Chapter 5

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The present study is an evaluation of the anthropometric, hematological, biochemical parameters and the somatic DNA damage in type 2 diabetic patients during Ramadan fasting. Only male subjects were included in this study because females break their fast during their menstrual period which will alter the duration of the fast. All the subjects participated in the present study were from middle class socioeconomic backgrounds who were working in sectors where physical exertion was necessary. Smokers and those who consume alcohol were excluded from the present study to avoid confounding effects of smoking and alcohol.

In the present study we have not interrupted the regular diet of the test subjects in order to assess the metabolic changes under their usual diet of Ramadan fasting season. In many previous studies there was regulation of food intake by the diabetics which can cause disparities in the results of their study. All the subjects were comparable by means of age, BMI, arterial blood pressure (table 4.1.1). The results are discussed below.

5.1 ALTERATIONS IN ANTHROPOMETRIC PARAMETERS

5.1.1 Body Weight

The love towards non-vegetarian dishes makes Keralites different from the other parts of India. The choice of Keralites has moved to chicken from fish. A complete non vegetarian diet on a daily basis may lead to the development of obesity (Newby, 2005). Diet has an important role in maintaining the body weight under normal healthy standards. Overeating can be considered as one of the major causes of morbidity and mortality in humans. Caloric restriction can be beneficial to improve the health of non obese individuals. The nutritional factors, energy intake and energy expenditure plays a key role in the body weight management (Stote, 2007). In Kerala, during Ramadan, both the meals (predawn and after sunset) consisted of fruits, vegetable salad, meat and cereal items including rice and roti. While breaking the fast like other Muslims in the world, Kerala Muslims also use dates and sweetened drinks. Usually Keralites have their breakfast and
dinner heavy while a minimal lunch, because many will be outside for their work and most of the Keralites prefer homely food. So in Keralites the change in the meal time during Ramadan does not affect drastically as there are two main meals during this period.

Reduction in the meal frequency and spending more time in spiritual activities reduces Ramadan observers’ appetite and has found effective in decreasing the weight in diabetic males (Khatib and Shafagoj, 2004). Rathor et al., 2003 and Mafauzy et al., 1990 reported a significant decrease in the body weight of diabetic patients after Ramadan fasting. The present study is in agreement with the findings of Rathor et al., (2003), Khatib and Shafagoj (2004) and Mafauzy et al., (1990) as there was a significant reduction in the body weight of diabetic males after Ramadan fasting. Similar result was observed in healthy males also which supports previous studies where there was a significant reduction in the body weight and BMI in healthy adults after Ramadan fasting (Ara et al., 2015; Unalacak et al., 2011). Kamal et al., (2012) observed a decrease in body weight of healthy adults of India between 14th and 26th day of Ramadan which was regained during post Ramadan period. This decrease may be due to a change in the timing of meals and decrease in the frequency of meals. Along with change in pattern of diet, all the subjects in the present study continued their job which required physical exertion, during Ramadan fasting. So diet and physical activities may have contributed to reduction in the body weight.

5.1.2 Arterial Blood Pressure

Human body has a group of efficient system for the regulation of arterial blood pressure which ranges from operating rapidly or slowly and operating at low pressure, normal pressure or high pressure to attain a state of equilibrium of the body. A long-term regulation of blood pressure involves the balance between intake and output of water and salt along with the involvement of kidneys (Guyton et al., 1972). Elevated blood pressure is an important risk factor for stroke, coronary heart disease, heart failure and kidney failure. Variation from the normal range of blood pressure results from environmental and genetic factors. The major environmental factors which lead to hypotension or hypertension include diet,
physical activities, phychosocial factors and alteration in sleeping patterns. Among these, dietary factors and sleeping patterns have a predominant role in blood pressure homeostasis (Appel et al., 2006). In the present study the mean value of the blood pressure of the test subjects was 127.9/83.8 (table 4.1.1) which is in a normotensive range. In an Indian study on healthy males (Kamal et al., 2012), a significant decrease in the systolic blood pressure was observed during 2nd week of Ramadan but on the 26th day no significant difference was reported in the values from the pre-Ramadan values. M’guil et al., (2008) has observed an unaltered systolic and diastolic blood pressure in type 2 diabetic patients at the end of Ramadan fasting. In the present study blood pressure was not significantly altered in diabetic males as well as healthy males after Ramadan fasting, but there was a slight decrease in the systolic and a slight increase in the diastolic blood pressure which is not statistically significant. This is in line with the observations by M'guil et al., (2008) in diabetic adults and Saiyad et al., (2014 ) who reported stable blood pressure in healthy adults in after Ramadan fasting.

A study conducted in Jammu-Kashmir, by Gupta et al., (2013) also made similar observation regarding arterial blood pressure. Kamal et al., (2012) and Sayedda et al., (2013) also reported a stable blood pressure with a decrease in body weight after Ramadan fasting, whereas Arabi et al., (2015) observed a decrease in systolic and diastolic blood pressure in healthy males after Ramadan fasting in NAFLD patients. Human body’s regulatory mechanisms might have activated to maintain the normal blood pressure level. An increase in the dietary fibre intake has reported to result in a non-conclusive reduction in arterial BP in normotensive people (Whelton et al., 2005). An animal model study has reported that a rise in vasopressin with a fall in body weight has maintained the normal pressure during long-term dehydration hours (Woods and Johnston, 1983). Some weight loss intervention has produced benefits on blood pressure regulation which had persisted long time. Maintaining normal body weight, regular physical activities, avoiding alcohol consumption, diet rich in fruits and vegetables can maintain normal blood pressure (Whelton et al., 2002; Whelton et al., 2005; Elmer et al., 2006).
5.2 MICROALBUMINURIA

Excretion of albumin less than 30 mg/day through urine can be considered as normal in case of diabetic patients and albumin excretion of 30-300 mg/day through urine is known as microalbuminuria and further clinical albuminuria. Microalbuminuria occurs in 30-40% diabetic patients, which may lead to renal complications and cardiovascular complications ultimately leading to coma and death (Mohan et al., 2015). Microalbuminuria is one of the first markers of diabetic nephropathy. Development of minimally increased albuminuria has a prognostic implication on diabetic nephropathy (Fantus, 2016) and this always cluster with cardiovascular risk (Winocour et al., 1992; Zambon et al., 1994). In the present study subjects of both diabetic fasting and diabetic nonfasting group had a microalbumin level within the biological limit. There was a significant (p = 0.001, table 4.2.1) decrease in the microalbumin level in diabetic patients after Ramadan fasting. No such significant change has been observed in non fasting subjects. Similar observation has been found in patients with micro-vascular complications (Kamar et al., 2015) as well as healthy adults (Asegaonkar et al., 2014) after Ramadan fasting. Reduction in bodyweight, maintaining normal blood pressure and normalization of blood glucose in diabetic patients may reduce the microalbumin level in urine (Koroshi et al., 2007). Reduction in the red meat also found to have beneficial effect on microalbuminuria (de Mello et al., 2006). The present observation may be due to the fact that there was a change in meal timing and diet pattern which has led to significant reduction in the body weight.

5.3 HEMATOLOGICAL PARAMETERS

Glucose control generally affects hematological parameters in diabetic people (Varm et al., 2014). A high WBC count is considered as a nonspecific marker for immune system activation and inflammatory response. The chronic activation of immune system proposes a risk of pathogenesis of diabetes (Vozarova et al., 2002). Report says an increase in WBC count is clinically relevant for estimating the risk of diabetic complications (Shimizu et al., 2015). Hosseini and Hejazi, (2013) concluded that Ramadan fasting had no effect on the WBC count in healthy individuals. M’guil, et al., (2008) reported an increase in WBC count
within the normal range in diabetic adults. Contradictory to this in the present study no significant change in total count of WBC was observed in the diabetic males as well as healthy males after a month long intermittent fasting (table 4.3.1). Regarding healthy adults the observations in the present study is in agreement with the observation of Hosseini and Hejazi (2013). The results of the present study indicate that there is no aggravation of diabetic complication after fasting with regard to the WBC count. Intermittent fasting helps internal energy system of the body to focus more towards the immunity (David, 2013). Intermittent fasting enhances defense mechanism of antioxidant system and reduces proinflammatory cytokines (Sandhya et al., 2016). Develioglu et al., (2013) also observed that Ramadan fasting has not altered the immunological status of the body.

The low RBC count, decreased hemoglobin content and decreased hematocrit are considered as indicators of anemia. Scott, (1981) observed a decrease in the number of RBCs after fasting in healthy subjects. In contrast Al Hourani et al., (2009) observed that there was no change in the number of circulating red blood cells after fasting in healthy individuals. The present study supports the findings of Al Hourani et al., (2009) as there was a no change in the number of RBCs in diabetic patients as well as healthy adults after Ramadan fasting. Even though the frequency of food intake reduces during Ramadan, the quality of diet is well maintained with rich in vegetable salads and fruits. The high nutritional support with reduced meal frequency might have helped the fasting observers to maintain the normal RBC count.

There are conflicting results on the change in the concentration of hemoglobin before and after fasting. Dewanti et al., (2006) and Chaouachi et al., (2008) reported a decreased hemoglobin content whereas Al Hourani et al., (2009), Tayebi et al., (2010) and Mohammed, (2011) had observed no alteration in the hemoglobin concentration in healthy subjects after fasting. M'guil et al., (2008) has found an increase of hemoglobin in females but in males no alteration was found. The present study is in agreement with M'guil et al., (2008) because the hemoglobin remained unchanged after an intermittent fasting in diabetic males. In healthy adults also hemoglobin was not altered after Ramadan fasting which supports the previous studies (Al Hourani et al., 2009; Tayebi et al., 2010;
Mohammed, 2011). It can be concluded that since the RBC count was well balanced and maintained by the diet and food timings, the hemoglobin concentration was also not affected by fasting.

Hematocrit (packed cell volume) gives an assessment of the RBC concentration in the blood. As observed in the number of RBC and hemoglobin concentration in the present study, no significant difference was detected in hematocrit value after fasting in diabetic patients. The same result was also observed in healthy adults. There are different studies on the effect of fasting on hematocrit and the results were contradictory (Dewanti et al., 2006; Chaouachi et al., 2008; Tayebi et al., 2010). An increase in the hematocrit was observed by Chaouachi et al., (2008) and a decrease was noted by Dewanti et al., (2006). The hematocrit value remained unchanged in the study conducted by Tayebi et al., (2010) and is in congruent with the observations of the present study. The disparity in the results may be due to the difference in demographic factors of the subjects, diet and the ethnicity (Hosseini and Hejazi, 2013). The test subjects in the present study were diabetic patients where as in the aforementioned studies, participants were non-diabetic people which may also add to the disparity of the results.

Platelets are essentials for hemostasis and its dysfunction is associated with abnormalities of macro-vascular and micro-vascular circulations. Multifactorial induced high sensitivity of platelet aggregation has been seen in diabetic patients (Mustard et al., 1977). Significant decrease in platelet count was observed in healthy females (Al Hourani et al., 2009 ) and males (Aybak et al., 1996; Ramadan et al.,1999) after Ramadan fasting. In contrary Mohammed et al., (2011) and Hosseini and Hejazi (2013) noticed that platelet count has not been altered by Ramadan fasting. The present study agrees with Mohammed et al., (2011) and Hosseini and Hejazi, (2013) as there was no alteration in the platelet count after fasting in diabetic patients as well as healthy adults.
5.4 ALTERATIONS IN BIOCHEMICAL PARAMETERS

5.4.1 Alterations in the blood glucose level

Monitoring blood glucose level has prime importance in assessing the diabetic status of a person. Poor glycemic control in due course leads to the development of diabetic complications. Table 4.3.2 describes the different parameters of the glycemic status. Fasting blood glucose (FBG) is widely used to diagnose diabetes mellitus because in this there is no chance of the influence of recent food intake. There are reports on a decrease in the fasting blood glucose level in healthy (Fakhrzadeh et al., 2003; Hind and Awad 2006) and diabetic patients (Rathor, 2003; Khatib and Shafagoj, 2004; Bouzid et al., 2016) after Ramadan fasting. M'guil et al., (2008) reported a decrease in FBG and PPBG only in diabetic females, but in diabetic males there was no significant difference in pre-Ramadan values of fasting and post-prandial blood glucose level compared to values of 29th day of Ramadan. Kamar et al., (2014) also observed an improvement in beta cell secretory efficiency and a decrease in the FBG after Ramadan fasting compared to the pre-Ramadan values. In the present study there was no statistically significant change in the fasting blood glucose level in diabetic patients after Ramadan fasting, but the fasting blood glucose on the 30th day was lower than the FBG value of pre-Ramadan. This supports the observations of M'guil et al., (2008) and Kamar et al., (2014). The well controlled diet along with modified pattern of drug intake with moderate physical activity followed by the diabetic patients who observed fasting, might have contributed to the unaltered level of FBG. In the present study in contrast to diabetic males, the decrease in the fasting blood glucose in healthy males after Ramadan fasting was statistically significant. This supports the previous studies which observed a significant reduction of fasting blood glucose in healthy individuals after Ramadan fasting (Fakhrzadeh et al., 2003; Hind and Awad, 2006). The restricted food consumption along with increased physical activities such as additional prayers during night time which require moderate physical involvement might have played a role in reducing the blood sugar level.

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Post-prandial hyperglycemia harmfully affects the vasculature and aggravates the diabetic complications. The traditional diet during Ramadan month includes high calorie sugary fluids and food stuffs. This may lead to an acute post-prandial hyperglycemia in healthy individuals and also in diabetic individuals, eventhough they have a baseline hyperglycemia. It has been reported an increased trend towards the worsening of hyperglycemia in diabetic patients after Ramadan fasting (Salti et al., 2004). Some other studies reported Ramadan fasting does not alter blood glucose levels of diabetic individuals (Sari et al., 2004). Vasan et al., (2012) had found a decrease in PPBS in diabetic adults after Ramadan fasting. M’guil et al., (2008) has found reduction in PPBS in females but not in males. The present study is in agreement with M’guil et al., (2008) as there was no significant alteration in the post-prandial blood glucose level in diabetic as well as non-diabetic subjects. This may be probably due to decrease in the frequency of meal time along with an altered diet which has been resulted in a weight loss in the present study and weight reduction is reported to have role in the enhancement of insulin sensitivity which improves the glycemic status (Khatib and Shafagoj, 2004).

HbA1c is the most practical and reliable single index of blood glucose control (Saudek et al., 2005). HbA1c values depend on the diet composition and the food intake regulation (Nomani et al., 1988). Several studies demonstrated the effect of Ramadan fasting on HbA1c in different contexts. In the current study a non significant decrease in the HbA1C values in diabetic patients and healthy adults was observed after fasting. HbA1c in diabetic patients can be reduced by due consideration of the quality and quantity of carbohydrate foods (Brand-Miller, 2003). An increase in the HbA1c was observed by Belkhadir et al., (1993) and Uysal et al., (1998). The present study supports the findings of Azizi and Siahkolah (1998) who reported unalterd HbA1c of diabetics during Ramadan.

Mean Blood Glucose level (MBG) adds to the ability of HbA1c to predict the risk of hyperglycemia (Kilpatric et al., 2007) and MBG is better in predicting cardiovascular risk (Kilpatric et al., 2008). In the present study in diabetic males as well as in healthy males no significant difference in MBG was observed after fasting. To the best of our knowledge there are no previous studies to substantiate
the results of the present study. The improved glycemic status after Ramadan fasting, as observed in the levels of FBG, PPBG and HbA1c, is reflected in the status of mean blood glucose as well.

5.4.2 Alterations in lipid profile

Dislipidemia is a prime contributor to the risk of cardiovascular disease and cardiac autonomic neuropathy in diabetic patients (Gordon et al., 2008; Mooradian, 2009). Raised oxidative stress as a result of dislipidemia adds to the progression of diabetic neuropathy (Vincent et al., 2004; Pop-Busui et al., 2010). Table 4.3.3 describes the alterations in the markers of lipid metabolism in the study and control groups. Current study evaluated lipid profile of the diabetic and non-diabetic individuals before and after Ramadan fasting. The previous results on the effect of fasting on lipid profile are conflicting. The dietary habits, percentage of fat and its saturation in the daily food can alter an individual’s lipid profile (Ziaee et al., 2006). In the present study it was observed a non-significant increase in the mean value of total cholesterol in diabetic individuals who observed fasting. Laajam, (1990) also found a similar observation in which there was a non-significant increase in the total cholesterol level in diabetic patients after fasting. A recent study observed a significant decrease in total cholesterol after fasting by healthy male adults (Ara et al., 2016). But a significant increase in the total cholesterol of healthy male adults after fasting has been observed in the present study and this result support observations of Hallak and Nomani, (1988) and Jenkins et al., (1989). The change in the lipid profile is closely linked to the composition of diet and response to the biochemical changes to the post fasting state of the body.

The inherent metabolic changes during Ramadan may decrease the serum triglycerides (Al Hourani et al., 2009). In the present study triglycerides were observed to be decreased significantly in diabetic subjects after fasting and it was understandable that Ramadan fasting has favorable effect on triglyceride level of a diabetic individual. This is in agreement with a previous result (Khatib and Shafagoj, 2004). M'guil et al., (2008) observed triglycerides are not altered in diabetic females during Ramadan fasting and significantly decreased in diabetic
males. On the other hand in the present study, a significant increase in triglycerides was observed in non diabetic subjects after fasting. Similar results regarding healthy individuals were found in other studies also (Shoukry et al., 1986; Adlouni et al., 1997). A significant decrease in TG of healthy adults after Ramadan fasting was also reported in some studies (Mahboob et al., 1999; Asgary et al., 2000). No change in triglycerides was also observed in healthy individuals after fasting (Ziaee et al., 2006). The increase of TG in healthy individuals may be due mobilization of body fat as well gorging of a single large meal rich in fat contents (Markel et al., 1985; Shoukry, 1986). The high literacy standard of Kerala people might have contributed to the awareness of diabetic people about the harmful effects gorging fat rich food and they might have become conscious about their diet. The well balanced diet and the manual works might add to the reduction of TG in diabetic individuals.

High density lipoprotein cholesterol (HDLc) was found to be increased in diabetic patients after Ramadan fasting (Dehghan et al., 1994; Khatib 1997; Uysal et al., 1998). A decrease in HDLc has been observed by Belkhadir et al., 1993 and Ziaee et al., 2006. The present study support the findings of Dehghan et al., (1994) and Uysal et al., (1998) as there was a slight but statistically significant increase in the HDL cholesterol in type 2 diabetic patients. Consumption of a single large meal per day increases the HDL cholesterol (Adlouni et al., 1997). The healthy individuals of present study has shown no significant change in the HDL cholesterol level after Ramadan fasting. Many previous studies have reported an increase in HDL cholesterol (Maislos, 1993; Mohammed, 2011; Ara et al., 2016) as well as a decrease in cholesterol HDLc in healthy individuals after fasting (El-Taher and Zabut 2015). This discrepancy in results may be due the difference in the type food materials and physique of individuals because these previous studies were conducted in Middle East part of the world.

Akanji et al., (2000) and Afrasiabi et al., (2003) conclude that Ramadan fasting improves lipid profile of the fasting observers. In the present study a significant decrease in low density lipoprotein (LDL) cholesterol and very low density lipoprotein (VLDL) cholesterol in type 2 diabetic patients was observed. Belkhadir et al., (1993) and Ziaee et al., (2006) reported an increase in LDL
cholesterol in diabetic patients. These observations are not supporting the present result. The decrease in LDLc might be due to shift in substrate utilization for energy with an increased utilization of fat. Indeed reports have shown an increase in fat oxidation during Ramadan fasting (El Ati et al., 1995). Dowod (2004) and Adlouni et al., (1997) reported a decrease in LDL cholesterol in healthy individuals after fasting. Yarahmadi et al., (2003) observed an increase of LDL in healthy individuals after Ramadan fasting. The present study also observed a significant increase of LDL in healthy fasting individuals with a stable VLDL. Usually in Kerala breaking of fast, the Iftar, is rather a time for get-together with family and friends where a variety of dishes rich fried items. Being healthy, individuals become careless in gorging food which may lead to an increase in cholesterol. The decrease in total cholesterol had to lead to significant decrease in Cholesterol/HDL ratio in diabetic patients who observed fasting while an increase in the total cholesterol after Ramadan fasting resulted in the increase of Cholesterol/ HDL ratio in healthy individuals.

5.4.3 Alterations in renal function markers

Dehydration during daytime imposes a stress on the concentration ability of kidneys (Leiper et al., 2003). The renal functions of kidney transplant patients were not affected harmfully by Ramadan fasting (Abdalla et al., 1998). Azizi (2013) observed a small insignificant changes in serum urea, creatinine and uric acid after observing Ramadan fasting. When normal eating and drinking habits alters, individual tends to reduce fluid intake and may result in hypohydration. Hypohydration can lead to relative increase in blood urea concentration (Leiper et al., 2003). In the current study blood urea has shown a significant increase in diabetic patients after fasting and the change was not significant when compared with diabetic controls as well as fasting controls, but was significant when compared with the healthy non-fasting individuals. This finding does not support the observations of M’guil et al., (2008) who observed a significant decrease in urea and creatinine in male and female diabetics while a significant decrease of urea only in females. Azwany et al., (2004) noticed a non-significant increase of blood urea and he suggested the fluid restriction could not induce dehydration in the subjects of his study. El-Gendy et al., (2012) also reported a non-significant
decrease in serum urea and creatinine in diabetic and no-diabetic subjects after fasting. The disparities in results may be due to the differences in the study groups and seasons. In the present study the subjects were working outside which cause water loss through perspiration additional to the osmotic diuresis in diabetic patients. No significant changes in serum urea were observed in healthy fasting adults in the present study and a similar result was found by Boobes et al., (2009).

In the present study Serum creatinine level in diabetic patients increased significantly but within biological limit after Ramadan fasting. The increase was significant compared with the other control group subjects. This change in serum creatinine has observed only in diabetic fasting but not healthy fasting group. The present finding is in line with Bouguerra et al., (2003) who observed a significant increase in serum creatinine. El-Gendy et al., (2012) reported a significant decrease of serum creatinine in diabetic patient when supplemented with vitamin E, but no change in serum creatinine was observed without vitamin E supplement. This result does not support the present observations. The reduced intake of fluids along with perspiration and osmotic diuresis might have contributed to the increase in creatinine level in the type 2 diabetic patients observed present study.

Uric acid is a product of purine metabolism. High level of serum uric acid is considered as a marker for the development of type 2 diabetes. It is also found to be associated with oxidative stress (Hayden and Tyagi, 2004; Dehghan et al., 1994). It was postulated that dehydration during fasting may lead to a raise in uric acid level (Mohammed et al., 2002). In the present study serum uric acid found increased and was statistically significant but not biologically significant. The change was significant when compared with diabetic non fasting group. Al-hader et al., (1994) and Forghani et al., (2001) in their study observed significant increase in uric acid and supports the present study. Nomani et al., (1990) found that the weight loss was positively associated with the uric acid levels. The present study also observed a decrease in body weight after Ramadan fasting which might have also contributed to the increase in uric acid levels in the study subjects.
5.4.4 Alterations in markers of liver function

Table 4.3.5 gives a light on the status of liver function markers in diabetic and non-diabetic groups before and after Ramadan fasting. Liver plays a major role in glucose homeostasis by storing glucose as glycogen as well as by producing glucose by gluconeogenesis. Liver function tests provide information about the functional status of an individual’s liver. The most common liver function tests include serum aminotransferases, alkaline phosphatase, bilirubin and albumin (Harris, 2005). The alterations in liver enzymes are common in type 2 diabetes. There are reports on the biochemical alterations in LFT in relation to type 2 diabetes especially in India (Bora et al., 2016). The major contributor for elevated liver enzymes in type 2 diabetes is nonalcoholic fatty liver disease (Harris, 2005). Altered portal insulin level and insulin-glucagon ratio may influence hepatocyte function and integrity in diabetic patients (Salmela et al., 1984). The association of insulin resistance and serum concentrations of liver enzymes has been established very well (Bora et al., 2016).

A high bilirubin level, which is a waste product of erythrocyte destruction, can be associated with an obstruction in the bile flow. This may happen in cases of liver diseases or any biliary system diseases (Gwaltney-Brant, 2016). Increased level of bilirubin in healthy people, after continuous fasting has been reported earlier (Owens and Sherlock, 1973; Barret, 1975). In the present study no significant changes in the total bilirubin (Bil. T) and direct bilirubin (Bil. D) was observed in diabetic individuals and healthy individuals after fasting. This is not concurrent with the previous studies which reported an increase in Bil.T after fasting and it stayed high even after two weeks of Ramadan (Azizi and Rasouli, 1992; M'guil et al., 2008;).

The sum of albumin and globulin represents total protein. More than half of the total protein is made up by albumin. Liver uses dietary protein to synthesis albumin. A low level of albumin indicates the deteriorating health status of the liver. Globulins are produced by liver as well as immune system. An elevated globulin level indicates worsened inflammation in liver or in immune system. In the current study no significant alteration in the total protein was observed in
diabetic and healthy individuals after fasting. This supports the observations of Mohammed et al., 2011. Stable total protein levels gives a light on the good nutritional status during Ramadan fasting.

In spite of liver being the only site of synthesis of albumin, it can be affected by nutritional status, hormonal balance and osmotic pressure. Protein malnutrition can be the major causative factor for hypoalbumineamia rather than liver abnormalities (Thapa and Walia, 2007). In the present study no significant changes in the albumin and globulin levels were observed in type 2 diabetic individuals after fasting. This supports the observations of Elhazmi et al., (1987). The same change has been observed in non fasting individuals as well. This result is consistent with the result of Ibrahim et al., (2008), who reported an unaltered level of albumin and globulins after fasting in healthy individuals. The A/G ratio was unaltered as there was no significant alteration in albumin and globulin values.

Hepatocellular health can be well demonstrated by the level of aminotransferases (ALT and AST) in serum. It is a well established fact that hepatocellular dysfunction is strongly associated with type 2 diabetes, insulin resistance and obesity (Vozarova, 2002). Ramadan is beneficial for non alcoholic fatty liver patients as it decreases ALT enzymes (Arabi et al., 2015). In the present study there were no significant alterations in the serum ALT and AST levels after fasting in diabetic patients and this goes along with the previous studies (Unalacak, 2011; Furuncuoglu, 2007). But M’guil et al., (2008) studied liver function in diabetics during Ramadan fasting and observed a significant decrease in serum ALT and AST levels.

ALP is one of the biomarker of Liver diseases, but it is not specific as it is secreted from other tissues like bone and kidney. But along with the liver enzymes and bilirubuin, ALP also helps to assess the status of liver function. Elevations of ALP occur in obstructive biliary disease. Malnutrition is also one of cause that leads to imbalances of ALP. In the previous study on the liver function during Ramadan in diabetics has revealed that ALP has not changed significantly from the pre-Ramadan value (M’guil et al., 2008). In contrast, the present study observed a significant increase in the ALP level after fasting in diabetic individuals.
Eventhough increase in ALP observed in non-fasting and healthy fasting individuals also, the increase occurred in diabetic patients was significant compared to the other groups. Since the values were under the biological range and there was no increase in other markers of liver function, ALP increase may not be an indication of altered liver function and may not be clinically relevant. Since the changes in urea, creatinine and uric acid indicated a hypohydration in the diabetic Ramadan observers of the present study, there may be chances of obstruction in the hepatic duct system and subsequent elevation in ALP. Further studies are required to correlate the increased ALP level and fasting.

5.4.5 Changes in baseline DNA damage after fasting

Several line of evidence indicates that diabetes mellitus can accelerate the generation of ROS to the level which cannot be foraged by the normal antioxidant system present in the body. The high level of ROS leads to oxidative stress which has a crucial role in diabetic complications and DNA damage (Eyfjord and Bodvarsdottir, 2005; shilpa et al., 2012; Pandarekandy et al., 2015). The high glucose level has been shown to cause or promote mutagenesis and probably even can lead to carcinogenesis (Lee and Chan, 2015). In the present study, we examined the frequency of micro-nucleated WBC cells in 1000 WBC cells, as a marker for somatic DNA damage. The results indicate that frequency of the DNA damage is more in diabetic subjects than the healthy individuals (Table-4.36). Diabetes patients have low antioxidative defense which makes them prone to onset of oxidative stress (Simone et al., 2008). The increased DNA damage in type 2 diabetic individuals compared to the healthy subjects observed in the present study could explain the possibility of potential DNA damage in fasting individuals. However, the present study reveals that in diabetic patients, there was no significant augmentation in the number of micronuclei after fasting and this observation supports the earlier studies which reported that intermediate fasting reduce oxidative stress in type 2 diabetes patients (Al-Shafei, 2014) and enhances the ability of nerve cells to repair DNA (Mattson, 2015). The high serum glucose, free fatty acids and insulin levels lead to production of ROS (Lee and Chan, 2015). The markers of oxidative stress are reported to be significantly high in diabetic patients with microalbuminuria (Simone et al., 2008).
The observed decrease in body weight, serum glucose, triglycerides, LDLc and microalbumin level in diabetic subjects after fasting, in the present study, might have contributed to the enhancement of antioxidant defense as well as to the prevention in the progression of free radical formation and thus an alleviation of oxidative stress. The present study also found that in healthy fasting individuals the DNA damage was not significantly altered after fasting. In healthy adults no alteration in the markers of oxidative stress was observed in some previous studies (Ibrahim et al., 2008; BaHammam et al., 2016), however, alleviation in oxidative stress in healthy individuals after fasting was noted by Faris et al., (2012). Fasting has beneficial role in the prevention of cancer as well by protecting the cells from DNA damage and promoting the programmed death of the damaged cells (Longo and Mattson, 2014). The beneficial effect of Ramadan fasting on the oxidative stress and the prevention of further DNA damage in diabetic individuals can be explained by the recent observations (Sandhya et al., 2016). This also can be considered beneficial to prevent mutagenesis and carcinogenesis as well as atherosclerosis.

Intermittent fasting is considered as an intervention for improving human health. Modified fasting regimens appear to promote weight loss and may improve metabolic health (Patterson et al., 2015). The observations of the present study demonstrate the beneficial effects of Ramadan fasting by diabetic as well as healthy individuals. The favorable health outcomes by intermittent fasting in the current study is reflected by a reduction in the body weight, betterment in the glycemic status, improvement of lipid profile and an alleviation in the oxidative stress. The arterial blood pressure was not affected seriously by the fasting. A reduction in the low density lipoprotein cholesterol and triglycerides was also observed. These effects of fasting can contribute to get a hold of better cardiac function in diabetic individuals. The reduction in the body weight and improved glycemic status of diabetic individuals can provide a better platform for the improvement of insulin resistance. In the current study, the observed improvement of metabolic profile after fasting eventually supports the antioxidant system for scavenging free radical formation and thus prevented the progression of diabetes induced DNA damage. In conclusion the Ramadan fasting does not harmfully affect the metabolism of diabetic as well as healthy individuals.