Infections with opportunistic pathogens have been one of the hallmarks of the acquired immunodeficiency syndrome since the beginning of the epidemic. It is now recognized that bacterial pneumonia occur at a higher frequency among HIV-infected patients compared to age-matched controls [10, 14]. Diseases caused by bacteria are responsible for a significant proportion of the morbidity and mortality seen in this population. The course of certain bacterial infections does not differ from that in the immunocompetent host, whereas other bacterial infections are notable for an increased incidence, a more fulminant course, invasive disease, and unusual rates of relapse. It is important to remember that HIV-infected patients may not present with an acute onset of symptoms, fever or elevated white blood cell count, which are characteristic of bacterial infections in the normal host [4].

In-house preparation and evaluation of Tc-99m-HIG kit for detection of infection in AIDS patients

Mohammed A. Bakir, Raid Mansour, Ali Mohammad, Khozama Habil, Taufik Yassine

Abstract. $^{99m}$Tc-HIG can be used for early detection of inflammation and infection foci especially in immunodeficiency patients. The present study involves in-house preparation and evaluation of human immunoglobulin-hydrazinonicotinic acid (HIG-HYNIC) kit to be labeled with $^{99m}$Tc. In this indirect labeling method, the structure and biological activity of the protein can be maintained. The kit consists of two vials A and B, the vial A contains 2 mg of HIG-HYNIC, and the vial B contains 10 mg of tricine and 1 mg of stannous chloride dihydrate (SnCl$_2$.2H$_2$O). The results revealed that the lyophilized kits were stable for at least 10 months, and showed a high quality from the points of physical, chemical, radiochemical and biological purities, and its validity for human injection. The labeling yield of HIG-HYNIC compound exceeded 98%. The application of the prepared kit was experimentally illustrated in animal models with artificially induced inflammatory lesions after E. coli injection into rat leg muscle. The biodistribution and imaging studies in rats have shown good localization in infected muscle (T) in comparison to the normal muscle (M) 4 and 24 h after injection. Ratios of 17 and 18 were achieved 4 and 24 h post injection, respectively. The results of biodistribution of $^{99m}$Tc-HIG in human and initial clinical evaluation show that this agent is a good radiopharmaceutical for infection imaging.

Key words: $^{99m}$Tc-HIG • HYNIC • infection imaging

Introduction

Infections with opportunistic pathogens have been one of the hallmarks of the acquired immunodeficiency syndrome since the beginning of the epidemic. It is now recognized that bacterial pneumonia occur at a higher frequency among HIV-infected patients compared to age-matched controls [10, 14]. Diseases caused by bacteria are responsible for a significant proportion of the morbidity and mortality seen in this population. The course of certain bacterial infections does not differ from that in the immunocompetent host, whereas other bacterial infections are notable for an increased incidence, a more fulminant course, invasive disease, and unusual rates of relapse. It is important to remember that HIV-infected patients may not present with an acute onset of symptoms, fever or elevated white blood cell count, which are characteristic of bacterial infections in the normal host [4].

The early detection of infection foci is very important especially for AIDS patients since it leads to more effective management to those populations if the clinical history and physical examination are indecisive, the clinical physician can choose from several diagnostic modalities to determine the localization [4], extent and severity of the disease, one can also choose some radiochemical methods (CT – computed tomography; MRI – magnetic resonance imaging; X-rays) that rely on morphological changes, however they are less ac-
curate in early stages of infection. In nuclear medicine a radionuclide [4] (radiopharmaceutical) is injected and the distribution of the radioactivity throughout the body is visualized using a dedicated gamma camera. The radiopharmaceutical used for imaging infection and inflammation lesion due to the locally changes physiological conditions. An infectious inflammatory focus is characterized by enhanced blood flow-enhanced vascular permeability and influx of white blood cells. Therefore, scintigraphic techniques can also visualize infectious foci in their early phases, when morphological changing are not yet apparent. In addition, scintigraphic imaging is an excellent non-invasive method of whole body scanning that can determine the extent of the infectious or inflammatory disease throughout the body.

Currently, there are several radiopharmaceuticals in use for infection and inflammation detection, e.g. gallium-67 citrate [8, 17], radiolabeled polymorphic-nuclearleucocyte (PMN) with indium-111 or Tc-99m [3, 9, 18], radiolabeled anti-granulocyte antibodies [22], radiolabeled liposomes with Ga-67, In-111 or Tc-99m [7, 8], interleukins [20], and chemotactic peptides [1, 2], flurine-18 fluorodeoxyglucose (FDG). Human polyclonal immunoglobulin (HIG) may be labeled with In-111 or Tc-99m. They accumulate [8] at sites of inflammation mainly via capillary leakage, with smaller contribution from binding of the Fc portion of IgG to Fc receptors on granulocytes and depending on the source of the pooled human immune globulin, binding of IgG to bacteria. Because HIG localize at sites of bacterial infection, sterile inflammation, and inflammatory tumors, it is a good “catch all” agent in FUO (fever of unknown origin), especially in neutropaenic and immunocompromised patients. In a comparative study [4], it was shown that Tc-99m-IgG labeled via chelator hydrazinonicotinamid, has in vivo characteristics highly similar to those of In-111-IgG, and in most cases can replace the In-111-labeled compound. Buttram et al. [5] had labeled IgG using Tc-99m with better imaging according to a method depending on the reduction of whole antibodies with the reducing agent diithiothreitol (DTT) to produce sulfhydryl containing antibodies, under non-oxidizing conditions, reducing antibodies containing free sulfhydryl groups. Paik et al. [16] provide a method for labeling antibodies with radionuclides like technetium to obtain stable labeled formulation. It was noticed that diethylenetriamine-pentaacetic acid (DTPA) acts as a competitive ligand, which binds reduced technetium more tightly than the non-stable binding site of the antibody, but less tightly than the stable binding site (whole IgG contains both stable and non-stable labeling sites for reduced technetium ions). The “non-stable site” (which may be one or more sites) has a high capacity but a low affinity for reduced technetium. Labeling of antibodies in the presence of DTPA, in a sufficient molar excess, can substantially completely inhibit labeling of the non-stable site, so that substantially only a stable labeled antibody is produced. Labeling of DTPA conjugated antibodies permits a higher percentage of incorporation of stable technetium radioisotope. Labeling in the further presence of DTPA also prevents formation of unstable label, and permits production with a high radiochemical yield of stably labeled DTPA-conjugated antibody. This method avoids the formation of colloid during the course of the labeling process, and labeling in the presence of free DTPA produces technetium-labeled antibody and DTPA-complexed technetium. Abrams et al. [15] prepared Tc-99m labeled IgG by using a sufficient molar access of hydrazinonicotinamide derivative (succinimidy-6-hydrazino nicotinate hydrochloride) which binds to the amino end of polyclonal antibody, the nicotinyl hydrazine modified IgG labeled by using a freshly prepared Tc-99m-glucopentanate kit. The radiochemical purity for all preparations was at least 90% of the radioactivity bound to IgG.

The aim of this study was to locally prepare a kit for Tc-99m human immunoglobulin labeled via HYNIC as a bifunctional molecule and Sn-tricine co-ligand, optimizing the labeling conditions, studying the biodistribution of the labeled compound in infected animal model, and conducting an initial human study.

**Experimental**

**Materials**

Human immunoglobulin-G (HIG) was obtained from GAMMARASS (China), succinimidyl hydrazinonicotinic acid (S-HYNIC) from Solulink (USA), tricine from Sigma (Germany), dried DMSO (Merck, Germany), sodium carbonate, tween 80 (polysorbate 80), culture media FTM, CSD (Merck, Germany), ammonia solution 25% (BDH, Germany), slide A-lyzer cassette, Sephadex G-25 M column (Sigma, Germany), LAL kits (Pyrotell, Germany).

**Preparation of Tc-99m-HIG**

**Preparation of HIG-HYNIC compound (vial A)**

- Purification of HIG. 10 ml of 5% HIG were injected in a slide A-lyzer cassette, dialyzed in a sterile dialysis container against 0.9% NaCl solution at +4°C under constant stirring (the outer solution was refreshed four times).
- Conjugation of HYNIC with HIG. 3 ml of the purified HIG were drawn up from the slide A-lyzer cassette and diluted to 5 ml with 0.9% NaCl (in 8 ml vial), followed by adding 0.5 ml of 1 M NaHCO₃ solution, and 10 portions of 10 µL HYNIC solution (1 mg HYNIC dissolved in 100 µL dried DMSO (dimethyl sulphoxide)), the reaction mixture being incubated for 30 min at room temperature in a dark place. The final preparation was purified in two methods: the slide A-lyzer cassette and a Sephadex G-25 M column. In the slide A-lyzer cassette method, the volume of the prepared solution was adjusted to 8.6 ml using 0.15 M acetic acid buffer, pH 6.4, and injected into another slide A-lyzer and kept overnight against 0.15 M acetic acid buffer, pH 6.4, at 4°C under constant stirring (the buffer was refreshed 4 times). The content was then transferred to a 50 ml beaker. In the Sephadex G-25 M column method, the prepared solution was loaded onto the surface of a previously activated Sephadex G-25 M column, eluted using 0.15 M acetic acid buffer,
pH 6.4). Elutes 2, 3, 4 and 5 were collected (1 ml of each fraction), and transferred to 50 ml beaker.

After purification, the final HYNIC-HIG solution volume was adjusted to 38 ml with 0.15 M acetate buffer, pH 6.4. The solution was dispensed into 0.5 ml aliquots through a 0.22 µm low protein binding filter, freeze-dried, sealed under nitrogen and stored at +4°C. In another preparation, in order to study its effect, the tween 80 was added in a concentration of 0.18 mg/100 ml of the final HIG-HYNIC solution.

**Preparation of Sn-tricine (vial B)**

Sn-tricine kits were prepared as follows: 0.4 g tricine was dissolved in 25 ml of double distilled water, pH 2.75, the oxygen being removed by applying a gentle stream of nitrogen. Then, 0.04 g of SnSO₄ was dissolved in 1 ml 2 M HCl and added to the tricine solution, the pH was then adjusted to 4.5 by adding 1.5 ml of NaOH solution and to 5.30 by adding 1.7 ml of NaOH solution. The final volume of the solution was adjusted to 40 ml with 0.9% NaCl and filtered directly into cold vials sealed under nitrogen using the 0.22 µm filter (1 ml fraction in each vial). The vials were freeze-dried and closed under nitrogen.

**Labeling of HIG-HYNIC conjugate with Tc-99m**

For radiolabeling of HIG-HYNIC conjugate with Tc-99m, the content of HIG-HYNIC vial was dissolved in 0.5 ml of saline, and the content of the Sn-tricine vial was dissolved in 5 ml of sterile 0.9% saline solution. 50 µl of the tricine-Sn solution was added to the HIG-HYNIC vial, followed by adding 30–35 mCi of Tc-99m and incubated for 30 min at room temperature.

**Quality control**

The following quality control tests were performed:

**pH**

pH values were measured using pH paper strips or a pH meter after dissolving the vial content in 3–5 ml of normal saline or water for injection.

**Endotoxin test**

The endotoxin test was performed on all batches and for both prepared vials HIG-HYNIC and Sn-tricine, and all vials and covers used in the preparation. For the prepared vials, the contents of the vials (HIG-HYNIC and Sn-tricine kits) were dissolved in 1 ml of water for injection and aliquots of 0.2 ml were then added to 3 LAL vials (0.25 unit sensitivity), incubated at 37°C for 60 min.

**Sterility test**

The sterility test was performed on all prepared batches and for both vials: HIG-HYNIC and Sn-tricine, according with the pharmacopoeial method using a soya-bean casein digest (SCD) and fluid thioglycollate media (FTM) and incubation of the content of the tested vial for 14 days at a temperature ranging between 30 and 35°C.

**Radiochemical purity of Tc-99m-HIG-HYNIC**

The radiochemical purity (RCP) of Tc-99m-HIG-HYNIC was determined by instant thin-layer chromatography (ITLC) as follows: 5 µl of the prepared compound was spotted on 10 cm ITLC-SG strips. The strips were then run with 0.15 M citrate buffer at pH 5.5 as mobile phase. The Rf values of the Tc-99m-HIG and the colloid ranged between 0 and 0.25, and that of the Tc-99m-tricine and Tc-99m ranged between 0.7 and 1.0. For colloid determination, 5 µl of the prepared compound was spotted on 10 cm ITLC-SG strips saturated with 1% human serum albumin (HSA). The strips were then run with a freshly prepared mobile phase of water:ethanol:ammonia 25%, the ratio being 1:2:5. Tc-99m, Tc-99m-tricine, and Tc-99m-HIG were moved to the top of the strip with Rf values of 0.7–1.0, whereas, the colloid remained at Rf of about 0–0.25.

**Stability study**

Both tween-contained and tween-free radiolabeled HIG-HYNIC was tested for stability at room temperature at 0, 1, 2, 4, 8, and 24 h. Also, the stability of the labeled compound was determined in a serum and PBS (phosphate-buffered saline) at 0, 1, 3 and 6 h at 37°C. Briefly, 5 ml of blood were withdrawn from a normal volunteer in a dry tube and centrifuged for 15 min at 3500 rpm and then the serum was separated. 100 µl of Tc-99m-HIG was added to 1 ml of the serum or PBS and incubated in a water bath at 37°C up to 6 h. The chromatographic system employed in the determination of the radiochemical purity of the preparations at various time points is described in the previous paragraph.

**Shelf-life study**

The shelf-life of the prepared kits was tested by determining radiochemical purity of the radiolabeled HIG-HYNIC for one year at 1.5 month intervals for the tween free kits, and for 9 months in one month intervals for tween 80 contained kits. The radiochemical purity was checked at each time point.

**Animal studies**

Thigh muscle abscesses were induced in Albino rats, about 150 g in weight, by injection of 0.1 ml of a 50:50 suspension of blood with 1 × 10⁶ colony/ml E. coli. The animals were monitored for local swelling between 24 h and 8 days after abscess induction. In order to confirm the abscess formation, stained samples of the abscess were studied after routine formalin fixed paraffin-embedded tissue sections. A hyperemic lesion with infiltration of granulocytes without pus formation is shown in Fig. 1.

Biodistribution studies were carried out in healthy and infected Albino rats, weighing about 150 g. The biodistribution in healthy rats was done to see...
the normal pharmacokinetics of the radiolabeled HIG, while the study on the infected rats was done to see the effectiveness of making an infection and the uptake in the induced lesion. Briefly, 0.1 ml of the radiopharmaceutical was administered at a dose of 37 MBq/0.1 ml in a tail vein of Albino rats. After intervals of 1, 4, and 24 h, aliquots of blood were collected from the hearts. The rats were then sacrificed, the organs were removed (heart, lungs, liver, spleen, kidneys, stomach, intestine, bone, infected thigh muscle, contralateral normal muscle), washed in normal saline, dried on a filter paper, weighed and the radioactivity in the organs and blood was measured in a well type gamma counter. The percentage of the administered dose in the different organs and in the blood was calculated as a function of administered dose and expressed as % dose/gram organ. Also, the infected-to-normal muscle ratios taken 1, 4, 24 h post injection were calculated.

To localize the induced lesion, rats were injected with 37 MBq Tc-99m-HIG-HYNIC, anesthetized with ether, and imaged 4, 24 h, post injection using a Simens gamma camera equipped with a low-energy high-resolution (LEHR) collimator. 1,000,000 counts were accumulated. In conclusion of imaging, the anesthetized rats were sacrificed; infection was confirmed by histopathological examination (Fig. 1).

**Initial clinical study with Tc-99m-HIG**

In order to initially evaluate the locally prepared Tc-99m-HIG compound in humans, two groups of patients were imaged using Tc-99m-HIG: a group of none-AIDS, non-infected patients, and a group of patients with AIDS test positive (latent phase), none of them showed clear specific clinical signs or symptoms of the disease. Briefly, patients were intravenously injected with 740 MBq of Tc-99m-HIG containing between 0.5 and 1 mg of HIG, and imaging were acquired 4–6, and 24 h post injection with a dual head gamma camera. Imaging parameters typical for $^{99m}$Tc were applied. Anterior and posterior whole body planar images were obtained. The following contraindications related to HIG were taken into consideration while selecting the patients for the study. These include, previous systemic reaction to IV HIG, gammaglobulinemia, and sever hypogammaglobulinemia.

**Results and discussion**

In the formulation of a new Tc-99m radiopharmaceutical kit, several parameters are required for the acceptable clinical applications. The new kit should have a long self-life, a high radiochemical purity (preferably > 90%), a high stability after preparation of the radiopharmaceutical (6 h or more), also stability of the radiopharmaceutical in vivo is one of the important aspects of the labeled compound after injection into the patient. For diagnostic applications, Tc-99m remains the radionuclide of choice in scintigraphic imaging as it has a number of favorable characteristics including low cost, low radiation dose to the patient, optimum gamma-energy profile, and a wide commercial availability via generators. Radiolabeling of protein or peptide radiopharmaceuticals using bifunctional chelating agents proved to be the most appropriate labeling method [13], where, a high specific activity of Tc-99m labeling can be achieved. 6-hydrazinonicotinic acid (HYNIC) as a bifunctional molecule is typically attached to the amino group of the protein. HYNIC-protein conjugates can be ready labeled with Tc-99m using a number of co-ligand systems. In this work, the tricine was used as co-ligand for radiolabeling of HIG-HYNIC conjugates. Larsen [12] has shown that Tc-99m-tricine is a useful precursor complex for radiolabeling of HYNIC-protein conjugates. This complex has improved efficacy for the Tc-99m labeling of HYNIC-protein compared to Tc-99m-glucoheptonate.

In this work, HIG kit for preparation of the radiopharmaceutical Tc-99m-HIG has been successfully prepared for the detection of infection and inflammation. HIG was conjugated with HYNIC as described above in a class-100 laminar bench. Purification of the conjugated HIG was done either by gel filtration or dialysis. Filtration of the formulation on a Sephadeoxy A-25 M column led to radiochemical purity of about 99%. The dialysis of the conjugated HIG formulation on the slide A-lyzer cassette led to a similar radiochemical purity. pH values for all freeze-dried HIG-HYNIC batches were between 6.4 and 7.4. Radiochemical purity of the labeled HIG-HYNIC when using Sn-tricine kit with pH ranging between 4.5 and 5.3 was determined. The results ranged between 96 and 99%.

Investigation of the stability of the labeled HIG-HYNIC showed that the radiochemical activity decreased marginally with time, but remained above 95% after 2 h and above 90% after 8 h and 85% after 24 h for both tween-free and tween-contained batches. The results are shown in Table 1. Also the labeled formulation has shown a high stability in both PBS and serum where more than 90% of the activity was chromatographed with the HIG fraction after 6 h. (Table 2). Shelf-life investigation of the prepared HIG-HYNIC kits was carried out for one year in 1.5 month intervals for batches.
prepared without tween 80, and for 9 months in one month intervals for batches prepared with tween 80. However, similar results were obtained for both cases. The results are showing a high quality of the prepared kit since the labeling efficiency remained about 95% in both cases, and for a period of more than 9 months. Table 3 is showing the results of the tween-free batches. In a recent study conducted by Rajabifar et al. [19] the authors prepared and evaluated 99mTc-HIG-HYNIC showing a similar radiochemical purity and stability. However, this study did not investigate the shelf-life of the prepared HIG-HYNIC kits. Rennen et al. [21] reported the radiochemical purity of 99mTc-HIG-HYNIC exceeding 92% and the stability in rat serum up to 4 h as determined in ITLC. Karczmarczyk et al. [11] studied the stability of 99mTc-HIG-HYNIC prepared using ethylenediaminetetraacetic acid (EDTA) as a co-ligand; they noticed that the stability of the labeled compound was decreased rapidly in the serum and this forced them to find proper conditions for improving the stability of the labeled HIG in circulating serum for 6 h using a mixture of four compounds.

The histological sections of the formalin fixed, paraffin-embedded abscess section from the infected rats are shown in Fig. 1. This figure is showing a hypereemic lesion with infiltration of granulocytes without major puss formation. The organ biodistribution of 99mTc-HIG-HYNIC 1, 4, and 24 h post injection for infected rats is shown in Fig. 2. The biodistribution pattern in healthy and infected rats was almost similar (only data from infected rats are shown). The highest %ID/g at 4 h uptake was observed in the blood, liver, kidneys, lungs, and spleen, respectively and the values were: 4.73, 2.83, and 2.62; 1.72 and 1.91. In a study conducted by Dams et al. [6] in granulocytopenic rats showed percent injected dose per gram (%ID/g) in the spleen, blood, lungs, liver, and kidneys as follows: 7.3, 5.9, 5.6, 4.1, and 3.2, respectively. In our study much lower uptake in the

Table 1. The stability of both tween-contained and tween-free Tc-99m-HYNIC-HIG at room temperature

<table>
<thead>
<tr>
<th>Time (h)</th>
<th>Reduced and collloid 99mTc (%)</th>
<th>Reducted and collloid 99mTc (%)</th>
<th>Reduced and collloid 99mTc (%)</th>
<th>Reduced and collloid 99mTc (%)</th>
</tr>
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<tbody>
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<td>99mTcO4 + 99mTc-tricine RACP*</td>
<td>99mTcO4 + 99mTc-tricine RACP*</td>
<td>99mTcO4 + 99mTc-tricine RACP*</td>
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</tr>
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<td>1.02</td>
<td>2.36</td>
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<td>1.34</td>
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<tr>
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<td>2.24</td>
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<td>24</td>
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<td>10.80</td>
<td>86.45</td>
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</tbody>
</table>

* RCP – radiochemical purity.

Table 2. The stability of the tween-free HYNIC-HIG at 37°C in PBS and plasma

<table>
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<tr>
<th>Time (h)</th>
<th>PBS (%)*</th>
<th>Plasma (%)*</th>
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<td>93.57 ± 3.37</td>
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</table>

* Radiochemical purity.

Table 3. The shelf-life of the tween-free HYNIC-HIG

<table>
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<tr>
<th>Time (months)</th>
<th>Reduced and collloid 99mTc (%)</th>
<th>Reduced and collloid 99mTc (%)</th>
<th>Reduced and collloid 99mTc (%)</th>
<th>Reduced and collloid 99mTc (%)</th>
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<td>99mTcO4 + 99mTc-tricine RACP*</td>
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</table>

* RCP – radiochemical purity.

Fig. 2. Biodistribution of tween-free Tc-99m-HYNIC-HIG in an E. coli infected rats 1, 4, and 24 h post injection (%ID/g, mean of 5 values per time point).
spleen, liver, and lungs was observed. Karczmarczyk et al. [11] showed the highest %ID/g at 4 h in the blood, liver, kidneys, and lungs as follows: 9, 3, 2.5, and 1.75. By comparing Karczmarczyk results to our results, much lower blood uptake and similar liver, kidneys, and lungs uptake can be observed. In a recent study performed by Rajabifar et al. [19] it was shown that in the biodistribution in the normal rats, the highest %ID/organ were observed in the blood, liver, kidney, spleen, stomach, lungs, thyroid and muscle, respectively at 4 and 24 h intervals. However, in the Rajabifar et al. [19] study no infected rats were included. In the current work a study on the infected rats was done to see the effectiveness of making an infection and the uptake in the induced lesion. The results showed %ID/g 1, 4 and 24 h post injection in the thigh muscle abscess of 0.40, 1.19 and 2.48, respectively. Also, these results indicated that the target (infected muscle)-to-(normal muscle) (T/M) 1, 4 and 24 h post injection were 4.04, 17, 18. The results are shown in Fig. 3. This figure is showing the gradual accumulation of the labeled compound in the abscess with an optimal ratio of 18 at 24 h post injection. However, there was no significant difference in this ratio 4 and 24 h post injection, raising the possibility of imaging at an earlier time than 4 h. Karczmarczyk et al. [11] showed in an artificially induced inflammatory lesions of mouse thigh muscle (by injection of turpentine oil) of only four times higher accumulation of the tracer 6 h after injection, and six times higher after 24 h. A comparison study has been conducted between tween 80 contained and tween 80 free HIG-HYNIC kits in both the normal and infected rats. The results showed a similar biodistribution pattern in both cases (data not shown). The images of an abscess in the left thigh muscle in a rat 4 and 24 h post injection using a gamma camera are shown in Fig 4. These images are showing that the prepared radiolabeled HIG readily accumulated in the induced abscess to an extent sufficient to yield excellent external images.

The group of none-AIDS, non-infected patients has showed the normal distribution of Tc-99m-HIG in both early and late images in humans. The distribution was mainly in the liver, spleen, kidneys, bladder, heart, blood vessel. Figure 5 is showing the normal distribution of Tc-99m-HYNIC-HIG (4 h post injection) in humans.

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![Fig. 3. Target-to-muscle ratios (T/M) 1, 4 and 24 h post injection.](image1)

![Fig. 4. Gamma camera imaging of Tc-99m-HYNIC-HIG in rat with induced infection through an E. coli injection in the left thigh muscle 4 h (A) (A1: anterior, A2: posterior) and 24 h (B) (B1: anterior, B2: posterior) post injection.](image2)

![Fig. 5. The normal distribution of Tc-99m-HYNIC-HIG (4 h post injection) in humans.](image3)
In-house preparation and evaluation of Tc-99m-HIG kit for detection of infection in AIDS patients

Distribution Tc-99m-HIG 4 h post injection. Ten out of twelve studied AIDS positive test patients, showed negative results (normal Tc-99m-HIG distribution). All these patients were apparently clinically normal at this time of the study (latent phase). One female patient showed a focal small area of increased uptake in the pelvis at both early and late images, (Fig. 6). This lesion was confirmed later as a little abscess. Another patient showed a slight homogeneous increased uptake of the Tc-99m-HIG in the chest.

Conclusion

This work involved the development of a Tc-99m radiopharmaceutical kit using HIG with a HYNIC chelating group, to be applied in the diagnostic of infection and inflammation lesions in general nuclear medicine with special interest in the early detection of infection foci in AIDS patients. HYNIC-HIG kit to be labeled with Tc-99m-labeled to human immunoglobulin G through the nicotinyl hydrazine derivative: a clinical study. J Nucl Med 39:119–124

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Fig. 6. Focal uptake in the pelvis of Tc-99m-HYNIC-HIG (4 h post injection) in a patient.