A Comparison of Peripheral and Centrally Collected Cyclosporine A Blood Levels in Pediatric Patients Undergoing Stem Cell Transplant

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Purpose/Objectives: To measure differences in cyclosporine A (CSA) trough concentrations from blood collected as a peripheral sample and from a CSA-uncontaminated (naive) lumen of a double-lumen central line.

Design: Prospective, comparative study.

Setting: Pediatric university teaching hospital in metropolitan Australia.

Sample: 71 paired central and peripheral CSA blood samples from a convenience sample of 14 pediatric allogeneic stem cell transplant recipients receiving IV CSA as prophylaxis or treatment for graft-versus-host disease. Ages ranged from 2 months to 14 years, 5 months.

Methods: Comparing blood samples collected from a peripheral site and a CSA-naive lumen of a double-lumen central line. Data were analyzed using a paired student t test and calculation of the 95% confidence interval of the concentration ratio from different sampling sites.

Main Research Variables: Site of blood sampling and CSA trough concentrations.

Findings: No significant difference existed between CSA concentration in samples collected from the different sites in children receiving intermittent infusions of CSA ($p = 0.13$). The 95% confidence interval of the CSA concentration ratio was 0.92–1.04.

Conclusions: When CSA is administered on an intermittent dosing schedule, comparable CSA trough concentrations can be determined from blood collected via the CSA-naive lumen of a double-lumen central line or at a peripheral sampling site.

Implications for Nursing: Pediatric allogeneic stem cell transplant recipients who require regular CSA trough concentrations no longer will require peripheral blood samples when receiving an intermittent dosing schedule.

Key Points . . .

- Blood sampling from the cyclosporine A- (CSA-) naive lumen of a double-lumen central line is appropriate for monitoring CSA trough concentrations when a patient is receiving intermittent-dose CSA.
- If CSA is administered as a continuous infusion, the accuracy of CSA trough concentrations cannot be ensured; therefore, the authors recommend collection for CSA trough concentrations via a peripheral sample.
- Whenever possible, painful procedures should be avoided in children.

Cyclosporine A (CSA) is an immunosuppressant agent used either alone or in combination with other therapies in allogeneic stem cell transplant (SCT), for prophylaxis and treatment of graft-versus-host disease (GVHD). GVHD is a potentially serious complication of SCT, and subtherapeutic blood CSA concentrations may increase a patient's risk for developing GVHD (Yee et al., 1988). CSA concentration monitoring is essential in the clinical management of patients undergoing SCT to ensure adequate dosing and to minimize the toxicity of the medication (Kami et al., 2000; Morris et al., 2002). Significant variability among patients in the metabolism of CSA, medication interaction, and clinical condition requires regular monitoring.

To ensure reliable CSA concentrations, the standard of practice at the authors' institution was changed from monitoring CSA trough concentrations in blood collected from the double-lumen tunneled central venous line to peripheral blood (venipuncture or capillary sample) sample. This change occurred because of an apparent variability in CSA trough concentrations when collected via the central line.

Several investigations have evaluated the administration and therapeutic monitoring of CSA with attention to the method of blood collection. Blifeld and Ettenger (1987) reported their experiences with two renal allograft recipients who received IV CSA via an indwelling, single-lumen, polyurethane catheter and subsequently had trough CSA concentrations collected from the same catheter. These researchers reported unusually elevated CSA trough concentrations and subsequently collected peripheral and indwelling catheter trough levels that showed a significant difference between the two samples. They postulated that CSA adheres to the intraluminal plastic in the central venous line. This finding highlighted a potential problem with the reliability of the CSA trough concentration blood samples that were collected from the same lumen by which the CSA was administered. Leson, Bryson, Giesbrecht, and Saunders

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Digital Object Identifier: 10.1188/05.ONF.73-77

(1989) studied four pediatric bone marrow transplant recipients with single-lumen silicone central venous lines through which medication administration and sample collection were performed. The children initially received IV CSA through the single-lumen line and subsequently received oral CSA therapy. The children had peripheral capillary samples collected simultaneously with central line samples for determination of CSA trough concentrations. CSA trough concentrations collected while the children were on IV therapy showed a significant difference between the peripheral and central blood samples. This difference seemed to dissipate once a child was on oral therapy. However, only 16 paired samples were analyzed.

Soto, Alsar, Avendano, Sacristan, and Zubizarreta (1992) attempted to compare CSA trough concentrations that had been collected simultaneously from a single-lumen central line and a venipuncture. The central line was used for medication administration and blood sampling. The 11 SCT recipients had either silicone (n = 4) or polyurethane (n = 7) catheters that were inserted for their clinical care. These researchers found that CSA trough concentrations measured from the central line blood samples were significantly higher ($p < 0.001$) in comparison to concentrations in peripheral blood samples. The results were independent of the type of catheter (Soto et al.). These researchers recommended repeating this study but collecting CSA blood samples from a CSA-naive (uncontaminated) lumen (Soto et al.). A limitation of all of these studies is the relatively small sample sizes, but the findings support the need to investigate the effect of different sites of blood sample collection on CSA blood concentration.

Only one study has evaluated the effect of continuous-infusion CSA on blood concentration levels in relation to site of blood sampling. Shulman, Ou, Reed, and Gardner (1998) studied patients who received a continuous infusion of CSA via a single-lumen central line with six CSA blood concentration samples collected from the catheter. Despite this small sample, the researchers concluded that a significant difference existed between peripheral and central CSA trough concentrations.

More recently, Claviez, Glass, Droger, and Suttorp (2002) found that adsorption of CSA in the inner surface of central line catheters could contribute to spurious CSA concentrations in samples collected from these lines. As expected, the error was greatest when the same lumen was used for drug infusion. Shibata et al. (2000) found that infusion sets made of polyvinyl chloride markedly adsorbed CSA but catheters made of polyethylene or polybutadiene showed no adsorption of CSA. No studies have examined the reliability of CSA blood samples when a double-lumen catheter was used and one lumen remained uncontaminated by CSA.

Venipuncture is associated with pain and distress in all patients with cancer, but it is well documented in pediatric patients (Van Cleve, Johnson, & Pothier, 1996). In children, venipuncture requires increased nursing time to support a child through the procedure using distraction and holding as well as additional clinical laboratory time and waiting for the child to be prepared. These lead to additional costs and potentially delay therapeutic interventions. Removal of as many of these types of procedures as possible improves the experience of children undergoing SCT.

The aim of this study was to demonstrate that when CSA is administered solely in one lumen of the double-lumen central line, the CSA blood concentrations obtained from the CSA-naive lumen accurately would reflect the circulating blood concentration of CSA. If the study were to show that no significant difference existed between the paired CSA trough concentrations, it would support a change in practice, eliminating the need for regular peripheral blood sampling from the care of the pediatric SCT recipients.

Methods

Study Design

A prospective comparative study was conducted using a convenience sample of blood CSA concentrations from pediatric SCT recipients undergoing an allogeneic transplant who received IV CSA for GVHD prophylaxis and treatment. The patients were recruited prospectively from a 16-bed pediatric hematology/oncology unit, with four designated SCT beds in a pediatric university teaching hospital in New South Wales, Australia. The study had the approval of the Southeast Sydney Area Health Research Ethics Committee. All patients (when age appropriate) and their guardians signed an informed consent to participate in the study. All patients who were approached agreed to participate in the study.

Subjects

The unit of analysis in this study was the CSA samples collected from pediatric allogeneic SCT recipients. All patients had a double-lumen tunneled silastic central venous line inserted prior to the commencement of conditioning therapy. The patients in this study all were enrolled prior to the initiation of their transplant conditioning therapy. The CSA collection began with their initial dose of CSA (day –2) and continued for the duration of their IV CSA therapy, which was throughout their post-transplant period. CSA sample collection ceased when IV therapy was discontinued in preparation for discharge, when a patient was transferred to the intensive care unit, or when death occurred.

Measures

All blood samples were analyzed for CSA concentration using the Behring Diagnostics Emit® immunoassay (Dade Behring, Deerfield, IL). The CSA target trough concentration for SCT recipients is 150–300 ng/ml. The peripherally drawn sample (standard of care) was processed in the same manner as other clinical blood samples. A patient's identifying details as well as the date and time collected were placed on the tube and laboratory form. Each central line sample (research sample) and the corresponding laboratory form were labeled as being from the central line and indicate the site, date, and time of collection. The laboratory staff did not enter the central line sample results into the clinical laboratory computer, and the clinicians remained blinded to the study sample results. The researcher obtained the central line sample results directly from the laboratory staff for later data analysis. The results of the central line sample remained blinded to the medical team providing direct patient care until the completion of the study. The peripheral blood samples were available for the clinical management of the patient as per the practice prior to the study.

Because choice is an important component of pediatric nursing practice, children selected which method they wanted their healthcare providers to use when their peripheral blood samples collected: capillary sample or venipuncture. Sufficient data existed to support a high correlation between the two sites (Merton, Jones, Lee, Johnston, & Holt, 2000;

Pettersen, Driscoll, Moyer, Dearani, & McGregor, 1999; Profumo, Foy, & Kane, 1995). All samples were analyzed as a single data set described as peripheral sampling.

Procedure

CSA was administered via IV, either as twice-daily short infusions or as a continuous infusion via one lumen of the double-lumen central line. Each patient had blood samples for CSA measurement collected when clinically necessary; these blood samples were taken at the end of the dosing interval prior to the next dose and were considered trough levels. The standard of practice was to collect CSA trough concentrations on Monday and Thursday mornings prior to the morning dose of CSA and at other times as clinically necessary. Patients had either a Broviac® (Bard Access Systems, Murray Hill, NJ) or a LuMax® (Cook Inc., Bloomington, IN) catheter inserted. Earlier research identified that CSA adheres to the internal lumen of catheter of silastic (silicone) and polyurethane catheters (Blifeld & Ettenger, 1987; Leson et al., 1989; Soto et al., 1992).

This study was performed within the constraints of standard clinical care. Ideally, the peripheral and central blood samples would be collected simultaneously. This was not feasible because of the workload of the nursing and phlebotomy staff. For this reason, the time of collection for each sample (peripheral and central) was included in the data. The documented time lapse between the paired samples permitted the CSA blood concentration data to be corrected for sampling time to allow a comparison of the observed and expected concentration at the same time. This correction is based on the knowledge that after a bolus infusion, blood concentrations of CSA will decline in a time-dependent manner. Concentration data were corrected for sampling time using the expected half-life of CSA of 6.5 hours, assuming an exponential decline in the concentration-time profile.

The RNs on the hematology/oncology and SCT unit did not routinely perform venipuncture or capillary sampling; therefore, phlebotomy nurses collected the first blood sample for CSA measurement via a peripheral sample. As soon as possible after the collection of the peripheral sample, the second blood sample was collected from the CSA-naive lumen by the nursing staff. The double-lumen catheters that were used in this population had different-colored lumens, generally red and white. The RNs were alerted to the correct lumen for administration and collection of CSA blood samples in the following manner. After a child and parent consented to participate in the study, a sign was placed on the patient's door identifying which lumen was to be used for administration of CSA and which was to be used for the collection of the research blood sample. These signs did not indicate whether a recipient was in the study. The morning of the first dose of CSA, the researcher or staff nurse marked on the sign which lumen was used to infuse the CSA (generally the white lumen). The sign also documented which was the CSA-naive lumen (generally the red lumen), which was to be used for collecting the blood sample for the CSA concentration measurement. Another sign was placed inside the patient's room in the area where the RNs reconstituted the CSA, and additional documentation was recorded on the patient's daily flow sheet. The RN did not administer the morning dose of CSA until the peripheral blood and research samples were collected. All RNs employed on the hematology/oncology and SCT unit received education on the management of central lines and are assessed to ensure compliance with the hospital guidelines. Prior to the initiation of the study, the researchers reviewed with the nursing staff the standardized blood collection procedure as well as the importance of proper blood collection for the purposes of the study. The central line sample was collected using the institution's guidelines (see Figure 1).

Blood taken from the extension tube, which was directly connected to the cyclosporine A- (CSA-) naive lumen of the central line. If difficulty occurred when withdrawing the blood sample, the specimen was collected directly from the hub of the CSA-naive lumen.

1. Clamp CSA infusion lumen of the double-lumen central line.
2. Withdraw and discard 5 ml blood.
3. Change syringe; collect standard daily blood samples (complete blood count [CBC], urea, creatinine, electrolytes, and liver function studies).
4. Change syringe; collect blood sample for CSA level (1 ml in ethylenediaminetetra-acetic acid [EDTA] tube).
5. Change syringe; flush with 5 ml of normal saline.
6. Unclamp line and restart infusions.

If the patient was receiving a continuous infusion of CSA,

1. Stop infusion while blood collection process is initiated (e.g., when the RN begins to set up to withdraw the blood samples).
2. Clamp CSA infusion lumen of the double-lumen central line.
3. Flush central line with 5 ml of normal saline prior to withdrawing and discard.
4. Change syringe; collect standard daily blood samples (CBC, urea, creatinine, electrolytes, and liver function studies).
5. Change syringe; collect blood samples for CSA level (1 ml in EDTA tube).
6. Change syringe; flush with 5 ml of normal saline.
7. Unclamp lines and restart infusions.

Figure 1. Protocol for Collection of Blood Samples From the Naive Lumen of the Central Line for Cyclosporine A Monitoring

Data Analysis

Seventy-one paired CSA trough concentrations were collected from the 14 pediatric SCT recipients enrolled in this study. Of the 71 paired samples, 50 samples had documented collection times. CSA concentration data pairs for each patient, from different sampling sites, were compared using a two-tailed, student t test at the $p \leq 0.05$ level of significance and generating the 95% confidence interval of the ratio between CSA blood concentration from different sampling sites. This later parameter allowed an assessment of the size of the difference observed between sampling sites. The paired student

Table 1. Patient Demographics

Variable	n	%
Age		
$\bar{X}$ = 71.4 months (6 years)	–	–
Range = 2 months–14 years, 5 months	–	–
Gender		
Female	7	50
Male	7	50
Diagnosis		
Leukemias	9	65
Hematologic disorders	2	14
Inborn error of metabolism	1	7
Primary immunodeficiency	1	7
Histiocytic disorder	1	7

N = 14

Table 2. Uncorrected and Corrected Cyclosporine A Concentration Observations From Peripheral and Central Sampling Sites

Observations	Peripheral IV (PIV) Sample $\bar{X} \pm SD$	Central Venous Sample $\bar{X} \pm SD$	Ratio PIV/Central Line (95% Confidence Interval)
Uncorrected (n = 71)	190.5 ± 104.5 ng/ml	208.0 ± 137.8 ng/ml*	0.98 (0.92–1.04)
Corrected (n = 50)	170.0 ± 96.0 ng/ml	196.8 ± 137.3 ng/ml**	0.94 (0.86–1.01)

* p = 0.129

** p = 0.120

t test was performed to determine whether a significant difference existed in the means of the results obtained by peripheral sample versus blood sampling from the central line.

Results

Subject demographic profiles of the 14 patients are listed in Table 1. The mean CSA concentration from each sampling site was in close agreement, suggesting that no significant difference existed in CSA concentration determined by the sampling sites (p = 0.13). This was confirmed by the 95% confidence interval of the ratio of the CSA concentrations (0.92–1.04) (see Table 2).

The time lapse between each sample collected in this study ranged from 0–50 minutes, with an average time lapse between samples being 16.3 minutes. Based on the knowledge of the expected pharmacokinetics and how the CSA concentration declines with time, the CSA concentrations were corrected depending on the timing of the specimen collection. Fifty sample pairs with completed times were analyzed, and the 95% confidence interval of concentration ratio included unity. Furthermore, a paired t test to compare the concentration data collected from the peripheral and central sampling sites concluded that the CSA concentrations were not significantly different (p = 0.12).

Discussion

Monitoring CSA blood concentrations plays an important role in the management of patients on immunosuppressive therapy (Morris et al., 2002; Yee et al., 1988). The narrow safety margin for CSA means that a concentration range is targeted to achieve adequate immunosuppression without significant unwanted side effects (Kami et al., 2000). The clinical care of children must use the least invasive method of sampling possible, and this study has attempted to address this important question in the context of CSA concentration monitoring.

The reliability of the central line sample could not be ensured in the continuous-infusion CSA recipients. The CSA concentrations in three paired samples were significantly elevated in comparison to the corresponding peripheral samples. All three of these samples were from patients who were receiving a continuous infusion of CSA. The other pathology samples collected at the same time as these three CSA samples were grossly abnormal without the corresponding symptomatology (e.g., elevated serum potassium and elevated serum glucose in patients receiving parenteral nutrition). The authors believe that this is evidence that these samples were contaminated by blood from the continuous infusions via the other lumen, leading to erroneously high CSA concentrations. These samples were collected at the initiation of the study, when continuous-infusion CSA was a relatively new practice compared to intermittent infusion, and sample collection procedures may have contributed, in part, to this variability. Therefore, the authors recommend that patients who receive continuous-infusion CSA should continue to require venipuncture for monitoring of CSA trough concentrations.

The study results showed that blood sampling from the naive lumen of a double-lumen central line is appropriate for CSA monitoring in children receiving IV intermittent doses of CSA. However, this study was not able to establish that it was accurate to collect CSA trough concentrations from a double-lumen central line if the patient was receiving continuous-infusion CSA. The nursing staff has incorporated the central line collection of CSA into their standard of care. Replication of the study and evaluating just continuous-infusion CSA trough collection would be beneficial to the last group of pediatric SCT recipients who are receiving CSA as a component of their care.

The authors would like to thank the nursing staff on the pediatric hematology/oncology and stem cell transplant unit for their assistance in the sample collections. They also would like to thank Daya Naidu, MD, MB, ChP, FRACPA, MAACB, associate professor and director in the Clinical Chemistry Laboratory at Prince of Wales Hospital in New South Wales, Australia, and the laboratory staff for their support in performing the assays and maintaining the blinded nature of the study, as well as Marcus Vowels, AM, MBBS, MD, FRACP, professor and director of the Pediatric Stem Cell Transplant Unit at Sydney Children's Hospital, Randwick, Australia, for his support and access to his patients to perform this study.

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