chemistry proceedings

MDPI

Proceedings

Study of ADMET Descriptors of Novel Chlorinated *N*-Arylcinnamamides †

Dominika Pindjakova [1,*], Tomas Strharsky [2,*], Jiri Kos [2], Lucia Vrablova [1], Milan Hutta [1] and Josef Jampilek [1,2]

[1] Department of Analytical Chemistry, Faculty of Natural Sciences, Comenius University, Ilkovicova 6, 84215 Bratislava, Slovakia; lucia.vrablova26@gmail.com (L.V.); milan.hutta@uniba.sk (M.H.); josef.jampilek@gmail.com (J.J.)

[2] Regional Centre of Advanced Technologies and Materials, Faculty of Science, Palacky University, Slechtitelu 27, 78371 Olomouc, Czech Republic; jiri.kos@upol.cz

* Correspondence: pindjakova.dominika@gmail.com (D.P.); tomas.strharsky01@upol.cz (T.S.)

† Presented at the 24th International Electronic Conference on Synthetic Organic Chemistry, 15 November–15 December 2020; Available online: https://ecsoc-24.sciforum.net/.

Abstract: A series of seven (2*E*)-3-(3,4-dichlorophenyl)-*N*-arylprop-2-enamides was prepared. Six compounds from this limited set were mono- and di-chlorinated not only on the aromatic ring of acid but also on the anilide ring. The compounds have been proposed as potential anti-infective, anti-inflammatory, and anti-cancer agents. Since lipophilicity significantly affects the biological activities and the pharmacokinetic profile of compounds, the hydro-lipophilic properties of these new highly chlorinated compounds were experimentally studied. At the same time, the overall ADMET profiles of the compounds were investigated to establish whether they comply with the Lipinski's Rule of Five and thus meet the qualitative concepts of "druglikeness" for new bioactive molecules. All the discussed compounds were analyzed using the reversed-phase high performance liquid chromatography method. The procedure was performed under isocratic conditions with methanol as an organic modifier in the mobile phase, using an end-capped non-polar C_{18} stationary reversed-phase column. In the present study, the structure-lipophilicity relationships of the studied compounds are discussed.

Keywords: *N*-arylcinnamamides; synthesis; lipophilicity; ADMET; structure-lipophilicity relationships

1. Introduction

The ADMET properties of compounds characterizing pharmacokinetics are as important as the biological effect of a drug [1–4]. Physicochemical properties affecting permeability and bioaccumulation of cells belong to the area of quantitative structure-property relationships (QSPR) and are influenced by chemical composition [5–7]. In this context, lipophilicity was recognized more than a hundred years ago as the most important parameter influencing ADMET and bioactivity (e.g., lipoid theory of narcosis formulated by Meyer and Overton) [3,4]. The lipophilicity parameter is also part of Lipinski's Rule of Five (Ro5) or Carr's Rule of Three (Ro3) [8,9]. The issue of lipophilicity was also addressed by Hansch, Fujita & Leo, who derived a set of empirical lipophilicity descriptors, so-called π-values [10].

Lipophilicity represents the affinity of a molecule or a moiety for a lipophilic environment. It is commonly measured by its distribution behavior in a biphasic system, either liquid-liquid or solid-liquid. In general, it is a thermodynamic parameter describing the partitioning of a compound between an aqueous and an organic phase and can be characterized by the partition coefficient (log *P*). Log *P* is defined as a logarithm of the partition coefficient of the compound between n-octanol and water at a pH where all of the compound molecules are in the in the neutral form [3,4]. Since

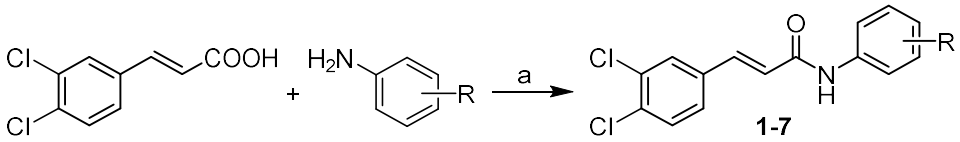
classical methods for the determination of these constants are time consuming and not always sufficiently reliable, reversed-phase high performance liquid chromatography (RP-HPLC) methods have become popular and widely used for lipophilicity measurement. A general procedure is the measurement of directly accessible retention time under isocratic conditions with varying amounts of an organic modifier in the mobile phase using end-capped non-polar C_{18} stationary RP columns and calculating the capacity factor *k*. Log *k*, calculated from the capacity factor *k*, is used as the lipophilicity index converted to log *P* scale [3,11].

Because most drugs are weak bases or acids that are ionized under physiological conditions, another parameters describing lipophilicity can be found, namely the distribution coefficient D_{pH} and its log D_{pH}, which is the logarithm of the distribution coefficient of the compound between n-octanol and an aqueous phase (buffer) at a specified pH. A portion of the compound molecules may be in the ionic form and a portion may be in the neutral form [3,4,12]. The distribution coefficient, which takes into account ionization, is a more reliable expression of lipophilicity at physiological pH, and log $D_{7.4}$ values (at pH 7.4) are of particular importance, because it resembles actual physiological values. Likewise, from the point of view of absorption after oral administration, the partition coefficient at pH 6.5 (log $D_{6.5}$) is important, because it is the pH in the small intestine. This descriptor is considered to be the most important lipophilicity descriptor and is preferred in the ADME study [3,4,13,14].

Recently, a large series of ring-substituted *N*-arylcinnamanilides together with their antimicrobial and anti-inflammatory activities as well as their activity related to the inhibition of photosynthetic electron transport in chloroplasts have been published [15–18]. Since early prediction of physicochemical properties, i.e., "druglikeness", is important for identification of a suitable candidate at the early drug discovery stage, several compounds from the new series of chlorinated *N*-arylcinnamanilides were investigated in relation to their ADMET profile and structure-lipophilicity relationships.

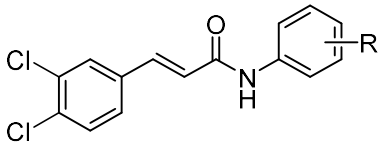
2. Results and Discussion

The reaction of 3,4-dichlorocinnamic acid using phosphorus trichloride with aniline in dry chlorobenzene in a microwave reactor provided a series of *N*-arylcinnamamides **1–7**, see Scheme 1 and Table 1.



Scheme 1. Synthesis of ring-substituted (2*E*)-3-(3,4-dichlorophenyl)-*N*-arylprop-2-enamides **1–7**. *Reagents and conditions*: (a) PCl_3, chlorobenzene, MW, 130 °C, 40 min.

Table 1. Structure of ring-substituted (2*E*)-3-(3,4-dichlorophenyl)-*N*-arylprop-2-enamides **1–7**, calculated lipophilicities (log *P*/Clog *P*), and experimentally determined log *k*, log $D_{7.4}$, and log $D_{6.5}$ values of investigated compounds.



Comp.	R	log *k*	log $D_{7.4}$	log $D_{6.5}$	log *P* [a]	log *P*/Clog *P* [b]
1	H	0.6199	0.6354	0.6669	4.42	4.30/4.9700
2	2-Cl	0.7764	0.8019	0.8203	5.10	4.86/5.0906
3	3-Cl	0.9071	0.8735	0.9453	5.31	4.86/5.9406
4	4-Cl	0.9009	0.8660	0.9381	5.19	4.86/5.9406
5	2,4-Cl	1.0932	1.0565	1.0985	5.68	5.41/5.8938
6	2,5-Cl	1.0840	1.0474	1.0887	5.72	5.41/5.8938

7	3,5-Cl	1.3043	1.3080	1.3336	5.90	5.41/6.7438

[a] ACD/Percepta ver. 2012, [b] ChemBioDraw Ultra 13.0.

The log *P*/Clog *P* data of all the investigated chlorinated *N*-arylcinnamamide derivatives were predicted using commercially available programs ChemBioDraw Ultra 13.0 and ACD/Percepta ver. 2012. The lipophilicity of the compounds was also examined by the RP-HPLC determination of capacity factors *k* followed by calculation of log *k* and the determination of distribution coefficients $D_{7.4}$ and $D_{6.5}$ with the subsequent calculation of log $D_{7.4}$ and log $D_{6.5}$. All the results are shown in Table 1. The HPLC procedure was performed under isocratic conditions with methanol as an organic modifier in the mobile phase using end-capped non-polar C18 stationary RP columns.

Parameters predicted by the ChemBioDraw software (log *P* and Clog) for individual positional isomers are not distinguished; therefore, these values are listed only in Table 1 without other discussion. On the other hand, lipophilicity data log *P* for compounds **1**–**7** predicted by ACD/Percepta showed high consensus with all the experimentally determined values log *k*, log $D_{7.4}$, and log $D_{6.5}$, as can be seen in graphs in Figure 1; the correlation coefficients r for n = 7 are as follows: 0.9609, 0.9420, and 0.9513, respectively. The mutual consensus of all the experimental parameters is also very high (r = 0.9931, 0.9981, and 0.9952, respectively), see Figure 2. Thus, based on the experimental and predicted results, it can be stated that unsubstituted (2*E*)-3-(3,4-dichlorophenyl)- *N*-phenylprop-2-enamide (**1**) is the least lipophilic compound, while (2*E*)-*N*-(3,5-dichlorophenyl)- 3-(3,4-dichlorophenyl)-prop-2-enamide (**7**) is the most lipophilic. The only difference between all experimental and predicted values was observed for compounds **5** (R = 2,4-Cl) and **6** (R = 2,5-Cl), where compound **5** actually shows a higher lipophilicity than compound **6**, which was predicted by the software vice versa: log *P* = 5.68 (**5**) and log *P* = 5.72 (**6**). This is caused by specific intra- and intermolecular interactions of the substituent in the *ortho* position with other spatially close moieties/fragments and the polar medium as was described recently [15,18,19–22]. Nevertheless, based on the results of this preliminary short study of several selected new anilides of 3,4-dichlorocinnamic acid, it can be assumed that the log *P* values predicted by ACD/Percepta recognized hydro-lipophilic properties in a good agreement with experimentally determined values, and thus, this software can be used for these simple chlorinated derivatives as a useful and mainly fast tool for the subsequent investigation of structure-activity relationships. The question remains what will be the inaccuracies in the prediction for anilide substituents capable of forming mainly hydrogen bonds (e.g., -F, -CF_3, -OCH_3) with the surrounding aqueous/buffered medium.

Distribution parameters π [23,24] were introduced to characterize the lipophilic contribution of individual substituents to the scaffold and they are calculated according to the relationship $\pi = \log k_S - \log k_U$, where log k_S is the determined logarithm of the capacity factor of the compound, and log k_U indicates the determined logarithm of the capacity factor of unsubstituted derivative **1**, whose π value is 0. The same applies to the values of the distribution coefficient D_{pH}. The π values of individual substituted anilide rings (π_{Ar}) of drivatives **1**–**7** are mentioned in Table 2, where there are differences (mutual order of values) between experimental and calculated π_{Ar} values of compounds **5** (R = 2,4-Cl) and **6** (R = 2,5-Cl). The differences between π_{Ar} values calculated by ACD/Percepta are due to the failure to include possible interactions of substituents in the *ortho* position with a spatially close carboxamide, while π_{Ar} values based on experimentally determined log *k*/log D_{pH} data carry these interactions in them. It should be noted that the π_{Ar} values calculated from the experimental log *k* and both log D_{pH} differ insignificantly from each other.

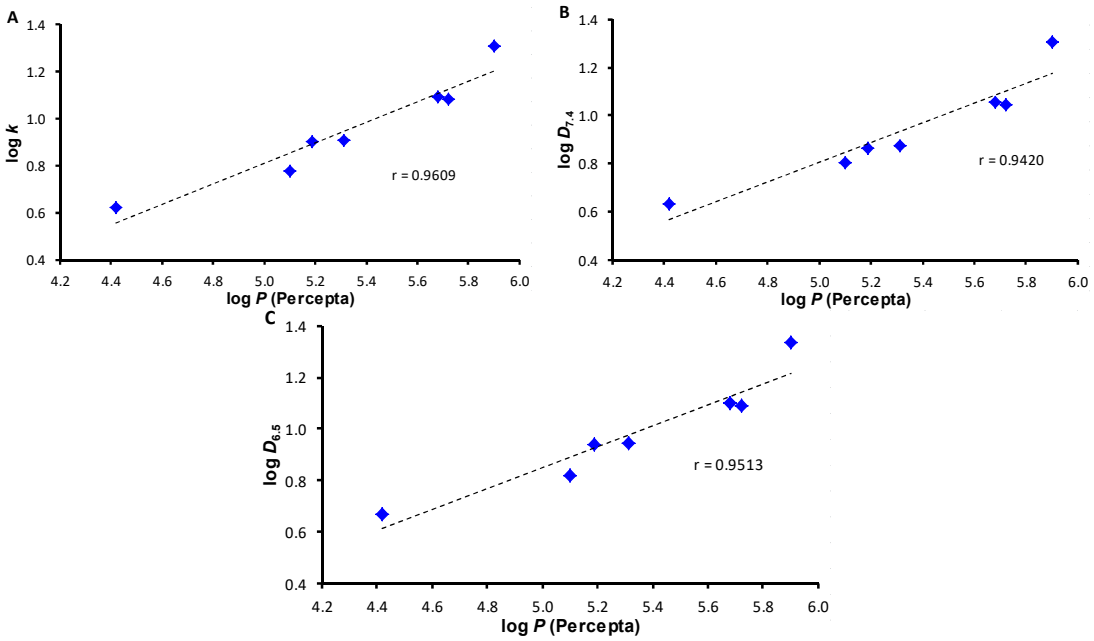


Figure 1. Comparison of predicted log *P* (ACD/Percepta) values with experimentally found log *k* (**A**), log $D_{7.4}$ (**B**), and log $D_{6.5}$ (**C**) values of ring-substituted (2*E*)-3-(3,4-dichlorophenyl)- *N*-arylprop-2-enamides **1–7**.

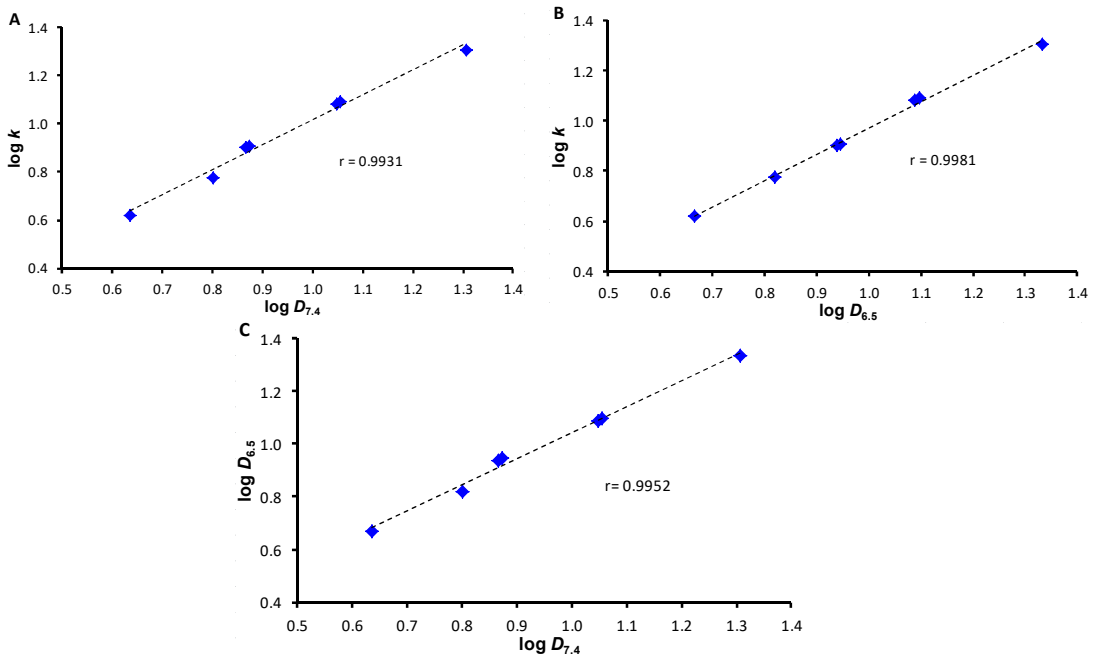


Figure 2. Comparison of experimentally found log *k* values with log $D_{7.4}$ (**A**) and log $D_{6.5}$ (**B**) values and log $D_{7.4}$ with log $D_{6.5}$ (**C**) of discussed compounds **1–7**.

Table 2. Comparison of determined distributive parameters π calculated from log *k* and log D_{pH} for each individual substituted anilide ring within the investigated series of compounds **1–7** and parameters π of individual substituted anilide rings predicted by ACD/Percepta.

Comp.	R	π_{Ar} (exp. log *k*)	π_{Ar} (exp. log $D_{7.4}$)	π_{Ar} (exp. log $D_{6.5}$)	π_{Ar} (ACD/Percepta)
1	H	0	0	0	1.76
2	2-Cl	0.16	0.17	0.15	2.23
3	3-Cl	0.29	0.24	0.28	2.32
4	4-Cl	0.28	0.23	0.27	2.33
5	2,4-Cl	0.47	0.42	0.43	2.82
6	2,5-Cl	0.46	0.41	0.42	2.73
7	3,5-Cl	0.68	0.67	0.67	2.90

The Ro5 [8,9] is one of the most accepted recommendations concerning the physicochemical parameters of biologically active compounds, and all medicinal chemists try to follow it when designing molecules [25]. The Ro5 contains the limits of specific molecular descriptors (see Table 3) set based on experimentally and statistically obtained results so that a compound that meets this recommendation has a higher chance of becoming a drug. Table 3 lists the parameters contained in Ro5 plus some of the other most used. But a suitable drug-like profile does not ensure that the molecule will become a drug and vice versa [26]. It is clear that ADMET-friendly properties, such as lipophilicity, polar surface area, etc., are important in the context of specific ligand-receptor interactions; therefore, the following Table 3 shows the profile of mainly Ro5 parameters characterizing the investigated set of compounds. Based on the data presented in Table 3, it can be stated that in general, the investigated compounds meet the Ro5 requirements. It should be mentioned that compounds **2–7** have a slightly higher lipophilicity (log *p* values) than recommended by Ro5. In addition to higher lipophilicity, the individual substituents on both the phenyl acid core and the anilide ring are characterized by electron-withdrawing properties (electronic σ parameters of anilide substituents ranged from 0.75–1.22 [27]), making them potentially interesting chemotherapeutics as well as agrochemicals [28]. On the other hand, these higher lipophilic compounds showed a log $D_{7.4}$ slightly higher than 1, indicating that the compounds are expected to have good solubility, good intestinal absorption (good balance of solubility and passive diffusion permeability), and minimized metabolism (lower binding to metabolic enzymes) [3,4].

Table 3. Values of parameters characterizing physicochemical properties calculated using ACD/Percepta ver. 2012 in relation to Lipinski's Rule of Five (Ro5).

Comp.	R	MW	log *P*	HBD	HBA	RB	TPSA	Parachor
1	H	292.16	4.42	1	2	3	29.10	581.26
2	2-Cl	326.60	5.10	1	2	3	29.10	617.13
3	3-Cl	326.60	5.31	1	2	3	29.10	617.13
4	4-Cl	326.60	5.19	1	2	3	29.10	617.13
5	2,4-Cl	361.05	5.68	1	2	3	29.10	653.00
6	2,5-Cl	361.05	5.72	1	2	3	29.10	653.00
7	3,5-Cl	361.05	5.90	1	2	3	29.10	653.00
Ro5		**<500**	**<5**	**<5**	**<10**	**–**	**–**	**–**

Molecular weight (MW), lipophilicity (log P), number of H-bond donors (HBD), number of H-bond acceptors (HBA), number of rotatable bonds (RB), topological polar surface area (TPSA).

3. Experimental

3.1. General

All reagents were purchased from Merck (Sigma-Aldrich, St. Louis, MO, USA) and Alfa (Alfa-Aesar, Ward Hill, MA, USA). Reactions were performed using an Anton-Paar Monowave 50 microwave reactor (Graz, Austria). The melting points were determined on a Kofler hot-plate apparatus HMK (Franz Kustner Nacht KG, Dresden, Germany) and are uncorrected. Infrared (IR) spectra were recorded on a Nicolet iS5 IR spectrometer (Thermo Scientific, West Palm Beach, FL, USA). The spectra were obtained by the accumulation of 256 scans with 2 cm^{-1} resolution in the region of 4000–450 cm^{-1}. All ^{1}H- and ^{13}C-NMR spectra were recorded on a JEOL JNM-ECA 600II device (600 MHz for ^{1}H and 150 MHz for ^{13}C, JEOL, Tokyo, Japan) in dimethyl sulfoxide-d_6 (DMSO-d_6). ^{1}H and ^{13}C chemical shifts (δ) are reported in ppm.

3.2. Synthesis

General procedure for synthesis of target compounds **1–8**: *General procedure for synthesis of target compounds* **1–8**: 3,4-Dichlorocinnamic acid (0.9 mM) was suspended in dry chlorobenzene (6 mL) at ambient temperature and phosphorus trichloride (0.45 mM, 0.5 eq.), and the corresponding substituted aniline (0.9 mM, 1 eq.) was added dropwise. The reaction mixture was transferred to the microwave reactor, where the synthesis was performed (40 min, 130 °C). Then the mixture was cooled to 40 °C, and then the solvent was removed to dryness under reduced pressure. The residue was washed with hydrochloride acid and water. The crude product was recrystallized from ethanol.

(2E)-3-(3,4-Dichlorophenyl)-N-phenylprop-2-enamide (**1**). Yield 63%; Mp 140–143 °C; IR (cm^{-1}): 3251, 3126, 3038, 1654, 1618, 1597, 1551, 1533, 1497, 1486, 1469, 1444, 1391, 1290, 1240, 1197, 1183, 1129, 1076, 1031, 1004, 969, 947, 922, 894, 868, 814, 784, 751, 735, 693, 684, 677, 661, 590, 564, 508, 485; ^{1}H-NMR (DMSO-d_6) δ: 10.23 (s, 1H), 7.90 (d, *J* = 2.1 Hz, 1H); 7.71–7.69 (m, 3H), 7.62 (dd, *J* = 8.9 Hz, *J* = 2.1 Hz, 1H); 7.57 (d, *J* = 15.1 Hz, 1H); 7.35–7.32 (m, 2H), 7.09–7.06 (m, 1H), 6.89 (d, *J* = 15.8 Hz, 1H); ^{13}C-NMR (DMSO-d_6), δ: 163.02, 139.08, 137.42, 135.67, 131.87, 131.74, 131.13, 129.61, 128.83, 127.37, 124.64, 123.51, 119.24.

(2E)-N-(2-Chlorophenyl)-3-(3,4-dichlorophenyl)prop-2-enamide (**2**). Yield 68%; Mp 154–156 °C; IR (cm^{-1}): 3293, 1659, 1626, 1592, 1533, 1468, 1441, 1387, 1337, 1289, 1276, 1242, 1198, 1183, 1148, 1127, 1058, 1034, 1026, 1001, 966, 956, 938, 915, 888, 865, 826, 743, 721, 713, 697, 679, 659, 615, 589, 532, 497, 461; ^{1}H-NMR (DMSO-d_6), δ: 9.66 (s, 1H), 7.96 (d, *J* = 8.2 Hz, 1H), 7.93 (d, *J* = 1.4 Hz, 1H), 7.72–7.71 (m, 1H), 7.64 (dd, *J* = 8.2 Hz, *J* = 2.1 Hz, 1H), 7.59 (d, *J* = 15.1 Hz, 1H), 7.52 (dd, *J* = 7.6 Hz, *J* = 1.4 Hz, 1H), 7.36 (td, *J* = 7.6 Hz, *J* = 1.4 Hz, 1H), 7.21 (d, *J* = 1.4 Hz, 1H), 7.19 (dd, *J* = 7.9 Hz, 1.7 Hz, 1H); ^{13}C-NMR (DMSO-d_6), δ: 163.45, 138.15, 135.63, 134.84, 132.01, 131.75, 131.14, 129.58, 129.52, 127.63, 127.49, 126.12, 125.51, 125.21, 124.15.

(2E)-N-(3-Chlorophenyl)-3-(3,4-dichlorophenyl)prop-2-enamide (**3**). Yield 63%; Mp 186–188 °C; IR (cm^{-1}): 3277, 3127, 1664, 1626, 1597, 1536, 1484, 1469, 1426, 1407, 1396, 1341, 1295, 1249, 1239, 1198, 1184, 1131, 1100, 1074, 1026, 1002, 996, 973, 923, 905, 881, 863, 818, 814, 784, 776, 729, 682, 677, 592, 575, 557, 498, 451; ^{1}H-NMR (DMSO-d_6) δ: 10.42 (s, 1H), 7.93–7.91 (m, 2H), 7.70 (d, *J* = 8.2 Hz, 1H), 7.63 (dd, *J* = 8.6 Hz, *J* = 1.7 Hz, 1H), 7.58 (d, *J* = 15.8 Hz, 1H), 7.51 (dd, *J* = 8.2 Hz, 1.4 Hz, 1H), 7.36 (t, *J* = 7.9 Hz, 1H), 7.13 (dd, *J* = 8.2 Hz, 1.4 Hz, 1H), 6.85 (d, *J* = 15.8 Hz, 1H); ^{13}C-NMR (DMSO-d_6), δ: 163.34, 140.52, 138.07, 135.49, 133.15, 132.07, 131.77, 131.15, 130.54, 129.75, 127.46, 124.13, 123.22, 118.70, 117.66.

(2E)-N-(4-Chlorophenyl)-3-(3,4-dichlorophenyl)prop-2-enamide (**4**). Yield 71%; Mp 158–160 °C; IR (cm^{-1}): 3291, 1660, 1623, 1590, 1554, 1528, 1489, 1473, 1397, 1338, 1294, 1282, 1244, 1203, 1181, 1135, 1092, 1030, 1012, 997, 973, 949, 904, 853, 818, 813, 788, 726, 709, 667, 637, 627, 560, 524, 509, 479, 442; ^{1}H-NMR (DMSO-d_6), δ: 10.37 (s, 1H), 7.90 (d, *J* = 2.1 Hz, 1H), 7.73–7.70 (m, 2H), 7.70 (d, *J* = 8.2 Hz, 1H), 7.62 (dd, *J* = 8.6 Hz, *J* = 1.7 Hz, 1H), 7.57 (d, *J* = 15.8 Hz, 1H), 7.40–7.37 (m, 2H), 6.85 (d, *J* = 15.8 Hz, 1H); ^{13}C-NMR (DMSO-d_6), δ: 163.14, 138.04, 137.79, 135.57, 131.99, 131.76, 131.14, 129.68, 128.75, 127.42, 127.08, 124.29, 120.77.

(2E)-N-(2,4-Dichlorophenyl)-3-(3,4-dichlorophenyl)prop-2-enamide (**5**). Yield 64%; Mp 190–192 °C; IR (cm^{-1}): 3276, 1658, 1626, 1579, 1553, 157, 1467, 1381, 1336, 1301, 1287, 1197, 1184, 1143, 1128, 1100, 1052,

1029, 1005, 963, 948, 920, 882, 868, 856, 831, 817, 797, 754, 720, 700, 684, 666, 610, 571, 558, 509, 472; NMR (DMSO-d_6) δ: 9.72 (s, 1H), 8.01 (d, J = 8.9 Hz, 1H), 7.92 (d, J = 2.1 Hz, 1H), 7.71 (d, J = 8.2 Hz, 1H), 7.68 (d, J = 2.1 Hz, 1H), 7.63 (dd, J = 8.6 Hz, J = 1.7 Hz, 1H), 7.59 (d, J = 15.8 Hz, 1H), 7.44 (dd, J = 8.9 Hz, J = 2.1 Hz, 1H), 7.19 (d, J = 15.8 Hz, 1H); ^{13}C-NMR (DMSO-d_6), δ: 163.54, 138.44, 135.55, 134.04, 132.09, 131.76, 131.14, 129.60, 129.08, 128.93, 127.64, 127.61, 126.24, 126.02, 123.88.

(2E)-N-(2,5-Dichlorophenyl)-3-(3,4-dichlorophenyl)prop-2-enamide (**6**). Yield 72%; Mp 203–205 °C; IR (cm^{-1}): 3398, 3115, 1696, 1633, 1581, 1554, 1512, 1474, 1444, 1408, 1329, 1308, 1259, 1236, 1201, 1159, 1133, 1091, 1047, 1026, 997, 975, 962, 923, 903, 873, 824, 802, 732, 685, 582, 571, 557, 548, 495, 458; ^{1}H-NMR (DMSO-d_6), δ: 9.74 (s, 1H), 8.15 (d, J = 2.7 Hz, 1H), 7.94 (d, J = 1.4 Hz, 1H), 7.72 (d, J = 8.2 Hz, 1H), 7.64 (dd, J = 8.2 Hz, J = 2.1 Hz, 1H), 7.60 (d, J = 15.8 Hz, 1H), 7.56 (d, J = 8.9 Hz, 1H), 7.26 (dd, J = 8.6 Hz, J = 2.4 Hz, 1H), 7.24 (d, J = 15.8 Hz, 1H); ^{13}C-NMR (DMSO-d_6), δ: 163.71, 138.73, 136.10, 135.51, 132.18, 131.78, 131.63, 131.18, 130.87, 129.65, 127.71, 125.50, 123.84, 123.46.

(2E)-N-(3,5-Dichlorophenyl)-3-(3,4-dichlorophenyl)prop-2-enamide (**7**). Yield 59%; Mp 169–171 °C; IR (cm^{-1}): 3449, 3182, 3114, 3083, 1659, 1620, 1587, 1544, 1476, 1442, 1410, 1387, 1341, 1300, 1269, 1193, 1151, 1139, 1116, 1097, 1032, 1012, 973, 953, 939, 865, 849, 815, 785, 724, 702, 867, 675, 666, 602, 581, 554, 530, 467,3 454; ^{1}H-NMR (DMSO-d_6) δ: 10.56 (s, 1H), 7.90 (d, J = 2.1 Hz, 1H), 7.73–7.72 (m, 2H), 7.69 (d, J = 8.9 Hz, 1H), 7.62 (dd, J = 8.9 Hz, J = 2.1 Hz, 1H), 7.59 (d, J = 15.8 Hz, 1H), 7.28–7.27 (m, 1H), 6.79 (d, J = 15.8 Hz, 1H); ^{13}C-NMR (DMSO-d_6), δ: 163.59, 141.37, 138.67, 135.29, 134.15, 132.24, 131.79, 131.15, 129.84, 127.52, 123.64, 122.71, 117.36.

3.3. Lipophilicity Determination by HPLC

A HPLC separation module Waters Alliance 2695 XE equipped with a Waters Dual Absorbance Detector 2486 (Waters Corp., Milford, MA, USA) was used. A chromatographic column Symmetry® C18 5 μm, 4.6 × 250 mm, Part No. W21751W016 (Waters Corp., Milford, MA, USA) was used. The HPLC separation process was monitored by Empower® 3 Chromatography Manager Software (Waters Corp.). Isocratic elution by a mixture of MeOH p.a. (72%) and H_2O-HPLC Mili-Q grade (28%) as a mobile phase was used for the determination of capacity factor k. Isocratic elution by a mixture of MeOH p.a. (72%) and acetate buffered saline (pH 7.4 and pH 6.5) (28%) as a mobile phase was used for the determination of distribution coefficient expressed as $D_{7.4}$ and $D_{6.5}$. The total flow of the column was 1.0 mL/min, injection 20 μL, column temperature 40 °C, and sample temperature 10 °C. The detection wavelength of 210 nm was chosen. A KI methanolic solution was used for determination of the dead times (t_D). Retention times (t_R) were measured in minutes. The capacity factors k were calculated according to the formula $k = (t_R - t_D)/t_D$, where t_R is the retention time of the solute, and t_D is the dead time obtained using an unretained analyte. The distribution coefficients D_{pH} were calculated according to the formula $D_{pH} = (t_R - t_D)/t_D$. Each experiment was repeated three times. The log k values of individual compounds are shown in Table 1.

3.4. Lipophilicity Calculations

Log P, i.e., the logarithm of the partition coefficient for n-octanol/water, was calculated using the programs ACD/Percepta (Advanced Chemistry Development. Inc., Toronto, ON, Canada, 2012) and ChemBioDraw Ultra 13.0 (CambridgeSoft, PerkinElmer Inc., MA, USA). Clog P values (the logarithm of n-octanol/water partition coefficient based on established chemical interactions) were calculated using ChemBioDraw Ultra 13.0 (CambridgeSoft) software. The results are shown in Table 1. The distributive parameters π_{Ar} of individual substituted anilide rings of individual compounds were predicted using ACD/Percepta and are shown in Table 2, while other physicochemical and topological descriptors are mentioned in Table 3.

Acknowledgments: This study was supported by the Slovak Research and Development Agency (APVV-17-0373) and by Palacky University Olomouc (IGA_PrF_2020_023).

References

1. Veber, D.F.; Johnson, S.R.; Cheng, H.Y.; Smith, B.R.; Ward, K.W.; Kopple, K.D. Molecular properties that influence the oral bioavailability of drug candidates. *J. Med. Chem*. **2002**, *45*, 2615–2623.
2. Van de Waterbeemd, H.; Gifford, E. ADMET in silico modeling: Towards prediction paradise? *Nat. Rev. Drug Discov*. **2003**, *2*, 192–204.
3. Kerns, E.H.; Di, L. *Drug-Like Properties: Concepts. Structure Design and Methods: From ADME to Toxicity Optimization*; Academic Press: San Diego, CA, USA, 2008.
4. Wermuth, C.; Aldous, D.; Raboisson, P.; Rognan, D. *The Practice of Medicinal Chemistry*, 4th ed.; Academic Press: San Diego, CA, USA, 2015.
5. Fukunishi, Y.; Nakamura, H. Definition of drug-likeness for compound affinity. *J. Chem. Inf. Model.* **2011**, *51*, 1012–1016.
6. Lipinski, C.A.; Lombardo, F.; Dominy, B.W.; Feeney, P.J. Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings. *Adv. Drug Deliv. Rev.* **2001**, *46*, 3–26.
7. Lipinski, C.A. Lead- and drug-like compounds: The rule-of-five revolution. *Drug Discov. Today Technol*. **2004**, *1*, 337–341.
8. Lipinski, C.A.; Lombardo, F.; Dominy, B.W.; Feeney, P.J. Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings. *Adv. Drug Deliv. Rev.* **1997**, *23*, 3–25.
9. Carr, R.A.; Congreve, M.; Murray, C.W.; Rees, D.C. Fragment-based lead discovery: Leads by design. *Drug Discov. Today* **2005**, *10*, 987–992.
10. Martin, Y.C. Hansch analysis 50 years on. *Wiley Interdiscip. Rev. Comput. Mol. Sci.* **2012**, *2*, 435–442.
11. Pliska, V.; Testa, B.; van der Waterbeemd, H. *Lipophilicity in Drug Action and Toxicology*; Wiley-VCH: Weinheim, Germany, 1996.
12. Caron, G.; Vallaro, M.; Ermondi, G. Log P as a tool in intramolecular hydrogen bond considerations. *Drug Discov. Today* **2018**, *27*, 65–70.
13. Andres, A.; Roses, M.; Rafols, C.; Bosch, E.; Espinosa, S.; Segarra, V.; Huerta, J.M. Setup and validation of shake-flask procedures for the determination of partition coefficients (log *D*) from low drug amounts. *Eur. J. Pharm. Sci.* **2015**, *76*, 181–191.
14. Culen, M.; Rezacova, A.; Jampilek, J.; Dohnal, J. Designing dynamic dissolution method: Review of instrumental options and corresponding physiology of stomach and small intestine. *J. Pharm. Sci.* **2013**, *102*, 2995–3017.
15. Pospisilova, S.; Kos, J.; Michnova, H.; Kapustikova, I.; Strharsky, T.; Oravec, M.; Moricz, A.M.; Bakonyi, J.; Kauerova, T.; Kollar, P.; et al. Synthesis and spectrum of biological activities of novel *N*-arylcinnamamides. *Int. J. Mol. Sci.* **2018**, *19*, 2318.
16. Pospisilova, S.; Kos, J.; Michnova, H.; Strharsky, T.; Cizek, A.; Jampilek, J. *N*-Arylcinnamamides as Antistaphylococcal Agents. In Proceedings of the 4th International Electronic Conference on Medicinal Chemistry (ECMC-4), 2018; Volume 5576. Available online: https://sciforum.net/manuscripts/5576/slides.pdf.
17. Hosek, J.; Kos, J.; Strharsky, T.; Cerna, L.; Starha, P.; Vanco, J.; Travnicek, Z.; Devinsky, F.; Jampilek, J. Investigation of anti-inflammatory potential of n-arylcinnamamide derivatives. *Molecules* **2019**, *24*, 4531.
18. Kos, J.; Bak, A.; Kozik, V.; Jankech, T.; Strharsky, T.; Swietlicka, A.; Michnova, H.; Hosek, J.; Smolinski, A.; Oravec, M.; et al. Biological activities and ADMET-related properties of novel set of cinnamanilides. *Molecules* **2020**, *25*, 4121.
19. Hansch, C.; Leo, A.; Unger, S.H.; Kim, K.H.; Nikaitani, D.; Lien, E.J. "Aromatic" substituent constants for structure-activity correlations. *J. Med. Chem.* **1973**, *16*, 1207–1216.
20. Kos, J.; Ku, C.F.; Kapustikova, I.; Oravec, M.; Zhang, H.J.; Jampilek, J. 8-Hydroxyquinoline-2-carboxanilides as antiviral agents against avian influenza virus. *ChemistrySelect* **2019**, *4*, 4582–4587.
21. Strharsky, T.; Jankech, T.; Kos, J.; Maricakova, K.; Pramukova, A.; Hutta, M.; Devinsky, F.; Jampilek, J. Preparation and Hydro-Lipophilic Properties of Selected Novel Chlorinated and Brominated *N*-arylcinnamamides. In Proceedings of the 23rd International Electronic Conference on Synthetic Organic Chemistry (ECSOC-23), 2019; Volume 41, p. 11. Available online: https://www.mdpi.com/2504-3900/41/1/11/pdf.

22. Bak, A.; Kos, J.; Michnova, H.; Gonec, T.; Pospisilova, S.; Kozik, V.; Cizek, A.; Smolinski, A.; Jampilek, J. Similarity-driven pharmacophore mapping for series of *N*-(disubstituted-phenyl)-3-hydroxynaphthalene- 2-carboxamides. *Int. J. Mol. Sci.* **2020**, *21*, 6583.
23. Norrington, F.E.; Hyde, R.M.; Williams, S.G.; Wootton, R. Physicochemical-activity relations in practice I. A rational and self-consistent data bank. *J. Med. Chem.* **1975**, *18*, 604–607.
24. Dearden, J.C. Partitioning and lipophilicity in quantitative structure-activity relationships. *Environ. Health Perspect.* **1985**, *61*, 203–228.
25. Jampilek, J. Potential of agricultural fungicides for antifungal drug discovery. *Expert Opin. Drug Dis.* **2016**, *11*, 1–9.
26. Ertl, P.; Schuffenhauer, A. Estimation of synthetic accessibility score of drug-like molecules based on molecular complexity and fragment contributions. *J. Cheminform.* **2009**, *1*, 8.
27. Gonec, T.; Kos, J.; Pesko, M.; Dohanosova, J.; Oravec, M.; Liptaj, T.; Kralova, K.; Jampilek, J. Halogenated 1-hydroxynaphthalene-2-carboxanilides affecting photosynthetic electron transport in photosystem II. *Molecules* **2017**, *22*, 1709.
28. Lipinski, C. To the Rule of 5 and Beyond? How Dr. Chris Lipinski Gained New Insights into Drug Discovery. CAS, American Chemical Society. 2019. Available online: https://www.cas.org/blog/rule-5-and-beyond-how-dr-chris-lipinski-gained-new-insights-drug-discovery (accessed on 13 October 2020).

Publisher's Note: MDPI stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.

© 2020 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (http://creativecommons.org/licenses/by/4.0/).