Polymeric Microspheres for Radionuclide Synovectomy Containing Neutron-Activated Holmium-166

Russell J. Mumper, Betty Jo A. Mills, U. Yun Ryo and Michael Jay

Division of Medicinal Chemistry and Pharmaceutics, Center of Membrane Sciences, and Division of Nuclear Medicine, University of Kentucky, Lexington, Kentucky

Poly-L-lactic acid (PLA) microspheres containing neutron-activated ^{166}Ho were investigated as potential agents for radionuclide synovectomy. Stable ^{165}Ho, complexed to acetylacetone (AcAc), was incorporated into PLA spheres by the solvent evaporation technique. Spheres prepared with the optimal mean particle size of 7.2 μm (range 2–13 μm) containing 25.4% ^{165}Ho-AcAc (9.1% ^{165}Ho) were irradiated in a high neutron flux to produce 31.1–36.0 mCi ^{166}Ho. In vitro human plasma studies showed that the irradiated spheres retained 99.0 ± 0.01% of the ^{166}Ho at 314 hr. In-vivo retention studies were conducted by administering irradiated PLA spheres with 257–591 μCi ^{166}Ho into the joint space of normal rabbits (n = 6). Biodistribution analysis and gamma camera analysis showed ^{166}Ho retention in the joint space after 120 hr of 97.7% ± 0.8% and 98.2% ± 2.4%, respectively, with no uptake by the lymph nodes. The ease with which the PLA spheres can be made in the optimal size range for later irradiation and their ability to retain the ^{166}Ho make them attractive agents for radionuclide synovectomy.

J Nucl Med 1992; 33:398–402

Radionuclide synovectomy is a noninvasive therapy for rheumatoid arthritis that has been investigated as an alternative to surgical synovectomy. Although surgical synovectomy can provide temporary relief lasting 2–5 yr (*1–3*), it is limited by its technical difficulty and requires prolonged hospitalization and rehabilitation for the patient (*4–5*). Radionuclide synovectomy consists of the intra-articular administration of a therapeutic radionuclide such as a beta emitter into a diseased joint to reduce inflamed synovium. This procedure was first reported by Fellinger et al. (*6*) in 1952 and has been used extensively in Europe (*7–12*). Early attempts to prepare colloidal or particulate agents of ^{90}Y (*13*) and ^{198}Au (*14*) were shown to be effective; however, undesirable leakage of radioactivity from the joint (25%–60% of the injected dose) resulted in radiation doses to the regional lymph nodes as high as 15,000 rads for ^{198}Au and 4,500 rads for ^{90}Y. It was found that excessive leakage of the colloids was due to the instability of the ^{90}Y and ^{198}Au systems with subsequent extra-articular leakage as well as the relatively small size of the colloids (*13–15*). Similar results were also obtained with colloids of ^{169}Er, ^{32}P, and ^{186}Re (*11–12, 16*). Larger colloids (1–5 μm) of ferric hydroxide macroaggregates (FHMA) labeled with ^{165}Dy were reported by Hnatowich et al. (*17*) in 1978 and since then, FHMA complexes with ^{90}Y (*18*) and ^{166}Ho (*19*) have been reported. Variable extra-articular leakage of 1%–13% was observed for the FHMA colloids and was also found to be dependent on the effective size of the radiocolloids, and also the stabilities of the FHMA and radionuclide-FHMA complexes. In addition, it was concluded that radioactive leakage from the joint can be reduced with the utilization of a radioisotope with a short half-life since a greater fraction of the decay occurs before possible leakage from the joint. Chinol et al. (*19*) concluded that the FHMA particles were suitable as carriers for shorter lived radioisotopes such as ^{165}Dy ($t_{1/2}$ = 2.3 hr) and ^{166}Ho ($t_{1/2}$ = 26.9 hr), however, Hosain et al. (*20*) concluded that ^{166}Ho was a more practical choice based on its longer half-life. Recently reported agents for radionuclide synovectomy including rhenium heptasulfide (*21*) and liposomes as carriers for radionuclides (*22*) have shown retentions of the radionuclides in normal rabbit knees of only 88.3% in three days and 63%–80% in one day, respectively. Currently, several new colloidal or spherical agents containing ^{153}Sm and ^{186}Re are being investigated in attempts to minimize extraarticular leakage (*23–25*). The results, to date, have been varied. In addition to the physical problems with stability, particulate size, and leaching, a practical consideration that hinders the future applicability of most of these agents is that high levels of radioactivity must be handled in their preparation. Their preparation involves several hands-on manipulations such as aliquoting, mixing, vortexing, centrifuging, and ultrasonication that are time consuming and increase the radiation exposure to the formulator. Furthermore, activities up to five times those needed for therapy must be used to

Received May 31, 1991; revision accepted Sept. 26, 1991.

For reprints contact: Michael Jay, PhD, Division of Medicinal Chemistry and Pharmaceutics, College of Pharmacy, University of Kentucky, Lexington, Kentucky 40536-0082.

prepare the colloids due to decay and preparation yield considerations.

We have reported on the potential use of biodegradable neutron activated poly-L-lactic acid (PLA) microspheres (10–40 μm) containing ^{166}Ho for the treatment of hepatic tumors (*26*). In this novel application, PLA microspheres containing sufficient amounts of stable ^{165}Ho, in the form of ^{165}Ho-acetylacetonate (^{165}Ho-AcAc), can be prepared in the optimal size range, fully characterized and later made radioactive by neutron bombardment. This paper reports on the application of smaller PLA spheres (2–13 μm) containing neutron activable ^{165}Ho for radionuclide synovectomy.

MATERIALS AND METHODS

Preparation and Characterization of PLA Spheres Containing Ho-AcAc

Holmium-166-AcAc was synthesized as described previously (*26*). PLA spheres containing ^{165}Ho-AcAc were prepared by the solvent evaporation technique (*26*). Briefly, 1.5 g PLA (57,000 MW; Henley Company, Montvale, NJ) and 0.9 g Ho-AcAc were dissolved in 30.0 ml chloroform and added to a continuous phase of 3% Polyvinyl alcohol (78,000 MW; Aldrich Chemical Company, Milwaukee, WI). Stirring at 1140 rpm was continued for 8 hr after which time the precipitated spheres were centrifuged at 2500 rpms for 10 min to remove the viscous continuous phase and filtered using a 3.0 μm filter. The filtered spheres were resuspended in 800 ml 0.1 N HCl for 2 min to remove the unincorporated ^{165}Ho-AcAc, refiltered, and washed with 800 ml of deionized H_2O. Six subsequent filtrations using fresh 3.0 μm filter paper assured optimal particle size. The particle size distribution of the microspheres was determined by optical microscopy (Nikkon Optiphot; Scientific Instruments, Carpentersville, IL). A ^{252}Cf neutron source (neutron flux = 10^6 n/cm^{2}s; University of Kentucky Chemistry Department) was used to determine the percentage of ^{165}Ho-AcAc incorporated in the microspheres by irradiating standards and prepared PLA spheres, and counting the irradiated samples in a gamma counter (60–100 keV window; Packard Model No. 101750, Minaxi Auto-Gamma 5000 series). The density of the PLA spheres with ^{165}Ho-AcAc was measured by volume displacements in water and air.

High Neutron Flux Irradiations of PLA Spheres

For in-vitro and in-vivo analysis, 50 mg PLA spheres and 150 mg inositol, used as a diluent, were irradiated in the TRIGA Reactor at the University of Illinois (reactor power = 600 kW; thermal neutron flux = 8.88×10^{12} n/cm^{2}s with 8% epithermal neutron flux) for 128 min to produce 31–37 mCi ^{166}Ho. The ^{166}Ho activities of irradiated samples were determined by a radioisotope calibrator (Model CRC-12, Capintec Inc., Ramsey, NJ).

In-Vitro Studies

Irradiated samples containing inositol and PLA spheres with 1120–1680 μCi ^{166}Ho were added to 1.0 ml human plasma in which the inositol was dissolved and the spheres easily suspended. These 1.0 ml suspensions were then contained in 4-in. segments of pure regenerated natural cellulose dialysis membrane (Fisher, Spectra/Por 7 membranes 50,000 MW cut-off) and placed in 24.0 ml human plasma maintained at constant temperature of 37°C. Aliquots of plasma were removed up to 410 hr and later counted for ^{166}Ho activity. At the end of the release study, the percentage of ^{166}Ho retained in the spheres was calculated by dividing the activity in the spheres by the total of the activities in the spheres, plasma, and adhering to the dialysis membrane.

Injection of PLA Spheres into Rabbit Knees

Six normal New Zealand White rabbits weighing 3.7–4.8 kg were used for in-vivo retention studies within the guidelines of the protocol approved by the Institutional Animal Care and Use Committee (IACUC). 4.5 mCi ^{99m}Tc-hydroxymethylene diphosphonate (^{99m}Tc-HDP) were injected intravenously into two rabbits and allowed to accumulate in the skeletal system 3 hr prior to anesthetization. The purpose of the ^{99m}Tc agent was to verify that injections of spheres deposited in the synovial space. All six rabbits were anesthetized with an intramuscular injection of ketamine HCl U.S.P. (35 mg/kg; Aveco). The right leg of the rabbits was shaved from mid thigh to below the knee and Clinidine solution (Povidone Iodine 10% U.S.P.; Clinipad Corp.) was applied to the knee area. Irradiated PLA sphere samples with 1.7–2.4 mCi ^{166}Ho and inositol were suspended in 2.5 ml of normal saline with 4 drops of Tween-80 (Aldrich Chemical Company, Milwaukee, WI), vortexed for 10 sec, and counted in the dose calibrator to determine the ^{166}Ho activity in the administration vial. From the 2.5 ml suspension of PLA spheres with ^{166}Ho, 1.0 ml was drawn using a sterile 3.0 ml syringe with a 23 g needle. A separate sterile 23 g needle attached to a sterile syringe containing normal saline was used to enter the knee joint capsule via various approaches (anterior, anteromedial, or anterolateral). The injection technique did not appear to alter the distribution of the spheres nor their leakage from the joint space. Correct localization was verified by easy withdrawal of synovial fluid. Without disturbing needle placement, the syringe containing saline was exchanged for the syringe containing the suspended PLA spheres with ^{166}Ho. The spheres were administered into the joint capsule and the syringe and needle were withdrawn and counted in a dose calibrator.

Imaging

Immediately after administration, rabbits were imaged using a Siemens Digitrac 37 Gamma Camera (Siemens Gammasonics Inc., Des Plaines, IL) fitted with a pinhole collimator. A 20% window was centered around the 81 keV gamma ray of ^{166}Ho and for detection of ^{99m}Tc, a 15% window around the 140 keV gamma ray of ^{99m}Tc was used. For all images, the rabbits were placed in restrainers with their right leg extended and the knee bent at a 30-degree angle. The pinhole collimator was positioned laterally above the right knee of the rabbit and 128 × 128 digital matrix images were obtained over 5 min. The position of the rabbit below the collimator was carefully measured and reproduced prior to each image. Regions of interest were defined using ACE Version 1.15m (Cardiac Medical Systems, Corp.) as well as background area outside the body of the rabbit. After injection of the PLA spheres, the two rabbits that had ^{99m}Tc-HDP administered were first imaged in the ^{99m}Tc window, and then in the ^{166}Ho window (without moving the rabbit). The remaining four rabbits that received PLA spheres were imaged only in the ^{166}Ho window. Background and decay-corrected counts per pixel acquired at time of injection, 24, 48, 72, 96, and 120 hr were determined and calculated as percent injected activity retained in the knee over time.

Biodistribution Studies

The rabbits were kept in metabolism cages so that their urine and feces could be collected. One milliliter of blood was drawn daily from an ear vein of each rabbit and counted for ^{166}Ho activity. It was assumed that total blood volume for the rabbit was 57.7 ml/kg (*27*). Biodistribution data for ^{166}Ho were acquired by killing the rabbits at 44 hr (n = 1) or 120 hr (n = 5) after administration of PLA spheres. The liver, spleen, gall bladder, heart, lungs, femur, right and left inguinal lymph nodes, the right knee, and a sample of blood were removed, weighed, and counted for activity in a NaI(Tl) well-counter.

RESULTS

Analysis of the Prepared and Irradiated Microspheres

^{252}Cf low neutron flux irradiation of PLA spheres and ^{165}Ho-AcAc standards showed that the prepared PLA spheres consisted of 25.4% ± 0.3% w/w ^{165}Ho-AcAc or 9.1% ± 0.1% w/w of the stable neutron activable ^{165}Ho (Ho-AcAc is 35.7% w/w ^{165}Ho). Optical microscopy analysis (40X magnification) of pre-irradiated spheres revealed a monodisperse distribution with a diameter range of 2–13 μm. Particle diameter distribution of a representative batch is shown in Figure 1. Volume displacement measurements showed that PLA spheres with ^{165}Ho-AcAc had a density of 1.4 g/ml.

In-Vitro Analysis

In Figure 2, the in-vitro retention profiles of five irradiated PLA sphere batches are shown. These types of retention profiles were consistent with previous studies where an initial release of 0.2%–0.5% ^{166}Ho activity in the first 8 hr was observed followed by stabilization with variable final degradation occurring at about 320–350 hr (*26*). After 314 hr (11.7 half-lives of ^{166}Ho; only 0.03% of the initial ^{166}Ho study activity remained), irradiated PLA spheres retained >99% of the initial ^{166}Ho activity.

In-Vivo Biodistribution and Imaging Analysis

Bone images of the lateral right knee were taken and markers were placed on the computer screen at the joint space and, parallel to that, above the condyles. Joint images following PLA sphere injection, taken without moving the rabbit at the time of image or the markers on the computer screen at time of photography, proved that activity was in the suprapatellar pouch and the posterior pouch.

The net activity injected into the rabbit knees was 257–891 μCi ^{166}Ho. Utilizing density and average particle size information for the PLA spheres, the mass and number of spheres administered was calculated to be 4.2–16.6 mg and (3.0 ± 1.6) × 10^7 particles, respectively. The mean data acquired with the gamma camera (Table 1) show a loss of 1.5% ^{166}Ho in the first 24 hr of the study with stabilization over the next 4 days. For all images, no organ or bone uptake of leached ^{166}Ho was observed. This excellent retention compares very well with in-vitro results. Blood samples drawn daily and corrected for total rabbit blood volume showed that 93.4% ± 4.2% of the total blood activity was collected by the end of day 2. Only one of the five rabbits killed at 120 hr had detectable ^{166}Ho in the blood which corresponded to only 0.04% of the initial activity administered. Urine collected over the 120-hr study had a similar trend with 83.4% ± 3.7% of total urine activity collected by the end of Day 3. Biodistribution analysis of the rabbits killed at 44 hr (n = 1; rabbit did not fully recover from anesthesia) and 120 hr (n = 5) are depicted in Table 2. It appears that the majority of ^{166}Ho leached from the joint occurs in the first 44 hr after administration which was similar to the small initial release observed in the in-vitro retention studies. In all rabbits killed at 120 hr, no ^{166}Ho activity was observed in the feces or inguinal lymph nodes. Of the 2.3% ^{166}Ho that leached from the knee (n = 5) over 120 hr, greater than two-thirds was found in the kidneys or urine. Radiation doses to the liver and kidney of rabbits were calculated based on the method of Neves et al. (*28*) assuming the following: the source and target regions are overlapping volumes, uniform distribution of the radionuclide, effective half-life equals the physical half-life, and the absorbed dose is due to beta particles only. After 120 hr, the rabbit (n = 5) liver and kidney dose per mCi of ^{166}Ho injected was 1.9–3.7 rads/mCi and 1.6–8.9 rads/mCi, respectively. Radiation doses to other organs were significantly less.

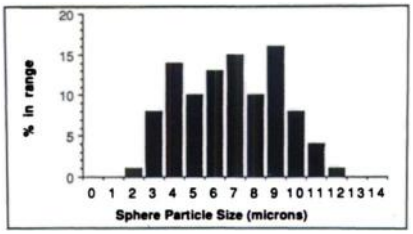

FIGURE 1. Particle size distribution of a representative batch of PLA spheres with Ho-AcAc.

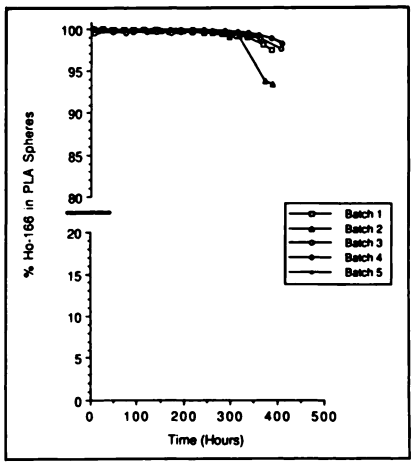

FIGURE 2. In-Vitro retention profiles of ^{166}Ho in PLA spheres in human plasma at 37°C.

TABLE 1
Percentage of Administered ^{166}Ho Remaining in Normal Rabbit Knees by Gamma Camera Analysis

Time after administration (in days)					
0 (n = 6)	1 (n = 6)	2 (n = 6)	3 (n = 5)	4 (n = 5)	5 (n = 5)
100	98.5 ± 1.7	98.6 ± 1.9	98.1 ± 2.9	99.0 ± 1.2	98.2 ± 2.4

TABLE 2
Biodistribution of ^{166}Ho After Administration of PLA Spheres in the Knees of Normal Rabbits

	Time after administration	
	120 hr (n = 5)	44 hr (n = 1)
	% Administered dose	
Right knee	97.7 (97.9–98.3)	98.30
Liver	0.6 (0.4–0.7)	0.36
Lung	0.02 (0.0–0.07)	0.01
Heart	0.001 (0.0–0.003)	0.02
Spleen	0.01 (0.003–0.01)	0.02
Gallbladder	0.001 (0.0–0.002)	0.33
Kidney	0.2 (0.1–0.4)	0.44
Urine	1.40 (0.8–1.5)	0.02
Right lymph nodes	ND	0.002
Femur/Bone marrow	0.11 (0.04–0.2)	0.49
Left lymph nodes	ND	0.0009
Blood	0.01 (0.0–0.04)	ND
Feces	ND	ND

ND = none detected.

DISCUSSION

The reported use of ^{166}Ho as a therapeutic radionuclide has increased in the last few years due to its favorable nuclear properties (*26,28–29*). Its half-life of 26.9 hr is long enough to eliminate logistic problems encountered with the short-lived ^{165}Dy, but is sufficient to provide a high radiation dose rate. The advantages of high dose rates for radiotherapeutic treatments are well known (*30–31*). The maximum soft-tissue penetration of a beta particle (E_{max} = 1.84 MeV) emitted from ^{166}Ho is 8.4 mm with an average of 3.3 mm. This appears ideal for treatments of inflamed knee synovium, which may become 1–7 mm thick depending on the severity of the disease (*20*).

Stable ^{165}Ho (100% natural abundance; neutron capture cross-section = 64 barns), in the form of ^{165}Ho-AcAc, can easily be incorporated into PLA spheres under non-hazardous conditions for later irradiation in a high neutron flux. Previous studies have shown that high neutron flux irradiations of PLA spheres have no effect on particle size and that irradiated spheres demonstrated in-vivo biodegradability (*26*). Noble et al. (*15*) found that particles in the 2–5 μm range were most suitable for injection into an inflamed joint. Davis (*32*) and Ratcliffe et al. (*33*) reported, however, that radiocolloids <5 μm were phagocytized by synovial cells. Particles larger than cells would be less effective, so therefore, spheres in the range ~5–10 μm were designed for optimal retention in the joint space.

Normal rabbit knees were used for our study since prior work by Venkatesan et al. (21) showed no significant difference in the mean retention of $^{186}Re_2S_7$ in normal and arthritic rabbit knees. Also, Ratcliffe et al. (*34*) concluded that there was no difference in the retention of ^{131}I-albumin microspheres (3.5 ± 1.7 μm) in normal or arthritic joints.

Analysis of the biodistribution data provides good evidence for the mechanism by which activity leaves the knee. Absence of ^{166}Ho in the lymphatic system after 120 hr is indicative of the fact that intact PLA particles do not escape the joint space. Free ^{166}Ho, a known bone-seeking element, could leave the joint space and deposit in the skeleton system, however, femur and bone marrow uptake of ^{166}Ho accounted for only 0.04%–0.2% of the injected dose and only 4.8% of the total leached activity. This low bone uptake led to overall biodistribution data that correlated well with the data obtained by gamma-camera analysis. A probable mechanism for ^{166}Ho leaving the joint space involves the binding of cationic ^{166}Ho to biological macromolecules with subsequent deposition in the body where the molecules are known to associate. Therefore, if $^{166}Ho^{3+}$ chelated with large molecules such as albumin, the ^{166}Ho would be found in the vascular system. If small compounds chelated with $^{166}Ho^{3+}$, activity would be excreted rapidly in the urine. The data did, in fact, show that 61.4% of the leached ^{166}Ho or 1.4% of the injected dose was found in the urine and that the majority of the activity leached was excreted in the first 3 days of the study.

Radiation doses to human knee, liver, and kidney can be estimated using the method of Neves if the rabbit data is extrapolated to humans. An 18.6-mCi dose of PLA spheres into the knee would deposit 10,000 rads to a 100-g synovial space corresponding to 537 rads/mCi ^{166}Ho injected. Leakage of activity to the liver (1702 g) and kidneys (303 g) would result in total radiation doses of only 0.2 rads/mCi and 0.3 rads/mCi ^{166}Ho injected, respectively. This calculation of the radiation dose to the synovial space correlates very well with previous dose estimates for other radiocolloids (*17,28–29*). In addition, the estimated radiation dose to the regional lymph nodes, considered a size effect of the particles, and bone marrow, considered a leaching effect, is small.

Neutron activable PLA spheres with ^{165}Ho may provide a potential improvement over ^{166}Ho-FHMA as an agent for synovectomy since the former: (1) may easily be prepared in the optimal particle size under non-hazardous conditions and later made radioactive and (2) is better able to retain the ^{166}Ho and remain intact in the joint space. A study involving the injection of ^{59}Fe and ^{166}Ho dual labeled FHMA into normal rabbit knees has been reported (*19*). Extra-articular leakage of 18.5% of the initial ^{59}Fe activity was observed after 5 days and it was concluded that the instability of FHMA is accelerated when complexed to ^{166}Ho.

In arthritic joints of fingers and toes, where the inflamed synovial membranes are thinner, a radionuclide emitting a lower-energy beta-particle would be more appropriate to minimize radiation doses to bone and cartilage (*20*). We have already reported on the potential use of microspheres containing neutron-activated ^{165}Dy ($t_{1/2}$ = 2.3 hr; E_{max} = 1.29 MeV) for therapy (*34*). This work has been extended to include spheres containing the neutron-activated rare-

earths ^{153}Sm ($t_{1/2}$ = 46.8 hr; E_{max} = 0.8 MeV) and ^{140}La ($t_{1/2}$ = 40 hr; E_{max} = 1.31 MeV). These activated rare-earths have nuclear properties that would enable them to serve as suitable alternatives in the case where less penetrating beta-particles are needed for therapy.

CONCLUSIONS

PLA spheres containing sufficient masses of neutron-activable ^{165}Ho can be prepared under non-hazardous conditions and irradiated at a later time to produce therapeutic amounts of ^{166}Ho. The irradiated particles show excellent ability to retain the excapsulated ^{166}Ho and theoretically can deliver a large radiation dose to diseased synovium while minimizing doses to other organs. The potential clinical use of this agent is very promising since its preparation does not require the handling of large amounts of activity and the half-life of ^{166}Ho eliminates the necessity for being close to a reactor.

ACKNOWLEDGMENTS

This work was supported in part by grant 1N-163 from the American Cancer Society, the National Science Foundation grant R11-8110671, and the Commonwealth of Kentucky through the Kentucky EPSCoR Program. The authors wish to thank Craig S. Pohlod and Mark Kaczor at the University of Illinois for sample irradiations.

REFERENCES

1. Geens S, Clayton M, Leidholdt J. Synovectomy and debridement of the knee in rheumatoid arthritis. *J Bone Joint Surg* 1969;51A:626–642.
2. Sledge CB, Zuckerman JD, Zalutsky MR, et al. Treatment of rheumatoid synovitis of the knee with intraarticular injection of dysprosium-165-ferric hydroxide macroaggregates. *Arthritis and Rheumatism* 1986;29:153–159.
3. Taylor AR, Harbison JS, Pepler C. Synovectomy of the knee in rheumatoid arthritis: results of surgery. *Ann Rheum Dis* 1972;31:159–161.
4. Camp AV. Synovectomy of the knee in rheumatoid arthritis: range of motion and quadriceps lag after surgery. *Ann Rheum Dis* 1972;31:155–158.
5. Laurin CA, Desmarchais J, Daziano L, Gariepy R, Derome A. Long-term results of synovectomy of the knee in rheumatoid patients. *J Bone Joint Surg* 1974;56A:521–531.
6. Fellinger K, Schmid J. Die Lokale Behandlund der Rheumatischen Erkraankungen. *Wien Z Inn Med* 1952;33:351.
7. Ansell BM, Crook A, Mallard JR, Bywaters EGL. Evaluation of intraarticular colloidal Au-198 in the treatment of persistent knee effusions. *Ann Rheum Dis* 1963;22:435.
8. Gumpel JM, Beer TC, Crawley JC, Farran HE. Yttrium-90 in persistent synovitis of the knee: a single center comparison of four radiocolloids. *Br J Radiol* 1975;48:377–381.
9. Delbarre F, Menkes CJ, Aignan M, Roucayrol JC, Ingrand J, Sanches A. Une nouvelle preparation radioactive pour la synoviorthese: le rheumium-186 colloidal. Advantages par rapport aucolloid d'or-98. *Nouv Presse Med* 1973;2:1372.
10. Oka M. Radiation synovectomy of the rheumatoid knee with yttrium-90. *Ann Clin Res* 1975;7:205–210.
11. Menkes CJ, Go AL, Verrier P, Aignan M, Delbarre F. Double-blind study of erbium-169 injection (synoviorthesis) in rheumatoid digital joints. *Ann Rheum Dis* 1977;36:254–256.
12. Onetti CM, Gutierrez E, Hleba E, Aguirre CR. Synoviorthesis with ^{32}P-colloidal chromic phosphate in rheumatoid arthritis. *J Rheumatol* 1982; 9:229–238.
13. Chappelle A, Oka M, Rekonen A, Ruotsi A. Chromosome damage after intra-articular injection of radioactive yttrium. *Ann Rheum Dis* 1972;31:508.
14. Topp JR, Cross EG, Fain AG. Treatment of persistent knee joint effusions with intra-articular radioactive gold. *Can Med Assoc J* 1975;112:1085–1089.
15. Noble J, Jones AG, Davies MA, Sledge CB, Kramer RI, Livni E. Leakage of radioactive particle systems from a synovial joint studied with a gamma camera: its application to radiation synovectomy. *J Bone Joint Surg* 1983;65A:381–389.
16. Deckart H, Tamaschke J, Ertl S, Tautz M, Knop G. Radiosynovectomy of the knee joint using ^{198}Au-colloid, ^{90}Y-ferric hydrate colloid and ^{186}Re-sulfide colloid. *Radiobiol Radiother* 1979;3:363.
17. Hnatowich DJ, Kramer RI, Sledge CB, Noble J, Shortkroff S. Dysprosium-165-ferric hydroxide macroaggregates for radiation synovectomy. *J Nucl Med* 1978;19:303–308.
18. Davis MA, Chinol M. Radiopharmaceuticals for radiation synovectomy: evaluation of two yttrium-90 particulate agents. *J Nucl Med* 1989;30:1047–1055.
19. Chinol M, Vallabhajosula S, Zuckerman JD, Goldsmith SJ. In-vivo stability of ferric hydroxide macroaggregates (FHMA). Is it a suitable carrier for radionuclides used in synovectomy? *Nucl Med Biol* 1990;17:479–486.
20. Hosain F, Haddon MJ, Hosain H, Drost JK, Spencer RP. Radiopharmaceuticals for diagnosis and treatment of arthritis. *Nucl Med Biol* 1990;17:151–155.
21. Venkatesan PP, Shortkroff S, Zalutsky MR, Sledge CB. Rhenium heptasulfide: a potential carrier system for radiation synovectomy. *Nucl Med Biol* 1990;17:357–362.
22. Zalutsky MR, Noska MA, Gallagher PW, Shortkroff S, Sledge CB. Use of liposomes as carriers for radiation synovectomy. *Nucl Med Biol* 1988;15:151–156.
23. Ehrhardt GJ, Lattimer JC, Day DE, and Stapleton J. Evaluation of activable, degradable glass Sm-153 microspheres for radiation synovectomy. *J Nucl Med* 1991;32:945.
24. Chinol M, Vallabhajosula S, Goldsmith SJ, Deutsch K, Chinen L, Deutsch E. Evaluation of new radiopharmaceuticals for radiation synovectomy. *J Nucl Med* 1991;32:963.
25. Ehrhardt GJ, Zinn K, Jia W, et al. Microspheres of human serum albumin labelled by a specific linker technology for use in radiation synovectomy. *J Nucl Med* 1991;32:964.
26. Mumper RJ, Ryo UY, Jay M. Neutron-activated Ho-166 poly(L-lactic acid) microspheres: a potential agent for the internal radiation therapy of hepatic tumors. *J Nucl Med* 1991;32:2139–2143.
27. Courtice FC, Gunton RW. The determination of blood volume by carbon monoxide and dye (T-1824) methods in rabbits. *J Physiol* 1949;108:405–417.
28. Neves M, Waerenborgh F, Patricio L. Palladium-109 and holmium-166 potential radionuclides for synoviotherapy—radiation absorbed dose calculations. *Appl Radiat Isot* 1987;38:745–749.
29. Lieberman E, Bordoni M, Thorton K, and Cadema Medical Products, Inc. Treatment of arthritis, including rheumatoid arthritis with ^{166}Ho radionuclide. United States Patent Number 4,849,209. July 18, 1989.
30. Spencer RP. Applied principles of radiopharmaceutical use in therapy. *Nucl Med Biol* 1986;13:461–463.
31. Spencer RP. Short-lived radionuclides in therapy. *Nucl Med Biol* 1987;14:537–538.
32. Davis M. In: *Radiopharmaceutical Science Council Newsletter.* May 1989. p. 6.
33. Ratcliffe JH, Hunneyball IM, Smith A, Wilson CG, Davis SS. Preparation and evaluation of biodegradable polymeric systems for intra-articular delivery of drugs. *J Pharm Pharmacol* 1984;36:431–436.
34. Ratcliffe JH, Hunneyball IM, Wilson CG, Smith A, and Davis SS. Albumin microspheres for intra-articular drug delivery: investigation of their retention in normal and arthritic knee joints of rabbits. *J Pharm Pharmacol* 1987;39:290–295.
35. Jay M, Mumper RJ, Khare SS, Ryo UY. Biodegradable radiotherapeutic microspheres containing activable isotopes of holmium, rhenium and dysprosium. *J Nucl Med* 1988;29:903–904.