ELECTRONIC SUPPLEMENTARY INFORMATION

Druggability profile of stilbene-derived PPAR agonists: determination of physicochemical properties and PAMPA study

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Determination of log *P* by RP-UPLC studies

In a typical experiment, the retention time (t_r) of each compound is determined on a minimum of five different organic modifier-water mobile phase ratios. For each mobile phase composition, the retention factor is calculated according to the formula:

$$logk = \log\left(\frac{t_R - t_{del} - \frac{V_{ext}}{F}}{t_0 - t_{del} - \frac{V_{ext}}{F}} - 1\right) \qquad \text{[eq. SI-1]}$$

where t_r and t_0 are the retention time of the solute and the unretained compound (uracil), respectively, t_{delay} is the injection delay, V_{ext} is the extra-column volume and F is the flow rate of the mobile phase.

Extrapolated retention factors (logk_w) is obtained by linear extrapolation to 100% water, from the isocratic log*k* values plotting as a function of the mobile phase composition.

The correlation between log*k* and the composition of mobile phase for **clofibric acid**, **gemfibrozil** and compounds **1-5** is reported in **Figure SI-1**.

A calibration curve was previously determined for the adopted chromatographic system using methanol as mobile phase based on the logk_w measurements of 52 compounds with well-known log P resulting in the following linear correlation:

$$logK_w = 0.83(\pm\ 0.02) * logP + 0.25(\pm\ 0.07) \qquad \text{[eq. SI-2]}$$

$$N = 52;\ R^2 = 0.99;\ S = 0.17;\ F = 4968$$

Thus, accordingly to this equation, the log P for our tested compounds was determination from the experimental measured logk_w (**Table SI-1**).

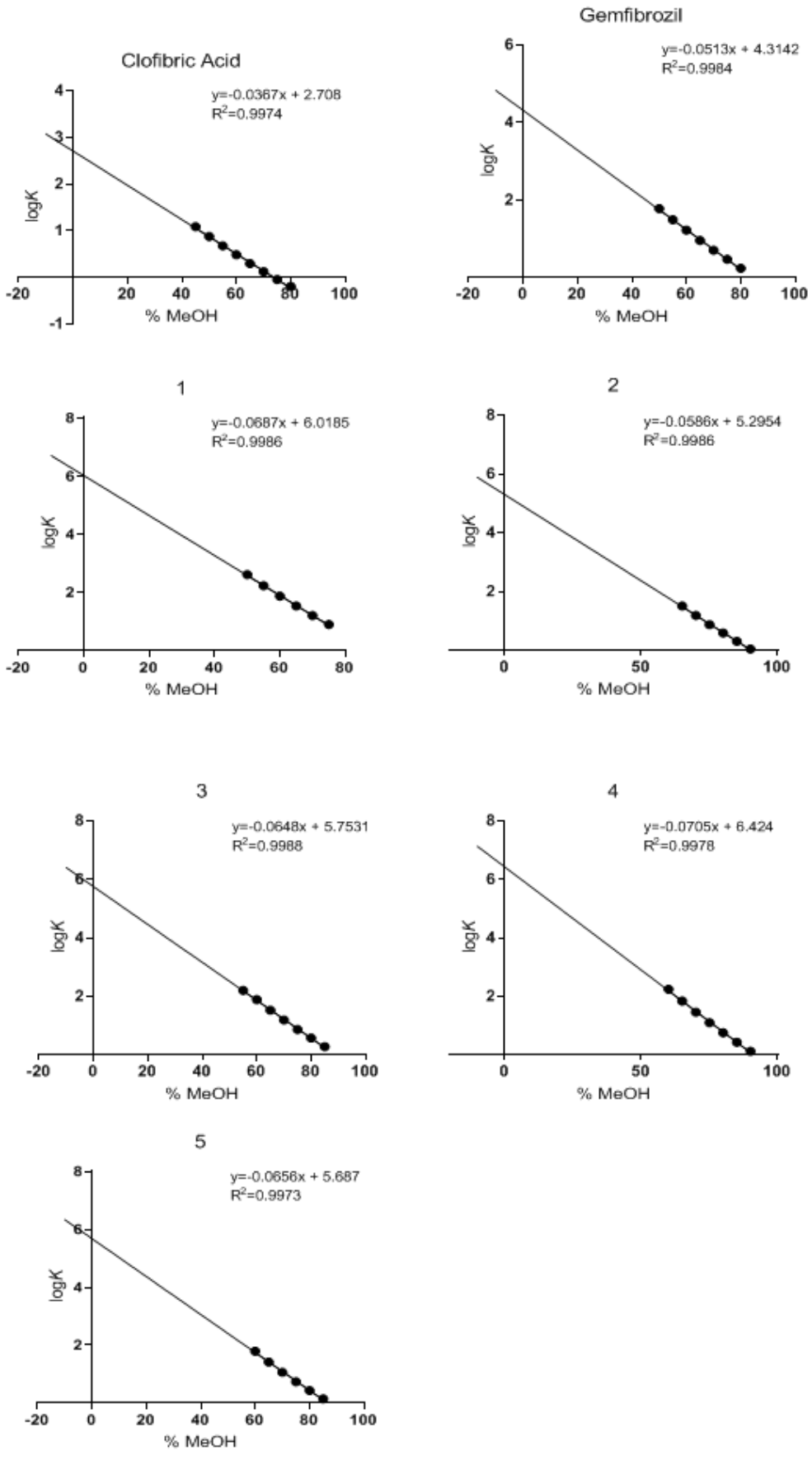

Figure SI-1. Extrapolation of the logk_w from the correlation between log$k$ and different composition of mobile phase by UPLC analysis.

Table SI-1. Determination of log *P* from logk_w for clofibric acid, gemfibrozil and compounds **1-5,** accordingly to **eq. SI-2.**

Cmpd	logk_w	log *P*	SD
Clofibric Acid	2.707	2.96	0.22
Gemfibrozil	4.314	4.90	0.28
1	6.018	6.95	0.36
2	5.295	6.08	0.35
3	5.753	6.63	0.34
4	6.425	7.44	0.41
5	5.687	6.55	0.40

Quantification of clofibric acid, gemfibrozil and compounds 1-5 concentrations by UV spectroscopy

The quantification of clofibric acid, gemfibrozil and compounds **1-5** concentrations in solubility and PAMPA assays was performed by UV spectroscopy.

UV spectra were acquired on a UV Reader Powerwave TM spectrometer (BioTek Instrument, Inc., Winooski, VT, USA) using 96-well quartz plate (Hellma GmbH&co, Müllheim, Germany).

The full UV spectra for each compound was acquired in the 200-600 nm range and the λ_{max} was determined (Figure SI-2).

The calibration curves were constructed by plotting the absorbance values determined at the respective λ_{max} against the micromolar concentration of the analyte in water, containing the 5% of DMSO. The concentration ranges were found to be linear in the 2.0–250.0 μM range for clofibric acid, gemfibrozil and compound **3**, in the 2.0-125.0 μM range for compounds **2** and **4** and in the 2.0–62.9 μM range for compounds **1** and **5** (Figure SI-3).

Calibration curves were used for the determination of the concentration of the analytes in the solubility and PAMPA assays.

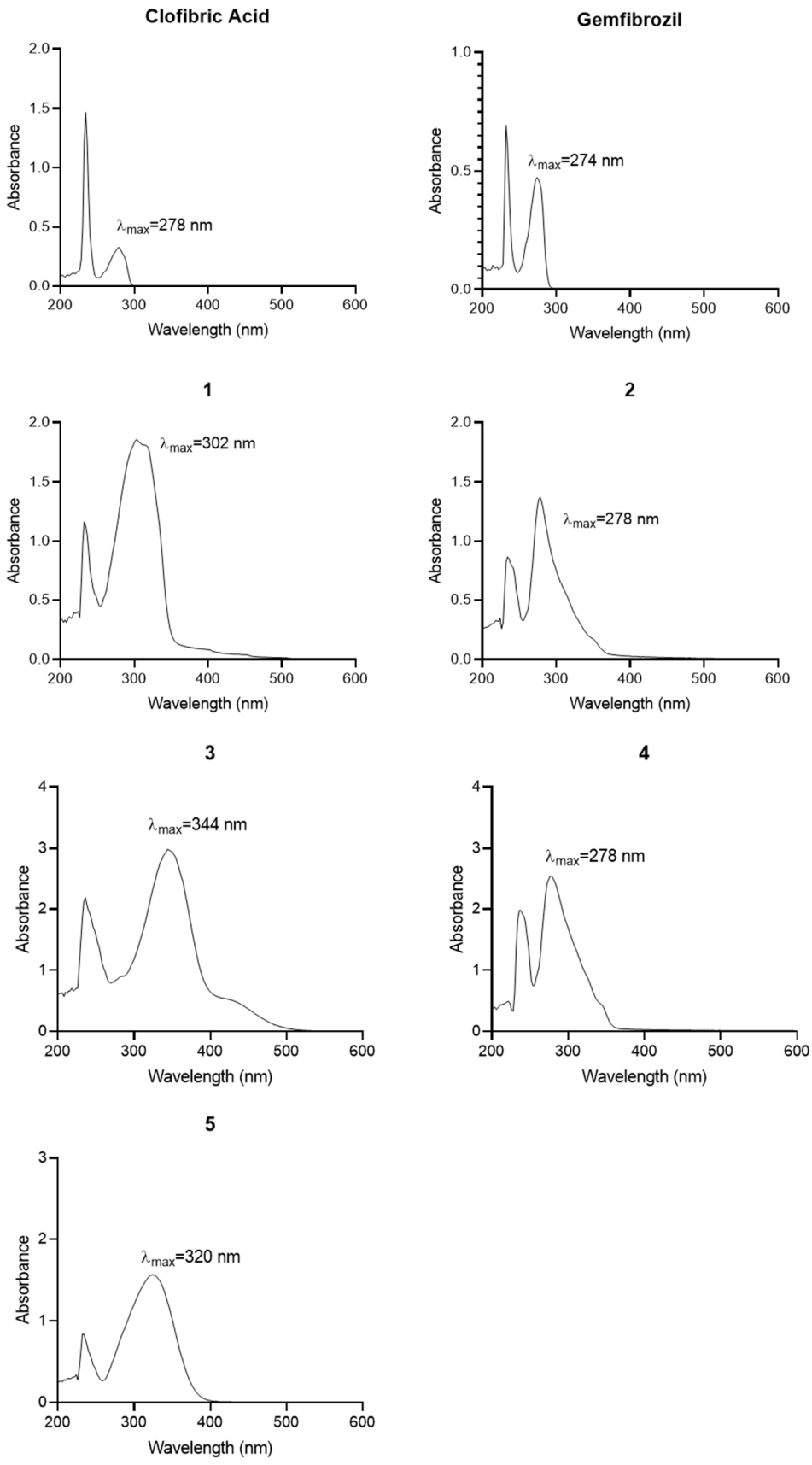

Figure SI-2. UV spectra of clofibric acid, gemfibrozil and compound **1-5** in aqueous solution containing 5% of DMSO.

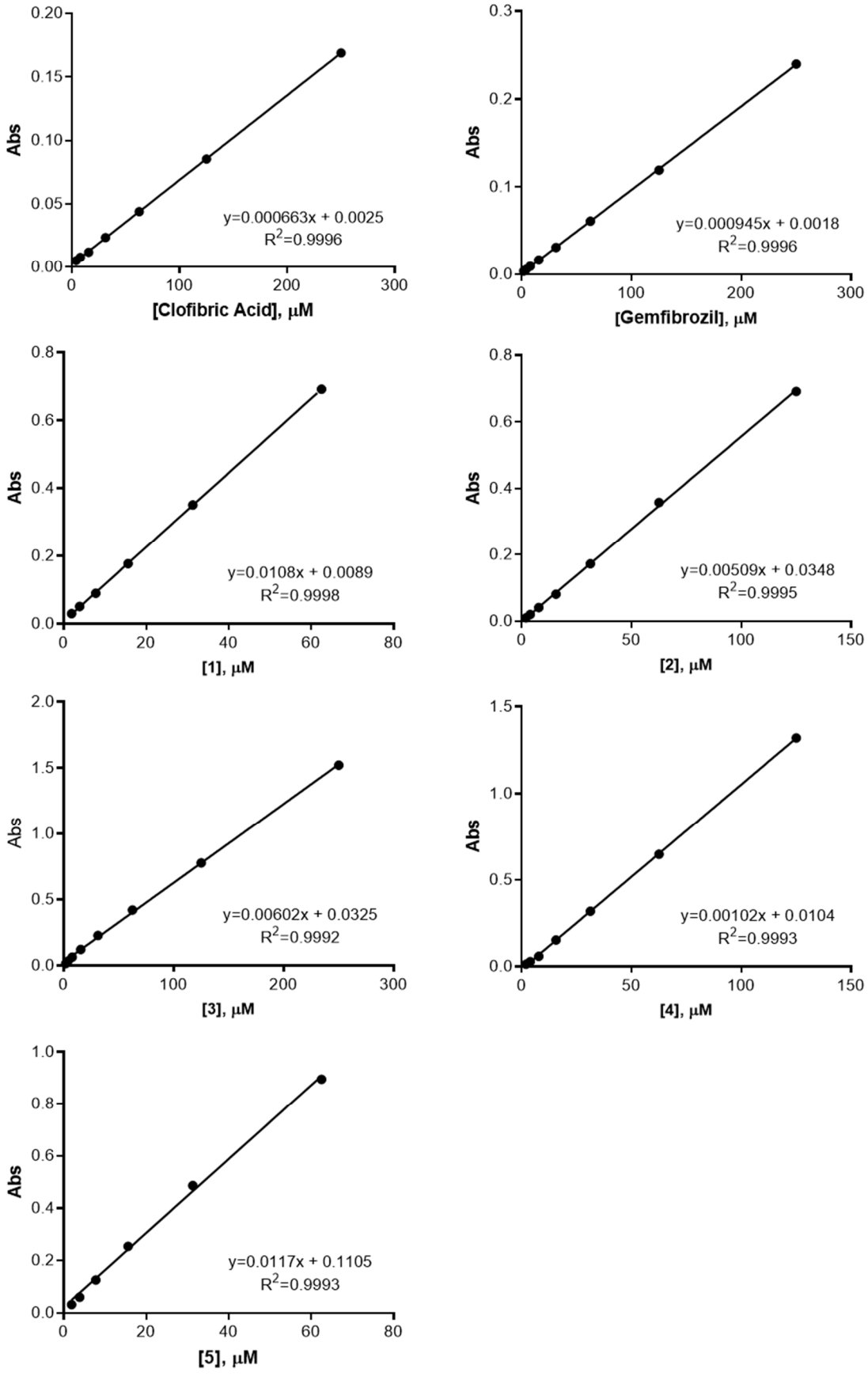

Figure SI-3. UV calibration curve for clofibric acid, gemfibrozil and compounds **1-5** determined at their λ_{max}.