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Automated analysis of quetiapine and other antipsychotic drugs in human blood by high performance-liquid chromatography with column-switching and spectrophotometric detection☆

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Abstract

An automated HPLC method with column switching is described for the determination of quetiapine, clozapine, perazine, olanzapine and metabolites in blood serum. After clean-up on silica C8 material (20 μm particle size) drugs were separated on ODS Hypersil C18 material (5 μm; column size 250 mm × 4.6 mm i.d.) within 25 min and quantified by ultraviolet (UV) detection at 254 nm. The limit of quantification ranged between 10 and 50 ng/ml. At therapeutic concentrations of the drugs, the inter-assay reproducibility was below 10%. Analyses of drug concentrations in serum of 75–295 patients treated with therapeutic doses of the antipsychotic drugs revealed mean ± S.D. steady state concentrations of 139 ± 136 ng/ml for quetiapine, 328 ± 195 ng/ml for clozapine, 48 ± 27 ng/ml for olanzapine and 71 ± 52 ng/ml for perazine. The method was thus suitable for routine therapeutic drug monitoring and may be extended to other drugs.

Keywords: Quetiapine; Clozapine; Norclozapine; Olanzapine; Perazine; High-performance liquid chromatography; Ultraviolet detection; Column-switching; TDM; Therapeutic drug monitoring

1. Introduction

Quetiapine was introduced in the clinic as a new antipsychotic drug for the treatment of schizophrenia and other psychotic [1,2] or schizoaffective disorders [3]. Quetiapine is in the same family as clozapine and olanzapine, which are classified as "atypical" antipsychotics [4] and do not cause major extrapyramidal side effects. Each is effective in the treatment of schizophrenia, treating both the positive and negative symptoms [1–3]. These new antipsychotics have markedly improved the quality of life in many schizophrenic patients and have consequently become first line antipsychotics.

Blood levels of psychoactive drugs resulting under a given dose are highly variable between individual patients. This is primarily due to inter-individual variations in compliance and in the activities of drug metabolizing enzymes. This leads to poor predictability of drug concentrations at a given dose. Dose correction is therefore required in about 50% of patients. For a number of typical and atypical antipsychotics, evidence has shown that therapeutic drug monitoring (TDM) improves response rates, minimizes side effects and reduces the risk of relapses under chronic treatment [5–8]. A pre-requisite to use TDM for treatment optimization is the availability of a valid and robust method that is suitable in a clinical routine. Since approximately 30 antipsychotic drugs are currently available in the clinic, it is advantageous to use an assay that is suitable for determination of multiple drugs. A current method using electrospray ionization mass spectrometry with off-line extraction before chromatographic separation has been reported for the measurement of clozapine, olanzapine, risperidone and quetiapine in plasma [9]. In the clinical routine, however, HPLC-UV may be advantageous because of lower cost and robustness. A fully automated on-line quantification of quetiapine in human serum using UV-detection was reported by Hasselstrøm and Linnet [10]. Using column switching with an on-line sample clean up may be even more advantageous, since it enables automated sample analysis in clinical routine [11–15]. However, these HPLC methods are restricted to few drugs. Here we describe the validation and

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application of an automated high performance liquid chromatographic method with column switching for rapid determination of quetiapine, which enables also the quantification of clozapine and its metabolite norclozapine as well as of olanzapine, perazine and which may be extended to other drugs.

2. Experimental

2.1. Chemicals

Quetiapine (free base, CAS No. 111974-72-2) by Astra Zeneca (London, United Kingdom), clozapine and norclozapine (free base) were kindly supplied by Novartis (Nürnberg, Germany), olanzapine (free base) by Lilly (Indianapolis, IN, USA) and perazine (dimalonate) by Lundbeck (Hamburg, Germany). Fluperlapine (free base, CAS No. 67121-76-0) was obtained from Biomol (Hamburg, Germany), acetonitrile (HPLC grade) from Merck (Darmstadt, Germany). TEMED (*N*,*N*,*N*,*N*-tetramethylethylenediamine) and acetic acid were obtained from Sigma (Taufkirchen, Germany). Water was deionized and filtered through a Milli-Q water processing system (Millipore, Eschborn, Germany).

2.2. Calibration and control samples

Stock solutions for quality control (QC) and calibration (C) samples were prepared by dissolving 10 mg of quetiapine, clozapine, norclozapine and olanzapine (free base) and 10 mg of perazine (dimalonate) in 10.0 ml methanol each. They were diluted with deionized water and mixed with drug free plasma from healthy volunteers to obtain standards of low to high concentrations.

The stock solution of the internal standard was prepared by dissolving 26.4 mg of fluperlapine in 10.0 ml methanol. This stock solution was diluted with deionized water to obtain a standard with a concentration of 40 μg/ml.

2.3. Patient samples

Blood samples, for the analysis of blood serum, were taken from patients who had been treated with quetiapine, clozapine, perazine or olanzapine. Blood was collected in the morning immediately before the first daily dose of the antipsychotics, in order to analyze the trough serum concentrations. Serum was prepared by centrifugation of blood samples at $4000 \times g$ for 10 min and, if not assayed on the same day, stored at $-20\,^{\circ}C$. Samples could be stored for several months without relevant decomposition of the drugs. After thawing, samples were centrifuged at $1500 \times g$ for 5 min before analysis.

Only samples sent to our laboratory for clinical monitoring were included in this study. All patient identifiers were removed before entry into the statistical database.

2.4. Instrumentation

The HPLC system consisted of an Agilent 1100 series with a binary pump, an autosampler, a thermostatted column compartment (containing an electric six-port switching valve) and a variable wavelength detector set at 254 nm. All HPLC components were purchased from Bio-Rad (München, Germany). Data acquisition and integration was performed by means of the HP ChemStation (Version A.06.01). The analytical column (250 mm × 4.6 mm i.d.) was packed with Hypersil ODS C18 (5 μm particle size) by MZ-Analysentechnik (Mainz, Germany). The clean-up column (10 mm × 4.0 mm) was filled with 20 μm particles of Silica C8 material (MZ-Analysentechnik).

2.5. Chromatographic procedure

Sample clean-up and chromatographic separation were performed at room temperature as follows:

2.5.1. 0–5 min

After re-centrifugation of serum ($1500 \times g$ for 5 min), 99 μl of the supernatant and 1 μl standard solution of the internal standard (fluperlapine) were mixed by the autosampler and injected at 0 min onto the clean-up column. Proteins and other interfering compounds were washed to waste using deionized water containing 8% (v/v) acetonitrile at a flow rate of 1.3 ml/min for 5 min.

2.5.2. 5–15 min

At 5 min, the electric six-port valve was switched to start the analytical run which used acetonitrile–water–tetramethylethylenediamine (37.5:62.1:0.4, v/v/v), adjusted to pH 6.5 by acetic acid (95%) to elute the analytes from the clean-up column onto the analytical column (back flush mode) for separation at a flow rate of 1.3 ml/min.

2.5.3. 15–25 min

At 15 min, the switching valve was reset and the precolumn rinsed with the clean-up eluent (8% acetonitrile in water). Separation on the analytical column continued until 25 min.

The clean-up column was replaced after injection of about 100 serum samples (=10 ml serum).

2.6. Interferences

To control for possible interferences with drugs that may be used in combination the suggested interfering compounds were prepared in blank serum as described for QC- and C-samples.

2.7. Calculations

Peak heights obtained from the analysis of spiked plasma, containing known amounts of drugs, were subjected to linear regression analysis for the calculation of correlation coefficients, slopes and intercepts. Drug concentrations in samples containing unknown amounts of drug were calculated on the basis of the computed regression lines.

2.8. Precision, accuracy, recovery and limit of detection

To validate the HPLC assay, data on precision, linearity, recovery, limit of quantification and interferences were raised in

accordance with the guidelines of the Clinical and Laboratory Standards Institute, formerly National Committee for Clinical Laboratory Standards (NCCLS, check out http://www.nccls.org for more information), for preliminary evaluation of quantitative clinical laboratory methods as follows.

Precision was expressed as coefficient of variation (CV) of plasma samples that had been spiked with low, middle and high concentrations of each drug and analyzed four times/day on 20 different days. The sequence of sample analysis was as follows: drug free plasma – calibration sample (middle) – control sample (low) – control sample (high) – low – middle – high – high – middle – low – patient plasma – patient plasma. After a 60 min interrupt, the entire sequence was repeated. Each sequence with repetition was conducted twice on each day of the validation. Linearity was determined for every drug using five different concentrations obtained by mixing a low and a high concentration in a ratio of 100/0, 25/75, 50/50, 75/25 and 0/100. The five concentrations were measured four times together with one calibration sample. Absolute recovery was determined by comparing peak-heights of plasma samples after column switching with samples of the same amount diluted in eluate (8%, v/v acetonitrile) and injected directly onto the analytical column. Analytical recovery was determined by comparing calculated and supplemented amounts of the analytes. Each sequence was conducted twice. For determination of the limit of quantification a mixture of the five compounds was analyzed 10 times at a concentration range of about 50% of the lowest common therapeutic concentration.

To control the assay for interferences with endogenous compounds samples were spiked with triglyceride, hemoglobin and bilirubin. Blank serum used to prepare calibration and control samples contained 70 mg/dl of triglyceride. Contaminations with triglycerides were imitated by adding an intralipid fat emulsion at five different concentrations of triglyceride to the blank serum supplemented with low, middle and high concentrations of the four drugs. Each spiked sample ($n = 15$) was measured twice. To control the applicability of the assay to hemolytic serum, sera containing 1 or 5% hemoglobin were supplemented with drugs and analyzed twice. Bilirubin was added at five different concentrations to serum samples containing low or high concentrations of clozapine, norclozapine, perazine, olanzapine and quetiapine and also measured in duplicate.

3. Results and discussion

The HPLC method enabled the analysis of serum samples containing quetiapine and in addition of clozapine, norclozapine, perazine and olanzapine within less than 25 min on a Hypersil C18 ODS analytical column. The on-line clean-up procedure efficiently removed matrix constituents, and the five drugs to be quantitated and fluperlapine, which had been added as internal standard, were well separated within 25 min (Fig. 1A–D).

Data on intra- and inter-assay precision are summarized in Table 1. Coefficients of variance were calculated by analyses of plasma samples that had been supplemented with low, middle and high concentrations of the drugs, i.e. 20, 80 and 170 ng/ml for quetiapine 200, 400 and 600 ng/ml for clozapine, 125, 250 and 375 ng/ml for norclozapine, 80, 160 and 230 ng/ml for perazine and 20, 40 and 80 ng/ml for olanzapine. Overall day-to-day imprecision ranged between 0.77 and 7.64%.

Table 1
Coefficients of variance (CV, %) for the determination of clozapine and norclozapine, perazine, olanzapine and quetiapine by the described HPLC method

Coefficients of variance (%)			
Quetiapine (ng/ml)	20	80	170
Within-run precision	3.55	2.46	4.67
Total precision	5.69	3.45	5.61
Between-day precision	3.52	1.19	1.07
Between-run precision	2.72	2.1	3.28
Clozapine (ng/ml)	200	400	600
Within-run precision	2.46	3.04	3.13
Total precision	4.19	4.25	4.52
Between-day precision	1.95	2.66	2.59
Between-run precision	2.77	1.33	1.98
Norclozapine (ng/ml)	125	250	375
Within-run precision	2.33	3.17	3.18
Total precision	3.76	4.28	4.24
Between-day precision	1.17	2.1	1.68
Between-run precision	2.71	1.97	2.26
Olanzapine (ng/ml)	20	40	80
Within-run precision	2.87	3.16	3.14
Total precision	6.98	4.94	7.64
Between-day precision	5.6	2.06	5.52
Between-run precision	3.04	3.18	4.23
Perazine (ng/ml)	80	160	230
Within-run precision	3.85	2.6	4.62
Total precision	5.14	3.89	5.73
Between-day precision	2.73	2.08	0.77
Between-run precision	2.02	1.99	3.3

Assay characteristics were obtained by the analysis of quality control plasma samples spiked with low, medium or high concentrations of the drugs. Analyses were conducted four times (two replicates) on 20 different days.

Linearity of the assay was tested using five concentrations of each drug 20–370 ng/ml of quetiapine, 105–1260 ng/ml of clozapine, 100–1210 ng/ml of norclozapine, 50–460 ng/ml of perazine and 10–170 ng/ml of olanzapine. Linear regression analysis revealed linearity over the whole range for all analytes with correlation coefficients (R^2) close to 1. At concentration zero the calculated regression line was close to zero (Table 2).

The recovery was determined for different concentrations of each drug: quetiapine 20, 45, 185 and 370 ng/ml, clozapine 105, 370, 630 and 1260 ng/ml, norclozapine 100, 350, 605 and 1210 ng/ml, perazine 50, 100, 230 and 460 ng/ml and olanzapine 10, 20, 85 and 170 ng/ml. The absolute recoveries were calcu-

Table 2
Correlation coefficients (R^2) and linear functions for the determination of quetiapine, clozapine and norclozapine, perazine and olanzapine by the described HPLC method

	R^2	$y = ax + b$
Quetiapine	≥0.9816	$y = 0.0949x - 0.1014$
Clozapine	≥0.978	$y = 0.4314x - 0.3847$
Norclozapine	≥0.9793	$y = 0.4655x - 0.4899$
Olanzapine	≥0.9807	$y = 0.1153x - 0.1745$
Perazine	≥0.9765	$y = 0.0337x - 0.0248$

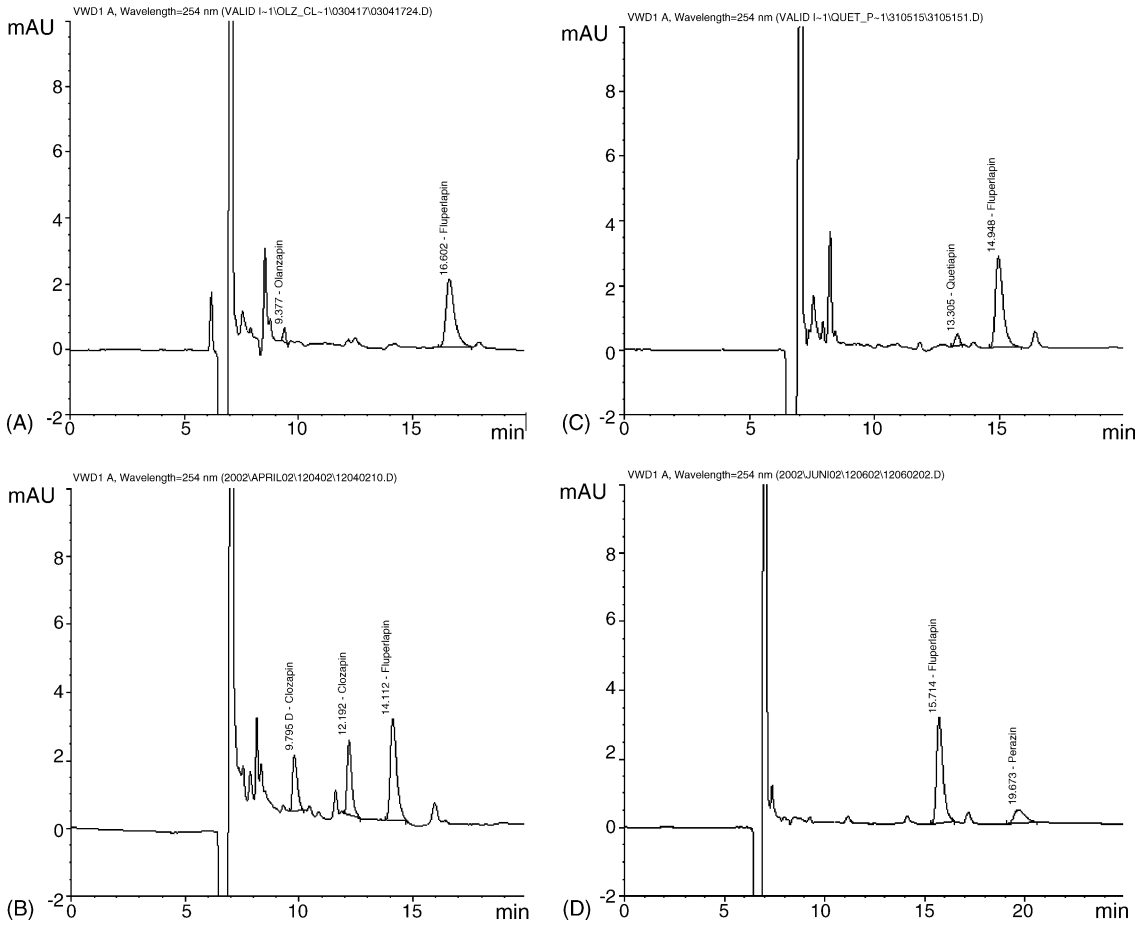

Fig. 1. Representative chromatograms obtained from the analysis of authentic human plasma of four representative patients who had ingested daily doses of (A) 20 mg olanzapine, (B) 250 mg clozapine, and (C) 175 mg quetiapine or (D) 200 mg perazine. Resulting drug concentrations found by the reported HPLC method were 21 ng/ml for olanzapine, 126 and 132 ng/ml for clozapine and norclozapine (D-clozapin), respectively, 42 ng/ml for quetiapine and 59 ng/ml for perazine.

lated from the ratio of peak heights resulting from direct injection of drug solutions on the analytical column and from the analysis of plasma samples by the described HPLC-column switching assay. They were similar for the different drug concentrations ranging from and 90 to 138% (mean ± S.D., 118 ± 17%) for quetiapine, 85 to 122% (mean ± S.D., 103 ± 16%) for clozapine, 95 to 126% (mean ± S.D., 111 ± 13%) for norclozapine and 76 to 133% (mean ± S.D.: 104 ± 18%) for perazine, 83 to 138% (mean ± S.D., 103 ± 17%) for olanzapine. The analytical recovery was calculated by comparing the amounts determined by the HPLC assay and the nominal values. Mean ± S.D. analytical recovery rates were 110 ± 14% (range 87–138%) for quetiapine, 98 ± 3% (range 95–105%) for clozapine, 99 ± 5% (range 93–105%) for norclozapine, 112 ± 6% (range 103–123%) for perazine and 103 ± 6% (range 91–111%) for olanzapine.

Analysis of samples containing drug concentrations suggested to be in the range of the lowest therapeutically expected concentration revealed CVs of 3.5% for 5 ng/ml of olanzapine, 1.4% for 10 ng/ml of quetiapine, 3.9% for 25 ng/ml of perazine, 2.4% for 50 ng/ml norclozapine and 2.0% for 50 ng/ml of clozapine. Since the CV was always below 5%, even lower concentrations may be analyzed.

Drugs could be stored in the dark at −20 °C for several months without measurable decomposition. A measurable decomposition of olanzapine over 7 days at room temperature was found (−54%), concentrations of an olanzapine plasma standard measured immediately after preparation and three months after storage at −20 °C differed by only 2% [16].

To control interferences with triglyceride, serum samples containing 20, 170 or 230 ng/ml of quetiapine, 100, 600 or 1200 ng/ml of clozapine, 100, 600 or 1200 ng/ml of norclozapine, 50, 230 or 460 ng/ml of perazine and 20, 80 or 160 ng/ml of olanzapine were supplemented with a triglyceride emulsion to a final concentrations of 1570, 3070, 4570 or 6070 mg/dl triglyceride. High triglyceride concentrations (≥1570 mg/dl) significantly reduced the resulting drug concentrations of perazine, clozapine and norclozapine whereas minor effects were observed for quetiapine and olanzapine (Fig. 2). In the presence of ≥1570 mg/dl of triglyceride concentrations of perazine (109 ng/ml) amounted to 45% of non-supplemented serum samples (mean ± S.D., 61 ± 11%). Those of clozapine (369 ng/ml) and norclozapine (530 ng/ml) decreased to 70 ± 10 and 85 ± 8%, respectively. No effect was seen for quetiapine (157 ng/ml) and for olanzapine (95 ng/ml) with a reduction to 87 ± 7% and 99 ± 9%, respectively.

To control effects of hemolysis on the assay, hemolytic serum containing 1 or 5% hemoglobin were prepared and analyzed. In the presence of 1% hemoglobin the resulting serum

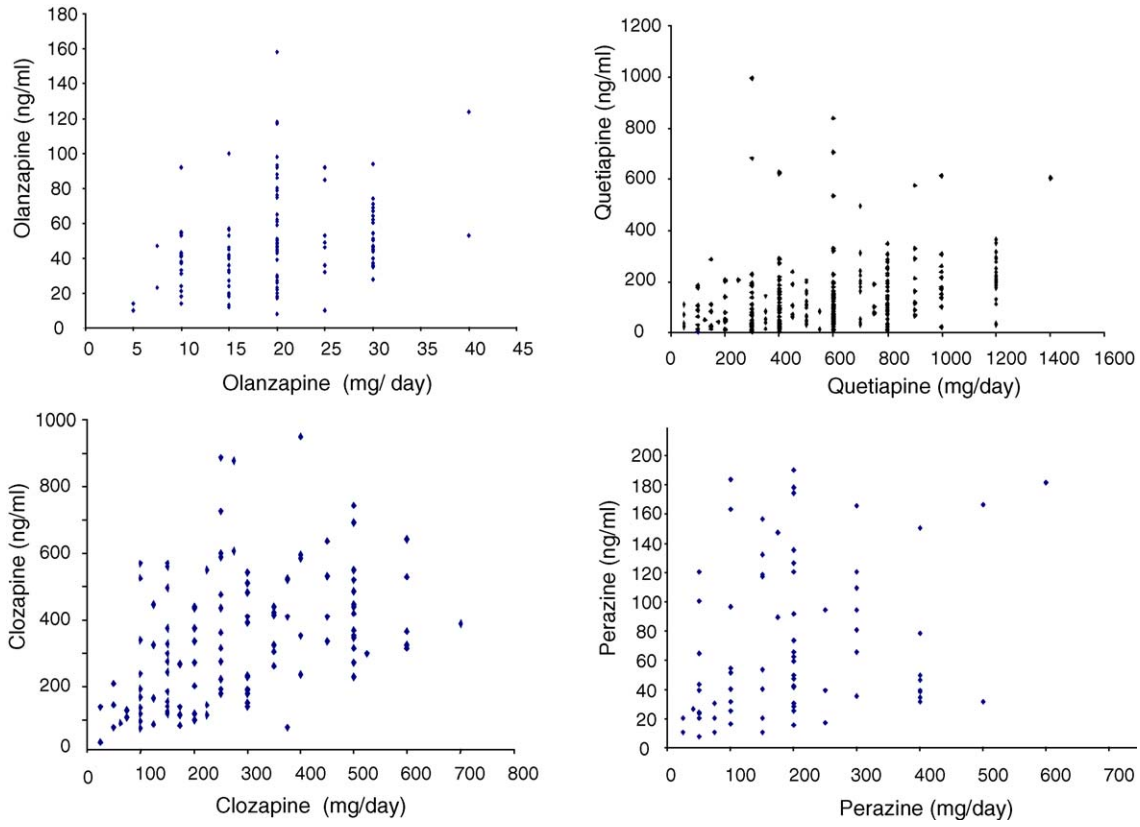

Fig. 2. Dose dependent concentrations of olanzapine ($n = 120$), quetiapine ($n = 295$), clozapine ($n = 120$), and perazine ($n = 75$) in blood plasma of schizophrenic patients. Values given are those of individual patients treated with stable daily doses of the antipsychotic drugs for at least 7 days.

concentrations were almost identical to non-hemolytic serum (97–100%) and attained 84–102% of controls in the presence of 5% hemoglobin.

Bilirubin was added to obtain final concentrations of 5, 10, 15 and 20 mg/dl of serum samples containing low or high concentration of clozapine (105 and 1260 ng/ml), norclozapine (100 and 1210 ng/ml), perazine (50 and 460 ng/ml), olanzapine (10 and 170 ng/ml) and quetiapine (20 and 370 ng/ml). In comparison to non-supplemented serum the concentrations of the drugs amounted to (mean ± S.D.) 101 ± 3%, 102 ± 5%, 100 ± 3%, and 101 ± 4% of control samples in the bilirubin supplemented samples.

To control for possible interferences with drugs that may be used in combination with quetiapine, clozapine, perazine or olanzapine the suggested interfering compounds were prepared in blank serum as described for QC- and C-samples. Testing standard solutions containing other psychotropic drugs that may be applied in combination with clozapine, perazine, olanzapine and quetiapine, interferences could be observed (Table 3).

Under the conditions used here, ghost peaks were not observed. This may be different when using other instruments.

In samples of schizophrenic patients who had taken fixed doses of quetiapine, clozapine, olanzapine or perazine for at least 7 days, drug concentrations were highly variable between individual patients (Fig. 2). Resulting ranges and standard deviations of serum concentrations were markedly higher than those of the dose ranges (Table 4).

The aim of this study was to establish a method that is suitable for determination of quetiapine and also of other antipsychotic drugs within a single TDM setting. The developed method is based on a method published by Weigmann et al. [14,17]. It is necessary to use C8 instead of CN material for sample clean up. Using C8 material it was able to run 150 instead of 30 samples. For sample clean up an eluent consisting of 8% acetonitrile was found to be optimal. This eluent was more stable than the eluent used before.

The modified HPLC assay enabled the determination of quetiapine, clozapine, norclozapine, olanzapine and perazine in serum. Column-switching techniques are useful for automated and rapid analysis of drugs in complex matrices [13,15,18–21]. In addition to a conventional isocratic HPLC system, a pre-column for sample clean up, a second HPLC pump and a six-port switching valve are needed. The method described here was found to be suitable for the TDM of at least four antipsychotic drugs. It enabled processing of more than 150 serum samples per clean-up column before column replacement was necessary. At low, middle and high drug concentrations the imprecision was always below 10%. An inter-assay imprecision up to 15% is considered acceptable for therapeutic drug monitoring of psychotropic drugs [22] and of antiepileptic drugs [23].

The endogenous compounds tested for interference were triglycerides, bilirubin and hemoglobin. With the exception of triglycerides, the presence of high concentrations of hemoglobin or bilirubin did not interfere with the assay. It therefore seemed

Table 3
Drugs tested for interference and retention times

Substance	Retention time (min)
Norperazine	n.d.
O-Desmethylenlafaxine	n.d.
Norsertraline	n.d.
Pimozide	n.d.
Sertindole	n.d.
Sulpiride	n.d.
Tianeptine	n.d.
Amisulpride	7.36
Tacrine	7.76
9-Hydroxyrisperidone	8.01
Pantoprazole	8.16
Norvenlafaxine	8.24
Venlafaxine	8.64
Risperidone	8.74
Carbamazepine-10,11-epoxide	8.84
Olanzapine	9.16
Dipiperone	9.25
Melperone	9.37
Donepezil	10.33
Norcitalopram	10.38
Norclozapine	10.40
Oxazepam	10.59
Pipamperone	10.71
Red. Haloperidol	10.81
Carbamazepine	10.92
Citalopram	10.99
Reboxetine	11.15
Spiperone	11.42
Nordoxepin	11.61
Lorazepam	12.94
Doxepin	12.36
Clozapine	13.15
Sertraline	13.34
Haloperidol	13.44
Temazepam	13.78
Quetiapine	13.86
Paroxetine	14.24
Fluperlapine (ISTD)	15.57
Desipramine	15.65
Domperidone	16.13
Nordiazepam	16.30
Protriptyline	16.40
Imipramine	16.75
Fluvoxamine	17.09
Normaprotiline	17.27
Norfluoxetine	17.83
Nortriptyline	18.18
Maprotiline	18.45
Nortrimipramine	18.48
Perazine	19.57
Amitriptyline	19.75
Ziprasidone	19.87
Fluoxetine	20.09
Trimipramine	21.09
Norclomipramine	23.26
Perphenazine	23.30
Diazepam	23.06
Clomipramine	26.15
Zotepine	33.01

n.d: no detectable peak from 0 to 25 min.

likely that hemolytic or hyperbilirubinic serum can be used for drug analysis. In presence of high amounts of triglycerides (≥1570 mg/dl), however, the concentrations of perazine and clozapine were lower than expected. The physiological range of triglycerides is in females (age 30–60) 44–185 mg/dl and in men (age 30–60) 40–250 mg/dl. A limit of 528 mg/dl is alarming. When patient sample thus amount to 1570 mg/dl of triglycerides or even higher, there is a problem to quantify the blood levels of clozapine or perazine.

Interferences with other drugs that might be used in combination with the antipsychotic drugs were minimal. There was only oxazepam (RT 10.6 min) that interfered with the detection of norclozapine (RT 10.4 min) and temazepam (RT 13.8 min) interfering with quetiapine (RT 13.9 min). These interferences should be taken into account when these drugs are used as comedication, though the combination of clozapine and benzodiazepines should be used cautiously, because severe adverse reactions have been reported for the combination [24].

The method described here analyzed plasma or serum samples automatically. It allows measurement of single samples. A sample arriving in the morning can thus be processed the same day. The described method thus exhibits both, sufficient accuracy and sufficient speed to report valid results within an appropriate time schedule. It has been proven to be suitable for a therapeutic drug monitoring service. In comparison with other methods reported in the literature and established for determination of quetiapine or other antipsychotic drugs [9,10,14,15,17–20] the described method seemed advantageous. Previously published own methods [13,14,17–20] could not be used for quetiapine, since the cyanopropyl-bonded clean-up material did not retain the drug. C8-material was suitable which was consistent with the findings reported by others [10]. Another column-switching method described for perospirone [25], a new experimental antipsychotic drug, used fluorescence detection which is rather indifferent to interferences. Quetiapine and the other drugs, however, could not be measured by fluorescence detection. Hasselstrøm and Linnet [10] used HPLC coupled with on-line solid-phase extraction on C2 material for quantification of quetiapine with sufficient precision and accuracy. Their clean-up procedure, however, was more complex than ours. It consisted of deproteinization of serum, cartridge conditioning and three washing steps before drug elution and HPLC analysis. A method that enables the quantification of a wide range of drugs is liquid chromatography (LC) coupled with mass spectrometry (MS) as reported for example by Zhou et al. [9]. LC methods coupled with MS are also optimal regarding sensitivity and specificity. HPLC with column switching and UV detection, however, is less expensive. The sensitivity and specificity of the described HPLC method was found to be sufficiently sensitive and selective. It could be applied in clinical practice. Using the described method, highly variable serum concentrations were found in the serum of psychiatric patients though the same dose was applied (Fig. 2). Mean levels (Table 4) were in the range of concentrations reported by others at therapeutic doses of the drugs [6,26,27].

In conclusion, HPLC with on-line column switching as described here is useful for therapeutic drug monitoring and also

Table 4
Daily doses of antipsychotic drugs and resulting concentrations of schizophrenic patients who had been treated for at least 7 days

	Quetiapine 295[a]	Clozapine 120[a]	Norclozapine	Olanzapine 120[a]	Perazine 75[a]
Daily dose					
Mean ± S.D. (mg/day)	595 ± 306	278 ± 156		20 ± 8	196 ± 127
Median	600	250		20	200
25–75th percentile	400–800	150–375		15–25	100–250
Range	50–1400	25–700		5–40	25–600
Plasma concentration					
Mean ± S.D. (ng/ml)	139 ± 136	328 ± 195	219 ± 126	48 ± 27	71 ± 52
Median	102	315	204	45	50
25–75th percentile	49–187	169–438	119–285	30–59	30–111
Range	3–996	25–945	27–581	8–158	7–190

Range of plasma concentrations indicate minimal and maximal values found in samples containing measurable amounts of the drugs.
[a] Number of patients.

for pharmacokinetic studies of quetiapine, clozapine, perazine and olanzapine. The method was found to be suitable in clinical practice and may be extended in the future to other psychoactive drugs like ziprasidone.

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