Abstract

According to the recent Diabetes Atlas published by the International Diabetes Federation, globally there are 366 million people with diabetes. The number projected for India was 61.3 million people with diabetes. The recent Indian Council of Medical Research-India Diabetes (ICMR-INDIAB) study confirmed that there are 62.4 million people with diabetes in India. Moreover, the study showed that there are 77.2 million people with pre-diabetes, an earlier stage of diabetes, a large percentage of whom will convert to diabetes in the near future.

Obesity is a global health problem and India is facing a rising epidemic of obesity. The prevalence of generalized obesity (as defined by BMI $\geq 25$ kg/m$^2$) and abdominal obesity (as defined by waist circumference $\geq 90$ cm in men and $\geq 80$ cm in women) in the INDIAB study were 21% and 24.8% respectively. Combined obesity (generalized plus abdominal) was present in 28.6% of the population, which translates to 199 million people with obesity in India. The combined diabetes and obesity epidemic is referred to as ‘diabesity’. Diabesity in Indians are characterized by the increased insulin resistance, along with the environmental factors particularly associated with industrialization and urbanization followed by altered food habits and lifestyle. Appropriate nutrition measures will help in reducing the risk of not only diabetes and obesity, but also hypertension, dyslipidemia and hyperinsulinemia. Thus, effective preventive programmes need to be urgently implemented to tackle the diabesity epidemic threatening the country.

Intake of high fat diet together with sedentary lifestyle leads to the metabolic overburden in the human body. This can lead to an increase in the production of reactive metabolites like methylglyoxal and glycoaldehyde which can cause the glycation of tissue protein, lipid and DNA. Advanced glycation end product (AGE) is produced in such conditions and gets deposited in different organs. Glycation causes impairment in the structural and functional properties of the target protein. The production of AGES is accelerated in diabetes mellitus due to the hyperglycemic environment and has been found responsible for the development of diabetes-specific micro vascular system (retinopathy, neuropathy...
and nephropathy) and macro-vascular system (atherosclerosis which leads to heart disease, stroke and peripheral blood disease). AGEs has also been found to be deposited in increased amount in the aging brain and also in the brain of people suffering from Alzheimer’s disease is one of the important factors for the cause of cognitive decline in diabetics.

AGEs interact with it receptors, mainly receptor for advanced glycation end product (RAGE) and induce oxidative stress and increased the expression of adhesion molecules in cultured endothelial cells and in mice models. Glycated albumin also promotes the secretion of inflammatory mediators in the arteries and endothelial cell apoptosis with glomerular hyperfilteration in the kidney.

Given the importance of AGE in the development of metabolic syndrome and neurodegenration, this research work was initiated in order to study and understand the various AGE-mediated cellular responses that induce the development of metabolic syndrome and cognitive decline associated with diabetes, and obesity. AGE-mediated cell signaling pathways were studied in different cell lines.

Work done during my PhD is organized and divided into five chapters.

**Chapter-I** reviews the current understanding and attempts to present a comprehensive understanding of the various aspects of the development of metabolic syndrome. Diabetes and obesity involves the combined manifestation of both the immune system and metabolic system. It discusses the different cellular responses and their underlying cell signaling pathways related to the innate immune system (inflammation) and the metabolic system and the extensive cross-talk associated which is responsible for the development of metabolic syndrome. The process of formation of advanced glycation end product (AGE) and its role in the development of various disorders associated with diabetes is discussed in detail. Lastly a comprehensive picture of the use of herbal drugs (phytochemicals) as therapeutic intervention for combating metabolic syndrome is presented.
Chapter-II gives a detailed account of the various material used and the experimental procedures undertaken for the investigations discussed in the result section.

Chapter-III describes the initial AGE-mediated cellular responses which comprises of oxidative stress and the sustained activity of stress responsive kinases such as IKK and ERK. AGE induced the activity of NF-kB, a transcription regulator of inflammation which was abrogated by the use of antioxidants. Surprisingly, AGE caused a decrease in the transcriptional activity of PPARγ, a transcription regulator of glucose and lipid metabolism and is often found to be down-regulated in diabetes which in turn can cause insulin resistance. This suggests the role of AGE in inducing insulin resistance. Among the different anti-oxidants used, only mangiferin was able to protect the AGE-mediated decrease in the PPARγ transcriptional activity which was partly due to the inhibition of ERK and partly due to it, acting as a PPARγ agonist. This chapter presents the molecular mechanism for the action of mangiferin in protecting against diabetes.

Chapter-IV, describes the role of AGE in increasing intracellular lipid content. Decrease in PPARγ transcriptional activity causes impairment in lipid transport in cells and causes lipid accumulation in non-adipose tissue as seen in diabetes and fatty liver condition. As AGE caused the down-regulation of PPARγ transcriptional activity, it was necessary to study its effect on the cellular lipid content. Moreover it was found that AGE induces the transcription of SREBP, a transcription regulator of lipid synthesis, leading to an increase in lipogenesis. This cellular response was found to be dependent on IKK and ERK kinases. Mangiferin which was found to be effective in the induction of PPARγ transcriptional activity was also effective in inhibiting AGE-mediated lipid accumulation by inhibiting increased SREBP transcriptional activity by preventing the activity of IKK and ERK kinases. This chapter presents the effect of mangiferin in combating dyslipidemia.

Chapter-V, describes the cellular responses upon prolonged exposure to AGE. It was found to cause 30-40% cell death especially in neuronal and monocytic cell lines. This response was found to be independent of AGE-mediated oxidant stress. The detailed
signaling pathway is presented which suggest that AGE induced cell death is caused by the increased intracellular calcium which in turn activates the NF-AT transcriptional factor followed by an increase in the expression of its dependent gene, FasL and ultimately FasL-mediated cell death. AGE has been found to have role in neurodegeneration and this chapter presents the probable molecular events.

Overall the present study helps to understand the different AGE-mediated cellular responses characterized by the perturbations in cell signaling and transcriptional machinery of the cells which can cause the development of metabolic syndrome and cognitive decline due to increased metabolic burden. It also suggest that by the combined modulation of various sub-systems and the different cell signaling pathways involved, a better therapeutic regime can be designed and implemented.