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Expression of the Lipogenic Enzyme Fatty Acid Synthase (FAS) as a Predictor of Poor Outcome in Nephroblastoma: An Interinstitutional Study

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Background. Treatment of nephroblastoma (Wilms tumor) has presently achieved a greater than 80% cure rate. Pathologic stage and grade are considered the most reliable prognostic parameters, but other biologic factors are under study in order to improve patient stratification into risk groups. Correlation of elevated levels of the lipogenic enzyme fatty acid synthase (FAS) with aggressiveness of some cancers has drawn attention to this enzyme as a possible marker of poor prognosis. ***Procedure.*** To determine the predictive strength of FAS expression in Wilms tumor (with particular emphasis on intermediate risk, i.e., non anaplastic tumors, the vast majority of nephroblastomas), we evaluated immunostaining expression in archival specimens from 94 neoplasms. The degree of expression was correlated with stage, grade, clinical course and administration of prenephrectomy chemotherapy. ***Results.*** Expression of FAS increased in anaplastic tumors ($P = 0.043$) and higher stages ($P = 0.029$). FAS expression correlated with OS and DFS at both univariate and multivariate analysis. Comparable results were obtained when analyzing the intermediate risk population separately. Pretreatment resulted in an increased FAS expression, without reaching significance level ($P = 0.059$). ***Conclusions.*** Expression of FAS might be an independent prognostic factor, particularly for intermediate-risk patients. The blockade of fatty acid synthesis by inhibition of FAS enzymatic function by means of metabolic analogues might prove a novel target pathway for the treatment of nephroblastoma. Med Pediatr Oncol 2003;40:302–308.

Key words: nephroblastoma; fatty acid synthase; prognosis; favorable histology; immunohistochemistry

INTRODUCTION

Nephroblastoma (Wilms tumor, WT) is the most common urinary tract malignancy in children [1] and multimodality therapy has achieved a greater than 80% cure rate. Now that most patients are becoming long-term survivors, the aim of cooperative trials has shifted to optimizing effectiveness while minimizing treatment toxicity [2]. Currently, pathologic stage and histology still remain the most important predictors of outcome [3]. Nevertheless, there are some low and intermediate risk patients (Table I), who fail to respond to treatment for no apparent reason, while a few high-risk patients unexpectedly do well. For these reasons, additional bio-pathologic and molecular predictors of outcome might improve risk group stratification [1,2].

Fatty acid synthase (FAS) is a multi-enzyme molecule that plays a central role in the de novo biosynthesis of fatty acids [4]. It consists of two identical subunits containing domains for acyl-carrier peptide and the seven different catalytic activities needed for the conversion of acetyl-CoA and malonil-CoA to palmitate [5,6].

FAS is expressed at low levels in most normal human tissues, since cells preferentially utilize circulating lipids for the synthesis of new structural lipids [4,7]. In the adult, FAS is distributed mainly in cells involved in lipid metabolism and in hormone-sensitive cells. It is, for example, active in the liver to produce lipids for export to metabolically active or storage tissues, in the lactating breast to produce milk lipids, in endometrial glands during the proliferative phase of menstrual cycle, in lung pneumocytes to provide surfactant [8]. In fetal tissues, it is closely associated with cell proliferation and is expressed at high

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TABLE I. Pathological Staging of Nephroblastoma (Simplified) According to the SIOP Nephroblastoma Clinical Trial (1993) and Grading According to the Stockholm Working Classification of Renal Tumors of Childhood (1994)

Stage	
I	Tumor limited to the kidney and completely excised
II−	Tumor extending outside the kidney, but probably completely excised, negative lymph nodes
II+	Tumor extending outside the kidney, but probably completely excised, positive lymph nodes
III	Incomplete excision, without hematogenous metastases (including previous biopsy and tumor rupture)
IV	Distant metastases
V	Bilateral renal tumors
Grade	
Low risk	Cystic partially differentiated nephroblastoma; nephroblastoma with fibroadenomatous-like structures; nephroblastoma of highly differentiated epithelial type; nephroblastoma completely necrotic (after preoperative chemotherapy)
Intermediate risk (standard)	Non-anaplastic nephroblastoma with its variants; nephroblastoma–necrotic but some features left (< 10%)
High risk	Nephroblastoma with anaplasia

levels in epithelial cells of respiratory, digestive and urinary tracts and in mesenchymal and hematolymphoid cells [8,9].

Recent studies have demonstrated that high levels of FAS expression and fatty acid synthetic activity also occur in a subset of human cancers, such as breast, ovary, and prostate [10–21]. In some of them, high degree of activation has been correlated with poor outcome, suggesting a relationship between FAS expression and tumor aggressiveness [10,11,13,16,18,20].

The aim of this study was to investigate the expression of FAS in nephroblastoma and the possible correlation with clinical course and histopathological features. In order to evaluate its prognostic relevance, with special attention to standard histology nephroblastomas (the largest subgroup of patients), we separately examined the whole WT population and the intermediate risk group. In addition, since our WT series included both pretreated and non-pretreated patients, we investigated the possible influence of neoadjuvant chemotherapy (CT) on FAS activation in neoplastic cells.

MATERIALS AND METHODS

Patients

We retrospectively identified 94 children affected by nephroblastoma with at least 12 months of follow-up and available tissue from the files of ''Bambino Gesù'' Children's Hospital. Data regarding sex, age, treatment and clinical course were recorded. Criteria for staging and grading were according to the protocol of the Société Internationale d'Oncologie Pédiatrique (SIOP) 93-01, December 1995 revision [3] (Table I).

Patients receiving preoperative CT were analyzed separately from those directly undergoing surgery. Clinical course was considered as overall survival (OS) and disease free survival (DFS) from date of diagnosis, as based on the initial diagnostic imaging, according to SIOP guidelines.

Immunohistochemistry

Four-micrometer sections from formalin-fixed paraffin-embedded tissue were de-waxed in xylene, rehydrated in ethanol dilution series, and then incubated for 20 min in 3% hydrogen peroxide to block endogenous peroxidase. After washes in TBS, the sections were treated with 20% normal bovine serum and then incubated at room temperature for 60 min with an affinity-purified rabbit polyclonal antiserum raised against human FAS (anti-FAS antibody was a gift from F.P. Kuhajda, MD, Department of Pathology, Johns Hopkins Medical Institutions, Baltimore, MD. Concentrated serum dilution 1:3,000). After washing, the slides were re-incubated with biotinylated anti-rabbit IgG (DAKO A/S, Glostrup-Denmark) at room temperature for 30 min, followed by further incubation with an avidin and biotinylated horseradish peroxidase complex (DAKO) at room temperature for 30 min. Chromogenic development was obtained using 3,3′-diaminobenzidine tetrahydrochloride with 0.03% hydrogen peroxidase (DAKO). Finally, sections were counterstained with hematoxylin, cleared and mounted. Sections without primary antibodies served as negative controls, while perirenal fat tissue adjacent to tumor served as positive control.

Two pathologists (F.D.C. and R.B.) independently performed light microscopic evaluation of stained sections without knowledge of patient characteristics. As FAS reaction was not uniformly distributed, they randomly selected 15 tumor fields and obtained a final mean value for each tumor. The staining was scored using the following scale: 0: negative (0–5% positive cells), 1+: weak (6–15%), 2+: moderate (16–30%), 3+: strong (31–50%), and 4+: very strong (>50%). Perirenal fat tissue, as internal positive control, was arbitrarily considered as 4+.

Statistical Analysis

For statistical analysis, stages were divided in low (I, II/negative lymph nodes), high (II/positive lymph nodes, III, IV), and bilateral (V).

The Chi square test was used to evaluate the association between FAS expression and categorical variables (stage and pretreatment). The Kendall's rank correlation coefficient was calculated to evaluate the correlation between FAS expression and grade. The Kaplan–Meier method (univariate analysis) and the Cox proportional hazard model (multivariate analysis) were applied to investigate the relationship between FAS, grade, stage and pretreatment with OS and DFS. In the Cox model, used to assess the effect of each variable on OS and DFS while adjusting for the others, only FAS was included as a continuous variable, while all the other variables were considered categorical (reference categories: low grade, stage I, pretreatment = no). The Log-rank test was used to compare the Kaplan–Meier curves.

RESULTS

Clinico-Pathological Features

Of 94 children, 50 were male and 44 female. Mean age at diagnosis was 42.6 months (range: 2–183). In all cases the diagnosis of nephroblastoma and the clinical stage were initially established by means of diagnostic imaging (abdominal ultrasound and computed tomography of chest and abdomen with intravenous contrast).

Patients were treated according to SIOP-6, SIOP-9, and SIOP-93-01 protocols which recommend preoperative CT and vary little in terms of therapeutic agents and duration of treatment [22]. Preoperative CT was administered to 73 patients: 59 with localized tumor received a 2-drug regimen (dactinomycin and vincristine) and 14 with metastatic disease at diagnosis received a 3-drug regimen (dactinomycin, vincristine, and doxorubicin/epirubicin). Previously untreated patients included the remaining 21 children who had undergone surgery first for the following reasons: 6 were less than 6 months of age, 7 had been referred after prior surgery, 4 had a doubtful radiologic diagnosis, and 4 presented as a surgical emergency. Nephrectomy was performed in all patients, with intraoperative rupture occurring in two cases.

Pathologic stage distribution was as follows: 44 stage I, 19 stage II/negative lymph nodes, 7 stage II/positive lymph nodes, 10 stage III, 3 stage IV, and 11 stage V. Microscopically, intermediate risk histology was predominant (77%), while low and high risks represented 9 and 14%, respectively. Postoperative treatment was based on surgical and histopathologic staging, with patients receiving either no further treatment, 2-drug, 3-drug, or 4-drug regimen ± radiotherapy [3].

Twenty-five patients presented one or more recurrences and metastatectomy was performed in 20 of them, with incomplete excision and/or tumor rupture occurring in 5 cases.

At the end of the study, 16 patients had died of disease, 4 were completing therapy, and the remaining 74 were off-therapy and in complete remission, with a mean follow-up of 79 months.

Immunohistochemistry

In the majority of cases, the two observers concurred in grading the percentage of FAS positivity; in a few cases there were minor variations in grading, and the final value was reached after mutual agreement.

Eighty-three percent of tumors expressed FAS, with a variable degree of immunostaining (Table II and Fig. 1). FAS was homogeneously distributed in the cytoplasm of neoplastic cells and showed a granular pattern in intensely stained cells. Generally, the epithelial component was always positive in tubular/cystic structures, but did not stain within pseudoglomeruli. Blastema stained in about half the cases, and less intensely. Mesenchymal cells were negative except for lipoblastic and chondroid differentiated elements, which strongly stained. Anaplastic tumors showed a stronger staining with positivity in all three components. Pretreated tumors generally stained more intensely than previously untreated ones.

TABLE II. FAS Expression According to Grade and Stage

	WTs (=94) 73 + 21 (16)	Negative 9 + 7 (1)	Weak 17 + 5 (2)	Moderate 23 + 6 (4)	Strong 18 + 2 (7)	Very strong 6 + 1 (2)
pStage						
I	44	5 (1) + 3	9 + 3	13 (1) + 3 (1)	7 (3) + 1	—
II−	19	3 + 1	1 + 1	4 (1) + 2	6 (2)	1
II+	7	—	2 (1)	0 + 1	2 (1)	2 (2)
III	10	0 + 3	1 + 1	1	0 + 1	2 + 1
IV	3	—	—	2	—	1
V	11	1	4 (1)	3 (1)	3 (1)	—
Risk grade						
Low	7	1 + 1	—	1 + 3	1	—
Intermediate	74	8 (1) + 6	15 (1) + 4	20 (2) + 2 (1)	13 (5) + 2	3 (1) + 1
High	13	—	2 (1) + 1	2 (1) + 1	4 (2)	3 (1)

Patients (= 94) were divided basing on pretreatment (yes = 73 + no = 21) and immunostaining was graded in five levels (deaths are reported in brackets).

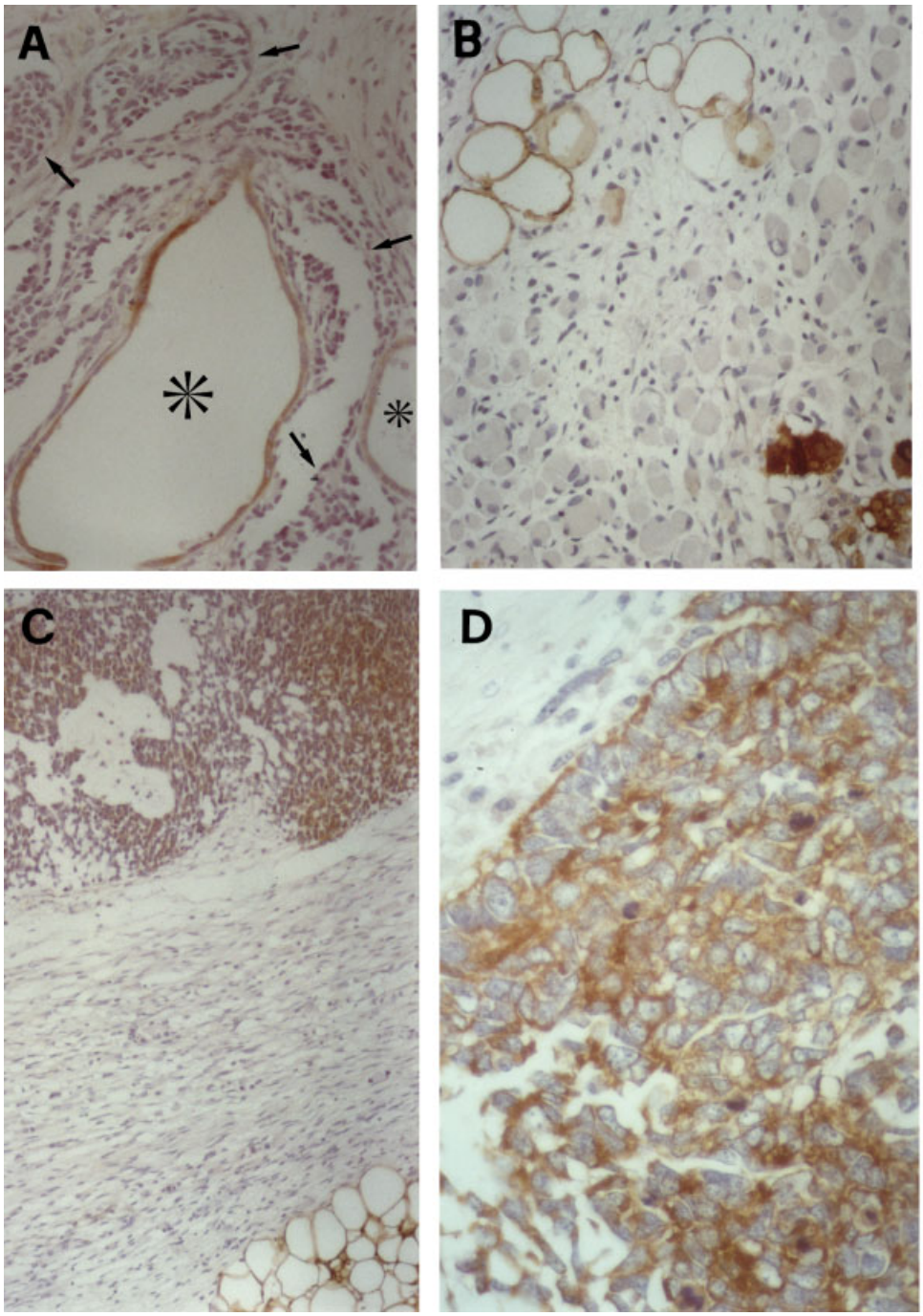

Fig. 1. Anti-FAS immunostaining patterns of positivity. **A**: Anti-FAS antibodies reacted against epithelial component of cystic and tubular structures (asterisks) but not against that of surrounding glomeruloid buds (arrows). **B**: Lipoblastic (right) and more differentiated lipocytic (left) elements resulted intensely positive, while rhabdomyoblastic background was negative. **C**: Diffuse immunostaining of a pretreated blastematous tumor. Thick fibrous pseudocapsule separated it from perirenal fat tissue (bottom) which represented the positive control. **D**: Strong and diffuse immunoreactivity of a high risk (anaplastic) Wilms tumor.

Statistical Results

At univariate analysis (Tau Kendall test), FAS expression showed a statistically significant association with grade ($P = 0.043$). Fisher exact test showed association with stage ($P = 0.029$) but not with pretreatment ($P = 0.059$).

The Kaplan–Meier analysis including all patients disclosed a correlation of FAS with both OS ($P = 0.022$) and DFS ($P < 0.0001$). Similar results were obtained at

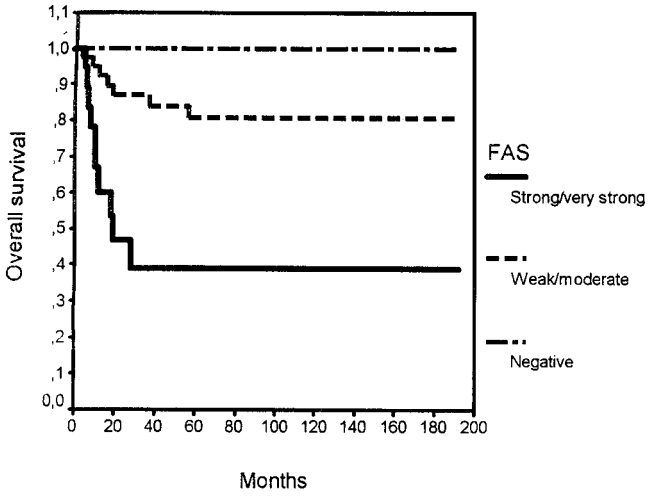

Fig. 2. Overall survival (OS, months) curves stratified by different FAS expression of intermediate-risk nephroblastomas. The OS at 5 years was 100, 81, and 39%, respectively.

separate analysis of the intermediate-risk group: FAS expression determined both OS ($P = 0.004$) and DFS ($P < 0.0001$) (Fig. 2).

At multivariate analysis, the Cox proportional hazard model fitted with FAS expression, stage, grade and pretreatment as independent variables and OS and DFS as dependent variables, disclosed that increasing FAS expression was a statistically significant negative prognostic factor with a hazard rate of 1.6 ($P = 0.05$) and 2.5 ($P < 0.001$), respectively. Moreover, stage was a statistically significant prognostic factor for DFS only, as the hazard of patients with high stage was 4.3-fold the hazard of patients with low stage ($P = 0.006$). The same results were obtained in the intermediate-risk population: increasing FAS expression was a statistically significant negative prognostic factor with a hazard rate of 2.6 ($P = 0.009$) for OS, and with a hazard rate of 3.0 ($P = 0.002$) for DFS. Similarly, for DFS only, patients with high stage showed a hazard rate of 4.1 ($P = 0.002$), as compared with low stage.

DISCUSSION

Wilms tumor is the most common intra-abdominal solid tumor of childhood [1]. Treatment includes chemotherapy and surgical resection for virtually all affected children and additional radiotherapy for those with advanced disease or adverse prognostic features [2,22]. This approach, based on tumor histology and stage, leads to cure rates that are the highest among pediatric solid tumors. Presently the goal is to identify patients at low and intermediate risk for relapse. This will allow a reduction in treatment burden in them and subsequent long-term toxic sequelae. Identification of high-risk patients is important, too, so that attempts to improve cure rates can be made by intensifying therapy.

Morphologic, cytogenetic, cytofluorimetric, and morphometric studies have been performed in order to delineate additional prognostic parameters [1,2]. During the last decade, there have been a number of advances in the understanding of Wilms tumor biology. Its development, for example, involves several genes, including WT1 (located at gene 11p13) and WT2 (located at gene 11p15). Mutations in certain chromosomal regions, most notably 16q and 1p, have been associated with poorer outcome [2]. As a result of such investigations, novel biological markers will be incorporated into therapeutic protocols for Wilms tumor.

Recent studies in adult populations have shown an association between high FAS levels and a poorer prognosis in a substantial subset of breast, colorectal, ovarian, pulmonary, endometrial, and prostate cancers [10–21]. For some of them, FAS expression has resulted to be a reliable prognostic factor [10,11,13,16,18,20].

FAS expression has never been investigated in nephroblastoma and in pediatric tumors in general. In human kidney, activation of FAS is high during fetal life (peak at 20 weeks) [8,9] and progressively decreases to a weak immunohistochemical staining observed in proximal tubules of the adult.

In our study, most tumors expressed FAS to a variable degree and expression increased with higher histologic grade and stage. FAS positivity was correlated with unfavorable outcome both at univariate and multivariate analysis. In addition, we obtained similar results when analyzing the intermediate-risk population (standard histology, which includes about 80% of all WTs) separately. The intermediate risk histology group exhibits heterogeneous histopathologic features, consisting of proliferating blastemal and epithelial cells and a variable amount of stromal component. All histologic subtypes within this group lack the features of anaplastic cells, but it is still unclear whether a subtype is more aggressive than the other and if it is appropriate to include them all within a single category. For this reason, we believe that FAS prognostic strength might be useful in this subgroup, in which some patients unexpectedly do not respond to therapy. As there was no statistically significant difference in FAS expression when comparing patients who received preoperative chemotherapy with those who did not, this parameter could apply equally to both patient groups.

The activation of FAS in normal tissues is complex and is regulated by carbohydrate metabolism, dietary fatty acid intake, sex steroid/thyroid hormones, and insulin [6]. In some cancers, hormones probably drive the activation [10–15,17], but in hormone-receptor negative neoplasms, like nephroblastoma, the mechanism involved in increased FAS expression is still unclear. It might be the consequence of a constitutional dysregulation of a

transcription factor, or could be linked to increased fatty acid demand by intensely proliferating neoplastic cells. As we observed a trend towards more intense immunostaining in pretreated patients (strong/very strong positivity 33% among the pretreated group vs. 14% in the untreated group), one could postulate that increased FAS expression reflected the presence of more aggressive neoplastic clones selected by treatment or that some yet unknown pharmacological stimuli might have induced its activation.

Since cancer cells are dependent on fatty acid synthetic activity and pharmacologic inhibitors of this enzyme are selectively cytotoxic to them [23–28], expression of FAS may also provide a potential target for intervention in the neoplastic process. Recently, FAS has been inhibited in vitro with cerulenin, a potent noncompetitive inhibitor of the enzyme [24]. The result was that neoplastic cells were unable to grow even when physiologic amounts of fatty acids were available. This suggests that the fatty acid synthetic pathway might be a promising target for drug development, since the enzyme appears to be activated in aggressive cancer cells while occurring at low levels within normal cells. Phase I studies await the development of a chemically stable FAS inhibitor, suitable for clinical investigations.

CONCLUSIONS

This study demonstrated that anti-FAS immunostaining was a reliable prognostic indicator in WT and might contribute to an improved stratification of intermediate-risk patients. Moreover, a better understanding of FAS pathway role in nephroblastoma biology might represent a starting-point in the design of new therapeutic strategies for this disease.

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REFERENCES

1. Murphy WM, Beckwith JB, Farrow GM. Tumors in infancy and childhood. In: Tumors of the kidney, bladder and related urinary structures. Washington D.C.: AFIP; 1994. (3° series), fascicle 11:12–92.
2. Wiener JS, Coppes MJ, Ritchey ML. Current concepts in the biology and management of Wilms tumor. J Urol 1998;159:1316–1325.
3. Nephroblastoma Clinical Trial. n° 93-01, Protocol, June 1993. (revised December 1995).
4. Kuhajda FP. Fatty-acid synthase and human cancer: New perspectives on its role in tumor biology. Nutrition 2000;16:202–208.
5. Ohlrogge J, Pollard M, Bao X, et al. Fatty acid synthesis: From CO_2 to functional genomics. Biochem Soc Trans 2000;28:567–573.
6. Latasa MJ, Moon YS, Kim KH, et al. Nutritional regulation of the fatty acid synthase promoter in vivo: Sterol regulatory element binding protein functions through an upstream region containing a sterol regulatory element. Proc Natl Acad Sci USA 2000;97:10619–10624.
7. Weiss L, Hoffman G, Schreiber R, et al. Fatty-acid biosythesis in man, a pathway of minor importance. Biol Chem 1986;367:905–912.
8. Kusakabe T, Maeda M, Hoshi N, et al. Fatty acid synthase is expressed mainly in adult hormone-sensitive cells or cells with high lipid metabolism and in proliferating fetal cells. J Histochem Cytochem 2000;48:613–622.
9. Wilentz RE, Witters LA, Pizer ES. Lipogenic enzymes fatty acid synthase and acetyl-coenzyme A carboxylase are coexpressed with sterol regulatory element binding protein and Ki-67 in fetal tissues. Pediatr Dev Pathol 2000;3:525–531.
10. Alò PL, Visca P, Marci A, et al. Expression of fatty acic synthase (FAS) as a predictor of recurrence in stage I breast carcinoma patients. Cancer 1996;77:474–482.
11. Alò PL, Visca P, Trombetta G, et al. Fatty acid synthase (FAS) predictive strength in poorly differentiated early breast carcinomas. Tumori 1999;85:35–40.
12. Milgraum LZ, Witters LA, Pasternack GR, et al. Enzymes of the fatty acid pathways are highly expressed in in situ breast cancer. Clin Cancer Res 1997;3:2115–2120.
13. Shurbaji MS, Kuhajda FP, Pasternack GR, et al. Immunohistochemical detection of a fatty acic synthase (OA-519) as a predictor of progression in prostate cancer. Hum Pathol 1996;27:917–921.
14. Myers RB, Oelschlager DK, Weiss HL, et al. Fatty acid synthase: An early molecular marker of progression of prostatic adenocarcinoma to androgen independence. J Urol 2001;165:1027–1032.
15. Swinnen JV, Vanderhoydonc F, Elgamal AA, et al. Selective activation of the fatty acid synthesis pathway in human prostate cancer. Int J Cancer 2000;88:176–179.
16. Rashid A, Pizer ES, Moga M, et al. Elevated expression of fatty acid synthase and fatty acid synthetic activity in colorectal neoplasia. Am J Pathol 1997;150:201–208.
17. Pizer ES, Lax S, Kuhajda FP, et al. Fatty acid synthase expression in endometrial carcinoma: Correlation with cell proliferation and hormone receptors. Cancer 1998;83:528–537.
18. Gansler TS, Hardman W, Hunt DA, et al. Increased expression of fatty acid synthase (OA-519) in ovarian neoplasms predicts shorter survival. Hum Pathol 1997;28:686–692.
19. Alò PL, Visca P, Framarino ML, et al. Immunohistochemical study of fatty acid synthase in ovarian neoplasms. Oncol Rep 2000;7:1383–1388.
20. Vlad LD, Axiots CA, Merino MJ, et al. Fatty acid synthase is highly expressed in aggressive thyroid tumors. Mod Pathol 1999;12:70A.
21. Piyathilake CJ, Frost AR, Manne U, et al. The expression of fatty acid synthase (FASE) is an early event in the development and progression of squamous cell carcinoma of the lung. Hum Pathol 2000;31:1068–1073.
22. Suryanarayan K, Marina N. Wilms tumour. Optimal treatment strategies. Drugs 1998;56:597–605.
23. Kuhajda FP, Pizer ES, Li JN, et al. Synthesis and antitumor activity of an inhibitor of fatty acid synthase. Proc Natl Acad Sci USA 2000;97:3450–3454.
24. Pizer ES, Wood FD, Heine HS, et al. Inhibition of fatty acid synthase delays disease progression in a xenograft model of ovarian cancer. Cancer Res 1996;56:1189–1193.
25. Pizer ES, Chrest FJ, Di Giuseppe JA, et al. Pharmacological inhibitors of mammalian fatty acid synthase suppress DNA

replcation and induce apoptosis in tumor cell lines. Cancer Res 1998;58:4611–4615.
26. Furuya Y, Akimoto S, Yasada K, et al. Apoptosis of androgen-indipendent prostate cell line induced by inhibition of fatty acid synthesis. Anticancer Res 1997;17:4589–4593.
27. Pizer ES, Jackisch C, Wood FD, et al. Inhibition of fatty acid synthase induces programmed cell death in human breast cancer cells. Cancer Res 1996;56:2745–2747.
28. Gabrielson EW, Pinn ML, Testa JR, et al. Increased fatty acid synthase is a therapeutic target in mesothelioma. Clin Cancer Res 2001;7:153–157.