Gadolinium-Based Contrast Exposure, Nephrogenic Systemic Fibrosis, and Gadolinium Detection in Tissue

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OBJECTIVE. The objective of our study was to retrospectively review one institution’s cases of nephrogenic systemic fibrosis (NSF), evaluate possible associated factors, determine the prevalence of NSF, and search for gadolinium in skin samples obtained from patients with NSF.

MATERIALS AND METHODS. A retrospective review of our dermatopathology database from 1997 to 2007 was performed to search for patients with NSF. The records of patients with NSF were reviewed for factors suspected to be associated with NSF such as acidosis, low hemoglobin levels, low serum calcium levels, inflammatory conditions, serum antibodies, pharmaceutical erythropoietin, angiotensin-converting enzyme inhibitors, gadolinium-based contrast agents (GBCAs), renal failure, and dialysis. The biopsy samples from NSF patients and from control subjects were examined with energy-dispersive X-ray spectroscopy to detect gadolinium. Retrospective chart reviews of patients evaluated at our local dialysis center and our dermatology clinic were conducted to identify patients who underwent MRI, who had NSF managed exclusively by our tertiary referral centers, or both from 1997 to 2007.

RESULTS. Seven cases of NSF were found in the dermatopathology database. Two of the seven patients were also followed up at our outpatient dialysis clinic. No other cases of NSF were discovered within the dialysis clinic’s population exclusively followed within our institution. All seven dermatopathology database NSF patients developed symptoms of NSF after receiving GBCAs during renal failure and showed concomitant proinflammatory conditions. No other proposed risk factors were uniformly present in these NSF cases. All four NSF patients with chronic renal failure developed NSF after hemodialysis, with one patient dialyzed 12 hours after receiving a contrast dose. Gadodiamide was the only GBCA that all seven NSF patients received before symptom onset. Symptom onset was from 3 weeks to 18 months after GBCA exposure, with cumulative GBCA doses ranging from 0.16 to 0.43 mmol/kg. Gadolinium was detected in six of seven NSF patients’ skin biopsies. Seven of eight random control specimens obtained from three healthy control subjects, three patients with renal insufficiency who had not been exposed to gadodiamide, and two patients without renal disease who had been exposed to gadodiamide were negative. Seventy-two dialysis clinic patients underwent 127 contrast-enhanced MR examinations from 1997 to 2007. Eighteen patients received gadopentetate, none of whom developed NSF. Sixty-three patients received gadodiamide, two of whom developed NSF (prevalence of NSF in patients exposed to GBCA, 2.8%; odds ratio, 0.82 [95% CI, 0.04–18.10]; likelihood ratio, 1.16 [95% CI, 1.06–1.26]). Nine patients received both contrast agents.

CONCLUSION. An association with GBCAs in the development of NSF is suggested in the setting of renal insufficiency, but other factors seem to play a role. Dialysis did not prevent the development of NSF. Gadolinium was detected in skin samples from NSF patients.
until September 2000, gadopentetate was used in almost all of the contrast-enhanced MR examinations. Our institution began using gadodiamide almost exclusively from September 2000 until the present. The local outpatient dialysis clinic associated with our institution is somewhat geographically isolated. These patients therefore received their imaging and clinical care almost exclusively in our institution (Figs. 1 and 2).

The purpose of this investigation was to retrospectively review NSF cases diagnosed and followed up in our institution, evaluate the proposed risk factors and associated patterns, search for the presence of gadolinium in skin samples of patients with NSF, and determine the prevalence of the disease in a specified population of patients who undergo dialysis and MR examinations.

Materials and Methods

Institutional review board approval was obtained, and the study was compliant with HIPAA. Patient consent was not necessary because of the retrospective, anonymous nature of the investigation.

A retrospective review of our institution’s dermatopathology database was conducted to discover all cases of NFD or NSF diagnosed from 1997 to 2007. Other subjects with stored samples previously diagnosed as scleromyxedema before inception of the NFD and NSF diagnoses were reexamined with the current diagnostic criteria to determine whether they were actually additional cases of NSF.

The criteria for the diagnosis of NSF included the appropriate clinical presentation in the setting of renal dysfunction, thickening or hardening of the skin in a typical distribution (lower legs, forearms) with sparing of the face, and histologic confirmation. Histologic criteria included the presence of a spindle cell, fibroblastic proliferation of the dermis intervening between thickened collagen bundles with a distinct lack of significant inflammation. Histologic stains and immunohistochemistry supporting the NSF diagnosis included increased mucin and CD34 positivity. All of the biopsy specimens from these patients were analyzed and diagnosed by a fellowship-trained dermatopathologist with 15 years of experience.

A retrospective chart review of the NSF cases from the dermatopathology database was conducted to search for the presence of proposed associated risk factors described in the literature such as hemoglobin levels, serum calcium levels, concomitant inflammatory conditions, recent surgical procedures, erythropoietin administration, lack of angiotensin-converting enzyme (ACE) inhibitors, presence and type of renal insufficiency, creatinine clearance (as calculated by the Cockroft-Gault method [6]), and dialysis. These patients’ records were then searched in our institution’s PACS for prior MR examinations, the examination dates, and whether the examinations were performed with GBCAs. If a GBCA was used, the type and volume of GBCA were recorded. Each patient’s weight obtained near the time of each MR contrast injection was noted, and the contrast dose was calculated. These doses were summed for each NSF patient to obtain the cumulative dose, which was plotted against the time from first exposure in renal failure to symptom onset. By calculating the Spearman’s rank correlation coefficient, these data were examined for any possible linear correlation between cumulative dose and elapsed time to symptom onset.

To investigate the prevalence of NSF in a well-defined patient population, a retrospective chart review was conducted of a local outpatient dialysis clinic’s patients evaluated in our institution from January 1997 to February 2007. This time frame was chosen to correlate with the inception of the institution’s dermatopathology database. These patients were treated almost exclusively in our institution. Hospital record numbers were recorded, and the institution’s PACS system was searched for all MRI studies performed with or without contrast material in this population. The type of contrast material was recorded for each study. The dermatopathology database and patient charts were then searched for cases of NSF. The results were tabulated with a 2 × 2 table and Haldane’s estimator to determine the prevalence, odds ratio, and likelihood ratio of developing NSF after receiving gadolinium compared with patients who underwent MRI without receiving contrast material.

Eleven paraffin blocks from prior biopsies of the seven patients diagnosed with NSF or NFD in the dermatopathology department were collected for evaluation of tissue gadolinium with scanning electron microscopy (SEM) energy-dispersive X-ray spectroscopy. Eight blocks from seven control patients without a diagnosis of NSF or NFD
were also chosen. These control cases were selected from the dermatopathology database to include healthy patients, those with renal insufficiency and a history of recent MRI or MR angiography, and two patients with a diagnosis of calciphylaxis. The available control samples therefore included three healthy control subjects, three patients with renal insufficiency who had not been exposed to gadodiamide, and two patients without renal disease who had been exposed to gadodiamide.

The paraffin blocks were mounted on a specimen holder and examined in a scanning electron microscope (S-4700, Hitachi) at 15–20 kilovolts with condenser lens 5 and objective lens 1 and a working distance of 12 mm. Energy-dispersive X-ray spectroscopy was conducted on the surface of each specimen and was targeted at 10 randomly selected, evenly dispersed sites throughout each specimen using a scanning electron microscope. If tumor was present in the samples, both tumoral and nontumoral areas were evenly targeted. Data were recorded and interpreted on a Genesis 2000 analyzer system (EDAX). Sensitivity, specificity, likelihood ratio, and CIs for this test were then calculated from a 2 × 2 table.

Results
The results of the data collected from the NSF cases diagnosed in our institution are summarized in Table 1.

Demographics
Seven biopsy-confirmed cases of NSF have been diagnosed at our institution to date, all occurring since 2001, with the latest case diagnosed in January 2007. All seven identified cases were documented in the dermatopathology database. Two of these cases were also followed up in our institution’s outpatient dialysis clinic. No other cases of NSF were identified in our dialysis clinic population. Two of the remaining five NSF cases were treated at dialysis centers in other cities, whereas the other three never required chronic treatment at a dialysis center.

The age of the patients with NSF ranged from 14 to 87 years, with a mean age of 47 years. Four were female and three were male. Two were African American, three were Hispanic, one was white, and one was from India.

Symptoms at Presentation
Of the seven patients with NSF, three presented with plaques on their forearms. One patient experienced sudden onset of swollen, stiff upper extremities and subsequent limited mobility. Three patients presented with raised plaques on both legs, with similar findings on the forearms and palms of one patient.

GBCA Exposure
All seven NSF patients received gadodiamide contrast material before the onset of symptoms and while in renal insufficiency. None received gadopentetate before developing symptoms.

Three patients developed symptoms of NSF after one exposure to gadodiamide. These patients developed NSF symptoms 3 weeks to 3 months (mean, 11 weeks) after the first gadodiamide exposure.

Four patients developed NSF after more than one gadodiamide-enhanced MR examination. All but one of these doses were administered while the patient was in renal failure. Patient 1 received two boluses of gadodiamide before symptom onset and developed symptoms 18 months after the first exposure and 2 months after the second exposure (with doses of 0.16 and 0.12 mmol/kg, respectively). Patient 7 was not in renal failure when given the first dose, but developed symptoms of NSF within 3 months after the second dose (0.12 mmol/kg) while in renal failure. Patient 4 received the first two boluses of gadodiamide within 1 week at doses of 0.13 mmol/kg each and developed symptoms within 3 months of the first exposure. Patient 2 received two 0.08 mmol/kg doses within 3 months, and symptoms appeared 3 months after the first bolus.

All of our NSF patients developed symptoms related to NSF after no more than three contrast-enhanced MR examinations, although a significant linear correlation between cumulative dose of GBCA and timing of symptom onset was not found (Spearman’s rank correlation coefficient = –0.13) (Fig. 3).

Gadolinium Detection in Skin Biopsy Samples
Gadolinium was detected in six of the seven NSF patients’ skin biopsies in both diseased and healthy skin (Table 2). Eight control samples were also analyzed including two random specimens from patients recently exposed to gadodiamide and three from patients with renal insufficiency. Two patients with calciphylaxis were randomly included because of the disease’s histologic similarities to NSF. Seven of the eight control samples were negative for gadolinium (Table 3). One control sample from a patient with calciphylaxis was positive for gadolinium. This patient had no record of MR contrast exposure in our institution, but the patient was known to have received treatment at multiple institutions and may have received GBCAs elsewhere. Sensitivity for this test was 86%; specificity, 88%; and likelihood ratio, 6.9, with a 95% CI of 1.07–43.97.

Interestingly, in one NSF patient (patient 7), gadolinium was detected not only in a typical NSF plaque but also in skin inflamed by a drug eruption not found to show NSF.

Dialysis and Renal Failure
All seven of the NSF patients developed renal failure before developing symptoms of NSF. All NSF cases developed after patients had been exposed to GBCAs, with creatinine clearances of less than 36 mL/min. Of the three patients who developed symptoms in the setting of acute renal failure, two did not receive dialysis. Patient 7 was dialyzed once before the onset of NSF symptoms, 2 days after the patient’s first enhanced MR examination in renal failure (creatinine clearance = 13.6 mL/min). This patient progressed to chronic renal failure after the onset of NSF symptoms.

Four patients had chronic renal failure at the time MR contrast material was administered before the onset of NSF symptoms. Two were on regularly scheduled hemodialysis when they received their first dose of gadodiamide. None was on peritoneal dialysis. Patient 1 received dialysis 48 hours after the second exposure. Patient 2 was dialyzed 12 hours postdose. Patient 3 began dialysis 9 days after receiving a bolus. Patient 6 started dialysis 23 days postdose.

Hemoglobin
Six patients had hemoglobin values obtained at the time of first gadolinium-based contrast injection during renal failure. Of these, five had decreased hemoglobin levels. Four of the five tested patients with multiple exposures had decreased hemoglobin during the second gadolinium dose.

Serum Calcium
Five of the seven NSF patients had chronically low serum calcium levels before their first GBCA injection. The other two NSF patients had no hypocalcemia before or after receiving GBCAs. No significant change in calcium levels was observed in any of the seven patients after receiving GBCAs.

Inflammatory Conditions
All seven NSF patients had coexisting proinflammatory conditions, such as recent surgery, positive inflammatory antibodies,
### TABLE 1: Findings in and Characteristics of Patients with Nephrogenic Systemic Fibrosis (NSF)

<table>
<thead>
<tr>
<th>NSF Patients</th>
<th>Age, Sex, Race</th>
<th>Cumulative Gd Dose Before Symptom Onset (mmol/kg)</th>
<th>Time from First Gd Exposure to Symptom Onset</th>
<th>Type of Renal Failure at Symptom Onset</th>
<th>Dialysis</th>
<th>Cause of Renal Failure</th>
<th>Time from Gd Exposure to Hemodialysis</th>
<th>Calcium Acid–Base</th>
<th>Hgb</th>
<th>Antibodies</th>
<th>Medications of Interest at Symptom Onset</th>
<th>Surgical Procedures</th>
<th>Outcome</th>
<th>Gd in Skin Biopsy Samples</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>33, F, AA</td>
<td>0.28</td>
<td>18 mo</td>
<td>Chronic Lupus nephritis</td>
<td>QOD</td>
<td></td>
<td>13.48 (mmol/mL)</td>
<td>Normal</td>
<td></td>
<td>ANA+</td>
<td>EPO</td>
<td>Laparotomy</td>
<td>Stable</td>
<td>Yes</td>
</tr>
<tr>
<td>2</td>
<td>62, M, AA</td>
<td>0.16</td>
<td>3 mo</td>
<td>Chronic Diabetic nephropathy</td>
<td>QOD</td>
<td></td>
<td>21.80 (mmol/mL)</td>
<td>Low</td>
<td>Low</td>
<td>ANA–</td>
<td>EPO</td>
<td>Decubitus ulcer, débridement</td>
<td>Stable</td>
<td>Yes</td>
</tr>
<tr>
<td>3</td>
<td>42, F, H</td>
<td>0.26</td>
<td>3 w</td>
<td>Chronic Chronic hypertension</td>
<td>QOD</td>
<td></td>
<td>11.89 (mmol/mL)</td>
<td>Low</td>
<td>Normal</td>
<td>ANA–</td>
<td>EPO</td>
<td></td>
<td>Mild improvement</td>
<td>Yes</td>
</tr>
<tr>
<td>4</td>
<td>14, F, H</td>
<td>0.26</td>
<td>3 mo</td>
<td>Acute Lupus nephritis</td>
<td>None</td>
<td></td>
<td>26.91 (mmol/mL)</td>
<td>Low</td>
<td>Low</td>
<td>ANA+, anti-SM+, ACL+</td>
<td></td>
<td></td>
<td>Marked improvement</td>
<td>No</td>
</tr>
<tr>
<td>5</td>
<td>38, F, H</td>
<td>0.19</td>
<td>3 mo</td>
<td>Acute SLE with diffuse proliferative nephritis</td>
<td>None</td>
<td></td>
<td>35.89 (mmol/mL)</td>
<td>Low</td>
<td>Acid</td>
<td>Low</td>
<td>ANA+, anti-dsDNA+</td>
<td></td>
<td>Stable</td>
<td>Yes</td>
</tr>
<tr>
<td>6</td>
<td>87, M, W</td>
<td>0.43</td>
<td>2 mo</td>
<td>Chronic End-stage renal disease from AAA repair</td>
<td>QOD</td>
<td></td>
<td>10.54 (mmol/mL)</td>
<td>Low</td>
<td>Normal</td>
<td>ANA–</td>
<td>EPO, ACE I, AAA repair and lower extremity embolectomy</td>
<td></td>
<td></td>
<td>Yes</td>
</tr>
<tr>
<td>7</td>
<td>56, M, I</td>
<td>0.24</td>
<td>8 mo</td>
<td>Acute Aneurysm repair</td>
<td>Once</td>
<td></td>
<td>11.20 (mmol/mL)</td>
<td>Low</td>
<td>Base</td>
<td>Low</td>
<td>EPO</td>
<td>Aneurysm repair and left iliac bypass</td>
<td>Stable</td>
<td>Yes</td>
</tr>
</tbody>
</table>

Note—Gd = gadolinium, HD = hemodialysis, Hgb = hemoglobin, AA = African American, H = Hispanic, W = white, I = from India, SLE = systemic lupus erythematosus, AAA = abdominal aortic aneurysm, QOD = every other day, ANA = antinuclear antibodies, anti-SM = anti-Smith antibodies, ACL = anticardiolipin, dsDNA = double-strand DNA, plus sign (+) = positive, minus sign (–) = negative, EPO = erythropoietin, ACE I = angiotensin-converting enzyme inhibitor, ECP = extracorporeal photopheresis, CAD = coronary artery disease.
or concurrent granulomatous disease at the time of symptom onset. However, none had undergone liver transplantation.

Three of the seven tested NSF patients were antinuclear antibody (ANA)–positive. Patient 4 was also positive for anti-Sm and anticardiolipin antibodies, although none of the other patients was tested for these antigens. Patients 6 and 7 underwent repair of aortic aneurysms within 6 months before symptom onset. Patient 6 also underwent an arterial embolectomy within that time frame. Patient 2 underwent surgical débridement of a large decubitus ulcer with secondary osteomyelitis approximately 3 weeks before symptom onset. Patient 1 underwent laparotomy 2 months before symptom onset. The remaining NSF patients had not recently undergone surgical procedures before symptom onset.

Acidosis

One patient was in metabolic acidosis at the time of the first GBCA exposure. Patient 7 was alkalotic during the first GBCA bolus received in the setting of renal failure. The remaining five patients did not have serum chemistry values sufficient to evaluate acid–base state in this time frame.

Medications

Six of the seven patients were not taking ACE inhibitors at the time of symptom onset. Five of the seven patients were taking erythropoietin at the time of symptom onset. No medications were common among all of the NSF patients except gadodiamide.

Patient Outcome

Five patients had no documented changes in their cutaneous lesions. One reported increased bilateral upper extremity stiffness. One showed mild symptomatic improvement after undergoing extracorporeal photophoresis, whereas another showed steady improvement after renal function returned to normal. Another patient died as a consequence of extensive atherosclerotic disease with no change in the cutaneous lesions.

Prevalence

One hundred thirty-eight MR studies were conducted in our institution’s local outpatient dialysis clinic from January 1997 to February 2007 on 83 patients, 72 of whom underwent 127 contrast-enhanced MR examinations. Eighteen of the 72 patients underwent 23 studies enhanced with gadopentetate, none of whom developed NSF. Nine of this group of 18 patients also underwent studies enhanced with gadodiamide. Sixty-three patients underwent 104 MR examinations enhanced with gadodiamide. Two of these patients developed NSF, both having received gadodiamide exclusively; both of these patients were diagnosed at our institution and are therefore part of the seven total cases of NSF described herein. Eleven patients underwent unenhanced MR examinations only.

Using a 2 × 2 contingency table, we correlated these data with the number of NSF cases (two NSF cases of 72 gadodiamide-exposed cases compared with no cases in the 11 unexposed who underwent MRI), showing an odds ratio (via Haldane’s estimator) of

![Fig. 3—Plot of total time to develop nephrogenic systemic fibrosis (NSF) symptoms after onset of gadolinium contrast exposure in renal failure versus cumulative dose of gadolinium-based contrast agent before symptom onset in each patient (●) shows no linear correlation between two factors \( r = 0.01 \).](image-url)

### TABLE 2: Energy-Dispersive X-Ray Spectroscopy Test Results Among Patients Diagnosed with Nephrogenic Systemic Fibrosis (NSF)

<table>
<thead>
<tr>
<th>NSF Patient</th>
<th>Origin of Skin Biopsy Samples</th>
<th>Gadolinium Detected?</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Left lateral foot</td>
<td>Yes</td>
</tr>
<tr>
<td>1</td>
<td>Right medial ankle</td>
<td>No</td>
</tr>
<tr>
<td>2</td>
<td>Left forearm</td>
<td>Yes</td>
</tr>
<tr>
<td>3</td>
<td>Right palm</td>
<td>Yes</td>
</tr>
<tr>
<td>4</td>
<td>Right leg</td>
<td>No</td>
</tr>
<tr>
<td>5</td>
<td>Right abdomen</td>
<td>Yes</td>
</tr>
<tr>
<td>5</td>
<td>Right arm</td>
<td>Yes</td>
</tr>
<tr>
<td>6</td>
<td>Left arm</td>
<td>Yes</td>
</tr>
<tr>
<td>6</td>
<td>Abdomen</td>
<td>No</td>
</tr>
<tr>
<td>7</td>
<td>Right leg</td>
<td>Yes</td>
</tr>
<tr>
<td>7</td>
<td>Left thigh(^a)</td>
<td>Yes</td>
</tr>
</tbody>
</table>

\(^a\)Uninvolved skin.

### TABLE 3: Energy-Dispersive X-Ray Spectroscopy Test Results Among Control Subjects

<table>
<thead>
<tr>
<th>Control Patients</th>
<th>Origin of Skin Biopsy Samples</th>
<th>Kidney Disease?</th>
<th>Gadolinium Exposure?</th>
<th>Gadolinium Detected?</th>
<th>Diagnosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Left leg</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>Subacute spongiotic dermatitis</td>
</tr>
<tr>
<td>2</td>
<td>Right upper back</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>Psoriasiform dermatitis</td>
</tr>
<tr>
<td>3</td>
<td>Left shin</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>Leukocytoclastic vasculitis</td>
</tr>
<tr>
<td>4</td>
<td>Left shoulder</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>Superficial basal cell carcinoma</td>
</tr>
<tr>
<td>4</td>
<td>Right mid back</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>Basal cell carcinoma</td>
</tr>
<tr>
<td>5</td>
<td>Left forehead</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>Squamous cell carcinoma</td>
</tr>
<tr>
<td>6</td>
<td>Right thigh</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>Calciaphylaxis</td>
</tr>
<tr>
<td>7</td>
<td>Left inner thigh</td>
<td>Yes</td>
<td>Unknown</td>
<td>Yes</td>
<td>Calciaphylaxis</td>
</tr>
</tbody>
</table>
Gadolinium-Based Contrast Exposure, NSF, and Gadolinium Detection in Tissue

0.82 (95% CI, 0.04–18.10) with a likelihood ratio of 1.16 (1.06–1.26) and a prevalence of 2.8% among all GBCA-exposed outpatient dialysis clinic patients.

Discussion
Gadolinium in Tissues
Gadolinium deposits can be found in the body after GBCA-enhanced MR examinations. White et al. [7] described the detection of gadolinium in the bone of subjects in the postcontrast state. Other researchers have detected gadolinium in biopsy samples obtained from skin lesions and from normal skin of patients diagnosed with NSF, although only one control sample was tested in that study [8]. Six of the seven NSF patients in our study tested positive for gadolinium in skin biopsies from various sites. Two specimens of uninvolved skin from patients with NSF were also positive for gadolinium by energy-dispersive X-ray spectroscopy, suggesting systemic deposition. One patient’s biopsy (patient 4) did not show evidence of gadolinium via energy-dispersive X-ray spectroscopy. This patient had acute renal failure that had resolved by the time the biopsy sample was obtained. The patient continued to have marked symptom improvement after renal function returned. The gadolinium may have no longer been present in her tissue at the time of the biopsy. Alternatively, the gadolinium may not have been detected because our technique used a random bombardment of electrons on only the surface of the paraffin block. The use of serial ultrathin sections would increase the surface area examined and may therefore increase the sensitivity of the test.

Our study incorporated eight control samples. Although no samples were from patients with both renal insufficiency and known gadolinium exposure, five of the eight samples had one of these factors. Three of the eight control subjects had neither renal insufficiency nor a known history of gadolinium exposure. These patients were not expected to have gadolinium detected via energy-dispersive X-ray spectroscopy, but they were included to further evaluate the validity of the test by increasing the number of control subjects. Seven of the skin samples from the control subjects were negative for gadolinium via energy-dispersive X-ray spectroscopy. The positive result in one control sample was from a patient with calciphylaxis and no history of exposure to GBCAs at our institution. Unfortunately, the patient died 1 year before our investigation, and any further history of possible GBCA exposure at an outside institution was unobtainable. This patient, however, was known to have received care at other hospitals in our state and the possibility that he was exposed to gadolinium at one of the outside institutions cannot be excluded.

Our technique of gadolinium detection used energy-dispersive X-ray spectroscopy directly on the paraffin block visualized by SEM. Energy-dispersive X-ray spectroscopy records characteristic X-ray spectra emitted from heavy metals that have been encountered by an electron beam [9]. Others have used 3-µm sections and have had concerns about contamination with the water bath, planchets, microtome blade, and tissue stains [8, 10]. Our technique does not require sectioning, which decreases foreign metal contamination while sparing tissue.

The sensitivity of our technique can only be estimated. Both the concentration and distribution of gadolinium in the sampled tissues are likely the most critical factors affecting sensitivity because detection depends on random sampling for microanalysis of the specimens. The technique does not, however, differentiate among the various chemical states of those metals, including gadolinium. The presence or absence of gadolinium can be detected, but whether it is in the chelated, elemental, or ionic state cannot be determined. Elster [9] found the upper limit of the minimum concentration of gadolinium was 0.005 mol/L, or approximately 0.005 mmol per gram of tissue. Sampling error is always possible because we bombarded only 10 areas per specimen and we used only one histologic section at the surface of the paraffin-embedded block.

Some investigators have suggested that gadolinium may accumulate at sites of inflammation, preexisting trauma, or stasis [8, 10]. Our findings might support this theory. In one of our NSF patients, gadolinium was detected not only in a biopsy sample of a typical NSF plaque but also in a biopsy sample of skin inflamed by a drug eruption not found to show NSF. Accumulation of gadolinium at sites of inflammation may explain the finding of NSF plaques occurring near IV catheters, dialysis access or fistula sites, or surgery sites, as described by other investigators [8, 10]. The predefinition of NSF for the lower extremities may reside in the fact that stasis dermatitis and capillaritis, characterized by mild perivascular lymphocytic infiltrate and extravasation of RBCs, are commonly present in these areas even in clinically normal skin, perhaps leading to more diffuse tissue deposition of gadolinium.

GBCAs and NSF
The U.S. Food and Drug Administration (FDA) approved five different GBCAs for patient use, with an approved standard dose of 0.1 mmol/kg. The adverse effects at this dosage have proven negligible. Gadodiamide and gadoteridol are also approved for use at up to 0.3 mmol/kg with no evidence of increased toxicity [11–13] and are now commonly used at that dose for MR angiography [12].

Linear chelate complexes are inherently less stable in vivo than many other GBCAs on the market because their gadolinium ions are more loosely bound to the chelate ligands than with other agents. Gadodiamide (Fig. 4) and gadopentetate are both linear chelate complexes. Other marketed GBCAs are macrocyclic compounds, which more completely encompass their gadolinium ions, forming more bonds with ionic gadolinium (Gd³⁺). Gadolinium bound to linear chelate ligands is therefore more likely to be released in vivo because other biochemical ions compete for the chelating agent’s binding site [14].

Fig. 4—Chemical structure of gadodiamide. Chelate ligand partially encompasses gadolinium ion. (Courtesy of GE Healthcare)
Even so, none of the patients in our study exposed exclusively to gadopentetate developed NSF. Conversely, all seven NSF cases in this investigation were in patients who developed NSF only after exposure to gadodiamide. Although these findings initially suggest that one gadolinium chelate is more commonly associated with NSF, the prevalence of each available GBCA in our patient population should be considered. It is possible that no cases of NSF were observed in our population who received only gadopentetate simply because too few patients received that GBCA to observe an associated case of NSF. Nevertheless, although no public statistics are available regarding the exact prevalence of each marketed GBCA used in imaging, multiple authors assert that most of the diagnosed NSF cases have been associated with gadodiamide, even to a level that is disproportionate to gadodiamide’s share of the market [14, 15].

One possible explanation for this trend is that gadodiamide has a lower thermodynamic stability constant than gadopentetate. Transmetallation—a process by which gadolinium ions leave the chelate complex and bind to molecules elsewhere—depends on the thermodynamic stability of the GBCA. The lower the thermodynamic stability, the more likely transmetallation will occur in vivo. The values for thermodynamic stability for gadodiamide and gadopentetate are 10^{44.9} and 10^{8.1}, respectively, meaning gadodiamide is more likely than gadopentetate to undergo transmetallation, releasing its gadolinium ions in vivo [12, 16]. This, augmented by a persistence of GBCAs in the circulation of patients with renal failure, has been speculated to lead to an increased incidence of gadolinium deposition after receiving gadodiamide, thus increasing the likelihood of a response by circulating fibrocytes in the tissues of patients receiving this GBCA [8, 12, 14].

The chelate complex is cleared almost entirely by renal excretion [16]. Thus, a markedly increased half-life is present in patients with renal insufficiency that is directly proportional to the degree of renal dysfunction [17]. This increased time in vivo has been proposed to increase the likelihood that ionic gadolinium will become free in tissues [14]. Cowper et al. [18] proposed a model of NSF pathogenesis that suggests peripherally deposited elemental or ionic gadolinium may act as an antigen that is targeted by bone marrow–derived circulating fibrocytes (CD34+ and procollagen positivity). Such cells are known to be stimulated by affected areas of the skin, perhaps by trauma or inflammation, to begin a woundlike healing process with the production of many cytokines, particularly transforming growth factor-β. Researchers have speculated that chelate molecules may encounter an in vivo chemical environment, such as acidosis, that causes gadolinium ions to be released [8, 19]. Interestingly, one of our patients was alkalotic at the time of gadolinium exposure. Even so, insufficient acid–base laboratory values were available in that patient’s records to effectively address this theory. Others have failed to substantiate a link to acidosis [12, 17]. Researchers have also suggested that the chelating agent itself may trigger NSF by interacting with biologic substrates once its active site is unbound [17].

Four of our patients with NSF had more than one gadodiamide bolus before developing NSF. All but one of those exposures occurred while the patient was in renal failure, with one patient remaining symptom-free until 18 months after the firstrenally impaired exposure. Such a protracted interval has not been previously reported in the literature, to our knowledge. Also of importance, 70 of our 72 outpatient dialysis clinic patients with renal failure exposed to gadodiamide, gadopentetate, or both did not develop NSF. Moreover, even though almost all reported cases of NSF have an associated exposure to GBCAs, two recent studies raise the possibility that GBCA exposure may rarely be absent [20, 21]. These findings seem to reinforce the suggestion by Marckmann et al. [17] that other factors may play a role in the pathogenesis of NSF.

### Other Proposed Risk Factors

None of the other proposed risk factors that we examined was present in all patients and therefore may not be necessary for the development of NSF. Individually, however, some of these factors may represent a “hit” that may tip the process toward a more suitable environment for fibrosis. Figure 5 illustrates the points at which some of these additional potential risk factors may interact with the previously described model of NSF development proposed by Cowper et al. [18]. Clearly, continued investigation into the pathogenesis of the fibrosis associated with NSF and the importance of these individual potential risk factors is needed.

Erythropoietin, for example, may, as Swaminathan et al. [22] pointed out, increase the number of bone marrow–derived CD34+ fibrocytes in the circulation that are available for recruitment into tissues. In almost all of their patients, symptoms of NSF started within 1 month of erythropoietin initiation or a significant dose increase. They also reported improvement in many of their patients who discontinued or decreased their dose of erythropoietin. Even so, the literature reports many cases of NSF in patients with no exposure to erythropoietin, as was also the case with two of our NSF patients. We can theorize that erythropoietin may increase the risk of developing NSF, but it may not necessarily be crucial for the development of the fibrosis associated with NSF.

Although dialysis itself was previously implicated, no common type of dialysis equipment, membrane cleaning method, or dialysis fluid has shown an association with the development of NSF [1, 23]. Two of our NSF patients were never dialyzed before

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**Fig. 5**—Diagram of possible mechanism of fibrosis in nephrogenic systemic fibrosis (NSF) illustrates points at which various potential risk factors may interact to potentiate development of fibrosis. Note that angiotensin-converting enzyme (ACE) inhibitors may inhibit fibrosis found in NSF. TGF–β = transforming growth factor–β.
symptom onset. Others have reported findings [12, 24–26] similar to ours.

Sadowski et al. [27] recently reviewed the records of 13 NSF patients and concluded that those who receive GBCAs in the setting of decreased renal function with a concurrent proinflammatory condition, such as recent surgery, inflammatory disease, or granulomatous disease, are more likely to develop NSF. All of our NSF patients had recently undergone surgery or had positive inflammatory antibodies at the time of symptom onset (or both). Cowper [26] emphasized an apparent pattern of developing NSF soon after vascular and other surgical procedures, including arteriovenous fistula formation and renal transplantation, with the total reported NSF cases with this combination approaching 90%. Multiple investigators have also described an apparent increased incidence of NSF among liver transplant recipients, although all of these patients also had renal failure [12, 28, 29]. None of our NSF patients underwent liver transplantation.

One team of investigators noticed an absence of ACE inhibitors in the medication regimens of renal patients at their institution who developed NSF. ACE inhibitors are known to deter the formation of fibrosis after tissue damage [30]. Only one of our patients was taking an ACE inhibitor. Other investigators have reported an absence of commonalities in NSF patients’ medications [12, 17, 19]. Mackay-Wiggan et al. [24] described four cases of NSF that were positive for anticardiolipin antibodies, two of which were also positive for ANA. Three of our seven NSF patients were ANA-positive. One was also positive for anticardiolipin antibodies.

Five of our seven NSF patients showed mild hypocalcemia that was present before, during, and after receiving the GBCA bolus in renal insufficiency. No similar pattern has been reported in the literature, although one investigation described a patient who developed mild hypocalcemia after receiving a GBCA [31]. Interestingly, hypocalcemia did not develop in the two NSF patients with normal calcium levels before receiving GBCA.

Five of six NSF patients with blood counts drawn at the time of the first contrast bolus in renal failure had low hemoglobin levels. Sadowski et al. [27] reported a preponderance of anemia among their patients, but they found no statistical significance of this pattern.

Mendoza et al. [23] suggested that perhaps even renal failure might not be a prerequisite for NSF because no similar disease was identified before 1997 in patients with renal dysfunction. However, Cowper et al. [18] assured the medical community that the term “nephrogenic” is not a misnomer because there is a demonstrable correlation with renal disease. None of our patients developed symptoms until after they had initially developed renal dysfunction.

A genetic predisposition to develop NSF that manifests in the setting of renal failure has also been postulated, but the epidemiologic data to date show no definite predilection of NSF for any age group, sex, geographic location, or race, although a slight increase in incidence exists among middle-aged patients [2, 32].

All of the cases of NSF in our study developed after patients were exposed to GBCAs and had creatinine clearances of less than 36 mL/min. In an alert updated in May 2007, the FDA’s previously published recommendation to withhold GBCAs in patients with a creatinine clearance of less than 30 mL/min was reassessed. The FDA also recommends withholding GBCAs in any patient with hepatorenal syndrome or liver transplant recipients in the perioperative state [33]. In Europe, the Committee for Medicinal Products for Human Use (CHMP) has issued similar guidelines [14], which strongly discourage the use of gadodiamide in the setting of glomerular filtration rates (GFRs) of < 30 mL/min or liver transplantation.

In light of our findings that show NSF can develop in patients with creatinine clearance values above the supposed safe threshold for renal function published by the FDA and CHMP, our institution is currently following the recommendations of Kanal et al. [15]. Those researchers suggested that all MR requisites include a questionnaire screening for renal dysfunction. If a patient with decreased renal function has mild disease (i.e., GFR > 60 mL/min), contrast-enhanced MR studies might be performed, although gadodiamide should be substituted with another contrast agent. If the disease is more severe (i.e., GFR < 60 mL/min), acute renal failure has developed, or the patient is on dialysis or has end-stage renal disease, they suggest withholding GBCAs unless the benefits of a gadolinium-enhanced MR examination can be clearly documented to outweigh the risks. Also, as asserted by Bongartz [14], GBCAs should be withheld in any diagnosed cases of NSF. Both the FDA [33] and Kuo et al. [34] recommend that all dialysis patients undergo emergent dialysis after receiving a GBCA. Even so, our research shows that patients who undergo dialysis 12 hours or more after GBCA exposure may still develop NSF. Indeed, no other current evidence has shown a decrease in incidence of NSF with dialysis [12].

Prognosis
Various treatments for NSF have shown variable success, including extracorporeal photopheresis, plasmapheresis, interferon-α, cyclophosphamide, thalidomide, and prednisone, but to date no single factor has proven as effective as restoring renal function [2, 32]. One of our patients experienced some symptom improvement with extracorporeal photopheresis, whereas another had complete resolution of NSF after renal function returned. NSF resolves only sporadically in the small population of patients who experience restored renal function. Patients with persistent NSF can have a wide range of outcomes, ranging from mild disease to severe debilitation [2, 10, 18, 26].

Limitations were present in our investigation. Acid–base and hemoglobin laboratory values were not uniformly available in the NSF patients’ records. The patient population examined for prevalence of NSF was somewhat small, although all cases of chronic renal failure were identified and researched in that population and the findings were statistically significant. Nevertheless, the calculated odds ratio and likelihood ratio should be considered with caution because of the very low number of patients in our population who were not exposed to GBCAs (n = 11). When detecting gadolinium, no ideal control samples (renal failure with skin eruptions and gadolinium exposure without NSF) were available in the skin samples already stored at our institution.

Further investigation of the prevalence of gadolinium in skin samples of GBCA-exposed renally impaired patients who do not develop NSF using ideal control samples is needed. Also, many of the patterns of proposed risk factors observed in NSF have no control observations available from a matched population who did not develop NSF because of the retrospective nature of this investigation.

In conclusion, in addition to the epidemiologic evidence reaffirming an association of GBCA exposure with the development of NSF, gadolinium is detectable in the skin samples of these patients. Aside from GBCA exposure, no other investigated risk factor was present in all of our NSF patients. Our
research also confirms that hemodialysis 12 hours or more after GBCA exposure does not protect from the development of NSF. In addition, our findings provide more evidence to support the suggestion that other factors may be associated with the development of NSF.

Further investigation is needed to determine the pathogenesis of the fibrosis observed in NSF. Research investigating the importance of individual potential risk factors for developing NSF is also needed.

References


