Department of Pharmaceutical Chemistry[1], Faculty of Pharmacy, Hacettepe University, Ankara, and Department of Pharmacology[2], Faculty of Medicine, Osmangazi University, Eskişehir, Turkey

4-Aryl-6,6-dimethyl-1,2,3,4,5,6,7,8-octahydroquinazoline-2,5-diones: synthesis, chromatographic resolution and pharmacological activity

S. Saraç[1], M. Yarim[1], M. Ertan[1], F. S. Kiliç[2], and K. Erol[2]

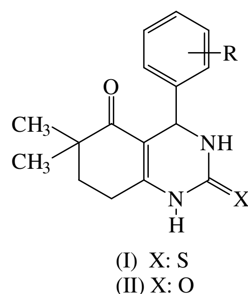
In this study, a series of 4-aryl-6,6-dimethyl-1,2,3,4,5,6,7,8-octahydroquinazoline-2,5-diones were synthesized by condensing urea with 4,4-dimethyl-1,3-cyclohexanedione and appropriate aromatic aldehydes according to the Biginelli reaction. The structures of the compounds were characterized by spectroscopic methods. The racemic compounds were resolved into the enantiomers by HPLC on amylose tris(3,5-dimethylphenylcarbamate) (Chiralpak AD). The compounds were tested *in vitro* for their calcium antagonistic activities. $BaCl_2$-induced contractions of rat ileum were inhibited dose-dependently. Compounds **3** and **5** exerted weak calcium antagonistic activity on smooth muscles compared with the standard, nicardipine.

1. Introduction

In our previous studies, the synthesis and calcium antagonist activities of some condensed tetrahydropyrimidine derivatives, such as 4-aryl-5-oxo-1,2,3,4,5,6,7,8-octahydroquinazoline-2-thione (**I**) and 4-aryl-1,2,3,4,5,6,7,8-octahydroquinazoline-2,5-diones (**II**) have been described [1, 2]. Some of these compounds exhibited calcium antagonist activity on rats in preliminary pharmacological investigations. In continuation of these investigations, we became interested in the synthesis and calcium antagonist activities of new annulated tetrahydropyrimidine derivatives having methyl groups on the 6-position of the octahydroquinazoline ring structure.

These compounds have a chiral center and, as stereoisomers often show different pharmacological activities, it seems advisable to resolve the racemic mixtures to investigate the biological properties of each enantiomer. The most widely used technique for the separation and quantification of enantiomers is HPLC. In the development of an HPLC method, a chiral stationary phase (CSP) is used to separate the enantiomers directly because of the simplicity and ease of operation resulting from this approach. Separation of enantiomers by chiral HPLC is now well established with over 50 different CSPs commercially available. Among them cellulose and amylose carbamate derivatives coated onto silica gel have proved to be extremely useful CSPs for chiral resolution in normal-phase mode [3–11].

In this paper, the enantioseparation of 4-aryl-6,6-dimethyl-1,2,3,4,5,6,7,8-octahydroquinazoline-2,5-diones was also studied on tris(3,5-dimethylphenylcarbamate) derivatives of cellulose and amylose.



2. Investigations, results and discussion

2.1. Chemistry

The synthetic pathway employed in the preparation of 4-aryl-6,6-dimethyl-1,2,3,4,5,6,7,8-octahydroquinazoline-2,5-diones is outlined in Scheme 1. The condensation of 4,4-dimethyl-1,3-cylohexanedione, substituted benzaldehyde and urea in absol. ethanol, accelerated by HCl, proceeded according to the Biginelli reaction [12]. Yields ranged from 23 to 38% (Table 1).

Table 1: Melting points, yields and empirical formulae of the compounds synthesized

Compd.	R	M.p. (°C)	Yield (%)	Empirical Formula
1	3-Cl	301–2	24.11	$C_{16}H_{17}ClN_2O_2$
2	4-Cl	306–7	23.78	$C_{16}H_{17}ClN_2O_2$
3	4-Br	310–1	33.56	$C_{16}H_{17}BrN_2O_2$
4	3-CH_3	294–5	29.45	$C_{17}H_{20}N_2O_2$
5	4-CH_3	>315	37.76	$C_{17}H_{20}N_2O_2$
6	3-OCH_3	260–1	22.79	$C_{17}H_{20}N_2O_3$

Scheme 1

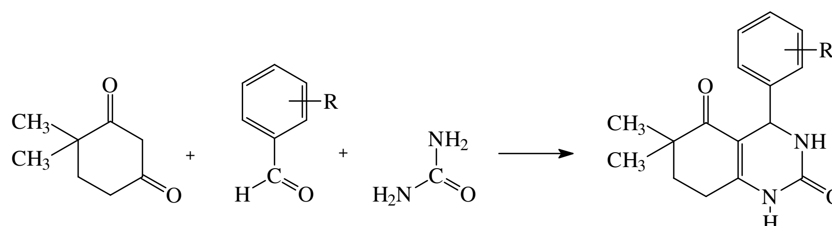


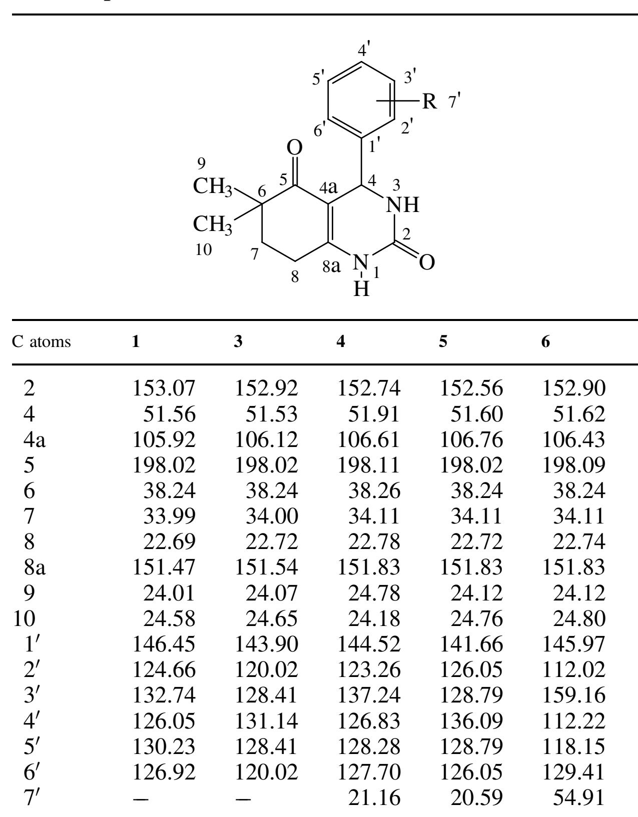
Table 2: UV, IR, ^{1}H-NMR spectral data of the compounds

Compd.	UV_{max}^{MeOH} (log ε)	IR (KBr) υ (cm^{-1})	^{1}H-NMR ([D_6]DMSO) δ (ppm)
1	293 (3.98) 207 (4.08)	3264 (N–H) 1715 (5-C=O) 1614 (2-C=O) 759, 691 (1,3-disubst. benzene)	0.92 (3 H, s, CH_3), 0.99 (3 H, s, CH_3), 1.76 (2 H, t, 7-H_2), 2.48 (2 H, t, 8-H_2), 5.13 (1 H, d, J = 3 Hz, 4-H), 7.14–7.34 (4 H, m, phenyl protons), 7.76 (1 H, s, 3-NH), 9.48 (1 H, s, 1-NH)
2	294 (3.94) 221 (4.08)	3256 (N–H) 1713 (5-C=O) 1620 (2-C=O) 831 (1,4-disubst. benzene)	0.93 (3 H, s, CH_3), 0.99 (3 H, s, CH_3), 1.78 (2 H, t, 7-H_2), 2.49 (2 H, t, 8-H_2), 5.14 (1 H, d, J = 2.9 Hz, 4-H), 7.18–7.40 (4 H, m, phenyl protons), 7.77 (1 H, s, 3-NH), 9.49 (1 H, s, 1-NH)
3	293 (4.00) 221 (4.06)	3268 (N–H) 1714 (5-C=O) 1615 (2-C=O) 836 (1,4-disubst. benzene)	0.90 (3 H, s, CH_3), 0.99 (3 H, s, CH_3), 1.75 (2 H, t, 7-H_2), 2.49 (2 H, t, 8-H_2), 5.10 (1 H, d, J = 2.9 Hz, 4-H), 7.14–7.52 (4 H, m, phenyl protons), 7.75 (1 H, s, 3-NH), 9.47 (1 H, s, 1-NH)
4	293 (4.01) 210 (4.07)	3261 (N–H) 1716 (5-C=O) 1612 (2-C=O) 761, 707 (1,3-disubst. benzene)	0.91 (3 H, s, CH_3), 0.99 (3 H, s, CH_3), 1.76 (2 H, t, 7-H_2), 2.25 (3 H, s, Ar–CH_3), 2.48 (2 H, t, 8-H_2), 5.07 (1 H, d, J = 3 Hz, 4-H), 6.98–7.21 (4 H, m, phenyl protons), 7.67 (1 H, s, 3-NH), 9.38 (1 H, s, 1-NH)
5	293 (3.92) 213 (3.96)	3263 (N–H) 1678 (5-C=O) 1611 (2-C=O) 840 (1,4-disubst. benzene)	0.90 (3 H, s, CH_3), 0.99 (3 H, s, CH_3), 1.75 (2 H, t, 7-H_2), 2.23 (3 H, s, Ar–CH_3), 2.48 (2 H, t, 8-H_2), 5.07 (1 H, d, J = 3.1 Hz, 4-H), 7.08 (4 H, s, phenyl protons), 7.67 (1 H, s, 3-NH), 9.38 (1 H, s, 1-NH)
6	292 (4.03) 206 (4.09)	3248 (N–H) 1697 (5-C=O) 1612 (2-C=O) 768, 709 (1,3-disubst. benzene)	0.93 (3 H, s, CH_3), 0.99 (3 H, s, CH_3), 1.76 (2 H, t, 7-H_2), 2.48 (2 H, t, 8-H_2), 3.70 (3 H, s, OCH_3), 5.09 (1 H, d, J = 3 Hz, 4-H), 7.17–7.25 (4 H, m, phenyl protons), 7.71 (1 H, s, 3-NH), 9.41 (1 H, s, 1-NH)

The structures of the compounds were confirmed by spectral data. All UV spectra showed two absorption maxima in the 206–221 and 292–294 nm regions, respectively. In the IR spectra all the compounds displayed strong absorption bands at 3248–3268 cm^{-1} (N–H), 1678–1716 cm^{-1} (5-C=O) and 1611–1620 cm^{-1} (2-C=O), respectively. Additionally, disubstituted benzene deformation bands were observed in the expected wave number regions. In the ^{1}H NMR spectra, methyl protons, 7-H_2 and 8-H_2 protons of octahydroquinazoline ring and C-4 phenyl protons resonated at the expected regions. The 4-H proton exhibited signals at the δ = 5.07–5.14 region as doublets. Two downfield one-proton singlets were assigned to the N–H protons of the octahydroquinazoline structure (Table 2). In the ^{13}C NMR spectra of the compounds the most deshielded carbon atoms are C-5 and C-2 (Table 3). Other carbon atoms were seen at the expected chemical shift values. 6,6-Dimethyl-1,2,3,4,5,6,7,8-octahydroquinazoline-2,5-dione structure was also confirmed by NOE-difference measurements (Table 4).

The EI MS of the compounds showed molecular ion $[M]^{+\cdot}$ peaks of different intensity which confirmed their molecular weights. The major fragmentation pathway was formed by the cleavage of the $C_6H_4R^\cdot$ or $R^\cdot$ radical from the molecular ion (Scheme 2).

Table 3: ^{13}C-NMR ([D_6]DMSO) spectral data of the compounds



C atoms	1	3	4	5	6
2	153.07	152.92	152.74	152.56	152.90
4	51.56	51.53	51.91	51.60	51.62
4a	105.92	106.12	106.61	106.76	106.43
5	198.02	198.02	198.11	198.02	198.09
6	38.24	38.24	38.26	38.24	38.24
7	33.99	34.00	34.11	34.11	34.11
8	22.69	22.72	22.78	22.72	22.74
8a	151.47	151.54	151.83	151.83	151.83
9	24.01	24.07	24.78	24.12	24.12
10	24.58	24.65	24.18	24.76	24.80
1′	146.45	143.90	144.52	141.66	145.97
2′	124.66	120.02	123.26	126.05	112.02
3′	132.74	128.41	137.24	128.79	159.16
4′	126.05	131.14	126.83	136.09	112.22
5′	130.23	128.41	128.28	128.79	118.15
6′	126.92	120.02	127.70	126.05	129.41
7′	–	–	21.16	20.59	54.91

2.2. *Enantioseparation of compounds 1–6*

In general tetrahydropyrimidine derivatives are formed as racemates in the classical Biginelli synthesis or related methods [13–16]. Since individual enantiomers of chiral tetrahydropyrimidines have opposing pharmacological effects on the calcium channel [17], the use of enantiomerically pure compounds is a requirement for the development of drugs of this type. Enantiomerically pure compounds were obtained by resolution of the corresponding racemic carboxylic acids via diastereomeric ammonium salts [18], by separation of diastereomeric derivatives bearing chiral auxiliaries at N-3 or by enantioselective HPLC [19, 20]. Recently, the chromatographic resolution of dihydropyrimidines by HPLC has been reported [21–23].

Table 4: NOE-difference experimental results of compounds 1, 3 and 5

1	3	5
1-NH to 8-H_2 (s) 3-NH to 4-H (s) 6-$(CH_3)_2$ to 7-H_2 (s)	1-NH to 8-H_2 (w) 3-NH to 4-H (s) 6-$(CH_3)_2$ to 7-H_2 (w) 7-H_2 to 6-$(CH_3)_2$ (s), 8-H_2 (s)	1-NH to 8-H_2 (w) 3-NH to 4-H (s) 8-H_2 to 1-NH (s), 7-H_2 (w) Ar-H to 4-H (w), Ar-CH_3(w)

s: strong, w: weak

Table 5: Separation characteristics of compounds 1–6 on amylose tris(3,5-dimethylphenylcarbamate) CSP (Chiralpak AD)

Compd.	R	k_1'	k_2'	α	R_s	N_1	N_2
1	3-Cl	0.33	1.07	3.24	4.33	1600	1886
2	4-Cl	0.62	0.86	1.39	1.23	1600	1510
3	4-Br	0.75	1.01	1.35	1.06	992	1024
4	3-CH_3	0.41	1.43	3.49	4.63	1156	1239
5	4-CH_3	0.65	1.20	1.85	2.53	1215	1600
6	3-OCH_3	0.64	1.80	2.81	5.13	1600	1632

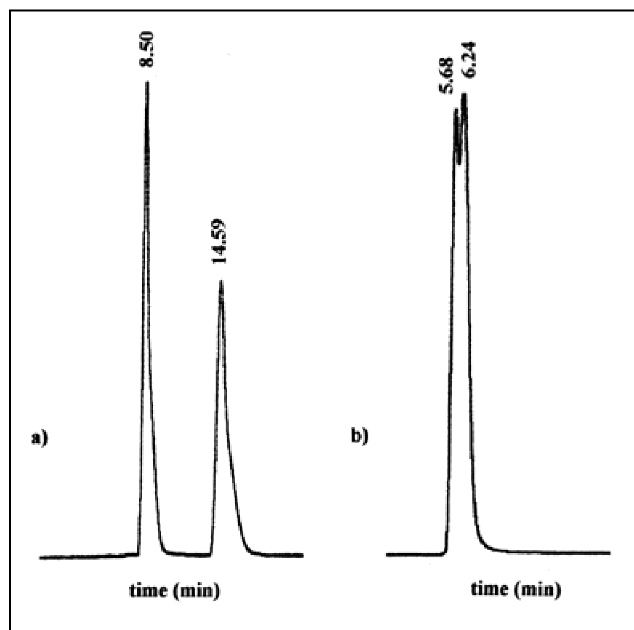


Fig.: Chromatograms of the separation of compound **4** on (a) Chiralpak AD column (25 cm × 4.6 mm) and (b) Chiralcel OD column (25 cm × 4.6 mm) using ethanol : n-hexane (90 : 10, v/v) as the mobile phase. Flow rate: 0.5 ml/min at room temperature; UV detection: 320 nm

The two chiral stationary phases used in our study for the enantiomeric separation of condensed tetrahydropyrimidines were cellulose tris(3,5-dimethylphenylcarbamate) (Chiralcel OD) and amylose tris(3,5-dimethylphenylcarbamate) (Chiralpak AD) coated on silica gel. It has been assumed that separation of enantiomers on cellulose- and amylose-based CSPs was due to the formation of solute-CSP complexes between the enantiomers and the chiral cavities in the higher order structures of the CSPs [8, 24, 25]. In polysaccharide phenylcarbamate-type CSPs, such as the CSPs used in our study, the binding of the solutes to the CSPs results from interactions between the solutes and the polar carbamate groups on the CSPs [8, 9, 25]. The carbamate groups on the CSP can interact with solutes through hydrogen bonding using the C=O and NH groups and through dipole-dipole interactions using the C=O moiety. In the cases of compounds **1**–**6**, the NH and C=O groups on the solute could form hydrogen bonds with the C=O and NH groups on the CSPs. Dipole-dipole interactions could also occur between the C=O groups on the solutes and C=O groups on the stationary phases. Wainer et al. [24] have reported that the solute-CSP complex, formed between solute aromatic functionalities and a cellulose-based CSP, can be stabilized by insertion of the aromatic moiety of the solute into the chiral cavity. This type of stabilization interaction might also occur in our case. Chiral discrimination between the enantiomers was due to the differences in their steric fit in the chiral cavities [8, 24, 25].

The chromatographic data for the separation of compounds **1**–**6** on Chiralpak AD column are given in Table 5. Under the conditions chosen, the enantiomers of compounds **1**, **4**, **5** and **6** were baseline separated on Chiralpak AD in a short time. The substitution of the phenyl ring in the 3-position led to a strongly improved separation on the amylose selector. The good separation of optical isomers of 3-chloro (compound **1**), 3-methyl (compound **4**) and 3-methoxy (compound **6**) derivatives not only makes this chromatographic method suitable for quantifying optical purity but also opens the way to the rapid preparative HPLC isolation of individual enantiomers.

The separation of compounds **1**–**6** was also examined on a Chiralcel OD column, the cellulose-based counterpart of

Scheme 2

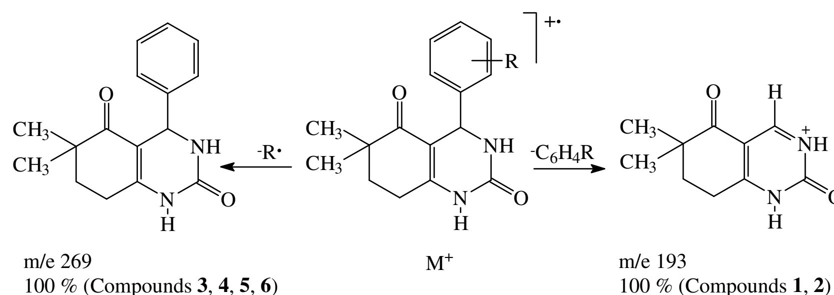


Table 6: The relaxant effects of nicardipine and compounds 1–6 on induced contraction of isolated rat ileum

Compd.	10^{-6} M	10^{-5} M	10^{-4} M
1	0	0	21.83 ± 5.29
2	0	0	11.17 ± 3.28
3	0	9.83 ± 2.27	44.83 ± 4.04
4	0	0	11.50 ± 3.92
5	0	28.33 ± 6.17	33.00 ± 8.18
6	0	0	13.67 ± 4.40
Nicardipine (10^{-8} M)	71.50 ± 5.54		

n (number of heads used) = 6

the Chiralpak AD column. On the OD column the derivatization groups on the cellulose were the same [tris(3,5-dimethylphenyl(carbamate)] as those on the amylose-based AD column. However, apart from the 3-CH_3 derivative (compound **4**), none of the compounds were separated on the OD column with ethanol : n-hexane (90 : 10) as the mobile phase. The resolution of compound **4** on OD column was poor ($\alpha = 1.10$, $R_s = 0.31$) compared to that on the AD column ($\alpha = 3.49$, $R_s = 4.63$). The separation of compound **4** on the AD and OD columns is shown in the Fig.
The amylose-based Chiralpak AD column and its cellulose-based counterpart, Chiralcel OD, retained the solutes differently although the derivatization groups on both CSPs were the same. This suggests that the retention of solutes depends not only on the derivatization group on the CSP, but also on the higher order structure of the CSP.

2.3. Pharmacology

Bioassay preparations such as isolated right (chronotropy) and left (inotropy) atria of guinea pig [26–28], rabbit portal vein, aortic strips of rabbit, isolated papillary muscle of guinea pig [29], guinea pig taenia coli in K^+-depolarizing Tyrode solution [30], isolated guinea pig ileum (Ba^{+2} stimulation) [29, 31], isolated rat ileum (Ca^{+2} stimulation) [32], and the radioligand binding method [33] are appropriate for pharmacological screening tests for calcium antagonists. In the present preliminary assay, the calcium antagonist activity of the racemic compounds **1–6** was evaluated *in vitro* by suppressing $BaCl_2$-induced contractions in rat ileum in comparison with the activity of nicardipine under the same experimental conditions. The maximum relaxation values of all these compounds were negligible when compared to those of nicardipine. Compounds **3** and **5**, having 4-bromo and 4-methyl substituents on the aromatic ring, respectively, are the most active derivatives (Table 6). Furthermore, the relaxant effects of these compounds were smaller than that of nicardipine.

3. Experimental

3.1. Chemistry

3.1.1. Equipment

All chemicals used in this study were supplied by E. Merck (Darmstadt, Germany). M.p.'s were determined with a Thomas-Hoover Capillary apparatus and are uncorrected. UV absorptions were measured on a Shimadzu UV-160A UV-visible spectrophotometer. The IR spectra were taken with a Perkin Elmer FT-IR spectrophotometer 1720 × (KBr disc). ^{1}H and ^{13}C NMR spectra were recorded on a Bruker AC 200 MHz FT NMR spectrometers using tetramethylsilane as the internal standard. All chemical shift values were recorded as δ (ppm). MS were obtained with a VG analytical 70–250S mass spectrometer with electron ionization (EI). The purity of the compounds was checked by TLC (Merck, silica gel, $HF_{254-361}$, Type 60, 0.25 mm). The elementary analysis of the compounds was performed on a Leco CHNS 932 analyzer at the Scientific and Technical Research Council of Turkey, Instrumental Analysis Laboratory at Ankara. The result of elementary analysis of the compounds also supported the postulated structure.

3.1.2. Preparation of 4-aryl-6,6-dimethyl-1,2,3,4,5,6,7,8-octahydroquinazoline-2,5-diones

A mixture of urea (0.3 g, 0.005 mol), benzaldehyde or substituted benzaldehyde (0.005 mol), 4,4-dimethyl-1,3-cyclohexanedione (1.107 g, 0.0075 mol), abs. ethanol (20 ml) and 37% HCl (4 drops) was heated under reflux for an appropriate period and the reaction solution was allowed to cool. The product which appeared as a precipitate was filtered off and washed with 50% ethanol (25 ml). Then it was recrystallized from ethanol.

3.2. HPLC studies

3.2.1. Chromatographic conditions

HPLC analysis was performed at room temperature with a Knauer HPLC Pump 64, a Rheodyne 7125 Injector, a Hitachi 655 A Variable Wavelength UV Monitor set at 320 nm and a Hewlett Packard HP 3396 Integrator. The stainless steel columns (25 cm × 4.6 mm) packed with Chiralcel OD [cellulose tris(3,5-dimethylphenylcarbamate) coated on silica gel (10 μm particle size)] and Chiralpak AD [amylose tris(3,5-dimethylphenylcarbamate) coated on silica gel (10 μm particle size)] were obtained from Daicel Chemical Industries, Tokyo, Japan. The separation was achieved with ethanol: n-hexane (90 : 10 v/v) as the mobile phase. The flow rate was 0.5 ml/min. The elution time of 1,3,5-tri-*tert*-butylbenzene was used as the dead time (t_0).

3.2.2. Chromatographic characteristics

The separation factor (α), which represents a measure of relative peak separation is expressed as follows:

$$\alpha = k_2'/k_1'$$

where k_1' and k_2' are capacity factors for the first and second eluting enantiomers, respectively. The capacity factor (k') is calculated as follows:

$$k_1' = (t_1 - t_0)/t_0 \text{ and } k_2' = (t_2 - t_0)/t_0$$

where t_0, t_1 and t_2 refer to the retention time in seconds for the nonretained compound, and the first and second eluting enantiomers, respectively.
The stereochemical resolution factor (R_s) of the first and second eluting enantiomer peaks is calculated by the ratio of the difference between the retention times (t_1 and t_2) to the arithmetic mean of the two peak widths

$$R_s = 2(t_2 - t_1)/(w_1 + w_2).$$

3.3. Calcium antagonist activity

Male and female albino rats weighing between 200–220 g were used in this study. Animals entered the test having fasted overnight. After the animals had been sacrified by cervical dislocation, the ileum (10–15 cm terminal portion) was immediately removed, discarding the 5–8 cm segment proximal to the ilio-caecal junction. Segments 1.5–2 cm long were mounted vertically in a 10 ml organ bath containing Tyrode solution of the following composition (mM): NaCl: 136.87, KCl: 2.68, $CaCl_2$: 1.80, $MgSO_4$: 0.81, NaH_2PO_4: 4.16, $NaHCO_3$: 11.9, Glucose: 11.1. The bath contents were maintained at 37 °C and aerated by 95% O_2 and 5% CO_2.
A tension of 2 g was applied and isometric recording was done using an isometric transducer (T-FDT_{10}-A). Responses were recorded with a MAY TDA95 transducer data acquisition system.
The preparations were allowed to equilibrate for 60 min with regular washes every 15 min. In order to check for antagonistic effects, contractions were induced with barium chloride (4×10^{-3} mol/l, bath concentration). After thorough washing out, this process was repeated until the amplitude of the contraction became constant. The substances to be tested were investigated using the single-dose technique. Barium chloride contractions were induced after addition of the test substances at different concentrations (10^{-6}, 10^{-5} and 10^{-4} M) and 5 min exposure time. Only one compound was tested in each preparation. Because of solubility problems, the compounds were dissolved in dimethylsulfoxide (DMSO) and the control responses were taken after the addition of 0.1 ml DMSO.
Results were expressed as a percentage of the maximum relaxation of the contractions of the compounds (Table 6). The responses of the compounds were compared the those of nicardipine. Student's t test was used for statistical analysis. p Values < 0.05 were considered to be statistically significant.

Acknowledgements: The authors would like to thank Prof. Dr. G. Blaschke from Westfälische Wilhelms-University of Münster (Germany) for spectral analysis and HPLC studies.

References

1 Saraç, S.; Yarım, M.; Ertan, M.; Erol, K.; Aktan, Y.: Boll. Chim. Farm. **136**, 657 (1997)
2 Saraç, S.; Yarım, M.; Ertan, M.; Boydağ, S.; Erol, K.; Pharmazie **53**, 91 (1998)

3 Okamoto, Y.; Kawashima, M.; Hatada, K.: J. Am. Chem. Soc. **106**, 5357 (1984)
4 Okamoto, Y.; Kawashima, M.; Hatada, K.: J. Chromatogr. **363**, 173 (1986)
5 Okamoto, Y.; Aburatani, R.; Fukumoto, T.; Hatada, K.: Chem. Lett. 1857 (1987)
6 Okamoto, Y.; Aburatani, R.; Hatano, K.; Hatada, K.: J. Liq. Chromatogr. **11**, 2147 (1988)
7 Ikeda, K.; Hamasaki, T.; Kohno, H.; Ogawa, T.; Matsumoto, T.; Sakai, J.: Chem. Lett. 1089 (1989)
8 Okamoto, Y.; Kaida, Y.: J. Chromatogr. A **666**, 403 (1994)
9 Yashima, E.; Okamoto, Y.: Bull. Chem. Soc. Jpn. **68**, 3289 (1995)
10 Chankvetadze, B.; Yashima, E.; Okamoto, Y.: J. Chromatogr. A **694**, 101 (1995)
11 Yashima, E.; Yamamoto, C.; Okamoto, Y.: SYNLETT, 344 (1998)
12 Biginelli, P.: Chem. Ber. **24**, 1317 (1891)
13 Kappe, C. O.: Tetrahedron **49**, 6937 (1993)
14 O'Reilly, B. C.; Atwal, K. S.: Heterocycles **26**, 1185 (1987)
15 Atwal, K. S.; O'Reilly, B. C.; Gougoutas, J. Z.; Malley, M. F.: Heterocycles **26**, 1189 (1987)
16 Atwal, K. S.; Rovnyak, G. C.; O'Reilly, B. C.; Schwartz, J.: J. Org. Chem. **54**, 5898 (1989)
17 Rovnyak, G. C.; Kimball, S. D.; Beyer, B.; Cucinotta, G.; DiMarco, J. D.; Gougoutas, J.; Hedberg, A.; Malley, M.; McCarthy, J. P.; Zhang, R.; Moreland, S.: J. Med. Chem. **38**, 119 (1995)
18 Kappe, C. O.; Uray, G.; Roschger, P.; Lindner, W.; Kratky, C.; Keller, W.: Tetrahedron **48**, 5473 (1992)
19 Atwal, K. S.; Swanson, B. N.; Unger, S. E.; Floyd, D. M.; Moreland, S.; Hedberg, A.; O'Reilly, B. C.: J. Med. Chem. **34**, 806 (1991)
20 Rovnyak, G. C.; Atwal, K. S.; Hedberg, A.; Kimball, S. D.; Moreland, S.; Gougoutas, J. Z.; O'Reilly, B. C.; Schwartz, J.; Malley, M. F.: J. Med. Chem. **35**, 3254 (1992)
21 Stalcup, A. M.; Chang, S-C.; Armstrong, D. W.; Pitha, J.: J. Chromatogr. A **513**, 181 (1990)
22 Armstrong, D. W.; Chang, C. D.; Lee, S. H.: J. Chromatogr. A **539**, 83 (1991)
23 Kleidernigg, O. P.; Kappe, C. O.: Tetrahedron: Asymmetry **8**, 2057 (1997)
24 Wainer, I. W.; Stiffin, R. M.; Shibata, T.: J. Chromatogr. **411**, 139 (1987)
25 Okamoto, Y.; Yashima, E.: Angew. Chem., Int. Ed. Engl. **37**, 1020 (1998)
26 Rose, U.: Arzneim.-Forsch./Drug Res. **39** (II), 1393 (1989)
27 Rose, U.: J. Heterocycl. Chem. **27**, 237 (1990)
28 Rose, U.: J. Heterocycl. Chem. **28**, 2005 (1991)
29 Kazda, S., Garthoff, B.; Meyer, H.; Schloßmann, K.; Stoepel, K.; Towart, R.; Vater, W.; Wehinger, E.: Arzneim.-Forsch./Drug Res. **30**(II), 2144 (1980)
30 Şafak, C.; Erdemli, I. Sunal, R.: Arzneim.-Forsch./Drug Res. **43**(II), 1052 (1993)
31 Rose, U.; Dräger, M.: J. Med. Chem. **35**, 2238 (1992)
32 Şafak, C.; Özkanlı, F.; Erol, K.; Aktan, Y.: Arzneim.-Forsch./Drug Res. **45**(II), 1154 (1995)
33 Spedding, M.; Cavero, I.: Life Sci. **35**, 575 (1984)

Received June 26, 2000
Accepted September 1, 2000

Assoc. Prof. Dr. Selma Saraç
Hacettepe University Faculty of Pharmacy
Department of Pharmaceutical Chemistry
06100 Sıhhiye-Ankara
Turkey