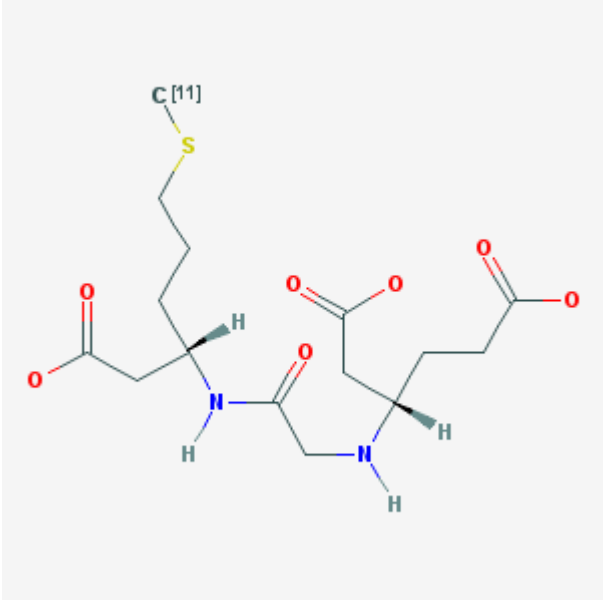


## *N*-[*N*-[(*S*)-1,3-Dicarboxypropyl]carbamoyl]-*S*-[<sup>11</sup>C]methyl-L-cysteine [<sup>11</sup>C]DCMC

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<b>Chemical name:</b>	<i>N</i> -[ <i>N</i> -[( <i>S</i> )-1,3-Dicarboxypropyl]carbamoyl]- <i>S</i> -[ <sup>11</sup> C]methyl-L-cysteine	
<b>Abbreviated name:</b>	[ <sup>11</sup> C]DCMC	
<b>Synonym:</b>	[ <sup>11</sup> C]MCG	
<b>Agent Category:</b>	Amino acid	
<b>Target:</b>	Prostate-specific membrane antigen (PSMA)	
<b>Target Category:</b>	Binding to PSMA	
<b>Method of detection:</b>	PET	
<b>Source of signal:</b>	<sup>11</sup> C	
<b>Activation:</b>	No	
<b>Studies:</b>	<ul style="list-style-type: none"> <li><i>In vitro</i></li> <li>Rodents</li> <li>Non-human primates</li> </ul>	

## Background

[PubMed]

The prostate-specific membrane antigen (PSMA) is a cell-surface glycoprotein with a molecular weight of ~100 kDa. It is expressed predominantly in the prostate and has also been detected in other tissues such as the kidney,

the proximal small intestine, and the salivary glands. Minimal expression has also been observed in the brain (1). It is believed that PSMA plays an important role in the progression of prostate cancer, glutamatergic neurotransmission, and in the absorption of folate (2). This glycoprotein is specifically upregulated several-fold in prostate cancers (PCAs), metastatic disease, and hormone-refractory PCAs because androgen levels inversely modulate PSMA levels (3). Interestingly, PSMA also has an elevated expression in the neovasculature of solid tumors, but is absent in normal neovasculature and prostate cancers tumors (4). Although its function in the prostate is unknown, in the central nervous system PSMA metabolizes *N*-acetyl-aspartyl-glutamate (NAAG), and in the proximal small intestine it removes  $\gamma$ -linked glutamates from poly- $\gamma$ -glutamate folate and folate hydrolase (FOLH1), and also acts as a carboxypeptidase, glutamate carboxypaptidase II (GCPII) (1). Due to its unique distribution, PSMA can be used as a marker for the detection of metastatic cancers with imaging agents. Although a commercially available monoclonal antibody [ $^{111}\text{In}$ -Capromomab pendetide (ProstaScint)] is in clinical use for the detection of prostate cancer, the results obtained with this antibody are not entirely reliable (5). In addition, the antibodies have limited access to tumors and may produce low signal-to-noise ratios because the target is intracellular, whereas PSMA is present on the cell surface (6, 7). Therefore, a small molecule  $^{11}\text{C}$  ligand, *N*-[*N*-[(*S*)-1,3-dicarboxypropyl]carbamoyl]-*S*-[ $^{11}\text{C}$ ]methyl-*L*-cysteine ([ $^{11}\text{C}$ ]DCMC), was synthesized and evaluated for the imaging of PSMA-positive and -negative tumors using positron emission tomography (PET) in an *in vivo* rodent model for prostate cancer (8).

## Synthesis

[PubMed]

*N*-[*N*-[(*S*)-1,3-Dicarboxypropyl]carbamoyl]-*L*-cysteine was used as a precursor for the synthesis of [ $^{11}\text{C}$ ]DCMC (9, 10). The precursor (9) was dissolved in *N,N*-dimethylformamide (DMF), and a fresh DMF/ammonia solution was added to the solution and then followed by water (10). The solution was cooled to 20°C and  $^{11}\text{C}$ -labeled iodomethane was bubbled through it. The reaction contents were subsequently heated for 60 seconds in a 45°C bath, and the reaction was terminated with acetonitrile/water/trifluoroacetic acid (6:94:0.075), followed by the addition of 20% trifluoroacetic acid (10). [ $^{11}\text{C}$ ]DCMC was separated by high-performance liquid chromatography, reformulated in 0.9% saline, and sterile filtered. The specific radioactivity of [ $^{11}\text{C}$ ]DCMC was determined and sterile sodium bicarbonate was added to the radioactive compound to bring the pH of the final formulation to ~7.0. The time for synthesis, including formulation, was ~30 min. Based on the amount of  $^{11}\text{C}$ -labeled iodomethane used for the synthesis, the yield of [ $^{11}\text{C}$ ]DCMC was calculated to be 16% at the end of synthesis, with a purity of >97% (10) and specific radioactivity of 167 Gbq/ $\mu\text{mol}$  (4,000 Ci/mmol) within 30 min after bombardment.

## In Vitro Studies: Testing in Cells and Tissues

[PubMed]

The relative affinity of DCMC for PSMA was determined using the NAAG peptidase assay in membranes isolated from Chinese hamster ovary cells transfected with rat GCPII (11). For this assay, 4  $\mu\text{mol/L}$  NAAG that contained trace amounts of  $^3\text{H}$ -NAAG was incubated with the membranes in the presence of 0.1, 0.3, 1, 3, 10, 30, 100, 300, and 1,000 nmol/L DCMC. The NAAG hydrolysis product was separated by ion-exchange chromatography and determined by scintillation spectrophotometry (8). The  $K_i$  of DCMC for the NAAG activity was determined to be 1.9 nmol/L.

## Animal Studies

### Rodents

[PubMed]

The *in vivo* biodistribution of  $[^{11}\text{C}]\text{DCMC}$  was investigated in healthy (10) and xenograft-bearing mice (8).

Male CD-1 mice weighing 20 to 25 g received an injection of 100  $\mu\text{Ci}$  (3700 kBq)  $[^{11}\text{C}]\text{DCMC}$  in 200  $\mu\text{l}$  saline through the tail vein (12). The mice were killed 5, 15, 30, 60, and 120 minutes after the injection. Brains were removed from the animals, and the cerebellum, olfactory bulb, hypothalamus, hippocampus, striatum, parietal cortex, brainstem, and the thalamus were harvested. The various tissues were weighed and the radioactivity was determined with an automated  $\gamma$  counter. For binding specificity, the mice were pretreated with doses of 1, 10, and 100 mg/kg 2-(phosphonomethyl)pentanedioic acid (PMPA), a potent inhibitor of GCPII (12), 5 minutes prior to the injection of  $[^{11}\text{C}]\text{DCMC}$ . The uptake of  $[^{11}\text{C}]\text{DCMC}$  was highest (61.0% of total) in the kidneys (which also have the highest GCPII concentration in the body) and was completely washed out by 120 minutes after administration. The prostate had a maximum of 2.55% uptake, probably because the mouse prostate has low levels of PSMA (10, 13). In this study, the brain, cerebellum, hippocampus, and cortex showed minimal incorporation of radioactivity. A target binding specificity of  $[^{11}\text{C}]\text{DCMC}$  was indicated because excess unlabelled DCMC or PMPA (up to 1 mg/kg) blocked uptake in the kidneys (10). In this study, the metabolites were determined at 5, 15, 30, and 60 minutes after administration. The plasma and kidneys showed the highest metabolite levels, 9.2% and 10.4% of the total, respectively, at 60 minutes. This is probably because the metabolites bind to and are excreted through the kidneys.

The human LNCaP cell line (PSMA-positive) of human prostate cancer was used to develop the xenograft-bearing mouse (8). For PSMA-negative controls, animals with PC-3 (prostate cancer) and MCF-7 (breast cancer) cell xenografts were used. For the study, animals bearing xenografts (17–20 g) were injected through the tail vein either with 6.49 pmol, 200  $\mu\text{Ci}$  (7400 kBq)  $[^{11}\text{C}]\text{DCMC}$  alone or in combination with 50 mg/kg PMPA. Blood was collected immediately after animals were killed; select tissues were harvested and weighed, and absorbed radioactivity was counted in a  $\gamma$  counter. The biodistribution of  $[^{11}\text{C}]\text{DCMC}$  was highest in the LNCaP tumors, and tumors with MCF-7 or PC-3 cells exhibited significantly lower incorporation of radioactivity (8). Using excess amounts of PMPA as a blocking agent, these observations were confirmed with PET and planer  $\gamma$  scintigraphy imaging studies, respectively. Similar observations, as described above, were made with imaging of the kidneys (8).

## Other Non-Primate Mammals

[\[PubMed\]](#)

No publications are currently available.

## Non-Human Primates

[\[PubMed\]](#)

Renal cortical uptake and clearance studies of  $[^{11}\text{C}]\text{DCMC}$  were performed using PET in an ~30-kg adult male baboon, *Papio Anubis* (10). PET scanning was initiated immediately after an intravenous injection of 10 mCi (370 MBq) high specific activity  $[^{11}\text{C}]\text{DCMC}$ . Thirty-five simultaneous, contiguous, sequential, high-resolution quantitative tomographic slices of the brain were obtained over a 90-minute period. In addition, the animal was positioned so that the renal cortex was in complete view. This study showed there was significant uptake of  $[^{11}\text{C}]\text{DCMC}$  in the renal cortex of the animal, which was decreased by pretreatment with 2 mg/kg PMPA. Uptake of  $[^{11}\text{C}]\text{DCMC}$  in the brain was low and the authors concluded that  $[^{11}\text{C}]\text{DCMC}$  is not suitable to measure GCPII activity in the brain.

## Human Studies

[\[PubMed\]](#)

No publications are currently available.

## References

1. Ghosh A, Heston WD. Tumor target prostate specific membrane antigen (PSMA) and its regulation in prostate cancer. *J Cell Biochem.* 2004; **91** (3):528–39. PubMed PMID: 14755683.
2. O'Keefe, D, D Bachich, and WD Heston, Prostate specific membrane antigen. Prostate cancer, biology, genetics, and the new therapeutics., ed. L Chung, W Issacs, and J Simons. 2001, New Jersey: Humana Press. 307-326.
3. Wright GL, Grob BM, Haley C, Grossman K, Newhall K, Petrylak D, Troyer J, Konchuba A, Schellhammer PF, Moriarty R. Upregulation of prostate-specific membrane antigen after androgen-deprivation therapy. *Urology.* 1996; **48** (2):326–34. PubMed PMID: 8753752.
4. Chang SS. Monoclonal antibodies and prostate-specific membrane antigen. *Curr Opin Investig Drugs.* 2004; **5** (6):611–5. PubMed PMID: 15242249.
5. Ponsky LE, Cherullo EE, Starkey R, Nelson D, Neumann D, Zippe CD. Evaluation of preoperative ProstaScint scans in the prediction of nodal disease. *Prostate Cancer Prostatic Dis.* 2002; **5** (2):132–5. PubMed PMID: 12497003.
6. Jain RK. Transport of molecules, particles, and cells in solid tumors. *Annu Rev Biomed Eng.* 1999; **1** :241–63. PubMed PMID: 11701489.
7. Sundaresan G, Yazaki PJ, Shively JE, Finn RD, Larson SM, Raubitschek AA, Williams LE, Chatziioannou AF, Gambhir SS, Wu AM. 124I-labeled engineered anti-CEA minibodies and diabodies allow high-contrast, antigen-specific small-animal PET imaging of xenografts in athymic mice. *J Nucl Med.* 2003; **44** (12):1962–9. PubMed PMID: 14660722.
8. Foss CA, Mease RC, Fan H, Wang Y, Ravert HT, Dannals RF, Olszewski RT, Heston WD, Kozikowski AP, Pomper MG. Radiolabeled small-molecule ligands for prostate-specific membrane antigen: in vivo imaging in experimental models of prostate cancer. *Clin Cancer Res.* 2005; **11** (11):4022–8. PubMed PMID: 15930336.
9. Kozikowski AP, Nan F, Conti P, Zhang J, Ramadan E, Bzdega T, Wroblewska B, Neale JH, Pshenichkin S, Wroblewski JT. Design of remarkably simple, yet potent urea-based inhibitors of glutamate carboxypeptidase II (NAALADase). *J Med Chem.* 2001; **44** (3):298–301. PubMed PMID: 11462970.
10. Pomper MG, Musachio JL, Zhang J, Scheffel U, Zhou Y, Hilton J, Maini A, Dannals RF, Wong DF, Kozikowski AP. 11C-MCG: synthesis, uptake selectivity, and primate PET of a probe for glutamate carboxypeptidase II (NAALADase). *Mol Imaging.* 2002; **1** (2):96–101. PubMed PMID: 12920850.
11. Fuhrman S, Palkovits M, Cassidy M, Neale JH. The regional distribution of N-acetylaspartylglutamate (NAAG) and peptidase activity against NAAG in the rat nervous system. *J Neurochem.* 1994; **62** (1):275–81. PubMed PMID: 8263527.
12. Jackson PF, Cole DC, Slusher BS, Stetz SL, Ross LE, Donzanti BA, Trainor DA. Design, synthesis, and biological activity of a potent inhibitor of the neuropeptidase N-acetylated alpha-linked acidic dipeptidase. *J Med Chem.* 1996; **39** (2):619–22. PubMed PMID: 8558536.
13. Tiffany CW, Lapidus RG, Merion A, Calvin DC, Slusher BS. Characterization of the enzymatic activity of PSM: comparison with brain NAALADase. *Prostate.* 1999; **39** (1):28–35. PubMed PMID: 10221263.