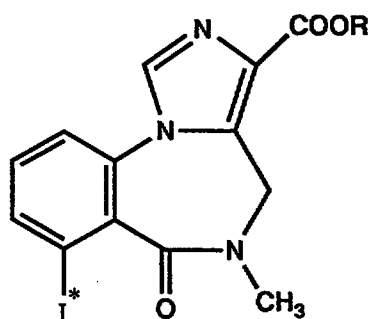


Description

The present invention is concerned with radioiodinated benzodiazepine derivatives of the formula



I

wherein I* signifies a radioactive iodine and R signifies an alkyl group with 1-4 C-atoms, and their use in the diagnosis of diseases and disorders of the brain through their ability to image the brain.

The radioactive iodine is preferably iodine 123. The alkyl group R is preferably methyl, ethyl, isopropyl, sec.butyl or tert.butyl, most preferably ethyl.

The most preferred compound in accordance with the present invention is ethyl 7-¹²³iodo-5,6-dihydro-5-methyl-6-oxo-4H-imidazo[1,5-a][1,4]benzodiazepine-3-carboxylate.

The compounds of formula I are useful for imaging the brain. Accordingly, the compounds of formula I show a rapid accumulation in the brain, which indicates that they have the ability to penetrate the so-called blood/brain barrier. The compounds of formula I show a rapid localization of the radioactivity in the brain after intravenous administration.

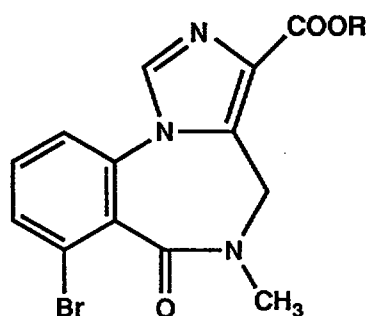
The compounds of formula I can be used for the diagnosis of diseases or disorders of the brain by their capacity to image changes in the distribution of the benzodiazepine receptors in the brain. In this manner the compounds of formula I can be used for the diagnosis of brain diseases or disorders such as cerebrovascular diseases (e.g. strokes), neurological disorders (e.g. epilepsy) and psychotic diseases.

The compounds of formula I can be prepared according to methods known per se. Thus, e.g. a compound of formula I can be prepared from a cold compound of formula I, i.e. a compound in

which I signifies a stable iodine, by exchange with a radioactive iodine, preferably iodine 123.

Iodine 123 in a 0.1N sodium hydroxide solution can be used for the exchange of cold iodine by hot iodine. This solution is heated with a solution of the cold compound of formula I for approximately a quarter of an hour to approximately two hours. This iodine exchange is carried out in the presence of a solvent such as e.g. glacial acetic acid.

A further and preferred method for the preparation of the compounds of formula I comprises reacting a compound of formula II



II

with a radioactive iodine.

Most preferably, iodine 123 is used.

The same reaction conditions are used as in the case of the exchange of the non-radioactive iodine by iodine 123.

The cold compounds of formula I and the compounds of formula II are either known from European Patent Publications Nos. 27214 or 59389 or from U.S. Patent No. 4,316,839. These cold compounds can be prepared according to the methods which are described in the aforementioned European Patent Publications and in the aforementioned U.S. Patent.

As mentioned above, the radioiodinated compounds in accordance with the present invention localize rapidly in the brain after intravenous administration. In almost all cases a sufficient amount of the administered dose will accumulate in the brain within 2 to 10 min., which permits the taking of scintiphotos. The compounds in accordance with the present invention show sufficient presence in the brain for at least 60 min., so that significant studies may be carried out.

The radioiodinated compounds in accordance with the present invention can be administered in an aqueous or aqueous/alcoholic medium. Such media can also contain conventional pharmaceutical adjuvants such as e.g. pharmaceutically acceptable salts to adjust the osmotic pressure, buffers, preservatives and the like.

A preferred carrier for parenteral administration of the compounds of formula I is a normal saline solution which contains from approximately 0.5% by weight to approximately 2% by weight of a suitable preservative.

The radioactive benzodiazepines in accordance with the present invention can be injected intravenously into a patient for the diagnostic imaging of the brain. In accordance with the present invention, the radioactive benzodiazepine of formula [I] is administered in an injectable unit dose. Any conventional carrier such as sterile saline, plasma etc. can be used for the preparation of the injectable solutions which are usable for the diagnostic imaging in accordance with the present invention. In general, the unit dose to be administered contains a radioactivity of approximately 2 mCi to approximately 10 mCi, preferably approximately 4 to 5 mCi. However, any amount of compound in accordance with the present invention which is effective for imaging the brain can be injected in accordance with the present invention. The solution to be injected is preferably a unit dose of approximately 0.1 ml to approximately 10 ml, especially of approximately 1 to 5 ml and particularly of 4 to 5 ml. After intravenous administration, the radioactive benzodiazepine of formula I will image the organs in vivo. Any conventional method of visualizing or imaging for diagnostic purposed can be used in accordance with the present invention. In this connection, scintigraphic methods for visualizing or imaging the brain can be used.

In accordance with the present invention, the compound of formula I and particularly ethyl 7-¹²³I-5,6-dihydro-5-methyl-6-oxo-4H-imidazo[1,5-a][1,4]benzodiazepine-3-carboxylate is administered to humans intravenously in normal saline solution containing 5% glucose. The dose injected into humans contains 4 to 5 mCi of the radioactive compound. Within 40 to 60 min. after injection, scintiphotos are taken for 25 min. with a Gamma Spect camera. In accordance with the present invention the compound is preferably injected at a dose of 0.0543 mCi per kilogram.

The compound of formula I can be administered as the free base or as a pharmaceutically acceptable acid addition salt thereof.

The following Examples illustrate the invention. Unless indicated otherwise, all temperature are given in degrees Celsius.

Example 1

The labelling procedure for the halogen exchange (bromide-iodide) is carried out in a conical reaction vessel which is tightly closed by a Teflon-laminated silicon septum. Iodine 123 activity (up to 300 mCi) in 0.1N NaOH is evaporated to dryness by means of a gentle stream of nitrogen at 90°. Thereupon, 1 mg of ethyl 7-bromo-5,6-dihydro-5-methyl-6-oxo-4H-imidazo[1,5-a][1,4]-benzodiazepine-3-carboxylate dissolved in 200 µl of glacial acetic acid is added, whereupon the reaction mixture is heated at 150° for 1 hr. After cooling, the mixture is dissolved in 5 ml of water and purified by HPLC. The purification was effected according to methods known per se. Labelling and purification were carried out in a suitable lead box container. The labelling was virtually quantitative.

Example 2

During the HPLC separation, the product of the main peak was collected and subsequently evaporated to dryness. The residue was dissolved in a solution containing 5% glucose and passed through a silver powder column in order to adsorb iodine liberated during the evaporation. After sterile filtration and adjustment of the activity concentration to 1 mCi/ml, the ethyl 7-¹²³iodo-5,6-dihydro-5-methyl-6-oxo-4H-imidazo[1,5-a][1,4]benzodiazepine-3-carboxylate was ready for use. The product quality was monitored by thin-layer chromatography on silica gel, developed with ethyl acetate/ammonium sulphate 200:1.

Example 3

The starting material used in Example 1 was prepared as follows:

A mixture of 167.95 g (624 mmol) of 6-bromo-3,4-dihydro-4-methyl-2H-1,4-benzodiazepine-2,5(1H)-dione, 600 ml of N,N-dimethyl-p-toluidine and 800 ml of chloroform (filtered over aluminium oxide) is treated dropwise at boiling temper-

ature with 160.80 g (1.05 mol) of phosphorus oxychloride and boiled under reflux for 4 hours. The solution obtained is poured into a pre-cooled mixture of 500 g of sodium bicarbonate and 2 l of water and stirred for 40 minutes. The inorganic phase is separated and extracted three times with chloroform. The combined chloroform extracts are dried over magnesium sulphate and the chloroform is evaporated.

In the meanwhile, a solution of 76 g (677 mmol) of potassium t-butyrate in 200 ml of dimethylformamide is cooled to -45° , treated firstly with 71 g (625 mmol) of ethyl isocyanacetate and subsequently dropwise at -50° to -20° with the above imine chloride solution. The cooling is removed, the mixture is stirred for 1.5 hours, treated with 13 ml of acetic acid, poured into 1800 ml of water and extracted five times with 500 ml of methylene chloride each time. The combined organic extracts are washed three times with water, dried over magnesium sulphate and evaporated. The crude product is recrystallized from methylene chloride and ethyl acetate and yields 156.80 g of ethyl 7-bromo-5,6-dihydro-5-methyl-6-oxo-4H-imidazo[1,5-a][1,4]-benzodiazepine-3-carboxylate of melting point $214-215^{\circ}$ (69% of theory).

Example 4

Iodide-iodide exchange labelling was carried out in exactly the same manner as the bromide-iodide exchange, but ethyl 7-iodo-5,6-dihydro-5-methyl-6-oxo-4H-imidazo[1,5-a][1,4]benzodiazepine-3-carboxylate was used as the starting material. This labelling was carried out to assure the identity of the labelled product in HPLC and thin layer chromatography.

Example 5

The starting material used in Example 4 was prepared as follows:

A mixture of 185.5 g (586.8 mmol) of 3,4-dihydro-6-iodo-4-methyl-2H-1,4-benzodiazepine-2,5(1H)-dione, 670 ml of N,N-dimethyl-p-toluidine and 800 ml of chloroform (filtered over aluminium oxide) is treated dropwise at boiling temperature with 91.6 ml (979.9 mmol) of phosphorus oxychloride and boiled under reflux for 2 hours. The solution obtained is poured into a pre-cooled mixture of 490 g of sodium carbonate and 2 l of water and stirred for 40 minutes. The inorganic phase is separated and extracted three times with methylene chloride. The combined

methylene chloride extracts are dried over magnesium sulphate and the methylene chloride is evaporated. In the meanwhile, a solution of 76.5 g (626.6 mmol) of potassium t-butyrate in 500 ml of dimethylformamide is cooled to -50° , treated firstly with 65.2 ml (586.8 mmol) of ethyl isocynoacetate and subsequently dropwise at -50° to -15° with the above imine chloride solution. The cooling is removed, the mixture is stirred for 1 hour, treated with 20 ml of acetic acid, poured into 1900 ml of water and extracted five times with methylene chloride. The combined organic extracts are washed three times with water, dried over magnesium sulphate and evaporated. After chromatography of the residue on silica gel and recrystallization from ethyl acetate there are obtained 96.02 g of ethyl 5,6-dihydro-7-iodo-5-methyl-6-oxo-4H-imidazo[1,5-a][1,4]benzodiazepine-3-carboxylate of melting point $244-246^{\circ}$ (39.8% of theory).

Example 6

3 rats (female, Wistar, spf) are measured at seven different points in time (2', 10', 20', 40', 1h, 6h, 15h). The weight of the animals lay between 113 and 144 g. The feeding was effected ad libitum. The i.v. injected doses varied from 184 to 355 μ Ci in each 0.2 ml.

Percentage of the injected activity per gram of the corresponding organ (in the rat)

	2'	10'	20'	40'	60'	6h	15h
Blood	0.70	0.87	0.46	0.17	0.11	0.04	0.013
Brain	2.12	3.22	2.99	2.70	1.80	0.05	0.001
Thyroid gland	0.60	0.54	1.22	0.08	0.06	0.03	0.007
Liver	2.38	5.66	2.62	0.58	0.19	0.02	0.016
Spleen	0.60	0.53	0.29	0.14	0.07	0.02	0.010
Kidneys	2.68	7.06	5.26	1.35	0.68	0.03	0.010
Stomach	0.19	0.41	0.35	0.27	0.48	0.73	0.080
Bowel	0.75	1.10	1.73	2.02	1.97	1.55	0.143
Lung	1.22	0.87	0.44	0.15	0.10	0.03	0.011
Heart	0.86	0.66	0.30	0.10	0.06	0.02	0.007
Ovary	0.88	0.73	0.53	0.22	0.16	0.03	0.011
Femur	0.54	0.45	0.24	0.10	0.07	0.02	0.007
Bladder	0.59	1.07	0.69	0.24	0.42	0.57	0.023
Muscle	0.68	0.42	0.20	0.09	0.04	0.01	0.003

Percentage of the injected activity in the corresponding organs
(in the rat)

	2'	10'	20'	40'	60'	6h	15h
Brain	3.11	5.38	4.20	4.02	2.42	0.08	0.0015
Thyroid gland	0.18	0.15	0.18	0.03	0.02	0.008	0.0015
Liver	16.90	34.37	17.61	3.90	1.29	0.15	0.107
Spleen	0.26	0.21	0.11	0.06	0.03	0.008	0.0035
Kidneys	3.15	7.48	6.25	1.53	0.79	0.04	0.009
Stomach	0.81	1.20	1.28	1.37	1.96	1.35	0.346
Bowel	9.28	12.70	22.53	25.54	28.48	19.50	1.012
Lung	1.04	0.81	0.40	0.15	0.09	0.03	0.0095
Heart	0.58	0.37	0.17	0.06	0.03	0.009	0.0035
Ovary	0.09	0.06	0.05	0.02	0.02	0.003	0.0008
Femur	0.39	0.25	0.14	0.06	0.04	0.01	0.004
Bladder	0.03	0.06	0.03	0.01	0.03	0.09	0.0022
Remaining body	63.94	29.92	25.92	11.04	7.09	2.88	1.30
Total body	99.75	92.96	78.87	47.80	43.17	24.16	2.80
Faeces + urine	0.05	6.17	19.39	48.72	51.71	48.81	42.69

Claims

1. A radioiodinated benzodiazepine derivative of the formula

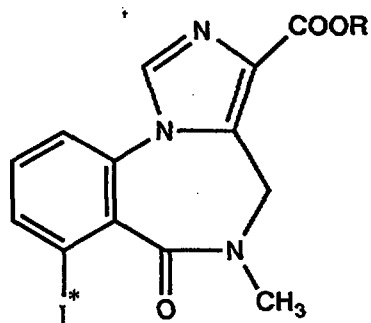


Percentage of the injected activity in the corresponding organs
(in the rat)

	2'	10'	20'	40'	60'	6h	15h
Brain	3.11	5.38	4.20	4.02	2.42	0.08	0.0015
Thyroid gland	0.18	0.15	0.18	0.03	0.02	0.008	0.0015
Liver	16.90	34.37	17.61	3.90	1.29	0.15	0.107
Spleen	0.26	0.21	0.11	0.06	0.03	0.008	0.0035
Kidneys	3.15	7.48	6.25	1.53	0.79	0.04	0.009
Stomach	0.81	1.20	1.28	1.37	1.96	1.35	0.346
Bowel	9.28	12.70	22.53	25.54	28.48	19.50	1.012
Lung	1.04	0.81	0.40	0.15	0.09	0.03	0.0095
Heart	0.58	0.37	0.17	0.06	0.03	0.009	0.0035
Ovary	0.09	0.06	0.05	0.02	0.02	0.003	0.0008
Femur	0.39	0.25	0.14	0.06	0.04	0.01	0.004
Bladder	0.03	0.06	0.03	0.01	0.03	0.09	0.0022
Remaining body	63.94	29.92	25.92	11.04	7.09	2.88	1.30
Total body	99.75	92.96	78.87	47.80	43.17	24.16	2.80
Faeces + urine	0.05	6.17	19.39	48.72	51.71	48.81	42.69

Claims

1. A radioiodinated benzodiazepine derivative of the formula



I

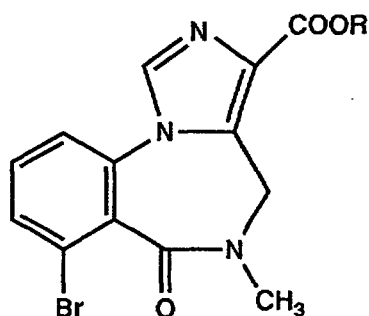
wherein R represents lower alkyl with 1-4 C-atoms and I* represents a radioactive iodine.

2. A compound in accordance with claim 1, characterized in that the radioactive iodine is iodine 123.

3. Ethyl 7-¹²³iodo-5,6-dihydro-5-methyl-6-oxo-4H-imidazo[1,5-a][1,4]benzodiazepine-3-carboxylate.

4. A process for the preparation of a compound of formula I of claim 1, characterized by either

- a) labelling a compound corresponding to formula I, but having a stable non-radioactive isotope of iodine, with a radioactive iodine, or
- b) reacting a compound of the formula

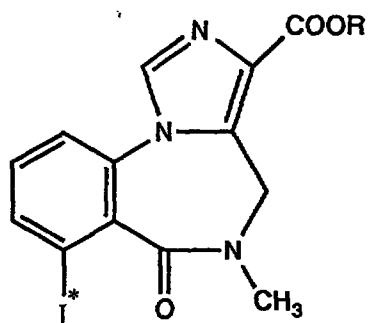


II

with a radioactive iodine.

5. A process according to claim 4, characterized in that iodine 123 is used as the radioactive isotope of iodine.

6. A composition for imaging the brain containing a compound of the formula



I

wherein I* signifies a radioactive iodine and R signifies an alkyl group with 1-4 C-atoms, and a suitable carrier for injection purposes.

7. A composition in accordance with claim 6, characterized in that the compound is ethyl 7-¹²³iodo-5,6-dihydro-5-methyl-6-oxo-imidazo[1,5-a][1,4]benzodiazepine-3-carboxylate.

8. A composition in accordance with claim 6, containing approximately 2 to approximately 10 mCi of the compound of formula I.