''In Medicine one must pay attention not to plausible theorizing but to experience and reason together. . . . I agree that theorizing is to be approved, provided that it is based on facts, and systematically makes its deductions from what is observed. . . . But conclusions drawn from unaided reason can hardly be serviceable; only those drawn from observed fact.'' Hippocrates: *Precepts. (Short communications of factual material are published here. Comments and criticisms appear as letters to the Editor.)*

"You Are Hereby Commanded to Appear": Pediatrician Subpoena and Court Appearance in Child Maltreatment

ABSTRACT. ***Objective.*** **To determine the frequency of court appearance by pediatricians evaluating child abuse and neglect cases and to identify case characteristics associated with actual court appearance or case adjournment.**

Design. **Retrospective review of subpoenas received between 1995 and 1999 for child maltreatment cases personally evaluated by 2 pediatricians during the years 1995 to 1998. Information was collected regarding patient age, gender, race/ethnicity, type of suspected maltreatment, date of evaluation, date of subpoena, type of court hearing, whether the pediatrician actually testified in court, and legal outcomes. Case characteristics were compared between pediatricians and were used to predict physician appearance and case continuance or adjournment in logistic regression models.**

Results. **Four hundred forty-five subpoenas concerning 260 patients were received. Although significant differences were noted between the pediatricians in type of abuse, no differences were found in patient age, gender, ethnicity or legal outcomes. The pediatricians received subpoenas in <15% of child maltreatment cases, and <5% of children seen resulted in the physician being required to actually appear in court. No case characteristics significantly predicted court appearance or case continuance or adjournment.**

Conclusions. **Although pediatricians are sometimes subpoenaed to appear in court to explain the medical evaluation and the needs of the child in cases of child abuse and neglect, most court cases were continued, adjourned, or settled before physician testimony. Most subpoenas did not result in the pediatrician going to court, and it is unclear which child factors may predict court involvement. Pediatricians can take steps to minimize (but not eliminate) the potential dissatisfaction and inconvenience associated with receiving and responding to subpoenas in child maltreatment cases.** ***Pediatrics*** **2001; 107:1427–1431;** ***subpoena, court appearance, child maltreatment, child abuse.***

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Professionals who care for children who are suspected victims of abuse and neglect are sometimes required to appear in court to explain the results of their evaluation and the needs of the child.[1–3] Court testimony, however, has not traditionally been considered part of routine pediatric practice although Abraham Jacobi noted the physician's role as "legitimate advisor to the judge and jury" in 1904.[4] Despite their important role in protecting children, pediatricians have reported uneasiness evaluating and reporting suspected abuse and have cited their apprehension with the adversarial process in court for a variety of reasons, including their lack of familiarity with legal procedures and the inconvenience of court attendance.[5–9] Others have postulated that physicians "fear" participation in the legal system because of professional liability or countertransference issues.[10,11]

It has been suggested that pediatricians should come to understand the legal process if they are to fulfill their role as advocates for the child's needs.[12] Involvement in the legal system begins with the issuance of a subpoena by an attorney for a litigant, the clerk of the court, or the judge. A subpoena is a document "commanding a person to appear before a court or other tribunal, subject to a penalty for failing to comply."[13] Subpoenas are issued to produce the factual or expert testimony that enables the court to make an informed determination either in a criminal matter (a trial of a defendant accused of a crime) or in a civil court dependency matter (the determination of abuse, neglect, parental supervision, rights or custody). Physicians and other health professionals who care for children with suspected abuse and neglect have historically provided invaluable information to courts regarding medical diagnoses, prognosis, and statements made by the child patient. Although subpoenas note the date and time of required court appearance, attorneys and judges may continue, adjourn, or cancel hearings with little or no notice to witnesses because of last minute agreements, physician expertise or availability, past record of the defendant, or other court procedures. The impact of subpoenas and court testimony on pediatric practice in child maltreatment cases has not been reported.[14]

We hypothesized that many subpoenas are issued that do not actually result in physician court appearance and that the need for the pediatrician in court depends on several factors, including case characteristics and type of court proceeding.

METHODS

We retrospectively reviewed all subpoenas received between 1995 and 1999 requesting court appearance related to child maltreatment cases evaluated between 1995 and 1998 by 2 pediatricians with expertise in the evaluation of child abuse and neglect. Information regarding patient age, gender, ethnicity, type of alleged maltreatment, timing of subpoena, timing of court appearance, type of court, and location was collected. Court outcomes were also recorded, including whether the physician had actually been required to appear in court and the disposition of the matter necessitating the subpoena (postponement, finding of guilt, plea of guilt, finding of not guilty, dismissal, or acceptance of responsibility). Cases that are postponed without being dismissed are considered to be adjourned or continued; these are interchangeable terms that are variably used in different jurisdictions.

Subpoenas were collated based on individual patients, and multiple subpoenas were sorted based on type of court (civil versus criminal) and nature of hearing (adjudication, determination of supervision or custody, deposition, preliminary examination or grand jury indictment, or trial of the accused). Time delays were calculated from actual patient visit to issuance of subpoena and need for appearance in court. Baseline information concerning the number of maltreatment cases evaluated by these physicians was used for comparison. Child maltreatment types were compared with patient characteristics, and rates of court appearance were calculated based on the total number of subpoenas received. Case characteristics and court outcomes were compared for the 2 pediatricians using χ^2 and 2-sample independent *t* test analyses; they were then used as independent variables with court appearance and case adjournment/continuance as dependent variables in multiple logistic regression models using the SAS statistical program (SAS Institute Inc, Cary, NC). We then conducted power analyses for these regression models using the SAS and PASS 2000 programs (NCSS Statistical Software, Kaysville, UT).

The study protocol was approved by the applicable human subjects research committees at our institutions.

RESULTS

Four hundred forty-five subpoenas were received between 1995 and 1999, pertaining to 13% of the 2018 patients personally evaluated for child maltreatment by the 2 physicians in 2 different states. Significant differences were noted between the physicians in patients' type of suspected maltreatment, but not age, race/ethnicity, or gender (Table 1). Children were primarily evaluated for suspected sexual (51%) and physical abuse (46%), although the type of maltreatment was different between the physicians ($P = .025$).

Most of the subpoenas were issued in the county where the physician's practice was located, although both physicians received subpoenas from >15 counties from a distance of over 250 miles. Most (63%) of subpoenas received were related to criminal proceedings (preliminary hearings or criminal trials), but 37% were related to civil (probate or family court) hearings designed to determine parental custody, adjudicate abuse, or dependency issues (Table 2). Most court proceedings resulted in a plea or finding of guilt of the perpetrator or responsibility of the family (52%), but 38% of the proceedings were adjourned or continued to a later date. On average, 1.7 subpoenas were issued per child (range: 1–8) over a period of 0 to 50 months after evaluation. Mean time delay from patient evaluation to subpoena issuance was 4.3 months, with shorter time periods for preliminary examinations (1.6 months) and civil actions (3.4 months), and there were longer time delays in criminal trials (6.7 months). Physician appearance in court was actually required for 21% of the subpoenas issued, only being needed for <5% of children evaluated for possible maltreatment.

Despite differences in patient abuse type, no difference in court outcomes or appearance in court were noted between the physicians ($P > .05$). Patient age and gender were significantly related with maltreatment type, with more physical abuse and neglect seen in younger, male patients (71% of males were referred for physical abuse or neglect, mean age 2.5 years, standard deviation = 3.7), and more sexual abuse was noted in older, female children (67% of females were referred for sexual abuse, mean age 7.31 years, standard deviation = 4.1, $P < .0001$). Rate of appearance in court was not related to receiving more subpoenas in a case for a particular type of court.

Actual physician court appearance and case adjournment or continuance were not predicted by patient characteristics or type of suspected maltreatment in logistic regression models, although significant differences for maltreatment type and case adjournment/continuance were noted in univariate analysis. Using a method modified from O'Brien[15] for log-linear models, the power of this sample of 260 children for detecting significant effects of maltreat-

TABLE 1. Patient Characteristics, 1995–1998

	Physician A	Physician B	Total	*P*
Number patients with subpoena	118	142	260	
Mean age, y standard deviation†	5.8 (4.8)	4.2 (4.5)	4.6 (4.5)	NS
Gender (%)*				NS
Male	46 (39)	58 (41)	104 (40)	
Female	72 (61)	82 (58)	154 (59)	
Race/ethnicity (%)*				NS
White	68 (63)	89 (64)	157 (63)	
Black	31 (28)	43 (31)	74 (30)	
Latino, Asian, biracial	9 (8)	8 (5)	17 (7)	
Type of suspected maltreatment (%)*				.025
Physical abuse	45 (38)	75 (53)	120 (46)	
Sexual abuse	67 (57)	65 (46)	132 (51)	
Neglect	6 (5)	2 (1)	8 (3)	

NS indicates not significant. $P > .05$.
* Numbers may not equal totals and percentages may not total to 100 because of rounding or missing data.
† Two-sample *t* test.

TABLE 2. Legal Outcomes, 1995–1999

	Physician A	Physician B	Total	*P*
Total subpoenas received	189	256	445	
Number of court appearances (%)	46 (24)	46 (18)	92 (21)	NS
Court type (%)				NS
Civil	78 (41)	85 (33)	163 (37)	
Criminal	111 (59)	171 (67)	282 (63)	
Mean number of subpoenas				
Per patient (SD)*	1.7 (1.1)	1.8 (1.1)	1.7 (1.1)	NS
Mean time to first subpoena, mo (standard deviation)	4.7 (7.2)	4.1 (6.2)	4.3 (6.7)	NS
Civil			3.4 (5.2)	
Preliminary hearing			1.6 (1.8)	
Criminal Trial			6.7 (7.0)	
Court appearance (%)				NS
Yes	46 (24)	46 (18)	92 (21)	
No	143 (76)	209 (82)	352 (79)	
Court outcome (%)†				.01
Adjournment/continuance	86 (46)	83 (32)	169 (38)	
Finding of guilt‡	99 (52)	132 (52)	231 (52)	
Not guilty/not responsible§	4 (2)	16 (6)	20 (5)	

NS indicates not significant. $P > .05$.
* Two-sample *t* test.
† Numbers may not equal totals and percentages may not total to 100 because of rounding or missing data.
‡ Includes pleas of guilt or no contest, indictment or finding of sufficient evidence for trial in criminal proceedings and finding or acceptance of responsibility in civil proceedings.
§ Includes dismissal or no determination.

ment type and gender differences in court appearance or case adjournment/continuance was 0.7 and 0.6, respectively, with $\alpha = 0.05$.

DISCUSSION

This review of subpoenas and court appearance by 2 pediatricians in 2 different states offers a glimpse into the participation of health care professionals in the legal system. Courts and litigants have come to rely on physician-generated evidence and testimony in settling on litigation strategy and making their determinations regarding the handling of cases, yet most subpoenas will not result in the need for physician court appearance. It is surprising that <15% of these patients resulted in the issuance of a subpoena. Furthermore, only 4.3% of patient visits for medical evaluation of child abuse and neglect resulted in the physician going to court. Most cases resulted in adjournment/continuation or were adjudicated with a plea of guilt or the family's acceptance of responsibility without physician testimony. This is different from the experience of forensic pathologists, for example, who in one study were subpoenaed in 64% of their homicide cases, only one third of which required court testimony.[16]

Why are so many subpoenas issued that do not result in physician appearance in court? Sometimes a legal finding is made before or during a hearing without the physician's participation, and we suspect that many courts determine that the physician's testimony is not necessary after subpoena because the medical issues are not in question and/or the physician is not available to appear in court. The vast majority of criminal cases are resolved when the accused pleads guilty thereby eliminating the need for a trial. In 1996, 91% of all felony charges filed in state courts and 93% of such cases filed in the federal system were resolved by a plea of guilt in 1997.[17,18] Less physician testimony may also be needed with good medical documentation or in states that permit the admission of medical records without the testimony of a live witness, if the records are accompanied by a certification that they are complete.[19] Court procedures can also require subpoenaing all potential witnesses regardless of whether their testimony is actually important to the legal case. Comparisons with the experience of other professionals in child maltreatment cases may be helpful, but little published information is available.

Predicting court appearance is problematic. Although some have suggested that physicians will appear more often in court with younger patients or with certain types of abuse, these were not predictive in our study.[3] Although the majority of our subpoenas were issued in the first year after a patient visit, some subpoenas were issued more than 3 years after medical evaluation. Time elapsed was related to the type of court in that preliminary hearings or probate or family matters occurred sooner after the patient visit than do criminal trials. It can take many months for a subpoena to be issued, thereby increasing the practitioner's uncertainty as to a court date. We hypothesize that time delay to subpoena is related to the purpose of the hearing, with issues concerning protection of the child necessitating more prompt hearings, and the statutory procedures related to the defendant in the criminal justice system which can cause greater delay.

Physicians received >1 subpoena per child in many cases when a subpoena was issued, and cases with multiple court proceedings or multiple adjournments generated several subpoenas. More subpoenas, however, did not increase the likelihood of court appearance in a particular case. Although the power of this study was limited, no factors significantly predicted appearance in court, and there was no

significant association with patient age, gender, ethnicity, type of abuse, or type of court. In our experience, the most effective way to predict actual court appearance is to contact the court officer issuing the subpoena to discuss the matter and to assess our potential role in the legal determination. When available, court-sponsored victim-witness units can also provide invaluable information.

There are significant differences between the 2 physicians in this study with respect to type of suspected maltreatment and rates of adjournment or continuance that undoubtedly reflect population and practice differences. Differing referral patterns and availability of additional community resources may affect patient characteristics. Gender-based differences associated with types of alleged abuse have been noted nationally, with more physical abuse among boys seen and more sexual abuse seen among girls.[20] Neglect cases represented <5% of all subpoenas, which may reflect fewer criminal prosecutions, less disagreement of the parties, less need for physician interpretation, or a less well-defined role for physicians in the evaluation of neglect.[21]

When controlled in a regression model, physician differences did not explain the low frequency of court appearance. The specific circumstances surrounding subpoena issuance and a decision by court officers to not compel physician appearance requires additional study. It is unfortunate that so many unnecessary subpoenas are issued because receiving a subpoena and planning to attend a court hearing may significantly disrupt a physician's professional schedule and inconvenience patients, families, colleagues, and institutions, particularly when there are multiple adjournments. It is our experience that each case requires 1 to 3 hours to schedule, prepare, and testify. We sometimes have to wait an hour or more outside the court in addition to any travel time. This requires us to cancel and reschedule 4 or more hours of patient care; this is worsened when last-minute changes are made or when we arrive at court only to find the case adjourned or continued. Inconvenience to patients and physicians could be improved by providing sufficient time (>72 hours) and a time certain for testimony to allow proper patient scheduling. We suspect that the process as is currently practiced of receiving subpoenas, scheduling court appearance and adjournment has contributed to physicians' dissatisfaction with the legal system and that significant improvements could be made to improve attorney-physician communication before and after subpoena issuance. Some have suggested the use of pretrial meetings and the need for additional research in this area.[14]

There are several limitations to this study. It reflects the experience of 2 pediatric practitioners specializing in the care of children with concerns of abuse and neglect. "Forensic pediatrics" has been noted to include expertise in the medical evaluation and treatment of maltreated children and court appearance, and we can only speculate what the experience would be of pediatric generalists.[22] Nonpediatric health professionals may receive a substantially different number of subpoenas, with some (such as forensic pathologists and emergency physicians) routinely receiving subpoenas for a variety of concerns beyond child maltreatment. This study also reflects physician experience and practices in a limited geographic area; wide variations in court practices have been noted in different jurisdictions.[17]

The nature of any differences between general and forensic pediatricians in the rates of subpoena or court appearance requires additional study. Although most child maltreatment cases are adjudicated within 1 year, additional subpoenas may yet be issued in our cases; this could increase the percentage of cases resulting in subpoenas and court appearance. It appears that subpoenas are often issued at the discretion of the prosecutor or defense attorney who may demonstrate wide variation in their use of this power. Because information was not available regarding patient characteristics of those cases that did not result in a subpoena, we are unable to predict whether a subpoena will be issued in the first place. These are areas for future research as we refine our knowledge of the role of medical professionals in the legal system.

CONCLUSION

Pediatricians and other health care providers may become concerned about the possibility of being required to appear in court at the time of initial patient encounter and may wonder if and when this case will go to court. Based on the experience of 2 pediatricians with expertise in the evaluation of child abuse and neglect, the proportion of child maltreatment cases requiring court appearance by the pediatrician is low. The issuance of a subpoena indicates that the physician's involvement is recognized by litigants or the court as being important to resolution of the legal issues in the case, but the pediatrician should not interpret the issuance of a subpoena to mean that court appearance will eventually be required. Case characteristics of the child in question do not reliably predict court outcome or whether actual court appearance will be mandated.

We recommend contacting the issuing court officer directly after receiving a subpoena to determine if the physician's court appearance will actually be required and to determine a mutually convenient time for that appearance. Additional research is required to understand why subpoenas are issued in particular cases, the impact of the subpoena process on different physician and professional groups, and whether changes in the legal system could result in improved subpoena issuance with more efficient use of physician time and testimony in child maltreatment cases.

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A Randomized Trial Comparing Povidone-Iodine to a Chlorhexidine Gluconate-Impregnated Dressing for Prevention of Central Venous Catheter Infections in Neonates

ABSTRACT. **Neonates who require a central venous catheter (CVC) for prolonged vascular access experience high rates of catheter-related bloodstream infection (CRBSI).**

Purpose. **A multicenter randomized clinical trial was undertaken to ascertain the efficacy of a novel chlorhexidine-impregnated dressing (Biopatch Antimicrobial Dressing) on the CVC sites of neonates for the prevention of catheter tip colonization, CRBSI, and bloodstream infection (BSI) without a source.**

Setting. **Six level III neonatal intensive care units.**

Patients Studied. **Neonates admitted to study units who would require a CVC for at least 48 hours.**

Methods. **Eligible infants were randomized before catheter placement to 1 of the 2 catheter site antisepsis regimens: 1) 10% povidone-iodine (PI) skin scrub, or 2) a 70% alcohol scrub followed by placement of a chlorhexidine-impregnated disk over the catheter insertion site. A transparent polyurethane dressing (Bioclusive Transparent Dressing) was used to cover the insertion site in both study groups. Primary study outcomes evaluated were catheter tip colonization, CRBSI, and BSI without an identified source.**

Results. **Seven hundred five neonates were enrolled in the trial, 335 randomized to receive the chlorhexidine dressing and 370 to skin disinfection with PI (controls). Neonates randomized to the antimicrobial dressing group were less likely to have colonized CVC tips than control neonates (15.0% vs 24.0%, relative risk [RR]: 0.6 95% confidence interval [CI]: 0.5–0.9). Rates of CRBSI (3.8% vs 3.2%, RR: 1.2, CI: 0.5–2.7) and BSI without a source (15.2% vs 14.3%, RR: 1.1, CI: 0.8–1.5) did not differ between the 2 groups. Localized contact dermatitis from the antimicrobial dressing, requiring crossover into the PI treatment group, occurred in 15 (15.3%) of 98 exposed neonates weighing ≤1000 g. No neonates in the PI group developed contact dermatitis.**

Conclusion. **The novel chlorhexidine-impregnated dressing, replaced weekly, was as effective as cutaneous disinfection with 10% PI and redressing the site every 3 to 7 days for preventing CRBSI and BSI without a source in critically ill neonates requiring prolonged central venous access. The risk of local contact dermatitis under the chlorhexidine dressing limits its use in low birth weight infants who require prolonged central access during the first 2 weeks of life. *Pediatrics* 2001;107:1431–1437; *bacteremia, bloodstream infection, catheter-related bloodstream infection, central venous catheter, chlorhexidine gluconate, cutaneous antisepsis, neonate.***

ABBREVIATIONS. BSI, bloodstream infection; CVC, central venous catheter; CRBSI, catheter-related bloodstream infection; PI,

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povidone-iodine; CFU, colony-forming unit; CNS, coagulase-negative *Staphylococcus*; RR, relative risk; CI, 95% confidence interval; PFGE, pulsed field gel electrophoresis.

Critically ill neonates have a high incidence of nosocomial bloodstream infection (BSI), which most often derives from central venous cathethers (CVCs) needed for prolonged central access.[1–7] CVC-related BSIs increase exposure of neonates to potentially toxic antibiotics and greatly increase length of stay and hospital costs.[8,9] Many of these infections derive from invasion of the transcutaneous catheter tract by microorganisms from the cutaneous flora, particularly during the first 2 weeks of catheterization.[10–15] Suppressing catheter site colonization with local antisepsis is an effective means of reducing the risk of catheter-related bloodstream infection (CRBSI).[16,17]

Although 10% povidone-iodine (PI) is widely used for skin antisepsis before placement of CVCs in neonates, systemic absorption of iodine by premature neonates after iodophor scrubs with development of laboratory findings of hypothyroidism, has been reported.[18–20] Recent trials in adults suggest that chlorhexidine gluconate is a more effective cutaneous antiseptic than PI for the prevention of CVC-related BSI in adults[16,17] and peripheral intravenous catheter tip colonization in neonates.[21] We report the results of a multicenter prospective, randomized trial undertaken to ascertain the efficacy of a novel chlorhexidine-impregnated dressing for the prevention of catheter colonization and CRBSI in critically ill neonates.

METHODS

Subjects

This study was conducted in 6 level III neonatal intensive care units, 4 in university teaching hospitals (Children's Hospital of Wisconsin, Children's Hospital of Philadelphia, Boston Children's Hospital, and University of Massachusetts Memorial Medical Center) and in 2 community hospitals (St Joseph's Hospital and Sinai-Samaritan Medical Center). The study protocol was approved by the investigational review board at each site and informed consent was obtained from the parents of each participating neonate. Study units ranged from 16 to 50 beds. Neonates admitted to units who would likely require a CVC for at least 48 hours were eligible for the study.

Treatment

After obtaining parental consent, neonates were block randomized to 1 of 2 treatment groups. Computer-generated randomization codes developed by the study statistician were maintained by center pharmacists. Neonates randomized to the control skin care regimen had the catheter insertion site cleansed for at least 30 seconds with PI (Purdue Frederick, Norwalk, CT); after the PI was allowed to dry, the CVC was inserted and then dressed with a polyurethane dressing (Bioclusive Transparent Dressing, Johnson and Johnson Medical, Division of Ethicon, Inc, Arlington, TX). Neonates randomized to the novel chlorhexidine dressing (Biopatch Antimicrobial Dressing, Johnson and Johnson Medical) had the insertion site cleansed for at least 30 seconds with 70% isopropyl alcohol; after the alcohol dried, the catheter was inserted, and the insertion site was covered with the chlorhexidine dressing (1.9-cm diameter for infants ≤1500 g, 2.5 cm for infants >1500 g). The site was then dressed with the same type of polyurethane dressing used in the control group.

The chlorhexidine antiseptic dressing used in the trial is a hydrophilic polyurethane absorptive foam impregnated with 250 μg/mg of chlorhexidine gluconate. Chlorhexidine gluconate is released continuously onto the underlying skin surface over a 10-day period,[22] with the greatest concentrations released onto the skin during the first 3 days. At steady-state release, approximating zero-order kinetics, occurs from day 4 through day 10.

At 5 centers, percutaneously placed CVCs were placed by neonatologists or nurse practitioners. A dedicated group of staff nurses inserted catheters at the sixth center. Broviac (Evermed, Inc, Cranston, RI) catheters were used when surgically placed CVCs were required. All catheters were placed by personnel wearing a mask, hat, sterile gloves, and gown. Sterile drapes were used to protect the field during catheter insertion.

Percutaneously placed CVC dressings were changed every 7 days. Surgically placed Broviac catheter dressings were changed every 7 days in the chlorhexidine dressing group and twice weekly in the control group. At each dressing change in both treatment groups, the site was recleansed using the same antiseptic used at catheter placement. A new chlorhexidine-impregnated dressing was placed at the time of dressing changes in the antiseptic dressing group. Decisions to remove catheters were made independently by primary nurses and attending physicians. Blood cultures were obtained in neonates with signs of sepsis. Blood cultures were obtained at the discretion of the attending neonatologist.

Culture Techniques

At the time the catheter was removed, a 1 × 1-cm area of skin surrounding the catheter was swabbed with a sterile cotton swab saturated with Stuart's media (Becton Dickinson Microbiology Systems, Sparks, MD). The skin swab was cultured semiquantitatively.[23] Heavy skin colonization was defined as ≥50 colony-forming units (CFUs).[21] CVCs were removed aseptically and the last 5 cm of the catheter tip was cultured using the semiquantitative method of Maki et al.[24] Catheter hubs were also cultured quantitatively.[25] Skin and hub cultures were performed to help determine route of CRBSI or BSI. Specimens not immediately cultured were refrigerated at 4°C. All cultures were inoculated within 8 hours of catheter removal. Standard laboratory methods were used to identify microorganisms colonizing the skin, hub, and CVC tips.[26] Species of coagulase-negative S*taphylococcus* (CNS) grown from the blood, skin, catheter tip, and catheter hub of patients with CRBSI were identified using standard laboratory biochemical analyses and antibiotic susceptibility profiles.[27] In addition, restriction-fragment subtyping[28,29] of isolates of CNS was performed to confirm concordance between strains of CNS grown from the blood cultures and catheter tips or hubs in neonates with CNS BSIs.

Primary Outcomes

Catheter tip colonization was defined by a semiquantitative catheter colony count ≥15 CFUs.[24] Catheter colonization was considered a primary outcome because of its strong association with CRBSI and BSI without a source.[24,30,31]

BSI without a source was defined as: a positive blood culture during the time the catheter was in situ or within 24 hours of removal; clinical signs or symptoms of a BSI within 6 hours of the positive culture; antibiotic therapy for ≥7 days, and no other documented primary site of infection; catheter tip and hub cultures were either not colonized or colonized with organisms different from those grown from the blood.

Signs and symptoms of BSI, defined before initiation of the trial included: an increase or decrease in the white blood cell count by 3×10^3 per mm^2 or ≥0.15 immature neutrophils ratio on a complete blood count, new-onset apnea, glucose intolerance or hypoglycemia, metabolic acidosis, tachycardia or hypotension, mottled or ashen appearance with a normal hematocrit, new onset of feeding intolerance, lethargy, or fever. A BSI without a source was considered a primary outcome because CVCs are often left in place if the episode of BSI can be cleared with antibiotic therapy given through the CVC and, thus, concomitant catheter tip and catheter hub cultures cannot be obtained.

CRBSI was defined as a clinically relevant BSI without an identifiable primary source other than a CVC colonized by the same strain grown from blood cultures. Hub cultures, if obtained, were negative for the organism grown from the blood.

Data Collection and Analysis

Data were extracted prospectively from maternal and neonatal charts of neonates enrolled in the study by a study nurse at each study center and sent to the primary center (St Joseph's Hospital, Milwaukee, WI). Data collectors at each center were trained by the principal investigator (J.S.G.) before initiation of the trial. Data obtained on each patient included maternal and neonatal demographics, 24-hour and 7-day Score for Neonatal Acute Physiology scores,[32] day of life of catheterization, catheter type, medications, invasive therapies, other sites of infection while the catheter was in situ, anatomic location of the catheter, number of attempts to insert the catheter, person placing the catheter, and laboratory and clinical findings used to assess infection.

A primary study nurse (C.P.A.) communicated monthly with each center to deal with study questions, ensure study compliance, and timely data collection. After study center data collection sheets were checked for potential errors and missing items by the primary study nurse, data were double entered into a data set for analysis using *Statistical Analysis System 6.1 for the PC* (SAS Institute, Cary, NC).

All findings were based on intention-to-treat analyses. To preserve statistical independence, only 1 catheter per patient was enrolled in the trial. Baseline differences between treatment groups were analyzed using χ^2 analysis for dichotomous variables and Student's *t* test for continuous variables. Where appropriate, Wilcoxon rank sum test was used for nonparametric analyses. Differences in primary outcomes stratified by catheter type (percutaneous or Broviac) were compared using Mantel-Haenszel stratified analysis. Mantel-Haenszel common relative risk (RR) ratios and 95% confidence intervals (CIs) were determined for all primary outcomes using the PI treatment group as the control group. The log-rank test was used to compare length of time until the first episode of a BSI among treatment groups.

Sample size was calculated for a power of 80% and an α error of 0.05. It was estimated that to detect a 50% reduction in CRBSI risk, from 9%[33,34] in the PI group to 4.5% in the antimicrobial dressing group, would require approximately 490 neonates in each treatment group. To detect a 50% reduction in catheter colonization from approximately 20%[6,34] in PI patients to 10% in chlorhexidine dressing neonates would require approximately 200 neonates in each treatment group.

RESULTS

During the study period (June 1994 to August 1997), 919 neonates in study nurseries required CVC. Two hundred fourteen of these eligible infants were not enrolled in the trial because their parents refused consent, study members were not available for patient enrollment, or the primary physician did not permit study enrollment. Enrollment was halted after 705 neonates were enrolled in the trial (335 chlorhexidine dressing and 370 PI).

The 2 groups were very similar with respect to baseline demographic characteristics, measures of illness severity, patient characteristics at the time of catheter placement, and patient characteristics and treatment during the time CVCs were in situ (Table 1). A comparable small proportion of study patients' catheters were not cultured at removal because of accidental extrusion, contamination, or deviation from the study protocol (control 8%, chlorhexidine dressing 6%, $P = .40$). Among neonates whose CVCs were not cultured there was no difference between treatment groups with respect to baseline characteristics shown in Table 1 (data not shown).

TABLE 1. Baseline Characteristics of Treatment Groups Before Central Catheter Placement

Characteristic	Chlorhexidine Dressing ($n = 335$)	10% PI ($n = 370$)	*P* Value
Sex (male), %	59	55	.38
Race (white), %	62	65	.39
Inborn, %	64	68	.26
Prolonged rupture of membranes (≥24 h), %	15	17	.77
Gestational age (wk), mean ± SEM	30.9 ± 0.3	30.7 ± 0.2	.56
Birth weight (g), mean ± SEM	1640 ± 50	1620 ± 50	.80
Score for Neonatal Acute Physiology (24 h), mean ± SEM	12.2 ± 0.4	12.2 ± 0.4	.97
Score for Neonatal Acute Physiology (7 d), mean ± SEM	6.8 ± 0.3	6.7 ± 0.3	.91
At time of catheter placement			
Day of life of catheter placement, mean ± SEM	10.6 ± 1.3	8.3 ± 0.7	.14
Surgically placed, %	6	6	.65
Site of insertion, %			
Arm	62	64	
Leg	10	9	.89
Head/neck	26	25	
Other	2	2	
Attempts to place catheter, mean ± SEM	2.0 ± 0.08	1.8 ± 0.07	.11
While catheter in situ			
Days ventilated, mean ± SEM	6.2 ± 0.5	6.9 ± 0.5	.35
Days catheter in situ, mean ± SEM	17.7 ± 0.9	17.4 ± 0.6	.79
Medications given through catheter, %:			
Ampicillin	1.8 ± 0.2	1.8 ± 0.2	.86
Vancomycin	3.1 ± 0.3	3.1 ± 0.3	.95
Cephalosporin	1.3 ± 0.2	1.3 ± 0.2	.80
Hyperalimentation	13.9 ± 0.6	14.2 ± 0.6	.68
Lipid emulsion	14.5 ± 0.6	14.3 ± 0.6	.90
Number of times line entered/day, mean ± SEM	4.5 ± 0.1	4.4 ± 0.1	.62
Reasons for catheter removal, %:			
Not required	65	64	
Documented sepsis	4	5	
Suspected sepsis	12	9	.51
Infiltration/malfunction	12	15	
Accidental removal	3	1	
Other	4	6	
Catheter tip not cultured, %	6	8	.40

Catheter Tip Colonization

A comparison of primary outcomes is shown in Table 2. Three hundred fourteen (94%) of neonates randomized to the chlorhexidine dressing group had CVC tips cultured compared with 341 (92%) of 370 control neonates. Catheter tip colonization occurred in 129 (19.7%) of 655 neonates whose catheter tips were cultured. Neonates with Broviac catheters were more likely to have colonized catheter tips, 11 (31%) of 35, than neonates with percutaneously placed CVCs, 118 (19.0%) of 620, although the difference did not reach statistical significance ($P = .08$). After stratifying by catheter type, among neonates with cultured catheters, catheter tip colonization was significantly less frequent in chlorhexidine dressing neonates when compared with control neonates (15.0% vs 24.0%, RR: 0.6, CI: 0.5–0.9, Table 2). Among neonates with cultured catheters, incidence density of catheter tip colonization was also lower in antimicrobial dressing treatment group (8.5 vs 14.1 episodes per 1000 catheter-days, $P = .005$).

In subcohort analyses, differences in catheter tip colonization rates between the treatment groups were most evident for neonates whose catheters were in situ ≤14 days (11% vs 25%, $P = .0007$). There was no difference detected between treatment groups' colonization rates in neonates whose catheters were in situ >14 days (23% vs 20%, $P = .53$).

Heavy cutaneous colonization of the insertion site was more common with colonized catheter tips than with noncolonized catheter tips (62% vs 15%; $P = .001$) and in PI-treated neonates than in neonates in the chlorhexidine dressing group (28% vs 20%; $P = .02$). CNS was the most common organism cultured from colonized catheter tips (Table 3).

Catheter Tip-Related BSIs

CRBSI occurred in 23 (3.5%) of 655 of neonates whose catheters were cultured. No difference in CRBSI rates was detected between the treatment groups (Table 2). Incidence density of CRBSI per 1000 catheter days in the 2 groups was also similar (1.9 vs 2.2 episodes per 1000 catheter-days, $P = .60$). There was no difference in rates of CRBSI in subcohort analyses based on length of catheterization time (data not shown).

Organisms causing CRBSI are shown in Table 3. Distribution of organism responsible for CRBSI among treatment groups was similar. CNS was the most common organism responsible for CRBSI in each treatment group. Subtyping by pulsed field gel electrophoresis (PFGE) was done on CNS isolates from eight of 15 neonates who had a CNS CRBSI. Restriction-fragment DNA subtyping showed concordance in isolates from catheter tips and blood cultures in 3 of the 5 presumed CRBSI in the control group and 2 of 3 in the chlorhexidine dressing group.

BSI Without a Source

The first episode of cryptogenic BSI without an identifiable source was also evaluated on the premise that most primary BSIs in patients with CVCs originate from the intravascular device.[35] There was no detectable difference between BSI without a source between the 2 treatment groups (15.2% vs 14.3%, RR: 1.1, CI: 0.8–1.5; Table 2). Incidence density of BSI without a source was also similar (2.5 vs 2.6 episodes per 1000 catheter-days; $P = .70$).

Most of the episodes of BSI without a source were caused by CNS (Table 3). Distribution of organisms responsible for episodes of BSIs among treatment groups was similar.

There was also no difference between the treatment groups with respect to days to the first episode of BSI without a source (log-rank test; $P = .70$). There was no difference in rates of BSI without a source in subcohort analyses based on length of catheterization time (data not shown).

Thirteen episodes (5 chlorhexidine dressings) of BSI without a source occurred in neonates whose catheter tip and catheter hub were not cultured when the CVC was removed. Six of the 13 episodes occurred at least 10 days before removal of the catheter.

Adverse Reactions

During the first 15 months of the study, 7 (5.9%) of 118 of neonates randomized to the antimicrobial dressing developed a severe localized contact dermatitis under the chlorhexidine dressing; 2 additional neonates developed an area of pressure necrosis under the chlorhexidine dressing. Two reactions, which led to scar formation at the site, have been reported previously.[36] Mean gestational age of neonates with contact dermatitis was 24.5 weeks (range: 22.5–26.5

TABLE 2. Comparison of Overall and Catheter Specific Primary Outcome Rates in the Two Treatment Groups

Outcome	Chlorhexidine Dressing (n = 335)	10% PI (n = 370)	RR (95% CI)	P Value
Catheter colonization	15.0% (47/314)*	24.0% (82/341)*	0.6† (0.5–0.9)	.004
Broviac	33.3% (5/15)	30.0% (6/20)	1.1 (0.4–3.0)	.83
Percutaneous	14.1% (42/299)	23.7% (76/321)	0.6 (0.4–0.8)	.002
Catheter tip-related BSI	3.8% (12/314)*	3.2% (11/341)*	1.2† (0.5–2.7)	.65
Broviac	6.7% (1/15)	5.0% (1/20)	1.3 (0.1–20)	.99
Percutaneous	3.7% (11/297)	3.1% (10/323)	1.2 (0.5–2.8)	.68
BSIs without a source	15.2% (51/335)	14.3% (53/370)	1.1† (0.8–1.5)	.69
Broviac	26.3% (5/19)	37.5% (9/24)	0.7 (0.3–1.7)	.44
Percutaneous	14.6% (46/316)	12.7% (44/346)	1.1 (0.8–1.7)	.49

* Cultured catheters.

† Mantel-Haenszel analysis stratified by catheter type. PI was used as the reference group.

TABLE 3. Microorganisms Associated With Catheter Colonization, CRBSI, and BSI Without A Source in the Two Treatment Groups

Organism	Catheter-Related Colonization		CRBSI		BSI Without a Source	
	Chlorhexidine Dressing	PI*	Chlorhexidine Dressing	PI	Chlorhexidine Dressing	PI
CNS	39	72	6	9	36	33
Enterococcus	4	3	3	1	3	3
Staphylococcus aureus	2	2	2	0	3	2
Klebsiella	1	0	0	0	6	6
Enterobacter	0	0	1	0	0	2
Candida	0	5	0	1	3	7
Malassezia furfur	1	0	0	0	0	0

* 10% PI.

weeks) and mean birth weight was 720 g (range: 560–880 g). All CVCs had been placed on or before the eighth day of life.

After these reactions, criteria for study enrollment were changed. Infants <26 weeks gestational age were enrolled only if the CVC was inserted after the first week of life. After the change in the protocol, there were 12 more episodes of contact dermatitis from the chlorhexidine dressing among 217 neonates randomized to the antiseptic dressing group. During the entire study period, 15 (15%) of 98 neonates <1000 g and 4 (1.5%) of 237 neonates ≥1000 g randomized to the antiseptic dressing group developed a contact dermatitis under the dressing ($P < .0001$). Contact dermatitis did not occur in any control neonates.

DISCUSSION

Ill neonates often require prolonged access with a CVC. In this trial, disinfection of the insertion site with 70% alcohol before catheter placement followed by application of a novel chlorhexidine-impregnated dressing to the site reduced CVC tip colonization when compared with the use of 10% PI cutaneous antisepsis. There were no differences between the 2 site care regimens in rates of CRBSI and BSI without a source.

Chlorhexidine gluconate is a cationic biquanide that provides rapid antisepsis because of its broad spectrum germicidal activity against most neonatal pathogens.[37,38] Microbial resistance to chlorhexidine gluconate is rare.[39] Although chlorhexidine gluconate can be absorbed through the skin of neonates, minimal absorption has been detected in prospective trials.[40–43] Toxic effects have not been seen, and the agent has been well-tolerated as a skin antiseptic in term and preterm infants.[21,40–45] The novel chlorhexidine-impregnated dressing reduces heavy cutaneous colonization at the insertion site and, as a consequence, reduces catheter tip colonization. Cutaneous antisepsis with chlorhexidine gluconate has been proven to be more efficacious than antisepsis with PI in preventing peripheral intravenous catheter tip colonization in neonates,[21] and CVC tip colonization and CRBSI in adults.[16,17]

Differences in catheter tip colonization were most evident in neonates whose catheters were in situ ≤14 days. In adult patients, the risk of catheter tip colonization and CRBSI increases with catheterization time.[12,13,46] Adults with percutaneously placed CVCs that are in place >7 days are more likely to have CRBSI than those whose catheters remain in place <7 days.[12,13,15,24,47–49] Infection control strategies aimed at reducing catheter tract colonization may be less effective in preventing CRBSI in adults who require prolonged catheterization because the intraluminal route of infection—the catheter hub—becomes a more important portal of entry for microorganisms as catheterization time increases.[14,46,50]

Although the antiseptic dressing reduced catheter tip colonization when compared with PI antisepsis, there were no differences in CRBSI or BSI without a source between the 2 treatment groups. Study power may have precluded finding a difference in rates of CRBSI and BSI. The study was halted after 705 neonates were enrolled because of funding constraints and the low rate of CRBSI in the study population. Given the low rate of CRBSI in both groups, it is unlikely that significant differences between the 2 treatment groups would have been detected even if enrollment had continued to the initial goal of 980 neonates.

Hub contamination causing BSI is an important mechanism of CRBSI in neonates,[5] and likely is the most important mechanism of BSI in adult patients whose percutaneously placed CVCs remain in situ >7 days.[14,46,50] Because mean length of time CVCs were in situ exceeded 17 days in each treatment group, hub contamination not detected by the investigators and not preventable by cutaneous antisepsis, may have been responsible for a large proportion of the cases of BSI without a source.

As in other CVC studies,[6,11,12,16,51] CNS was the most common organism responsible for CVC tip colonization, CRBSI, and BSI without a source. CNS isolates grown from catheter tips and the blood of neonates with CRBSI were compared using biotypes and antibiotic sensitivity profiles. A more reliable means of determining isolate concordance using DNA restriction-fragment subtyping by PFGE was performed.[28] Eight of 15 episodes of CRBSI attributable to CNS could be evaluated with PFGE, 5 of 8 showed concordance between isolates grown from the catheter tip and blood cultures. Peripheral and CVCs are often colonized with multiple CNS species.[15,21,52] If multiple CNS isolates from catheter or hub cultures are not subjected to DNA typing, potential matches can be missed.[15,21] This may have

been the case in the 3 catheter-associated CNS BSIs that did not show DNA concordance.

The chlorhexidine-impregnated dressing can reduce the potential risk of hypothyroxemia in low birth weight infants by reducing exposure to iodine-based antiseptics.[18–20] However, local reactions at the site of the chlorhexidine dressing occurred in 5.7% of the antiseptic dressing-treated neonates. Most reactions occurred in neonates ≤28 weeks gestational age and ≤1000 g. Local contact dermatitis from the chlorhexidine dressing may limit its use in acutely ill low birth weight neonates.

In a recent randomized trial of adult patients with CVCs or arterial catheters, patients randomized to the novel chlorhexidine-impregnated dressing group had a significantly lower incidence of CRBSI than controls.[53] We believe that the chlorhexidine dressing was more effective in preventing CRBSI in this trial than in our trial because adult patients studied by Maki et al[53] had catheters in place for an average of 6 days, far less than the 17 days in our patients. The largest proportion of CRBSIs in the trial of Maki et al[53] derived from skin colonization rather than hub colonization and the antiseptic dressing was most effective for preventing CRBSIs by this route.

CONCLUSION

The use of alcohol for cutaneous antisepsis followed by the placement of a chlorhexidine-impregnated dressing over the insertion site of CVCs, left on for up to 7 days between dressing changes, provides protection against catheter tip colonization. CRBSI and BSI without a source rates were similar amount treatment groups. However, a substantial risk of contact dermatitis at the dressing site may limit its use in low birth weight infants in the first 2 weeks of life.

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Exposure to Lead Appears to Selectively Alter Metabolism of Cortical Gray Matter

ABSTRACT. ***Objective.*** **The effects of lead poisoning on the development of children have been examined primarily in the context of behavioral and neuropsychological studies. The purpose of this study was to examine the in vivo use of magnetic resonance spectroscopy (MRS) for the evaluation of the neurotoxic effects of lead on the nervous system. MRS has the ability to monitor brain metabolism by detecting a number of neurochemicals among which is *N*-acetylaspartate, a metabolite shown to decrease in processes that involve neuronal loss.**

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Methods. **In the present study we evaluated the metabolism of gray and white matter of frontal cortex using MRS in individuals with elevated blood lead levels and compared the results with those obtained on nonlead-exposed controls.**

Results. **Although all of the participants had normal MRI examinations of the brain, the lead-exposed individuals exhibited a significant reduction in the N-acetylaspartate/creatine and phosphocreatine ratios in frontal gray matter compared with the nonlead-exposed controls.**

Conclusions. **The findings of this study suggest that lead has an effect on brain metabolites as detected by MRS in vivo. More specifically, we have found statistically significant reduced levels of brain metabolites in gray but not white matter in lead-exposed individuals. These results imply that MRS is able to detect metabolic abnormalities in individuals with lead poisoning.** ***Pediatrics*** **2001;107:1437–1443;** ***lead, neuronal loss, proton MRS, brain cortex.***

ABBREVIATIONS. MRS, magnetic resonance spectroscopy; MRI, magnetic resonance imaging; NAA, N-acetylaspartate; Cr, creatine and phosphocreatine; Cho, choline-containing compounds; 3DSPGR, 3-dimensional spoiled GRASS, gradient acquired in the steady state; TR, repetition time; TE, echo time; VOI, voxel of interest; 1D, 1-dimensional; mI, myoinositol.

During the past several years there has been a growing interest in the effects of exposure to lead on the developing nervous system as well as the mechanisms by which lead disrupts brain function in children. The effects of elevated blood lead levels on the development of children have been examined primarily in the context of behavioral and neuropsychological evaluations, as debate continues on the effects low to moderate lead levels (10–40 μg/dL) have on general behavioral and cognitive functioning. One of the most consistently reported impairments associated with lead exposure at levels as low as 25 μg/dL involves its negative impact on general intellectual functioning.[1–6] A number of centers have conducted longitudinal as well as cross-sectional studies, reporting inverse relations between IQ and dentine or blood lead levels, as well as diminished academic achievement and psychomotor development following low to moderate lead levels.[1–6] Recent studies have been particularly careful at adjusting for confounding variables such as parental intelligence, socioeconomic status, education and home environment, with a general finding that the detrimental effect on IQ remained significant.[6–9] Other neuropsychological impairments reported in conjunction with lead exposure, although less robust, include impaired memory and learning,[10] impaired perceptual integration,[11] slower reaction time[12,13] impaired motor development,[3] as well as visual–motor integration and serial choice reaction performance.[14]

A number of investigators challenged the findings of lowered intellectual and cognitive ability in children, particularly at lower lead levels (10–25 μg/dL), claiming other variables, such as social class, family size and marital relationship,[15] maternal education, quality of care, prenatal and postnatal stressors,[16] and iron deficiency[17] may have caused a detrimental effect on the cognitive functioning of these children. Difficulties in attributing a causal relationship between elevated lead levels and cognitive deterioration also stem from the fact that most studies used epidemiologic and statistical methods to establish the effects of lead, with differences in sample selection, peak and duration of lead levels, differences in measurement of lead levels (blood, dentine, bone) and differences in procedures and statistical methods. Because of these limitations we suggest that there is a strong need for a more objective method with which to evaluate of the effects of lead on the development of children.

Little is known regarding the effects of lead on brain metabolism in vivo, and on structural and functional correlates of brain function. In the human brain, magnetic resonance spectroscopy (MRS) provides a noninvasive risk-free method with which to monitor the biochemistry of acute and chronic stages of disease.[18–20] The development of spatial localized spectroscopic methods that sample the relative levels of mobile metabolites from a volume of tissue defined from a magnetic resonance image has provided a basis for integrating the biochemical information obtained by MRS with the anatomic and pathologic information obtained from magnetic resonance imaging (MRI). This combination of metabolic and anatomic information affords a new means of understanding the origins and time course of progression in variety of diseases. In the brain, MRS has gained widespread acceptance as a method for assessing both neuronal viability as well as demyelination. This acceptance is based on the fact that one of the metabolites identified in proton spectra of the brain, N-acetylaspartate (NAA), is largely confined to neurons,[21,22] and has been proposed as a neuronal marker. In the cortex, NAA is located in neuronal cell bodies whereas in the white matter it is located largely in axons. A decrease in NAA has been proposed as an indicator of neuronal and axonal damage and loss.[23] Proton MRS has been used to study neurodegenerative processes where decreases in NAA have been shown as common findings in patients with Alzheimer's disease,[24,25] Parkinson's disease,[26] and Huntington's disease.[27] In practice, the decrease in NAA is measured relative to the level of creatine (Cr), a stable metabolite whose level is constant following neuronal loss. As an example, van der Knapp et al[28] have demonstrated that increased cerebral atrophy was accompanied by lower ratios of NAA to creatine in patients with demyelination disorders. In children, Kimura et al[29] reported abnormally low NAA/Cr ratios in neurologically delayed infants as compared with children with no known developmental delays. Grodd et al[30] reported a marked decrease of NAA in children with focal or generalized demyelination. Because there is evidence showing reduced NAA peaks in disease processes involving intellectual deterioration, it is reasonable to hypothesize a decrease in NAA in the brain of children and adults with clinical evidence of lead neurotoxicity. Lopez-Villegas et al[31] developed a technique to obtain metabolic information differentially from gray and white matter using proton MRS.

They reported that spectra from frontal gray matter showed choline-containing compounds (Cho)/Cr and NAA/Cr ratios significantly lower than those from white matter in healthy young adults. They also reported lower Cho and higher Cr content in gray matter. Trope et al[32] used this technique to examine a child with elevated blood lead levels, as compared with a first cousin with no lead exposure, as a first step in determining whether this method might serve as a new technique for evaluating the effects lead has on the central nervous system. This study demonstrated that the lead-exposed boy showed a significant alteration in brain metabolites, with a reduction in NAA/Cr ratio for both gray and white matter compared with his nonlead-exposed cousin. Neuropsychological evaluation demonstrated areas of impairment in the lead-exposed child, including difficulties in academic skills of reading, writing and arithmetic, deficient linguistic skills and attention. By contrast, neuropsychological examination of the cousin was within normal limits. As a next step, we have used the methods described previously by Trope et al[32] and Lopez-Villegas et al[31] in the present study to examine proton spectra obtained from individuals with elevated lead levels and compare them to spectra obtained from healthy, nonexposed individuals.

METHODS

Subjects

Sixteen individuals with documented blood lead levels ranging from 23 to 65 μg/dL (mean lead = 39.93 μg/dL, standard deviation [SD] = 13.39) were included in the subject group. There were 5 boys and 11 girls. The mean age at the time of testing was 8 years, 9 months (range: 4–21 years, SD = 4.36). All of the individuals in the subject group came to medical attention before the age of 5 years (range: 10–60 months, mean = 27.68 months, SD = 13.82). The control group consisted of 5 individuals (siblings/cousins of Subject Group) whose reported lead levels as indicated in their pediatrician records never exceeded 10 μg/dL. There were 3 boys and 2 girls. The mean age at the time of testing was 8 years, 6 months (range: 6–11 years, SD = 1.62). All of the individuals were evaluated using the MRI and MRS methods at the University of Pennsylvania Medical Center. The only significant difference between the 2 groups is the exposure to lead in the subject but not the control group. None of the participants was known to have neurologic deficits.

Table 1 summarizes subject characteristics for the two groups.

Procedures

Informed consent was obtained from the parents/guardians of all participants. All of the magnetic resonance studies were performed at the Hospital of the University of Pennsylvania in Philadelphia, on a 1.5T Signa system (GE Medical Systems, Milwaukee, WI). MRI was performed with a standard quadrature head coil. The children were not sedated and participated willingly.

After conventional MRI, the standard quadrature head coil was replaced by a 3-inch surface coil that was positioned over the left frontal region immediately supraciliary. A sagittal localizer was obtained followed by axial 3-dimensional spoiled gradient acquired in the steady state (GRASS) (3D-SPGR) images (256 × 256 matrix, 8-cm field of view, repetition time [TR] 22.4 ms, echo time [TE] 7.5 ms, 45 flip angle, 2 acquisitions, 1.5-mm thickness and 28 slices). The 3D-SPGR images provide high contrast between gray and white matter and were used to choose the voxel of interest (VOI) for the spectroscopic study. Immediately after high resolution MRI, 1-dimensional (1D)-proton spectra were obtained with the stimulated-echo acquisition mode for localization. Water suppression was achieved by using 3 chemical shift-selective radiofrequency pulses followed by a dephasing gradient applied on each of the 3 axes. The sequence parameters included the following: 19-cm field of view, spectral bandwidth 2500 Hz, 32 phase-encoding steps, TR 2000 ms, TE 31 ms, mixing time 10.6 ms, 2048 complex points, 8-step phase cycling and 16 acquisitions. We selected a VOI of 30–40 × 6 × 10 mm including cortical gray and white matter. Spectra from contiguous 6 × 6 × 10-mm voxels were obtained from the VOI by 1D phase-encoding. Cortical sulci were included in the VOI in all cases. Because the thickness of cortical gray matter is about 3 mm[33] the inclusion of cortical sulci in the VOI guarantee about 6 mm-thickness of gray matter. To avoid partial volume effects, the spatial distribution of gray and white matter included in the VOI has been checked to be relatively invariant in at least 6 of the MR images (1.5 mm-contiguous slices) that contributed to the MRS slice (10 mm-thickness). Scalp and marrow were excluded from the VOI to prevent contamination from lipids. Gradient shimming on the VOI and optimization of

TABLE 1. Characteristics of Individuals Undergoing MRS

Participant	Gender	Highest Pb Level (μg/dL)	Age at Highest Pb	Age at MRS
Pb1	M	54	3 y	10 y
Pb2	F	30	10 mo	6 y
Pb3	F	45	2 y	17 y
Pb4	M	65	1 y	6 y
Pb5	F	33	1 y 11 mo	7 y
Pb6	F	56	11 mo	21 y
Pb7	F	43	1 y	13 y
Pb8	F	51	2 y	6 y
Pb9	M	23	4 y	9 y
Pb10	F	25	3 y	8 y
Pb11	F	25	5 y	9 y
Pb12	M	24	1 y 6 mo	4 y
Pb13	F	57	2 y 6 mo	7 y
Pb14	F	26	3 y 5 mo	6 y
Pb15	F	43	2 y 10 mo	8 y
Pb16	M	39	2 y	6 y
C1, cousin of Pb1	M	—	—	9 y
C2, sister of Pb2	F	—	—	9 y
C3, sister of Pb4, Pb5	F	—	—	11 y
C4, brother of Pb*	M	—	—	8 y
C5, brother of Pb15	M	—	—	6 y

Pb indicates lead; C, control; M, male; F, female.
* C4's brother who had high lead levels was not a participant.

solvent suppression were performed before the start of the acquisition. The spectral acquisition time was 17 minutes and the total examination time, including MRI and MRS studies, was ~55 minutes. The MR procedure was well-tolerated by all subjects.

The spectral processing was performed with ProNMR (Softpulse Software, Guelph, Ontario, Canada) using zero filling to 4K data points, 1.5 Hz line broadening applied in the time domain, 2-dimensional Fourier transformation, and zero-order phase. Areas under the peaks were determined using a Marquardt fitting routine to Lorentzian line shapes in the frequency domain and peak area ratios were calculated. MRI and MRS were evaluated blind to the status of the subjects. The MRI films were reviewed by an attending radiologist at the Department of Radiology of the University of Pennsylvania.

The peak assignments were made based on the published literature and the chemical shifts were determined using NAA as a chemical shift standard. The following resonances were assigned: NAA, 2.0 ppm, 2.6 ppm; Cr, 3.0 ppm, 3.9 ppm; Cho, 3.2 ppm; and myoinositol (mI), 3.5 ppm. The region between 2.1 and 2.5 ppm contains peaks from glutamate, glutamine, gamma-amino butyric acid and NAA. These peaks could not be resolved because of the overlap of resonances. Other peaks from glutamate and glutamine are contained in the region between 3.6 and 3.8 ppm. Residual lipid signals were identified in the region between 0.5 and 1.5 ppm. The peak at 2.01 ppm and 3.0 ppm were used for the quantification of NAA and Cr, respectively.

The mean and SD for the mean values determined from each individual in each of the 2 groups were calculated for each metabolite ratio. Statistical comparisons between metabolite ratios from gray and white matter were made using 2-tailed unpaired Student's *t* test for each of the metabolite shifts using *Statview Version 5.01* (SAS Institute, Cary, NC).

RESULTS

All of the MRI examinations were reported normal with no evidence of structural abnormalities for any of the participants.

A representative study showing the VOI prescription in the left prefrontal lobe along with the stack-plot of proton spectra from adjacent voxels obtained by 1D phase-encoding is shown in Fig 1.

The signal-to-noise ratio from spectra coming from the margins of the VOI was lower compared with intermediate voxels probably resulting from partial volume effects. Typical individual spectra from frontal gray matter and white matter with the principal metabolites identified are shown in Fig 2 for the subject group and the control group.

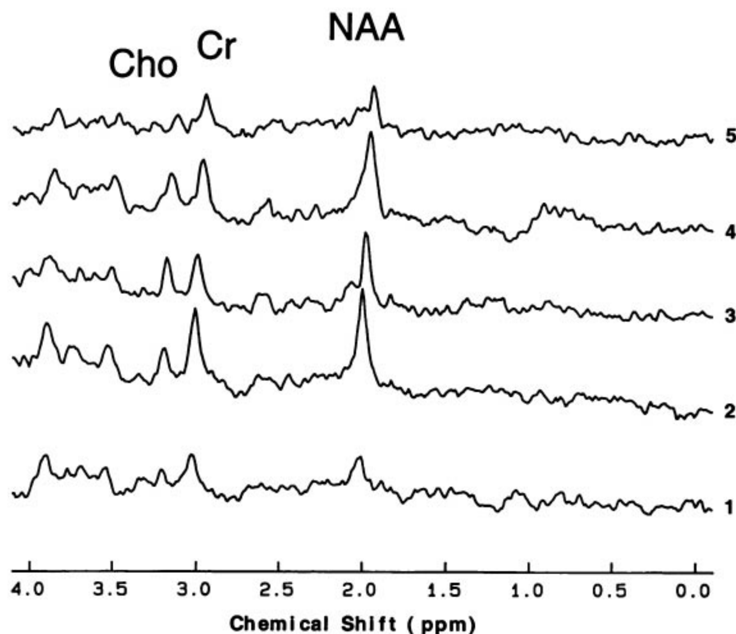
The results of an analysis of peak area ratios for gray and white matter are summarized in Table 2 for the 2 groups.

The NAA/Cr ratio in gray matter was significantly lower for the subject group as compared with the control group (unpaired t test, $P = .0345$). In contrast, none of the other ratios in gray matter or the ratios obtained in white matter (see Table 2) was statistically significantly different.

Figure 3 shows a plot of the variation in the NAA/Cr ratio in gray matter with the highest lead level in the blood of each individual in the subject group. The best-fit linear regression showed an r value of 0.05 indicating that there is no correlation between these 2 variables.

DISCUSSION

The present study examined in vivo metabolic differences in the brains of individuals exposed to lead as compared with healthy controls. The individuals who participated in this study and included in the control group were relatives (siblings, cousins) of the individuals comprising the subject group. Both subjects and controls in this study came from the same socioeconomic and home environments, thus eliminating a number of variables usually regarded as factors in behavioral lead studies. The only significant difference between them was elevated lead levels in the subject but not in the control group. The MRS study of the healthy, nonlead-exposed individuals resulted in spectra entirely consistent with the spectral pattern reported in previous studies for healthy individuals[31,34,35] documenting the levels of these metabolites in the normal adult brain, as well



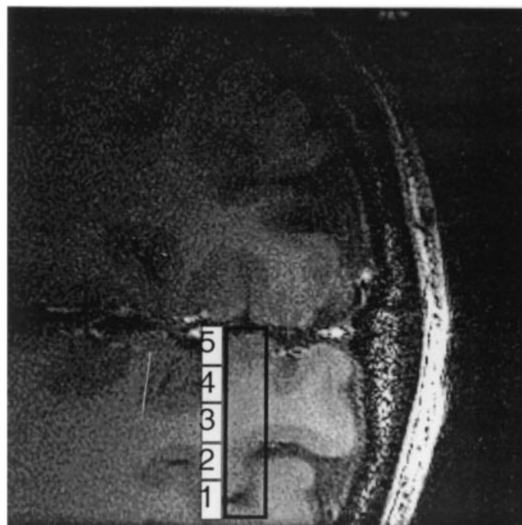


Fig 1. A representative study showing the VOI prescription in the left prefrontal lobe along with the stack-plot of proton spectra from adjacent voxels obtained by 1D phase-encoding.

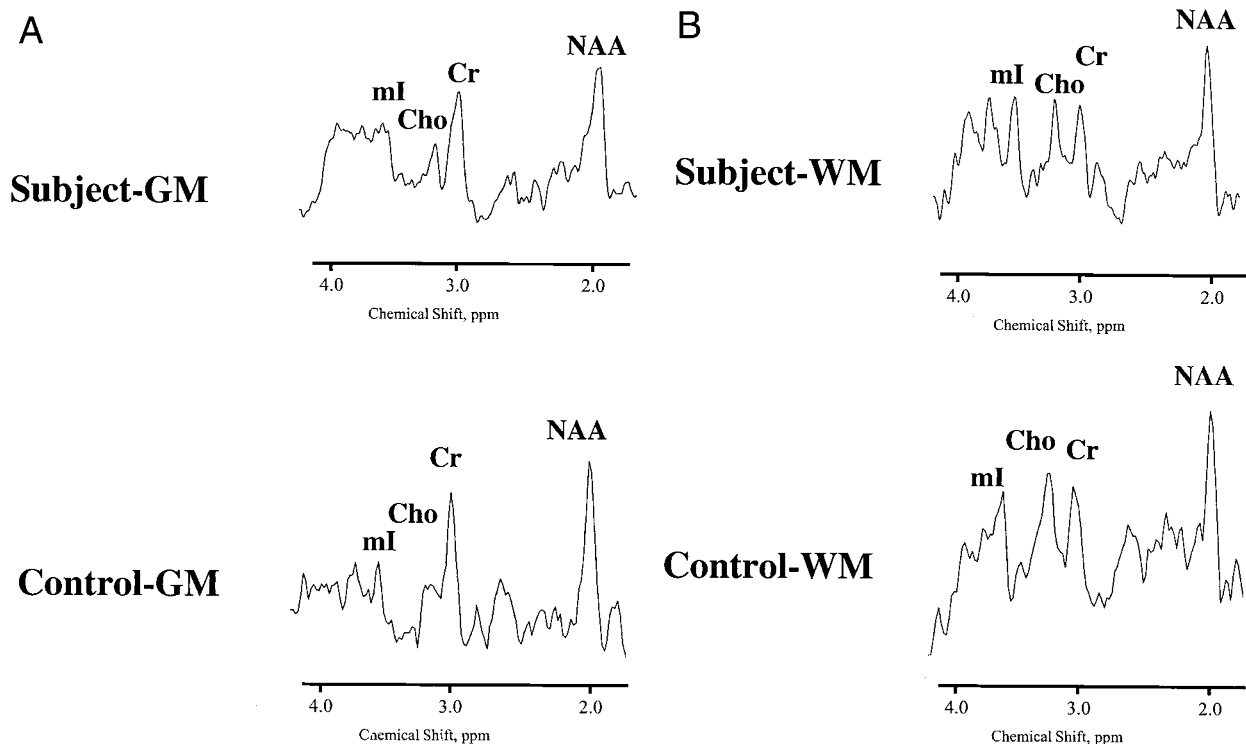


Fig 2. A, Spectra from frontal gray matter with the principal metabolites identified for a subject (top) with high blood lead levels and normal control (bottom). B, Spectra from frontal white matter with the principal metabolites identified for a subject with high blood lead levels (top) and normal control (bottom).

TABLE 2. A Summary of the Means and Standard Deviations in the Metabolite Ratios Obtained for Controls and Subjects Exposed to Lead

Metabolite Ratio	Subjects (n = 16)	Controls (n = 5)	P Value*
NAA/Cr GM	1.10 ± 0.19	1.30 ± 0.13	.035
Cho/Cr GM	0.38 ± 0.16	0.45 ± 0.08	.35
mI/Cr GM	0.42 ± 0.12	0.46 ± 0.09	.50
NAA/Cr WM	1.28 ± 0.25	1.40 ± 0.27	.34
Cho/Cr WM	0.80 ± 0.17	0.83 ± 0.15	.70
mI/Cr WM	0.58 ± 0.15	0.64 ± 0.07	.47

* The P value was computed using an unpaired Student's t test.

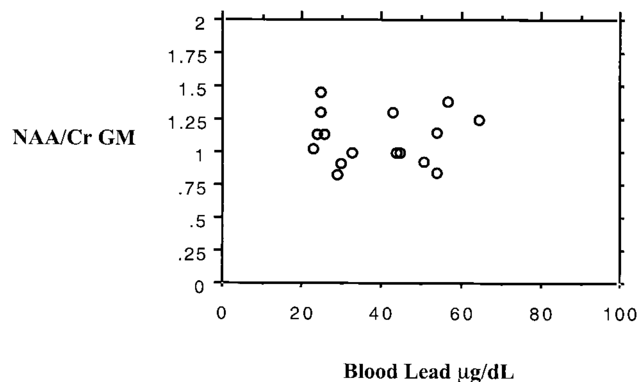


Fig 3. A plot of the variation in the NAA/Cr ratio in gray matter with the highest lead level in the blood of each individual in the Subject group.

as the estimated metabolite concentrations. These studies demonstrated that spectra from frontal gray matter are characterized by a lower Cho/Cr ratio and NAA/Cr ratio as compared with those obtained from white matter. Using the same technique as used in the present study, Lopez-Villegas et al[31] also reported that in healthy young adults, there were no differences in mI/Cr ratios for gray and white matter. The spectra obtained from individuals in the control group showed the same pattern of metabolite ratios. It is appropriate to compare the spectra obtained from the children who participated in the present study with those obtained in the study of Lopez-Villegas et al of young adults, because it has been reported that the levels of metabolites in the brains of children reach their adult levels by the age of 3 years.[29]

In contrast to the spectra obtained from the control group, the spectra obtained from the lead-exposed individuals showed lower NAA/Cr ratios for gray matter. Previous studies have linked lowered NAA/Cr ratios to neuronal loss and decline in intellectual functioning.[21,24,28,29] Therefore, the lowered NAA/Cr ratio in the lead-exposed individuals is suggestive of neuronal loss in the region examined. There is no indication in any of the individuals examined of any developmental history for any event other than his/her lead exposure. It is therefore possible that the reduction in NAA/Cr ratio may be a direct result of his/her elevated lead levels. Alternatively, in the absence of premorbid MRS measurements, the loss in NAA/Cr in these subjects may reflect a selective vulnerability to lead exposure rather than a direct effect of lead neurotoxicity.

It is of note that we did not find a significant correlation between the highest reported blood lead level and the NAA/Cr ratio in frontal gray matter.

The reason for this is unclear. We acknowledge the possibility that the highest blood lead level reported in this study may not reflect the highest exposure to lead. It is possible that the NAA/Cr ratio may reflect other aspects of lead exposure such as the duration of exposure or time since the highest level of exposure. These possibilities could be tested in a cohort with more detailed lead exposure data.

The relationship between NAA/Cr and neuronal viability was demonstrated by Cheng and coworkers.[36] This group found an inverse linear correlation between surviving pyramidal neurons per unit area on microscopic examination with the NAA/Cr ratio determined by MRS in brain samples obtained from patients with Pick disease. To our knowledge, this is the first study to demonstrate a histopathological validation of MRS decreases in NAA.

In our previous case study, we found that the NAA/Cr ratio was lower in white matter as well as gray matter. In the larger group, there was a trend for a lower NAA/Cr ratio in white matter in the lead-exposed group, which did not reach statistical significance. It is possible that this trend might become significant when a larger cohort is studied. We were able to obtain high quality spectra from voxels as small as 0.36 cm^3 at 1.5T. The spatial resolution used in the present study is sufficient to obtain spectra from voxels almost exclusively comprising gray matter. The 1D phase-encoding approach used has the advantage of obtaining several spectra simultaneously in a relatively short period of time. The present study has allowed us to examine the spectroscopic patterns of frontal gray and white matter after lead exposure relative to the normal pattern seen in healthy children and adults. This provides opportunities for the investigation of the brain of children and adults with lead poisoning to determine more precisely the effects of lead on the brain, and to examine any regional metabolic abnormalities.

We have demonstrated differences in metabolites in regions in the frontal lobe, which are particularly relevant, as the signature effects of lead neurotoxicity involve functions of the prefrontal and frontal lobes such as attention and executive functions, social-behavioral conduct, and impulse control. Additional studies confirming these differences as well as sampling different regions in the brain will be helpful in establishing whether lead affects specific brain regions or, alternatively, affects the brain more diffusely. The potential for this technique in determining the specific effects of lead on the central nervous system appears feasible and significant.

CONCLUSION

Both subjects and controls in this study came from the same socioeconomic and home environments, thus eliminating a number of variables usually regarded as cofounders in behavioral lead studies. Although MRI examinations were normal for both groups, MRS metabolites in the lead-exposed subjects were significantly reduced as compared with the controls, suggesting interference with neuronal functioning after lead exposure. More specifically, this study showed that individuals with a history of moderate elevations in blood lead levels show reduced NAA/Cr ratios in frontal gray matter. It was demonstrated that MRS can be used as a technique to measure brain metabolites in vivo, and provides us with a measure of neuronal viability. This finding is important in relation to children exposed to lead, as it enables us to evaluate the degree of neuronal loss.

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Tubulointerstitial Nephritis and Uveitis Syndrome With Autoantibody Directed to Renal Tubular Cells

ABSTRACT. The pathogenesis of tubulointerstitial nephritis and uveitis (TINU) syndrome remains unknown, but T cell-mediated immune response has been postulated to play a role. On the other hand, TINU syndrome is characterized by hypergammaglobulinemia and high serum immunoglobulin G (IgG) levels, suggesting an involvement of humoral immunity. We describe a case of TINU syndrome in a 13-year-old girl with multiple tubular dysfunctions including renal glucosuria, tubular proteinuria, phosphaturia, uricosuria, and concentrating and acidifying defect. IgG antibody from her serum was reactive against 125-kDa human kidney protein. Immunofluorescence study using mouse kidney revealed that the antibody was against cortical renal tubular cells. The antibody disappeared as the renal symptoms resolved. We suggest that IgG antibody may contribute to tubular dysfunction in some patients with TINU syndrome. *Pediatrics* 2001;107:1443–1446; *tubulointerstitial nephritis, uveitis, antibody, kidney, renal tubule, hypergammaglobulinemia.*

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ABBREVIATIONS. TINU, tubulointerstitial nephritis and uveitis; IgG, immunoglobulin G; PBS, phosphate-buffered saline; SDS-PAGE, sodium dodecyl sulfate polyacrylamide gel electrophoresis.

Tubulointerstitial nephritis and uveitis (TINU) syndrome was originally described by Dobrin et al[1] in 1975. Since then, more than 50 patients have been reported. Although its cause remains unknown, evidence suggests that TINU syndrome is a cell-mediated immune disease. On the other hand, TINU syndrome has sometimes been associated with hypergammaglobulinemia and elevated serum immunoglobulin (IgG) levels, suggesting a humoral-mediated immune response.[2] Recently, antitubular cell antibodies have been demonstrated in patients with Sjögren syndrome and systemic lupus erythematosus who have renal tubular acidosis.[3,4] In the present study, we examined whether a similar antibody may be detected in a patient with TINU syndrome.

CASE REPORT

A 13-year-old girl presented with photophobia. A diagnosis of bilateral anterior nongranulomatous uveitis was made. Because her serum creatinine was 1.1 mg/dL, and glucosuria and proteinuria were noted, she was referred to our hospital. She reported fatigue, abdominal pain, and a 1.8-kg weight loss over the previous month. She denied having fever, joint pain, nocturia, preceding infection, or medication use. Her medical history was unremarkable. On examination, her height was 157 cm, weight was 42.2 kg, and blood pressure was 116/74 mm Hg. Physical examination was unremarkable.

Laboratory findings showed 10.4 g/dL hemoglobin, 31.6% hematocrit, and white blood cell count 5900/mm^3 with 76.7% neutrophils, 15.2% lymphocytes, and 2.7% eosinophils. The erythrocyte sedimentation rate was 73 mm in the first hour. C-reactive protein was 1.61 mg/dL; serum creatinine, 0.9 mg/dL; blood urea nitrogen, 11.4 mg/dL; uric acid, 1.8 mg/dL; total protein, 8.5 g/dL; albumin, 4.6 mg/dL; sodium, 140 mEq/L; potassium, 3.3 mEq/L; chloride, 103 mEq/L; calcium, 9.4 mg/dL; and phosphorus, 2.6 mg/dL. Fasting blood glucose was 96 mg/dL. Venous blood gas analysis revealed pH 7.338, P_{CO_2} 46.1 mmHg, and HCO_3^- 25.0 mEq/L. Serum gammaglobulin was 1600 mg/dL (normal: 800–1500 mg/dL), and IgG 1860 mg/dL (normal: 635–1770 mg/dL), immunoglobulin A, 395 (normal: 190–340 mg/dL), and immunoglobulin M 237 mg/dL (normal: 37–154 mg/dL). Serum complement levels were normal, and antinuclear antibody, anti-DNA antibody, rheumatoid factor, antineutrophil cytoplasmic antibody, circulating immune complexes, and Coombs' test were negative. Anti-streptolysin O titer was <57 IU/mL. Antibodies for hepatitis B and C, toxoplasma, *Treponema pallidum*, and cytomegalovirus were not detected. Tuberculin test was negative. Serum angiotensin converting enzyme was normal.

Urinalysis showed a pH of 7.5, specific gravity 1.010, 2+ glucosuria (7.5 g/d) and 2+ proteinuria (0.93 g/d); there were 21 to 50 white blood cells/high-powered field and 3 to 5 red blood cells/high-powered field with sparse granular casts. Thirty to forty percent of urine white blood cells were eosinophils as detected by Hansel's stain. Urinary β2-microglobulin was 77.8 mg/L (normal: $<$1.0 mg/L), α1-microglobulin 146 mg/L (normal: $<$10 mg/L), and N-acetyl-β-D-glucosaminidase/creatinine 41.4 (normal: $<$5). Urine protein electrophoresis showed a tubular pattern. Creatinine clearance was 108 mL/min per 1.73 m^2. Fractional excretion of sodium was 0.59% (normal: $<$2%), fractional excretion of potassium, 15.6% (normal: 4%–16%); urinary calcium excretion, 0.5 mg/kg/day; fractional excretion of uric acid, 36.9% (normal: 7.6 $\pm$ 3.75%); and the percentage of tubular reabsorption of phosphate, 69.7%. There was no aminoaciduria. Urine osmolality after 14-hour water deprivation was 469 mOsm/L. Furosemide 1 mg/kg was administered intravenously, and urine pH was followed until 4 hours. The minimal urine pH was 5.8 at 2 hours. Chest radiograph and cardiac echogram were normal. Gallium scintigraphy revealed uptake in the eyes and kidneys.

A percutaneous renal biopsy was consistent with acute interstitial nephritis. The edematous interstitium was infiltrated with lymphocytes, plasma cells, and eosinophils (Fig 1). There were breaks in the tubular basement membranes with infiltration of the tubular epithelial cell layer by lymphocytes and plasma cells. Glomeruli were normal. Immunofluorescence studies showed no immunoglobulins, fibrinogen, or complement.

She was treated with topical steroids. Serum creatinine normalized (0.6 mg/dL) spontaneously, and anemia, blood abnormalities, and most of the urine abnormalities disappeared within 6 months. Urine concentration defect and β2-microglobulinuria resolved after 1 year.

METHODS

Samples

Sera from the reported patient (acute phase and after complete recovery), a patient with sarcoidosis who had similar multiple tubular dysfunctions and hypergammaglobulinemia as the reported case, and a healthy participant were tested.

Purification of IgG

Eight hundred μL of serum was incubated with 200 μL antihuman IgG agarose (Sigma, St Louis, MO) at 4°C overnight. After washing with phosphate buffered saline (PBS) 3 times, half of the agarose was used for assay for antibody binding kidney proteins as described below. IgG was eluted from the remaining agarose with 300 μL eluting buffer (0.1 M glycine, 0.15 M NaCl, pH 2.4) for immunohistochemical studies. Eluted fractions were immediately neutralized by the addition of sodium hydroxide. Purification of IgG was confirmed by sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE).

Assay for Antibody Binding Kidney Proteins

Normal kidney tissue was obtained from a kidney explanted because of tumor nephrectomy. Tissue was homogenized in solubilization buffer containing 20 mmol/L HEPES (pH 7.2), 1% Triton-100, 10% glycerol, 20 mmol/L sodium fluoride, 1 mmol/L sodium orthovanadate, 1 mmol/L phenylmethylsulfonyl fluoride, 10 μg/mL aprotinin, and 10 μg/mL leupeptin. Insoluble material was removed by centrifugation (10 500 $\times$ g, 10 minutes). The protein content in lysate was measured with a DC protein assay (Bio-Rad Laboratories, Tokyo, Japan). One mg kidney lysate was incubated with 100 μL IgG bound agarose at 4°C overnight. After washing with PBS 3 times, 25 μL Laemmli's buffer was added and boiled. Bound proteins were separated by SDS-PAGE and visualized by Coomassie blue staining.

Immunohistochemistry

Mouse cryostat kidney sections were incubated with IgGs. For the secondary antibody, affinity purified goat antihuman IgG (H & L) (American Qualex, San Clemente, CA) was used. The fluorescence signal of labeled specimens was observed first with a Zeiss Axivert microscope and then analyzed by a laser confocal microscope (Zeiss LSM 510, Zeiss, Germany). Digitized images were produced with a Mirus Film Printer Galleria (Mirus Industries Corporation, Santa Clara, CA) using Raster Plus 95 software (version 1.01) (Graphx, Inc, Woburn, MA).

RESULTS

Figure 2 shows SDS-PAGE gels showing kidney proteins reacting with IgGs. Antibody recognizing approximately 125-kDa protein was present in IgG from the reported patient, but was barely detectable in a healthy participant and a patient with sarcoidosis (Fig 2A). This antibody disappeared after recovery (Fig 2B).

Localization of this antibody in the kidney was detected by indirect immunofluorescence with the use of mouse renal tissue. IgG from the reported patient reacted with renal tubular cells in the cortex but not with those in the medulla (Fig 3A, B). The staining was diffuse in the proximal tubules and cortical distal tubules. Secondary antibody alone showed yellow staining attributable to autofluorescence (Fig 3C). The intracellular localization of the antigen was in the cytoplasma. IgG from a patient with sarcoidosis or a healthy participant did not show any specific staining (data not shown).

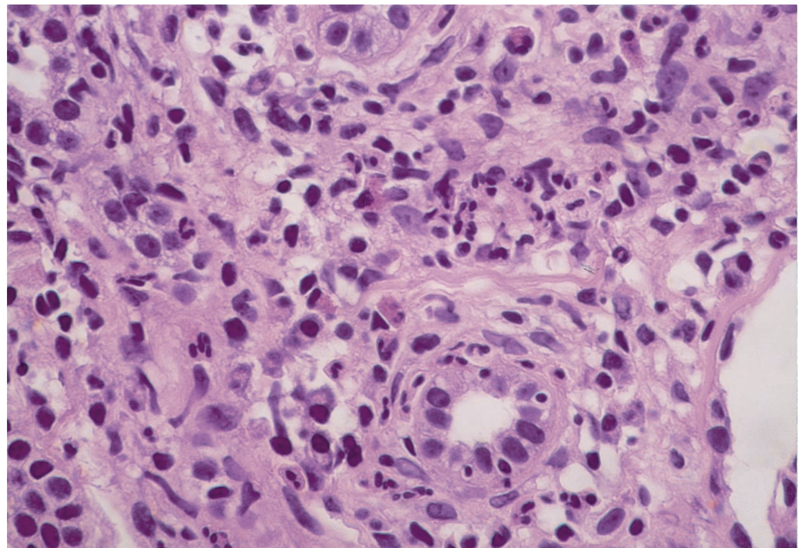

Fig 1. Renal biopsy specimen showing infiltration of lymphocytes, plasma cells, and eosinophils in the interstitium (magnification $\times$ 400).

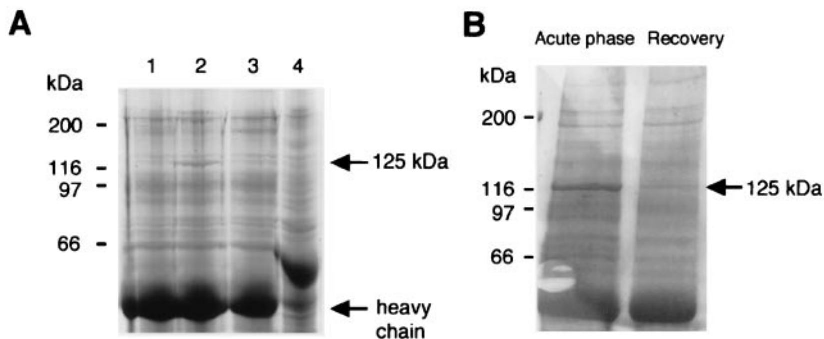

Fig 2. Kidney proteins reacting with IgGs. Patient serum was incubated with anti-human IgG agarose. After washing, 1 mg kidney lysate was incubated with 100 μL IgG-bound agarose. Bound proteins were separated by SDS-PAGE and visualized by Coomassie blue staining. A, Antibody recognizing approximately 125-kDa protein was present in IgG from the reported patient (lane 2), but was barely detectable in a healthy subject (lane 3) and a patient with sarcoidosis (lane 1). Lane 4 is kidney lysate. B, The antibody reacting with 125-kDa protein disappeared after recovery.

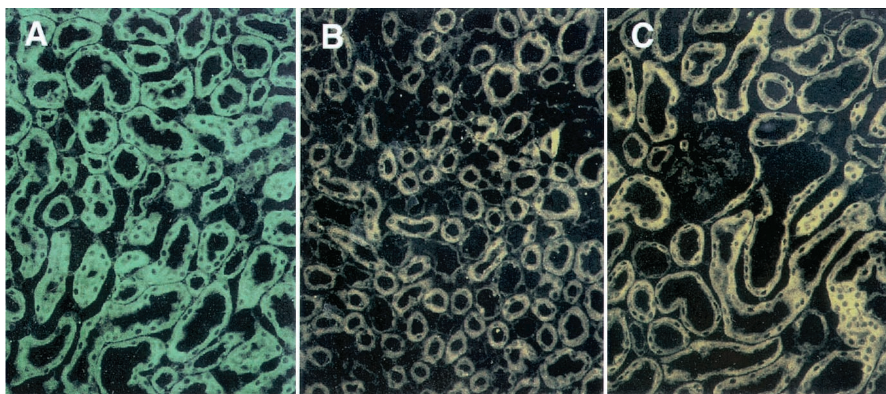

Fig 3. Immunohistochemical studies. IgG from the reported patient reacted with renal tubular cells in the cortex (A) but not with those in the medulla (B). Secondary antibody alone showed yellow staining due to autofluorescence (C).

DISCUSSION

We reported a patient with TINU syndrome who had antitubular cell autoantibody. The antibody disappeared at recovery. T cell-mediated immune response has been suggested to underlie the pathogenesis of the disease. Thus, granuloma formation is reported in various organs including bone marrow, lymph nodes, and the liver.[1,5] The presence of T cells in the renal interstitial infiltrates has been demonstrated.[6,7] In contrast to increased immune reactivity at the inflammatory sites, patients with TINU syndrome exhibit anergy to skin tests. Furthermore, decreased lymphokine secretion by peripheral blood mononuclear cells of patients with TINU syndrome has been demonstrated.[5] These findings are consistent with active local immunity and subsequent peripheral suppressed immune response. On the other hand, circulating immune complexes[8,9] and autoantibodies such as rheumatoid factor,[1,10] antinuclear antibody,[11] and antineutrophil cytoplasmic antibodies[12,13] have been reported in patients with TINU syndrome. These reports, together with our finding, suggest that humoral-mediated process may be operative at least in some patients with TINU syndrome.

Autoantibodies to tubular cells have been demonstrated in the sera from patients with Sjögren syndrome,[3] systemic lupus erythematosus,[4] thyroid disease,[14] and vasculitis.[15] We reported for the first time the presence of antitubular antibody in a patient with TINU syndrome. It remains to be clarified whether the antitubular cell antibody may have developed after tubular antigen exposure by injury, or may have participated in the pathogenesis of TINU syndrome by inducing tubular injury.

The identity of 125-kDa protein also remains to be clarified. It was present in the cytosol but not in the nucleus of cortical tubular cells. Whether this 125-kDa protein can be found in the eye is not known. It has been postulated that autoimmune reaction against common antigens in the kidney and uvea leads to tubulointerstitial nephritis and uveitis. Also, the role of autoimmunity in the pathogenesis of uveitis has been suggested. Thus, antibodies to RNP, Ro, dsDNA, ssDNA, histones, and others have been demonstrated in sera from patients with uveitis.[16]

The reported patient had both proximal (tubular glucosuria, proteinuria, uricosuria, phosphaturia) and distal tubular dysfunctions (concentrating and acidifying defect), whereas the antibody reacted with only tubular epithelia in cortex. Therefore, the observed distal tubular dysfunctions might be attributable to injury to cortical distal tubules, a segment participating in both urine concentration and acidification.

Immunofluorescence study of renal biopsy specimen was negative for IgG. Although in previous studies, autoantibodies to tubular cells have been recognized by localization of immunoglobulins, this traditional technique is probably not sensitive. With the assay used in the present study, more patients with TINU syndrome may be detected who have antitubular cell antibodies.

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PAID TO BURY BAD NEWS

. . . A university pharmacologist acted as a reviewer for medical journals considering whether to publish reports on phenylpropanolamine (PPA) while he was on the payroll of the leading diet pill manufacturer, Thompson Medical Company. His anonymous critiques helped relegate some articles that questioned PPA's safety to little-known journals.

Gerth J, et al. Another part of the battle. *New York Times.* December 13, 2000

Submitted by Student