Original paper

EGOT lncRNA in head and neck squamous cell carcinomas

Tomasz Kolenda[1,2,3], Magda Kopczyńska[1,2*], Kacper Guglas[1,2,3*], Anna Teresiak[1], Renata Bliźniak[1], Izabela Łasińska[4], Jacek Mackiewicz[2,5,6], Katarzyna Lamperska[1]

*These authors have contributed equally to this work.

[1]Laboratory of Cancer Genetics, Greater Poland Cancer Centre, Poznan, Poland
[2]Department of Cancer Immunology, Chair of Medical Biotechnology, Poznan University of Medical Sciences, Poznan, Poland
[3]Postgraduate School of Molecular Medicine, Medical University of Warsaw, Warsaw, Poland
[4]Department of Medical and Experimental Oncology, Heliodor Swiecicki Clinical Hospital, Poznan University of Medical Sciences, Poznan, Poland
[5]Department of Biology and Environmental Sciences, Poznan University of Medical Sciences, Poznan, Poland
[6]Department of Diagnostics and Cancer Immunology, Greater Poland Cancer Centre, Poznan, Poland

Head and neck squamous cell carcinomas (HNSCCs) are one of the most challenging cancers to cure. In this study, we focused on eosinophil granule ontogeny transcript (EGOT), a transcriptional regulator of granule protein expression during eosinophil development that has been previously associated with cancers. Expression levels of EGOT and other selected genes as well as clinical pathology data from HNSCC samples, were obtained from The Cancer Genome Atlas (TCGA) and analysed using GraphPad Prism 5. Our results indicated that the expression of EGOT is slightly down-regulated in HNSCC, depending on tumour grade and location, and is only up-regulated in grade 4 tumours and those located in the pharynx. EGOT expression levels were found to vary according to age, N-stage, grade, lymph node dissection and human papillomavirus (HPV) infection. Patients with higher levels of EGOT expression have longer disease-free survival and overall survival outcomes. Further analysis revealed that EGOT targets are associated with cell division, proliferation, protein modification, drug response and cell motility. Taken together, our findings suggest that the EGOT is involved in the progression of HSNCC and seems particularly associated with virus-related forms of HNSCC.

Key words: eosinophil granule ontogeny transcript, lncRNA, head and neck cancers, biomarker.

Introduction

Head and neck squamous cell carcinoma (HNSCC) is the fifth most common cancer and the sixth most common cause of cancer-related mortality worldwide. Tobacco smoking, alcohol consumption and human papillomavirus (HPV) infection are the main causes of these malignancies [1, 2]. Some progress has been made towards effectively treating HNSCC, however, it remains largely unsatisfactory because of still high mortality [3, 4]. Biomarkers that have been associated with different treatment responses are needed to better combat this deadly disease.

Previous studies have correlated the regulation of microRNAs and long non-coding RNAs (lncRNAs) with tumour progression, lymph node metastases and poor prognosis in HNSCC. lncRNAs are a class of functional RNA molecules over 200-nucleotides long that modulate the activity of transcription factors and regulate changes in the chromatin structure despite not being translated into proteins. Previous studies suggest that lncRNAs have the potential to greatly improve the diagnosis, prognosis and targeted treatment of HNSCC [5, 6, 7, 8].

In this study, we focused on the expression of the eosinophil granule ontogeny transcript (EGOT) lncRNA. Human EGOT is located on the antisense strand of the intron of the ITPR1 gene. It has two known isoforms – EGO-A (unspliced) and EGO-B (spliced) – that share the same transcriptional start site and both are polyadenylated. EGOT regulates eosinophil granule protein expression during eosinophil cells developmental process and functions as a non-coding RNA [9, 10]. Numerous recent studies have been dedicated to understanding the role of EGOT in glioma [11], breast cancer [12], gastric cancer [13], haematological malignancies [14] and in viral infections [15, 16, 17] and in cardiology [18, 19]. However, the function of lncRNA EGOT remains unknown in HNSCC. Therefore, we used data made available by The Cancer Genome Atlas (TCGA) to further characterize the role of EGOT in the biology of HNSCC and determine its utility as a new biomarker in clinical practice.

Material and methods

TCGA data

The TCGA expression data for lncRNA EGOT and other selected genes was downloaded from cBioPortal (Head and Neck Squamous Cell Carcinoma, TCGA, Provisional, 530-sample dataset) [20]. Clinical data on tumour and healthy control samples were obtained from the UALCAN database (http://ualcan.path.uab.edu) [21]. All data are available online and access is unrestricted.

Data analysis

The clinical pathology parameters analysed for associations with EGOT expression levels in all localizations of the HNSCC samples include age (below vs. above 61 years), sex (women vs. men), T-stage (T1 + T2 vs. T3 + T4), N-stage (N0 + N1 vs. N2 + N3), cancer grade (G1 + G2 vs. G3 + G4), cancer stage (I + II vs. III + IV), HPV p16 marker (negative vs positive), perineural invasion (negative vs positive), angiolymphatic invasion (negative vs positive), disease surgical margin status (negative vs positive) and lymphoid neck dissection status (negative vs positive). Next, subgroups using the median of expression level of EGOT as cut-off: 1) EGOT low and 2) EGOT high were generated. Disease-free survival (DFS) and overall survival (OS) were determined in these subgroups during 180 and 210 months, respectively.

Target prediction

EGOT target prediction was performed using an online tool (http://rtools.cbrc.jp/cgi-bin/RNARNA/index.pl) [22]. Expression levels of predicted genes were compared between the EGOT low- and high-expression groups. Next, the identified target genes were classified according to biological process and cellular pathway using the Functional Annotation Tool from the DAVID 6.7 Bioinformatics Resource [23].

Statistical analysis

All statistical analyses were performed using GraphPad Prism 5 (GraphPad, San Diego, CA, USA). The Shapiro-Wilk normality test, t-test and Mann-Whitney U test were used for EGOT level depending on clinical parameters and for genes expression depending on EGOT subgroups. The expression level of EGOT depending on the cancer locations was checked using one-way ANOVA obtained using Dunn's multiple comparisons test. For DSF and OS analyses, the Log-Rank (Mantel-Cox) test was used and median survival, Hazard Ratio (Mantel-Haenszel; HR) and 95% Confidence Interval (CI) of ratio were calculated. In all analyses, $p < 0.05$ was used to determine statistical significance.

Results

Using the TCGA data available from the cBioportal and UALCAN databases, the expression level of EGOT was evaluated in HNSCC tissue ($n = 520$) and healthy control ($n = 44$) samples. We found that EGOT expression depends on the grade and localization of the HNSCC. According to the database, the expression of EGOT was slightly down-regulated in HNSCC. However, no significant difference was found in EGOT expression of the cancer and controls samples (median expression 0.097 vs. 0.221 transcripts per million respectively; $p = 0.5143$; Fig. 1A). An analysis of cancer stage (1-4) also failed to find any significant differences ($p > 0.05$; Fig. 1B). However, notable differences were observed in terms of tumour grade. Relative to healthy controls, EGOT expression was down-regulated in grade 1 ($p = 0.00002$), unchanged in grades 2-3 ($p = 0.0813$ and $p = 0.2456$, respectively) and significantly up-regulated in grade 4 ($p = 0.0049$; Fig. 1C).

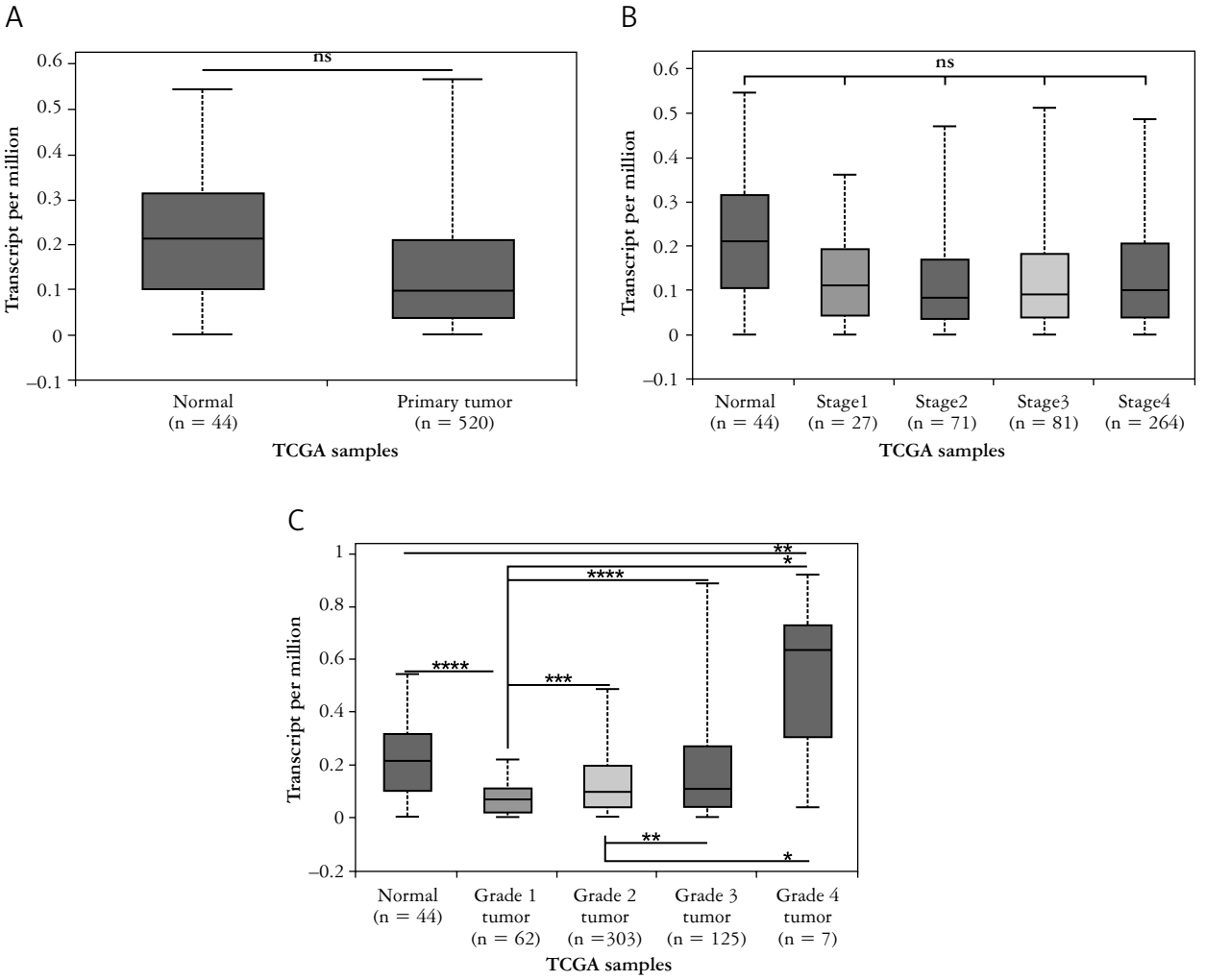

Fig. 1A-C. Expression level of EGOT in HNSCC. A) Expression in normal (healthy) tissue and primary tumour. B) Expression in disease stages 1-4. C) Expression in cancer grades 1-4. Graphs from UALCAN database, modified; paired t-test; the graphs represent the median of the value presented as transcripts per million; * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, **** $p < 0.0001$

The samples from patients with HNSCC (n = 522) were divided into three groups according to the National Institutes of Health (NIH) classification of tumour localization: oral cavity (n = 316), pharynx (n = 90) and larynx (n = 116). Expression analysis revealed that EGOT is down-regulated in tumours from the oral cavity (–0.4228 ±0.02226; $p < 0.0001$) and larynx (–0.4097 ±0.05293; $p = 0.0001$) compared to the pharynx (0.0566 ±0.1692). No differences were observed between the oral cavity and the larynx ($p = 0.2468$; Fig. 2).

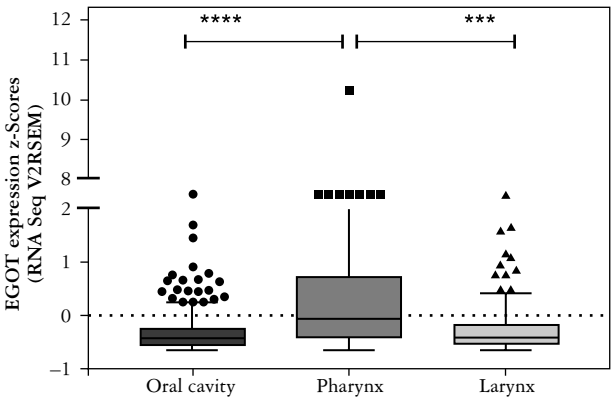

Fig. 2. The expression level of EGOT in the oral cavity (n = 316), pharynx (n = 90) and larynx (n = 116) locations of HNSCC; one-way ANOVA obtained using Dunn's multiple comparisons test; *** $p < 0.001$, **** $p < 0.0001$

EGOT levels differ depending on clinical pathology parameters

Next, EGOT expression levels were analysed according to clinical pathology parameters. Significant differences were found in terms of patients'

Table I. EGOT expression levels and clinical pathology parameters for all three HSNCC localizations. t-test; $p < 0.05$ considered as significant

Parameter	Group	Mean ± SEM	n	p-value
Age	< 61	–0.07733 ±0.05089	280	
	> 61	–0.2614 ±0.05225	240	0.0003
Sex	Female	–0.2015 ±0.09194	137	
	Male	–0.1496 ±0.03740	384	0.0948
Alcohol	Yes	–0.1534 ±0.03914	348	
	No	–0.2077 ±0.07647	162	0.0756
Smoking	Yes	–0,1821 ±0,04264	394	
	No	–0.1599 ±0.06174	334	0.7493
Cancer stage	I + II	–0.2645 ±0.04684	98	
	III + IV	–0.1959 ±0.04459	348	0.7242
T Stage	T1 + T2	–0.1406 ±0.05588	184	
	T3 + T4	–0.2352 ±0.04911	274	0.1700
N Stage	N0 + N1	–0.2603 ±0.02764	327	
	N2 + N3	0.03429 ±0.09318	172	0.0173
Grade	G1 + G2	–0.2483 ±0.03924	367	
	G3 + G4	–0.02305 ±0.07696	132	0.0111
Perineural invasion	Positive	–0.2247 ±0.07497	168	
	Negative	–0.1908 ±0.04764	195	0.5186
Lymph Node Neck Dissection	Positive	–0.2072 ±0.03856	421	
	Negative	0.04041 ±0.1012	97	0.0055
Angiolymphatic invasion	Positive	–0.1415 ±0.07194	124	
	Negative	–0.2289 ±0.05575	225	0.5111
Disease surgical margin status	Positive	–0.002577 ±0.1163	61	
	Negative	–0.2549 ±0.03000	342	0.0613
HPV p16 status	Positive	0.7089 ±0.2262	39	
	Negative	–0.3351 ±0.04416	72	< 0.0001

age ($p = 0.0003$), N-stage ($p = 0.0173$), grade ($p = 0.0111$), lymph node neck dissection ($p = 0.0053$) and were also associated with HPV p16 status ($p < 0.0001$); however, there were no differences in terms of sex ($p = 0.0948$), alcohol consumption ($p = 0.0756$), tobacco smoking ($p = 0.7493$), cancer stage ($p = 0.7245$), T-stage ($p = 0.1699$), angiolymphatic ($p = 0.5111$) and perineural invasions ($p = 0.5186$) nor disease surgical margin status ($p = 0.0612$). These data are summarised in Table I.

EGOT expression levels influence on DFS and OS

The HNSCC samples ($n = 522$) were divided into two groups according to EGOT expression using the median of EGOT expression level as a cut-off. The low expression group was defined as expression levels to –0.392 ($n = 261$) and the high expression group included all of the samples with expression levels above –0.392 ($n = 261$). We found that patients with high EGOT expression levels had longer median DFS periods than those with low levels – 71.22 vs 46.81 months respectively ($p = 0.0452$; HR = 0.7128, 95% CI: 0.5117-0.9931; Fig. 3A). Furthermore, patients in the high expression group had longer median OS times than the low expression group – 46.81 vs. 71.22 months ($p = 0.0096$; HR = 0.7008, 95% CI: 0.5352-0.9175; Fig. 3B).

Predicted EGOT targets and their function

Next, we used the available database to predict lncRNA-RNA interactions in human transcriptome for EGOT. We found 300 genes with a sumenergy of interaction between –26374.2 and –2838.8 and compared their expression levels in the low and high expression groups. Analysis using the DAVID

Bioinformatics Resource revealed that, among patients with high EGOT expression levels, 107 genes were up-regulated. These genes are associated with the regulation of many cellular processes (e.g., differentiation, adhesion, development, cell communication, signal transduction, division and proliferation), protein phosphorylation and other modifications, cellular component organization, cellular homeostasis, drug response and cell motility. The 17 down-regulated genes indicated in the analysis were associated with cell cytoskeleton and filaments, localization/binding, cellular transport and protein activity (Tables II and III).

Discussion

Poor prognosis and high resistance to radio- and chemotherapy are characteristic of HNSCC. Various treatment strategies have been employed in the past couple of decades but none have been great breakthroughs. Improved clinical outcomes will occur once molecular diagnostics and personalized treatments are determined for the different sub-types of this cancer [3, 4]. To date, great effort has been dedicated to the discovery of clinical biomarkers, particularly in oncology, and lncRNAs have been identified as one of the new promising classes of molecules [7, 8]. A previous study investigated the role of lncRNAs, including HOTAIR, UCA1, LET, MEG3, MALAT1, H19 and NAG7, in the biology of HNSCC in addition to their suitability as biomarkers [7]. This study as the first revealed the biological role of lncRNA EGOT in HNSCC. We found that EGOT expression levels depend on tumour grade and location. EGOT is slightly down-regulated in lower grades (1-3) relative to healthy tissue but up-regulated in the case of grade 4. The expression of EGOT is lower in the oral cavity and larynx but higher in the pharynx. Similarly, Xu *et al*. found that EGOT expression was lower in breast cancer cells compared to non-cancerous samples and varies according to the molecular subtypes of breast cancer. Furthermore, they showed that low EGOT expression levels significantly correlate positively with tumour size, lymph node metastasis and Ki-67 expression. This evidence shows that the down-regulation of EGOT is involved in the progression of more invasive types of cancer [12]. In glioma, the expression of EGOT is significantly lower in the cancer than in the adjacent non-cancerous tissues [11]. An *in vitro* study determined that the over-expression of EGOT inhibits cell proliferation and migration and promotes cell apoptosis by increasing protein expression levels of caspase-3, caspase-9 and cytochrome c in U251 and U87 glioma cell lines [11].

Similarly, in renal cell carcinoma, the expression of EGOT is down-regulated in tumour samples compared to paired, healthy tissues. *In vitro* study found that the up-regulation of EGOT expression suppresses proliferation, migration and invasion and induces apoptosis in 786-O and ACHN renal cell lines [24].

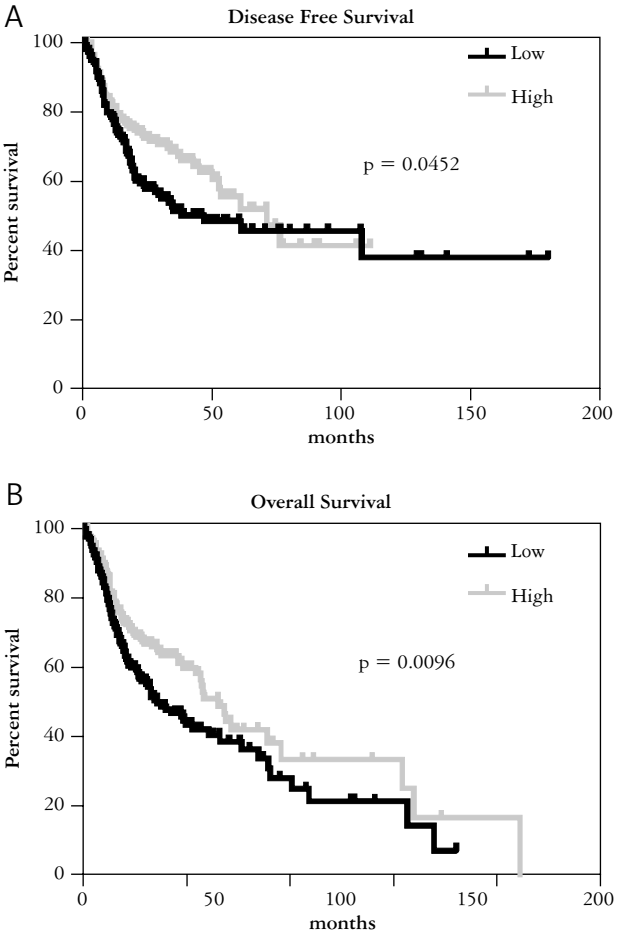

Fig. 3A-B. Association between expression level and patient outcome. Disease-free survival (A) and overall survival (B) in cases of HNSCC with low and high EGOT expression levels

Taken together, these results suggest that EGOT serves as a suppressor gene.

Surprisingly, our analysis of clinical pathology parameters in patients with HNSCC indicated that a low expression level of EGOT is observed in the group of patients with lower N-stage, lower grade, with a lymph node neck dissection and with higher age of patients. However, some evidence has shown that EGOT might also have oncogenic properties. For example, a study of samples from patients with gastric carcinoma and an MKN-45 gastric cancer cell line revealed that EGOT is up-regulated in this cancer and high expression levels are associated with lymphatic metastasis and higher TNM stage. Furthermore, down-regulated EGOT expression *in vitro* results in the inhibition of the hedgehog signalling pathway, cell proliferation and cycle progression arrest in the case of gastric cancer cell line [13].

Table II. The expression level of predicted targets of EGOT in the group of HNSCC patients with high and low expression of EGOT. T-test; p-value < 0.05 considered as significant

Target	p-value	High vs. Low	Target	p-value	High vs. Low
MUC16	0.0006	UP	NSD3	< 0.0001	UP
SYNE1	0.0254	UP	FRK	< 0.0001	UP
NEAT1	0.0003	UP	FAT4	0.0007	UP
DNAJC10	0.0213	UP	HERC1	0.0009	UP
ABI2	0.0033	UP	MBNL3	0.0159	UP
PCLO	< 0.0001	UP	TENM1	0.0383	UP
CNKSR3	0.011	UP	RYR3	0.0037	UP
KCNJ6	0.0076	UP	CBX5	< 0.0001	UP
MUC4	< 0.0001	UP	ALMS1	0.0005	UP
XKR4	< 0.0001	UP	PSD3	< 0.0001	UP
FAT3	< 0.0001	UP	LIFR	0.0003	UP
KSR2	0.0012	UP	BRWD3	0.008	UP
ABCA13	< 0.0001	UP	ATM	0.0014	UP
MUC17	0.0005	UP	CLMN	0.0035	UP
USH2A	< 0.0001	UP	ATRX	0.0159	UP
ONECUT2	< 0.0001	UP	TEX15	< 0.0001	UP
PPIP5K2	0.0041	UP	PGAP1	0.0002	UP
KIAA1109	0.0042	UP	ANKRD12	0.0004	UP
KCTD16	0.0026	UP	MON2	0.001	UP
INO80D	0.0103	UP	SMAD2	0.0007	UP
AKAP9	0.0026	UP	NF1	0.0328	UP
CEP350	0.018	UP	B3GALT5	< 0.0001	UP
TTL	0.0462	UP	DCHS2	0.0001	UP
SOD2	0.0076	UP	SCAI	< 0.0001	UP
USF3	0.0189	UP	EPG5	0.0421	UP
HYDIN	0.0089	UP	ERBB4	0.0036	UP
DNAH11	< 0.0001	UP	APC	0.0057	UP
KMT2A	0.0378	UP	NEXMIF	< 0.0001	UP
CFLAR	0.0132	UP	GOLGB1	0.0002	UP
RYR2	0.0063	UP	PTBP2	< 0.0001	UP
DMD	< 0.0001	UP	NBEA	< 0.0001	UP
TNR	0.0246	UP	RSF1	0.0062	UP
CAMK4	0.0243	UP	FAXC	0.0012	UP
AFF2	0.0309	UP	SLC5A3	0.0412	UP
ENTPD1	0.0205	UP	MED28	0.0036	UP
UNC80	0.0064	UP	VWC2	0.0181	UP
HOOK3	0.0036	UP	SLC1A2	< 0.0001	UP
ARFGEF3	0.0003	UP	CENPF	0.0111	UP
DNAH8	0.0019	UP	ACVR2B	0.0003	UP
PLXNA4	0.0429	UP	ZBTB8B	0.0025	UP
NUFIP2	0.0213	UP	ASXL3	0.0006	UP
SLC24A2	0.0495	UP	DBT	0.0205	UP

Table II. Cont.

Target	p-value	High vs. Low
BNC2	0.0295	UP
ZC3H6	< 0.0001	UP
GPRIN3	0.0267	UP
BRWD1	0.0002	UP
LONRF2	0.0016	UP
PPP1R12B	< 0.0001	UP
PHC3	0.0162	UP
PGR	0.0108	UP
DNAH5	0.0083	UP
DNAH6	< 0.0001	UP
FUT9	< 0.0001	UP
MGA	0.0029	UP
CPLANE1	0.004	UP
INTU	< 0.0001	UP
AGO1	< 0.0001	UP
DNAH7	< 0.0001	UP
PRLR	0.0248	UP
PLCE1	0.0173	UP
KLF12	0.0029	UP
SESTD1	0.0074	UP
DMXL1	0.0105	UP
CFAP44	< 0.0001	UP
CEP290	0.0211	UP
MACF1	0.0032	DOWN
SUGT1	0.002	DOWN
LYST	0.0332	DOWN
FZD3	0.0025	DOWN
PTCHD1	0.014	DOWN
RNF217	0.0022	DOWN
RBM28	0.022	DOWN
FAM83F	0.0028	DOWN
AHNAK2	0.0068	DOWN
ENAH	0.0005	DOWN
MYO5A	< 0.0001	DOWN
TTC39B	0.031	DOWN
PTPRT	0.0023	DOWN
VPS53	0.0009	DOWN
NAV1	< 0.0001	DOWN
PTPN4	0.0051	DOWN
CUX1	0.041	DOWN

We observed that patients with high EGOT expression levels have better prognoses (longer DFS and OS) than those to those with low expression. The same association was identified in breast cancer patients: lower EGOT expression levels are indicators of lower OS times [12]. However, the opposite pattern was observed for gastric cancer in which higher EGOT expression was associated with shorter survival times [13].

Analysis of the predicted EGOT targets indicated an association between this lncRNA and mRNAs the of genes connected with many important cellular processes. We analysed the changes in these genes between two groups of patients divided according to expression level of EGOT. Our results showed that the genes that were up-regulated in the high expression group are involved in the regulation of cellular processes (differentiation, adhesion, developmental process, cell communication, signal transduction, division and proliferation), protein phosphorylation and other modifications, cellular component organization, cellular homeostasis, drug response and cell motility. The genes that were down-regulated in this group related to cell cytoskeleton and filaments, localization/binding, cellular transport and protein activity. The changes in these processes have either an indirect or direct influence on the treatment response and survival of patients with HNSCC.

It must also be noted that the location of HNSCC is a crucial clinical factor in terms of treatment strategy and survival prediction. The observed up-regulation of EGOT expression in the pharyngeal cancers is probably due to HPV infection, which is characteristic of the oropharynx (tonsils and base of tongue) but is sometimes also associated with other locations [25]. Indeed, our observations confirmed that EGOT expression is mostly up-regulated in HPV p16 positive HNSCC. It has been shown that the expression of some other lncRNAs is associated with viral infections [15, 26]. It has also been shown that EGOT expression is up-regulated in HPV positive HNSCC [17] but no role and exact mechanism for its involvement in HPV infection was proposed [17]. Carnero *et al*. found that the high levels of EGOT expression are required for hepatitis C virus replication in samples from patients with hepatocarcinoma (HCC) and in HCC cell lines; moreover, cells with lower EGOT expression produced fewer viral genomes [15]. Cytoplasmatic viral replication is probably required to induce EGOT expression and EGOT level dramatically decreases after viral inhibition in the cells e.g., by exposure to sofosbuvir, daclatasvir or ribavirin [15]. The expression of EGOT is up-regulated in response to pathogen-associated molecular patterns (dsRNA or synthetic analogues and viral RNA) and upon

Table III. Classification of the predicted targets of EGOT changed in HNSCC patients (p-value < 0.05) into specific biological processes and cellular pathways based on DAVID 6.7 database

Up-regulated genes			
GO identifier	Process	p-value	Number of genes
GO:0045595	regulation of cell differentiation	9.82E-5	12
GO:0030155	regulation of cell adhesion	1.248E-4	7
GO:0051239	regulation of multicellular organismal process	2.16E-4	16
GO:0050793	regulation of developmental process	3.88E-4	13
GO:0006468	protein amino acid phosphorylation	0.001	12
GO:0045841	negative regulation of mitotic metaphase/anaphase transition	0.001	3
GO:0007094	mitotic cell cycle spindle assembly checkpoint	0.001	3
GO:0016310	phosphorylation	0.001	13
GO:0048523	negative regulation of cellular process	0.001	20
GO:0045839	negative regulation of mitosis	0.002	3
GO:0051784	negative regulation of nuclear division	0.002	3
GO:0010810	regulation of cell-substrate adhesion	0.002	4
GO:0048519	negative regulation of biological process	0.002	21
GO:0007017	microtubule-based process	0.003	7
GO:0042592	homeostatic process	0.003	12
GO:0048468	cell development	0.003	11
GO:0030154	microtubule-based movement	0.003	5
GO:0007018	cell differentiation	0.004	19
GO:0016043	cellular component organization	0.004	25
GO:0030030	cell projection organization	0.004	8
GO:0019725	cellular homeostasis	0.004	9
GO:0048869	cellular developmental process	0.006	14
GO:0030071	regulation of mitotic metaphase/anaphase transition	0.006	3
GO:0006464	protein modification process	0.006	17
GO:0050678	regulation of epithelial cell proliferation	0.007	4
GO:0045596	negative regulation of cell differentiation	0.007	6
GO:0042493	response to drug	0.007	6
GO:0010948	negative regulation of cell cycle process	0.008	3
GO:0001952	regulation of cell-matrix adhesion	0.010	3
GO:0045786	negative regulation of cell cycle	0.010	4
GO:0010648	negative regulation of cell communication	0.013	6
GO:0010646	regulation of cell communication	0.013	13
GO:0030334	regulation of cell migration	0.015	5
GO:0051093	negative regulation of developmental process	0.015	6
GO:0006996	organelle organization	0.016	15
GO:0032879	regulation of localization	0.021	9
GO:0040012	regulation of locomotion	0.023	5
GO:0051270	regulation of cell motion	0.023	5
GO:0007093	mitotic cell cycle checkpoint	0.024	3
GO:0009966	regulation of signal transduction	0.026	11
GO:0060284	regulation of cell development	0.029	5

Table III. Cont.

Up-regulated genes			
GO identifier	**Process**	**p-value**	**Number of genes**
GO:0051674	localization of cell	0.030	6
GO:0048870	cell motility	0.030	6
GO:0009987	cellular process	0.036	68
GO:0051641	cellular localization	0.036	11
Down-regulated genes			
GO identifier	**Process**	**p-value**	**Number of genes**
GO:0046907	intracellular transport	0.002	5
GO:0051649	establishment of localization in cell	0.006	5
GO:0051641	cellular localization	0.008	5
GO:0030705	cytoskeleton-dependent intracellular transport	0.046	2
GO:0006810	transport	0.076	6
GO:0051234	establishment of localization	0.079	6
GO:0043228	non-membrane-bounded organelle	0.003	8
GO:0043232	intracellular non-membrane-bounded organelle	0.003	8
GO:0005856	cytoskeleton	0.005	6
GO:0015630	microtubule cytoskeleton	0.082	3
GO:0030055	cell-substrate junction	0.094	2
GO:0008092	cytoskeletal protein binding	0.001	5
GO:0003779	actin binding	0.040	3
GO:0051015	actin filament binding	0.051	2
GO:0004725	protein tyrosine phosphatase activity	0.098	2
GO:0008092	cytoskeletal protein binding	3.90E-4	5
GO:0003779	actin binding	0.024	3
GO:0004725	protein tyrosine phosphatase activity	0.077	2

TNFα (tumour necrosis factor α) treatment. TNFα induces NF-kB (nuclear factor-κB), which likely stimulates EGOT expression by binding to its promoter in the case of liver cells [15].

Our analysis suggests that EGOT is involved in the progression of HNSCC. Furthermore, it seems likely that the role of EGOT is connected to HPV infection, given the association between high EGOT expression levels and pharyngeal tumours, younger patients, better DFS and OS and p16 expression, as these are all characteristic of HPV positive HNSCC cases. We supposed that EGOT could be potentially a new biomarker of HPV infection and probably has important role in viral response and biology of HPV positive HNSCC cancers.

Availability of data and materials section

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request. Raw data are available on the cBioPortal and UALCAN databases.

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The authors declare no conflict of interest.

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Address for correspondence

Tomasz Kolenda
Laboratory of Cancer Genetics
Greater Poland Cancer Centre
Garbary 15
61-866 Poznan, Poland
e-mail: kolenda.tomek@gmail.com