

## FAS-detect™ ELISA

A Quantitative Enzyme-Linked Immunosorbent Assay for the Detection of Fatty Acid Synthase (FAS) in Human Serum

### Product Insert

Catalog No. E-1001

**FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES.**

#### Background/Description:

Fatty Acid Synthase (FAS) is an enzyme with cytoplasmic distribution that plays a central role in the de novo biosynthesis of fatty acids (1). Using immunohistochemical techniques, FAS expression has also been observed in a wide variety of tumors, including many common human carcinomas such as breast (2,3), colon (4), prostate (5,6), and endometrial (7) carcinomas (Figure 1). Using a two-site ELISA technique, Wang, et al (8) have demonstrated the ability to quantitatively measure FAS in the serum of individuals with several different types of cancer, including carcinoma of the breast, prostate, colon and ovary.

FAS-detect™ ELISA is a highly sensitive, specific, and quantitative enzyme-linked immunosorbent assay that can be used to detect soluble FAS in human serum, secondary to production and release by FAS-positive tumors.

#### Kit Components:

1. FAS Antigen Capture Plate (96 tests)-1 ea.
2. FAS Sample Diluent containing Biotinylated FAS Detection Antibody (12 ml).
3. FAS Enzyme Conjugate. Horseradish Peroxidase-labelled Streptavidin (12 ml)
4. FAS Standards (5 ea.). 1 ml each with concentration indicated on vial label
5. FAS TMB Substrate (12 ml)
6. FAS Substrate Stop Solution (12 ml)
7. FAS 20x Wash Buffer (30 ml)
8. FAS Sample Dilution Tray-1 ea.

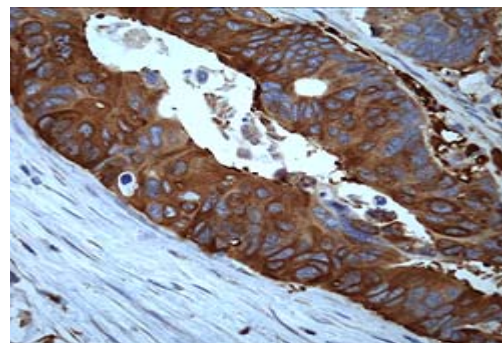


UMB Research Park, Bldg One  
800 W. Baltimore Street, Suite 150  
Baltimore, MD 21201  
(P) 410.558.9200 • (F) 410.558.9300

#### Storage:

Store all kit components at 2-8° C. During prolonged storage, crystal formation may be observed in the 20x Wash Buffer concentrate. The crystals can be re-dissolved by swirling the bottle in warm tap water.

Figure 1: Immunohistochemical staining of FAS in colon cancer using FAS-detect™ IHC (original magnification 400x).



#### Procedure:

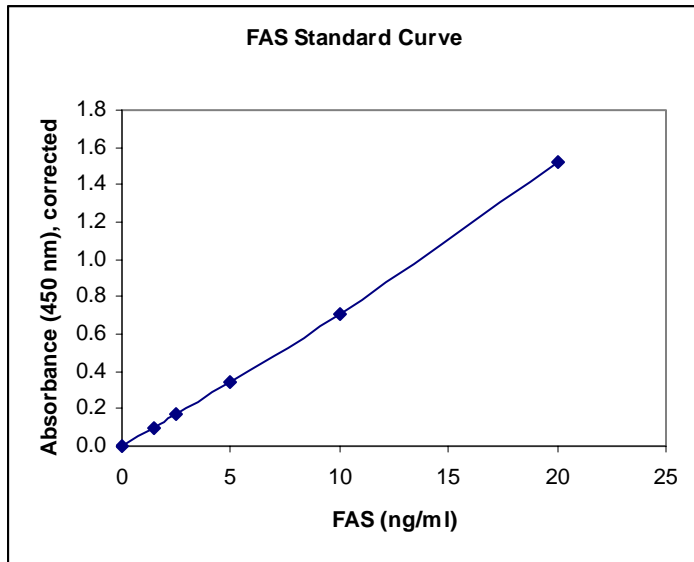
1. Remove the kit components from storage and allow to warm to room temperature.
2. Transfer 100 ul of samples and standards to the sample dilution tray in duplicate.
3. Dilute each sample and standard with 100 ul of Sample Diluent/Detection Antibody.
4. Transfer 150 ul of the diluted samples and standards to the FAS Capture Plate.
5. Place the plate on a plate shaker and incubate for 90 min. at room temperature.
6. Wash the plate 5x with 1x Wash buffer.
7. Add 100 ul of FAS Enzyme Conjugate to each well and incubate on a plate shaker for 60 min. at room temperature.
8. Wash the plate 5x with 1x Wash Buffer.
9. Add 100 ul of TMB Substrate to each well and incubate on a plate shaker for 15 min. at room temperature.
10. Stop the reaction by adding 100 ul of Substrate Stop Solution to each well; shake for an additional 10-15 sec.
11. Read the absorbance of each well at 450 nm.

#### Quality Control:

1. Absorbance value for Standard 1 should be less than 0.3.
2. Absorbance value for Standard 5 should <1.0
3. If these parameters are not met, please contact FASgen Diagnostics for technical assistance, (410) 558-9200.

#### Evaluation of Results:

1. Calculate the average absorbance values for each FAS Standard and unknown sample.
2. Subtract the mean value of Standard 1 (0ng/ml) from the mean absorbance of each standard and unknown.
3. Prepare a standard curve by plotting the corrected absorbance for each standard on the y-axis versus the standard concentration (printed on the label of the vial) on the x-axis.
4. Determine the concentration of FAS for each unknown specimen by interpolation from the standard curve (manual or software).
5. Results will be obtained in ng/ml.
  - a. NOTE: If a sample contains FAS levels that exceed the upper portion of the curve, it may be necessary to prepare serial 2-fold dilutions of the sample and re-test.



#### Bibliography:

1. Wakil, S. Fatty acid synthase, a proficient multifunctional enzyme. *Biochemistry* 28: 4523-4530, 1989.
2. Alo, P.L., Visca, P., Marci, A., Mangoni, A., Botti, C., and Di Tondo, U. Expression of fatty acid synthase(FAS) as a predictor of recurrence in stage I breast carcinoma patients. *Cancer* 77: 474-482, 1996.
3. Alo, P., Visca, P., Trombetta, G., Mangoni, A., Lenti, L., Monaco, S., Botti, C., Serpieri, D.E., and Di Tondo, U. Fatty acid synthase (FAS) predictive strength in poorly differentiated early breast carcinomas. *Tumori* 85: 35-40, 1999.
4. Rashid, A., Pizer, E.S., Moga, M., Milgraum, L.Z., Zahurak, M., Pasternack, G.R., Kuhajda, F.P., and Hamilton, S.R. Elevated expression of fatty acid synthase and fatty acid synthetic activity in colorectal neoplasia. *Am. J. Pathol.* 150: 201-208, 1997.
5. Epstein, J.I., Carmichael, M., and Partin, A.W. OA-519 (fatty acid synthase) as an independent predictor of pathologic state in adenocarcinoma of the prostate. *Urology* 45: 81-86, 1995.
6. Swinnen, J.V., Roskams, T., Joniau, S., Van Poppel, H., Oyen, R., Baert, L., Heyns, W., and Verhoeven, G. Overexpression of fatty acid synthase is an early and common event in the development of prostate cancer. *Int. J. Cancer* 98: 10-22, 2002.
7. Pizer, E.S., Lax, S.F., Kuhajda, F.P., Pasternack, G.R., and Kurman, R. Fatty acid synthase expression in endometrial carcinoma: correlation with cell proliferation and hormone receptors. *Cancer* 83: 528-537, 1998.
8. Wang, Y., Kuhajda, F.P., Sokoll, L.J., and Chan, D.W. Two-site ELISA for the quantitative determination of fatty acid synthase. *Clinical Chimica Acta* 304: 107-115, 2001.

#### Distributed by:

ImmTech, Inc.  
206 High St.  
New Windsor, MD 21776  
(410) 775-7060 (Phone)  
(410) 775-7061 (FAX)  
customerservice@immtech.net  
www.immtech.net



UMB Research Park, Bldg One  
800 W. Baltimore Street, Suite 150  
Baltimore, MD 21201  
(P) 410.558.9200 • (F) 410.558.9300