Clinical Biochemistry 49 (2016) 1390–1395

Contents lists available at ScienceDirect

Clinical Biochemistry

journal homepage: www.elsevier.com/locate/clinbiochem

Sampling on ice will not yield reliable uric acid monitoring in rasburicase-treated patients

Barbara Depreter [a,c], Veronique Stove [a,b], Joris Delanghe [a,b,*]

[a] Department of Laboratory Medicine, Ghent University Hospital, Ghent, Belgium
[b] Department of Clinical Chemistry, Microbiology and Immunology, Ghent University, Ghent, Belgium
[c] Department of Paediatrics and Medical Genetics, Ghent University, Ghent, Belgium

ARTICLE INFO

Article history:
Received 8 March 2016
Received in revised form 22 April 2016
Accepted 24 April 2016
Available online 27 April 2016

Keywords:
Fasturtec®
Kinetics
Rasburicase
Uric acid
Uricolysis
Uricolytic activity

ABSTRACT

Objectives: Rasburicase is administered to prevent hyperuricemia and counteract the consequences of tumour lysis syndrome (TLS). The benefit of monitoring uric acid (UA) concentrations in rasburicase-treated patients is questionable as spuriously low values are most frequently encountered. The manufacturer recommends a cold sample handling procedure to arrest ex vivo uricolysis. Contrariwise, it was recently considered that the temperature does not significantly affects rasburicase uricolysis. We here present a thorough investigation on rasburicase kinetics in clinical samples.
Design and Methods: UA was spiked in sera from rasburicase-treated patients at varying concentrations, divided in three fractions for incubation at 4 °C, 22 °C or 37 °C and measured at fix time points. The Michaelis–Menten constant (K_m) and activation energy (E_{act}) were estimated by linear regression and the Arrhenius equation, respectively. Additionally, UA concentrations retrieved in sera of rasburicase-treated patients were retrospectively studied (3.5 years period).
Results: Although uricolysis increased at a higher temperature, incubation at 4 °C did not arrest uricolysis entirely. The yielded K_m of 128 μmol/L highlights that maximum uricolytic activity is reached at UA concentrations lower than those observed for TLS patients. Furthermore, the E_{act} of 27 kJ/mol corresponds to only a modest logarithmic decrease of the uricolytic capacity by 4–5% per −1 °C. In routine practise, 'negative' UA concentrations were observed during 88.5% of the rasburicase therapy episodes, even when samples were stored at 4 °C.
Conclusion: In contrast to manufacturer's guidelines, simple cooling of the sample will not arrest the temperature-dependent uricolysis provoked by rasburicase and therefore not yield reliable UA monitoring.
© 2016 The Canadian Society of Clinical Chemists. Published by Elsevier Inc. All rights reserved.

1. Introduction

The therapeutic use of urate oxidase (rasburicase) is reserved for paediatric and adult patients with leukaemia, lymphoma or solid tumours undergoing chemotherapy [1–3]. Due to massive (malignant) cell destruction, an excess of intracellular components leaks into the blood stream, which is referred to as tumour lysis syndrome (TLS). Uric acid (UA) is formed during the final catabolic step of the purine metabolism by hypoxanthine oxidase [4]. The limited solubility (402–420 μmol/L at 37 °C) and low acid dissociation constant (pKa 5.4) of UA will lead to precipitation of monosodium urate crystals, especially in low pH regions such as the renal tubules [4,5]. Kidney damage by urolithiasis, tubular necrosis and renal failure are hence major threats in acute setting [6].

The therapeutic enzyme rasburicase will enhance catalyzation of UA into the more readily excretable allantoin [7]. Rasburicase is currently distributed in Europe (Fasturtec® [Sanofi-Synthelabo, Paris, France]) and the U.S. (Elitek™ [Sanofi-Synthelabo Inc., New York, NY]) [8]. The European Medicines Agency (EMA) and U.S. Food and Drug Administration (FDA) recommend a standard dose of 0.15–0.20 mg/kg/day intravenously administered over 30 min with a maximum of 15 mg/day up to five days [9,10]. Therapy duration is adjusted based upon clinical and biochemical monitoring of the patient.

However, in routine practise, the benefit of monitoring UA concentrations in serum/plasma is questionable as spuriously low values are most frequently encountered [11]. This pre-analytical phenomenon is due to the continuous ex vivo uricolysis provoked by rasburicase after the blood samples are taken. According to the manufacturer, a cold sample handling procedure should be able to arrest the ex vivo uricolytic activity of rasburicase entirely [3,11–16]. Hence, blood should be collected in pre-chilled tubes and immediately placed in an ice water

Abbreviations: TLS, tumour lysis syndrome; UA, uric acid; pKa, acid dissociation constant; EMA, European Medicines Agency; FDA, Food and Drug Administration; min, minutes; h, hour; RT, room temperature; LOD, limit of detection; PCCC, PreciControl ClinChem; K_m, Michaelis–Menten constant; E_{act}, activation energy; R^2, correlation coefficient; V, uricolytic activity; V_{max}, maximum uricolytic activity; S, UA concentration; CI, confidence interval.

* Corresponding author at: Ghent University Hospital, De Pintelaan 185 (2P8), B-9000 Ghent, Belgium.
E-mail address: joris.delanghe@ugent.be (J. Delanghe).

http://dx.doi.org/10.1016/j.clinbiochem.2016.04.011
0009-9120/© 2016 The Canadian Society of Clinical Chemists. Published by Elsevier Inc. All rights reserved.

bath (4 °C) during transport to the laboratory. Moreover, the use of pre-cooled centrifuges and cold preservation of the sera prior to analysis (mandatory within 4 h) is advised.

Contrariwise, a recently published paper is claiming that the impact of temperature on the uricolysis provoked by rasburicase (Elitek™) is negligible, provided that analysis is performed promptly upon arrival in the laboratory [18]. This latter study reported no significant differences in UA concentrations between paired samples that were either collected, transported or processed at room temperature (RT) or were collected in pre-chilled tubes, transported on ice and centrifugated at 4 °C, respectively. Most interestingly, UA concentrations never dropped below the limit of detection (LOD) after handling at RT. Hence, refrigeration of the samples may be presumed to be dispensable, making the current pre-analytical amendments for UA measurements in samples from rasburicase-treated patients questionable.

Until now, a thorough investigation of rasburicase kinetics, focussing on the ex vivo uricolysis, is lacking. Here, we present a kinetic study of rasburicase at 4 °C, 22 °C and 37 °C using UA-spiked sera from rasburicase-treated patients. Next, these results are correlated to the UA concentrations retrieved in sera of rasburicase-treated patients, who were routinely monitored at the University Hospital of Ghent (Belgium) during a 3.5 years period.

2. Materials and methods

2.1. Serum UA measurements

Serum UA measurements were performed by an enzymatic, colorimetric test on a Roche Cobas 8000 c701 platform (Roche Diagnostics, Basel, Switzerland). A 2-point end linear calibration was performed using Roche diagnostics' calibrator for automated systems, according to manufacturer's instructions [19]. The method showed an analytical measuring range between 11.9–1487 μmol/L and a LOD of 11.9 μmol/L [19]. Performance characteristics were evaluated using commercial certified control material (PreciControl ClinChem (PCCC) 1 (678 μmol/L) and PCCC2 (1267 μmol/L)). Between-run percent coefficient of variation (CV%) ranged between 0.919–1.19% (PCCC2 – PCCC1) and within-run CV% was 0.790%. Bias and total error of the method varied between 3.96% - 4.25% and 5.92% - 5.76%, respectively (PCCC1 – PCCC2).

2.2. Study of rasburicase kinetics

We studied the uricolysis of varying UA concentrations spiked in sera from rasburicase-treated patients ($n = 5$). All patients suffered from a haematological malignancy (M/F ratio: 4; age: 27–79 years and received a standard rasburicase dose of 15 mg (equivalent to a body weight of 75 kg). Time between rasburicase dosing and sample collection varied between 1 and 23 h (median 9 h 24 min, mean 7 h 4 min). The UA measurements performed prior to spiking consistently showed concentrations below the LOD. A UA solution was prepared for spiking by dissolving crystalline UA (Sigma-Aldrich, St.Louis, Missouri, USA) in 1 mol/L sodium hydroxide (J.T.Baker®, Deventer, The Netherlands) to obtain a final concentration of 297 mmol/L. All spiked sera were divided in three fractions for incubation at 4 °C, 22 °C or 37 °C prior to UA measurements. An ice water bath and a heating block were used to obtain the desirable incubation temperatures of 4 °C and 37 °C, respectively, and samples were kept on the bench for storage at 22 °C. The sera were measured instantaneously (time point 0 min) after spiking, followed by analysis at fix time points.

First, sera from two rasburicase-treated patients were spiked with two different UA concentrations within the adult reference range (202–416 μmol/L). Therefore, 0.5–2 μL of the UA solution was spiked into 1 mL of patient sera, leading to a final concentration of 412.2 μmol/L and 262.3 μmol/L for the first patient and 252.8 μmol/L and 119.0 μmol/L for the second patient. The spiked samples ($n = 4$) were divided in three fractions for incubation at the specified temperatures and UA measurements were performed on each aliquot after 20 min, 40 min and 60 min of incubation.

Next, sera (2 mL) from three other rasburicase-treated patients were spiked with 8 μL UA solution, so that a final high UA concentration of approximately 1200 μmol/L was obtained, as one could encounter in severe TLS patients [20]. These spiked samples ($n = 3$) were also promptly divided in three fractions for incubation at the defined temperatures. Here, the uricolytic activity and UA concentrations were measured every 10 min during a 130 min incubation period. We applied linear regression on the double-reciprocal Lineweaver–Burk plot of the uricolytic activity versus UA concentrations at 22 °C ($n = 2$) and 37 °C ($n = 3$) to calculate the mean Michaelis–Menten constant ($K_{m,}$ equals -1/x-intercept) and the correlation coefficients (R^2). The mean activation energy (E_{act}, kJ/mol) was estimated based on the uricolytic activity in the temperature range 4–37 °C using the Arrhenius equation [21].

In addition, we investigated the uricolytic activity of rasburicase during the fix 5 min pre-incubation at 37 °C on a Roche Cobas 8000 c701 analyser. Serum from a rasburicase-treated patient (UA concentration < 11.9 μmol/L) and a healthy donor (UA concentration 89.2 μmol/L) were both spiked in order to achieve a similar starting UA concentration of approximately 1200 μmol/L and immediately placed on the analyser for measurement.

2.3. Study of rasburicase-treated patients in routine practise

A retrospective data analysis was performed on all hospitalized patients treated with Fasturtec® between 1 January 2011 and 30 June 2014 at the University Hospital of Ghent (Belgium). The study was approved by the Ethical Committee of the Ghent University Hospital and was in accordance with the Declaration of Helsinki. Patients were identified from the historical electronic records and laboratory results and clinical information were retrieved from the hospital's information system. Patients received Fasturtec® 1.5 mg or 7.5 mg, reconstituted in 1 or 5 ml diluent respectively, depending on the amount prescribed by the clinician. If a patient had received rasburicase more than once with an interval of at least four days, each period was enumerated, analysed separately and referred to as a distinct therapy episode.

Within this 3.5 years period, we enrolled 174 rasburicase therapy episodes (155 patients) with adequate biochemical monitoring of the rasburicase therapy, defined as UA concentrations determined within three days after start and stop of therapy. Patients without documented UA concentrations within this time frame were excluded, as accurate evaluation of the speed of declination and regress of UA concentrations was hence impossible. Duration of undetectable UA concentrations was based on the last day showing UA concentrations below the LOD after stop of therapy. Patients (M/F ratio: 1.25; age: 1–90 years) were mainly adults (87.7%), treated because of a haematological malignancy (72.3%). Based on the EMA/FDA dose recommendations (0.15–0.20 mg/kg), patients were divided into five categories: underdosed (<0.10 mg/kg), suboptimal (0.10–0.14 mg/kg), optimal (0.15–0.20 mg/kg), supraoptimal (0.21–0.25 mg/kg) and overdosed (>0.25 mg/kg). During all therapy episodes, blood samples were collected as part of routine clinical care and transported immediately to the laboratory.

2.4. Statistical analysis

Lineweaver–Burk (reciprocal) plots and linear regression were performed by GraphPad Prism (version 5.00 for Windows, GraphPad Software, San Diego California USA). MedCalc for Windows, version 12.3.0.0 (MedCalc Software, Mariakerke, Belgium) was used for the remaining illustrations and the detection of outliers (Tukey). Data considering the retrospective data analysis were processed by IBM SPSS version 22 (SPSS Inc., Chicago, IL, USA).

Download English Version:

https://daneshyari.com/en/article/5509973

Download Persian Version:

https://daneshyari.com/article/5509973

Daneshyari.com