AXUMIN™
Fluciclovine F 18 Injection

Axumin™ (fluciclovine F 18) Scientific and Clinical Background

Indication and Important Safety Information

INDICATION

Axumin™ (fluciclovine F 18) injection is indicated for positron emission tomography (PET) imaging in men with suspected prostate cancer recurrence based on elevated blood prostate specific antigen (PSA) levels following prior treatment.

IMPORTANT SAFETY INFORMATION

- Image interpretation errors can occur with Axumin PET imaging. A negative image does not rule out recurrent prostate cancer and a positive image does not confirm its presence. The performance of Axumin seems to be affected by PSA levels. Axumin uptake may occur with other cancers and benign prostatic hypertrophy in primary prostate cancer. Clinical correlation, which may include histopathological evaluation, is recommended.
- Hypersensitivity reactions, including anaphylaxis, may occur in patients who receive Axumin. Emergency resuscitation equipment and personnel should be immediately available.
- Axumin use contributes to a patient's overall long-term cumulative radiation exposure, which is associated with an increased risk of cancer. Safe handling practices should be used to minimize radiation exposure to the patient and health care providers.
- Adverse reactions were reported in ≤1% of subjects during clinical studies with Axumin. The most common adverse reactions were injection site pain, injection site erythema and dysgeusia.

To report suspected adverse reactions to Axumin, call 1-855-AXUMIN1 (1-855-298-6461) or contact FDA at 1-800-FDA-1088 or www.fda.gov/medwatch.

Full Axumin prescribing information is available at: www.axumin.com.

Disclaimer: Please note that Blue Earth Diagnostics does not recommend the use of Axumin™ (fluciclovine F 18) injection in any manner inconsistent with that described in the full prescribing information.

AXUMIN™
Fluciclovine F 18 Injection

Axumin™ (fluciclovine F 18) Scientific and Clinical Background

BACKGROUND

Axumin (generic name: fluciclovine F 18, and previously known as F-18 FACBC), is a diagnostic radiopharmaceutical consisting of a synthetic amino acid (an analog of leucine) for Positron Emission Tomography (PET) imaging. Axumin was approved by the FDA on May 27, 2016. The approved clinical indication is as follows:

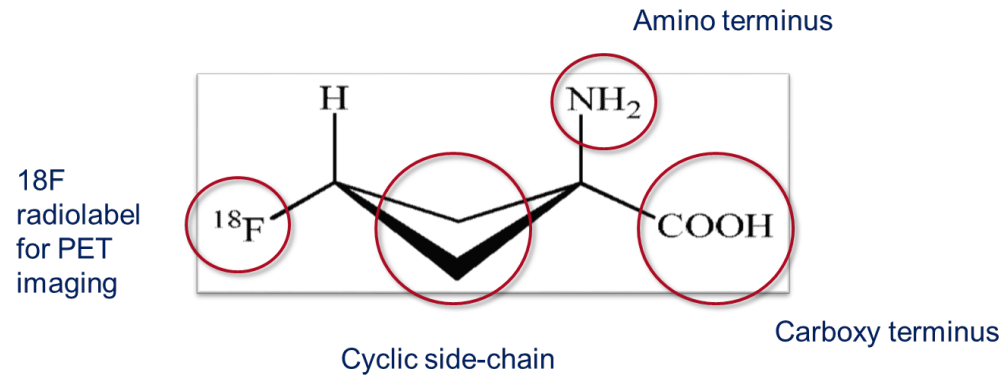
Axumin is a radioactive diagnostic agent indicated for positron emission tomography (PET) imaging in men with suspected prostate cancer recurrence based on elevated blood prostate specific antigen (PSA) levels following prior treatment.[1]

The compound was originally invented by Mark Goodman and Timothy Shoup at Emory University, in a search for an agent with characteristics that might enable improved imaging of brain and genitourinary malignancies. The resultant Emory University patents were originally licensed to Nihon Mediphysics (NMP), a Japanese radiopharmaceutical company. GE Healthcare licensed fluciclovine from NMP for development outside of Japan and has since sub-licensed these rights to Blue Earth Diagnostics (BED).

BED is funded by Syncona LLP, an investment company of The Wellcome Trust. BED's initial research focus for the product has been in the detection and localization of recurrent prostate cancer and assessment of metastases in selected patients with primary prostate cancer.

CHEMICAL COMPOSITION

The molecular structure of Axumin is shown below in Figure 1. Its molecular weight is 132 Daltons.



*anti*1-amino-3-18F-fluorocyclobutane-1-carboxylic acid (also known as FACBC)

Figure 1: Molecular structure of Fluciclovine F 18 (Axumin)

AMINO ACID TRANSPORT IN CANCER

Cancer cells proliferate rapidly and may have aberrant cell energetics, increasing the demand for amino acids for both anabolism & catabolism. Mammalian cells have many amino acid transporters which may be over-expressed in cancer cells and certain amino acids (leucine, glutamine) are also involved in pro-oncogenic signaling via mTOR, the activity of which in prostate cancer cells is linked to androgen driven LAT1 & LAT3 amino acid transporter expression, as shown in Figure 2 below.[2,3]

MECHANISM OF ACTION

Fluciclovine has been shown to be transported into prostate cancer cell lines, predominantly via ASCT2 (like glutamine) or LAT1 transporters, with the role of the latter increasing under conditions of acidic pH, such as may develop intra-tumorally once cancers reach a certain size.[4] Fluciclovine F 18 imaging has also been demonstrated in orthotopic prostate cancer models.[5,6] Fluciclovine F 18 is not metabolized nor is it incorporated into newly synthesized proteins.

The uptake of fluciclovine into prostate cancer cells has been investigated in cell lines expressing androgen receptors (LNCaP) or not (DU145), with and without 5α-dihydrotestosterone (DHT) and with and without bicalutamide.[7] DHT stimulated the expression of amino acid transporters in LNCaP but not DU145 cells. [^{14}C] labeled Fluciclovine uptake was enhanced, in a DHT-dependent manner, in LNCaP cells only and the increased uptake could be reversed following co-incubation with bicalutamide. This shows that in a hormone sensitive cell line, androgen dependent signaling causes an increased uptake of fluciclovine F 18, which can be reversed following administration of an androgen receptor blocker such as bicalutamide. Further exploration of the link between therapy induced modulation of fluciclovine (^{18}F) and subsequent tumor responses may therefore be warranted.

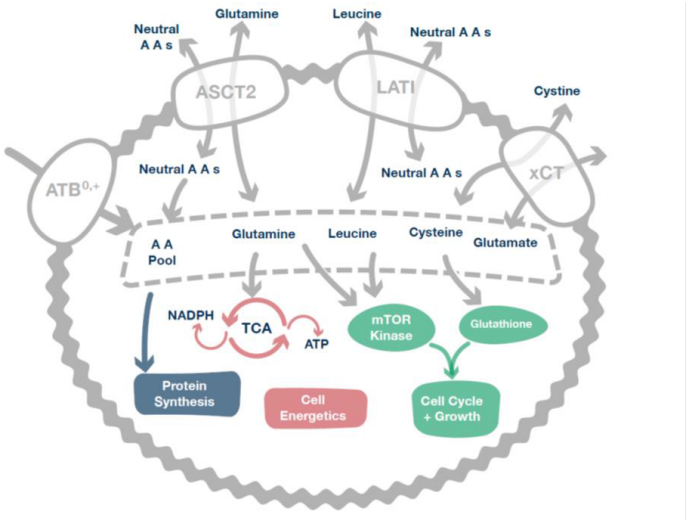


Figure 2: Role of amino acids and their transporters in cellular metabolism

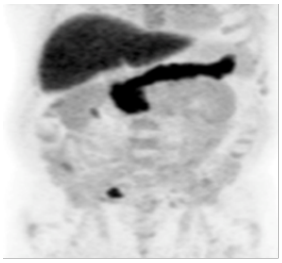
BIODISTRIBUTION[1]

After intravenous administration, the highest amounts of fluciclovine F 18 are found in the following organs and tissues (as a percent of administered radioactivity):

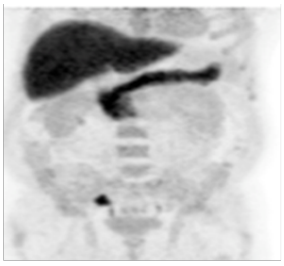
Liver: 14%
Red bone marrow: 12%
Lung: 7%
Myocardium: 4%
Pancreas: 3%

With increasing time, fluciclovine F 18 also distributes to skeletal muscle. Urinary excretion: Across the first four hours post-injection, 3% of administered radioactivity was excreted in the urine. Across the first 24 hours post-injection, 5% of administered radioactivity was excreted in the urine.

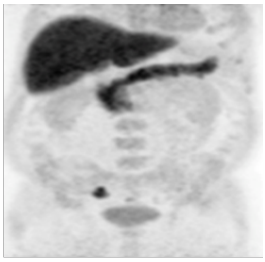
The following images represent fluciclovine F 18 biodistribution. In these images, liver and pancreatic uptake are clearly seen, with some noticeable uptake in bone marrow. Note the extremely low initial urinary bladder activity, which increases gradually over time.[8]



5-16 min. post-injection[10]

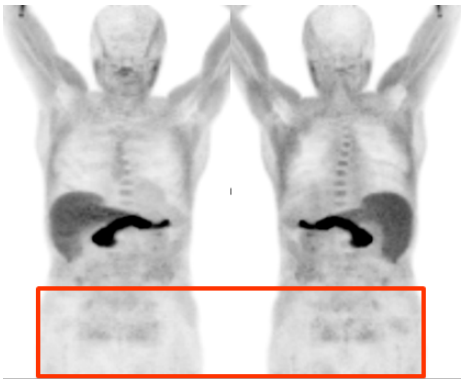


17-28 min. post-injection[10]



29-40 min. post-injection[10]

Note the very limited pelvic activity soon after injection:



Early (5 mins.) post-injection[10]

DOSING, ADMINISTRATION, IMAGE ACQUISITION, IMAGE INTERPRETATION[1]

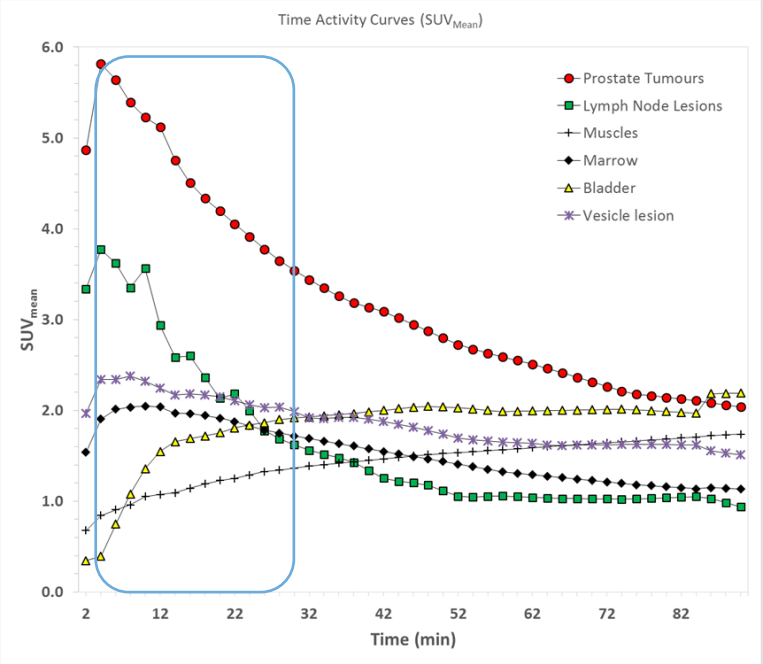
The recommended dose of Axumin is 370 MBq (10 mCi) administered as an intravenous (IV) bolus injection followed by IV saline flush. The patient should avoid any significant exercise for at least one day prior to PET imaging to minimize skeletal uptake, and the patient should fast for at least 4 hours prior to administration (although small amounts of water are permissible).

The patient should be injected on the PET scanner table, and positioned supine with their arms above their head. CT scanning for attenuation correction and anatomic correlation may then be initiated. PET scanning should begin 3 to 5 minutes after completion of injection. Image acquisition should begin at mid-thigh and proceed to the base of the skull. Typical total scan time is between 20 to 30 minutes.

Localization of recurrence in sites typical for prostate cancer is based on fluciclovine F 18 uptake in comparison with tissue background. For larger lesions (≥1 cm), uptake equal to or greater than bone marrow is considered suspicious for prostate cancer recurrence. For small lesions (<1 cm), focal uptake greater than blood pool should be considered suspicious for prostate cancer recurrence.

PHARMACODYNAMICS

The uptake and clearance of fluciclovine F 18 as a function of time are represented in the time activity curves below. The optimal image time window is delineated by the box.[1,9]



AXUMIN™
Fluciclovine F 18 Injection

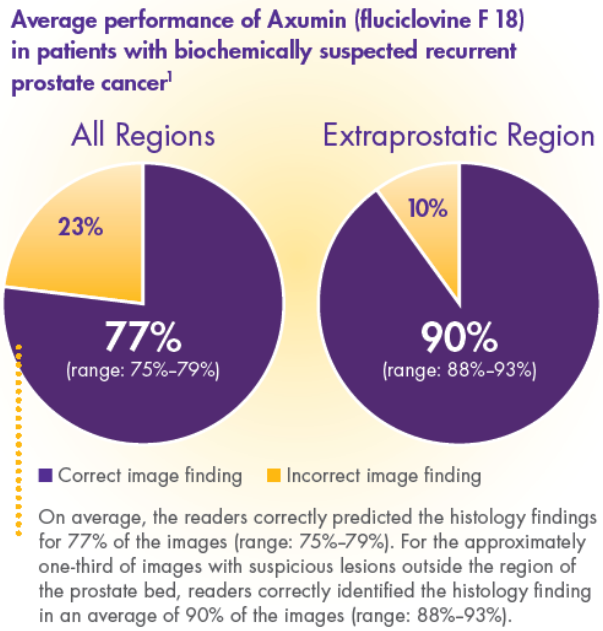
CLINICAL EXPERIENCE – BIOCHEMICALLY RECURRENT PROSTATE CANCER[1]

The safety and efficacy of Axumin were evaluated in two studies (Study 1 and Study 2) in men with suspected recurrence of prostate cancer based on rising PSA levels following radical prostatectomy and/or radiotherapy.

Study 1 evaluated 105 fluciclovine F 18 PET scans in comparison to histopathology obtained by biopsy of the prostate bed and biopsies of lesions suspicious by imaging. PET/CT imaging generally included the abdomen and pelvic regions. Images were originally read by on-site readers.

In Study 2, 96 fluciclovine and C11 choline scans were compared. The choline scans were read by on-site readers and the fluciclovine F 18 scans were read by the Study 1 blinded readers.

Clinical Summary: 105 Axumin images were evaluated by three independent readers who were unaware of the clinical details of each patient or whether the biopsy of the prostate gland was positive or negative for cancer. On average, a correct image finding was identified in 77% of patients (range: 75%-79%). For cancer outside the region of the prostate, a correct image finding for cancer was identified in an average of 90% of patients (range: 88%-93%). The results seem to be affected by PSA levels with, in general, lower PSA levels in patients with negative scans than in those with positive scans. In patients with PSA levels ≤ 1.78 ng/mL, 15 of 25 had a positive scan, with 11 confirmed as positive by histology; 71 of 74 patients with PSA levels > 1.78 ng/mL had a positive scan, of which 58 were confirmed as positive.



Charts illustrate findings from Study 1 only. Information adapted from Section 14, Table 4, of Prescribing Information.

AXUMIN™
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Performance of Axumin in Patients with Biochemically Suspected Recurrent Prostate Cancer, at the Patient Level and at the Prostate Bed and Extra-prostatic Region Levels[1]

N = number of patient scans evaluated

	Reader 1	Reader 2	Reader 3
Patient	N = 104	N = 105	N = 99
True Positive	75	72	63
False Positive	24	23	13
True Negative	5	7	15
False Negative	0	3	8
Prostate Bed	N = 98	N = 97	N = 96
True Positive	58	56	47
False Positive	29	26	15
True Negative	10	12	24
False Negative	1	3	10
Extraprostatic	N = 28	N = 28	N = 25
True Positive	25	26	22
False Positive	2	2	2
True Negative	0	0	0
False Negative	1	0	1

Note: extra-prostatic confirmation was normally by fluciclovine F 18 guided biopsy, and negative areas were rarely biopsied. This limits the utility of the information that can be obtained on patients with negative images.

Axumin: Study 1: Imaging vs PSA vs histology[8]

	PSA (ng/mL)			
	≤1.78	**>1.78 - ≤4.48**	**>4.48 - ≤9.25**	**>9.25**
No. patient scans	25	25	25	24
True Positive	11	17	21	20
False Positive	4	5	4	4
True Negative	9	3	0	0
False Negative	1	0	0	0

Note: The detection rate of Axumin seems to be affected by PSA levels, as can be seen from these data.

AXUMIN™
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USER TRAINING

All Axumin customers will be provided product training through a program being offered by Blue Earth Diagnostics, Inc. Training will include the following:

- Imaging and Interpretation Manual
    - Manual including the Axumin Prescribing Information (PI) and detailed background and data substantiating acquisition and interpretation techniques

- Image Acquisition Training
    - Blue Earth Medical Affairs training presentation via Live Meeting or Webinar

- Image Interpretation Training
    - Self-directed training for Imaging Physicians
        - Includes video of expert reader reporting on Axumin cases

Disclaimer: The Axumin™ Image Acquisition Training is provided to help familiarize imagers with techniques for the safe and effective usage of Axumin. The responsibility for the accurate and timely acquisition and interpretation of images using Axumin PET/CT scanning rests with the nuclear medicine physician or radiologist supervising the PET/CT imaging facility. The Axumin Image Acquisition Training is not intended to substitute for the independent medical judgment of the physician(s) responsible for the individual patient's management, nor is it a guarantee of any specific clinical results.

AXUMIN SAFETY PROFILE

The Axumin clinical studies database consists of 877 subjects, including 797 men with prostate cancer. Adverse reactions were reported in ≤1% of subjects during clinical studies with fluciclovine F 18. The most common adverse reactions were injection site pain, injection site erythema (redness) and dysgeusia (abnormal taste in the mouth).

There are no contraindications to the use of Axumin, per the product package insert. Please see page two for Important Safety Information.

CONCLUSION/SUMMARY

Axumin (generic name: fluciclovine F 18, and previously known as F-18 FACBC), is a diagnostic radiopharmaceutical consisting of a synthetic amino acid (an analog of leucine) for Positron Emission Tomography (PET) imaging. Axumin was approved by the FDA on May 27, 2016 with the following indication:

> Axumin is a radioactive diagnostic agent indicated for positron emission tomography (PET) imaging in men with suspected prostate cancer recurrence based on elevated blood prostate specific antigen (PSA) levels following prior treatment.[1]

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Axumin is taken up into prostate tumors by cell surface amino acid transporters which are upregulated in cancer cells. After intravenous administration, the highest amounts of fluciclovine F 18 are found in liver, bone marrow, lung, myocardium and pancreas, with uptake increasing in skeletal muscle over time. Urinary bladder activity is extremely low after injection, and increases gradually over time.

The imaging protocol and interpretive criteria are described herein as well as in the Prescribing Information (see "Dosing, Administration, Image Acquisition, Image interpretation", above). All Axumin customers will be provided product training through a program being offered by Blue Earth Diagnostics, Inc., which will include information on image acquisition and interpretation principles, image acquisition logistics, and image interpretation methodology.

The safety and efficacy of Axumin were evaluated in two studies in men with suspected recurrence of prostate cancer based on rising PSA levels following radical prostatectomy and/or radiotherapy. The results are described in "Clinical experience – Biochemically Recurrent Prostate Cancer", above.

The Axumin clinical database consists of 877 subjects, including 797 men with prostate cancer. Adverse reactions were reported in ≤1% of subjects during clinical studies with fluciclovine F 18. There are no contraindication to the use of Axumin. Please see page two of this document for Important Safety Information.

Further medical information about Axumin can be obtained by calling 1-855-AXUMIN1 (1-855-298-6461). Full Axumin prescribing information is available at: www.axumin.com.

To report suspected adverse reactions to Axumin, call 1-855-AXUMIN1 (1-855-298-6461) or contact FDA at 1-800-FDA-1088 or www.fda.gov/medwatch.

REFERENCES

1. Axumin (fluciclovine F 18) Injection; US Prescribing Information; Blue Earth Diagnostics, Ltd; August 2016
2. Ganapathy et al., Nutrient transporters in cancer: Relevance to Warburg hypothesis and beyond. Pharmacol & Therap. 2009;121
3. Wang et al. Androgen Receptor and Nutrient Signaling Pathways Coordinate the Demand for Increased Amino Acid Transport during Prostate Cancer Progression. Cancer Research 2011; 71(24), 7525-7536.
4. Oka et al Transport mechanisms of trans-1-amino-3-fluoro[1- 14C] cyclobutanecarboxylic acid in prostate cancer cells Nucl Med Biol 2012;39:109–119
5. Oka et al A Preliminary Study of Anti-1-Amino-3-18FFluorocyclobutyl-1-Carboxylic Acid for the Detection of Prostate Cancer J Nucl Med 2007;48:46–55
6. Oka S, Hattori R, Kurosaki F, Toyama M, Williams LA, Yu W, Votaw JR, Yoshida Y, Goodman MM, Ito O. A preliminary study of anti-1-amino-3-18f-fluorocyclobutyl-1- carboxylic acid for the detection of prostate cancer, J Nucl Med. 2007. 48: 46-55.
7. Okudaira H. et al. Accumulation of Trans-1-Amino-3-[18F]Fluorocyclobutanecarboxylic Acid in Prostate Cancer due to Androgen-Induced Expression of Amino Acid Transporters Mol Imaging Biol. 2014;16:756-64.
8. Schuster et al. Anti-1-Amino-3-18F-Fluorocyclobutane-1-Carboxylic Acid: Physiologic Uptake Patterns, Incidental Findings, and Variants That May Simulate Disease. J Nucl Med 2014; 55:1986-92
9. Data on File, Blue Earth Diagnostics, Inc.