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Research Article

# A new Turkish pharmacokinetics software program for therapeutic drug monitoring of theophylline

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**Abstract**

In this study, it was aimed to develop a Turkish software with pharmacokinetic (PK) data for therapeutic drug monitoring and IV dose adjustment of narrow therapeutic index theophylline.

The study involved three groups, each comprising two rabbits. The dose required for the target concentration (15µg/mL) was calculated with the developed program according to the weight of the rabbits in the first group. Blood samples taken at certain times were analyzed by validated HPLC method to calculate the elimination rate constant (ke) after IV bolus administration. The $r^2$ values for ke were found to be 0.86 and 0.95. The second dose calculated according to revised PK findings was administered and blood samples were taken. When the analyzed results and theoretical results were compared, the deviation was found to be 5.53% and 8.795%. The findings were taken as the population PK for other applications.

IV multiple dose bolus and IV fast-slow combined infusion were administered to the second group and the third group, respectively. The results obtained from the analysis of blood samples taken at the times determined according to the application were compared with the theoretical results.

As a result, although there is a high difference between theory and practice at low concentrations, there is very little variation at high concentrations. By using this program, it has been achieved to keep theophylline at the desired level without reaching the minimum toxic concentration and without falling below the minimum effective concentration. It is thought that deviations will be reduced with larger samples.

**Keywords:** TDM, theophylline, software pharmacokinetic programme

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## Teofilinin terapötik ilaç izlemi için yeni bir Türkçe farmakokinetik yazılım programı

**Öz**

Terapötik ilaç izlemi için farmakokinetik (FK) veriler ile Türkçe bir yazılım geliştirilmesi hedeflendi ve dar terapötik indeksli teofilinin iv doz ayarlaması yapılması amaçlandı.

Her grupta 2 tavşan olacak şekilde 3 grup oluşturuldu. İlk gruptaki tavşanlara kilosuna göre hedef derişimi 15µg/mL olması için gereken doz geliştirilen programa hesaplatılarak uygulandı ve eliminasyon hız sabitini (ke) hesap edecek şekilde belli zamanlarda alınan kan örnekleri valide edilen HPLC metodu ile analizlendi. ke için çizilen eğimin $r^2$ değerleri 0.86 ve 0.95 bulundu. Revize edilen FK bulguları ile gereken bireysel doz hesaplanarak 2.doz uygulaması yapılıp kan örnekleri alındı. Analizlenen sonuçlar ile teorik sonuçlar kıyaslandığında %sapma %5.53 ve %8.795 bulundu. 1. gruptan elde edilen FK bulguları, popülasyon FK'sı olarak alınarak diğer uygulamalarda kullanıldı.

Ikinci gruba iv çok doz bolus, üçüncü gruba ise iv hızlı-yavaş kombine infüzyon uygulaması yapıldı. Uygulamaya göre belirlenen zamanlarda alınan kan örneklerinin analizinden elde edilen sonuçlar teorik sonuçlarla kıyaslandı.

Sonuç olarak düşük derişimlerde teorik ile pratik arasında yüksek farklılık görülmek ile birlikte yüksek derişimlerde farklılanma çok azdır. Bu programı kullanarak teofilinin minimum toksik derişimine erişmeden ve minimum etkin derişiminin altına düşmeden istenilen düzeyde tutulması başarılmıştır. Daha büyük örneklemler ile sapmaların azaltılacağı düşünülmektedir.

**Anahtar Kelimeler:** TDM, teofilin, farmakokinetik yazılım programı

## 1 Introduction

Due to the individual variability of some drugs, not every patient can be treated with the standard drug method. Individual dose application is difficult for drugs whose clinical efficacy is unknown or that do not respond clinically until they have an irreversible toxic effect on the patient. In such cases, the therapy can be maintenaned through therapeutic drug monitoring (TDM). TDM can be defined as individual therapy to each patient for specific drugs (Freedman & Marshall, 1993a; Kang & Lee, 2009; Marks, 1985; Touw, Neef, Thomson, & Vinks, 2005). That's to adjust the dose needed to the patient and treated the disease. There is no certain solution for dose adjustment, but TDM especially utilizes if the relationship between plasma level and response can be computed mathematically (Touw et al., 2005). The aim of this strategy is to stay within the therapeutic index (TI, therapeutic window, or therapeutic range) and to continue the steady state concentration. TI encompasses the interval between a minimum level that causes the therapeutic effect (minimum effective concentration, MEC) and a minimum level that causes the toxicity (minimum toxic concentration, MTC) of a drug (Birkett, 1997; Marks, 1985; Whiting et al., 1984). This is especially important for drugs that have a narrow TI (NTI), such as theophylline. Theophylline has a dose-response relationship. It is used in asthma and chronic obstructive disease as a bronchodilator. Its NTI for adults is 10–20 µg/mL or 5–15 µg/mL depending on the source and changes according to the age of the patient (Hallworth & Watson, 2017; Hardman, Limbird, Gilman, Molinoff, & Ruddon, 1996; Pesce, Rashkin, & Kotagal, 1998). It is metabolized in the liver mostly by especially CYP1A2 and

CYP3A4. The clearance shows an alteration due to a variation in the rate of metabolism (Freedman & Marshall, 1993b; Hardman et al., 1996). The half-life is approximately 9 hours. However, it decreases in cigarette smokers because it induces the hepatic metabolism (Freedman & Marshall, 1993b; Jenne, Nagasawa, McHugh, Macdonald, & Wyse, 1975). The other pharmacokinetic (PK) parameters of theophylline are shown in Table 1 (Hardman et al., 1996).

**Table 1.** The pharmacokinetic parameters of theophylline

| **BA (oral) %** | **Excretion by urine %** | **Plasma protein binding %** | **Cl (mL/min/kg)** | **Vd (L/kg)** | **$t_{1/2}$ (hour)** | **TI (μg/mL)** | **MTC (μg/mL)** |
|---|---|---|---|---|---|---|---|
| 96±8 | 18±3 | 56±4 | 0.65±0.20 | 0.50±0.16 | 9.0±2.1 | 5-15 | 20 |

BA: Bioavailability, Cl: clearance, Vd: Volume of distribution, $t_{1/2}$: half-life of drug, TI: Therapeutic window, MTC: minimum toxic concentration; (±SD)

An important point for TDM is when and how much sample is collected from the patient. Many samples are required to create a PK profile. However, the patient's comfort requires medication monitoring with at least a few samples (Ghiculescu, 2008).

If area under the curve (AUC) and dose are correlated with each other, to determine the AUC, at least three blood samples are necessary. Taking more blood samples from patients is practically impossible.

Also, if there is a correlation between Ctrough (trough serum concentration is the pre-dose concentration) or Ct (the concentration of a determined time) and dose, a single sampling strategy is preferable and applicable for drug monitoring. If there is no relationship between Ctrough/Ct and dose, there is no other way except to determine AUC (Touw et al., 2005). Especially for drugs with NTI, peak concentration (Cmax) is an important parameter.

The analysis method is the most important step. It has to be safe and easy. To be cheap, fast, and available in laboratory environments is preferable in terms of application in practice. A kit may be preferable for this area in terms of ease of use, but there isn't any kit for theophylline.

Chromatographic methods are the best option because the simple sample preparation and small volume requirements make them especially suitable for monitoring theophylline therapy in a pediatric population (Freedman & Marshall, 1993c; Hendeles, Weinberger, & Johnson, 1978). In addition to chromatographic methods for the theophylline assay, electrochemical techniques with a faster response perspective can also be evaluated(Koçak, Nas, Kantekin, & Dursun, 2018; Wagnew et al., 2022). However, attention should be paid to the applicability of this method in the hospital. The analysis method must be validated according to guidelines set by the authorities. (Fda & Cder, 2018).

With a user-friendly program, monitoring as many drugs as possible is possible. For this reason, we aimed to develop a new software program for adjusting and individualizing the dose of

theophylline. *In vivo* studies on the rabbits were conducted under multiple bolus dose and IV infusion administration conditions to test the validity of the software program.

Currently, there are documents in the form of SOPs or online/setup programs that indicate what needs to be done for TDM, but there is no proven program with Turkish software (Fuchs, Csajka, Thoma, Buclin, & Widmer, 2013). In this way, theophylline will be a model substance for other IV drugs that will need to be followed. For this purpose, firstly, a Turkish program including PK algorithms was developed. Then, theophylline was analyzed by HPLC after the administration of an IV bolus, an IV multiple dose bolus, and an IV infusion to rabbits. Individual dose calculations were performed from the program developed using theophylline results obtained from the analysis. The accuracy and validity of the program were proved by comparing the theoretical results with plasma samples after the application of the calculated doses to rabbits.

## 2 Materials and Methods

### 2.1 Standards and Reagents

Neofleks 5% Dextrose Solution (Türktıpsan A.Ş., Turkey) and Teobag-200 (Eczacıbaşı-Baxter, Turkey) were purchased from the Drug Company. Teophylline (Cipla, India) and caffein (Cipla, India) were used for the analytical method as standards and internal standards, respectively. Heparin Sodium (Panpharma S.A., Turkey) was used as an anticoagulant agent in the cannula. All other chemicals and reagents were of analytical grade.

### 2.2 Execution of Software Program

The programming language C# was used as the host language for the software program in the Visiual Studio environment. For the data visualization, Component One Studio was used to make it convenient to prepare tables, and graphic procedures, and input media.

Firstly, the algorithms for PK calculations and the interface of the software program were prepared. Three screens, including patient, active substance, and administration route, were designed.

Patient screening involves the following parameters: that are name, surname, personal ID number, height, weight, sex, age, and blood group of the patient. For adjusting the dosage of theophylline, weight was used as a covariate.

Active substance screening involves the theoretical values of PK parameters obtained from references and pharmaceutical IV preparations on the market for the active substance. Beside the constant data, there is a numerical box to input the desired plasma concentration.

The administration route screen has five sub-screens: individual dose, IV bolus, IV multiple dose bolus, IV infusion, and IV combined with IV bolus and infusion. At first, a program user has to calculate the dose from the desired plasma concentration at the individual dose screen.

A patient, an active substance and its pharmaceutical form, and an administration route of drug are chosen, respectively, from the program. After the data of patient weight inputs into the patient screen, the desired plasma concentration to reach the target therapeutic level is written at the active substance screen. When the “Calculate the dose” button is pressed at the individual dose screen, the program gives the calculated dose on same screen in the related place. Following the drug administration to patients, three blood samples are taken at different times, and the drug concentrations in the blood samples analyzed by HPLC are inputted into interested text boxes at "calculating of individual dose screen" for estimating the elimination rate constant (ke), volume of distribution (Vd), and clearance (Cl) (Dhillon & Gill, 2006). While ke is calculated, it takes care of the coefficient of determination ($r^2$). Afterwards, the maintenance dose is calculated to reach the steady-state concentration.

### 2.3 *In vivo* Animal Study Design

Six white male rabbits weighing between 1.5 and 2 kg were used for this study. The drug was given at a calculated dose according to its weight by IV administration to the rabbit marginal vein of the left ear. The blood samples were taken from the rabbit marginal vein of the right ear by the 24G branule, and plasma was separated from whole blood by a vacutainer containing EDTA. After each blood sample was taken, 0.5 mL of the 1% heparin solution was injected into the branule. The investigations were performed after approval by the Animal Ethics Committee at the Faculty of Medicine of Ege University (No. 2007-37).

The study was designed as three groups with two rabbits in each group. The first group was used to find mean of their ke, Vd, and Cl parameters by administration of one dose bolus, and these results was considered the population PK.

For finding the population PK, the first bolus dose was calculated taking into account the weight of the rabbits and was administered to both rabbits in the first group. After injection, three blood samples taken at certain intervals from the rabbits were analyzed. The analyzing results were put in a concerned place in the program, and the second dose was calculated for each rabbit separately. After second dose administration, a last blood sample for measuring Ctrough was taken and analyzed to compare this analysis result with the corresponding to theoretical value for each rabbit.

The first doses for the second group used for examination of multiple dose bolus administration were calculated according to population PK, and the results from plasma samples were compared with theoretical values. The continuous infusion was applied to the third group. For the first infusion rate of the third group, the population PK was assumed to be their PK parameters.

As an expression of the closeness to the therapeutic target, deviation from the theoretical value of the practical value obtained was used. The practical values were compared with theoretical values on a deviation% from the theoretical to practical value by using Equation 1.

$$\text{Deviation\%} = \frac{\text{PC} - \text{TC}}{\text{TC}} \times 100$$

Equation 1- Calculation of deviation% from theoretical value to practical value. PC: The Practical Concentration Value Obtained; TC: The Theoretical Concentration Value calculated by the program.

### 2.4 The Strategy of Multiple Bolus Doses Administration

The bolus doses to be given to rabbits in the second group were separately calculated from the program using their weights. After being injected, three blood samples were taken at different hours and analyzed. The maintenance dose, Ctrough, and dose interval were estimated by the program for each rabbit, and the calculated maintenance dose was administrated. A last blood sample was taken to compare practical and theoretical values.

### 2.5 The Strategy of Continuous Infusion Administration

The infusion flow rate was determined by inputting the preferred time to reach the target concentration. After starting the infusion, three blood samples taken at different hours were analyzed. Then, when the preferred time was over, second infusion rates were adjusted to maintain steady-state concentrations. Before and after the second infusion rate administration, three blood samples were taken, and this study was completed by stopping the administration of drugs.

### 2.6 The Validation of Analytical Method of Theophylline

The analytical method was modified from Karasulu E. et al. (Karasulu, Apaydin, & Tuglular, 2006). An Agilent 1100 series HPLC was used at isocratic conditions. It was equipped with an injection valve of 20 µL, a UV detector set at 273 nm, and a $C_{18}$, 5 µm x 4.6 mm x 150 mm analytical column. The mobile phase composition was a mixture of acetonitrile, tetrahydrofuran, acetic acid, and distilled water (20: 20: 5: 955) (v/v/v/v) at a flow rate of 1.0 ml/min. The overall analysis time was 10 minutes.

The method validation was performed using the following parameters: selectivity, accuracy, precision and recovery, sensitivity, reproducibility, and stability of standard and test solution (Hendeles et al., 1978). The linearity of the method involved six points: 1, 4, 8, 12, 16, and 20 µg/mL.

Each sample (200 µL) was mixed with caffeine solution as an internal standard and shaken by vortex for 10 sec. 600 µL of methanol was added to it and shaken by vortex for 10 sec. After centrifuging at 10,000 rpm at 4 °C for 15 min, the supernatant was removed under the vacuum concentrator with nitrogen. The remainder was solved with 0.5 µL of mobile phase and injected into HPLC.

## 3 Results and Discussion

### 3.1 Execution of Software Program

The program has two parts: the source code, including the algorithms of PK, and the execution area for users. The source codes for the PK module of software were prepared in C#. The image of administration route screens is presented in Figure 1.

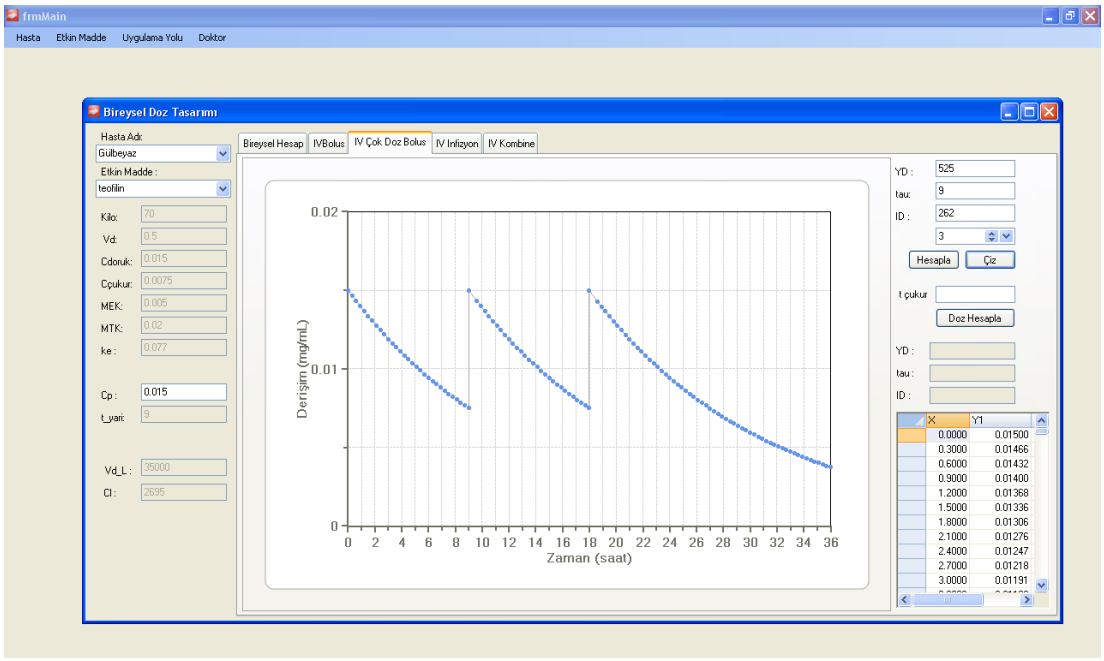


**Figure 1-** The administration route screen

### 3.2 The Strategy of Multiple Bolus Dose Administration

Three blood samples were taken after the first group received a bolus IV dose of the dose that was calculated from the source data. Vd and ke changed from 0.77 h-1 to 0.188 h-1 and 500 mL to 1242 mL, respectively (Table 2); $r^2$ was 0.86 and 0.95 for each rabbit. A blood sample was also taken immediately before the second dose. The deviation of the result from the theoretical result was -89.799% and 18.385% for each rabbit. The second dose was determined to reach the target concentration according to these values, which vary for each rabbit separately. When the theoretical results are compared with the sample results taken at a given time after the second dose, the deviation value was found to be 5.53% and 8.795%, respectively.

**Table 2-** Pharmacokinetic parameters of the rabbits at multiple bolus dose administration

| | Accepted values in common** | 1st rabbit | 2nd rabbit | The parameter of Population PK | 3th rabbit | 4th rabbit |
|---|---|---|---|---|---|---|
| **1. dose** | | 14 mg / 7 mL | 20 mg / 10 mL | - | 20 mg / 10 mL | 20 mg / 10 mL |
| **ke ($h^{-1}$)** | 0.077 | 0.252 | 0.124 | 0.188 | - | - |
| **Vd (mL)** | 500 | 1439 | 1045 | 1242 | - | - |
| **2. dose* theoretically** | | 20 mg | 11.652 mg | - | - | - |
| **2. dose practically** | | 20 mg / 10 mL | 11 mg / 5.5 mL | - | 20 mg / 10 mL | 20 mg / 10 mL |

* The second dose was given at 400 min and 340 min for the first and second rabbits, respectively.

** According to Hardman et al.(Hardman et al., 1996)

The values obtained from the first group were accepted as population PK. According to these values, the IV bolus dose of the second group, the time of administration of the second dose, the Ctrough before the administration of the second dose, and the maintenance dose were calculated according to their weight. The deviations from the theoretical results of the blood concentrations of rabbits before and after the second dose were 11.546% and -8.751% for the first rabbit, and 50.114% and 4.851% for the second, respectively.

The times of dose administrations, the time of Ctrough, and the deviation% of all samples belonging to the multiple bolus doses administration are presented in Table 3. The profiles of the plasma concentration time for practical and theoretical values at multiple bolus doses are presented as graphical in Figure 2.

**Table 3-** The deviation % from the theoretical value to the practical value at multiple bolus dose administration

| **First Group** | | | | **Second Group** | | | |
|---|---|---|---|---|---|---|---|
| **1st rabbit** | | **2nd rabbit** | | **3th rabbit** | | **4th rabbit** | |
| **t (min)** | **Deviation%** | **t (min)** | **Deviation%** | **t (min)** | **Deviation%** | **t (min)** | **Deviation%** |
| **0** | First dose time | **0** | First dose time | **0** | First dose time | **0** | First dose time |
| **5** | | **3** | | **6** | -5.672 | **10** | -0.473 |
| **30** | | **38** | | **21** | -24.137 | **20** | -21.564 |
| **60** | | **53** | | **40** | -25.579 | **50** | -18.945 |
| **385 (Ctrough)** | -89.799 | **360 (Ctrough)** | 18.385 | **416 (Ctrough)** | 11.546 | **340 ($C_{trough}$)** | 50.114 |
| **400** | Second dose time | **360** | Second dose time | **416** | Second dose time | **340** | Second dose time |
| **600** | 5.530 | **365** | 8.795 | **425** | -8.751 | **350** | 4.851 |

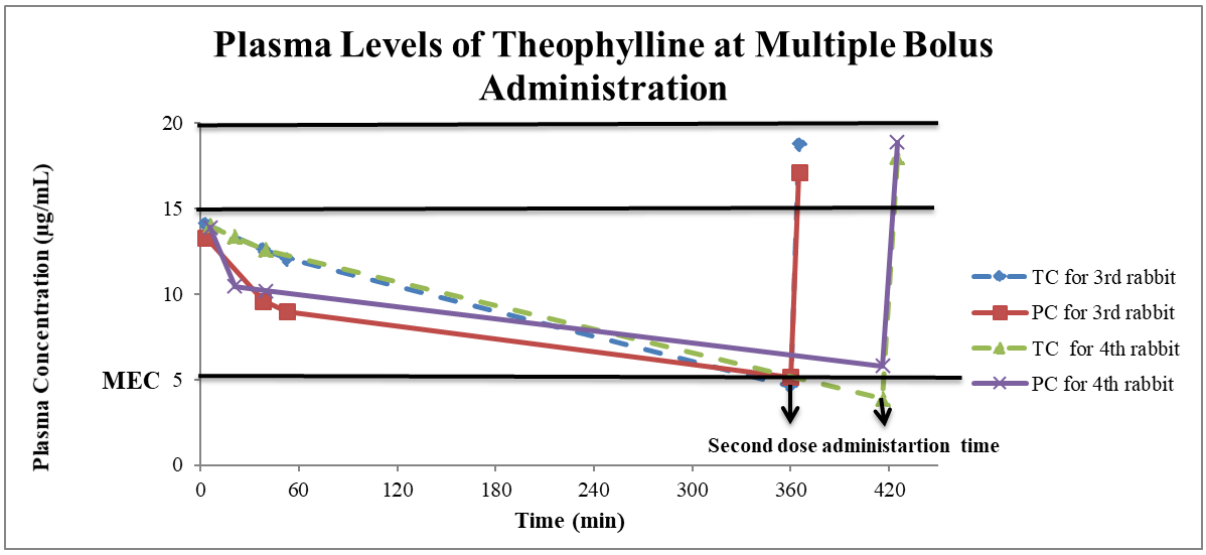


**Figure 2-** The plasma levels of theophylline at multiple bolus doses administration with practical and theoretical results of 3rd and 4th rabbits

### 3.3 The Strategy of Continuous Infusion Administration

The infusion rate required to reach the target level within 1 hour was calculated as 10 mL/hour (20 mg) based on the population PK parameters determined from the program, and three blood samples were taken after IV infusion administration. The first infusion was terminated with a blood sample taken at 60 minutes and the second infusion rate was calculated as 3.3 mL/hour (6.6 mg) from the program and administered for 30 minutes. Two blood samples were taken throughout the second infusion, and a final blood sample after the end of the second infusion was taken and analyzed. Theophylline level in the last blood sample of the first infusion was 15.718 µg/mL for the first rabbit and deviated from the theoretically calculated value by 2.331%. In the second rabbit, it was found to be 15.245 µg/mL and deviated from the theoretical calculated value by 17.269%. The deviations in the samples after the second infusion for both rabbits were between -0.846% and 18.664%.

The starting times of IV infusions, the time of $C_{target}$, and the deviation% of all samples are presented in Table 4. The deviations from the desired concentration of theophylline of results of samples taken during the second continuous infusion rate are shown in Table 5.

The profiles of the plasma concentration time of practical and theoretical values at IV infusion administration are presented as graphical in Figure 3.

**Table 4-** The Deviation % from the theoretical value to practical value at IV infusion administration

| **Third Group** | | | |
|---|---|---|---|
| **5th rabbit** | | **6th rabbit** | |
| **t (min)** | **Deviation%** | **t (min)** | **Deviation%** |
| **0** | First infusion rate time | **0** | First infusion rate time |
| **15** | 73.947 | **7** | 96.280 |
| **30** | 30.807 | **18** | 47.300 |
| **45** | 16.480 | **28** | 17.269 |
| **60** | 2.331 | **30** | Second infusion rate time |
| **60** | Second infusion rate time | **40** | -0.593 |
| **75** | 16.724 | **60** | -0.846 |
| **90** | 18.664 | **60** | The ended of infusion |
| **90** | The ended of infusion | | |

**Table 5-** The deviation from the desired concentration of theophylline in the results of samples taken during the second continuous infusion rate

| **5th rabbit** | | **6th rabbit** | |
|---|---|---|---|
| **t (min)** | **Deviation% (PC-15)/15x100*** | **t (min)** | **Deviation% (PC-15)/15x100*** |
| 60 | 4.786 | 28 | 3.633 |
| 75 | 5.386 | 40 | -14.460 |
| 90 | 11.306 | 60 | -12.860 |

* 15 value is the therapeutic target or TC

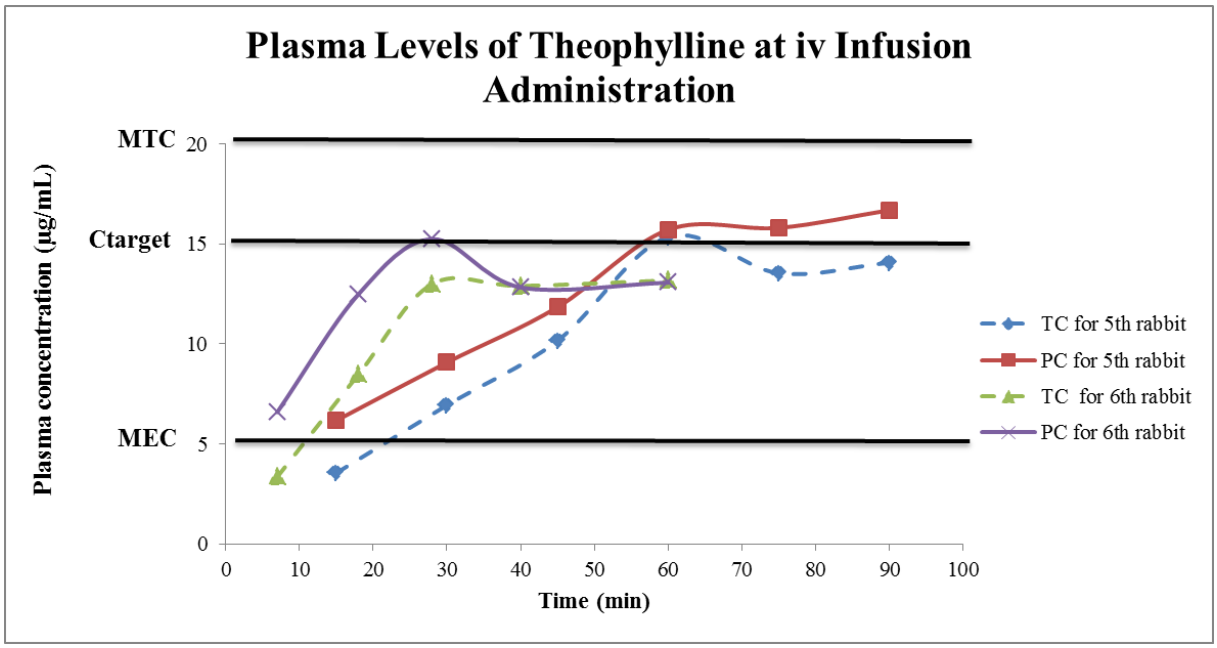


**Figure 3-** The plasma levels of theophylline at IV infusion administration with practical and theoretical results of 5th and 6th rabbits

## 3.4 The Validation of Analysis Method of Theophylline

In analytical validation, selectivity, determination of LOQ and LOD, repeatability of injection and extraction repeatability as precision criteria, solution stability, recovery studies, accuracy, and linearity studies were performed. The chromatogram with the blank plasma and matrix sample together is presented in Figure 4. The results of analytical method validation and specification limits according to CDER, FDA guidance (Fda & Cder, 2018) are shown comparatively in Table 6.

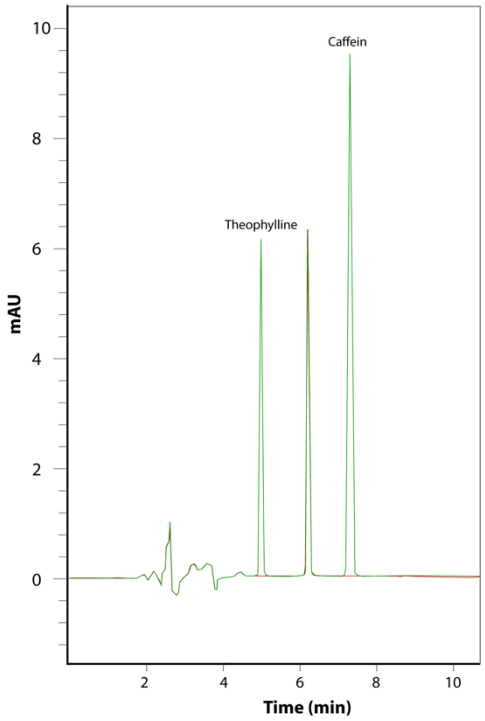


**Figure 4-** The chromatogram with the blank plasma (red line) and matrix sample (green line)

**Table 6-** The results of the theophylline's analytical validation

| **Validation Parameters** | | **Specification limits** (Hata! Başvuru kaynağı bulunamadı.) | | **Results** |
|---|---|---|---|---|
| **Selectivity** | **RT of Theophylline** | - | | 5.086 min |
| | **RT of Caffeine** | - | | 7.554 min |
| **Sensitivity** | **LOD** | - | | 0.043 µg/mL |
| | **LOQ** | - | | 0.026 µg/mL |
| **r ²** | | ≥ 0.95 | | 0.999 |
| **Calibration Curve** | | ± 20% for the LLOQ (1µg/mL);<br>± 15% for the other points (4, 8, 12, 16, 20 µg/mL) | | -13.6% - -0.1% for LLOQ<br>2.1% – 4.27 % for the other points |
| **Precision (Repeatability)** | | RSD% ≤ 2% | | 0.206% |
| **Reproducibility** | | Low concentration (1µg/mL) | RSD% ≤ 20% | 6.524% |
| | | Medium concentration (8µg/mL) | RSD% ≤ 15% | 0.895% |
| | | High concentration (20µg/mL) | | 1.095% |
| **Recovery** | | Low concentration (1µg/mL) | | 94.6% |
| | | Medium concentration (8µg/mL) | | 100.2% |
| | | High concentration (20µg/mL) | | 97.3% |
| **Stability of solutions at room temperature** | | Not more than 15% differences (during the 12 hours) | | 0.8% for lowest and highest concentrations |

Following FDA guidelines(Fda & Cder, 2018) the method validation of theophylline was checked and found to be correct within the limits of the validation parameters (Table 6). The analysis time of one sample, including the sample preparation, is approximately 30 min with this method. In terms of chromatographic analysis, the method is simple and fast.

The appropriateness of the Ctrough or AUC data calculated from the samples taken is evaluated with the correlation coefficient. In the sources where the AUC calculation was made based on at least 3 samples, correlation coefficient values varied between 0.54 and 0.96 and were accepted (Erdman, Rodvold, & Pryka, 1991; Kuypers et al., 2010). In this study, the high correlation (the coefficients of determination of the first and second rabbit samples were 0.86 and 0.95, respectively) between the analysis results of the three blood samples after the dose determined by the two rabbits in the first group shows the accuracy and reliability of the sampling time and measurement method.

In line with the results obtained with high correlation, a dose adjustment was made to the first group, and a second dose was administered. The deviations % of Ctrough and Ct after the second dose decreased from %-89.799 to %5.53 and from %18.385 to %8.795 for each rabbit, respectively (Table 2). The decrease in the deviation values, which is close to the therapeutic target, shows the applicability of the program.

When McClain et al. interpreted the results of the critical values of therapeutic drugs obtained from 36 laboratories without any standardization, they showed that the relative coefficient of variation was large (> 88%) for Ctrough and less (> 19%) for Cpeak (McClain, Owings, & Bornhorst, 2011). The relative coefficient variation for the Cpeak value of theophylline obtained from 33 laboratories was 19%. It has been shown that theophylline can maintain its effectiveness in a narrow therapeutic range by reducing the deviation from the theoretical value in the Ctrough values obtained from this program for IV bolus application (Table 2). In this way, the monitoring of theophylline could be standardized.

When the population PK was compared to the other rabbits in the second group, the deviation% in Ctrough was observed as 11.546 and 50.114, but it was observed that the elevation of deviations at low concentrations did not affect MTC, and also that the practical concentration remained above TI at 5 μg/mL.

In the second post-dose measurement, deviation% values were found to be -8.751 to 4.851, and the target concentration was reached. The program has also been shown to work reliably for multiple dose administration (Table 3).

The third application, infusion, showed that the target concentration was reached in a certain time and the theophylline plasma levels of each rabbit covered the therapeutic range of theophylline (theophylline concentration range of blood samples taken between 15 and 90 minutes between 6.175 and 16.696 μg/mL). The measurement results at the time of reaching the target concentration were 15.718 and 15.245 μg/mL, and the deviations of these values from the theoretical calculation were found to be 2.331% and 17.269%. Also, deviations after the

second infusion rate ranged from -0.846 to 18.664 (Table 4). The program worked successfully in infusion administration.

## 4 Conclusion

Thanks to this program, it has been shown that theophylline, which has a NTI, can be used safely by achieving values close to the therapeutic target. And thanks to this clinical benefit, a cost-effectiveness program was designed.

In the implementation of the program, it is seen that it is a user-friendly program in order to clearly indicate the location of the data to be entered and to guide the user. In addition to the mathematical results, the graphical presentation of the results has made the program more visually understandable and interpretable by the expert.

The software program is in operation and will be developed in the future. TDM is not just a program; it is a whole, from the selection of the sampling time to the analytical method. Developing faster analytical methods will increase the applicability of TDM. For now, the program is practicable for the TDM of theophylline.

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