Effect of adenosine deaminase activity on lipid profile in patients of type 2 diabetes mellitus

Amandeep Kaur1,∗, Sahiba Kukeraja2, Tejinder Singh3, Ishanjit Singh Sandhu4, Gurambir Singh5

1Assistant Professor, 2Professor & Head, 4,5Intern, Dept. of Bio-Chemistry, Sri Guru Ram Das Institute of Medical Science & Research, Amritsar, 3Major, Army Medical Corps

Abstract
Background: Type 2 diabetes mellitus is a worldwide endemic disease. Dyslipidemia is also a frequent disorder associated with diabetic patients. The present study is an attempt to link that elevated levels of adenosine deaminase in Type 2 diabetes mellitus patients augment hyperlipidemia.

Material and Methods: The subjects included in this study were divided into 3 groups. Group A consisted of 30 normal healthy individuals who served as controls with no history of DM. Group B consisted of 30 patients of Type 2 Diabetes Mellitus both males & females in the age group of 40-65 years on oral hypoglycaemic drugs with HbA1c <7%. Group C consisted of 30 patients of Type 2 Diabetes Mellitus both males & females in the age group of 40-65 years on oral hypoglycaemic drugs with HbA1c ≥7%. Serum levels of fasting blood sugar, HbA1c, ADA and lipid profile were estimated in all the subjects under study.

Results: All the three parameters, FBS, HbA1c and ADA levels were found to be increased in the patients of Type 2 DM as compared to controls. The mean S. Triglyceride and S. VLDL-C levels were significantly elevated in group C as compared to Group A. The mean serum levels of S. HDL-C showed a decreasing trend with increase in levels of HbA1c.

Conclusion: From the present study, it was concluded that dyslipidemia was found to be higher in diabetic patients with respect to healthy controls. The lipid abnormality (dyslipidemia) associated with type 2 diabetes typically consists of elevated triglyceride and decreased HDL-C levels.

Keywords: Diabetes Mellitus type 2, Adenosine deaminase (ADA), Glycated haemoglobin (HbA1c), Fasting blood Sugar (FBS)

Introduction
The incidence of Type 2 Diabetes Mellitus is increasing worldwide. Type 2 diabetes results from the interaction between a genetic predisposition, behavioural and environmental risk factors. (1) Although the genetic basis of type 2 diabetes has yet to be identified, there is strong evidence that such modifiable risk factors as obesity and physical inactivity are the main non-genetic determinants of the disease. (2)

Presence of lipoprotein disorders is a very common finding in diabetic patients and is the major contributor to the morbidity and mortality from cardiovascular diseases. According to ATP III guidelines, diabetic dyslipidemia is defined by the presence of high serum total cholesterol, high serum triglyceride, high LDL-C and low serum HDL in type 2 diabetic patients. (3)

The typical pattern of dyslipidemia present in type 2 patients is a raised triglyceride levels and low HDL cholesterol. Diabetic patients have a tendency of increased transport of large amounts of fatty acids to liver which are then reasssembled into triglycerides and secreted in VLDL, defective insulin action and hyperglycaemia could lead to these lipoproteins abnormalities. Control of hyperglycaemia and associated lipid abnormalities is very well identified as a modifiable risk factor among patients with type 2 diabetes. (4)

Adenosine acts directly to stimulate insulin activity via several processes such as glucose transport, lipid synthesis, pyruvate dehydrogenase activity, leucine oxidation and cyclic nucleotide phosphodiesterase activity. (5) Removal of endogenous adenosine by ADA resulted in an immediate rise in lipolytic activity. Consequently, elevated ADA levels in DM patients may augment hyperlipidemia by increasing lipolysis. (6)

Several studies were conducted to find out the lipid abnormalities in diabetes mellitus and to ascertain the effect of anti-diabetic treatment on these abnormalities. (7-10) In this prospective clinical study, we examined the serum lipid profile including total cholesterol, LDL-C, HDL-C, TG and VLDL; in addition, serum ADA activity among our type 2 diabetic patients with poor to good control.

Material and Methods
The subjects included in the present study were 60 patients of Type 2 diabetes mellitus in age group of 40-65 years of either sex, on oral hypoglycaemic drugs, attending the OPD of Department of Medicine of the Institute. A group of 30 normal healthy individuals, age...
and sex matched from the same population were served as controls.

These 90 subjects were divided into 3 groups:

- GROUP A comprised of 30 normal healthy individuals both males and females in the age group of 40-65 years from the general population who volunteered for getting included in the present study.
- GROUP B comprised of 30 patients of Type 2 Diabetes Mellitus (Non Insulin Dependent Diabetes Mellitus) both males & females in the age group of 40-65 on oral hypoglycaemic drugs with HbA1c<7%.
- GROUP C comprised of 30 patients of Type 2 Diabetes Mellitus (Non Insulin Dependent Diabetes Mellitus) age and sex matched on oral hypoglycaemic drugs with HbA1c≥7%.

Informed consent was taken from all the subjects included in the study. Patients with Type 1 diabetes mellitus, acute complications of diabetes mellitus, history of inherited disorder of lipid metabolism, liver disease, endocrine diseases affecting lipids (hypothyroidism, cushings syndrome), hypertriglyceridermia, hypertension, congestive heart failure, smoking, on drugs affecting lipid metabolism, history of acute infection, tuberculosis, gout, rheumatoid arthritis, skeletal muscle injury and renal failure were not included in this study.


Statistics: Results were analyzed by One way ANOVA and Post Hoc Turkey HSD and a probability of less than 5% (p<0.05) was considered to be statistically significant. The study was approved by the ethical committee of the institute.

Results

The statistical analysis showed sex and number distribution in these three groups was comparable. Table 1

<table>
<thead>
<tr>
<th>No.</th>
<th>FBS</th>
<th>HbA1c</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group</td>
<td>Mean±SD</td>
<td>Comparison</td>
</tr>
<tr>
<td>A</td>
<td>30</td>
<td>82.00±13.00</td>
</tr>
<tr>
<td>B</td>
<td>30</td>
<td>126.12±22.71</td>
</tr>
<tr>
<td>C</td>
<td>30</td>
<td>136.97±24.88</td>
</tr>
</tbody>
</table>

Table 2: Showing FBS & HbA1c in control and study groups

The mean FBS levels of Group A were 82.00±13.00 mg/dl, Group B were 126.12±22.71 mg/dl and the corresponding values among Group C subjects were 136.97±24.88 mg/dl. In the present study, the mean FBS levels of Group B and Group C were found to be highly significantly higher than Group A (p<0.001). Although the mean FBS levels of Group C were higher than Group B but the difference was statistically not significant (p=0.115). It was observed that mean HbA1c levels in Group A were 5.75±0.46%, in Group B were 6.09±0.56% and the corresponding values among Group C were 8.72±1.35%. From this study it was observed that the difference in the levels of HbA1c was found to be insignificant between Group B and Group A (p=0.300).

In the present study the mean serum ADA levels in Group A were 17.30±7.28 U/L, in Group B were 30.04±10.41 U/L whereas in Group C were 44.23±16.14 U/L. Statistical analysis showed that the mean serum ADA levels of Group C were significantly higher than Group B (p<0.001) and the levels of ADA were significantly higher in both Group B and Group C as compared to Group A (p<0.001).

Table 3
Table 3: Comparison of Serum Adenosine Deaminase (ADA) levels in three groups

<table>
<thead>
<tr>
<th>Group</th>
<th>N</th>
<th>Range (U/L)</th>
<th>Mean±SD (U/L)</th>
<th>95% CI</th>
<th>Comparison</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group A</td>
<td>30</td>
<td>2.5-30.0</td>
<td>17.30±7.28</td>
<td>14.58-20.02</td>
<td>Group A vs. B</td>
<td>&lt;0.001***</td>
</tr>
<tr>
<td>Group B</td>
<td>30</td>
<td>12.7-55.5</td>
<td>30.04±10.41</td>
<td>26.16-33.93</td>
<td>Group A vs. C</td>
<td>&lt;0.001***</td>
</tr>
<tr>
<td>Group C</td>
<td>30</td>
<td>11.6-82.2</td>
<td>44.23±16.14</td>
<td>38.21-50.26</td>
<td>Group B vs. C</td>
<td>&lt;0.001***</td>
</tr>
</tbody>
</table>

N: Number of cases; SD: Standard Deviation; CI: Confidence Interval; ***P<0.001; Highly Significant

Serum cholesterol ranged from 132-265 mg/dl with mean of 186.86±35.42 mg/dl in Group A. Among the subjects of Group B, the values of cholesterol ranged from 103-280 mg/dl with mean of 187.71±45.13 mg/dl and in Group C the values of cholesterol ranged from 120-275 mg/dl with mean of 188.59±44.48 mg/dl. The difference was statistically not significant between the above three groups (p > 0.05).

In Group A, the values of triglyceride ranged from 82-350 mg/dl with mean of 155.40±71.57 mg/dl. Among the subjects of Group B, the values of triglyceride ranged from 58-291 mg/dl with mean of 179.16±58.76 mg/dl and in Group C the values of cholesterol ranged from 120-770 mg/dl with mean of 244.45±161.94 mg/dl. The mean of triglyceride levels of Group C were significantly higher than Group A (p=0.005) while the difference between Group A and Group B was not significant (p=0.670). Similarly in case of Group B and Group C the difference was statistically not significant (p = 0.055).

In Group A, the values of HDL ranged from 32-72 mg/dl with mean of 50.65±11.25 mg/dl. Among the subjects of Group B, the values of HDL ranged from 28-61 mg/dl with mean of 41.52±7.11 mg/dl and in Group C the values of HDL ranged from 25-66 mg/dl with mean of 43.27±9.95 mg/dl. The mean values of HDL were significantly higher in Group A than Group B (p=0.001). Similarly the values of mean HDL in Group A were significantly higher than Group C (p=0.010). But in case of Group B and Group C the difference was statistically not significant (p=0.760).

The values of VLDL in Group A ranged from 16.5-77.1 mg/dl with mean of 32.13±15.65 mg/dl. Among the subjects of Group B, the values of VLDL ranged 11.6-58.2 from with mean of 35.77±11.81 mg/dl and in Group C the values of VLDL ranged from 21.2-154 mg/dl with mean of 48.90±32.38 mg/dl. The mean of VLDL levels of Group C were significantly higher than Group A (p=0.011) while the difference between Group A and Group B was not significant (p=0.795). The difference among Group B and Group C was statistically not significant (p=0.057).

Thus it was observed that values of S.TG and S.VLDL-C were significantly higher in Group C (HbA1c>7%) when compared with Group A. But the levels of S.HDL-C showed a decreasing trend with the increase in levels of HbA1c. Table 4

Table 4: Comparison of Serum Lipids and Lipoprotein Levels in three groups

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Range and Mean (mg/dl)</th>
<th>Group A</th>
<th>Group B</th>
<th>Group C</th>
<th>Comparison</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>S.TOTAL CHOL</td>
<td>Range</td>
<td>132-265</td>
<td>103-280</td>
<td>120-275</td>
<td>Group A vs. B</td>
<td>0.997NS</td>
</tr>
<tr>
<td></td>
<td>Mean</td>
<td>186.8±35.42</td>
<td>187.71±45.13</td>
<td>188.59±44.48</td>
<td>Group A vs. C</td>
<td>0.968NS</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Group B vs. C</td>
<td>0.996NS</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>S.TG</td>
<td>Range</td>
<td>82-350</td>
<td>58-291</td>
<td>106-770</td>
<td>Group A vs. B</td>
<td>0.670NS</td>
</tr>
<tr>
<td></td>
<td>Mean</td>
<td>155.40±71.57</td>
<td>179.16±58.76</td>
<td>244.45±161.94</td>
<td>Group A vs. C</td>
<td>0.005**</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Group B vs. C</td>
<td>0.055NS</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>S.HDL-C</td>
<td>Range</td>
<td>32 – 72</td>
<td>28 – 61</td>
<td>25 – 66</td>
<td>Group A vs. B</td>
<td>0.001**</td>
</tr>
<tr>
<td></td>
<td>Mean</td>
<td>50.65±11.25</td>
<td>41.52±7.11</td>
<td>43.27±9.95</td>
<td>Group A vs. C</td>
<td>0.010*</td>
</tr>
<tr>
<td>S.VLDL-C</td>
<td>Range</td>
<td>16.5-77.1</td>
<td>11.6-58.2</td>
<td>21.2-154</td>
<td>Group B vs. C</td>
<td>0.760NS</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Group A vs. B</td>
<td>0.795NS</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Group A vs. C</td>
<td>0.011*</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Discussion
The incidence of diabetes mellitus has been increasing rapidly in the past 2 decades and this is accompanied by the notably high prevalence of associated disorders, such as hypertension, atherogenic lipid profile and metabolic syndrome.\(^{(19)}\)

Our study provided the evidence for the presence of high prevalence of dyslipidemia in type 2 diabetic patients. Although our patients received oral hypoglycaemic agents for hyperglycemia, a significant proportion of them had abnormal lipid profile. The most frequent was hypertriglyceridemia and the least frequent was elevated total cholesterol.

In present study, the mean serum ADA levels in patients of diabetes mellitus type 2 were highly significantly higher than controls (Table 3) and the mean serum triglycerides levels were higher in patients of diabetes mellitus type 2 than controls but the difference was significant in Group C and Group A (Table 4). Hence it was observed that with increase in S. ADA levels there was an increase in S. Triglyceride levels in patients of Type 2 diabetes mellitus, the reason for this finding could be that adenosine inhibited adenylate cyclase through A1 adenosine receptors, resulting in inhibition of lipolysis. Removal of adenosine by ADA resulted in an immediate rise in lipolytic activity. Consequently, elevated ADA levels in DM patients may augment hyperlipidemia by increasing lipolysis.\(^{(6)}\)

Study by Schwabe U et al\(^{(20)}\) also supported this; they were able to show that adenosine is released by fat cells into the incubation medium in amounts which inhibit hormone-induced lipolysis and cyclic AMP accumulation. Adenosine and related compounds are known to mimic the action of insulin on glucose and lipid metabolism in adipose tissue.

Obesity and insulin resistance have been suggested to contribute to the pathogenesis of hypertriglyceridemia in type 2 diabetics. This was in accordance to study by Peterson CM et al.\(^{(21)}\)

Conclusion
In this study, it was shown that with increase in S. ADA levels there was an increase in S. TG levels in patients of type 2 DM. Removal of adenosine by ADA resulted in an immediate rise in lipolytic activity. However, despite some differences with the reported studies, as a general, S. TG, total cholesterol, LDL-C levels were higher and HDL-C levels lower than in the control subjects, consistent with the literature. Further studies are needed, especially the elucidation of the role of S.ADA levels on the lipid profile of type 2 DM patients with fair to poor control.

Bibliography

<table>
<thead>
<tr>
<th></th>
<th>Mean</th>
<th>Range</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>S.LDL-C</td>
<td>32.13±15.65</td>
<td>24.0-178</td>
<td>35.77±11.81</td>
<td>13.9-192</td>
<td>48.90±32.38</td>
</tr>
<tr>
<td></td>
<td>Group B vs. C</td>
<td>0.057(\text{NS})</td>
<td>Group A vs. B</td>
<td>0.760(\text{NS})</td>
<td>Group A vs. C</td>
</tr>
<tr>
<td>Mean</td>
<td>102.09±37.03</td>
<td>109.70±44.66</td>
<td>96.44±42.88</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group C vs.</td>
<td>0.437(\text{NS})</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

N: Number of cases; SD: Standard Deviation; CI: Confidence Interval; NS: p>0.05; Not Significant