participate as a subject in this research Project.

_____ _____

Participant **Date**

Witness to signature **Date**

Key to Master Chart

BMI	:	Body Mass Index.
HTN	:	Hypertension.
PCOD	:	Polycystic ovarian disease.
FBS	:	Fasting blood sugar.
Physical activity	:	L : Light work, M: Moderate work, H : Heavy work
PPBS	:	Post prandial blood sugar.
TG	:	Triglyceride.
TC	:	Total cholesterol.
HDL	:	High density lipoprotein.
LDL	:	Low density lipoprotein.
VLDL	:	Very low density lipoprotein.

MASTER CHART

	Name	Age	Sex	IP NO	BMI	First degree relative with DM	HTN	Physical activity	History of gestational diabetes mellitus or delivery of baby>4.1kg	Clinical condition with insulin resistance	History of hypertensive heart disease/ ischemic heart disease.	H/O PCOD	FBS	PPBS	TG	TC	HDL	LDL	VLDL
1	BASAPPA PATIL	48	M	4685	28.64	NO	YES	M		NO	YES		102	160	208	267	54	171	41.6
2	CHANDRASHEKHAR	53	M	3190	26.67	NO	YES	L		NO	NO		92	135	90	100	30	52	18
3	LAXMIBAI PATTAR	54	F	3660	30.6	NO	YES	M	NO	NO	YES	NO	119	154	580	205	30	59	116
4	SHRIKANTH WADAR	52	M	132879	27.7	YES	YES	M		NO	NO		102	134	85	246	38	191	17.2
5	HASHIMSAHAB	60	M	7755	29.2	NO	YES	M		NO	NO		72	130	133	210	45	138	26.6
6	WAHID KUTTI	45	M	73248	25.4	YES	NO	M		NO	YES	NO	102	138	115	224	41	160	23
7	HANMANTAPPA	48	M	7893	27	NO	YES	M		NO	YES		104	160	133	210	45	138	26.6
8	SHIVAPPA TALU	50	M	31160	40.08	YES	YES	M		YES	YES	NO	70	113	28	167	20	120	25.6
9	RAMESH PATIL	56	M	3644	26.7	YES	YES	H		NO	NO		108	130	173	182	48	99	34.6
10	AKSHATA GUBBI	45	M	3603	25.4	NO	NO	M		NO	NO		76	83	102	130	32	78	20.4
11	ELIZABETH	45	F	72484	35.2	YES	NO	M	NO	NO	NO	NO	106	126	153	246	34	181	30.6
12	MARALINGAPPA	46	M	60545	30.6	YES	NO	M		NO	NO		104	138	173	269	22	183	34.6
13	MALLAPA PATIL	48	M	3733	28.4	YES	NO	M		NO	NO		102	134	220	205	30	59	116
14	BASAPPA PATHAN	59	M	76310	30.74	YES	YES	M		NO	YES		110	150	85	269	51	183	34.6
15	TULJARAM	46	M	3130	27.7	YES	NO	M		NO	NO		108	154	85	246	38	191	17.2
16	GURUBASAPPA	56	M	2853	26.25	YES	NO	M		NO	YES		108	91	138	128	28	72	27.6
17	RAJENDRA HATTI	55	M	3174	28.7	NO	YES	M		NO	YES		113	160	173	182	48	99	34.6
18	MALLAPA KUMBAR	52	M	3733	25.7	NO	YES	M		NO	NO		104	154	116	158	40	95	23.2
19	DEVENDRAPPA.G	55	M	3754	29.31	YES	NO	L		YES	YES		108	154	116	158	40	95	23.2
20	VIRUPAKSHI.G	48	M	62484	31.2	YES	NO	M		NO	NO		104	156	157	217	40	146	31.4
21	BOURAMMA.B	55	F	3896	26.56	NO	YES	M		NO	YES		120	154	112	209	32	155	22.4
22	LAXMANNA MASALI	50	M	3899	28.4	YES	NO	M		NO	NO		122	158	126	160	42	95	23.2
23	BASWARAJ KALSAD	55	M	5050	28.7	YES	YES	M		NO	YES		106	143	173	269	51	183	34.6

24	BHAGIRATI PATIL	55	F	7873	25.3	YES	NO	M	NO	NO	NO	NO	94	136	110	258	63	173	22
25	GOUSAMUDDIN	56	M	3577	33.77	YES	YES	M		NO	YES		106	180	92	186	34	134	18.4
26	BASAMMA BAGALI	50	F	3603	28.06	YES	YES	M	NO	YES	NO	NO	76	88	102	130	32	78	20.4
27	HARVANT SINGH	55	M	77601	28.67	NO	N0	M		NO	YES	NO	101	130	115	193	30	46	39
28	BHIMRAO.J	72	M	4279	28.7	NO	YES	L	N	YES	YES		108	130	130	168	30	112	26
29	NAGAPPA HADAPAD	50	M	3636	28.4	YES	YES	M		NO	YES		119	160	130	168	30	112	26
30	LAXMAN MASALI	60	M	3899	26	NO	NO	M		NO	YES		122	160	157	217	40	146	31.4
31	GOURWWA H	50	F	60561	25.43	YES	NO	H	NO	NO	YES	NO	102	135	295	313	41	191	62.6
32	SHARNAPPA ANGADI	65	M	3893	26	NO	YES	M		YES	YES		118	188	108	159	33	114	21.6
33	DATTU JADHAV	50	M	4528	27.5	YES	YES	M		NO	YES		105	155	60	163	43	108	12
34	ISUF KUNABI	55	M	86238	31.3	YES	YES	M		NO	YES		102	122	94	210	35	156	18.8
35	BASAPPA H	60	M	3644	31.25	NO	YES	M		NO	NO		113	140	173	182	48	99	34.6
36	LAXMAN NAVI	55	M	4642	25	YES	YES	M		NO	YES		98	120	94	210	35	156	18.8
37	DUNDAWWA B	48	F	4685	26.7	NO	NO	M	NO	NO	YES	NO	96	130	190	189	50	112	37
38	CHANDRAKANTH.T	47	M	374280	25	YES	NO	M		NO	NO		98	115	89	155	30	109	16
39	IRANNA HANJAGI	45	M	36730	30.1	NO	NO	H	NO	NO	NO		88	134	118	200	30	146	23.6
40	DHIRESH PATEL	48	M	3931	30.4	YES	YES	L		NO	NO		106	148	133	210	45	138	26.6
41	SAROJA JOHN46	46	F	364720	26.25	YES	YES	M	NO	NO	NO	NO	106	126	153	246	34	181	30.6
42	SULEMAN CHISTI	50	M	4348	25.59	NO	YES	M		NO	NO		84	125	147	133	23	81	29.4
43	MAHADEVAPPA.V	45	M	3243	26.6	YES	YES	M		NO	NO		102	140	239	165	32	85	47.8
44	CHANAPPA.M	45	M	104057	26.23	YES	YES	M		NO	NO		118	199	127	189	30	134	25.4
45	GOURAWWA	46	F	60561	25.43	YES	YES	M	N0	NO	YES	NO	104	135	295	313	41	191	62.6
46	RAVIKUMAR	45	M	84326	26.4	YES	NO	M		NO	NO		75	110	200	230	34	128	68.4
47	BASAMMA PATIL	46	F	86238	25	YES	YES	M	NO	NO	NO	NO	125	180	230	156	30	80	46
48	ROUSHANBEE	55	F	3931	26.4	YES	NO	M	NO	NO	NO	NO	96	134	138	128	28	72	27.6
49	EMVANAL.D	58	M	3970	29	NO	YES	L		YES	NO		110	130	127	189	30	125	34
50	LAXMIBAI K	45	F	78468	27	YES	YES	H	NO	NO	NO	NO	102	150	200	228	31	157	40
51	IRANNA HARIJAN	56	M	3673	28	YES	YES	H		NO	NO		80	110	118	200	30	146	23.6
52	ABDUL KHAN	46	M	620611	31.62	YES	NO	M		NO	YES		82	108	200	228	31	157	40
53	BASAPPA MADAN	56	M	4232	23	NO	NO	M		NO	NO		90	162	210	133	32	74	27
54	KALLAPPA	46	M	4567	23.2	NO	NO	M		NO	NO		92	118	220	140	30	80	30
55	HUSAINE MALLA	50	F	3167	24.61	NO	NO	H	NO	NO	NO	NO	94	114	139	186	38	182	46

56	LOKAPPA MATHPATI	48	M	3256	28.8	NO	NO	M		NO	NO		96	120	129	160	23	101	36
57	SULEMAN KHAN	54	M	4348	23.59	YES	NO	M		NO	NO		96	148	133	210	45	138	26.6
58	DEVENDRAPPA PATIL	46	M	3269	24.21	YES	NO	M		NO	NO		84	125	147	133	23	81	29.4
59	MALLAPA	50	M	3179	28.7	NO	NO	M		NO	NO		90	160	140	180	20	140	22
60	JAYPALLAPA	60	M	3265	25	YES	NO	M		NO	NO		96	150	135	130	30	80	20
61	SHIVALINGAPPA	52	M	4541	24.8	NO	NO	M		NO	NO		80	111	155	166	32	103	31
62	CHENAPPA.M	48	M	3296	28.08	YES	NO	M		NO	NO		126	208	140	220	20	80	26
63	SHIVAPPA PATIL	55	M	4444	24.59	NO	NO	M		NO	NO		84	160	150	130	25	82	22.6
64	SHARNAPPA	50	M	2834	24.1	NO	NO	M		NO	NO		81	93	110	239	48	149	42
65	MALLAPA TELI	54	M	3138	24.7	YES	NO	M		NO	NO		90	113	130	168	32	106	30
66	SHANKARGOUDA G	50	M	77590	23.67	NO	NO	M		NO	NO		91	130	115	165	30	86	29
67	GURUSIDAPPA.H	50	M	4301	31.25	YES	NO	L		NO	YES		90	160	190	189	50	112	37
68	DEEPESH	45	M	3198	24.8	NO	NO	M		NO	YES		103	126	128	169	23	120	26
69	AMRUT LALJI	48	M	3204	25.96	YES	NO	M		NO	NO		103	146	127	160	38	80	42
70	SHIVSANGAPPA	48	M	4470	26.9	NO	NO	M		NO	YES		80	160	160	140	30	80	30
71	PARAPPA.B	53	M	4603	27	YES	NO	M		NO	NO		102	110	94	210	35	156	18.8
72	GUNDAPPA.B	60	M	4562	25.59	YES	NO	M		NO	NO		86	145	147	132	20	81	29.4
73	SAROJINI	50	F	4464	28.8	YES	NO	M	NO	NO	YES	NO	74	166	120	166	30	112	24
74	KASHIBAI BALI	55	F	3845	28.77	NO	NO	M	NO	NO	NO	NO	91	73	152	170	46	84	40
75	ABBID ALI	48	M	3325	32.92	NO	NO	M	NO	NO	YES		92	158	146	199	38	112	49.2
76	MAHADEVAPPA.V	45	M	4437	26.2	YES	NO	M		NO	NO		96	88	139	165	32	85	47.8
77	CHANDRAKANTH.P	52	m	3358	28.26	YES	NO	M		NO	NO		98	115	80	159	30	109	20
78	KHASIM SAHAB	55	M	4216	25.71	YES	NO	M		NO	NO		80	134	140	160	30	100	30
79	KAPIL.PATIL	50	M	3265	28.7	NO	N0	M		NO	NO		96	120	150	167	54	100	23
80	RAJENDRA KATTI	55	M	3174	28.7	YES	NO	M		YES	YES		102	134	100	167	54	100	23
81	BALASAHEB.T	58	M	10872	27	YES	NO	M		NO	NO		97	134	97	185	37	130	18.2
82	C.M.NUCCHI	48	M	115245	26	NO	NO	M		NO	YES		108	148	85	159	36	106	17
83	R.S.WALI	55	M	18304	27	YES	NO	M		NO	YES		78	130	88	138	29	91	17.6
84	GURUBAI	54	F	10494	26	NO	NO	H		NO	YES		101	154	91	185	37	130	18.2
85	SIDDANGOUDA	45	M	11087	26	YES	NO	M		NO	YES		98	134	200	228	31	157	40
86	GURAMMAA	51	F	11126	25	YES	NO	M	NO	NO	NO		100	148	55	160	40	100	20
87	PEETAMBARI.P	48	F	11022	28	YES	NO	M	NO	NO	YES	YES	108	138	113	245	50	172	22.6

88	PRAHLAD	50	M	11121	26	NO	NO	M		NO	NO		124	148	60	100	30	60	10
89	LAXMIBAI	48	F	120414	27	NO	NO	M	NO	NO	YES		108	144	103	157	39	97	20.6
90	MANSINGH	48	M	11180	26	YES	NO	M		NO	NO		90	110	84	115	32	66	16.8
91	KALAVATI	50	F	11202	29	NO	NO	M	NO	NO	YES	NO	98	136	97	183	38	126	19.4
92	P.B.JAJU	52	M	120430	25	YES	NO	M		YES	NO		110	156	118	200	30	146	23.6
93	GURUBAI	50	F	10494	26	NO	N0	H	NO	NO	YES	NO	92	140	20	228	31	157	40
94	MADURAO	50	M	11309	26	YES	NO	M		NO	YES		108	160	127	220	30	134	25.4
95	SANGAPPA	45	M	10745	28	NO	NO	M		NO	YES		98	128	138	128	28	72	27.6
96	BASAWARAJ.P	47	M	11274	26	YES	NO	M		NO	NO		82	130	95	133	27	87	19
97	MAHADEVAPPA.P	54	M	11304	28	NO	NO	L		NO	NO		90	120	50	154	49	95	10
98	SUSALAMMA.H	46	F	11249	25	YES	NO	M	NO	NO	YES	NO	93	126	230	156	30	84	46
99	SHIVALINGAPPA	54	M	123320	26	NO	NO	H		NO	NO		90	115	200	230	34	128	68.4
100	SIDAPPA.P	45	M	11283	27	YES	NO	M		NO	NO		123	158	295	313	41	191	62.6
101	RENUKA.P	45	F	11313	25	YES	NO	M	NO	NO	YES		99	126	135	228	45	156	27
102	MADIWALLAMMA.S	55	F	11366	26	NO	NO	M	NO	NO	YES	NO	86	126	55	135	28	96	11
103	SABU RATHOD	55	M	11281	25	YES	NO	M		NO	YES		99	114	64	174	35	126	12.8
104	VILAS.MARUTI	52	M	124751	26	YES	YES	M		NO	YES		92	130	64	174	35	126	12.8
105	MALLAPPA.S	52	M	11555	26	YES	NO	M		NO	NO		99	154	55	135	28	96	11
106	AMARESH	45	M	11354	29	NO	NO	M		NO	YES		111	134	103	157	39	97	20.6
107	SAVITRI	46	F	11334	25	YES	NO	M	NO	NO	YES	NO	66	102	88	138	29	91	17.6
108	DODAMMA C	50	M	11574	25	YES	NO	M		NO	NO		90	108	97	177	26	132	19.4
109	ZIBRIL.KUSUM	48	M	11540	26	NO	NO	M		NO	YES		88	122	60	198	25	161	12
110	GURAPPA IJERI	51	M	11340	26	YES	NO	M		NO	YES		93	85	85	134	30	80	24
111	BAASANN GURADDI	51	M	11613	28	NO	N0	M		NO	NO		96	134	114	110	26	61	22.8
112	SHRIHARI	46	M	11475	26	NO	NO	M		NO	YES		80	130	63	147	32	102	12.6
113	BALRAJ	48	M	11632	26	YES	NO	II		NO	YES		90	114	69	179	31	131	13.8
114	BASAVARAJ	55	M	11611	27	YES	NO	M		NO	NO		98	130	160	182	25	125	32
115	APPARAYA	48	M	11495	27	YES	NO	M		NO	NO		88	122	140	101	27	46	28.2
116	CHANAPPA.M	50	M	11623	27	NO	NO	M		NO	YES		88	114	146	128	28	71	29.2
117	JYOTI PATIL	45	F	11623	28	YES	NO	M	YES	NO	NO	NO	101	118	180	161	47	78	36
118	MAHADEVI	50	F	120729	25	NO	NO	M	NO	NO	NO	NO	82	108	92	196	45	133	18.4
119	ANAND	47	M	126853	28	YES	NO	M		NO	YES		88	114	115	191	49	119	23

120	S.M.MASALI	48	M	126929	28	YES	NO	M		NO	NO		99	136	155	160	48	81	31
121	MAHADEVI PATRI	45	F	11715	26	NO	NO	M		NO	NO		88	103	110	159	45	67	22
122	KAREPPA M	52	M	11717	27	NO	NO	H		NO	NO		89	110	90	120	40	62	18
123	SHOBHA	55	F	11788	26	NO	NO	M	N0	NO	YES	NO	91	130	106	149	39	86	21.2
124	YALLAWWA	50	F	11790	27	YES	NO	M	N0	NO	YES	NO	72	108	70	144	34	96	14
125	HARISCHANDRA	60	M	127945	28	NO	NO	L		NO	NO		89	114	71	149	40	95	14.2
126	LAXMI.S	45	F	129058	25	NO	NO	H	NO	NO	YES	NO	76	112	99	198	39	139	19.8
127	RAMESH	46	M	1E+06	26	NO	NO	M		NO	YES		88	108	92	148	36	94	18.4
128	SHANTADEVI	54	F	11826	25	YES	NO	H	NO	NO	NO	NO	72	110	68	136	38	64	34
129	PUTLABAI	48	F	11621	27	NO	NO	H	NO	NO	YES	NO	80	110	202	201	36	125	40.4
130	PARVATI	55	F	11473	28	YES	NO	H	NO	NO	YES	NO	88	120	151	179	47	102	30.2