

PRODUCT MONOGRAPH

CERETEC™

Kit For the Preparation of Technetium-99m Exametazime Injection

Reagent for Preparation of a Radiodiagnostic Agent

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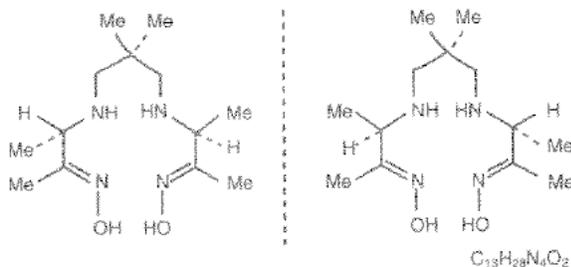
Reagent for Preparation of a Radiodiagnostic Agent

DESCRIPTION

The Ceretec kit is supplied as a five-unit package. Each unit consists of three vials: Ceretec reagent, methylene blue and phosphate buffered saline. These sterile, non-pyrogenic, non-radioactive ingredients are necessary to prepare Technetium-99m Exametazime intravenous injection with methylene blue stabilizer, or to prepare Technetium-99m Exametazime intravenous injection without methylene blue stabilizer.

Each Ceretec multi-dose reagent vial contains a pre-dispensed, sterile, non-pyrogenic, freeze dried mixture of 0.5 mg exametazime, 7.3 µg stannous chloride dihydrate and 4.5 mg sodium chloride. Following freeze-drying, the vial is filled with inert nitrogen atmosphere to a pressure just below atmospheric and is sealed with a rubber closure. The product contains no antimicrobial preservative.

The structural formula of exametazime is:



Prior to publication of the USAN, exametazime was formerly known as hexamethylpropylene amine oxime (HM-PAO). The name HM-PAO appears in many publications. Also known as (RR,SS)-4,8 diaza-3,6,6,9-tetramethyl undecane-2,10-dione bisoxime.

Each vial of sterile, non-pyrogenic Methylene Blue 1% Injection USP contains 10 mg of Methylene Blue USP in 1 ml of Sterile Water for Injection. The pH is adjusted to 3.0-4.5.

Each vial of 0.003M Phosphate Buffered Saline contains Monosodium Phosphate USP and Dibasic Sodium Phosphate USP in 4.5 ml of 0.9% Sodium Chloride Injection. Each milliliter

contains 0.276 mg monobasic sodium phosphate monohydrate, 0.142 mg dibasic sodium phosphate anhydrous and 9 mg sodium chloride, and provides 0.285 mg (3mM) of phosphate, 0.157 mEq of sodium and 0.154 mEq of chloride. The total calculated osmolarity is 317 mOsmol/L.

When used according to the preparation instructions (see Dosage and Administration), Methylene Blue, Sodium Phosphate/Sodium Chloride mixture acts as a stabilizer.

The Tc99m complex of exametazime is covered by Canadian patent 1243329.

Physical Characteristics

Technetium Tc99m decays by isomeric transition with a physical half-life of 6.02 hours⁽¹⁸⁾. Photons that are useful for detection and imaging studies are listed in Table 1.

Table 1. Principal Radiation Emission Data

Radiation	Mean %/ Disintegration	Mean Energy (KeV)
Gamma-2	87.87	140.5

External Radiation

The specific gamma ray constant for Tc99m is $206\mu\text{Ckg}^{-1}/37\text{MBq}\cdot\text{hr}.$ (0.78R/millicurie-hr.) at 1 cm. The first half-value layer is 0.02 cm of Pb. A range of values for the relative attenuation of the radiation emitted by this radionuclide that results from interposition of various thicknesses of Pb is shown in Table 2. For example, the use of a 0.25 cm thickness of Pb will attenuate the radiation emitted by a factor of about 1,000.

Table 2. Radiation Attenuation by Lead Shielding

Shield Thickness (Pb) cm	Coefficient of Attenuation
0.02	0.5
0.08	10^{-1}
0.16	10^{-2}
0.25	10^{-3}
0.33	10^{-4}

To correct for physical decay of this radionuclide, the fractions that remain at selected intervals after time of calibration are shown in Table 3.

Table 3. Physical Decay Chart: Tc99m, Half-Life 6.02 Hours

Hours	Fraction Remaining	Hours	Fraction Remaining
*0	1.000	5	0.562
1	0.891	6	0.501
2	0.794	8	0.398
3	0.708	10	0.316
4	0.631	12	0.251

*Calibration time

CLINICAL PHARMACOLOGY

Regional cerebral blood flow scintigraphy

Conventional radioisotope brain scanning uses polar radiopharmaceuticals such as Tc99m pertechnetate which do not penetrate the normal brain. They image brain pathology by crossing the damaged blood brain barrier but even using single photon emission computed tomography (SPECT) are unable to match the resolution and detailed morphology provided by X-ray computerized tomography (CT).

The development of positron emission tomography (PET) and novel radiopharmaceuticals based on the short-lived positron emitting nuclides ^{11}C , ^{18}F , ^{15}O and ^{13}N made functional imaging of the brain possible and produced spectacular results⁽¹⁾. This led to the search for agents which can cross the blood brain barrier and provide information on regional cerebral blood flow using conventional gamma camera and SPECT imaging.

In order to cross the blood brain barrier a substance must be uncharged, lipophilic and of low molecular weight. However, the ideal tracer must not only cross the blood brain barrier, but remain with a fixed distribution within the brain for a sufficiently long time to allow the acquisition of data for reconstruction of tomographic images. Although Xenon-133 has been widely used to measure regional cerebral blood flow it cannot be imaged without specialized instrumentation as it is rapidly washed out of the brain⁽²⁾.

Though Iodine-123 labeled amines have been shown to cross the blood brain barrier and be retained⁽³⁾, the ideal radiopharmaceutical product is one that complexes with a readily available radionuclide such as technetium-99m. The Tc99m complex of hexamethylpropylene amine

oxime (HM-PAO) is not only taken into the brain but also shows long term retention^(4,5,6). In particular the RR,SS (d,l) diastereoisomer of HM-PAO (exametazime) provides a Tc99m complex which exhibits near-ideal characteristics^(7,8,9).

When pertechnetate is added to the exametazime ligand in the presence of stannous reductant a lipophilic Tc99m complex is formed. This converts with time to a secondary complex which is less lipophilic. This secondary complex does not cross the blood brain barrier. A consequence of the conversion of lipophilic to secondary complex is that the useful life of the reconstituted agent is limited. The *in vitro* addition of methylene blue to the Tc99m exametazime will stabilize the complex for 4-6 hours. Methylene blue may be added to the Tc99m for cerebral imaging. Methylene blue must not be used in the preparation of Tc99m exametazime labeled leukocytes.

Studies in normal volunteers have shown that the Tc99m exametazime complex is rapidly cleared from the blood after intravenous injection. Uptake in the brain reaches a maximum of 3.5-7.0% of the injected dose within one minute of injection. Up to 15% of the cerebral activity washes out of the brain by 2 minutes post injection after which there is little loss of activity for the following 24 hours except by physical decay of Tc99m. The activity not associated with the brain is widely distributed throughout the body particularly in muscle and soft tissue. About 30% of the injected dose is found in the GI tract immediately after injection and about 50% of this is excreted through the gut over 48 hours. About 40% of the injected dose is excreted through the kidneys and urine over the 48 hours after injection resulting in a reduction in general muscle and soft tissue background.

***In Vitro* Tc99m leukocyte labeling**

Leukocytes are involved in a number of the body's responses to disease including infection, inflammation and infarction. Techniques have been developed to tag leukocytes with a radiolabel using In 111, in order to subsequently assess sites of localization and consequently pathology using a gamma camera. In 111 labeled leukocytes are an established noninvasive means of diagnosing a variety of inflammatory conditions in which granulocyte migration is a prominent feature.⁽¹⁰⁻¹³⁾

The superior imaging characteristics of Tc99m have led to a search for a suitable method to label leukocytes with this nuclide. Labeling by means of complexes such as Tc99m oxine, Tc99m pyrophosphate and medronate and the incorporation of Tc99m colloids by phagocytes have been proposed, but all suffer deficiencies either in label stability or in "activation" or damage to leukocytes during the labeling procedure, leading to an unnatural biodistribution on reinjection.⁽¹⁴⁻¹⁵⁾

The small lipophilic nature of the Tc99m exametazime complex facilitates its uptake into leukocytes, following which the Tc99m is selectively retained in neutrophils. Provided the recommended cell separation and labeling procedures are carried out, the Tc99m labeled leukocytes do not appear to suffer significant damage or "activation", as evidenced by their *in*

vivo recovery and lack of lung and liver uptake. Label elution rate is up to 10% in the first hour, declining thereafter.

Following cell separation and radiolabeling, according to the package insert instructions, a labeling efficiency of around 55% may be expected with around 78% of the label associated with the neutrophil population. Studies of elution rates indicate that Tc99m exametazime shows relative selectivity for granulocytes⁽¹⁶⁾ and acts as an effective radiolabeling agent. Following reinjection of the Tc99m labeled leukocytes the functional integrity of the granulocytes appears to be well maintained as the recovery of radiolabeled granulocytes (i.e., the circulatory granulocyte associated activity as a percentage of injected granulocyte associated activity) at 40 minutes after injection gave a mean value of 37%⁽¹⁷⁾ which compares favorably with pure granulocytes labeled with In 111 tropolonate.⁽¹²⁾ The initial biodistribution is similar to that of In 111 tropolonate labeled pure granulocytes. During the first hour following injection of Tc99m labeled leukocytes, activity is seen in the lungs, liver, spleen, blood pool and bone marrow as well as in the bladder. The kidneys (parenchyma and/or renal pelvis) and gallbladder may also be visualized. This pattern of activity continues to be seen at 4 hours post-injection except that lung activity is greatly reduced and faint bowel activity may be visible. At 24 hours post-injection substantial colonic activity is seen, in addition to the normal areas visualized in earlier scans.

TOXICOLOGY

Toxicity studies have been performed on intravenously administered Ceretec in male and female rats and rabbits.

Ceretec without methylene blue stabilizer

No adverse reactions or mortalities were observed at a dose level equivalent to the single injection of 1200 times the maximum human equivalent dose (MHD). Similarly, 14 day repeat-dose studies in rats and rabbits at a cumulative dose of up to 14,000 times the maximum human equivalent dose resulted in no adverse reactions or mortalities. At termination, thorough histopathology, hematology and blood chemistry revealed no abnormalities.

Ceretec with methylene blue stabilizer

Because of the diluting effect of the stabilizing solution, it was not possible to achieve doses as high as those attained using Ceretec alone. However, in single dose studies in male and female rats and rabbits using up to 700 times MHD, there were no mortalities. In the rats, at 350 and 700 times MHD, some evidence of the well-documented toxicity of methylene blue was seen. This included transient lethargy at both doses and decreased respiratory rate, mostly in the high dose group. No dose related effects were observed in rats receiving 100 times MHD. In the rabbits, no treatment-related changes were seen in any of the dosage groups.

INDICATIONS AND CLINICAL USES

Regional cerebral blood flow scintigraphy

Tc99m exametazime intravenous injection is used for regional cerebral blood flow scintigraphy. In stroke, reduced cerebral blood flow appears as photopenic areas on scintigrams. Tc99m exametazime scintigraphy may also be useful in investigations of transient ischemic attack, migraine and tumors of the brain.

In epilepsy, areas of both ictally increased and interictally decreased perfusion have been demonstrated. Characteristic areas of reduced perfusion have been demonstrated in Alzheimer's disease which may provide the basis for differential diagnosis of dementia.

***In Vitro* Tc99m Leukocyte Labeling**

Tc99m Exametazime is an effective agent for *in vitro* Tc99m leukocyte radiolabeling. Tc99m labeled leukocytes are useful in the detection of sites of focal infection, especially abdominal abscess and as an adjunct in the investigation of pyrexia of unknown origin (PUO), and in the evaluation of inflammatory conditions not associated with infection such as inflammatory bowel disease (IBD).

CONTRAINDICATIONS

There are no specific contraindications.

WARNINGS

Care should be taken when handling blood specimens to be labeled using this radiopharmaceutical. Even if the subject has been tested, no method can offer complete assurance that Hepatitis B Virus, Human Immunodeficiency Virus (HIV) or other infectious agents are absent. All human blood samples should be considered potentially infectious. Precautions for handling are as those for handling radioactive materials.

The contents of the Ceretec kit are intended for use in the preparation of Tc99m exametazime injection and are NOT to be directly administered to the patient.

The contents of the kit are not radioactive. However after the sodium pertechnetate Tc99m is added, adequate shielding of the final preparation must be maintained to minimize radiation exposure to occupational workers and patients.

Ideally, examinations using radiopharmaceuticals, especially those elective in nature, of women of childbearing capability should be performed during the first ten days following the onset of menses.

PRECAUTIONS

General

The Tc99m labeling reactions involved depend on maintaining the tin (stannous ion) in the reduced state. Hence, sodium pertechnetate Tc99m containing oxidants should not be employed.

Sodium Chloride Injection, USP must be used as the diluent. Do not use bacteriostatic sodium chloride as a diluent for sodium pertechnetate Tc99m injection because it will increase the oxidation products and adversely affect the biological distribution of Ceretec.

Radiopharmaceuticals should be used only by those medical practitioners who are appropriately qualified in the use of radioactive prescribed substances in or on humans.

As in the use of any other radioactive material, care should be taken to minimize radiation exposure to patients consistent with proper patient management, and to minimize radiation exposure to occupational workers.

The components of the reagent vials are sterile and nonpyrogenic. It is essential that the user follows directions carefully and adheres to strict aseptic technique.

The possibility exists that, following the administration of Tc99m exametazime injection with methylene blue stabilizer, urine discoloration may occur.

Carcinogenesis, Mutagenesis and Impairment of Fertility

Since adequate reproduction studies have not been performed in animals to determine whether this drug affects fertility in males and females, has teratogenic potential, or has other adverse effects on the fetus, this radiopharmaceutical preparation should not be administered to pregnant or nursing women unless it is considered that the benefits to be gained outweigh the potential hazards.

Nursing

Where an assessment of the risk/benefits ratio suggests use of this product in lactating mothers, nursing should be stopped.

Pediatric Use

Adequate studies do not exist to support the use in children. As in pregnancy and lactating mothers, the benefits to risk ratio should be assessed before consideration is given to the use of this product in this age group.

Use in Pregnancy

In women of childbearing age the possibility of pregnancy should always be taken into account. It would be prudent to treat as pregnant any woman of reproductive age presenting for a nuclear medicine examination at a time when a menstrual period is overdue or missed, unless there is information that precludes pregnancy. If the menstrual cycle is irregular, a pregnancy test may be indicated before proceeding.

ADVERSE REACTIONS

A very few cases of mild hypersensitivity evidenced by the development of an urticarial erythematous rash have been reported, following I.V. injection of the reconstituted product.

A very few reports have also been received of hypersensitivity reactions, possibly anaphylactic in nature, following administration of Tc99m labeled leukocytes prepared using Tc99m exametazime. It should also be noted that materials used in cell separation may cause hypersensitivity reactions. It is essential that cells are washed free of sedimentation agents before they are reinjected into the patient.

In case of side effects following administration of radiopharmaceuticals, users should ensure the availability of appropriate medical treatment at the time of administration of any radiopharmaceutical to the patient. Users are requested to report to GE Healthcare Canada Inc. any instances of suspected adverse drug reactions or side effects associated with the use of this product.

DOSAGE AND ADMINISTRATION

Regional cerebral blood flow scintigraphy

Shielding should be used at all times when handling both vial and syringes. The 0.45 µm syringe filter must be attached prior to injection of Ceretec with methylene blue stabilizer. Please refer to Cautionary Notes for injection procedure.

The normal adult (70kg) dose is 350-500 MBq (9.5-13.5mCi) by intravenous injection.

Brain imaging may commence from 2 minutes after injection.

Although gross abnormalities of regional cerebral blood flow may be visualized by planar imaging it is strongly recommended that SPECT imaging be carried out to maximize the value of the study.

***In Vivo* localization of Tc99m labeled leukocytes**

The normal adult (70kg) dose is 200MBq (5.4mCi) as Tc99m labeled leukocytes by intravenous injection. Administer the Tc99m labeled leukocyte suspension using a 19G needle as soon as possible after labeling. Dynamic imaging may be performed for the first 60 minutes after injection to assess lung clearance and to visualize immediate cell migration.

Static imaging at 0.5-1.5 hours, 2-4 hours and if necessary, at 18-24 hours post injection should be performed to detect focal accumulation of activity. Care should be taken to distinguish between leukocyte localization and normal biodistribution.

INSTRUCTIONS FOR PREPARATION

Procedure for the Preparation of Tc99m Exametazime Injection with Methylene Blue Stabilizer for Intravenous Injection

(Use aseptic technique and wear waterproof gloves throughout entire procedure)

NOTE: DO NOT USE THIS PROCEDURE FOR LEUKOCYTE LABELING. SEE PROCEDURE FOR THE PREPARATION OF Tc99m EXAMETAZIME INJECTION WITHOUT METHYLENE BLUE STABILIZER.

- 1) Withdraw 0.5 mL Methylene Blue Injection USP 1% into a sterile syringe and inject into 4.5 mL vial of 0.003 M Monobasic Sodium Phosphate USP and Dibasic Sodium Phosphate USP in 0.9% Sodium Chloride Injection USP. Gently swirl and withdraw 2 mL of Methylene Blue/Phosphate Buffer mixture into a syringe. This mixture must be used within 30 minutes of preparation.
- 2) Reconstitute Ceretec with Technetium Tc99m sodium pertechnetate according to the preparation procedure for Preparation of Tc99m Exametazime Injection; 0.37 GBq to 2.0 GBq (10 mCi to 54 mCi) technetium Tc99m may be added to the vial.
 - a) Place one of the vials in a suitable shielding container and sanitize the rubber septum with an isopropyl alcohol swab.
 - b) Using a 10 mL syringe, inject into the shielded vial 5mL of sterile additive-free eluate from a Tc99m generator (see Cautionary Notes 1 -5). Before withdrawing the syringe from the vial withdraw 5 mL of gas from

the space above the solution to normalize the pressure in the vial. Gently swirl the shielded vial for 10 seconds to ensure complete dissolution of the powder.

- c) Immediately proceed to Step 3.
- 3) Add stabilizing solution from Step 1 to the reconstituted Ceretec vial within 2 minutes of reconstitution.
- 4) Determine the radiochemical purity of the solution (see *Radiochemical Purity Measurement* section). A radiochemical purity greater than 80% is necessary for product acceptance.
- 5) Maintain the reconstituted product at 20-25°C, 68-77 °F. Visually inspect the vial for particulate matter prior to injection.
- 6) Maintain adequate shielding of the radioactive preparation.
- 7) The injection may be used for up to 4 hours in cerebral scintigraphy studies.
- 8) Prior to patient injection, the enclosed filter **must** be attached to the injection syringe. Please refer to Cautionary Note #7.
- 9) The pH of the prepared injection is 6.5-7.5.
- 10) The patient dose should be measured in a suitable radioactivity calibration system immediately prior to administration.

Cautionary Notes

- 1) 0.37-2.0 GBq (10 - 54 mCi) technetium Tc99m sodium pertechnetate may be added to the vial. Before reconstitution the technetium Tc99m generator eluate may be adjusted to the correct radioactive concentration to a volume of 5 mL by dilution with preservative-free, non-bacteriostatic saline for injection.
- 2) When using exametazime with the methylene blue stabilizer, generator eluate more than 30 minutes old should not be used. For radiolabeling of non-stabilized exametazime, generator eluate more than 2 hours old should not be used.
- 3) Use only eluate from a technetium Tc99m generator which was previously eluted within 24 hours. For the highest radiochemical purity, reconstitute with freshly eluted Tc99m generator eluate.
- 4) 5 mL of eluate is necessary due to the limited solubility of exametazime.

- 5) Oxidant-free Tc99m eluate must be used due to the minimal amount of stannous ion present in the product.
- 6) Radiochemical purity testing must be performed prior to patient administration. A radiochemical purity greater than 80% is necessary for product acceptance.
- 7) Prior to patient injection, the enclosed filter must be attached to the syringe. Secure the filter in place by attaching it to the luer lock syringe containing the stabilized Tc99m Exametazime, (then attach a needle if necessary).

Procedure for Preparation of Tc99m Exametazime Injection without Methylene Blue Stabilizer for Intravenous Injection or *In Vitro* Leukocyte Labeling

(Use aseptic technique and wear waterproof gloves throughout the entire procedure).

- 1) Place one of the vials in a suitable shielding container and sanitize the rubber septum with an isopropyl alcohol swab.
- 2) Using a 10mL syringe, inject into the shielded vial 5mL of sterile additive-free eluate from a Tc99m generator (see cautionary notes 1-5). Before withdrawing the syringe from the vial withdraw 5 mL of gas from the space above the solution to normalize the pressure in the vial. Gently swirl the shielded vial for 10 seconds to ensure complete dissolution of the powder.
- 3) Assay the total activity and calculate the volume to be injected. The patient dose should be measured in a suitable radioactivity calibration system immediately prior to administration.
- 4) Complete the label provided and attach to the vial shield. The technetium Tc99m exametazime injection is ready for quality control.
- 5) Maintain adequate shielding of the radioactive preparation.
- 6) Use within a maximum of **30 minutes** after reconstitution. Discard any unused material in accordance with Canadian radioactive waste regulations.
- 7) Visually inspect the reconstituted material and do not use if there is evidence of foreign matter.
- 8) The injection may be prepared for use in cerebral scintigraphy or in the preparation of Tc99m labeled leukocytes.
- 9) The pH of the prepared injection is 9.0 - 9.8.

- 10) The patient dose should be measured in a suitable radioactivity calibration system immediately prior to administration.

Cautionary Notes

- 1) 0.37 - 2.0 GBq (10 - 54 mCi) technetium Tc99m sodium pertechnetate may be added to the vial. Before reconstitution the technetium Tc99m generator eluate may be adjusted to the correct radioactive concentration to a volume of 5 mL by dilution with preservative-free, non-bacteriostatic saline for injection.
- 2) When using exametazime with the methylene blue stabilizer, generator eluate more than 30 minutes old should not be used. For radiolabeling of non-stabilized exametazime, generator eluate more than 2 hours old should not be used.
- 3) Use only eluate from a technetium Tc99m generator which was previously eluted within 24 hours. For the highest radiochemical purity, reconstitute with freshly eluted Tc99m generator eluate.
- 4) 5 mL of eluate is necessary due to the limited solubility of exametazime.
- 5) Oxidant-free Tc99m eluate must be used due to the minimal amount of stannous ion present in the product.
- 6) Radiochemical purity testing must be performed prior to patient administration. A radiochemical purity greater than 80% is necessary for product acceptance.

Procedure for Separation of Leukocytes and Subsequent *In Vitro* Labeling with Tc99m Exametazime without Methylene Blue Stabilizer

(Use aseptic technique throughout).

- i) Draw 9mL of acid-citrate-dextrose^(a) into each of two 60 mL plastic non-heparinized syringes.
- ii) Withdraw 51 mL of patient's blood into each syringe using a 19G Butterfly needle infusion set. Close the syringes with sterile hubs.
- iii) Dispense 2mL of sedimentation agent^(b) into each of 5 Universal containers or tubes.
- iv) Without attaching a needle to the syringes dispense 20mL of blood into each of the 5 Universal tubes containing sedimentation agent. Dispense the remaining 20 mL of blood into a tube without sedimentation agent.

TIP *To avoid bubbles and frothing run the blood **gently** down the sides of the tubes.*

- v) Mix the blood and sedimentation agent with one gentle inversion. Remove the cap of the Universal tube and burst the bubble formed at the top using a sterile needle. Replace the cap and allow the tubes to stand for 30-60 minutes for erythrocyte sedimentation to take place.

TIP *The period of time for erythrocyte sedimentation depends on the patient's condition. As a guideline it should be stopped when the blood has sedimented to give approximately half the volume as sedimented red cells.*

- vi) Meanwhile centrifuge the tube containing 20mL of blood and no sedimentation agent at 2000 G for 10 minutes. This will yield supernatant cell-free plasma (CFP) containing ACD which is retained, at room temperature, for use as a cell labeling and reinjection medium.
- vii) When sufficient red cell sedimentation has taken place (see v), carefully transfer 15mL aliquots of the cloudy straw-colored supernatant into clean Universal tubes. Take care to avoid withdrawing any sedimented erythrocytes. The supernatant is leukocyte-rich, platelet-rich plasma (LRPRP).

TIP *Do not use needles on sampling syringes to avoid unnecessary cell damage.*

- viii) Centrifuge the LRPRP at 150 G for 5 minutes to give supernatant, platelet-rich plasma (PRP) and a pellet of “mixed” leukocytes.
- ix) Remove as much of the PRP as possible into clean Universal tubes and further centrifuge at 2000 G for 10 minutes to give more supernatant, cell-free plasma (CFP) containing sedimentation agent. This will be used to wash the cells after labeling.
- x) Meanwhile, loosen the pellets of “mixed” leukocytes by **very gently** tapping and swirling the Universal tubes. Using a syringe, without an attached needle, pool all the cells into one tube, then, using the same syringe, add 1 mL of cell-free plasma containing ACD (from vi) and **gently** swirl to resuspend.
- xi) Reconstitute a vial of Ceretec® with 5mL of Tc99m generator eluate containing approximately 200MBq (5.4mCi) of Tc99m sodium pertechnetate (using the procedure described above).
- xii) Immediately following reconstitution add 4mL of the resulting Tc99m exametazime solution to the “mixed” leukocytes in CFP (from x).
- xiii) **Gently** swirl to mix and incubate for 10 minutes at room temperature.

- xiv) On completion of incubation **carefully** add 10mL of CFP containing sedimentation agent (from ix) to the cells, in order to stop labeling. Gently invert the cells to mix.
- xv) Centrifuge at 150 G for 5 minutes.
- xvi) Remove and retain all of the supernatant.
TIP *It is critical that all the supernatant which contains unbound Tc99m exametazime is removed at this stage. This can be best achieved using a syringe with a wide-bore(19G) needle.*
- xvii) Gently resuspend the Tc99m labeled mixed leukocyte preparation in 5-10mL of CFP containing ACD from (vi). Gently swirl to mix.
- xviii) Measure the radioactivity in the cells and in the supernatant from (xvi). Calculate the labeling efficiency (LE) which is defined as the activity in the cells as a percentage of the sum of the activity in the cells and the activity in the supernatant.
TIP *Labeling efficiency depends on the patient's leukocyte count and will vary according to the volume of the initial blood sample. Using the volumes in (ii), a LE of about 55% might be expected.*
- xix) Without attaching a needle, carefully draw up the labeled cells into a plastic, non-heparinized syringe and close it with a sterile hub. Measure the radioactivity.
- xx) Labeled cells are now ready for reinjection. This should be performed without delay.
- xxi) The patient dose should be measured in a suitable radioactivity calibration system immediately prior to administration.

NOTE:

- (a) Acid-citrate-dextrose (ACD) - commercial preparations are available.
- (b) 6% hydroxyethyl starch is recommended. Alternatively, a sterile preparation of 2% methyl cellulose in 0.9% saline may be used.

Radiochemical Purity Measurement

Three potential radiochemical impurities may be present in the prepared injection of the lipophilic complex Tc99m exametazime. These are secondary Tc99m exametazime

complex, free pertechnetate and reduced hydrolyzed Tc99m. A combination of 3 chromatographic systems is necessary for the complete definition of the radiochemical composition of the injection.

Test samples are applied by needle approximately 2.5 cm from the bottom of two GMCP - SA strips (2 cm (\pm 2 mm) x 20cm) and one Whatman No. 1 strip (2.5cm x 30cm) and then immediately placed in prepared ascending chromatography development tanks containing fresh solvent (1cm depth). The two GMCP-SA strips are run in butanone and 0.9% aqueous sodium chloride respectively and the Whatman No. 1 in 50% aqueous acetonitrile. After a 14 cm elution the strips are removed, solvent fronts marked, dried and the distribution of activity determined using suitable equipment.

Interpretation of Chromatograms

System 1 (GMCP-SA: butan-2-one(MEK))

Secondary Tc99m exametazime and reduced hydrolyzed-Tc99m remain at the origin.

Lipophilic Tc99m exametazime complex and Tc99m pertechnetate migrate at Rf 0.8-1.0

System 2 (GMCP-SA: 0.9% sodium chloride)

Lipophilic Tc99m exametazime complex, secondary Tc99m exametazime complex and reduced-hydrolyzed-Tc99m remain at the origin. Tc99m pertechnetate migrates at Rf 0.8-1.0

System 3

(Whatman No. 1: 50% aqueous acetonitrile)

Reduced-hydrolyzed-Tc99m remains at the origin.

Lipophilic Tc99m exametazime complex, secondary Tc99m exametazime complex and Tc99m pertechnetate migrate at Rf 0.8-1.0

- i) Calculate the percentage of activity due to both secondary Tc99m exametazime complex and reduced-hydrolyzed-Tc99m from System 1 (A% + C%). Calculate the percentage of activity due to Tc99m pertechnetate from System 2 (B%). Calculate the percentage of activity due to the reduced-hydrolyzed-Tc99m from System 3 (C%).
- ii) The radiochemical purity (as percentage lipophilic Tc99m exametazime complex) is given by: $100 - (A\% + B\% + C\%)$ where:
A% represents the level of secondary Tc99m exametazime complex.
B% represents the level of Tc99m pertechnetate.
C% represents the level of reduced-hydrolyzed-Tc99m.

A radiochemical purity of at least 80% may be expected provided the measurement has been carried out within 30 minutes of reconstitution for Ceretec without methylene blue stabilizer and within 4 hours for Ceretec with methylene blue stabilizer.

RADIATION DOSIMETRY

(1) Brain Scintigraphy

Based on human data, the absorbed radiation doses to an average human adult (70 kg) from an intravenous injection of this product are estimated in Table 4.

Table 4. Estimated Absorbed Radiation Dose* for Cerebral Scintigraphy

Absorbed radiation dose Tc99m exametazime injection

Target Organ	mGy/MBq	rads/mCi	mGy/740MBq	rads/20mCi
Adrenals	0.0053	0.020	3.92	0.392
Bladder	0.023	0.085	17.02	1.702
Bone surfaces	0.0051	0.019	3.77	0.377
Brain	0.0068	0.025	5.03	0.503
Breast	0.002	0.007	1.48	0.148
Gall-bladder	0.018	0.067	13.32	1.332
GI tract				
Stomach	0.0064	0.024	4.74	0.474
Small Intestine	0.012	0.044	8.88	0.888
Upper Large Intestine	0.018	0.067	13.32	1.332
Lower Large Intestine	0.015	0.056	11.10	1.110
Heart	0.0037	0.014	2.74	0.274
Kidneys	0.034	0.126	25.16	2.516
Liver	0.0086	0.032	6.36	0.636
Lungs	0.011	0.041	8.14	0.814
Muscles	0.0028	0.010	2.07	0.207
Esophagus	0.0026	0.010	1.92	0.192
Ovaries	0.0066	0.024	4.88	0.488
Pancreas	0.0051	0.019	3.77	0.377
Red marrow	0.0034	0.013	2.52	0.252
Skin	0.0016	0.006	1.18	0.118
Spleen	0.0043	0.016	3.18	0.318
Testes	0.0024	0.009	1.78	0.178
Thymus	0.0026	0.010	1.92	0.192
Thyroid	0.026	0.096	19.24	1.924
Uterus	0.0066	0.024	4.88	0.488
Remaining organs	0.0032	0.012	2.37	0.237

*International Commission on Radiological Protection, "Radiological Protection in Biomedical Research," ICRP 62,1993.

Effective Dose Equivalent (EDE) 7.8 mSv/740 MBq (International Commission on Radiological Protection, "Radiological Protection in Biomedical Research," ICRP 62,1993).

2) *In vivo* localization of Tc99m labeled leukocytes.

The estimated absorbed radiation doses to various organs following the intravenous administration of Tc99m labeled leukocytes given by ICRP 53** are as follows (bladder voiding every 3.5 hours)

Target Organ	Absorbed Radiation Dose	
	(mGy per 200MBq)	(rads/25mCi)
Spleen	30	13.89
Red marrow	4.4	2.04
Liver	4	1.85
Pancreas	2.8	1.3
Ovaries	0.84	0.39
Testes	0.34	0.16
Uterus	0.76	0.35

**International Commission on Radiological Protection, "Radiation Dose to Patients from Radiopharmaceuticals", ICRP 53, 1987.

Effective Dose Equivalent (EDE) 3.4 mSv/200MBq (International Commission on Radiological Protection, "Radiological Protection in Biomedical Research," ICRP 62, 1993).

HOW SUPPLIED

Each Ceretec Kit is supplied as a five-unit package containing:

5 multiple dose Ceretec vials containing a freeze-dried sterile, pyrogen-free mixture of exametazime, stannous chloride dihydrate and sodium chloride sealed under an inert nitrogen atmosphere.

5 vials of Methylene Blue Injection USP 1%.

5 vials of 0.003 M Monobasic Sodium Phosphate USP and Dibasic Sodium Phosphate USP in 0.9% Sodium Chloride Injection USP.

15 syringe filters (0.45µm low-binding Durapore® polyvinylidene difluoride [PVDF] membrane in a polypropylene housing)

5 labels for the reconstituted injection.

1 package insert.

Durapore® is a trademark of Millipore Corporation.

STORAGE

Store the kit at any temperature in the range of 15-25 °C, 59-77°F.

Store the reconstituted injection at 20-25 °C, 68-77 °F using appropriate radiation shielding.

EXPIRY

Use Tc99m exametazime injection with methylene blue stabilizer within 4 hours after reconstitution. Use Tc99m exametazime injection without methylene blue stabilizer within 30 minutes after reconstitution. Protect from freezing. Refer to carton for expiration date of kit.

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