Human Pathology (2013) **44**, 1508–1515

ELSEVIER

Human PATHOLOGY

www.elsevier.com/locate/humpath

Original contribution

Expression of the oncofetal protein IGF2BP3 in endometrial clear cell carcinoma: assessment of frequency and significance

Oluwole Fadare MD[a,b,*], Sharon X. Liang MD, PhD[c], Marta A. Crispens MD[b], Howard W. Jones III MD[b], Dineo Khabele MD[b], Katja Gwin MD, PhD[d], Wenxin Zheng MD[e], Khaled Mohammed MD[f], Vinita Parkash MD[g,h], Jonathan L. Hecht MD, PhD[i], Mohamed M. Desouki MD, PhD[a]

[a]*Department of Pathology, Microbiology and Immunology, Vanderbilt University School of Medicine, Nashville, TN 37232, USA*
[b]*Department of Obstetrics and Gynecology, Vanderbilt University School of Medicine, Nashville, TN 37232, USA*
[c]*Department of Pathology and Laboratory Medicine, North Shore–LIJ Health System and Hofstra North Shore–LIJ School of Medicine, New Hyde Park, NY 11030, USA*
[d]*Department of Pathology, University of Chicago, Chicago, IL 60637, USA*
[e]*Department of Pathology and Department of Obstetrics and Gynecology, University of Arizona College of Medicine, Tucson, AZ 85724, USA*
[f]*Department of Gastroenterology, University of Pittsburgh Medical Center, Pittsburgh, PA 15213, USA*
[g]*Department of Pathology, Yale University School of Medicine, New Haven, CT 06510, USA*
[h]*Department of Pathology, Bridgeport Hospital, Bridgeport, CT 06610, USA*
[i]*Department of Pathology, Beth Israel Deaconess Medical Center, and Harvard Medical School, Boston, MA 02215, USA*

Received 5 November 2012; revised 6 December 2012; accepted 7 December 2012

Keywords:
IMP3;
Clear cell carcinoma;
Endometrium;
Insulin-like growth factor-II mRNA-binding protein

Summary Insulin-like growth factor-II messenger RNA–binding protein 3 (IGF2BP3 or IMP3) is a biomarker whose expression has been found to be a negative prognostic factor in several neoplasms including ovarian clear cell carcinoma (CCC). In this study, we analyzed the frequency and clinicopathologic significance of IMP3 expression, as assessed by immunohistochemistry and as scored using a modified H-score system, in a cohort of 50 endometrial CCCs. Cases with scores of 0 to 100, 101 to 200, and 201 to 300 were classified as negative/mildly positive (n = 17), moderately positive (n = 20), and strongly positive (n = 13), respectively. A distinctive pattern of increased staining at the myoinvasive front (relative to the main tumor) was evident in 46% of the cases with evaluable foci of myometrial invasion. Moderate/strong IMP3 staining was associated with a tumor architectural pattern that has been reported to be of poor prognostic significance: at least 10% of the tumor composed of solid architecture or individual infiltrating tumor cells ($P = .01$). Increasing levels of IMP3 expression showed a trend toward decreasing relapse-free survival (RFS; median survival, 75.6, 81.3, and 48.4 months for the negative/mildly, moderately, and strongly positive groups, respectively [$P = .09$]). However, IMP3 expression was not significantly associated with reduced overall survival or RFS in a multivariate

* Corresponding author. Department of Pathology, Microbiology and Immunology, Vanderbilt University Medical Center, MCN C-2310D, 1161 21st Ave S, Nashville, TN 37232, USA.
E-mail address: oluwolefadare@yahoo.com (O. Fadare).

0046-8177/$ – see front matter
http://dx.doi.org/10.1016/j.humpath.2012.12.003

analytic model. The finding in a subset of our cases of increased IMP3 expression at the tumoral myoinvasive front is consistent with a role for IMP3 in invasiveness, as is the trend toward reduced RFS in cases expressing IMP3 at high levels. These preliminary findings suggest that IMP3 expression may be involved in the pathogenesis of CCC and is worthy of further exploration.

1. Introduction

The insulin-like growth factor-II messenger RNA (mRNA)–binding proteins IMP1 (IGF2BP1), IMP2 (IGF2BP2), and IMP3 (IGF2BP3) are an evolutionarily conserved family of mRNA-binding proteins that comprise 2 RNA recognition motifs and 4 K homology (KH) domains [1]. IGF2BPs are involved in the subcytoplasmic localization of mRNAs and hence translational control during embryogenesis [2,3]. Accordingly, they are primarily produced during the early stages of embryogenesis but may be expressed in some organs (such as the ovaries and testes) later in development [1,2]. The *IGF2BP3* gene is located on chromosome 7p11.5, was previously designated as *K homology domain-containing protein overexpressed in cancer*, and was cloned after a screen for differentially expressed genes in pancreatic cancer relative to the normal pancreas [4]. The resultant protein, a 580-amino-acid oncofetal RNA-binding protein (IMP3), has emerged during the last decade as a robust diagnostic marker whose immunohistochemical assessment can be used to reliably distinguish some malignancies from their benign histologic or cytologic mimics and to distinguish between histotypes of selected neoplasms [5]. Furthermore, preliminary lines of evidence indicate that IMP3 expression is a negative prognostic factor and/or a predictor of tumor progression in a remarkably high proportion of neoplasms in which this expression has been evaluated, including colorectal, gastric, renal, mammary, bladder, thyroid, and oral cavity carcinomas; cutaneous melanomas; osteosarcomas; meningiomas; neuroblastomas; and cervical intraepithelial neoplasias [5]. It was recently reported that IMP3 expression is an independent marker of reduced survival in patients with ovarian clear cell carcinomas (CCCs) [6]. Endometrial CCC is a significantly rarer neoplasm, and a comparable analysis of IMP3 expression in CCC has not heretofore been reported. The purpose of this study is to assess the frequency and prognostic significance of IMP3 expression in endometrial CCCs.

2. Materials and methods

2.1. Case selection and review

The 50 cases that comprised the final data set were retrieved from the archived files of multiple institutions. The cases were selected at these institutions by gynecologic pathologists, who all searched for tumors signed out as CCC at their respective institutions, subjected them to secondary review, and selected those cases that they considered to be unequivocally diagnostic of CCC. All cases were subsequently reviewed centrally by a panel of 3 gynecologic pathologists (J.H., V.P., and O.F.). Each panelist reviewed all cases independently. A case was included in the final data set only if at least 2 of the 3 central panelists agreed with a diagnosis of CCC. Nineteen percent of the cases that were in the prereview data set were excluded as non-CCC after central review. The 50 cases that comprised the final data set thus included cases that had been subjected to at least 3 layers of review. These cases included 5 biopsies and 45 hysterectomy specimens. The cases had been used, entirely or in subsets, in previously published studies from this group. Clinical data were extracted from the medical record. This study was approved by the institutional review board at Vanderbilt University (institutional review board no. 12606).

2.2. Immunohistochemistry

IMP3 immunohistochemical analyses were performed on a single section from all 50 cases in a Leica Bond Max immunohistochemical autostainer (Leica Microsystems, Buffalo Grove, IL). The proprietary Leica Epitope Retrieval 2 solution was used to facilitate heat-induced antigen retrieval. Slides were then incubated with the primary antibody, a monoclonal mouse antihuman IMP3, Clone 69.1 (dilution 1:750; Dako, Carpinteria, CA); the Bond Polymer Refine detection system was used for visualization. IMP3 is known to exhibit a predominantly cytoplasmic reactive pattern, although nuclear expression may also be uncommonly seen. Any immunoreactivity (nuclear and/or cytoplasmic) was interpreted as representing positivity. A case of endometrial serous carcinoma with known IMP3 immunoreactivity served as a positive control, whereas a section of normal endometrium in the proliferative phase served as a negative control. Immunohistochemical staining for IMP3 was jointly scored on all cases by 2 authors (O.F. and M.M.D.) using a semiquantitative system that is based on the H index [7]: 3 × percentage of strongly staining cells + 2 × percentage of moderately staining cells + percentage of weakly staining cells, giving "composite scores" that ranged from 0 to 300 (Fig. 1). Given the high frequency of immunoreactivity as outlined later, the cases were classified into 3 groups based on their composite scores. Cases with a