Microparticle Shedding by Erythrocytes, Monocytes and Vascular Smooth Muscular Cells Is Reduced by Aspirin in Diabetic Patients

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### Baseline Characteristics of the 43 Diabetic Patients and the 38 Control Subjects Studied

<table>
<thead>
<tr>
<th></th>
<th>PATIENTS (n=43)</th>
<th>CONTROLS (n=38)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>55±10</td>
<td>59±13</td>
<td>.133</td>
</tr>
<tr>
<td>Males [n (%)]</td>
<td>24 (55.8)</td>
<td>17 (44.7)</td>
<td>.217</td>
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<tr>
<td>Current smokers [n (%)]</td>
<td>9 (20.9)</td>
<td>7 (18.4)</td>
<td>.617</td>
</tr>
<tr>
<td>Diabetis Mellitus [n (%)]</td>
<td>43 (100)</td>
<td>0 (0)</td>
<td>&lt;.0001</td>
</tr>
<tr>
<td>Dyslipidemia [n (%)]</td>
<td>40 (93.0)</td>
<td>34 (89.5)</td>
<td>.722</td>
</tr>
<tr>
<td>Hypertension [n (%)]</td>
<td>40 (90.7)</td>
<td>25 (65.8)</td>
<td>.064</td>
</tr>
<tr>
<td>Body Mass Index (kg/m²)</td>
<td>25.70±3.04</td>
<td>26.89±3.29</td>
<td>.086</td>
</tr>
<tr>
<td>Body Mass Index &gt;25 kg/m² [n (%)]</td>
<td>21 (48.8)</td>
<td>22 (57.9)</td>
<td>.485</td>
</tr>
<tr>
<td>Systolic Blood Pressure (mmHg)</td>
<td>138±14</td>
<td>145±22</td>
<td>.072</td>
</tr>
<tr>
<td>Diastolic Blood Pressure (mmHg)</td>
<td>81±8</td>
<td>82±10</td>
<td>.510</td>
</tr>
<tr>
<td>Statins [n (%)]</td>
<td>33 (76.7)</td>
<td>32 (84.2)</td>
<td>.806</td>
</tr>
<tr>
<td>Acetylsalicylic acid [n (%)]</td>
<td>43 (100)</td>
<td>0 (0)</td>
<td>&lt;.0001</td>
</tr>
</tbody>
</table>

*P* value from one-way ANOVA for quantitative variables and from Chi-square analysis for qualitative variables.
Table 2 of the supplementary material.

Comparison of Circulating Microparticle Levels in the 38 Controls and the 43 Patients Before and After Acetylsalicylic Acid Intervention

<table>
<thead>
<tr>
<th></th>
<th>DIABETIC PATIENTS</th>
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<tbody>
<tr>
<td></td>
<td>AV⁺ MPs/µL PFP</td>
<td>Before ASA</td>
<td>After ASA</td>
<td>CONTROLS</td>
<td>P¹</td>
</tr>
<tr>
<td>Total</td>
<td>500.7 ± 141.2</td>
<td>479.1 ± 140.8</td>
<td>336.45 ± 169.04</td>
<td>&lt;.001</td>
<td>&lt;.001</td>
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Platelet-derived cMPs

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<tbody>
<tr>
<td>CD61⁺</td>
<td>150.2 ± 86.3</td>
<td>140.9 ± 79.4</td>
<td>103.57 ± 68.08</td>
<td>.005</td>
</tr>
<tr>
<td>CD61⁺/CD142⁺</td>
<td>31.4 ± 25.5</td>
<td>31.0 ± 27.3</td>
<td>15.26 ± 11.67</td>
<td>&lt;.001</td>
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<tr>
<td>PAC1⁺</td>
<td>12.2 ± 11.9</td>
<td>15.1 ± 13.1</td>
<td>6.8 ± 3.13</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>CD62P⁺</td>
<td>32 ± 26.6</td>
<td>32.8 ± 26.7</td>
<td>19.07 ± 17.25</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>PAC1⁺/CD62⁺</td>
<td>6.6 ± 8.1</td>
<td>7.8 ± 8.7</td>
<td>4.5 ± 6.8</td>
<td>.002</td>
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Endothelial-derived cMPs

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<tbody>
<tr>
<td>CD146⁺</td>
<td>10.3 ± 7.3</td>
<td>11.5 ± 9.2</td>
<td>6.07 ± 7.42</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>CD146⁺/CD62E⁺</td>
<td>9.7 ± 7.5</td>
<td>10.1 ± 8.7</td>
<td>5.07 ± 5.42</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>CD62E⁺</td>
<td>6.6 ± 8.1</td>
<td>7.8 ± 8.7</td>
<td>4.25 ± 3.79</td>
<td>&lt;.001</td>
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Erythrocyte-derived cMPs

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<tbody>
<tr>
<td>CD235a⁺</td>
<td>78 ± 48.9</td>
<td>64.5 ± 28.3*</td>
<td>74.53 ± 60.29</td>
<td>.424</td>
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Leukocyte-derived cMPs

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<tbody>
<tr>
<td>CD45⁺</td>
<td>130.3 ± 97.5</td>
<td>118.5 ± 59.8</td>
<td>43.09 ± 30.39</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>CD3⁺/CD45⁺</td>
<td>28.3 ± 30.9</td>
<td>24.1 ± 15.7</td>
<td>12.61 ± 8.21</td>
<td>&lt;.001</td>
</tr>
</tbody>
</table>
CD14+          39.5 ± 29.8  30.7 ± 19.8*  4.73 ± 4.9      <.001   <.001
CD11b+/CD14+   25.6 ± 25    19.9 ± 15*  17.19 ± 16.7   <.001   <.001
CD45+/CD3+/CD14- 64.5 ± 92.9  56.9 ± 44.3  33.67 ± 27.21  .043   .007
CD11b+         70.3 ± 52.4  65.3 ± 37.2  68.57 ± 48.16  .081   .434
CD142+/CD14+   23.1 ± 20.6  16.3 ± 12.6* 9 ± 12.99     <.001   <.001
CD142+         72.1 ± 53.6  73.1 ± 53.1  68.45 ± 40.49  .275   .240

Smooth muscle cell-derived cMPs

SMA+           6.5 ± 10.6  3.1 ± 4.6*  2.17 ± 2.85     .014   .270
CD142+/SMA-α+  2.8 ± 4.6  1.3 ± 2.4*  0.99 ± 1.85     .025   .456

*P values from the Student t test for unrelated samples between: P1: controls and patients before the ASA intervention; and P2: controls and patients after the ASA intervention. *P<.05 from the comparison between before and after the intervention (Student’s t test for paired samples) in diabetic patients.

Used markers were CD61 for platelets, CD146 for endothelial cells, CD235ab for erythrocytes, CD45 for total leukocytes, and CD3 for lymphocytes and CD14 for monocytes origins accounting for agranulocytes. Granulocytes were inferred subtracting agranulocytes subpopulation from leukocytes fraction and smooth muscle actin-α was used for smooth muscle cells. The other CDs were used as biomarkers of cell activation (see Table 1 from the manuscript).

AV, Annexin V; ASA, acetylsalicylic acid; CI, confidence interval, cMPs, circulating microparticles; PFP, platelet free plasma.