^{99m}Tc-[Cys^{3,4,10},D-Phe^{7},Arg^{11}]α-MSH$_{3-13}$

^{99m}Tc-(Arg^{11})CCMSH

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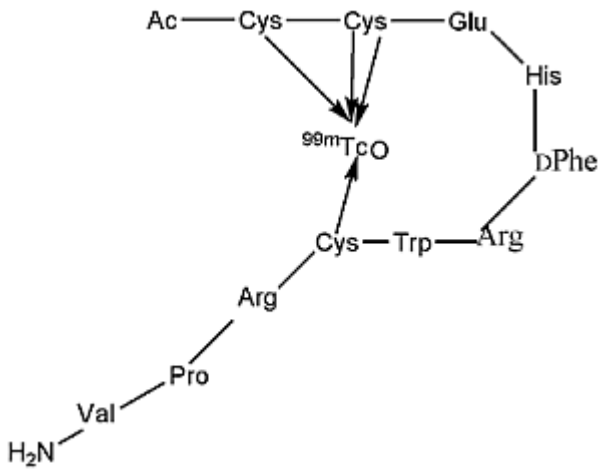
Chemical name:	^{99m}Tc-[Cys^{3,4,10},D-Phe^{7},Arg^{11}]α-MSH$_{3-13}$
Abbreviated name:	^{99m}Tc-(Arg^{11})CCMSH
Synonym:	^{99m}Tc-α-MSH, ^{99m}Tc-Labeled α-melanocyte –stimulating hormone peptides
Backbone:	Peptide
Target:	Melanocortin-1 (MC-1) receptor
Mechanism:	Peptide-receptor binding
Method of detection:	Single-photon emission computed tomography (SPECT) or gamma planar imaging
Source of signal:	^{99m}Tc
Activation:	No
Studies:	• *In vitro* • Rodents



^{99m}Tc-(Arg^{11})CCMSH structure.
Click on protein, nucleotide (RefSeq), and gene for more information about the melanocortin-1 receptor.

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Background

[PubMed]

^{99m}Tc-[Cys^{3,4,10},D-Phe^{7},Arg^{11}]αMSH$_{3-13}$ (^{99m}Tc-(Arg^{11})CCMSH) is a radioligand developed as a single-photon emission computed tomography (SPECT) imaging probe for primary and metastatic melanoma (1). ^{99m}Tc-(Arg^{11})CCMSH is an α-melanocyte –stimulating hormone (MSH) peptide labeled with ^{99m}Tc, a gamma emitter with a physical $t_{½}$ of 6 h.

Malignant melanoma is the sixth most common cancer in the United States (1). Early diagnosis and prompt surgical removal comprise the best approach for a possible cure (2). The melanocortin (MC) system is the best characterized neuropeptide network of the skin, and it is involved in pigmentation regulation, cortisol production and many other physiological processes (3). Most cutaneous cell types express MC receptors, proopiomelanocortin (POMC), prohormone convertases, and also release MCs. Five MC receptors (MC-1 to MC-5) have been cloned and characterized as receptors that belong to the G-protein–coupled receptors superfamily. MSHs (α-, β-, and γ-MSH) are derived from POMC by the proteolytic action of prohormone convertases. α-MSH (Ac-Ser^{1}-Tyr^{2}-Ser^{3}-Met^{4}-Glu^{5}-His^{6}-Phe^{7}-Arg^{8}-Trp^{9}-Gly^{10}-Lys^{11}-Pro^{12}-Val^{13}-NH$_2$) is a 13–amino acid peptide and is the most potent naturally occurring melanotropic peptide (4). The biological effects of α-MSH are mediated *via* MC receptors.

Although positron emission tomography (PET) imaging with [^{18}F]fluoro-2-deoxy-2-D-glucose ([^{18}F]FDG) is effective in the detection of melanoma, it is not melanoma-specific and some melanoma cells do not take up [^{18}F]FDG (5, 6). Radiolabeled α-MSH peptide analogs have been shown to specifically bind to MC-1 receptors which are overexpressed on human and mouse melanoma cells (5, 7-10). Some studies have successfully used 1,4,7,10-tetraazacyclodecane-*N*,*N*′,*N*″,*N*‴-tetraacetic acid (DOTA) coupled to the α-MSH peptide analogs for radiolabeling. These α-MSH derivatives (DOTA-α-MSH) can be labeled with a variety of radionuclides (5). To improve the *in vivo* pharmacokinetics of these radiolabeled peptides, a number of different α-MSH analogs have been designed (11, 12). Froidevaux et al. (11) showed that the kidney uptake of a short linear DOTA–α-MSH analog (DOTA-NAPamide) could be considerably reduced by neutralizing the charge of the Lys^{11} residue. Giblin et al. (13) used metal cyclization to design a new class of α-MSH peptide analogs that are resistant to chemical and proteolytic degradation *in vivo* (10). ^{99m}Tc-CCMSH analogs, cyclized through site-specific coordination of ^{99m}TcO, showed excellent tumor uptake and retention properties in B16/F1 murine melanoma-bearing C57 mice (13, 14). Miao et al. (12) replaced the Lys^{11} with Arg^{11} in the CCMSH analog to form (Arg^{11})CCMSH in an effort to reduce nonspecific kidney uptake and minimize the kidney radiation dose.

Synthesis

[PubMed]

Miao et al. (12) reported the synthesis of the (Arg^{11})CCMSH by using the standard fluorenylmethoxycarbonyl (Fmoc)/HBTU chemistry on amide resin. A solid-phase peptide synthesizer was used. The peptide was acetylated with glacial acetic acid at the N-terminus. The peptide was deprotected and cleaved from the resin by a mixture of trifluoroacetic acid, thioanisol, ethanedithiol, and water at room temperature for 3 h. The peptide was then purified by high-performance liquid chromatography (HPLC), and the identity was confirmed by electrospray ionization mass spectrometry. The ^{99m}Tc radiolabeling was prepared *via* a glucoheptonate transchelation reaction (14).In this procedure, stannous chloride ($SnCl_2$) was used as a reducing agent and glucoheptonate was used as a transfer ligand. Briefly, 10 μg of the peptide was added to the mixture of $SnCl_2$, ^{99m}Tc-pertechnetate, and glucoheptonate. The mixture was incubated at 75°C for 30 min. The final ^{99m}Tc-(Arg^{11})CCMSH product was purified by HPLC. The specific activity was 1.2135 x 10^{10} MBq/g (3.280 x 10^8 mCi/g). Based on a molecular weight of 1,467, the specific activity can be estimated to be 1.79 x 10^7 MBq/μmol (48.4 x 10^4 mCi/μmol).The radiochemical stability was evaluated in phosphate- buffered saline (pH 7.4). Over a 24-h period of incubation at 25°C in phosphate-buffered saline, only radiolabeled peptide was detected by HPLC analysis with no detectable free radioactivity (14).

In Vitro Studies: Testing in Cells and Tissues

[PubMed]

Giblin et al. (13) performed *in vitro* quantitative receptor-binding assays of unlabeled CCMSH on B16/F1 murine melanoma cells. The inhibition constant (IC_{50}) was determined to be 7.6 x 10–^{9} M. Miao et al. (12) determined the IC_{50} of unlabeled (Arg^{11})CCMSH to be 1.7 X 10–^{9} M, and the molecular weight was measured to be 1,476.

Animal Studies

Rodents

[PubMed]

Biodistribution and SPECT imaging studies with ^{99m}Tc-(Arg^{11})CCMSH were conducted in C57 mice bearing B16/F1 (flank melanoma tumors) and B16/F10 (pulmonary metastatic melanoma tumors) melanomas (1). Each mouse received 27.75 MBq (0.75 mCi) of ^{99m}Tc-(Arg^{11})CCMSH by i.v. administration. The radioactivity levels in the flank tumors (n = 4) were 14.03 ± 2.58 (1 h), 11.16 ± 1.77 (4 h), and 3.33 ± 0.50 (24 h) % injected dose per g (% ID/g). The tumor/muscle ratios were 61 (1 h), 372 (4 h), and 333 (24 h). The critical organ appeared to be the kidneys (% ID/g) with 11.66 ± 1.44 (1 h), 5.53 ± 1.17 (4 h), and 0.60 ± 0.08 (24 h). The other major organ radioactivity levels (% ID/g) at 1 h were 7.32 ± 1.28 (intestines), 3.00 ± 0.45 (stomach), 1.59 ± 0.19 (liver), 1.52 ± 0.39 (lung), 1.97 ± 0.37 (skin), 0.63 ± 0.17 (heart), 0.04 ± 0.02 (brain), 0.23 ± 0.06 (muscle), and 0.87 ± 0.12 (blood). With coinjection of 10 μg of a high- affinity unlabeled αMSH

analog, NDP ([NIe^4,D-Phe^7] αMSH), the tumor radioactivity was reduced by 90% (1.44 ± 0.23% ID/g) at 1 h.

In the pulmonary metastatic tumor model, the radioactivity levels (% ID/g) of ^{99m}Tc-(Arg^{11})CCMSH in the lung metastases (n = 4) were 2.77 ± 0.98 (2 h) and 2.30 ± 1.78 (4 h) (1). The radioactivity levels in the normal lung were 0.52 ± 0.22 (2 h) and 0.65 ± 0.13 (4 h). The lung metastases/normal lung ratios were 5.33 and 3.54 at 2 h and 4 h, respectively. The lung metastases/muscle ratios were 11.54 and 5.00 at 2 h and 4 h, respectively. In the imaging studies, the flank melanoma tumors were visualized clearly at 2 h. The pulmonary metastatic melanoma lesions were also clearly imaged at 2 h, and distinct metastatic focal deposits were visible.

Other Non-Primate Mammals

[PubMed]

No publication is currently available.

Non-Human Primates

[PubMed]

No publication is currently available.

Human Studies

[PubMed]

No publication is currently available.

Supplemental Information

[Disclaimers]

NIH Support

NCI P50-CA-103130

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