Quinaldine Derivatives: Preparation and Biological Activity

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Abstract: The series of quinaldine derivatives were prepared, some of them by means of novel synthetic methods. The synthetic approach, analytical and spectroscopic data of all newly synthesized compounds are presented. The prepared compounds were tested for their *in vitro* antifungal activity as well as for their photosynthesis-inhibiting activity (the inhibition of photosynthetic electron transport in spinach chloroplasts (*Spinacia oleracea* L.) and the reduction of chlorophyll content in *Chlorella vulgaris* Beij.). Structure-activity relationships among the chemical structure, the physical properties and the biological activities of the evaluated compounds are discussed in the article.

Key Words: Quinaldine derivatives, *in vitro* antifungal activity, photosynthesis inhibition, lipophilicity, structure-activity relationships, *chlorella vulgaris*, spinach chloroplasts.

1. INTRODUCTION

Over the last three decades there has been a dramatic increase in the incidence of fungal infection. Discovery of new drugs for the treatment of systemic mycoses is a major challenge in infectious disease research. There is an intensified need for new antifungal remedies with novel modes of action due to the rapid growth of the immuno-compromised patient population, the development of resistance to the present azole therapies, and high toxicity of polyenes [1].

The compounds possessing the quinoline pharmacophore in the molecule have been clinically used as antifungal, antibacterial and antiprotozoic drugs [2,3] as well as antineoplastics [4]. Some quinoline derivatives showed also antiasthmatic and antiplatelet activity [5-7]. Acetylcholine-sterase inhibitory activity of various quinoline derivatives has been tested for potential treatment of nervous diseases [8].

This presented study is a follow-up paper to the previous articles [9-15] and deals with the synthesis of quinaldine derivatives as substituted bicyclical pyridine derivatives [9-11] on the one hand and the fungistatic effect of this first series of substituted quinaldine derivatives on the other hand.

Our previous observation proved, that various compounds possessing *N*-hetarene in the molecule can inhibit photo-synthetic electron transport, it means, they showed herbicidal activity [9-15]. In addition, we have investigated the prepared compounds by testing them for their efficiency related to inhibition of photosynthetic electron transport (PET) in spinach chloroplasts (*Spinacia oleracea* L.) and reduction of chlorophyll content in *Chlorella vulgaris* Beij.

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Lipophilicity is one of the important physico-chemical properties of biologically active compounds, therefore, that it is the most studied property in drug design. Two methods were used for the lipophilicity determination: *i*) calculation Log *P*/CLog *P* using the two programs and *ii*) determination of the capacity factor K and subsequently Log *K* using the reversed phase high performance liquid chromatography (RP-HPLC). All values were compared and the influence of the biological effects on the lipophilicity parameters is shown too.

This work aimed at preparing quinaldine derivatives by means of novel synthetic pathways and consequently at searching for the structure-activity relationships in the mentioned series, i.e. to continue studying of the substituent variability influence on the biological activity.

2. EXPERIMENTAL

2.1. Instrumentation and Chemicals

All solvents used for the synthesis were of analytical grade. The solvents were dried and freshly distilled under argon atmosphere. 2-Methylquinoline, 2-methylquinolin-4-ol and 2-methylquinolin-8-ol were purchased from Sigma-Aldrich. Kieselgel 60, 0.040-0.063 mm (Merck, Darmstadt, Germany) was used for flash chromatography (F_C) and a silica gel was impregnated by TEA. TLC was performed on Silufol UV 254 plates (Kavalier, Votice, Czech Republic) and plates were impregnated by TEA. The plates were illuminated under UV (254 nm). Melting points were determined on Boetius PHMK 05 (VEB Kombinat Nagema, Radebeul, Germany) and are uncorrected. Elemental analyses were carried out on an automatic microanalyser EA1110CE (CE Instruments, Milano, Italy). Infrared spectra were recorded with neat oils (for non-crystalline materials) and in KBr pellets (for crystalline materials) on an IR-spectrometer Nicolet Impact 400. ^{1}H and ^{13}C NMR Spectra

1573-4064/05 $50.00+.00

were recorded on a Varian Mercury – Vx BB 300 (299.95 MHz for ^{1}H and 75.43 MHz for ^{13}C), Varian Comp. (Palo Alto, CA, U.S.A.). Chemical shifts are given relative to internal $Si(CH_3)_4$.

2.2. Synthesis

2-Methyl-7,8-dihydro-6H-quinolin-5-one (1)

3-Aminocyclohex-2-enone (5.6 g, 50.0 mmol) and but-3-en-2-one (7.0 g, 100.0 mmol) were refluxed in DMF for 1 h. Then the solvent was removed at reduced pressure. F_C (Et_2O/petroleum ether 6:1) gave a colourless oil. Yield: 4.2 g (52%). R_F: 0.38 (Et_2O/petroleum ether 6:1). ^{1}H and ^{13}C NMR spectra identical with Reimann [16].

5-Hydroxy-2-methyl-7,8-dihydroquinolin-1-oxide (2)

Compound **1** (2.0 g, 12.6 mmol) was dissolved in dry DCM and 50% MCPBA (6.0 g) was added. The mixture was stirred under room temperature for 16 h. The reaction mixture was transferred to a separation funnel and extracted with EtO_2 and saturated solution of Na_2CO_3. The combined organic extracts were dried over anhydrous $MgSO_4$ and filtered. The solvent was removed at reduced pressure. F_C (acetone/petroleum ether 3:2) gave a yellow crystalline compound. Yield: 1.2 g (55%). M.p. 152-154 °C. R_F: 0.33 (acetone/petroleum ether 3:2). Anal. Calc. for $C_{10}H_{11}NO_2$ (177.20): 67.78% C, 6.26% H, 7.90% N; found: 67.75% C, 6.26% H, 7.87% N. IR spectrum (KBr), cm^{-1}: 3436, 1186 (OH), 3027 (=CH-), 2958 (CH_3), 3085, 1567, 1103 (pyridine), 1442 (CH_2), 1259 (N-O). ^{1}H-NMR ($CDCl_3$), δ: 7.83 (d, 1H, *J*=8.25 Hz, H4), 7.36 (d, 1H, *J*=8.24 Hz, H3), 4.61 (dd, 1H, *J*=6.32 Hz, *J*=4.39 Hz, CH), 3.42-3.35 (m, 2H, CH_2), 2.61 (s, 3H, CH_3), 2.60-2.52 (m, 2H, CH_2). ^{13}C-NMR ($CDCl_3$), δ: 188.7, 154.3, 152.7, 127.2, 124.3, 123.7, 57.4, 29.0, 21.6, 18.9.

2-Methyl-5-oxy-5,6,7,8-tetrahydroquinolin-1-oxide (3)

See compound **2** for conditions. A yellow crystalline compound. Yield: 0.5 g (23%). M.p. 74.5-76 °C. R_F: 0.23 (acetone/petroleum ether 3:2). Anal. Calc. for $C_{10}H_{11}NO_2$ (177.20): 67.78% C, 6.26% H, 7.90% N; found: 67.81% C, 6.23% H, 7.86% N. IR spectrum (KBr), cm^{-1}: 2957 (CH_3), 3086, 1566, 1109 (pyridine), 1696 (C=O), 1441 (CH_2), 1257 (N-O). ^{1}H-NMR ($CDCl_3$), δ: 7.79 (d, 1H, *J*=7.83 Hz, H4), 7.29 (d, 1H, *J*=7.83 Hz, H3), 3.25 (t, 2H, *J*=6.46 Hz, CH_2), 2.66 (t, 2H, *J*=6.46 Hz, CH_2), 2.59 (s, 3H, CH_3), 2.27-2.15 (m, 2H, CH_2). ^{13}C-NMR ($CDCl_3$), δ: 195.9, 154.3, 153.5, 129.3, 123.7, 122.9, 37.4, 24.5, 20.3, 18.9.

2-Methyl-5,6,7,8-tetrahydroquinolin-5-ol (4)

Ketone **1** (1.2 g, 0.8 mmol) was dissolved in dry toluene (30 ml) and Synhydride (70% solution in toluene, 10 ml) was added dropwise when stirring. The mixture was refluxed for 2 h. After cooling, 15% aqueous HCl (30 ml) was added. The mixture was extracted with Et_2O, the combined Et_2O extracts were dried over anhydrous $MgSO_4$, filtered and the solvent was removed under reduced pressure. F_C (Et_2O/MeOH 4:1) gave a light yellow crystalline compound. Yield: 0.5 g (60%). R_F: 0.17 (Et_2O). M.p. 95-97 °C. Anal. Calc. for $C_{10}H_{13}NO$ (163.22): 73.59% C, 8.03% H, 8.58% N; found: 73.64% C, 8.06% H, 8.57% N. IR spectrum (KBr), cm^{-1}: 3416, 1091 (OH), 2934 (CH_3), 3056, 1592, 1101 (pyridine), 1465 (CH_2). ^{1}H-NMR (DMSO-d_6), δ: 8.60 (d, 1H, *J*=7.93 Hz, H4), 7.03 (d, 1H, *J*=7.93 Hz, H3), 4.37 (dd, 1H, *J*=5.00 Hz, *J*=5.00 Hz, CH), 3.46-3.39 (m, 2H, CH_2), 2.73-2.67 (2H, m, CH_2), 2.50 (3H, s, CH_3), 2.19-2.08 (2H, m, CH_2). ^{13}C-NMR (DMSO-d_6), δ: 156.2, 155.9, 136.6, 132.6, 120.7, 66.1, 41.1, 32.1, 24.1, 21.7.

8-Bromo-2-methyl-7,8-dihydro-6H-quinolin-5-one (5)

A solution of **1** (0.5 g, 3.1 mmol), NBS (0.6 g, 3.1 mmol), and dibenzoyl peroxide (0.016 g) in dry CCl_4 was refluxed for 8 h under argon. The mixture was cooled in an ice bath, filtered, and the filtrate was concentrated *in vacuo*. F_C (Et_2O/petroleum ether 2:3) gave a yellow crystalline compound. Yield: 0.03 g (5%). R_F: 0.70 (Et_2O). M.p. 107-109 °C. Anal. Calc. for $C_{10}H_{10}BrNO$ (240.10): 50.02% C, 4.20% H, 5.83% N; found: 50.07% C, 4.26% H, 5.81% N. IR spectrum (KBr), cm^{-1}: 2938 (CH_3), 3064, 1584, 1101 (pyridine), 1703 (C=O), 1463 (CH_2), 611 (C-Br). ^{1}H-NMR ($CDCl_3$), δ: 8.18 (d, 1H, *J*=7.85 Hz, H4), 7.24 (d, 1H, *J*=7.86 Hz, H3), 5.60 (t, 1H, *J*=7.25 Hz, CH), 3.22-3.09 (m, 2H, CH_2), 2.85-2.70 (m, 2H, CH_2), 2.60 (s, 3H, CH_3). ^{13}C-NMR ($CDCl_3$), δ: 195.9, 164.3, 160.4, 135.7, 124.3, 121.7, 49.3, 34.1, 30.9, 25.0.

2-Methylquinolin-5-ol (6)

Method A: See compound **5** for conditions. Yield: 0.4 g (71%).

Method B: Ketone **1** (1.0 g, 6.3 mmol) and DDQ (1.4 g, 6.3 mmol) were dissolved in dry dioxan and stirred for 24 h under argon. Then the mixture was concentrated in vacuum and saturated solution of Na_2CO_3 was added. The mixture was extracted with Et_2O, the combined Et_2O extracts were dried over anhydrous $MgSO_4$, filtered and the solvent was removed under reduced pressure. F_C (Et_2O/petroleum ether 5:1). Yield: 0.8 g (83%).

A white crystalline compound. R_F: 0.25 (Et_2O). M.p. 227-229 °C; M.p. 230-232 °C [17].

3-Bromo-2-methylquinolin-4-ol (8)

Compound **7** bromination. See compound **5** for conditions. The crude product was purified by crystallization from EtOH/H_2O, and a white crystalline compound was obtained. Yield: 0.8 g (98%). R_F: 0.55 (acetone/toluene 4:1). M.p. 322-324 °C; M.p. 326 °C [18].

7-Bromo-2-methylquinolin-8-ol (10)

Compound **9** bromination. See compound **5** for conditions. A white crystalline compound. Yield: 0.2 g (20%). R_F: 0.23 (Et_2O/petroleum ether 1:6). M.p. 137-139 °C; M.p. 134-136 °C [19].

5-Bromo-2-methylquinolin-8-ol (11)

Compound **9** bromination. See compound **5** for conditions. F_C (Et_2O/petroleum ether 1:6) gave a white crystalline

compound. Yield: 0.03 g (4%). R_F: 0.32 (Et_2O/petroleum ether 1:6). M.p. 68.5-70.0 °C; M.p. 68 °C [20].

5,7-Dibromo-2-methylquinolin-8-ol (12)

Compound **9** bromination. See compound **5** for conditions. A white crystalline compound. Yield: 0.1 g (11%). R_F: 0.12 (Et_2O/petroleum ether 1:6). M.p. 124-126 °C. M.p. 125 °C [20].

2-Bromomethylquinoline (14)

2-Methylquinoline (1.0 g, 6.9 mmol) was dissolved in dry CCl_4 and refluxed in the presence of NBS (1.3 g, 6.9 mmol) and AIBN (0.014 g). After cooling, the solution was filtered, evaporated and the residue was washed with MeOH to give a yellow crystalline compound. Yield: 0.9 g (55%). R_F: 0.57 (acetone/petroleum ether 1:4). M.p. 67-68 °C; M.p. 63-64 °C [21].

Quinoline-2-carboxylic acid (15)

A mixture of **13** (1.0 g, 7.0 mmol) and SeO_2 (2.5 g, 21.0 mmol) in dioxan was refluxed for 8 h. After cooling, the solution was evaporated and the residue was extracted with EtOAc and H_2O. The combined organic extracts were dried over anhydrous $MgSO_4$, filtered and the solvent was removed under reduced pressure. The crude product was purified by crystallization from EtOH/H_2O and then from H_2O only, and a light yellow crystalline compound was obtained. Yield: 0.7 g (57%). R_F: 0.47 (MeOH/isopropanol 1:1). M.p. 158-159 °C; M.p. 155 °C [22].

2-(2-quinolin-2-ylethyl)quinoline (16)

A stirred mixture of 4-hydroxyacetanilide (0.5 g, 3.5 mmol), 2-bromomethylquinoline (0.8 g, 3.5 mmol), anhydrous K_2CO_3 (3.0 g), KI (0.1 g) and DMF (50 ml) was refluxed for 3 h. The hot mixture was filtered, the filtration cake was washed with boiling DMF, and the solution was evaporated under reduced pressure. F_C (acetone/petroleum ether 1:4) gave a yellow crystalline compound. Yield: 0.4 g (42%). R_F: 0.35 (acetone/petroleum ether 1:4). M.p. 159-161 °C; M.p. 163 °C [23].

2.3. Lipophilicity HPLC determination (capacity factor K/calculated log K)

The HPLC separation module Waters Alliance 2695 XE and Waters Photodiode Array Detector 2996 (Waters Corp., Milford, MA, U.S.A.) were used. The chromatographic column Symmetry® C_{18} 5 µm, 4.6×250 mm, Part No. WAT054275, (Waters Corp., Milford, MA, U.S.A.) was used. The HPLC separation process was monitored by Millennium32® Chromatography Manager Software, Waters 2004 (Waters Corp., Milford, MA, U.S.A.).

For compounds **1** and **4-16** as a mobile phase the mixture of MeOH p.a. (55.0%) and H_2O-HPLC – Mili-Q Grade (45.0%) was used. The total flow of the column was 1.0 ml/min, injection 30 µl, column temperature 30 °C and sample temperature 10 °C. The detection wavelength 250 nm was chosen. The retention time (dead time) of the KI methanolic solution was T_D = 1.956 min. For compounds **2** and **3** as a mobile phase the mixture of MeOH p.a. (40.0%) and H_2O-HPLC – Mili-Q Grade (60.0%) was used. The total flow of the column was 0.8 ml/min, injection 30 µl, column temperature 30 °C and sample temperature 10 °C. The detection wavelength 210 nm was chosen. The retention time (dead time) of the KI methanolic solution was T_D = 2.238 min.

The capacity factors K was calculated using the Millennium32® Chromatography Manager Software and it was calculated by means of the equation $K = T_R\text{-}T_D/T_D$ (T_R is a retention time of the individual compounds). The calculated Log *K* of the individual compounds is shown in Table **1**.

2.4. Lipophilicity Calculations

Hydrophobicity of compounds (Log *P*/CLog *P* values) was calculated using the programs CS ChemOffice Ultra ver. 7.0 (CambridgeSoft, Cambridge, MA, U.S.A.) and ACD/Log P ver. 1.0 (Advanced Chemistry Development Inc., Toronto, Canada). Results are shown in Table **1**.

2.5. *In Vitro* Antifungal Susceptibility Testing

The broth microdilution test [24,25] was used for the assessment of *in vitro* antifungal activity of the synthesized compounds against *Candida albicans* ATCC 44859 (CA), *Candida tropicalis* 156 (CT), *Candida krusei* E28 (CK), *Candida glabrata* 20/I (CG), *Trichosporon beigelii* 1188 (TB), *Aspergillus fumigatus* 231 (AF), *Absidia corymbifera* 272 (AC), and *Trichophyton mentagrophytes* 445 (TM). Fluconazole (FLU) was used as a reference drug. The procedure was performed with twofold dilution of the compounds in RPMI 1640 medium (Sevapharma a.s., Prague, Czech Republic) buffered to pH 7.0 with 0.165 mol of 3-morpholino-propane-1-sulfonic acid. The final concentrations of the compounds ranged from 500 to 0.975 µmol/l. Drug–free controls were included. The minimal inhibitory concentrations (MICs) were determined after 24 h and 48 h of static incubation at 35°C. With *T. mentagrophytes*, the final MICs were determined after 72 h and 120 h of incubation. The results are summarized in Table **2**.

2.6. Herbicidal Activities

2.6.1 Study of Photosynthetic Electron Transport Inhibition in Spinach Chloroplasts

Chloroplasts were prepared by the procedure of Walker [26] from spinach (*Spinacia oleracea* L.). The inhibition of photosynthetic electron transport (PET) in spinach chloroplasts was determined spectrophotometrically (Kontron Uvikon 800, Kontron, Muenchen, Germany) using an artificial electron acceptor 2,6-dichlorophenol-indophenol (DCIPP) according to Kralova *et al*. [27] and the rate of photosynthetic electron transport was monitored as a photoreduction of DCPIP. The measurements were carried out in phosphate buffer (0.02 mol/l, pH 7.2) containing sucrose (0.4 mol/l), $MgCl_2$ (0.005 mol/l) and NaCl (0.015 mol/l). The chlorophyll content was 30 mg/l in these experiments and the samples were irradiated (~100 W/m^2)

Table 1. Comparison of Calculated Lipophilicities (Log *P*/CLog *P*) and Determined Log *K* of Compounds

Compound	Log *P*/CLog *P* CS ChemOffice Ultra	Log *P* ACD/Log P	Log *K*
1	1.64 / 1.523	1.88 ± 0.23	0.1580
2	*b* / -0.686	-0.26 ± 0.35	0.1128
3	*b* / -0.429	-0.84 ± 0.75	0.0190
4	1.91 / 1.079	1.02 ± 0.22	0.8233
5	2.03 / 2.076	2.04 ± 0.33	0.2443
6	2.43 / 2.577	1.91 ± 0.21	0.6061
7	0.62 / 0.781	1.18 ± 0.23	0.0029
8	0.93/ 1.722	2.34 ± 0.38	0.0996
9	2.43 / 2.577	2.33 ± 0.23	0.6838
10	3.26 / 3.282	3.38 ± 0.38	1.0759
11	3.26 / 3.562	3.54 ± 0.38	1.3142
12	4.09 / 4.188	4.55 ± 0.43	1.8054
13	2.82 / 2.528	2.54 ± 0.20	0.6649
14	2.10 / 2.603	2.17 ± 0.25	0.7125
15	3.31 / 2.811	2.78 ± 0.23	0.7301
16	5.28 / 4.362	4.43 ± 0.22	1.8850
FLU	0.99 / -0.440	0.31 ± 0.74	–
DCMU	2.76 / 2.691	2.78 ± 0.38	–

[a] for compounds **2** and **3** only, [b] impossible to compute due to charges.

from 10 cm distance with a halogen lamp (250 W) using a 4 cm water filter to prevent warming of the samples (suspension temperature 22 °C). The studied compounds were dissolved in DMSO due to their limited water solubility. The applied DMSO concentration (up to 4%) did not affect the photochemical activity in spinach chloroplasts (PET). The inhibitory efficiency (concentration) of the studied compounds has been expressed by IC_{50} values, i.e. by molar concentration of the compounds causing 50% decrease in the oxygen evolution relative to the untreated control. Comparable IC_{50} value for a selective herbicide 3-(3,4-dichlorophenyl)-1,1-dimethylurea, DCMU (DIURON) was about 1.9 μmol/l. The results are summarized in Table **2**.

2.6.2. Study of Chlorophyll Content Reduction in Chlorella Vulgaris Beij.

The green algae *C. vulgaris* Beij. was cultivated statically at room temperature according to Kralova *et al*. [28] (photoperiod 16 h light/8 h dark; photosynthetic active radiation 80 μmol/m^2.s; pH 7.2). The effect of the compounds on algal chlorophyll (Chl) content was determined after 4-day cultivation in the presence of the tested compounds. The Chl content in the algal suspension was determined spectrophotometrically (Kontron Uvikon 800, Kontron, Muenchen, Germany) after extraction into methanol according to Wellburn [29]. The Chl content in the suspensions at the beginning of the cultivation was 0.1 mg/l. Because of the low solubility of the studied compounds in water, these were dissolved in DMSO. DMSO concentration in the algal suspensions did not exceed 0.25% and the control samples contained the same DMSO amount as the suspensions treated with the tested compounds. The antialgal activity of compounds was expressed as IC_{50}. Comparable IC_{50} value for a selective herbicide DCMU was about 7.3 μmol/l. The results are summarized in Table **2**.

3. RESULTS AND DISCUSSION

3.1. Synthesis

The compounds synthesis is shown in Scheme **1**. We described new/more advantageous preparations of some compounds. The main starting material **1** was obtained by means of condensation of but-3-en-2-one with 3-aminocyclohex-2-enone [16]. Compounds **2** and **3** were generated *via N*-oxidation with *m*-chloroperoxybenzoic acid (MCPBA) in dichloromethane (DCM). Ketone **1** was reduced with Synhydride® (70% solution of bis(2-methoxyethoxy) dihydroalanate sodium in toluene) that gave 60% yield of racemic secondary alcohol **4**. Radical oxidative bromination

Table 2. ***In Vitro*** **Antifungal Activity (IC_{80}), Photosynthesis Inhibition (IC_{50})[a] of the Selected Compounds in Comparison with Standards (FLU, DCMU)**

Comp.	MIC/IC_{80} [µmol/l]								IC_{50} [µmol/l]	
	CA	**CT**	**CK**	**CG**	**TB**	**AF**	**AC**	**TM**	**spinach chloroplasts**	***Chlorella vulgaris***
	24h ***48h***	***24h*** ***48h***	***24h*** ***48h***	***24h*** ***48h***	***24h*** ***48h***	***24h*** ***48h***	***24h*** ***48h***	***72h*** ***120h***		
2	125 >125	125 >125	125 >125	125 >125	125 >125	125 >125	125 >125	31.25 31.25	89	*b*
3	>250 >250	>250 >250	>250 >250	>250 >250	>250 >250	>250 >250	>250 >250	250 250	258	*b*
4	125 >125	125 >125	125 >125	125 >125	125 >125	125 >125	125 >125	125 >125	*b*	83.4
6	>125 >125	>125 >125	>125 >125	>125 >125	>125 >125	>125 >125	>125 >125	125 125	397	*b*
8	*b*	*b*	*b*	*b*	*b*	*b*	*b*	*b*	322	*b*
9	>250 >250	>250 >250	>250 >250	>250 >250	>250 >250	>250 >250	>250 >250	250 250	*b*	13.0
10	3.91 7.81	31.25 31.25	7.81 7.81	31.25 31.25	15.63 62.5	15.63 31.25	31.25 62.5	31.25 62.5	*b*	40.3
11	3.91 3.91	15.63 31.25	3.91 7.81	15.63 15.63	31.25 31.25	15.63 15.63	15.63 15.63	15.63 15.63	504	67.9
12	<0.45 0.9	7.81 15.63	7.81 15.63	7.81 15.63	31.25 62.5	15.63 15.63	62.5 62.5	7.81 15.63	418	110.1
15	>250 >250	>250 >250	>250 >250	>250 >250	>250 >250	>250 >250	>250 >250	>250 >250	689	*b*
16	125 >125	125 >125	125 >125	125 >125	125 >125	125 >125	125 >125	125 >125	*b*	*b*
FLU	0.06 0.12	0.12 >125	3.91 15.62	0.98 3.91	0.24 0.48	>125 >125	>125 >125	1.95 3.91	–	–
DCMU	–	–	–	–	–	–	–	–	1.9	7.3

[a] IC_{50} values are related to PET inhibition in spinach chloroplasts and reduction of chlorophyll in *C. vulgaris*, [b] not tested due to precipitation of a dissolved drug.

of ketone **1** using *N*-bromosuccinimide (NBS) yielded compounds **5** (5%) and **6** (71%), nevertheless compound **6** was obtained also by means of oxidation with 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) in 83% yield. Literature described compound **6** preparation from 2-methylquinolin-5-ylamine in 32% yield [17] or from 5-amino-2,4-dibromophenol in 61% yield [30].

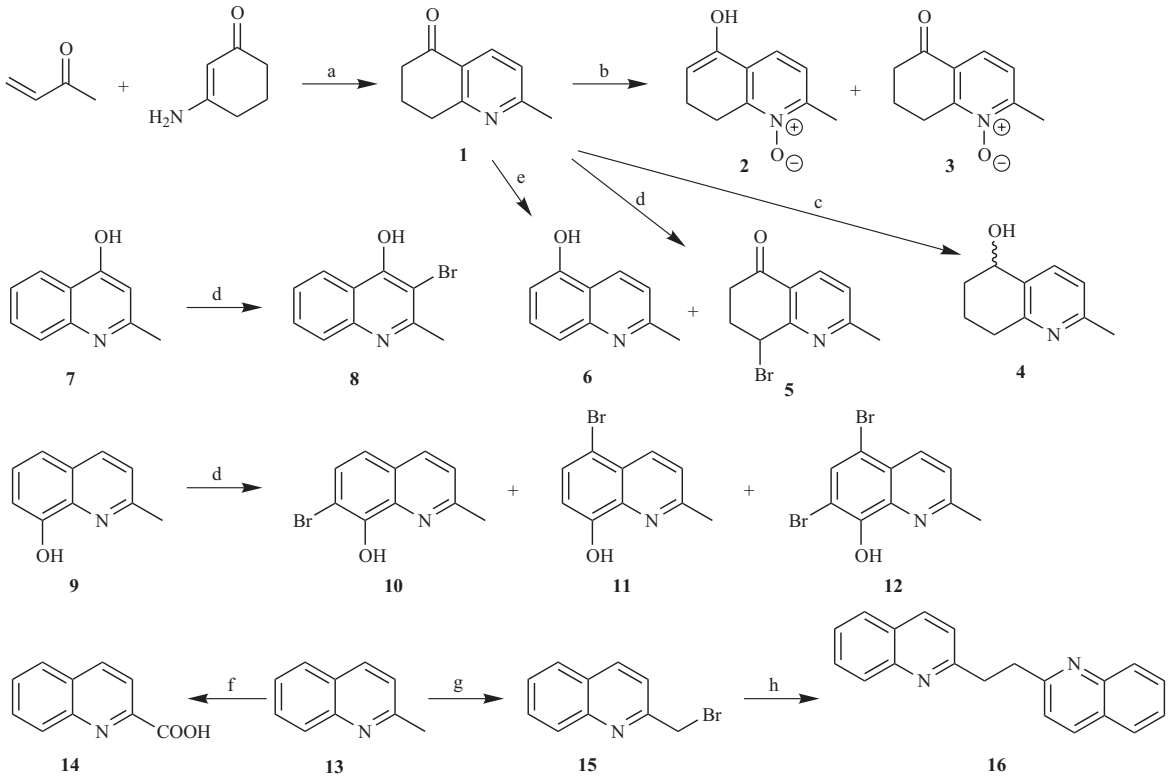
Compounds **7**, **9** and **13** were further starting materials and all were oxidatively brominated using NBS and dibenzoyl peroxide (compound **13** with NBS and 2,2′-azobisisobutyronitrile (AIBN) [31]) to give compounds **8**, **10**-**12**, **15**. Preparations of compounds **8**, **10**-**12** were described using water hydrobromic acid or by means of condensation of 2-amino-4-bromophenol with but-2-enal or bromination using NBS under homogeneous catalysis by sulfuric acid or copper(II)chelates from different precursors [18-20,32].

Quinaldine (**13**) was also oxidized using SeO_2 to acid **14** in 57%. An interesting reaction is dimerization of compound **15**. Product **16** was generated in 42% yield and quinaldine (**13**) in 49% yield *via* reduction with 4-hydroxyacetanilide. The latter was oxidized to *N*-acetyl-*p*-benzoquinoneimine, which was isolated as a yellow crystalline compound with M.p. 109-111 °C; M.p. 107-109 °C [33].

Keto-enol tautomerism of the quinolines possessing the phenol moiety as well as an intramolecular hydrogen bond between the quinoline nitrogen and the phenol moiety are describes in [34].

3.2. Lipophilicity

Hydrophobicities of compounds were calculated using the two programs (Log *P*/CLog *P* values) and measured by means of RP-HPLC determination of capacity factors K and subsequently calculated Log *K*. All values were compared.



Scheme 1. Quinaldine derivatives synthesis.
Conditions: a) DMF; b) MCPBA, DCM; c) Synhydride, toluene; d) NBS, dibenzoyl peroxide, CCl_4; e) DDQ, dioxan; f) SeO_2, dioxan; g) NBS, AIBN, CCl_4; h) 4-hydroxyacetanilide, K_2CO_3, KI, DMF.

The results are shown in Table **1**, illustrated in Fig. (**1**). It can be assumed, the computed Log *P*/CLog *P* values using the program CS ChemOffice Ultra (ChemDraw) ver. 7.0 and the calculated Log *K* values relatively correspond with the expected lipophilicity increasing within individual series of the compounds. The capacity factor K/calculated Log *K* values specify lipophilicity within individual series of the compounds.

The dependences between biological effects and lipophilicity parameters Log *K* of individual compounds is shown in Figs. (**2**,**3**). The dependences of compounds **2** and **3** (*N*-oxides) were not shown in Figs. (**2**,**3**) due to the fact, that Log *K* of these compounds was determined under different chromatographic conditions because of their low hydrophobicity.

3.3. *In Vitro* Antifungal Susceptibility Testing

Sixteen studied compounds were tested for their antifungal activity. Six compounds **2**, **4**, **10**-**12**, **16** showed medium or higher inhibitory activity, see Table **2**. Their MIC ranged from 0.45 to 125 μmol/l. Compounds **5**, **7**, **8** and **14** were not soluble in the testing medium and compounds **1**, **13**, **15** did not show any activity.

The results from this observation have exposed on the importance of phenolic moiety or hydroxylic group, which can be conjugated with heteroaromatic ring, and bromine for sufficient lipophilicity value for antifungal activity. Isomers **10**-**12** possess the necessary solubility and high antifungal activity against all evaluated fungal strains. Compound **12** (dibromo derivatives) and regioisomer **11** (bromine in the quinaldine $C_{(5)}$ position) were the most efficient compounds. Bromine in $C_{(5)}$ seems to be more advantageous than bromine in the $C_{(7)}$ position of quinaldine (compound **10**). An interesting activity showed *N*-oxide **2**, probably due to the conjugated hydroxylic group. Alcohol **4** showed higher activities than its aromatic analogues **6** and **9** as well.

The antifungal activity showed a linear increase with the lipophilicity increasing of the compounds within this series. The activity decreases with too high lipophilicity of compound **16**, see Fig. (**2**), which describes dependence between *in vitro* antifungal activity {log ($1/IC_{80}$ [mol/l])} and logarithm of retention factor (Log *K*) of studied compounds **4**, **6**, **9**-**12**, **16**. The activity of the studied compounds against the fungal strain *Trichophyton mentagrophytes* 445 was selected for illustration of this dependence.

3.4. Herbicidal Activities

Ten compounds were tested for their herbicidal activities. Compounds **14** and **16** were not tested for their herbicidal activities due to low solubility in the testing medium.

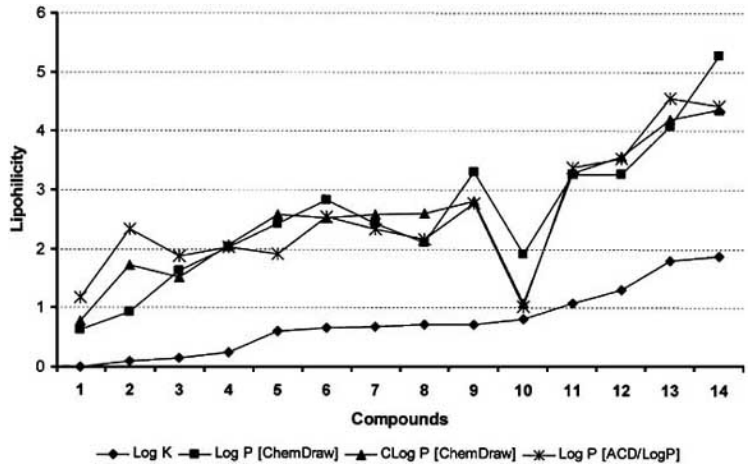


Fig. (1). Comparison of the computed Log *P*/CLog *P* values using the two programs and the calculated Log *K* values.

Compounds **1**, **5**, **7** and **13** did not show any activities in either evaluation. The activities of the other compounds are shown in Table **2**.

The studied compounds can be divided into three groups according to aromaticity of rings A (heterocycle) and B, (see Figure **1**): *i*) Group 1: ring A is aromatic or partly saturated and B is aromatic, compounds **6**, **8**, **9**-**12**, **15**; *ii*) Group 2: ring A is aromatic or partly saturated and B is partly unsaturated, compound **4**; *iii*) Group 3: ring A is *N*-oxide and B is partly unsaturated, compounds **2**, **3**.

3.4.1. PET Inhibition in Spinach Chloroplasts

Seven studied compounds inhibited photosynthetic electron transport in spinach chloroplasts, see Table **2**. The IC_{50} values ranged from 89 to 689 μmol/l. The inhibitory activity of the studied compounds was low, the most efficient inhibitor was compound **2** (IC_{50}: 89 μmol/l). The dependence between PET inhibition in spinach chloroplasts {log ($1/IC_{50}$ [mol/l])} and logarithm of retention factor (Log *K*) of compounds **6**, **8**, **11**, **12**, **15** (Group 1) are shown in Fig. (**3**). It could be assumed, according to the Fig. (**3**), that no activity of the compounds belonging to Group 1 practically depended on the lipophilicity.

Group 1 showed only very low biological activity and Group 2 did not show any biological activity. The most active PET inhibitor of Group 1 was **8** (IC_{50}: 322 μmol/l).

Group 3 (*N*-oxides) showed the highest biological activity. The addition of diphenylcarbazide (an artificial electron donor acting in the intermediate Z^+/D^+ on the donor side of photosystem II [35]) to spinach chloroplasts inhibited by **2** caused complete restoration of the photosynthetic electron transport. This indicates that the primary donor of PS II (P680) was not damaged by this compound. Previous EPR experiments showed that the site of action of the related compounds in the photosynthetic apparatus of spinach chloroplasts was intermediate D^+, i.e. tyrosine radical situated in the 161st position of the protein D_2 located on the donor side of photosystem II [12]. The same site of action could be supposed also for these studied compounds.

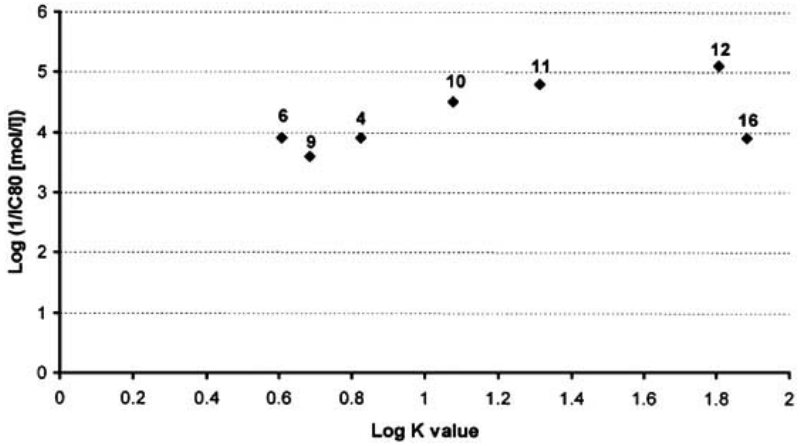


Fig. (2). Dependence between *in vitro* antifungal activity against the fungal strain *Trichophyton mentagrophytes* 445 {log ($1/IC_{80}$[mol/l])} and logarithm of retention factor (Log *K*) of the studied compounds **4**, **6**, **9**-**12**, **16**.

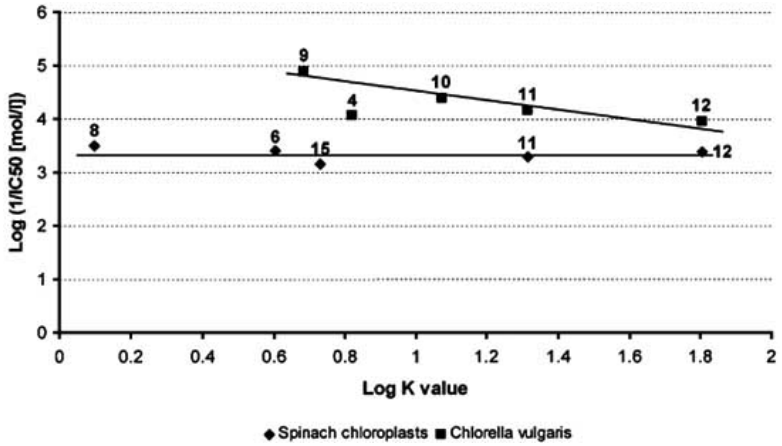


Fig. (3). Dependence between photosynthesis-inhibiting activities {log (1/IC_{50}[mol/l])} and logarithm of retention factor (Log *K*) of the studied compounds **4**, **6**, **8**-**12**.

3.4.2. Reduction of chlorophyll content in Chlorella Vulgaris

Five of the studied compounds inhibited chlorophyll production in *C. vulgaris*, see Table **2**. The interesting IC_{50} values varied in the range from 13.0 (**9**) to 67.9 μmol/l (**11**). Compound **9** (IC_{50}: 13.0 μmol/l) was the most efficient inhibitor.

The dependence between reduction of chlorophyll content in *C. vulgaris* {log (1/IC_{50} [mol/l])} and logarithm of retention factor (Log *K*) of compounds **4** (Group 2), **9**-**12** (Group 1) is shown in Fig. (**3**). The activity showed linear decrease with the lipophilicity increasing of the compounds (especially compounds **9**-**12**, Group 1) within this series, see Fig. (**3**).

Group 1 showed the highest biological activity. Substitution of ring B is more important than that of ring A. Substitution in $C_{(8)}$ by the phenolic group is the necessary condition for the inhibition activity. The inhibitory activity decreased with further substitution of ring B with bromine atoms (increase of lipophilicity).

Groups 2 and 3 showed no or moderate effect on chlorophyll content in *C. vulgaris*.

CONCLUSIONS

The sixteen compounds were tested for their *in vitro* antifungal effect and ten compounds for their herbicidal activities. All the evaluated compounds were characterized. Newly prepared compounds were determined by ^{1}H and ^{13}C NMR spectra, IR spectra, and by means of CHN analysis. 5,7-Dibromo-2-methylquinolin-8-ol (**12**), log *P*: 4.09 and 7-bromo-2-methylquinolin-8-ol (**11**), log *P*: 3.52 showed the highest *in vitro* activities against all eight fungal strains tested. The presence of the phenolic moiety in $C_{(8)}$ and substitution of the quinaldine $C_{(7)}$ and $C_{(5)}$ position by bromine atoms are necessary for the activity. 5-Hydroxy-2-methyl-7,8-dihydroquinolin-1-oxide (**2**) was the most efficient PET inhibitor in spinach chloroplasts; IC_{50}: 89 μmol/l. Sufficient water solubility (*N*-oxide), the conjugated hydroxylic group with heteroaromatic ring and low lipophilicity prove to be advantageous for the biological effect. The most intensive reduction of chlorophyll content in the green algae *C. vulgaris* showed methylquinolin-8-ol (**9**); IC_{50}: 13.0 μmol/l. The phenolic moiety in the quinaldine $C_{(8)}$ position and low lipophilicity are important for this activity.

ACKNOWLEDGEMENTS

This study was supported by the Ministry of Education of the Czech Republic No. MSM 0021620822, and by the Slovak Scientific Grant Agency VEGA No. 1/0089/03.

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Received: 21 March, 2005 Accepted: 04 July, 2005