Studies on Gallium Accumulation in Inflammatory Lesions: I. Gallium Uptake by Human Polymorphonuclear Leukocytes

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The mechanism of ionic gallium-67 localization in inflammatory lesions was studied. Human polymorphonuclear leukocytes (PMN) had higher Ga-67 uptake than lymphocytes, whereas red blood cells had no affinity for Ga-67. Uptake by PMN showed temperature dependence, was independent of Ga-67 concentrations, and was not inhibited by metabolic inhibitors. However, its binding to PMN could be removed by trypsin but not by neuraminidase. These results are consistent with the hypothesis that the plasma membrane serves as a diffusion barrier and Ga-67 only binds to the surface of the PMN plasma membrane. When this membrane's permeability barrier was disrupted, as in heat-killed PMN, Ga-67 uptake increased markedly.

Experimental abscesses were induced with* E. coli *or turpentine in rabbits. Twenty-four hours after i.v. injection, only 20% of Ga-67 in abscesses was in fractions containing intact PMN, cell debris or bacteria; the remainder was in a soluble, non-cellular fraction (2,500-g supernatant).

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Since the initial observation of Edwards and Hayes (*1*) that gallium-67 was concentrated in the involved lymph nodes of a patient with Hodgkin's disease, the accumulation of Ga-67 by a variety of tumors (*2–5*), as well as in abscesses and other inflammatory lesions (*3–6*), has been well documented. Currently, carrier-free Ga-67 citrate is widely used as a tumor- and abscess-scanning agent. Much effort has been focused on the elucidation of the mechanism of Ga-67 localization in tumors, but the exact mechanism remains unclear. Little has been done to elicit the mechanisms of Ga-67 accumulation in abscesses or inflammatory lesions. Arseneau et al. (*7*) showed that granulocytes from patients with chronic granulocytic leukemia had higher Ga-67 uptake than lymphocytes from patients with chronic lymphocytic leukemia. Gelrud et al. (*6*) provided evidence that binding of Ga-67 within polymorphonuclear leukocytes (PMN) is responsible for its localization in areas of inflammation. Nothing is known, however, about the mechanism of Ga-67 uptake by neutrophils. In this study, we demonstrated that at least 50% of Ga-67 taken up by PMN was bound to the plasma membrane. In non-viable PMN, Ga-67 diffused across the plasma membrane and contributed to the localization of Ga-67 in abscesses.

MATERIALS AND METHODS

Materials. Carrier-free Ga-67 citrate and C-14 inulin were obtained commercially. Gallium was present as a complex formed from 9 ng of Ga-67 chloride ($\approx$ 2 mCi), 2 mg of sodium citrate, and 6.8 mg NaCl in an aqueous volume of 1 ml. This was diluted with 2.5% Na citrate and modified Hanks' solution, with the final medium including the desired concentration of Ga-67 (usually 2 μC/ml) in modified Hanks' solution containing 0.25% citrate (final pH 7.3). Commercial sources were also used for neuraminidase* (from *Cl. perfringens*, Type V, E.C. 3.2.1.18), trypsin (bovine pancreas, Type IV), bovine serum albumin, KCN, N-ethylmaleimide (NEM),

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Na azide, iodoacetate, Ficoll, sodium diatriazoate (Hypaque), Freund adjuvants, tryptic soy broth, and agar.

Isolation of human blood cells. Isolation of human polymorphonuclear leukocytes (PMN) and lymphocytes was performed as described previously (*8,9*). Briefly, venous blood was obtained from normal individuals. Leukocytes were isolated by dextran sedimentation of red blood cells, differential centrifugation, and NH_4Cl lysis of contaminating red cells. Leukocytes were then suspended in 10 ml modified Hanks' solution (with 5 mM glucose) (*10*), placed on top of a 10-ml Ficoll-Hypaque mixture, and centrifuged at 400 g for 40 min at 20°C, according to Boyum (*11*). The pellet, which consisted of 97–99% PMN, and the cells at the interface, which consisted of 85–90% lymphocytes by differential counting, were washed twice with modified Hanks' solution. Lymphocytes and PMN were diluted to a final concentration of 10 million cells/ml. Red blood cells (RBC) were isolated from heparinized venous blood of normal individuals. Plasma and buffy coat were removed after the whole blood was centrifuged at 2,000 g for 20 min. The packed red cells were washed twice with modified Hanks' solution and diluted to a final concentration of 300 million RBC/ml.

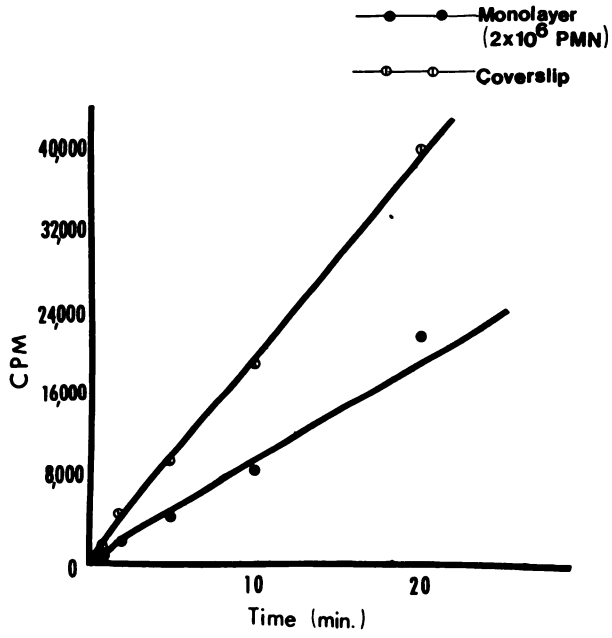

FIG. 1. Gallium-67 uptake by PMN monolayers and glass coverslips. PMN monolayers or glass coverslips were incubated with 2 μCi/ml Ga-67 for varying intervals.

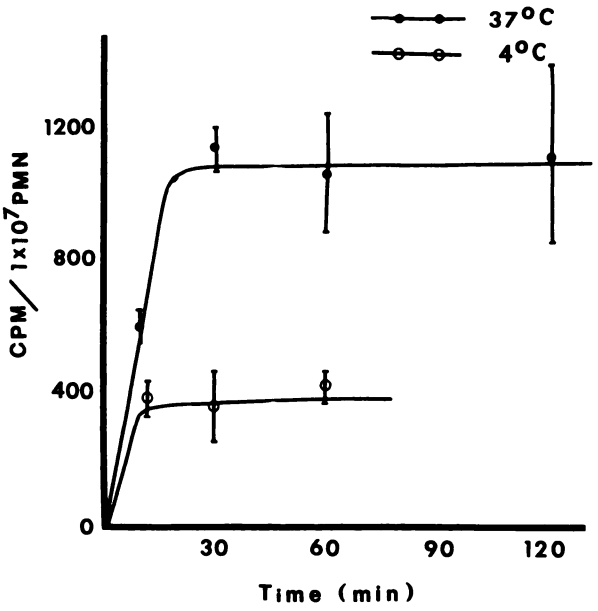

FIG. 2. Time course of Ga-67 uptake by human PMN. PMN were incubated with 2 μCi/ml Ga-67 for varying intervals at 4°C or 37°C. Results were expressed as Ga-67 uptake (cpm) per 10 million PMN. (Mean ± standard error of mean) No. of experiments = 4.

Measurement of Ga-67 uptake by human PMN. Three different techniques were evaluated for this uptake measurement. First, the cell-monolayer technique developed by Hawkins and Berlin (*12*) was studied. Briefly, cell monolayers were formed on round glass coverslips (22 mm in diam, 2 million PMN/coverslip). Cell monolayers were then incubated with 0.5 ml of Ga-67 (2 μCi/ml) for various intervals. At the end of incubation, cell monolayers were washed in four 50-ml beakers containing ice-cold modified Hanks' solution. Control experiments were performed with coverslips, but no PMN monolayer, in identical fashion. As shown in Fig. 1, Ga-67 was taken up by PMN monolayers, as well as by coverslips. This might be due to Ga-67 sticking to the coverslips because of negative charges of glass surface. Similar results were obtained when coverslips were incubated with Ga-67 in plasma. Thus, Ga-67 bound tightly to the glass surface, which invalidated the results obtained from PMN monolayers.

The second method evaluated was the Millipore-filter technique described by Merz et al. (*13*). Here PMN were incubated with Ga-67 in 50-ml metabolic flasks. At the end of incubation, the cells were collected by passing the cell suspension through a 0.45-μ Millipore membrane using plastic syringes. The cells were further washed twice with 10 ml ice-cold modified Hanks' solution. Control experiments were done in the same fashion except that no PMN were used. Again, significant amounts of Ga-67 bound to the Millipore filter membrane even in the absence of

PMN. There were 3,410 counts per minute (cpm) retained on the Millipore filter after passing 1 μCi of Ga-67 (9×10^5 cpm) followed by washing twice with 10 ml modified Hanks' solution. This observation is consistent with that of Driedger (*14*) who found that gallium-67 sticks to various kinds of Millipore filters.

Because of this nonspecific binding of Ga-67, we finally adopted a centrifugation technique. PMN were incubated with Ga-67 in disposable 15-ml plastic centrifuge tubes in a water bath at 37° C with constant shaking. At the end of incubation, PMN were spun down at 2,000 g and 4° C for 10 min, then washed twice with 10 ml modified Hanks' solution. Preliminary experiments indicated that further washes did not significantly change the uptake of Ga-67 by PMN. Independent experiments, using C-14 inulin as an indicator for extracellular-fluid contamination, indicated that only two washes were necessary to wash off the incubation medium contaminating the initial pellet. In the absence of PMN, no Ga-67 remained in the tubes, thus showing no binding to the centrifuge tubes. The radioactivity was counted in an automatic scintillation well counter with a window setting of 80-320 keV.

For the study of the effects of various metabolic inhibitors, PMN were preincubated for 20 min at 37° C in the presence or absence of 1 mM KCN, Na azide, Na iodoacetate, or of 2 mM N-ethylmaleimide, Gallium-67 uptake was then determined. For the study of the effects of enzymes on Ga-67 uptake, PMN were preincubated with 2 μCi/ml Ga-67 for 30 min at 37° C; they were then incubated in the presence or absence of trypsin (1 mg/ml) or neuraminidase (0.05 units/ml). The radioactivity retained with PMN after this enzymatic treatment was determined. Gallium-67 uptake by heat-killed PMN was determined by heating PMN for various intervals in a 56° C water bath. The number of nonviable human PMN was determined by uptake of trypan blue (0.2% in modified Hanks' solution). Gallium-67 uptake was then determined as described above.

Animal studies. Female New Zealand white rabbits, weighing between 2 and 4 kg, were used for this purpose. For the production of bacterial abscesses, 10^{10} *E. coli* in 1 ml of Freund adjuvant were injected into the muscle of the inner aspect of one thigh. Control animals were injected with 1 ml of Freund adjuvant. For the production of sterile abscesses, 0.2 ml of turpentine were used. After three days, abscesses could be detected in animals injected with *E. coli* in Freund adjuvant or with turpentine, but not those injected with Freund adjuvant alone. Four to five hundred μCi of Ga-67 citrate were injected intravenously through a marginal ear vein. Twenty-four hours later, the lower extremities of the animals were scanned with a rectilinear scanner. Blood was obtained by cardiac puncture and the animal was then killed by i.v. injection of 5 ml pentobarbital (65 mg/ml). The thigh was dissected and the abscess excised. Care was taken to avoid contamination of blood in the excised specimen. Several specimens of muscle were obtained from the contralateral thigh for controls. Blood, muscle, and abscess were counted in the well counter, as described above. A standard solution of Ga-67 was prepared at the time of injection and counted at the same time, with same geometry, as the above specimens. The results were expressed as percentage of dose injected per gram of tissue or blood, and an abscess-to-muscle or abscess-to-blood ratio was calculated accordingly. For the study of the distribution of Ga-67 in abscesses, specimens of abscess from one animal were suspended in 10 ml modified Hanks' solution in plastic centrifuge tubes. The suspension was agitated using a vortex mixer for 5 min to disperse leukocytes and/or bacteria from the fibrous tissue. It then was filtered through gauze to remove the fibrous tissue, and the filtrate was centrifuged at 600 g for 10 min at 4° C. The supernatant was further centrifuged at 2,500 g for 10 min at 4° C. The 600-g and 2,500-g pellets were each washed twice with 10 ml modified Hanks' solution, and were finally resuspended in 2 ml modified Hanks' solution and counted in the well counter. Gram staining of the 600-g and 2,500-g pellets was also done and examined with light microscopy.

The statistical significance was determined based on Student's t test (*15*).

RESULTS

Gallium-67 uptake by human PMN, lymphocytes and RBCs. Figure 2 shows the results of a time course uptake of Ga-67 by human PMN. PMN had higher Ga-67 uptake at 37° C than at 4° C. Moreover, Ga-67 uptake reached a maximum after 30

TABLE 1. GALLIUM-67 UPTAKE BY HUMAN PMN, LYMPHOCYTES, AND RBC

	PMN	Lymphocytes	RBC
Ga-67 uptake			
Mean	1,401 (16)	998 (8)	20 (3)
Range	1,000–3,342	448–2,154	17–23

The figures give Ga-67 uptake (in cpm) per 10 million cells. Cells were incubated with 2 μCi/ml Ga-67 for 30 min at 37°C and washed twice with 10 ml modified Hanks' solution. Numbers in parentheses indicate the number of experiments. P value (lymphocyte vs. PMN) < 0.05.

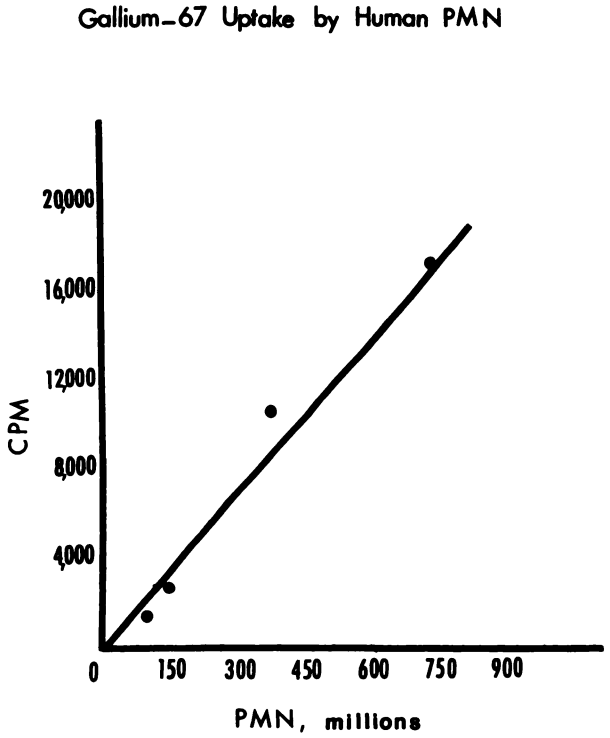

FIG. 3. Gallium-67 uptake by varying numbers of human PMN.

min of incubation. At this point, the total gallium uptake represents only 0.1% of the radioactivity present in the medium. All subsequent experiments were done with incubation for 30 min. Table 1 shows the results of Ga-67 uptake by PMN, lymphocytes, and RBC. PMN had higher Ga-67 uptake than lymphocytes, whereas RBCs had almost no affinity for Ga-67. Figure 3 shows that there was a linear relationship between the number of PMN and the amount of Ga-67 uptake. Since Ga-67 binds to plasma proteins (*16*), Ga-67 uptake by PMN also was studied using either serum or heparinized plasma instead of modified Hanks' solution. Similar results were obtained.

Effect of various concentrations of Ga-67 and metabolic inhibitors on Ga-67 uptake by human PMN. There are three possible mechanisms by which Ga-67 could be taken up by human PMN: (a) simple diffusion, (b) a carrier-mediated transport process, or (c) binding of Ga-67 to membrane components. In order to investigate these possibilities, PMN were incubated with various concentrations of Ga-67. As shown in Table 2, Ga-67 uptake by PMN was practically the same at Ga-67 concentrations from 0.2 to 5 μCi/ml. Thus the uptake by PMN is unlikely to be due to simple diffusion. The effect of various metabolic inhibitors on Ga-67 uptake by PMN was also studied. If Ga-67 is transported by an active transport process, its uptake by PMN should be inhibited by metabolic inhibitors. As shown in Table 3, KCN, N-ethylmaleimide (NEM), Na azide, or iodoacetate had essentially no inhibitory effect on Ga-67 uptake by PMN.

TABLE 2. EFFECT OF VARIOUS GALLIUM CONCENTRATIONS ON GALLIUM-67 UPTAKE BY HUMAN PMN

	0.2 μCi/ml	1 μCi/ml	2 μCi/ml	5 μCi/ml
Ga-67 uptake	1,714	1,982	1,644	1,634

The results are expressed on the same basis as in Table 1, and are the means of two experiments.

TABLE 3. EFFECT OF METABOLIC INHIBITORS ON GALLIUM-67 UPTAKE BY HUMAN PMN

	Uptake (cpm/10^7 PMN)
Control	1,236
+ KCN (1 mM)	1,326
+ NEM (2 mM)	1,671
+ Na Azide (1 mM)	1,215
+ Na Iodoacetate (1 mM)	1,129

The results are expressed on the same basis as in Table 1 and are means of two experiments. PMN were preincubated for 20 min at 37°C in the presence and absence of metabolic inhibitors. At the end of preincubation, Ga-67 was added (final concentration = 2 μCi/ml).

Effects of trypsin and neuraminidase on the binding of Ga-67 to human PMN. Mertz et al. (*13*) have shown that brief trypsinization of lymphocytes labeled with Ga-67 removes about 50% of the Ga-67, suggesting that Ga-67 binds to the plasma membrane of lymphocytes. Since Ga-67 is a strongly charged cation, it is possible that it binds to some negatively charged moiety of the cell surface, such as sialic acid. Accordingly, the effects of trypsin and neuraminidase on the binding of Ga-67 to PMN were studied. We used bacterial neuraminidase, which cleaves sialic acids at 2–3′ and 2–6′ glycosidic bonds, since more than 80% of the sialic acids present in the PMN plasma membrane are in 2–6′ linkage (*17*). [Neuraminidase from influenza virus only cleaves 2–3′ glycosidic bonds (*17*)]. As shown in Table 4, neuraminidase had no effect on the binding of Ga-67 to PMN. We have previously shown that, at the concentration used, neuraminidase removes 40–60% of surface sialic acids from human PMN (*18*). In contrast, trypsin removed almost 50% of the Ga-67 bound to PMN. This is not due to a nonspecific effect of protein, since at the same protein concentration, albumin had no such effect.

Distribution of Gallium-67 in bacterial- and tur-

TABLE 4. EFFECT OF TRYPSIN AND NEURAMINIDASE ON GALLIUM-67 UPTAKE BY HUMAN PMN

	Ga-67 uptake		P values
	Mean	Range	(vs control)
Control	1,163 (3)	756–1,563	
+ BSA* (1 mg/ml)	1,072 (3)	698–1,306	>0.8
+ Trypsin (1 mg/ml)	655 (3)	442– 801	<0.05
+ Neuraminidase (0.05 units/ml)	1,131 (3)	710–1,446	>0.8

The results are expressed on the same basis as in Table 1. PMN were preincubated for 30 min at 37°C with 2 μCi/ml Ga-67. They were then incubated in the presence or absence of enzyme for 20 min at 37°C. The number in parentheses indicates the number of experiments. P values were obtained based on paired difference.

* BSA; bovine serum albumin.

pentine-induced abscesses. In view of the evidence presented in the accompanying paper (*19*), it was hypothesized that both PMN and bacteria might contribute to the localization of Ga-67 in abscesses. In order to determine their roles in this accumulation, in-vivo experiments were carried out in rabbits. Three days after injection of *E. coli* or turpentine, abscesses were palpable in the injected thigh. Rectilinear scans of the abscesses, 24 hr after i.v. injection of Ga-67, are shown in Fig. 4. High concentrations of Ga-67 were present in abscesses induced by either E. coli (Fig. 4A) or turpentine (Fig. 4B). Quantitative data were obtained at necropsy (Table 5). Abscess-to-muscle ratio and abscess-to-blood ratio were variable but high, however, there seemed to be no difference between these two groups of abscesses. It was also noted that only a small fraction

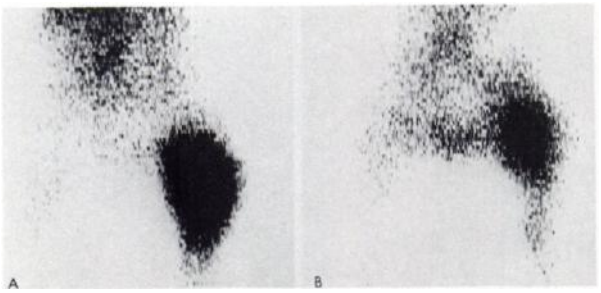

FIG. 4. Rectilinear scans of abscesses induced by *E. coli* (A) and turpentine (B). Abscesses were 3 days old and were scanned 24 hr after i.v. injection of Ga-67.

of Ga-67 present in the whole blood was associated with leukocytes (less than 14%). In *E. coli* abscesses, the 600-g fraction consisted predominantly of PMN, with some intracellular *E. coli* and some *E. coli* sticking to the surface of PMN. It contained 12–22% of the total Ga-67 present in the abscess (Table 6). The 2,500-g fraction consisted of free *E. coli* and PMN cell debris. It contained only 3.5–6% of the total Ga-67, the majority of Ga-67 being in the supernatant fraction. In turpentine-induced abscesses, the 600-g fraction consisted of PMN and it contained 15–16% of total Ga-67 present in the abscess. The 2,500-g fraction consisted of cell debris and contained only 2% of the total Ga-67. Again, the majority of Ga-67 was in the supernatant fraction. There are several possibilities for these observations: (a) abscesses might contain some as yet uncharacterized proteins, which bind Ga-67 avidly; (b) the soluble fraction containing most of the Ga-67 in the abscess might represent lysed PMN, since PMN reach the site of infection, ingest, and kill bacteria, and eventually die and are digested by their own lysosomal enzymes. In order to test this second possi-

TABLE 5. GALLIUM-67 UPTAKE BY BACTERIAL AND STERILE ABSCESSES

Rabbit No.	Dose injected (μCi)	Radioactivity (% dose/gram)			Ratio*		
		Blood	Muscle	Abscess	A/M	A/B	WBC/B
E. coli abscess							
1	586	0.081	0.005	0.244	49	3	0.136
				0.160	32	2	
2	442	0.090	0.005	0.126	25	1.4	0.023
3	443	0.012	0.0057	0.525	92	45	0.120
				0.371	65	31	
Turpentine abscess							
4	544	0.014	0.012	0.298	24	21	0.051
				0.214	17	16	
5	552	0.063	0.002	0.298	149	4.7	0.016
				0.229	114	3.6	

* A/M = abscess-to-muscle ratio; A/B = abscess-to-blood ratio. WBC/B = ratio of radioactivity present in white blood cells to that of whole blood.

TABLE 6. DISTRIBUTION OF RADIOACTIVITY IN ABSCESSES

	Radioactivity (%)		
Rabbit No.	600 g	2,500 g	Supernatant
E. coli abscess			
1	12	3.5	84.5
2	22	6	72
3	18.5	5.5	76
Turpentine abscess			
4	15	2	83
5	18	2	80

In *E. coli* abscesses, 600-g fraction consisted predominantly of PMN and some *E. coli*; 2,500-g fraction consisted of *E. coli* and cell debris.

In turpentine-induced abscesses, 600-g fraction consisted of PMN, and 2,500-g fraction consisted of cell debris.

bility, Ga-67 uptake by heat-killed PMN was studied.

Gallium-67 uptake by heat-killed human PMN. After heating in a 56° C water bath, the number of nonviable cells increased with increasing intervals of incubation, but the total number of PMN did not change. Thus, there was no loss of cells due to lysis.

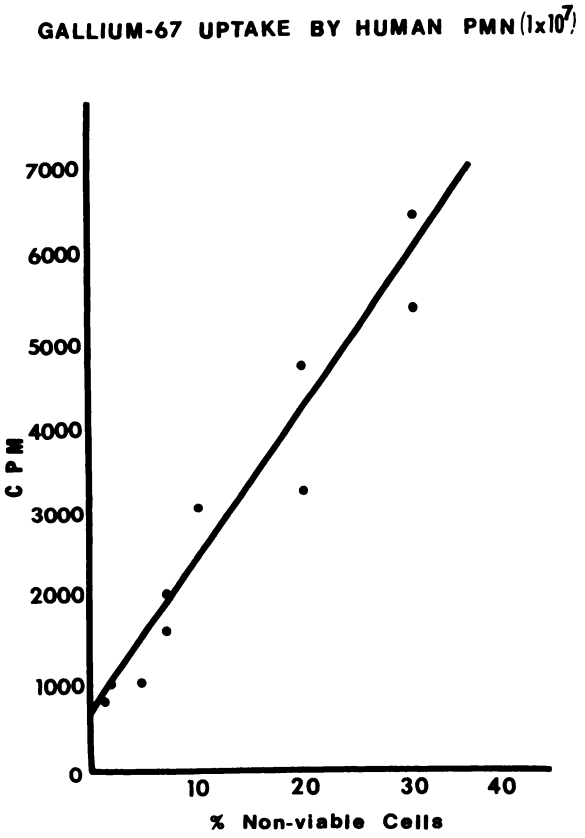

FIG. 5. Effects of nonviable PMN on Ga-67 uptake. Human PMN were heated at 56°C for various intervals and nonviable cells were determined by uptake of trypan blue. Gallium-67 uptake was then determined. Results are expressed as Ga-67 uptake (cpm) per 10 million PMN with various percentages of nonviable cells.

As shown in Figure 5, an increase in the number of nonviable cells was associated with a markedly increased uptake of Ga-67.

DISCUSSION

In-vitro study of Ga-67 uptake is hampered by the nonspecific binding of Ga-67 to various surfaces. As shown in this study, Ga-67 bound to glass surfaces as well as Millipore filter membranes. Gallium is a highly charged cation. In the absence of adequate concentrations of chelating agents such as citrate, it forms colloid and may precipiate out of solution on standing. Under our experimental conditions, the final concentration of citrate (0.25%) was far in excess of that required to keep gallium in solution. Because of the strong negative charge of the glass surface, it is possible that electrical charge is responsible for the retention of gallium on glass coverslips. Attempts to eliminate this nonspecific binding by using plasma were not successful. Our observation that Ga-67 also bound to Millipore filters is consistent with that of Driedger (*14*). Failure to recognize this nonspecific binding of Ga-67 may produce misleading information. We found that plastic centrifuge tubes did not have this undesirable property and were suitable for the study of Ga-67 uptake in vitro.

Uptake of Ga-67 was higher in human PMN than in the lymphocytes. In contrast, RBC had negligible affinity for Ga-67. These observations are consistent with those of Arseneau et al. (*7*) and Glickson et al. (*20*). In PMN, Ga-67 uptake was temperature-dependent. It was not inhibited by metabolic inhibitors. Its binding to PMN could be removed, however, by brief treatment with trypsin. These results are consistent with the hypothesis that the plasma membrane of PMN serves as a diffusion barrier and Ga-67 only binds to the surface of the plasma membrane. The fact that its uptake was not affected by different concentrations of Ga-67 (Table 2) suggests that at the concentrations tested (0.2 to 5 μCi), all the binding sites of the plasma membrane are saturated, since under these experimental conditions, less than 1% of Ga-67 present in the incubation medium was taken up by PMN. A similar conclusion was drawn by Mertz et al. (*13*) regarding the binding of Ga-67 to human lymphocytes. However, since they did not take into consideration the nonspecific binding of Ga-67 to a Millipore filter, their conclusion must be viewed with skepticism. The exact site of Ga-67 binding on the PMN plasma membrane is not clear, but we showed that it was not the surface sialic acid. In nonviable PMN, the plasma membrane diffusion barrier no longer exists and Ga-67 uptake increases markedly. It is postulated that under this condition

Ga-67 diffused across the disrupted plasma membrane and bound to intracellular components. A similar phenomenon is observed with p-chloromercury-benzene sulfonic acid (PCMBSA) (*8*). This sulfhydryl group inhibitor has a strong negative charge (sulfonyl group) and binds to surface -SH groups only. When the membrane permeability barrier of PMN is disrupted, however, PCMBSA binds to both intracellular and surface -SH groups, and its uptake by PMN increases thirtyfold (*8*). Intracellular localization of Ga-67 has been the subject of intensive investigations, but the results are conflicting. Investigators at Oak Ridge (*21*) have identified "lysosomal-like" granules as the primary sites of Ga-67 localization in a number of tumors and normal cells such as hepatocytes, Kupffer's cells, etc. In contrast, Orii (*22*) and Glickson et al. (*18*) fail to show preferential localization of Ga-67 in any subcellular organelle. Gel chromatography of homogenates of tumors reveals a spectrum of unidentified soluble proteins that appear to bind Ga-67 ionically (*23*). Anghileri (*24*), on the other hand, provided evidence for competitive binding of Ga-67 and Ca-45 by hydroxyapatite, phospholipids, and albumin.

After i.v. injection, Ga-67 binds to plasma proteins (*16*) as well as to leukocytes (*7*). As shown in this study, less than 14% of Ga-67 remaining in circulation 24 hr after i.v. injection was bound to leukocytes. Both free and protein-bound Ga-67 diffuse out of the circulation and are distributed throughout the entire body, notably in the liver, spleen, skeleton, and kidneys (*25*). In animals or patients with tumors or abscesses, high concentrations of Ga-67 are localized in these tissues.

Based on our present study, we propose the following mechanism for Ga-67 localization in abscesses and inflammatory lesions. After i.v. injection, Ga-67 binds to the surface membrane of PMN. These labeled PMN move out of circulation continuously to the site of inflammation. Gallium-67 also diffuses directly into the site of inflammation, where it is taken up by bacteria (see accompanying paper) (*19*). In addition, Ga-67 diffuses across the cell membrane of nonviable PMN and possibly binds to intracellular components. The relative roles of these processes in the localization of Ga-67 probably depend on the local factors within the abscess. It is also possible that some other components of the abscess bind Ga-67 and contribute to its uptake.

FOOTNOTE

* Neuraminidase, 0.5 U/mg protein. One unit liberates 1.0 μmol of N-acetyl-neuraminic acid per minute at pH 5.0, 37°C using N-acetyl neuramin-lactose as substrate. Trypsin, 10,000 BAEE U/mg. One BAEE unit = OD_{253} of 0.001/min with N-benzoyl-L-arginine ethyl ester as substrate at pH 7.6, 15°C.

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SNM ROCKY MOUNTAIN CHAPTER ANNUAL MEETING

April 6–8, 1977 **Hilton Inn** **Albuquerque, New Mexico**

CALL FOR ABSTRACTS

The Annual Scientific Meeting of the Rocky Mountain Chapter of the Society of Nuclear Medicine will be held Thursday through Saturday, April 6–8, 1977 at the Hilton Inn, 1901 University Boulevard, N.E., Albuquerque, NM 87125.

The program will include: Clinical Radiopharmacy; Investigation of IND Drugs; Special Technologist Session; Regular Scientific Session; Commercial Exhibits; Business Meeting; Tour of Los Alamos Scientific Laboratories.

The Clinical Radiopharmacy program will include exploring this new topic in depth and will include presentations by several guest speakers, as well as panels and discussions by participants. This part of the program is cosponsored by the University of New Mexico, College of Pharmacy and Radiopharmacy. Program coordinator is Buck A. Rhodes.

GUIDELINES FOR SUBMITTING ABSTRACTS

Abstracts accepted for the program will be published in the *Rocky Mountain Medical Journal* (except for those also accepted for publication in *JNM*). The deadline for submitting abstracts for the regular scientific session is:

FEBRUARY 15, 1978

Abstracts may be submitted simultaneously to both the Chapter and the National SNM offices. The original abstract and two copies (or three copies, if the original is submitted to national office) with supporting data attached to each should be sent to:

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