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# Radiochemical and biological characteristics of $^{99m}$Tc-piroxicam for scintigraphy of inflammatory lesions

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Piroxicam was labeled effectively with $^{99m}$Tc due to the presence of electron donating atoms such as sulfur, nitrogen, and oxygen in its structure. The labeling yield was found to be influenced by different factors such as the amount of piroxicam, stannous chloride dihydrate, pH of the reaction mixture, reaction time and reaction temperature. The suitable amount of stannous chloride dihydrate required to produce high labeling yield of $^{99m}$Tc-piroxicam was 50 μg, above this quantity (200 μg) a colloidal solution was formed. Another factor which plays a significant role in this labeling reaction is the pH of the reaction medium. The labeling reaction was done only at alkaline pH range from 9–11, because piroxicam was not soluble at acidic or neutral pH. The labeling reaction proceeded well at room temperature and the complex was decomposed by heat. The labeled piroxicam ($^{99m}$Tc-piroxicam ) showed good localization in inflamed foci and good imaging must be taken at 24-hour post injection, as the ratio of both types of inflammation (sterile and septic) to the background are 10.6 and 8.7, respectively.

## Introduction

One of the areas of clinical interest of radionuclides is the diagnosis of inflammation. Inflammatory diseases can be successfully detected by nuclear medicine imaging with some radiopharmaceuticals. Gallium [$^{67}$Ga] citrate is commonly used for this diagnosis, with whole body imaging at 24, 48 and sometimes 72 hours after injection of radiotracer.[1,2] $^{67}$Ga which entails high radiation exposure, is not routinely available and has unfavorable physical characteristics for gamma camera imaging. These features limit the use of $^{67}$Ga to certain indications such as fever of unknown origin, chronic osteomyelitis of the spine and lung infections, especially in immunocompromised patients. $^{111}$In-labeled leukocytes are also used for the detection of inflammatory diseases such as abscesses.[3] The physical characteristics of $^{111}$In, with its medium energy, long physical half-life and radiation burden to the patient, combined with its production in a cyclotron, causing limited availability, mean that the resolution achieved with gamma camera imaging is suboptimal and that $^{111}$In is not favoured in clinical practice.$^{99m}$Tc-citrate has been used for the scintigraphic visualization of inflammatory lesions and pancreatitis.[4] Promising new agents for scintigraphic detection of inflammation include labeled liposomes, which have been widely studied for achieving controlled drug delivery and for imaging purposes. Conventional liposomes are rapidly cleared from the circulation by phagocytic cells of the mononuclear phagocyte system (MPS),[5] therefore, their use for diagnostic imaging is limited. Since all routinely used agents have drawbacks that limit their application, there is a need for new and better radiopharmaceuticals.

Piroxicam is one of the oxim derivatives, a class of enolic acids that have anti-inflammatory, analgesic, and antipyretic activity. Piroxicam appears to be the equivalent of aspirin, or naproxen for the long-term treatment of rheumatoid arthritis or osteoarthritis. It may be tolerated better than aspirin. The structural formula of piroxicam is illustrated in Fig. 1.

There is an enterohepatic cycling of piroxicam, and estimates of the half-life in plasma have been variable, a mean value appears to be about 50 hours. After absorption, piroxicam is extensively (99%) bound to plasma proteins. At steady state (e.g., after 7 to 12 days), concentrations of piroxicam in plasma and synovial fluid are approximately equal. Less than 5% of piroxicam is excreted in the urine unchanged. Piroxicam is an effective anti-inflammatory agent, it is an inhibitor of prostaglandin biosynthesis in vitro. Piroxicam also inhibits activation of neutrophils even when products of cyclooxygenase are present at inflammatory sites, hence, additional modes of anti-inflammatory action have been proposed, including inhibition of proteoglycanase and collagenase in cartilages.[6,7] Due to the ability of piroxicam to localize itself in inflammatory sites, $^{99m}$Tc-piroxicam complex will be investigated as a useful radiopharma-ceutical to image experimentally induced inflammatory lesions in animals.

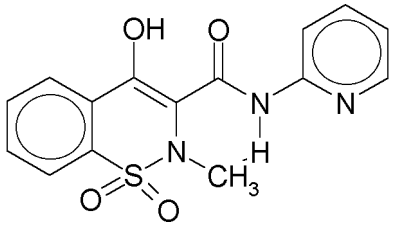


*Fig. 1.* Chemical structure of piroxicam

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## Experimental

All chemicals and laboratory reagents used in this work were of the highest purity grade. In all cases, the water used was deoxygenated bidistilled water. In addition piroxicam (M.wt. = 331.35) was obtained from Military Armed Forces Pharmaceutical Industry, Cairo, Egypt.

*Animals*

Albino type mice weighing 20–25 g, the mice were used for biological distribution study.

*Radioactive material*

Pertechnetate-99m solution was obtained by eluting from the sterile $^{99}$Mo/$^{99m}$Tc generator (Elutic, Brussels, Belgium) with sterile saline solution 30–40 mCi/ml (1110–1480 MBq/ml).

*Methods*

*Preparation of stock solution of stannous chloride dihydrate ($SnCl_2 \cdot 2H_2O$):* Exactly 400 mg of stannous chloride dihydrate was dissolved in 0.5 ml of conc. HCl by heating on a hot plate, then the volume was completed to 10 ml using nitrogen purged double distilled water (40 mg $SnCl_2 \cdot 2H_2O$/ml). One ml was again diluted to 10 ml with nitrogen purged double distilled water. Two ml of this solution was dispensed to 10 ml clean penicillin vials, flushed with nitrogen for 5 minutes, then stored at –20 °C for future use. The final concentration was 4 mg/ml ($336.8 \cdot 10^{-4}$ mmol).

*Technetium-99m labeling of piroxicam:* In a clean sterile 10 ml penicillin vial, the required amount of piroxicam was dissolved in bicarbonate buffer pH 11, stannous chloride dihydrate and sodium pertechnetate were added and the vial closed under a positive nitrogen pressure. The reaction vial was incubated for 15 minutes at room temperature. Unless otherwise stated the following quantities were always used: 0.5 mg piroxicam, 50 µl $SnCl_2 \cdot 2H_2O$ and 0.5 ml (~750 MBq ) $^{99m}TcO_4^-$.

*Determination of the radiochemical purity:* The radiochemical purity and the in-vitro stability were determined using electrophoresis. Electrophoresis was done using cellulose acetate strips and 0.2M phosphate buffer pH 7. Five micro-liters of the reaction mixture, after 0.22 µm millipore filtration, were spotted on the starting point and 300 V electric current was applied for one and half hours. Free pertechnetate was moved with distance equal to 7 cm while $^{99m}$Tc-piroxicam complex remained at the origin. No evidence was found for the presence of colloid as 0.22 µm millipore filtration removed it as stated by CELERIER et al.[8] The electrophoretic diagram of $^{99m}$Tc-piroxicam and other radiochemical species are presented in Fig. 2.

*Biodistribution studies*

Albino mice were used for quantitative biodistribution studies. Sterile inflammation was induced by the injection of sterile turpentine oil (200 µl), intramuscularly, into the right thigh muscle while abscesses were induced by the injection of a suspension of *Escherichia coli* (*E. coli*). When swelling of the muscle was apparent, $^{99m}$Tc-piroxicam was injected intravenously (i.v.). Groups of three mice were used for each experiment. The mice were sacrificed by decapitation under chloroform anesthesia at 0.5, 2, 4 and 24 hours after injection, respectively. Blood samples were collected at the time of decapitation. Both thighs (right thigh muscle as target, left thigh muscle as control) and organs were dissected, weighed and their radioactivity was measured using a well-type NaI(Tl) detector connected with a single channel γ-counter (SR 7). Results were expressed as percent of the injected dose per organ or body fluid.

## Results and discussion

Piroxicam is one of the oxim derivatives. The chemical structure is presented in Fig. 1. Due to the presence of atoms which are electron donors such as sulphur, nitrogen and oxygen in its structure it has the ability to form metal-donor complexes with transition metals like $^{99m}$Tc. The produced complex may be formed by sharing the negative charge or electron pairs of these atoms and groups with the reduced technetium when it is in the reduced states +1 or +3 according to:

$$^{99m}\text{Tc} + \text{Sn}^{+2} \rightarrow \text{Reduced } ^{99m}\text{Tc (+1 or +3)} \quad (1)$$

$$\text{Reduced } ^{99m}\text{Tc} + \text{Piroxicam} \rightarrow ^{99m}\text{Tc-piroxicam} \quad (2)$$

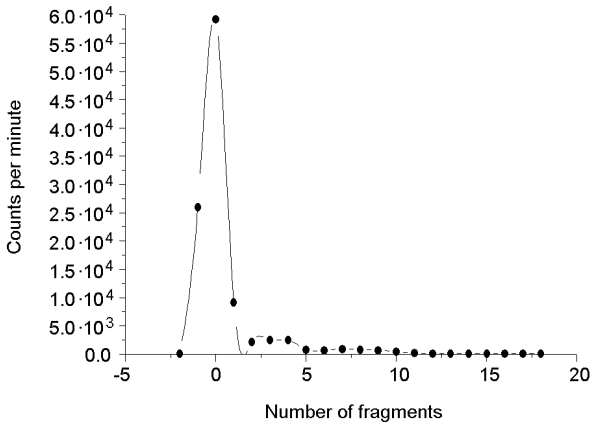


*Fig. 2.* Electrophoretic pattern of $^{99m}$Tc-piroxicam complex; conditions: solvent: 0.2M phosphate buffer pH 7, paper type: cellulose acetate; 300 V; running time: 1.5 hours

Also, due to the ability of piroxicam to localize in inflammatory sites, it was examined for labeling with $^{99m}$Tc. The produced complex, $^{99m}$Tc-piroxicam, will be examined as an inflammation localizing agent. Factors affecting the labeling process were studied and the results can be summarized as follows.

*Effect of the amount of piroxicam*

The influence of the amount of piroxicam on the percent labeling yield of $^{99m}$Tc-piroxicam complex was studied and the results are presented in Table 1. According to these data, it was clear that 0.5 mg of piroxicam was sufficient to produce $^{99m}$Tc-piroxicam complex with a radiochemical yield of >97%. Increasing the amount of piroxicam up to 2 mg does not affect the percent yield of $^{99m}$Tc-piroxicam. Below 0.5 mg of piroxicam the yield decreased and the free pertechnetate increased indicating the insufficiency of the amount of piroxicam to complex all the reduced $^{99m}$Tc ion. For this reason 0.5 mg of piroxicam was used in all this work as the optimum quantity required to attain high radiochemical yield.

*Effect of the amount of stannous chloride dihydrate*

In general, most of the $^{99m}$Tc-complexes are prepared by the reduction of pertechnetate with an inorganic reducing agent. Tin(II) is the most widely used reducing agent. The amount of tin(II) required to reduce pertechnetate from its hepta valent state to the reactive reduced states, must be adjusted to prevent the formation of undesirable radiochemical species, such as reduced hydrolyzed technetium,$^{99m}$Tc-tin colloids and free pertechnetate,$^{99m}TcO^{-}_{4}$, especially in alkaline pH medium. To study the effect of stannous chloride dihydrate content on the formation of $^{99m}$Tc-piroxicam complex, the following amounts of stannous chloride dihydrate were used, 10, 25, 50, 100 and 200 μg. The results obtained from the analysis of the reaction mixture after 15-minute reaction time at room temperature (25 °C) are shown in Table 2. As is clear from this data, 10 and 25 μg of $SnCl_2{\cdot}2H_2O$ produce $^{99m}$Tc-piroxicam complex with a yield of 80.7% and 89.1%, respectively. This attributed to the insufficiency of $SnCl_2{\cdot}2H_2O$ present in the reaction mixture to reduce all pertechnetate to lower reduced states and so, the free pertechnetate is the predominant species in the solution. By increasing the amount of $SnCl_2{\cdot}2H_2O$ to 50 μg, the radiochemical yield of $^{99m}$Tc-piroxicam increased to >97%. On the other hand, at 200 μg $SnCl_2{\cdot}2H_2O$, a colloidal solution was obtained and this may be due to the formation of $Sn(OH)_2$ which is insoluble in the reaction medium.

*Effect of pH of the reaction medium*

The effect of pH of the reaction mixture on the percent yield of the labeling of piroxicam with $^{99m}$Tc was studied. The test was carried out at pH values ranging from 1 to 11 using different buffer systems. Piroxicam was found not soluble at acidic pH values 1 and 4 and partially soluble at neutral pH values 5.5 and 7, so, no labeling was done in the pH range of 1–7. Labeling at alkaline pH values 9 and 11 leads to the formation of $^{99m}$Tc-piroxicam complex with radiochemical yields of 95.9% and 97.3%, respectively, as shown in Table 3.

*Table 1.* Effect of the amount of piroxicam on the percent labeling yield of $^{99m}$Tc-piroxicam

| Piroxicam amount, mg | $^{99m}$Tc-piroxicam, % | $^{99m}TcO^{-}_{4}$, % |
|---|---|---|
| 0.1 | 88.3 ± 2.2 | 11.7 ± 1.5 |
| 0.3 | 90.1 ± 1.9 | 9.9 ± 0.9 |
| 0.5 | 97.3 ± 1.8 | 2.7 ± 0.3 |
| 1.0 | 95.5 ± 1.2 | 4.5 ± 0.4 |
| 2.0 | 95.5 ± 0.9 | 4.5 ± 0.2 |

Mean ± S.D. (mean of three experiments).
Reaction conditions: X mg piroxicam in bicarbonate buffer at pH 11, 50 μg $SnCl_2{\cdot}2H_2O$, 0.5 ml (~750 MBq) $^{99m}TcO^{-}_{4}$. The reaction mixture was kept at room temperature (25 °C) for 15 minutes.

*Table 2.* Effect of the amount of stannous chloride dihydrate on the percent labeling yield of piroxicam with $^{99m}$Tc

| Stannous chloride dihydrate amount, μg | $^{99m}$Tc-piroxicam, % | $^{99m}TcO^{-}_{4}$, % |
|---|---|---|
| 10 | 80.7 ± 1.8 | 19.3 ± 1.1 |
| 25 | 89.1 ± 2.2 | 10.9 ± 0.8 |
| 50 | 97.3 ± 2.4 | 2.7 ± 0.3 |
| 100 | 93.5 ± 1.2 | 6.5 ± 0.3 |
| 200 | Colloidal solution was formed | |

Mean ± S.D. (mean of three experiments).
Reaction conditions: 0.5 mg piroxicam in bicarbonate buffer at pH 11, X μg $SnCl_2{\cdot}2H_2O$, 0.5 ml (~750 MBq) $^{99m}TcO^{-}_{4}$. The reaction mixture was kept at room temperature (25 °C) for 15 minutes.

*Table 3.* Effect of pH of the reaction medium on the percent labeling yield of $^{99m}$Tc-piroxicam complex

| pH | $^{99m}$Tc-piroxicam, % | $^{99m}TcO^{-}_{4}$, % |
|---|---|---|
| 1–4 | Piroxicam was not soluble with the formation of ppt | |
| 5–7 | Piroxicam was partially soluble | |
| 9 | 95.9 ± 0.9 | 4.1 ± 0.4 |
| 11 | 97.3 ± 1.6 | 2.7 ± 0.5 |

Mean ± S.D. (mean of three experiments).
Reaction conditions: 0.5 mg piroxicam in bicarbonate buffer at different pH, 50 μg $SnCl_2{\cdot}2H_2O$, 0.5 ml (~750 MBq) $^{99m}TcO^{-}_{4}$. The reaction mixture was kept at room temperature (25 °C) for 15 minutes.

*Effect of the reaction time and reaction temperature*

The results of the influence of both time and temperature of the reaction medium on the radiochemical yield of $^{99m}$Tc-piroxicam are presented in Tables 4 and 5. The results clearly show that the complexation between $^{99m}$Tc and piroxicam is kinetically inert as the $^{99m}$Tc-piroxicam complex is formed at room temperature after 15-minute standing time and decomposed gradually by heating in water bath as shown in Table 4. The rate of decomposition increased as the time of heating increased. At 5-minute post boiling, $^{99m}$Tc-piroxicam complex decomposed by 12% while it decomposed by 48% after 30-minute boiling time. Also, the data presented in Table 5 show that the result of the degradation of $^{99m}$Tc-piroxicam by heating is due to the formation of free pertechnetate as it is equal to 50% after 30-minute boiling time.

*Effect of pertechnetate activity and in-vitro stability of $^{99m}$Tc-piroxicam*

The results of the in-vitro stability of $^{99m}$Tc-piroxicam are presented in Table 6. The data clearly show that $^{99m}$Tc-piroxicam complex is stable for up to 8 hours which application is suitable for the most nuclear medicine. Also, Table 7 summarizes data obtained from the effect of different pertechnetate activities ranging from 10 to 50 mCi (370–1850 MBq) in different volumes. The data show that there was no effect of both pertechnetate activity and dilution on the $^{99m}$Tc-piroxicam complex in the volume and activity range studied, as the corresponding activity detected, after the addition of 9 ml (1850 MBq ) pertechnetate was 95.8%.

*Biodistribution of $^{99m}$Tc-piroxicam in mice*

Piroxicam acts as an antiinflammatory drug via two mechanisms: (1) as an inhibitor of prostaglandin biosynthesis, also, it inhibits activation of neutrophils even when products of cyclooxygenase are present, and (2) it inhibits proteoglycanase and collagenase. The second mode of action is specific for piroxicam over indomethacin, so one can expect a higher accumulation of piroxicam than indomethacin in inflammatory sites.

The biodistribution mode of the labeled piroxicam ($^{99m}$Tc-piroxicam) was examined in groups of healthy mice, each of 3 mice. The tracer was injected in the tail vein and the mice were sacrificed at 0.5-, 2-, 4- and 24-hour post injection. The data of this experiment are presented in Table 8. It was noticed that the activity circulated with the blood was still high even at 24-hour post injection and was equal to 12.5%.

*Table 4.* Effect of the temperature of the reaction mixture on the percent labeling yield of piroxicam with $^{99m}$Tc

| Reaction time and temperature | $^{99m}$Tc-piroxicam, % | $^{99m}TcO^-_4$, % |
|---|---|---|
| 15 min at 25 °C | 97.3 ± 1.4 | 2.7 ± 0.2 |
| 5 min at 100 °C | 84.9 ± 1.7 | 15.1 ± 0.8 |
| 10 min at 100 °C | 81.9 ± 2.5 | 18.1 ± 1.2 |
| 20 min at 100 °C | 64.3 ± 1.2 | 35.7 ± 1.8 |
| 30 min at 100 °C | 49.7 ± 2.6 | 50.3 ± 2.3 |

Mean ± S.D. (mean of three experiments).
Reaction conditions: 0.5 mg piroxicam, in bicarbonate buffer at pH 11, 50 μg $SnCl_2 \cdot 2H_2O$, 0.5 ml (~750 MBq) $^{99m}TcO^-_4$. The reaction mixture was kept at 100 °C for different lengths of time.

*Table 5.* Effect of reaction time on the percent labeling yield of piroxicam with $^{99m}$Tc at room temperature (25 °C)

| Reaction time, min | $^{99m}$Tc-piroxicam, % | $^{99m}TcO^-_4$, % |
|---|---|---|
| 5 | 91.2 ± 1.5 | 8.8 ± 1.2 |
| 15 | 97.3 ± 1.3 | 2.7 ± 0.3 |
| 30 | 98.1 ± 0.9 | 1.9 ± 0.2 |
| 60 | 97.5 ± 0.8 | 2.5 ± 0.4 |
| 120 | 98.3 ± 0.9 | 1.7 ± 0.2 |

Mean ± S.D. (mean of three experiments).
Reaction conditions: 0.5 mg piroxicam, in bicarbonate buffer at pH 11, 50 μg $SnCl_2 \cdot 2H_2O$, 0.5 ml (~750 MBq) $^{99m}TcO^-_4$. The reaction mixture was kept at room temperature (25 °C) for different lengths of time.

*Table 6.* The in vitro stability of $^{99m}$Tc-piroxicam complex determined by electrophoresis

| Time post labeling, hour | $^{99m}$Tc-piroxicam, % | $^{99m}TcO^-_4$, % |
|---|---|---|
| 0.5 | 98.2 ± 1.4 | 1.8 ± 0.2 |
| 1 | 97.8 ± 1.7 | 2.2 ± 0.3 |
| 2 | 98.0 ± 1.2 | 2.0 ± 0.3 |
| 4 | 98.1 ± 1.3 | 1.9 ± 0.2 |
| 8 | 97.3 ± 1.3 | 2.7 ± 0.1 |
| 12 | 96.9 ± 1.6 | 3.1 ± 0.2 |

Mean ± S.D. (mean of three experiments).
Reaction conditions: 0.5 mg piroxicam in bicarbonate buffer pH 11, 50 μg $SnCl_2 \cdot 2H_2O$, 0.5 ml (~750 MBq) $^{99m}TcO^-_4$. The reaction mixture was kept at room temperature (25 °C).

*Table 7.* Effect of pertechnetate activity on the in-vitro stability of $^{99m}$Tc-piroxicam complex

| $^{99m}$Tc-pertechnetate activity, MBq | $^{99m}$Tc-piroxicam, % | $^{99m}TcO^-_4$, % |
|---|---|---|
| 1 ml ~ 370 | 97.3 ± 1.5 | 2.7 ± 0.3 |
| 2 ml ~ 740 | 96.9 ± 0.9 | 3.1 ± 0.1 |
| 5 ml ~ 1110 | 96.0 ± 1.0 | 4.0 ± 0.2 |
| 9 ml ~ 1850 | 95.8 ± 0.7 | 4.2 ± 0.1 |

Mean ± S.D. (mean of three experiments).
Reaction conditions: 0.5 mg piroxicam in bicarbonate buffer at pH 11, 50 μg $SnCl_2 \cdot 2H_2O$, X ml (~750 MBq) $^{99m}TcO^-_4$. The reaction mixture was kept at room temperature (25 °C) for 15 minutes.

This may be attributed to the binding of piroxicam to plasma protein and also due to the high biological half life of piroxicam which is near to 50 hours.[9] Also, the activity in the liver and intestine was relatively high being equal to 29.4% and 29.7% at 2-hour post injection, respectively. This accumulation may be due to the enterohepatic cycle of piroxicam. The high activity localized in liver decreased gradually as time passed while the intestine activity increased with time as the liver evacuated its content into the intestine. At 24-hour post injection, the activity held by the collected urine and feces increased due to the normal excretion pathway of piroxicam which was via liver and kidney.

*Detection of infections and sterile inflammatory foci in mice*

Typical biodistribution data of $^{99m}$Tc-labeled piroxicam are presented in Table 9. The mice were infected by the injection of 0.2 ml of *E-coli* in thigh muscles (for septic inflammation induction) and 0.2 ml of sterile turpentine oil (for sterile inflammation induction). The data were collected at 4- and 24-hour post injection . By comparing these data with the data collected after the injection of the labeled piroxicam in normal mice one can predict that there is no difference in organ uptake specially excretory organs, liver, kidney, intestine, besides urine, and feces.

*Table 8.* Biodistribution of $^{99m}$Tc-piroxicam in normal mice

| Organs and body fluid | % I.D./organ and body fluid at different post injection times | | | |
|---|---|---|---|---|
| | 30 min | 2 hrs | 4 hrs | 24 hrs |
| Liver | 20.4 ± 1.4 | 29.4 ± 1.5 | 23.8 ± 1.2 | 8.8 ± 0.6 |
| Urine | 22.6 ± 0.9 | 27.4 ± 0.9 | 26.7 ± 1.5 | 31.6 ± 1.6 |
| Kidneys | 5.1 ± 0.5 | 4.1 ± 0.6 | 3.8 ± 0.4 | 1.2 ± 0.1 |
| Blood | 16.8 ± 1.2 | 16.2 ± 1.3 | 15.5 ± 1.0 | 12.5 ± 1.1 |
| Heart | 0.2 | 0.3 ± 0.1 | 0.3 ± 0.1 | – |
| Lung | 0.6 ± 0.1 | 0.4 ± 0.1 | 0.3 | – |
| Intestine and stomach | 12.0 ± 1.1 | 29.7 ± 1.4 | 39.1 ± 2.8 | 11.1 ± 1.1 |
| Spleen | 0.5 ± 0.1 | 0.3 | 0.3 ± 0.1 | – |
| Muscle | 1.5 ± 0.2 | 1.4 ± 0.1 | 0.9 ± 0,2 | 0.3 |
| Bone | 1.3 ± 0.1 | 1.9 ± 0.2 | 1.1 | 0.7 ± 0.1 |
| Collected feces | – | – | 6.2 ± 0.7 | 35.2 ± 2.9 |

Mean ± S.D. (mean of three experiments).
Vial content: 0.5 mg piroxicam in bicarbonate buffer at pH 11, 50 μg $SnCl_2 \cdot 2H_2O$, 0.5 ml (~750 MBq) $^{99m}TcO^-_4$. The reaction mixture was kept at room temperature (25 °C) for 15 minutes.

*Table 9.* Biodistribution of $^{99m}$Tc-piroxicam in inflamed mice (septic and sterile) at different post injection times

| Organs and body fluid | %I.D./ organ and body fluid at different post injection times | | | |
|---|---|---|---|---|
| | Turpentine oil | | *E-coli* | |
| | 4 hrs | 24 hrs | 4 hrs | 24 hrs |
| Liver | 20.8 ± 1.2 | 10.3 ± 1.2 | 18.4 ± 1.4 | 9.5 ± 0.7 |
| Urine | 18.2 ± 1.5 | 32.4 ± 2.3 | 23.6 ± 2.1 | 31.2 ± 2.4 |
| Kidneys | 4.2 ± 0.5 | 0.9 ± 0.2 | 2.1 ± 0.4 | 0.5 ± 0.1 |
| Blood | 10.6 ± 0.9 | 9.3 ± 0.9 | 13.5 ± 0.9 | 11.4 ± 1.2 |
| Heart | 0.4 | 0.2 | 0.5 ± 0.1 | 0.2 |
| Lung | 0.7 ± 0.1 | 0.4 | 0.3 | – |
| Intestine and stomach | 28.6 ± 2.1 | 8.2 ± 0.5 | 25.3 ± 2.5 | 5.5 ± 1.4 |
| Spleen | 0.6 ± 0.1 | 0.2 | 0.5 | 0.1 |
| Infected muscle | 4.2 ± 0.2 | 5.6 ± 0.6 | 5.3 ± 0.2 | 6.1 ± 0.7 |
| Non-infected muscle | 1.1 | 0.5 ± 0.1 | 1.4 ± 0.1 | 0.7 ± 0.2 |
| Bone | 1.2 | 1.0 ± 0.1 | 1.6 ± 0.1 | 0.9 |
| Collected feces | 4.5 ± 0.3 | 25.6 ± 1.3 | 3.6 ± 0.3 | 30.0 ± 3.5 |

Mean ± S.D. (mean of three experiments).
Vial content: 0.5 mg piroxicam in bicarbonate buffer at pH 11, 50 μg $SnCl_2 \cdot 2H_2O$, 0.5 ml (~750 MBq) $^{99m}TcO^-_4$. The reaction mixture was kept at room temperature (25 °C) for 15 minutes.

Both infected and non-infected inflamed thigh muscles had high activity at 4-hour post injection which was equal to 5.3% and 4.2%, respectively. This uptake increased as time passed to 6.1% and 5.6% at 24-hour post injection while other non excretory organs declined. The target/non target muscle for both sterile and septic inflamed mice was equal to 10.6 and 8.7 at 24-hour post injection. This means that this $^{99m}$Tc labeled compound is able to localize in inflammatory foci of both kinds septic and sterile and can be used as an inflammatory imaging agent specially after 24-hour post injection.

## Conclusions

The amount of piroxicam has no effect on the percent yield of $^{99m}$Tc-piroxicam and 0.5 mg is optimum for high labeling yield >97%. The use of 50 μg stannous chloride dihydrate was found sufficient to produce high radiochemical yield. Above this quantity (200 μg) a turbid solution was obtained due to the formation of the colloid, especially in highly alkaline reaction medium. Due to the insolubility of piroxicam in acidic medium and partial solubility in neutral pH, the labeling reaction was done at highly alkaline pH value 9 and 11. The labeling of piroxicam with $^{99m}$Tc proceeds well at room temperature (25 °C) but after 15 minutes it decomposes with heat.

Upon the injection of $^{99m}$Tc-piroxicam in both normal and inflammed mice, it was found that the complex distributed well all over the organs of the body and bound for long time with the plasma protein. About 12% of the injection activity was detected in the blood at 24-hour post injection. Also, the complex was taken with high percentage by both septic and aseptic foci which can be detected well at 24-hour post injection when all the activity of the other organs are cleared except that of excretory ones.

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