Molecular and Cellular Endocrinology 426 (2016) 22–32

Contents lists available at ScienceDirect

Molecular and Cellular Endocrinology

journal homepage: www.elsevier.com/locate/mce

Evidence for possible role of toll-like receptor 3 mediating virus-induced progression of pituitary adenomas

Xin Zheng [a, 1], Song Li [a, 1], Zhenle Zang [a], Jintao Hu [a], Jiayin An [a], Xiangdong Pei [a], Feng Zhu [c], Weihua Zhang [b, **], Hui Yang [a, *]

[a] Multidisciplinary Center for Pituitary Adenomas of Chongqing, Department of Neurosurgery, Xinqiao Hospital, Third Military Medical University, Chongqing 400037, China
[b] Department of Biology and Biochemistry, College of Natural Sciences and Mathematics, University of Houston, TX, USA
[c] Innovative Drug Research Centre, University of Chongqing, China

ARTICLE INFO

Article history:
Received 30 October 2015
Received in revised form
25 January 2016
Accepted 11 February 2016
Available online 15 February 2016

Keywords:
Pituitary adenoma
Progression
Toll-like receptor 3
Viruses

ABSTRACT

Tumor-related viruses are known to be involved in initiation and progression of certain tumors. However, the relationship between virus and pituitary adenomas (PAs) remains unknown. Here, we investigated infection status of three types of viruses (HPV16, HHV6B and HSV1) and expression level of toll-like receptor 3 (TLR3) in 60 human PA samples. We also determined the role of TLR3 signaling pathway on a PA cell line (GH3). We firstly found that positive rates of HPV16 and HHV6B infection were significantly higher in invasive PA samples than in noninvasive samples ($P < 0.01$). Similarly, TLR3 mRNA and protein expression also increased in invasive PA samples ($P < 0.01$). In vitro analysis indicated that GH3 cell proliferation and survival were enhanced by TLR3 activation, which was accompanied by NF-κB activation. Our data indicate that HPV16 and HHV6B viruses may be involved in promoting the progression of PA by activating the TLR3 signaling pathway.

1. Introduction

Pituitary adenomas (PAs) are usually benign tumors that account for 10–15% of all intracranial tumors (Aflorei and Korbonits, 2014). PAs can be classified according to the type of hypersecretion of pituitary hormones. Clinically non-functioning pituitary adenomas (NFPAs) and growth hormone-secreting pituitary adenomas (GH-PAs) are most common in the PAs whose first-line therapy is surgery (Colao et al., 2011). Despite their histologically benign nature, 25–55% of PAs can infiltrate adjacent structures such as bony parts of the sella turcica, sphenoid sinus and/or cavernous sinus. These adenomas are defined as “invasive PAs” (Meij et al., 2002; Scheithauer et al., 1986; Thapar et al., 1996). Invasive PAs often present a higher Ki-67 index and are related to significant morbidity, poor prognosis, and poor response to the different alternative therapies (Di Ieva et al., 2014). Understanding of the factors affecting tumor invasion is crucial for developing new adjuvant treatments and for prognosis of PA patients.

Viral infection plays an important role in initiation and development of certain types of tumors (Cerwenka and Lanier, 2001). In total, 15–20% of human cancers are estimated to be caused by oncogenic viruses (Butel, 2000). The first human tumor-associated virus was discovered in 1964 and was named Epstein–Barr virus (EBV); this virus was found to be involved in the tumorigenesis of Burkitt's lymphoma, Hodgkin's lymphoma and nasopharyngeal carcinoma (Poreba et al., 2011). DNA viruses (e.g., hepatitis B virus, human papillomaviruses) and RNA viruses (e.g., human T-cell lymphotropic virus 1 and hepatitis C virus) were found to be associated with tumorigenesis (Bergonzini et al., 2010). Accumulating evidence suggests that virus-induced chronic inflammation and oxidative stress could promote tumor development and that chronic inflammation was often mediated by pathogen-associated molecular patterns (Read and Douglas, 2014).

Activation of the Toll-like receptor (TLR) is one of the most common pathogen-associated molecular patterns that play important roles in tumor initiation and progression. The TLR family

* Corresponding author. Multidisciplinary center for pituitary adenomas of Chongqing, Department of Neurosurgery, Xinqiao Hospital, Third Military Medical University, 183 Xinqiao Main Street, Shapingba District, Chongqing, China.
** Corresponding author.
E-mail addresses: wzhang13@uh.edu (W. Zhang), yanghui64@hotmail.com (H. Yang).
[1] Xin Zheng and Song Li contributed equally to this work.

http://dx.doi.org/10.1016/j.mce.2016.02.009
0303-7207/

Table 1
Patient clinical characteristics.

Case	Age (yrs)	Sex	Knosp grade	Hormonal type	Ki-67 (LI ≥ 3%)[a]	HPV16[b]	HHV6B	HSV1
1	46	F	IV	NFPA	+	P	P	N
2	50	F	III	NFPA	+	N	N	P
3	56	F	III	NFPA	+	P	P	N
4	49	F	IV	NFPA	+	P	N	P
5	46	M	III	NFPA	+	P	N	N
6	51	M	III	NFPA	–	N	N	P
7	44	M	IV	NFPA	+	N	P	N
8	51	F	III	NFPA	–	P	N	P
9	47	M	IV	NFPA	+	P	P	N
10	43	F	III	NFPA	+	P	P	P
11	55	M	IV	NFPA	+	N	P	P
12	76	M	III	NFPA	+	N	N	N
13	53	M	IV	NFPA	+	P	P	P
14	47	M	IV	NFPA	+	P	N	P
15	56	F	IV	NFPA	–	P	N	N
16	43	M	II	NFPA	–	N	N	N
17	60	F	I	NFPA	+	N	P	N
18	46	F	II	NFPA	–	N	N	P
19	27	F	I	NFPA	+	P	N	P
20	53	M	I	NFPA	–	N	N	N
21	35	F	II	NFPA	–	P	P	P
22	66	M	I	NFPA	–	P	N	P
23	53	M	II	NFPA	–	P	P	P
24	48	F	I	NFPA	–	P	N	N
25	61	M	I	NFPA	–	N	N	N
26	51	F	I	NFPA	+	N	P	N
27	60	F	II	NFPA	–	N	N	N
28	50	F	I	NFPA	–	N	N	N
29	51	F	I	NFPA	–	N	N	N
30	63	M	I	NFPA	+	N	N	N
31	54	F	III	GH	+	N	P	P
32	70	F	III	GH	+	N	P	P
33	43	M	IV	GH	–	P	N	N
34	39	F	III	GH	+	P	P	P
35	48	M	IV	GH	+	P	N	N
36	46	F	III	GH	+	N	P	P
37	30	F	IV	GH	–	P	N	N
38	43	F	III	GH	+	P	P	P
39	51	M	IV	GH	–	P	N	N
40	46	M	III	GH	+	N	P	P
41	42	M	IV	GH	+	P	P	N
42	42	F	IV	GH	+	P	P	N
43	46	F	IV	GH	+	P	P	N
44	41	F	IV	GH	–	P	N	N
45	46	M	IV	GH	–	P	N	P
46	45	M	II	GH	–	N	N	N
47	45	M	I	GH	–	N	N	N
48	55	F	I	GH	–	P	N	N
49	34	M	I	GH	–	P	N	N
50	46	M	II	GH	–	N	P	N
51	46	M	I	GH	–	N	N	P
52	40	F	I	GH	+	P	P	N
53	44	F	II	GH	–	N	N	P
54	37	M	I	GH	–	N	N	N
55	46	M	II	GH	+	N	P	N
56	37	M	II	GH	–	N	N	P
57	51	F	II	GH	+	N	N	P
58	40	F	I	GH	–	N	N	N
59	37	M	I	GH	–	N	N	N
60	40	M	I	GH	–	N	N	N

[a] "+" indicates Ki-67 LI ≥ 3%; "–" indicates Ki-67 LI < 3%.
[b] "P" indicates positive detection of oncoviruses; "N" indicates negative detection of oncoviruses.

includes 10 members (TLR1-TLR10) in humans (Kawasaki and Kawai, 2014). TLR3, which is widely detected in human tumor cells, recognizes double-stranded RNA from viruses, mammalian RNA associated or released from necrotic cells, and synthetic ligand polyinosinic:polycytidylic acid [Poly(I:C)] (Huang et al., 2008; Amarante and Watanabe, 2010). TLR3 expression is related to dedifferentiated status, worse prognosis, lymph node invasion, recurrence and higher tumor metastasis probability (Chuang et al., 2012; Saint-Jean et al., 2011).

In this study, we evaluated the infection status of three oncoviruses (HPV16, HSV1, and HHV6B) and analyzed the expression of TLR3 mRNA and protein in PA tissues. Furthermore, we determined whether activation of the TLR3 signaling pathway by viral mimic Poly(I:C) could affect the proliferation, apoptosis, and invasion of as well as cytokine production by a PA cell line in vitro.