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Radioiodination, purification and bioevaluation of Piroxicam in comparison with Meloxicam for imaging of inflammation

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The present study is performed to compare the electrophilic substitution radioiodination reaction of two non-steroidal anti-inflammatory drugs namely, Piroxicam (Pirox) and Meloxicam (Melox) with ^{125}I where both chloramine-T (CAT) and iodogen were used as oxidizing agents. The factors affecting the percent of radiochemical yields such as drug concentration, pH of the reaction mixtures, different oxidizing agents, reaction time, temperature and different organic media were studied to optimize the conditions for labeling of Pirox and Melox and to obtain high radiochemical yields. The maximum radiochemical yield of ^{125}I-Piroxicam (^{125}I-Pirox) was 94% using 3.7 MBq of $Na^{125}I$, 0.4 mM of Pirox as substrate, 3.6 mM of chloramine-T (CAT) as oxidizing agent in acetone at neutral pH = 7 and at 60 °C within 20 min where the maximum radiochemical yield of ^{125}I-Melox was 92% using 0.7 mM of Melox as substrate, 0.62 mM of iodogen as oxidizing agent in acetone at neutral pH = 7 and at 25 °C within 30 min. The radiochemical yields were determined by TLC and high-pressure liquid chromatography (HPLC). Tracers showed good localization in inflamed muscle either septic or sterile. The collected data indicates that Pirox and Melox can be used as anti-inflammatory imaging agents at 24 and 2 h post injection, respectively.

Introduction

Even in most recent decades, infection and inflammation remain important reasons of mortality and morbidity globally. Infections, especially internal infections, were difficult to detect at early stages, resulting in delayed diagnosis, treatment and sometimes death. Clinicians use a variety of clues, e.g., clinical, laboratory, and radiological tests to give a good diagnosis of infection as early as possible. Piroxicam (Pirox) is one of the oxicam derivatives, a class of enolic acid that have anti-inflammatory, analgesic and antipyretic activity (Fig. 1). It is an inhibitor of prostaglandin biosynthesis, it also inhibit activation of neutrophils even when products of cyclooxygenase are present at inflammatory sites; hence additional mode of anti-inflammatory action have been proposed including inhibition of proteoglycanase and collagenase in cartilage.[1,2] Piroxicam was labeled effectively with ^{99m}Tc due to the presence of electron donating atoms such as sulfur, nitrogen, and oxygen in its structure.[3] Meloxicam (Melox) (Fig. 1) is a novel non-steroidal anti-inflammatory drug (NSAID) of the acidic enolcarboxamide class, structurally related to piroxicam. Meloxicam significantly decreased symptoms of pain, function, and stiffness in patients, with a low incidence of gastrointestinal side effects. In models, it exhibits anti-inflammatory, analgesic, and antipyretic activities. Meloxicam was radiolabeled by introducing a ^{14}C atom into the carboxamide group of the molecule. The concentrations of [^{14}C]Meloxicam and its metabolites in blood, plasma, tissue, urine, bile, and feces were measured using a liquid scintillation counter (Packard TriCarb 3385).[4]

The present work concerns on comparing the electrophilic labeling of Piroxicam and Meloxicam by ^{125}I and study the factors, affect the radiochemical yield. The labeled compounds are purified by using HPLC for studying the biodistribution in normal mice and inflamed mice.

Experimental

Materials and methods

All chemicals and laboratory reagents used during this work were of the highest purity analytical grade. Meloxicam and Piroxicam were kindly supplied by the Egyptian International Pharmaceutical Industries Company (EIPICO), Egypt. Acetone was used as a solvent without further purification. Double distilled and deoxygenated water was used for all experiments. Chloramine-T [N-chloro-p-toluene sulfonamide salt (CAT)] from Aldrich and iodogen (1,3,4,6-tetrachloro-3,6-diphenyl glycoluril) from Pierce Chemical Company. Thin layer chromatography (TLC) aluminum sheets (20×25 cm) SG-60 F_{254} (Merck). $Na^{125}I$ (185 MBq/5 μL) in diluted NaOH, pH 7–11 was purchased from Institute of Isotopes, Budapest, Hungary.

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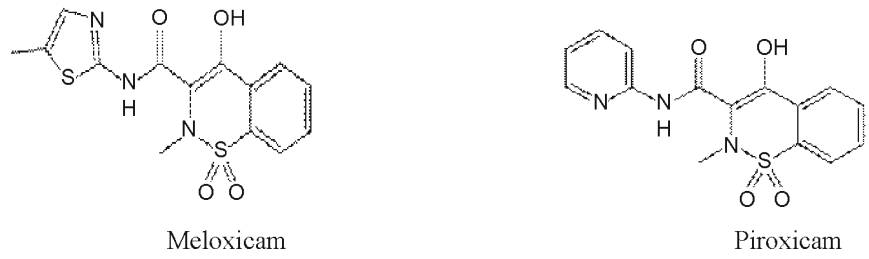

Fig. 1. Structure formula of the studied drugs Piroxicam and Meloxicam

Equipment

Radioactivity was measured by means of a γ-counter (Nucleus Model 2010) connected with a well type NaI(Tl) crystal. High performance liquid chromatography (HPLC) [Shimadzu Model], LC-9A pump, equipped with a Rheodyne injector (Syringe Loading Sample Injector-7125), UV spectrophotometric detector shimadzu SPD-6A and stationary phase comprising a reversed phase nucleosil phenyl column (250 mm×4.6 mm, 5 μm).

Animal

Mice: Albino type weighing 20–25 g, were used for biological distribution study.

Labeling of Piroxicam and Meloxicam with $Na^{125}I$

Chloramine-T method. In 1.0 mL volume reaction vial that could be tightly closed by a screw cap containing 100 μL of 0.2M phosphate buffer, pH 7.0 a suitable concentration of Pirox and Melox placed and an appropriate quantity of freshly prepared CAT were added. For labeling, 10 μL of $Na^{125}I$ (3.7 MBq) was added, and then the reaction mixture was kept in a water bath for different time. The reaction was quenched by the addition of sodium metabisulphite.

Iodogen method. Iodogen is essentially insoluble in water; 5.0 mg of iodogen was weighed in a clean glass culture tube and dissolved in 5.0 mL chloroform. An appropriate volume of this solution was added to clean glass culture tubes and dried under nitrogen atmosphere. Iodogen was deposited on the wall of the glass tubes as a thin film. The tubes were then closed and stored at 4 °C till use. Iodination reactions were directly carried out in these tubes, 100 μL of 0.2M phosphate buffer, pH 7.0 and suitable concentration of Pirox and Melox were placed. For labeling, 10 μL of $Na^{125}I$ (3.7 MBq) was added, and then, the reaction mixture was kept in a water bath for different times. The reaction was quenched by adding sodium metabisulphite.

Determination of radiochemical yield and purity

The radiochemical yields were determined by TLC and HPLC.

TLC analysis. This technique was done using thin layer silica gel coated on aluminum sheet (20 cm×20 cm). It was cut into strips each strip was 1.5 cm width and 13 cm length. The spotted point is placed 2 cm above the edge. The solvent used for developing was methylene chloride : ethyl acetate mixture (3 : 7 v/v). Radioiodide ^{125}I remained near the origin (Rf = 0–0.1), while the labeled compounds of Pirox (^{125}I-Pirox) and Melox (^{125}I-Melox) moved with the solvent front (Rf = 0.7–0.8). The radiochemical yields (%) at time (t), were calculated as the percent ratio of activity of labeled compounds relative to the total activity on the TLC-strip according to the following equation:

$$\text{Radiochemical yield, \%} = \frac{\text{Activity of labeled product} \times 100}{\text{Total activity}}$$

HPLC analysis. The radiochemical yields and the purities of ^{125}I-Pirox and ^{125}I-Melox were determined by direct injection of 10 μL of the reaction mixture at the optimum conditions for obtaining the highest radiochemical yields, into the column (Rp-18. 250 mm×4.6 mm, 5 μm) built in HPLC (Shimadzu model), which consists of pumps LC-9A, Rheohydron injector (Syringe Loading Sample Injector-7125) and UV spectrophotometer detector (SPD-6A) adjusted to the 230 nm wavelength using methanol : water (70 : 30 v/v) as mobile phase with a flow rate of 1.0 mL/min.[5] The labeled compound was collected separately by using a fraction collector up to 12, evaporated under reduced pressure, dissolved in saline solution and sterilized by Millipore (0.22 μm) under aseptic condition and its activity was counted by using well a type NaI(Tl) crystal connected to a single-channel analyzer. Figure 2 shows that the free radioiodide was separated at the retention time of 4 min while the labeled ^{125}I-Pirox was separated at 7.0 min. Figure 3 shows that the labeled ^{125}I-Melox was separated at 8.0 min.

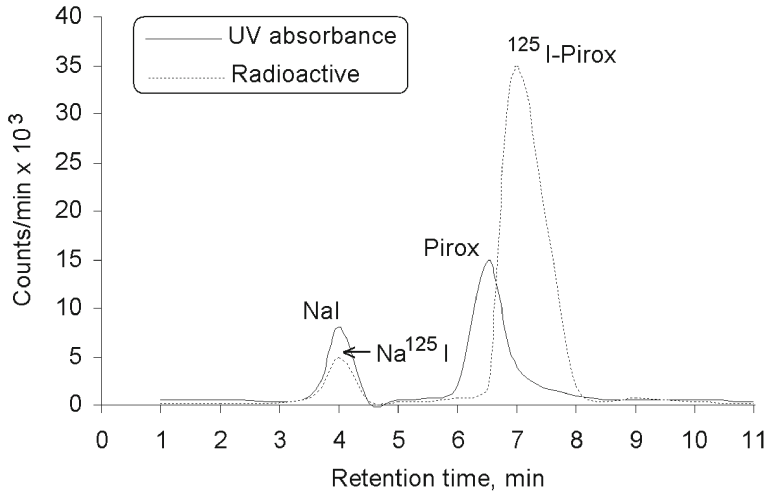

Fig. 2. HPLC radiochromatogram and UV profile of ^{125}I-Pirox; conditions: solvent: methanol : water (70 : 30 v/v) as mobile phase, column: RP18, flow rate: 1 mL/min and UV wavelength: 230 nm

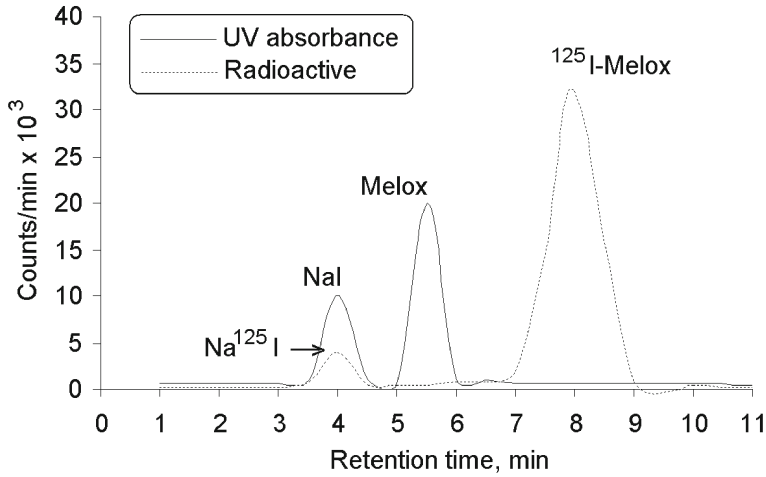

Fig. 3. HPLC radiochromatogram and UV profile of ^{125}I-Melox; conditions: solvent: methanol : water (70 : 30 v/v) as mobile phase, column: RP18, flow rate: 1 mL/min and UV wavelength: 230 nm

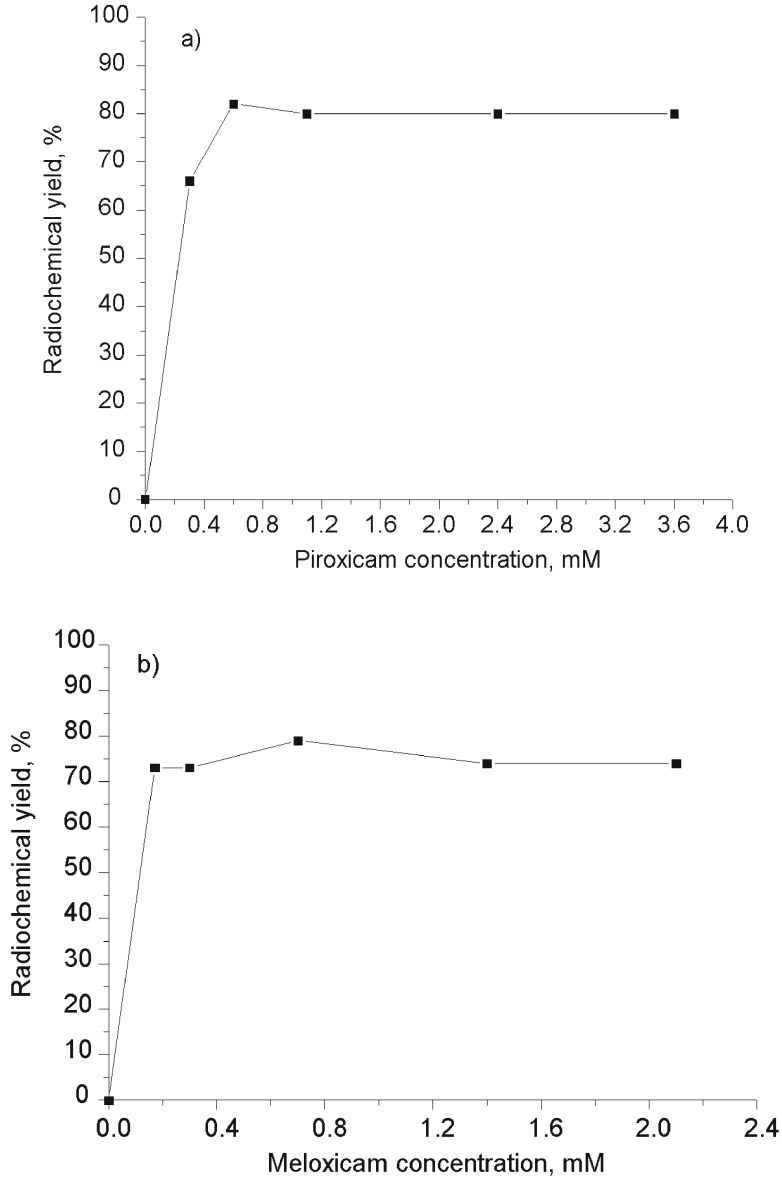

Fig. 4. Variation of the radiochemical yield of (a) ^{125}I-Pirox and (b) ^{125}I-Melox as a function of Pirox and Melox concentrations. 1.1 mM CAT and 10 μL of 3.7 MBq Na^{125}I in pH 7.0, x mM Pirox and Melox in acetone, at 60 °C for 20 min

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, ^{125}I-Pirox and ^{125}I-Melox 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, 3 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-counter (SR 7). Results are expressed as percent of the injected dose per organ or body fluid.

Results and discussion

Effect of substrate concentration

The influence of Pirox and Melox concentrations on the percent of radiochemical yields of ^{125}I-Pirox and ^{125}I-Melox by using 1.1 mM CAT at pH 7 and 60 °C for

20 min were investigated as shown in Fig. 4 (a, b). The results indicated that the radiochemical yields of both tracers increase with increasing the concentrations of Pirox to 0.4 mM and Melox to 0.7 mM up to 82% and 79%, respectively. This indicates that both of them are sensitive towards the electrophilic substitution reactions. Further increase in Pirox and Melox concentrations, show a slight decrease in the percent of radiochemical yields. This decrease in the yields may be due to the high excess of the molar concentration of tracers in comparison with the radioactive iodine atoms.[6]

Effect of pH

The variation of the radiochemical yields of ^{125}I-Pirox and ^{125}I-Melox with CAT as a function of pH were investigated at a pH range from 2.0 to 11. The reactions were carried out by using 1.1 mM CAT at 60 °C within 20 min and 0.4 mM Pirox or 0.7 mM Melox. Figure 5 shows that the maximum radiochemical yields of ^{125}I-Pirox and ^{125}I-Melox are obtained at pH=7.0. This is due to the fact that iodide oxidized easily to iodonium cation I^+ at pH 7.0 and the ring ionized to anion by loss H^+ ion and the iodination reaction occurs.[7] At acidic pH radiochemical yields of both tracers decreased, this may be attributed to the predominance of ICl species which have low oxidation potential less than HOCl.[8] At alkaline pH the yield also decreased markedly reaching to 52% and 51% of ^{125}I-Pirox and ^{125}I-Melox, respectively, as a result of decreasing HOI which was responsible for the electrophilic substitution reaction.[9]

Effect of CAT concentration

Radioiodination of organic molecules has been performed by using a mild oxidizing agent such as CAT, which decomposes to hypochlorite anion that acts as an oxidizing agent, transforming iodine from I^- to oxidative state I^+. The radioiodination of Pirox is highly dependent on the concentration of CAT. Figure 6 shows, at low concentration of CAT, radiochemical yield of ^{125}I-Pirox was very low. This can be mainly attributed to the insufficiency of CAT to oxidize iodide ions to the iodonium ions.[10] The maximum yield was 94% of ^{125}I-Pirox using 3.6 mM CAT at 60 °C within 20 min. In case of radioiodination of Melox the radiochemical yield increases with increasing concentration of CAT. The maximum yield was 85% of ^{125}I-Melox using 1.7 mM CAT at 60 °C within 20 min. Thereafter, the yield decrease with increasing concentration of CAT above 1.7 mM the yield decrease to 54%. This is due to the fact that the high concentration of CAT causes a number of undesirable oxidative side reactions including chlorination,[11] polymerization and denaturation of substrate.[9]

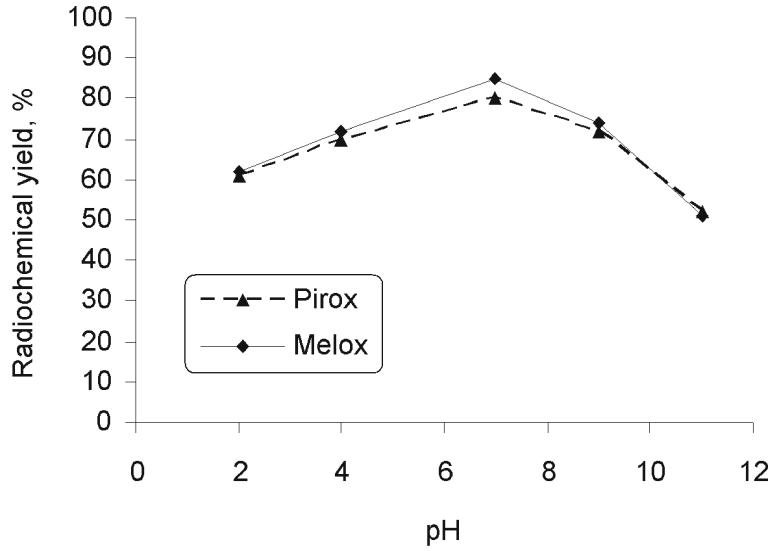

Fig. 5. Variation of the radiochemical yield of ^{125}I-Pirox and ^{125}I-Melox as a function of pH.
0.4 mM Pirox or 0.7 mM Melox, 1.1 mM CAT and 3.7 MBq Na^{125}I in 100 μL of buffer with different pH, at 60 °C within 20 min

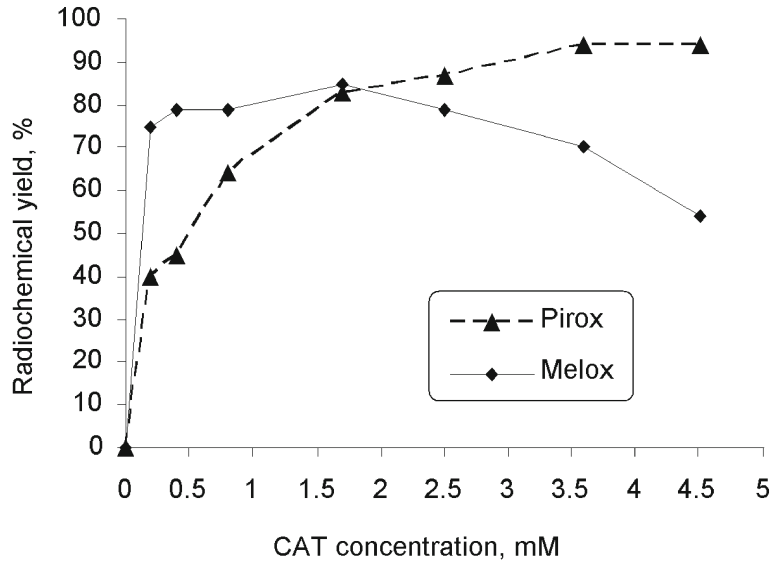

Fig. 6. Variation of the radiochemical yield of ^{125}I-Pirox and ^{125}I-Melox as a function of the Chloramine-T concentration. 0.4 mM Pirox or 0.7 mM Melox, x mM CAT and 3.7 MBq Na^{125}I in pH 7 at 60 °C within 20 min

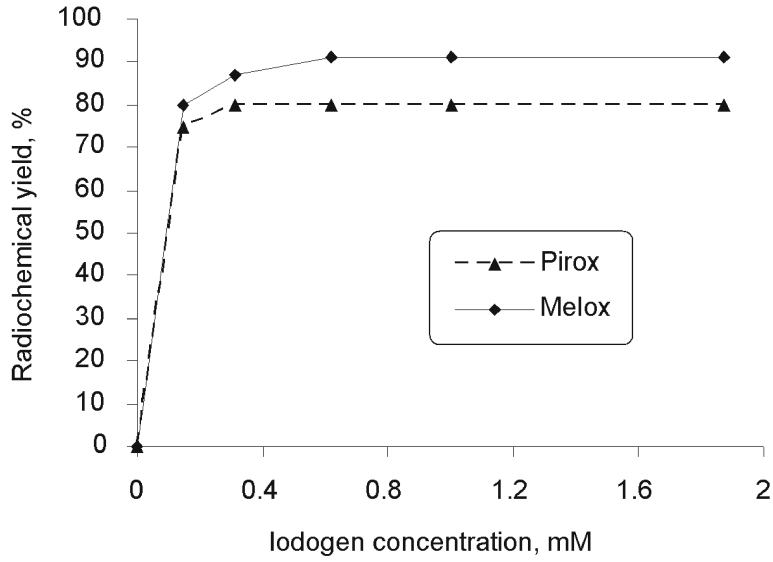

Fig. 7. Variation of the radiochemical yield of ^{125}I-Pirox and ^{125}I-Melox as a function of the iodogen concentration. 0.4 mM Pirox or 0.7 mM Melox, x mM iodogen and 3.7 MBq Na^{125}I in pH 7 at 60 °C within 20 min

Effect of iodogen concentration

Iodogen is a moderate oxidizing agent, resembling a 4-folds CAT, but with less oxidative damage effect.[12] Figure 7 illustrates the effect of iodogen concentration on the radiochemical yields of ^{125}I-Pirox and ^{125}I-Melox. The radiochemical yields of tracers increased with increasing concentration of iodogen. The maximum radiochemical yields reach to 80% and 91% of ^{125}I-Pirox and ^{125}I-Melox, respectively. Increasing the concentration of iodogen above 0.62 mM did not increase the yield.

Effect of the reaction temperature

The radioiodination of Pirox and Melox by ^{125}I were carried out by studying the effect of the reaction temperature (25–100 °C) using 0.4 mM Pirox and 3.6 mM CAT at pH 7.0 and 0.7 mM Melox and 0.62 mM iodogen. The data indicate that the reaction temperature is a significant factor that affects the labeling yield. As shown in Figs 8 and 9 the radiochemical yield of ^{125}I-Pirox was very low at room temperature (75%) while the radiochemical yield of ^{125}I-Melox is 92% at room temperature. By increasing the reaction temperature to 60 °C the radiochemical yield of ^{125}I-Pirox reaches 94% but the radiochemical yield of ^{125}I-Melox remains constant. On the other hand, increasing the reaction temperature above 60 °C up to 100 °C the radiochemical yields of both tracers decrease to 60% for Pirox and 61% for Melox within 20 min. This may be attributed to the thermal decomposition of the labeled compound or distortion of the oxidizing agent.[13]

Effect of solvent

By applying the optimum condition for labeling of Pirox (0.4 mM), 3.6 mM CAT and 10 μL 3.7 MBq Na^{125}I in pH 7.0 at 60 °C within 20 min and the optimum condition for labeling of Melox (0.7 mM), 0.62 mM iodogen and 10 μL 3.7 MBq Na^{125}I in pH 7.0 at 25 °C within 30 min different solvents such as ethanol, acetonitrile, dimethyl sulphoxide (DMSO), acetone and methylene chloride were also studied. The results given in Table 1 indicate that the most suitable solvent is acetone for both cases; this may be attributed to Pirox and Melox which are completely soluble in acetone. The radiochemical yields of Pirox and Melox reach to 94% and 92%, respectively. Using DMSO as a solvent gave poor radiochemical yields for both tracers in spite of the advantage characteristics of the dipolar aprotic which include a high boiling point, ability to solvate in a broad variety of solutes and to be useful in radioiodination reaction.[14,15]

Table 1. Effect of different solvents

Solvent types	Radiochemical yield of Pirox, %	Radiochemical yield of Melox, %
Acetone	94	92
Acetonitrile	74	48
Ethanol	46.5	52
Methylene chloride	52	75
Dimethyl sulphoxide	42	29

Table 2. Stability of ^{125}I-Pirox and ^{125}I-Melox

Time post labeling, h	Radiochemical yield of Pirox, %	Radiochemical yield of Melox, %
1	94.2	92.1
2	94	91
3	94.2	92
4	92.2	89
12	94.4	89.2
16	94	89
24	94.1	89.1

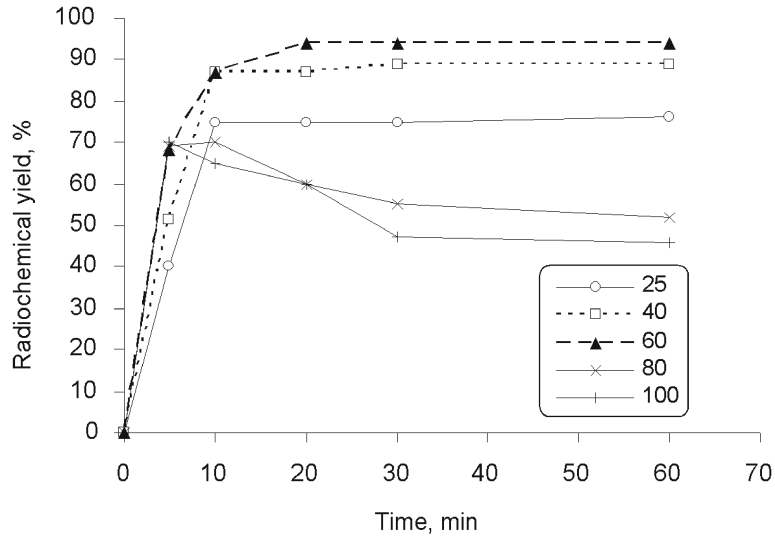

Fig. 8. Variation of the radiochemical yield of ^{125}I-Pirox as a function of the reaction time. 0.4 mM Pirox, 3.6 mM CAT and 3.7 MBq Na^{125}I in pH 7 at different temperature

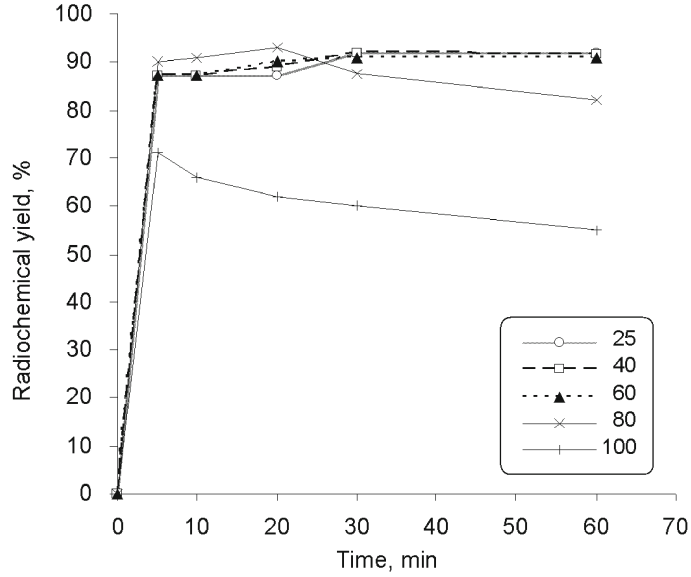

Fig. 9. Variation of the radiochemical yield of ^{125}I-Melox as a function of the reaction time.
0.7 mM Melox, 0.62 mM iodogen and 3.7 MBq Na^{125}I in pH 7at different temperature

In-vitro stability of ^{125}I-Pirox and ^{125}I-Melox

The stability of ^{125}I-Pirox and ^{125}I-Melox studied in order to determine the suitable injection to avoid the formation of the undesired products which were resulted from the radiolysis of the labeled compounds. These undesired radioactive products may be toxic or accumulated in undesired organ. Table 2 shows that ^{125}I-Pirox was stable up to 24 h, while ^{125}I-Melox was stable up to 4 h then the radiochemical yield slightly decreased.

Biodistribution studies

Normal mice. The biodistribution mode of labeled ^{125}I-Pirox and ^{125}I-Melox were examined in groups of healthy mice, each of three mice. The tracers were injected in the tail vain and the mice sacrificed at 0.5, 1.0 and 3.0 h post injection. The data of this study were presented in Tables 3 and 4. Table 3 shows that the activity circulated by labeled Pirox with the blood was still high even at 3.0 h post injection and equal to 12.6%. This may be attributed to the binding of Pirox to plasma protein and also due to the high biological half life of Pirox which is near to 50 h.[16] The activity in the liver and intestine was high and equal to 30.1% and 20.1%, respectively, at 2.0 h post injection, due to the enterohepatic cycle of Pirox.[3] The activity localized in liver, intestine and kidneys decreased at 3.0 h post injection and the activity held by the collected urine and feces due to the normal excretion pathway of Pirox which is via the liver and kidney. The other organs (heart, lung, spleen and bone) show no activity uptake. By comparing the data collected after the injection of labeled Melox presented in Table 4 with the data collected after the injection of labeled Pirox it appeared that there is no difference in organs uptake.

Infected and sterile inflamed mice

The mice were infected by injection of 0.2 mL of E. Coli in thigh muscle (for septic inflammation induction) and 0.2 mL of sterile turpentine oil (for sterile inflammation induction). Labeled Pirox and Melox was injected in the tail vain of infected and sterile inflammed mice. The data are presented in Tables 5 and 6. The data in Table 5 indicates that both infected and sterile inflammed mice when injected by ^{125}I-Pirox were similar where at 4 hours post injection the activity equals to 35.2% and 34.1% for infected muscle and sterile muscle, respectively. The activity decreased to 26.4% and 29.6% for infected muscle and sterile muscle, respectively, at 24 hours post injection. The injection of labeled Melox are presented in Table 6. The activity uptake in the blood was high at 0.5 h and reached up to 17.7% and 13.9% for infected and sterile mice, respectively, where at 24 hours the activity decreased to 4% and 3.1%. Also the infected and sterile muscle had high activity at 2 hours post injection and equals to 35.3% and 33%, respectively, then decreased at 24 hours to 9.6% and 9.3%. This loss in activity uptake is related to the introduction of methyl group in thizoloyl moiety of Melox has facilited of the formation of metabolites[17] that undergo fast elimination, leading to a short biological half life ($T_{1/2}$) equal to 23 hours in comparison with Pirox.

Table 3. Biodistribution of ^{125}I-Pirox in normal mice

Organs and body fluids	Injected dose/organ percent at different intervals of time post injection, h		
	0.5	2	3
Blood	14.2 ± 1.4	16.2 ± 1.5	12.6 ± 1.3
Bone	2.8 ± 0.9	1.4 ± 1.2	0.9 ± 0.7
Muscle	9.2 ± 0.5	5.0 ± 0.7	2.0 ± 0.6
Lung	0.3 ± 0.1	0.1 ± 0.2	0.6 ± 0.1
Heart	0.2 ± 0.2	0.1 ± 0.1	0.1 ± 0.1
Liver	20.2 ± 1.2	30.1 ± 1.4	20.8 ± 1.5
Kidneys	5.1 ± 0.8	3.1 ± 1.2	1.2 ± 0.9
Spleen	0.1 ± 0.2	0.1 ± 0.4	–
Stomach	14.4 ± 0.6	21.5 ± 2.3	8.8 ± 2.4
Intestine	16.2 ± 2.4	20.1 ± 2.7	26.3 ± 2.5
Urine	2.1 ± 0.9	19.2 ± 1.1	5.2 ± 0.8
Fecess	–	13.1 ± 0.4	22.4 ± 0.3

Mean ± S.D. (mean of three experiments).

Table 4. Biodistribution of ^{125}I-Melox in normal mice

Organs and body	Injected dose/organ percent at different intervals of time post injection, h		
	0.5	2	3
Blood	18.7 ± 1.2	20.9 ± 0.8	10.7 ± 0.4
Bone	0.6 ± 0.2	7.1 ± 0.2	4.9 ± 1.3
Muscle	6.8 ± 0.3	11.0 ± 0.2	10.2 ± 0.3
Lung	0.8 ± 0.1	0.4 ± 0.2	0.4 ± 0.2
Heart	1.0 ± 0.2	0.1 ± 0.1	0.1 ± 0.3
Liver	14.6 ± 0.2	20.4 ± 0.2	12.7 ± 1
Kidneys	17.1 ± 0.8	5.7 ± 1.2	1.2 ± 0.9
Spleen	0.3 ± 0.2	0.1 ± 0.3	–
Stomach	37.5 ± 0.5	8.1 ± 2.3	2.5 ± 2.1
Intestine	14.6 ± 2.6	26.6 ± 2.1	5.7 ± 2.7
Urine	14.4 ± 0.6	12.6 ± 1.6	20.3 ± 0.6
Fecess	–	13.1 ± 1.4	23.4 ± 0.1

Mean ± S.D. (mean of three experiments).

Table 5. Biodistribution of ^{125}I-Pirox in inflamed mice (septic and sterile)

Organs and body fluids	Injected dose/organ percent at different intervals of time post injection, h					
	Turpentine oil			E-coli		
	0.5	2	24	0.5	2	24
Blood	19.7 ± 1.2	21.8 ± 1.8	13.0 ± 0.5	12.8 ± 1.2	17.7 ± 1.5	17.0 ± 1.2
Bone	3.1 ± 0.2	1.5 ± 0.2	1.6 ± 0.1	2.8 ± 0.1	3.6 ± 0.1	1.1 ± 0.1
Lungs	2.8 ± 0.1	1.7 ± 0.2	0.8 ± 0.2	0.3 ± 0.1	0.3 ± 0.1	–
Heart	0.8 ± 0.1	0.7 ± 0.1	0.6 ± 0.1	0.2 ± 0.1	0.1 ± 0.1	–
Liver	10.4 ± 0.2	5.0 ± 0.3	7.0 ± 0.3	14.3 ± 0.2	20.2 ± 1.4	4.4 ± 0.1
Kidneys	7.9 ± 0.2	9.3 ± 0.4	13.3 ± 0.4	5.0 ± 0.1	4.3 ± 0.2	1.2 ± 0.2
Spleen	0.1 ± 0.1	0.9 ± 0.1	0.3 ± 0.1	0.5 ± 0.1	0.1 ± 0.1	0.05 ± 0.1
Stomach	4.0 ± 0.5	10.6 ± 0.4	8.4 ± 0.4	26.4 ± 0.5	30.2 ± 2.1	17.0 ± 1.1
Intestine	11.7 ± 0.4	11.3 ± 1.4	7.0 ± 0.9	0.2 ± 0.3	10.5 ± 1.1	1.4 ± 0.1
Urine	3.0 ± 0.2	4.7 ± 0.2	14.5 ± 1.2	8.7 ± 2.4	16.4 ± 1.5	27.3 ± 1.1
Feces	4.5 ±	12.6 ± 0.2	18.4 ± 0.5	–	26.2 ± 1.1	40.2 ± 1.3
Infected muscle	26.3 ± 1.5	34.1 ± 1.2	26.4 ± 1.4	28.9 ± 1.2	35.2 ± 1	29.6 ± 1.7
Non infected muscle	9.7 ± 0.5	2.5 ± 0.1	0.1 ± 0.1	7.0 ± 0.2	7.7 ± 1.4	2.2 ± 1.1

Mean ± S.D. (mean of three experiments).

Table 6. Biodistribution of ^{125}I- Melox in inflamed mice (septic and sterile)

Organs and body fluids	Injected dose/organ percent at different intervals of time post injection, h					
	Turpentine oil			E-coli		
	0.5	2	24	0.5	2	24
Blood	17.7 ± 1.2	12.5 ± 1.8	4.0 ± 0.5	16.7 ± 1.2	17.9 ± 1.5	3.1 ± 1.2
Bone	18.7 ± 0.2	6.2 ± 0.2	0.2 ± 0.1	24.7 ± 0.1	3.5 ± 0.1	0.9 ± 0.1
Lungs	0.3 ± 0.1	0.4 ± 0.2	0.05 ± 0.2	0.1 ± 0.1	0.1 ± 0.1	–
Heart	0.1 ± 0.1	0.1 ± 0.1	–	0.2 ± 0.1	0.1 ± 0.1	–
Liver	13.4 ± 0.2	17.5 ± 0.3	2.0 ± 0.3	14.3 ± 0.2	20.2 ± 1.4	4.4 ± 0.1
Kidneys	7.9 ± 0.2	4.0 ± 0.4	3.3 ± 0.4	9.1 ± 0.1	4.3 ± 0.2	1.2 ± 0.2
Spleen	0.3 ± 0.1	0.1 ± 0.1	–	0.3 ± 0.1	0.2 ± 0.1	–
Stomach	8.1 ± 0.5	14.6 ± 0.4	0.7 ± 0.4	9.5 ± 0.5	17.3 ± 2.1	7.0 ± 1.1
Intestine	3.1 ± 0.4	10.2 ± 1.4	1.5 ± 0.9	3.28 ± 0.3	21.7 ± 1.1	0.9 ± 0.1
Urine	0.4 ± 0.2	16.6 ± 0.2	31.6 ± 1.2	0.4 ± 2.4	16.0 ± 1.5	24.7 ± 1.1
Feces	4.5 ±	2.6 ± 0.2	8.4 ± 0.5	–	6.2 ± 1.1	37.2 ± 1.3
Infected muscle	22.8 ± 1.5	35.3 ± 1.2	9.6 ± 1.4	25.8 ± 1.2	33.0 ± 1	9.3 ± 1.7
Non infected muscle	2.8 ± 0.5	5.5 ± 0.1	2.5 ± 0.1	2.6 ± 0.2	8.3 ± 1.4	4.5 ± 1.1

Mean ± S.D. (mean of three experiments).

Conclusions

Piroxicam and Meloxicam are labeled easily with ^{125}I. The optimum condition for labeling of Pirox is 0.4 mM Pirox, 3.6 mM CAT and 3.7 MBq $Na^{125}I$ in pH 7 within 20 min at 60 °C and the optimum condition for labeling of Melox is 0.7 mM Melox, 0.62 mM iodogen and 3.7 MBq $Na^{125}I$ in pH 7 within 30 min at 25 °C. The labelled Pirox is able to localize in inflammatory foci of both kinds (septic and sterile) and can be used as inflammatory imaging agent till 24 hours post injection where labeled Melox is able to localize in inflammatory foci of both kinds (septic and sterile) but used as inflammatory imaging agent specially at 2 hours post injection.

References

1. A. J. LEWIS, D. W. FURST (Eds), Non-steroidal Anti-Inflammatory Drugs. Mechanisms and Clinical Use, Marcel Dekker, New York, 1987.
2. S. R. ABRAMSON, G. WEISSNMANN, Arthritis Rheum., 32 (1989) 1.
3. E. A. EL-GHANY, A. M. AMINE, A. S. EL-SAYED, M. T. EL-KOLALY, F. ABDEL-GELIL, J. Radioanal. Nucl. Chem., 125 (2005) 266.
4. U. BUSCH, J. SCHMID, G. HEINZEL, H. SCHMAUS, J. BAIERL, C. HUBER, W. ROTH, The American Society for Pharmacology and Experimental Therapeutics, (1998) 26.
5. M. S. ARAYNE, N. SULTANA, F. A. SIDDIQUI, Pak. J. Pharm. Sci., 18 (2005) 58.
6. E. A. EL-GHANY, A. M. AMINE, A. S. EL-SAYED, M. T. EL-KOLALY, F. ABDEL-GELIL, J. Radioanal. Nucl. Chem., 117 (2005) 266.
7. P. J. FRAKER, J. C. SPECK, Biochem. Biophys. Res. Common., 80 (1978) 849.
8. F. CYNTHIA BAERER, D. ROGER KNAPP, A. J. KAUMANN, L. S. THEODORE, B. LUTZ, J. Mol. Pharmacol., 12 (1979) 328.
9. J. C. SACCAVINI, C. BRUNEAU, IAEA. CN., 4519 (1984) 153.
10. R. EL-SHEIKH, E. A. EL-GHANY, A. S. EL-WETERY, Z. A. SALEH, H. HUSSIEN, Arab. J. Nucl. Sci. Appl., 40 (2007) 36.
11. W. G. WOOD, C. WACHTER, P. C. SCRIBA, Z. FRES, Anal. Chem., 301 (1980) 119.
12. G. PETZOL, H. H. COENEN, J. Labeled Comp. Radiopharm., 18 (1981) 139.
13. K. M. EL-AZONY, Arab J. Nucl. Sci. Appl., 37 (2004) 81.
14. V. I. STANKO, N. G. IROSHIKOVA, J. Gen. Chem. USSR, 49 (1984) 1823.
15. V. I. STANKO, N. G. IROSHIKOVA, A. F. VOLKOV, A. I. KLIMOVA, Int. J. Appl. Radiation Isotopes, 35 (1984) 1129.
16. J. G. HARDMAN, L. E. LIMBIRD, P. B. MOLIINOFF, R. W. RUDDON (Eds), The Pharmacological Basis of Therapeutics, 9th ed., Goodman & Gilman's, 1996.
17. J. SCHMID, U. BUSCH, G. HEINZEL, G. BOZLER, S. KASCHKE, M. KUMMER, Drug Metab Dispos., (1995) 23.