# Clinical Evaluation of a Scintigraphic Method for Diagnosing Inflammations/Infections Using Indium-111-Labeled Nonspecific Human IgG

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This study was undertaken as part of a Phase II study to assess the sensitivity and safety of $^{111}$In-DTPA-human IgG, an imaging agent for the detection of inflammations and/or infections. Forty patients with infection/inflammation on the basis of clinical findings, microbiologic results, and/or the basis of results from other imaging modalities were studied. For evaluation of sensitivity, whole-body images were obtained at 6–12 hr (early) and 20–28 hr (delayed) postinjection and occasionally at 48 hr. No adverse reactions were recorded in any of the 40 patients studied. Positive results were obtained in 37 of 37 evaluable subjects (100%). The test appears to be a promising method for the detection of inflammation and/or infection.

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A number of imaging techniques are available for detecting occult sites of infection in the body. In most situations, detection is achieved by demonstrating anatomical changes in a suspected region of the body using computerized tomography (CT), ultrasound or magnetic resonance imaging (MRI) or by using a radionuclide imaging technique ($^{67}$Ga-citrate, $^{111}$In-chloride or $^{111}$In-labeled white cells) to localize sites of infection/inflammation. Although these techniques have demonstrated good sensitivity and specificity, they each possess a number of limitations (*1–5*).

A noninvasive whole-body screening method, utilizing a simple, readily available, radiolabeled kit preparation, which would allow the early detection or follow-up of infection/inflammation and which overcomes some of the limitations imposed by the other imaging techniques, would be highly desirable.

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Recently, various newly developed specific radiolabeled murine monoclonal and human polyclonal immunoglobulins have been shown to localize at sites of infection or inflammation in animals and human subjects (*4–11*).

Rubin et al. conducted a series of studies comparing specific murine monoclonal antibody preparations directed against bacteria (Type I *Pseudomonas aeruginosa*) and a "nonspecific antibody" against a non-mammalian, nonbacterial haptene, p-arsanilic acid, and demonstrated that both agents could image infection (*12*).

Similarly, they demonstrated that human polyclonal IgG, derived from two different commercial sources, imaged various infections (*10–12*) and conducted a clinical trial reporting a sensitivity of 92% and specificity of 95% with their product (*14*). The results of these studies (*10–16*) showed that radiolabeled IgG localization at the infection site did not require a specific immunoglobulin. The authors hypothesized that the nonspecific nature of localization of the antibody at the infected site was due to the Fc portion of the IgG molecule. They supported this hypothesis by demonstrating that the Fab fragment of human IgG did not localize in the infected thigh of a rat while both the whole IgG molecule and the Fc segment did localize. They further supported the affinity of the IgG molecule for sites of inflammation by comparing the uptake of both $^{67}$Ga-citrate and $^{99m}$Tc-labeled human serum albumin to the uptake of $^{111}$In-labeled IgG in the infected thigh of rats. The localization of $^{111}$In-labeled IgG substantially exceeded the localization of the other two compounds,

This study reports on the results of a Phase II trial of a radiolabeled polyclonal immunoglobulin $^{111}$In-PRI#0001-A-1 ($^{111}$In-DTPA-IgG) currently being developed by McNeil Pharmaceuticals for the radioimmunodetection of focal sites of infection/inflammation. This study differs from previously published reports because it uses a polyclonal human immunoglobulin not subjected to pepsin digestion as part of its manufacturing procedure (*17*).

## MATERIALS AND METHODS

Forty patients with suspected inflammation or infection were entered into this Phase II study after the nature of the procedure was explained and informed consent obtained as approved by the University of Miami Institutional Review Board.

Patients signing an informed consent had to fulfill at least one of the following two criteria:

1. Have a positive culture from the site of known infection.
2. Have either focal collection seen on an imaging study performed within 1 wk of the $^{111}$In-DTPA/IgG scan or a fever greater than 100.5°F for more than 3 days duration and one of the following:
   a. Abdominal and pelvic pain of greater than 3 days duration that is not related to diarrheal disease.
   b. Presence of a vascular prosthesis or aneurysm or orthopedic prosthesis.
   c. Localized pain.

Patients were excluded who: were not medically stable or had current disease and/or therapy that might affect radioimaging; had a history of agamma or extreme hypogammaglobulinemia, selective IgA deficiency, or past history of severe reactions to intravenous or intramuscular administrations of human IgG; had a BUN greater than 50 or serum creatinine greater than 2.5 mg/dl; or were using an experimental drug or experimental device.

### Reagents

The human polyclonal immunoglobulin and the chelator diethylenetriamine pentaacetic acid (DTPA) were provided as a DTPA-IgG complex by McNeil Pharmaceutical Research Institute as a two-vial kit. The $^{111}$In-Cl was provided by Amersham (Amersham, Arlington Heights, IL). Briefly, 0.5 ml of 0.25 *M* sterile citrate buffer was added to DTPA-IgG complex vial followed by 1.4 to 2.6 mCi of $^{111}$Indium-chloride (10 mCi/ml), and the mixture was incubated for 15 min at room temperature. Radiochemical purity test was performed by thin-layer chromatography using ITLC silica gel impregnated glass fiber strips (Gelman Instrument Co., Product No. 61885) with 2–3 ml of 0.1 *M* nonsterile citrate buffer as solvent. Only products with a labeling efficiency equal or greater than 90% were used for infusion.

### Procedure

Medical histories, physical examinations, and laboratory tests that included complete blood count (CBC) with white blood cell differential and platelet count, chemistries, immunoglobulin quantitation and urinalysis were obtained from all patients prior to the study. These were repeated at 24 hr postinjection.

The $^{111}$In-DTPA-IgG solution was administered intravenously as a single bolus injection within 6 hr after radiolabeling. All patients received 1 mg of IgG radiolabeled with 1.4 to 2.6 mCi of $^{111}$In with a labeling efficiency of 90.1%–98.5%. Vital signs including blood pressure, pulse rate, respiratory rate and temperature were measured within 15 min prior to injection, and again at 5, 15, and 30 min and 1, 4, 6–12 and 18–24 hr postinjection.

### Imaging

Whole-body scans, including anterior and posterior and/or spots of head, neck, chest, abdomen, pelvis and lower extremities and SPECT of selected cases, were acquired at approximately 9 ± 3 hr, 24 ± 4 hr, and when possible 48 ± 4 hr postinjection. This was performed using a large field of view gamma camera with a medium-energy collimator and 20% energy windows set on the 174 and 247 keV photopeaks of $^{111}$In. Images were recorded and stored in both digital and analog form. Whole-body images were acquired for a preset time of 10 min or $1 \times 10^6$ counts, whichever occurred first. SPECT images of selected sites were obtained utilizing a 64 × 64 matrix, over 360° with step and shoot at 6° increments for 40 sec per stop.

### Scan Interpretation and Clinical Assessment

Scans were interpreted by two onsite nuclear medicine physicians without knowledge of the clinical condition of the patient. Disagreements were resolved by consensus opinion.

The scans were reported as: *positive* when areas of increased abnormal concentration of the tracer were seen in the early images and remained as such and/or became more intense subsequently; *negative* when no abnormal uptake and/or only normal physiologic organ uptake was seen; or *equivocal* when an unusual distribution not related to physiologic organ uptake was seen. Patients were followed-up by an independent clinical researcher until: their discharge from the hospital, the clinical resolution of their diseases in response to medical therapy, or until a positive confirmation of infection/inflammation was evi-

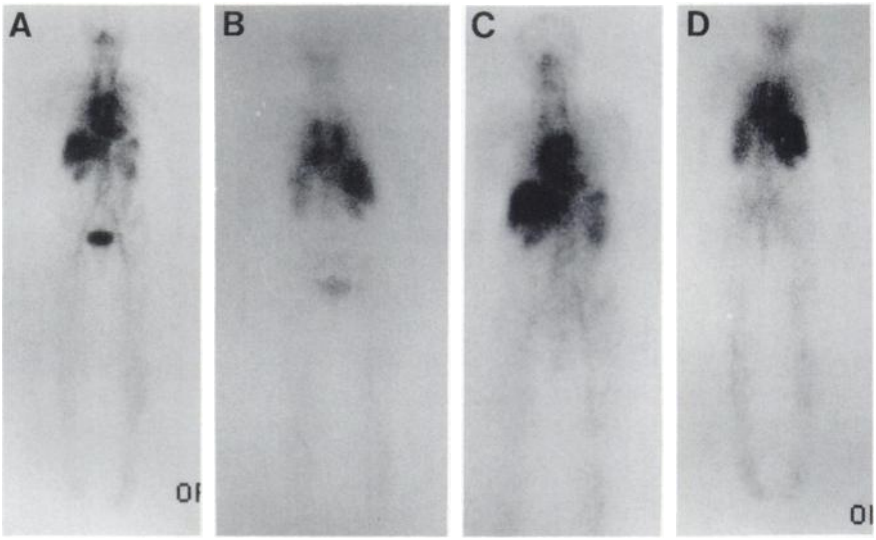


**FIGURE 1.** Whole-body $^{111}$In-IgG scans showing normal distribution of the radiotracer in early and delayed images. (A) Early anterior, (B) early posterior, (C) delayed anterior, and (D) delayed posterior.

dent by surgery, percutaneous drainage, positive culture of pertinent samples, conventional imaging techniques or postmortem examination.

Based on this analysis, the results were classified as follows:

*True-positive* when the scan showed an abnormal area of focally increased uptake of $^{111}$In-DTPA-IgG and demonstration of focal infection/inflammation at the same site within seven days of the scan. (Except Patients LG013 and JB016 who had lesions confirmed beyond 7 days based on the clinical course and diagnostic tests).

*False-positive* when the scan showed abnormal area of increased uptake of $^{111}$In-DTPA-IgG but no infection/inflammation was demonstrated within 1 wk of the scan.

*True-negative* when the scan showed no abnormal uptake of $^{111}$In-DTPA-IgG and no focal infection/inflammation was demonstrated at follow-up within 1 wk of the scan.

*False-negative* when the scan showed no abnormal uptake of $^{111}$In-DTPA-IgG, but there was demonstration of infection within 1 wk of the scan.

*Unevaluable* when the results of the scan were either normal or abnormal, but clinical information could not confirm a final diagnosis.

## RESULTS

In all patients studied, no adverse reactions were detected nor were there any significant change seen in the baseline laboratory studies.

Indium-111-DTPA-IgG normally distributes within the heart, lung, major blood vessels, the liver, kidneys, bladder, mucosae of the nose and vagina, and male external genitalia on early images. Activity in the heart, lung, major vessels, nose and genitalae decreases on delayed images if there is no infection/inflammation in these organs. No activity is normally seen within the gastrointestinal tract (Fig. 1).

Of the 40 patients studied, 22 were female and 18 were male, age range from 22 to 84 yr. The primary sites of inflammation/infection were: lung (Fig. 2), 14 patients; spine and/or extremities (Fig. 3), 11 patients; female reproductive organs (Fig. 4), 13 patients; paranasal sinuses, 2 patients; kidney and urinary tract, 1 patient; and peritoneum, 1 patient. Of the 40 patients, 3 were unevaluable due to: (a) dose infiltration, (b) lung biopsy nonconclusive of suspected sarcoidosis, or (c) activity from $^{67}$Ga study 6 days prior that interfered with $^{111}$In peaks. Of the 37 evaluable patients, DTPA-IgG uptake scan results were positive in 37 patients (100%). The final diagnosis and correlation is shown in Table 1.

The study demonstrated that infection or inflammation could be localized within 9 ± 3 hr in all but one patient (who was scanned only at 24 hr). Two patients showed slightly better localization at 24 ± 4 hr than that at 9 ± 3 hr. One patient (EP039) demonstrated this finding predominantly in the lungs (Fig. 2).

Delayed images beyond 24 hr were performed in one patient (040) who had a positive IgG scan at early imaging (6 hr), but in whom there was doubt if an abscess was present to explain her fever because she was 2 wk post-

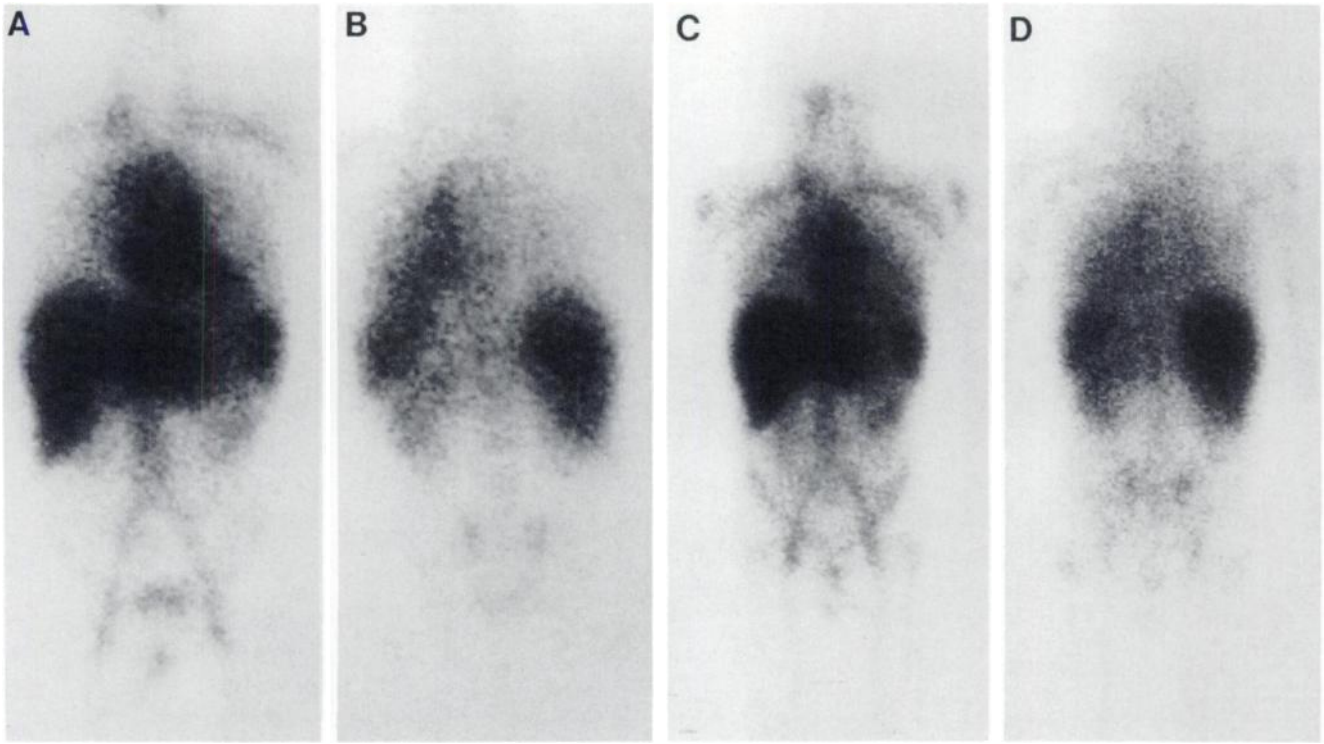


**FIGURE 2.** Early anterior (A) and posterior (B) whole-body scan of Patient 039 with pulmonary tuberculosis are compared to delayed anterior (C) and posterior (D) images to show increasing diffuse bilateral accumulation in the lungs over the 24-hr period.

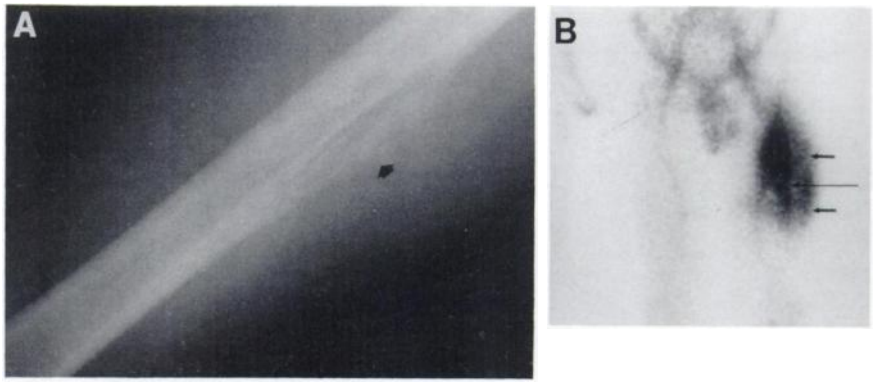


**FIGURE 3.** (A) X-ray of left femur showing lytic cortical lesion and ill defined periosteal reaction of the proximal and lateral aspect of the diaphysis of the femur consistent with osteomyelitis with overlying soft-tissue swelling. (B) Early $^{111}$In-IgG scan shows intense uptake in the soft tissues (short arrows) and longitudinally in the topography of the left femur (low arrow).

surgery for neocystectomy. In this case, the patient returned and a persistent positive scan promoted a transvaginal drainage of 200 cc of seropurulent material. This was followed by defervescence of the fever.

SPECT images were useful especially in the cases where pelvic inflammatory disease was the clinical diagnosis (Fig. 4). In these cases, although the planar images showed areas of increased uptake greater than the physiologic distribution, SPECT was better able to delineate the presence of abscess(es).

The radiopharmaceutical demonstrated the presence of infection under a variety of settings. This appears to be true for acute infections (14 patients with disease of less than 1 wk duration) subacute infections (14 patients with disease of 1–4 wk duration) and chronic infections (9 patients with illness of greater than 4 wk duration).

## DISCUSSION

In the 40 patients studied, no adverse reactions or side effects were encountered, confirming the previous high safety record for the product. Possible associated risks of PRI#0001-A-1 are those related to allergic reactions to the intravenous use of the IgG immunoglobulin. As such, it is contraindicated in patients with selective IgA deficiency who possess antibodies to IgG and in patients who have had severe systemic reaction to the intravenous or intramuscular administration of human immunoglobulin when therapeutic doses of 2 g or more are given. These reactions are rare, however, occurring in less than 1% of patients who are not immunodeficient. Moreover, they can be readily treated by stopping the infusion and giving an antihistamine or epinephrine if needed.

This study supports preliminary reports indicating a higher detection rate obtained by various authors utilizing a similar technique but different preparations (*10,16,21, 24*). The source of the IgG for the product used in this study is Gamaimmune® that has been evaluated extensively in humans for the treatment of a number of immunodeficiency syndromes and some autoimmune disorders, such as idiopathic autoimmune thrombocytopenic purpura, with a good safety record (*18,19*).

According to the manufacturer, a trial conducted on normal males demonstrated that an administered dose of 2.4 mCi of $^{111}$In-DTPA-IgG limits the exposure of sensitive organs (ovaries, testes, eyes and bone marrow) to less than 3 rads and other organs to less than 5 rads (*20*). When compared to $^{111}$In-labeled granulocytes, the dosimetry considerations for $^{111}$In-DTPA are more favorable, allowing a larger amount of $^{111}$In-chloride to be administered and resulting in reduced imaging time and better count statistics (*15,21*). In addition, a high ratio (6:1) of localization in abscesses relative to soft tissues has been shown that may account for improved image contrast (*22*).

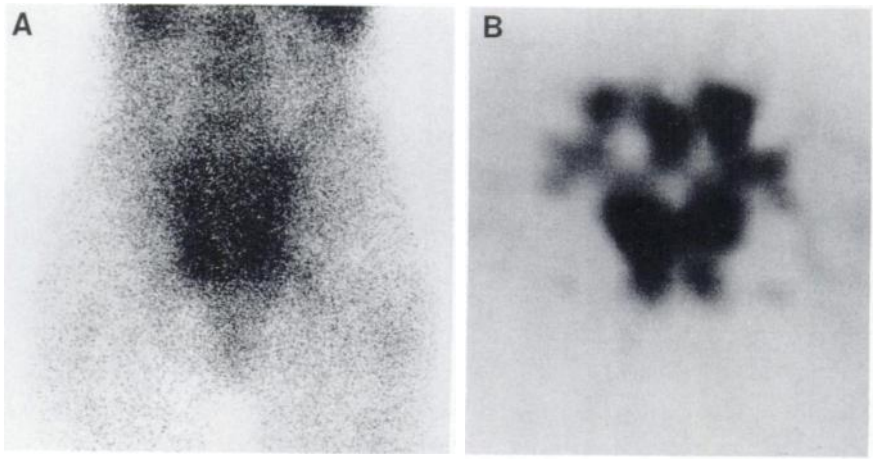


**FIGURE 4.** (A) Delayed posterior planar pelvic scan in Patient 001 shows focal bilateral increased uptake in the pelvis with pelvic inflammatory disease and bilateral tuboovarian abscesses. (B) Coronal SPECT done at approximately 18 hr postinjection shows the pelvis of a patient with well-defined bilateral focal uptake consistent with a clinical diagnosis of bilateral tuboovarian abscess.

**TABLE 1**
Results of Patients Studied

| Patient | Clinical diagnosis | Proof of diagnosis | $^{111}$In-IgG | Final assessment |
|---|---|---|---|---|
| HS 001 | Bilateral tuboovarian abscess | US, response to Rx | + | TP |
| WV 020 | Bilateral tuboovarian abscess | US, response to Rx | + | TP |
| BC 024 | Bilateral tuboovarian abscess | US, response to Rx | + | TP |
| SL 014 | bilateral tuboovarian abscess | US, response to Rx | + | TP |
| GG 017 | Pelvic inflammatory disease, Bilateral tuboovarian abscess | US, response to Rx | + | TP |
| VM 012 | Pelvic inflammatory disease, pelvic mass | US, response to Rx | + | TP |
| TJ 019 | Pelvic inflammatory disease, Pelvic mass | US, response to Rx | + | TP |
| FW 005 | Pelvic inflammatory disease | US, response to Rx | + | TP |
| RD 011 | Pelvic inflammatory disease | US, response to Rx | + | TP |
| LG 013 | Pelvic inflammatory disease | KUB, US, response to Rx | + | TP |
| WV 018 | Pelvic inflammatory disease | US, response to Rx | + | TP |
| CO 040 | Pelvic abscess | CT, surgery, response to Rx | + | TP |
| MR 004 | Endometritis post abortion | US, response to Rx | + | TP |
| QA 021 | *Pneumocystis carinii* pneumonia | Lung biopsy | + | TP |
| DB 038 | *Pneumocystis carinii* pneumonia | Lung biopsy, CxR | + | TP |
| PT 027 | *Pneumocystis carinii* pneumonia, tuberculosis | CxR | not done | Unevaluable |
| MJ 015 | Bronchopneumonia | CxR. sputum culture | + | TP |
| LS 025 | Pneumonia CMV, tuberculosis | CxR, lung Bx, AFB and culture | + | TP |
| CE 026 | Tuberculosis | Lung biopsy, CxR | + | TP |
| SM 029 | Tuberculosis | CxR, AFB and culture | + | TP |
| ET 031 | Tuberculosis | CxR, AFB and culture | + | TP |
| JM 033 | Tuberculosis | Pleural biopsy | + | TP |
| EP 039 | Tuberculosis | CxR, AFB and culture | + | TP |
| JA 030 | Tuberculosis, bronchiectasis | AFB culture, CxR | + | TP |
| TB 032 | Sarcoidosis | $^{67}$Ga scan, lung biopsy non-conclusive | − | Unevaluable |
| GF 006 | Osteomyelitis left femur | x-ray, drainage culture | + | TP |
| EB 009 | Osteomyelitis right femur | Bone scan | + | TP |
| JB 016 | Osteomyelitis left foot | x-ray | + | TP |
| RM 036 | Osteomyelitis right shoulder | CT | + | TP |
| GM 002 | Septic arthritis left hip | CT, drainage culture + surgical | + | TP |
| CJ 022 | Left subclavian and left buttock abscesses S/P facsiotomy right arm compartment syndrome | $^{67}$Ga scan, bone scan, drainage cultures | + | TP |
| CC 023 | Cellulitis right arm | Drainage culture, response to Rx | + | TP |
| LH 007 | Right calf hematoma | CT venogram | + | TP |
| HM 010 | Right ankle synovitis | Bone scan, x-ray, surgery | + | TP |
| AD 037 | Infected laminectomy | Drainage culture | + | TP |
| DF 028 | Compression fracture L4-L5 | MR of spine | + | Unevaluable |
| GJ 008 | Sinusitis (ethmoid and maxillary) | CT, surgery | + | TP |
| DY 034 | Sinusitis (maxillary) | x-ray response to Rx | + | TP |
| TG 003 | Budd-Chiari syndrome peritonitis | Liver biopsy, Ga scan | + | TP |
| TB 035 | Hydronephrosis, urinary tract infection | IVP, urine culture | + | TP |

CMV = cytomegalovirus, Rx = therapy, and AFB = acid fast bacilli stain.

The mechanism of localization of IgG at sites of infection/inflammation is yet to be defined and various hypotheses have been suggested. One hypothesis is that there could be specific binding of the Fc fragment of the immunoglobulin G to the Fc receptors (*7,11*). Another is that nonspecific binding of the immunoglobulin G may occur by oxidative cross-linkage of IgG by polymorphonuclear leukocytes (*23*). More recently, some experiments indicate that although the Fc segment plays a role microautoradiography has demonstrated that IgG primarily localizes in the intercellular matrix (Rauh D, *personal communication*, 1991).

These preliminary results showing a 100% sensitivity in this selected group of patients are encouraging and support the findings of other investigators that $^{111}$In-DTPA IgG has a high detection rate for infection or inflammation. A

Phase III study, which increases the number and variety of infections/inflammatory disorders studied, would be useful in further assessing this particular preparation and its sensitivity and specificity as compared to other preparations currently being evaluated (*24,26*).

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