Supporting Information to:

Flavonoids and Isoflavonoids from Sophorae Flos Improve Glucose Uptake in Vitro

Quan Cheng Chen\textsuperscript{1,2}, Wei Yun Zhang\textsuperscript{1}, WenYi Jin\textsuperscript{1}, Ik Soo Lee\textsuperscript{1}, Byung-Sun Min\textsuperscript{3}, Hyun-Ju Jung\textsuperscript{4}, MinKyun Na\textsuperscript{5}, SangMyung Lee\textsuperscript{6}, KiHwan Bae\textsuperscript{1}

Affiliation
\textsuperscript{1} College of Pharmacy, Chungnam National University, Daejeon, Korea
\textsuperscript{2} Institute for Biomedical Research, Xiamen University, Xiamen, P.R. China
\textsuperscript{3} College of Pharmacy, Catholic University of Daegu, Gyeongbuk, Korea
\textsuperscript{4} Department of Oriental Pharmacy, Wonkwang University, Jeonbuk, Korea
\textsuperscript{5} College of Pharmacy, Yeungnam University, Gyeongbuk, Korea
\textsuperscript{6} KT&G Central Research Institute, Daejeon, Korea

Correspondence

Prof. Dr. KiHwan Bae
College of Pharmacy
Chungnam National University
Daejeon 305-764
Korea
Tel.: +82-42-821-5925
Fax: +82-42-823-6566
baekh@cnu.ac.kr
Glucose uptake assay

HepG2 cells were cultured in Dulbecco’s modified Eagle’s medium (DMEM) with 10% fetal bovine serum (FBS) and maintained in an atmosphere of humidified 5% CO2 at 37 °C. Cells were plated at $1 \times 10^5$ cells/well into 96-well plates for 24 h and treated with or without tested samples in the absence or presence of 10 μM 2-NBDG for 1 h. After removal from medium and washing twice with pre-cold phosphate-buffered saline (PBS), cells were transferred into 5-mL polystyrene round-bottom tubes (BD Falcon) using 500 μL pre-cold fresh serum-free medium and maintained at 4 °C. The fluorescence intensity of 2-NBDG was recorded using a FACScalibur (Becton Dickinson) flow cytometer. Data from 1000 single-cell events were collected. Three independent experiments were performed, and the FCS (Flow Cytometry Standard) files were opened with WinMDI (version 2.9) software as dot-plot style and converted to text files. The 2-NBDG uptake of single cells was expressed as the average of fluorescence intensities recorded from 1000 cell events. The relative fluorescence intensity (FI) was calculated as $FI = FI_{2-NBDG} - FI_{background}$, where $FI_{2-NBDG}$ is the fluorescence intensity of a single cell treated with or without tested sample in the presence of 2-NBDG and $FI_{background}$ is the fluorescence intensity of a single cell treated with or without tested sample in the absence of 2-NBDG.

Table 1S Relative fluorescence intensities (FI) stimulated by isolated compounds at 10 μM

<table>
<thead>
<tr>
<th>Compound</th>
<th>FI</th>
<th>Compound</th>
<th>FI</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>37.6 ± 2.4*</td>
<td>6</td>
<td>58.0 ± 3.1*</td>
</tr>
<tr>
<td>2</td>
<td>8.8 ± 1.7</td>
<td>7</td>
<td>5.4 ± 2.1</td>
</tr>
<tr>
<td>3</td>
<td>5.3 ± 4.7</td>
<td>8</td>
<td>37.2 ± 4.6*</td>
</tr>
<tr>
<td>4</td>
<td>66.4 ± 2.0*</td>
<td>9</td>
<td>72.9 ± 1.6*</td>
</tr>
<tr>
<td>5</td>
<td>61.4 ± 2.3*</td>
<td>Rosiglitazone</td>
<td>42.9 ± 2.1*</td>
</tr>
</tbody>
</table>

Note: Data are expressed as mean ± SEM of three independent experiments. Analysis of variance was performed using ANOVA procedures followed by Dunnett’s post hoc test. *$P < 0.01$, compared with vehicle-treated control group.
Fig. 1S Representative histograms from fluorescence-activated cell-sorting analysis for glucose uptake in HepG2 cells. 2-NBDG (FL1) indicates fluorescence intensity. Black and red histograms represent cells strained with and without 2-NBDG, respectively. (A) vehicle-treated control group, (B) 10 μM rosiglitazone as positive control group, (C) 100 μg/mL MeOH extract, (D) 100 μg/mL EtOAc fraction, (E) 10 μM (solid lines) and 100 μM (dotted lines) tamarixetin (4), (F) 10 μM (solid lines) and 100 μM (dotted lines) pratensein (9).