Abstract: Aim: The study aimed to ascertain whether there is variation in the fibrinolytic/coagulation component of diabetes associated with disease progression to macro vascular complications and if D-dimer can discriminate such variation. Methods: 343 participants were selected based on clinical status and divided into 7 groups: control, family history of diabetes, pre-diabetes with/without CVD, diabetes with/without CVD and CVD only. Plasma D-dimer was tested. Statistical analysis was performed on log normalized data by ANOVA, Fisher’s LSD post hoc test. After the initial analysis, data was classified and re-analysed by quartiles, interquartile range and 95th-percentile. Results: An overall significant difference between groups (p < 0.002) and a steady rise in D-dimer levels that became increasingly higher than control as the disease progressed from prediabetes to cardiovascular complications was observed. Statistically significant difference was observed between control versus diabetes (p < 0.0005). Analysis of data by quartiles and percentiles gave qualitatively similar results, but greater significant difference between control versus preprediabetes at 3rd quartile and interquartile range (p < 0.014). Conclusion: We report changes in D-dimer levels that may indicate diabetes disease progression to macrovascular complications. Using D-dimer in conjunction with other biomarkers to identify stages of disease progression, commencing from prediabetes and continuing to development of asymptomatic and clinical cardiovascular disease in diabetes mellitus, is worthy of consideration.
D-dimer identifies stages in the progression of diabetes mellitus from family history of diabetes to cardiovascular complications

Short title: D-dimer changes in progression of diabetes

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Summary

Aim: The study aimed to ascertain whether there is variation in the fibrinolytic/coagulation component of diabetes associated with disease progression to macrovascular complications and if D-dimer can discriminate such variation.

Methods: 343 participants were selected based on clinical status and divided into 7-groups: control, family history of diabetes, pre-diabetes with/without CVD, diabetes with/without CVD and CVD only. Plasma D-dimer was tested. Statistical analysis was performed on log normalized data by ANOVA, Fisher’s LSD post hoc test. After the initial analysis, data was classified and re-analyzed by quartiles, interquartile range and 95th-percentile.

Results: An overall significant difference between groups (p < 0.002) and a steady rise in D-dimer levels that became increasingly higher than control as the disease progressed from prediabetes to cardiovascular complications was observed. Statistically significant difference was observed between control versus diabetes (p < 0.0005). Analysis of data by quartiles and percentiles gave qualitatively similar results, but greater significant difference between control versus preprediabetes at 3rd quartile and interquartile range (p < 0.014).

Conclusion: We report changes in D-dimer levels that may indicate diabetes disease progression to macrovascular complications. Using D-dimer in conjunction with other biomarkers to identify stages of disease progression, commencing from prediabetes and continuing to development of asymptomatic and clinical cardiovascular disease in diabetes mellitus, is worthy of consideration.
INTRODUCTION

The physiology of haemostasis involves a balance between coagulation and fibrinolysis, the mechanisms involved being complex. Fibrinolysis is a process whereby a covalently cross-linked fibrin clot is broken down by the proteolytic enzyme plasmin, producing cross-linked products of fibrin (D-dimer). Thus, fibrin D-dimer is a characteristic degradation product of cross-linked fibrin and an index of coagulation activity. Elevated plasma D-dimer levels have been reported in subjects with coronary and peripheral atherosclerosis. Hence D-dimer has been proposed as a potential diagnostic and management tool for cardiovascular disease (CVD), including atherosclerosis and coronary artery disease. However, whether there are significant differences in plasma D-dimer levels at different stages of diabetes associated with disease progression to macrovascular complications have not yet been reported.

Elevated plasma D-dimer level in diabetes mellitus

Sommeijer et al. have reported that some coagulation markers are elevated in DM, and our preliminary reports have shown increased D-dimer levels in DM. Yano et al 2003, however, suggested that in type 2 diabetes, there is hypo-fibrinolysis as thrombomodulin-thrombin complex, which is formed on intact vascular endothelium, may trigger thrombin-activatable fibrinolysis inhibitor (TAFI). Indeed, this is supported by the observation that hyperglycaemia and insulin resistance enhance the synthesis and secretion of plasminogen-activator inhibitor Type 1, as well as being associated with increased blood levels of TAFI. These results suggest that both fibrinolysis and therefore the generation of D-dimer are reduced in DM.

Nevertheless, when there is hypercoagulation, there must be commensurate hyperfibrinolysis as an inherent physiological response to prevent the development of thromboembolism. If hypercoagulation and hypofibrinolysis occur concurrently in DM as implied by Yano and his group,
there would be obvious immediate effects of thromboembolism in almost all people with DM especially among the undiagnosed population. Therefore, the cause of the high or low level of D-dimer in DM has yet to be adequately explained. Meanwhile, there is the observation that depletion of natural cellular antioxidants by hyperglycaemia leads to enhanced activation of the nuclear redox-sensitive transcription factor, which in turn up-regulates events at the gene level including pro-coagulant tissue factor.\textsuperscript{16} However, whether such up-regulation of the pro-coagulant tissue factor progresses or varies with the development of hyperglycaemia toxicity associated with cardiovascular complication of diabetes has yet to be investigated.

Summarily, D-dimer is a marker for fibrinolytic/coagulation processes associated with disease conditions such as deep vein thrombosis and atherosclerosis. Increased D-dimer values have been reported for diabetes mellitus, but the question as to whether there is variation in D-dimer levels associated with the progression of diabetes mellitus including progression to cardiovascular disease is yet to be addressed.

**Hypotheses and objective**

DM is part of the constellation of disorders referred to as metabolic syndrome whose characteristics include (among other things) fibrinolytic and coagulation changes. We know that hyperglycaemia enhances hypercoagulation\textsuperscript{16,17} and that cardiovascular complications in DM are speculated to start prior to diagnosis, which is the pre-diabetes stage.\textsuperscript{18} Therefore, we hypothesized that plasma D-dimer levels may differ in people at different stages of DM. The objective of this study was to investigate whether D-dimer levels vary with the progression of DM, and whether such variation identifies the different stages of DM.

**MATERIALS AND METHODS**
The study was approved by the Ethics in Human Research Committee of Charles Sturt University as part of a diabetes complications screening project. We measured plasma D-dimer levels in 343 participants (195 females and 148 males). The participants were selected based on medical history obtained from a health questionnaire and from recent pathology reports, where blood glucose levels, serum creatinine, urinary albumin excretion, any established diagnosis of diabetes, CVD and/or kidney disease were recorded. Family history of diabetes, obesity, age, gender, smoking, alcohol consumption and medication were also recorded. The participants were divided into seven groups as shown in Table 1.

PLACE TABLE 1 HERE

In order to avoid the confounding effects of family history of diabetes, individuals that were otherwise healthy but had primary or secondary relatives with diabetes were included in a separate group (FH-DM). In the control group, participants were included if their blood glucose readings were ≤ 5.5 mmol/L, body mass index ≤ 30 kg/m² and no CVD nor kidney disease was reported or identified. Pre-diabetes in this study was defined as persons with impaired fasting glucose (IFG), impaired glucose tolerance (IGT)¹⁹ or participants that are otherwise healthy but presented with fasting blood sugar levels ≥ 5.6 mmol/L and ≤ 7.0 mmol/L.²⁰ Other participants that were included were those with established diagnosis of CVD, high blood pressure (BP), DM or clinically established IFG/IGT.

Cardiovascular complication of diabetes in this study refers to coronary artery disease. General exclusion criteria included history of kidney disease (positive serum creatinine/urinary albumin) and other macrovascular diseases. Other exclusion criteria were a body mass index >30 kg/m², chronic alcoholism, and smoking. Participants with any other ongoing disease condition or medication were
excluded, except those in the CVD and/or DM groups who were being treated with prescription drugs that are primarily hypoglycaemic, anticoagulant, anti-inflammatory or hypolipidaemic. Any participant taking a non-steroidal anti-inflammatory drug (NSAID) and/or antidepressant was also excluded.\(^{21, 22}\)

A participant was considered to have CVD only if it was indicated on his/her health questionnaire to have established diagnosis of high BP or other forms of CVD and/or pathology reports. Blood pressure and 12-lead electrocardiogram were performed as part of the screening process to confirm absence or presence of CVD. Diastolic pressure > 90 mmHg and/or systolic pressure > 140 mmHg at lying position were taken as cut-off indicating hypertension. Any participant that showed abnormal readings in our screening and does not belong to either CVD or DM+CVD group was excluded. 12-lead ECG identified those with asymptomatic CVD and included arrhythmia and signs of coronary artery disease. Blood glucose level was performed to assess diabetes status and to recruit the prediabetes group members. For D-dimer determination, venous blood was collected from the cubital fossa into a heparinized tube and mixed thoroughly. The plasma was separated and stored frozen at -78 °C until tested using the MiniQuant\textsuperscript{®} procedure from Immunodiagnostics.\(^{23}\) Separate finger tip capillary blood was collected for the blood glucose levels that were determined immediately using the Accu-Chek\textsuperscript{®} system (Roche Australia Pty Ltd).

Our data were not normally distributed, similar to the observation of Lowe et al.\(^{24}\) Therefore, data were transformed to natural logarithms for analysis and back-transformed to obtain the values given in Table 2. Two phases of statistical analysis were performed by ANOVA followed by Fisher’s LSD post hoc test using SPSS (version ‘14 for Windows’) statistical package provided by the university. Firstly, data analysis was performed to observe differences between the plasma D-dimer levels for the different groups. Secondly, data was ranked and divided into quartiles and corresponding quartiles of the different groups were re-analyzed to observe consistency or variation with the first analysis, where the whole data was analyzed. Similar to the quartile analysis, the corresponding
interquartile ranges and 95th percentiles of the groups were also re-analyzed. Correlation analysis was performed to determine correlation for gender or age using the correlation analysis tool (Microsoft Excel’s Analysis ToolPak).

In our preliminary study for the determination of predictive importance of D-dimer for identifying prediabetes, we studied D-dimer results and total cholesterol (TC) in a group of 45 people identified with prediabetes (20 females and 25 males) with or without established diagnosis of high blood pressure. TC was determined using Cholestech® procedure.

RESULTS

We determined that there was no correlation between age and D-dimer in our data, although it has been speculated that D-dimer might be influenced by age. We also determined that gender difference was statistically not significant on plasma D-dimer levels in our data. Therefore, we did not discriminate between genders in the groups nor did we control for age and gender.

The results from our first analysis indicated that D-dimer gradually increases as disease progresses sequentially from normal to pre-diabetes, pre-diabetes with CVD, DM without CVD and DM with CVD (fig 1). ANOVA showed a significant difference between groups (p < 0.002), while Fisher’s LSD post hoc test showed that the D-dimer concentration (μg/L) was significantly lower in the control group compared to the DM+CVD group (p < 0.007), and the DM alone group (p < 0.0005).

PLACE FIGURE 1 HERE
To allow for some participants having normal or minimal changes in D-dimer levels with diabetes but good blood sugar control, we divided the D-dimer results into quartile ranges. ANOVA showed that the trends of plasma D-dimer level changes were the same. That is, there was consistent gradual increase in plasma D-dimer levels as disease progresses sequentially from normal to pre-diabetes, pre-diabetes with CVD, DM with CVD and DM without CVD (fig 2). Furthermore, we also observed that across the quartiles, the mean concentration of plasma D-dimer in the FH-DM group was not consistent in comparison with other groups (fig 2).

In all the aforementioned analyses, plasma D-dimer levels were higher in the prediabetes group compared with the control, but statistically significant only in the 3rd quartile range (p < 0.0001) and in the interquartile range (p < 0.014). The significant differences were greater between the control group and the preDM+CVD group, as plasma D-dimer was significantly lower in the control group in the 3rd quartile range (p < 0.0001) and in the interquartile range (p < 0.011). A summary of descriptive characteristics of the groups and statistical results of plasma D-dimer (μg/L) for the different groups as well as p-values indicating increasing significant difference in DM progression is presented in table 2. FH-DM and CVD groups have been discretionally located at both ends of the tables and figures, in order to appropriately visualize the trend in DM progression starting from the control group.
To determine whether D-dimer adds information for identifying DM and/or CVD, we compared the D-dimer results to total cholesterol for a group with prediabetes and either with our without hypertension. Four of forty-five participants proved negative for both tests. Among the remaining forty-one, TC identified twelve positives, out of which D-dimer identified eight. The eight that were picked by D-dimer were also positive for blood pressure (BP). That is, no participant showed positive for both TC and D-dimer without having high BP. Among the thirty-four that showed high BP, sixteen had raised D-dimer while ten had raised TC. This represents 17.8% of possible early CVD in this prediabetes cohort where hypertension is not identified by TC.

Our study highlighted that differentiating between stages of diabetes progression using the 1st and 4th quartile range can differentiate between thirteen pair-wise comparisons out of stages of progression of a possible twenty-one. This suggests a role for D-dimer in diabetes assessment (Table 3). Specifically of clinical interest is that high D-dimer differentiated between preDM, DM and DM + CVD, as well as between preDM + CVD, DM + CVD and DM. A statistically significant difference was also obtained between control, DM + CVD as well as between preDM and DM. Both low and high levels of D-dimer.

DISCUSSION

The results of this study support several previously published studies that reported increased D-dimer in DM as well as CVD. From the results presented (Fig 1), it can be seen that the
mean level of D-dimer is lower in the control group compared to the diabetes group (p < 0.001), which strengthens our earlier reported conclusion.\textsuperscript{11} We did observe that in all the analyses including the different quartile and percentile ranges, the levels of plasma D-dimer in the pre-diabetes group remained consistently higher compared to the control (figs 1 and 2), with statistically significant differences obtained at the 3\textsuperscript{rd} quartile range (p < 0.0001) and at the interquartile range (p < 0.014). Although the knowledge of coagulation/fibrinolysis imbalance in DM is not new, the observation that plasma D-dimer levels increase along with disease progression, from the pre-diabetes stage with cardiovascular complications, as shown in Fig 1 and Table 3 has not been reported.

The above observations suggest a unique, at least complementary role for D-dimer in identification of early CVD among pre-clinical DM population and assessing disease progression. Furthermore, the data draw attention to the postulate that cardiovascular complications of diabetes may start during the pre-diabetes stage and involve inflammatory activities.\textsuperscript{27} Figure 1 indicates that there may be different pathophysiological interactions involved when CVD is present in people with prediabetes compared to when CVD is present in those with diabetes compared to those with diabetes but no CVD. We observed that D-dimer levels are greater in the preDM + CVD group compared to the preDM and group but this relationship is reversed when comparing DM to DM + CVD.

CVD is common but under-diagnosed among diabetes patients, and the asymptomatic stage of CVD could be screened in the diabetes population.\textsuperscript{28} There are difficulties in accurately diagnosing CVD in the early stages. Thus, there is the need to identify new markers that would be useful for assessing asymptomatic or early stages of the disease in individuals at risk.\textsuperscript{29} We suggest that the use of plasma D-dimer along with conventional risk factors such as total cholesterol for early identification of pre-diabetes may be of value as indicated by in Figure 3.

There was no statistical difference between diabetes with/without CVD groups compared to CVD alone. However, it was interesting to observe that the group that has diabetes alone consistently
presented the highest D-dimer levels, despite all members of the CVD and DM+CVD groups having established CVD. As a vast majority of the CVD and DM+CVD members are being treated with a combination of hypoglycaemic and cardiovascular medications (some of which possess anticoagulant and/or anti-inflammatory properties), it is possible that the observed lower plasma D-dimer levels in DM+CVD and CVD groups compared to the DM group could be due to CVD medications. Yet, we also noted that between preDM and preDM+CVD, plasma D-dimer levels were consistently higher in the latter but not statistically different. This observation may demonstrate that the hypercoagulability in DM is associated with progression of the disease, as well as supporting the view that diabetes patients may benefit from anti-inflammatory therapy.\textsuperscript{30}

Furthermore, we observed that the mean plasma D-dimer level in the preDM group was consistently lower compared to DM+CVD group and much lower compared to DM group (fig 1 and fig 2). Using the interquartile range analysis, we obtained statistically significant difference between preDM group compared to both the DM+CVD group (p < 0.02) and the DM group (p < 0.001). Bearing in mind that most of the DM+CVD group members are in CVD management that could reduce thromboatherogenesis, it is likely that our observation is a function of the benefits of CVD management for diabetes patients.\textsuperscript{30}

Comparing stages of disease progression using the 1\textsuperscript{st} quartile range indicated that only six pair-wise comparisons were statistically significant. Of interest is that only using the 1\textsuperscript{st} quartile data we obtained statistically significant differences between the FH stage and later stages of disease progression. When we used the 4\textsuperscript{th} quartile range, which is most often accepted as indicating coagulation and fibrinolytic changes associated with diabetes, eight pair-wise comparisons were statistically significant (Table 3). The literature has suggested that both low and high levels of D-dimer are associated with diabetes and therefore using both the 1\textsuperscript{st} and 4\textsuperscript{th} quartile ranges we obtained thirteen of twenty-one statistically significant pair-wise comparisons. These differences
between D-dimer levels and disease progression may be of value when deciding which medication to prescribe. Thus some ACE inhibitors have anti-inflammatory activity and could be used in preference to other ACE inhibitors.

What this report adds to current knowledge is firstly that in diabetic progression, D-dimer level starts increasing at the pre-clinical stage of disease development (fig 1). We speculate that early intervention and risk reduction of atherothrombotic events that are indicated by D-dimer levels may represent good health care. Secondly, our report provides evidence to speculate that besides the established involvement of diabetic lipidaemia in cardiovascular complications, diabetic hypercoagulability is another pathological process in the development of atherosclerosis and coronary artery disease.

When we compared D-dimer with BP and TC, two of the traditional risk markers of CVD, we showed that no individual presented both high total cholesterol and high D-dimer without presenting high BP (Fig 3). This suggests that laboratory determination of both TC & D-dimer could reduce chances of making a false negative decision about CVD complication. We also observed that among the 34 that showed high BP, 16 had raised D-dimer while 10 had raised TC. This represents 17.8% of possible early CVD in this prediabetes cohort that was not identified by TC. This suggests a unique predictive value of D-dimer for early CVD among pre-clinical DM population. It has been reported that over 50% of all future vascular events occur in persons without dyslipidaemia, and that plasma D-dimer is a novel risk marker for the prediction of future cardiovascular events.33 Thus, we speculate that inclusion of D-dimer as a complementary test can improve identification of asymptomatic macrovascular events in prediabetes as well as reduce chances of making a false negative decision about CVD complication. However, a large prospective study will be necessary to validate this speculation.
We observed that FH-DM group was the only group that showed inconsistent relative comparison with other groups. Although the pathophysiology in people with family history of diabetes is not the focus of this study, we suggest that it may contribute additional knowledge to do further study that will explain this observation. Associated with the diversity of available analysis kits for the determination of D-dimer levels is the corresponding diversity of what is considered a normal or reference range.\(^{34} 35\) As most work has been carried out with deep vein thrombosis and pulmonary oedema patients, our study adds information in terms of providing reference values for controls and people with one or more of the traits of diabetes. A larger cohort that we are now analyzing will provide possible cut-off points for the identification of diabetes disease progression.

**Conclusion:**

We report that there is an identifiable, steady, unidirectional increase in D-dimer levels in the progression of diabetes mellitus in this study. A negative D-dimer result requires support of other tests to exclude cardiovascular events. Positive D-dimer level is salient in pre-diabetes conditions. On the basis of a known type of vascular event and the feasibility of follow-up, we speculate that using D-dimer determination to complement cholesterol profile could improve risk assessment for early CVD identification and intervention during pre-clinical diabetes.

**ACKNOWLEDGEMENT**

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REFERENCES


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Figure 1: Histogram plot of Mean ± SD values of the groups (ANOVA: P < 0.002)

Figure 2: Histogram plot showing pattern of variation in ‘means’ of plasma D-dimer values at different quartile and percentile ranges, in relation to disease progression. Key for groups: A = FH group; B = control; C = preDM; D = preDM+CVD; E = DM+CVD; F = DM; G = CVD
Figure 3: Venn diagram showing distribution of the 41 positives, i.e. number of positives for only one test and number of positive for different combinations of tests.

Tables
Table 1: Summary of grouping and selection criteria of participants

<table>
<thead>
<tr>
<th>Group</th>
<th>Health status or disease diagnosis</th>
<th>Positive tests at screening clinic</th>
<th>Ongoing medication</th>
<th>Other group-specific exclusion criteria, beside positive tests at screening clinic</th>
</tr>
</thead>
<tbody>
<tr>
<td>FH-DM</td>
<td>Persons with up to second degree family history of DM</td>
<td>-</td>
<td>-</td>
<td>Any ongoing medication or known disease condition</td>
</tr>
<tr>
<td>Control</td>
<td>Healthy persons</td>
<td>-</td>
<td>-</td>
<td>Family history of diabetes, any known disease condition</td>
</tr>
<tr>
<td>Pre-DM</td>
<td>Suspected pre-diabetes</td>
<td>FBS/RBS + Lipid profile ±</td>
<td>-</td>
<td>Family history of diabetes, any known disease condition</td>
</tr>
<tr>
<td>Pre-DM+CVD</td>
<td>Suspected pre-diabetes persons that has high BP</td>
<td>FBS + BP +</td>
<td>Anticoagulants Anti-inflammatory</td>
<td>Family history of diabetes, any known disease condition</td>
</tr>
<tr>
<td>DM+CVD</td>
<td>Both diabetes and high blood pressure (BP) or other forms of CVD present</td>
<td>FBS/RBS ± BP ± ECG ±</td>
<td>Anticoagulants Anti-inflammatory Beta-blockers or Hypoglycaemics Hypolipidaemics</td>
<td>Any other co-morbidity or medication</td>
</tr>
<tr>
<td>DM</td>
<td>Type 1 or type 2 diabetes mellitus persons</td>
<td>FBS/RBS ± Lipid profile ±</td>
<td>Hypoglycaemics Hypolipidaemics</td>
<td>HbA1c + Any co-morbidity or other medication</td>
</tr>
<tr>
<td>CVD</td>
<td>Persons already diagnosed as having known BP or other forms of CVD</td>
<td>BP ± ECG ± Lipid profile ±</td>
<td>Anticoagulants Anti-inflammatory Beta-blockers or</td>
<td>HbA1c + Any co-morbidity or other medication</td>
</tr>
</tbody>
</table>
Table 2: A descriptive characteristics and statistical results of plasma D-dimer (μg/L) for the different groups and p-values indicating increasing significant difference in DM progression

<table>
<thead>
<tr>
<th></th>
<th>FH-DM</th>
<th>Control</th>
<th>PreDM</th>
<th>PreDM + CVD</th>
<th>DM + CVD</th>
<th>DM</th>
<th>CVD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean (μg/L)</td>
<td>162</td>
<td>146</td>
<td>188</td>
<td>278</td>
<td>236</td>
<td>250</td>
<td>148</td>
</tr>
<tr>
<td>SD</td>
<td>24</td>
<td>23</td>
<td>25</td>
<td>29</td>
<td>28</td>
<td>23</td>
<td>33</td>
</tr>
<tr>
<td>Median</td>
<td>182</td>
<td>140</td>
<td>203</td>
<td>235</td>
<td>218</td>
<td>264</td>
<td>151</td>
</tr>
<tr>
<td>Minimum</td>
<td>23</td>
<td>23</td>
<td>23</td>
<td>23</td>
<td>11</td>
<td>34</td>
<td>23</td>
</tr>
<tr>
<td>Maximum</td>
<td>1913</td>
<td>1176</td>
<td>2511</td>
<td>3762</td>
<td>3756</td>
<td>1372</td>
<td>2990</td>
</tr>
<tr>
<td>N = 343</td>
<td>32</td>
<td>50</td>
<td>34</td>
<td>39</td>
<td>75</td>
<td>59</td>
<td>54</td>
</tr>
<tr>
<td>F/M = (195/148)</td>
<td>15/19</td>
<td>27/23</td>
<td>26/13</td>
<td>36/23</td>
<td>36/39</td>
<td>35/19</td>
<td>20/12</td>
</tr>
<tr>
<td>$X_{DM}$*</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>7 years</td>
<td>9 years</td>
<td>NA</td>
</tr>
<tr>
<td>ANOVA p-value</td>
<td>NS</td>
<td>NA</td>
<td>NS</td>
<td>NS</td>
<td>0.001</td>
<td>0.0005</td>
<td>0.015</td>
</tr>
<tr>
<td>3rd quartile p-value</td>
<td>NS</td>
<td>NA</td>
<td>0.0001</td>
<td>0.0001</td>
<td>0.0001</td>
<td>0.0001</td>
<td>0.0001</td>
</tr>
<tr>
<td>Int’quartile range</td>
<td>NS</td>
<td>NA</td>
<td>0.014</td>
<td>0.011</td>
<td>0.0001</td>
<td>0.0001</td>
<td>0.0001</td>
</tr>
</tbody>
</table>

* = Mean duration of DM; NA = not applicable; NS = not significant; p-values = compared to control group.
Table 3: Pair-wise comparisons for 1\textsuperscript{st} and 4\textsuperscript{th} quartiles for diabetes disease progression to macrovascular disease.

<table>
<thead>
<tr>
<th></th>
<th>Contr</th>
<th>FH</th>
<th>preDM</th>
<th>preDM+CVD</th>
<th>DM+CVD</th>
<th>DM</th>
<th>CVD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Contr***</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>FH</td>
<td>0.038**</td>
<td>NS</td>
<td></td>
<td>0.032</td>
<td>0.01</td>
<td>0.002</td>
<td>0.002</td>
</tr>
<tr>
<td>preDM</td>
<td>NS</td>
<td>NS</td>
<td></td>
<td>NS</td>
<td></td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>preDM+CVD</td>
<td>NS</td>
<td>NS</td>
<td></td>
<td>NS</td>
<td></td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>DM+CVD</td>
<td>0.002</td>
<td>NS</td>
<td>0.035</td>
<td>0.05</td>
<td></td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>DM</td>
<td>0.0001</td>
<td>NS</td>
<td>0.007</td>
<td>0.008</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>CVD</td>
<td>0.032</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td></td>
<td>NS</td>
<td>NS</td>
</tr>
</tbody>
</table>

*top right shows p values for group comparisons using 1\textsuperscript{st} quartile; bottom left shows group comparisons using 4\textsuperscript{th} quartile.

** p values significant for p < 0.05.

*** Contr = control; FH = family history; preDM = prediabetes; CVD = cardiovascular disease; DM = diabetes mellitus.

**** NS = non-significant.