Genomics Data 5 (2015) 9–12

Contents lists available at ScienceDirect

Genomics Data

journal homepage: http://www.journals.elsevier.com/genomics-data/

Data in Brief

Gene expression analysis of laryngeal squamous cell carcinoma

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ARTICLE INFO

Article history:
Received 9 April 2015
Received in revised form 27 April 2015
Accepted 29 April 2015
Available online 8 May 2015

Keywords:
Laryngeal squamous cell carcinoma tumor
Microarray
Gene expression profile

ABSTRACT

Laryngeal squamous cell carcinoma (LSCC) is one of the most common malignancies of the head and neck tumors Zhang et al., 2013 [1]). Previous studies have associated its occurrence with social activities, such as tobacco and alcohol consumption (Hashibe et al., 2007a [2]; Hashibe et al., 2007b [3]; Shangina et al., 2006 [4]). Here, we performed a genome-wide gene expression profiling in thirty-one patients positively diagnosed for LSCC, in order to investigate new targets involved in tumorigenesis.

Specifications	
Organism/cell line/tissue	*Homo sapiens*
Sequencer or array type	Whole Human Genome Oligo Microarray chips (Agilent, G4112F, USA)
Data format	Raw: CEL files, normalized data: SOFT, MINIML and TXT
Experimental factors	Twenty nine samples of laryngeal squamous cell carcinoma tumor vs. thirteen adjacent non-neoplastic tissue
Experimental features	We performed a transcriptome analysis in 31 LSCC patients in order to identify new targets involved in tumorigenesis.
Consent	Informed consent was obtained from all patients included in the study.
Sample source location	Ribeirao Preto, Sao Paulo, Brazil

1. Direct link to deposited data

http://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE59102.

2. Experimental design, materials and methods

2.1. Study population and clinical data

All the samples were collected from patients undergoing surgical ablation of larynx squamous cell carcinoma at the Head and Neck Surgery Division of the Department of Ophthalmology, Otorhinolaryngology and Head & Neck of Ribeirao Preto Medical School, USP (Brazil). Inclusion criteria were: histopathological diagnosis of LSCC, elective surgeries for LSCC in patients without previous treatments and patients' allowance to donate part of their tumor for genetic studies. Exclusion criteria were: doubtful diagnosis of LSCC, unavailable post-surgical follow-up, patients without complete clinical data or signed agreement for collection of samples.

A total of twenty nine LSCC tumor samples and thirteen adjacent non-neoplastic tissues proceed for the transcriptome analysis. Clinical information and TNM staging classification of the LSCC patients can be found in Table 1 and Supplemental Table 1. In our study most of the patients were male (96.8%), all of them were smokers and alcoholic, including both current and former. Thirty five percent of the tumors were originated from glottis, followed by larynx (29.0%) and supraglottis (22.6%). Patients were further classified according to the TNM system: 48.4% of the patients

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http://dx.doi.org/10.1016/j.gdata.2015.04.024

Table 1
Clinical information of LSCC patients.

Characteristics	Staging		Total ($n = 31$)
	Early ($n = 15$)	Late ($n = 16$)	
Mean ages			
Year (min–max)	60 (40–78)	62.2 (49–83)	61.1
Gender			
Male	14 (93.3%)	16 (100%)	30 (96.8%)
Female	1 (7.7%)	0 (0%)	1 (3.2%)
Smoking (%)			
Smoker	11 (73.3%)	6 (37.5%)	17 (54.8%)
Former smoker	4 (26.7%)	10 (62.5%)	14 (45.2%)
Non-smoker	0 (0.0%)	0 (0.0%)	0 (0.0%)
Alcoholism (%)			
Alcoholic	10 (66.7%)	7 (43.75%)	17 (54.8%)
Former alcoholic	5 (33.3%)	9 (56.25%)	14 (45.2%)
Non-alcoholic	0 (0.0%)	0 (0.0%)	0 (0.0%)
Tumor site			
Glottis	6 (40%0)	5 (31.3%)	11 (35.5%)
Larynx	3 (20.0%)	6 (37.5%)	9 (29.0%)
Supraglottis	3 (20.0%)	4 (25.0%)	7 (22.6%)
Subglottis & oropharynx	1 (6.7%)	0 (0%)	1 (3.2%)
Glottis & supraglottis	0 (0%)	1 (6.3%)	1 (3.2%)
Laryngopharynx	1 (6.7%)	0 (0%)	1 (3.2%)
Epiglottis	1 (6.7%)	0 (0%)	1 (3.2%)
Tumor relapse	4 (26.7%)	4 (25.0%)	8 (25.8%)
Metastasis	1 (6.7%)	2 (12.5%)	3 (9.7%)
Cured?			
Yes	7 (46.7%)	9 (56.3%)	16 (51.6%)
No	8 (53.3%)	7 (43.8%)	15 (48.4%)
Patient status			
Alive	9 (60.0%)	10 (62.5%)	19 (61.3%)
Dead	6 (40.0%)	6 (37.5%)	12 (38.7%)
Follow-up			
Month average	41	46	44

were in early staging and 51.6% in late staging. Both staging groups showed similar proportions of tumor relapse, cured and dead patients. This study was approved by the Ethics Committee of Ribeirao Preto Medical School, University of Sao Paulo (USP) (Proc. No. 9371/2003) and signed informed consent was obtained from all patients.

2.2. Microarray experiments

After LSCC histopathological confirmation and microdissection of the tumors from their non-neoplastic adjacent tissue, the samples were store in liquid nitrogen. Total RNA was extracted with TRIzol (Life Technologies, USA). After extraction, the RNA was purified with RNeasy Kit (Qiagen, USA) and quantified with Nanodrop spectrophotometer (Thermo Scientific, USA). Its quality was evaluated by 1.5% agarose gel electrophoresis (28S and 18S ribosomal RNA detection).

For the microarray analysis it was used the Whole Human Genome Oligo Microarray kit 4 × 44K (Agilent, G4112F, USA). cDNA was hybridized to the microarray chip in the Fluidics Station 450 system (Affymetrix, USA), using the Quick Amp Labeling one-color kit (Agilent, USA). As an internal mRNA control, the One Color RNA Spike-In Kit was used (Agilent, USA). The arrays were scanned by the Agilent Scanner G2505C. Agilent Feature Extraction software (version 9.5.3.1) was used to analyze acquired array images.

2.3. Quality control and signal pre-processing

Plain text files were loaded and processed into R with the bioconductor package Agi4 × 44Pre-Process [5]. The annotation package hgug4112a.db [6] was used to assign gene information to each probe. The CV.rep.probes function was used to estimate the percent of coefficient of variation (% CV) for replicated non-control

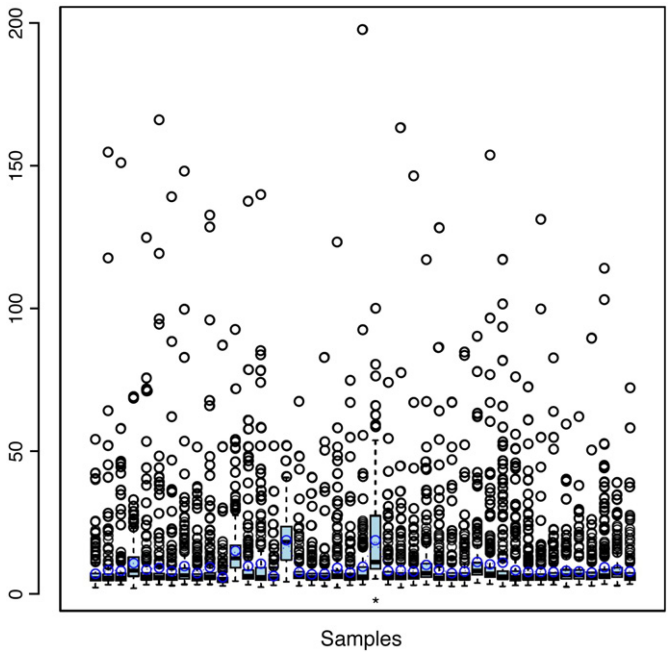

Fig. 1. Boxplot shows the percent of the coefficient of variation for non-control probes to each array. The asterisk represents the technical replicate array.

probes, inferring the reproducibility of the arrays (Fig. 1). The gProcessedSignal and the gBGUsed values of each array were loaded as foreground and background signals, respectively into an RGList object. Next, those arrays were renamed according to each sample clinical information. The following flags were used to filter probes: Control, Well Above BG, Is Found, Well Above NEG CTRLS, Is Saturated, Population Outlier, and Non Uniform Outlier. Additionally probe signals with at least 90% of good features in an experimental condition were selected. Logarithmic transformation of base 2 was applied for high quality probes. Quality control was accessed with package arrayQualityMetrics [7]. Outlier detection did not identify experimental problems. It was performed by looking for arrays (a and b), which the sum (S) of the distances (d) to all other arrays, $S_a = \Sigma_b d_{ab}$ was exceptionally large; calculating the Kolmogorov–Smirnov statistic (K_a) between each array and the pooled data distribution;

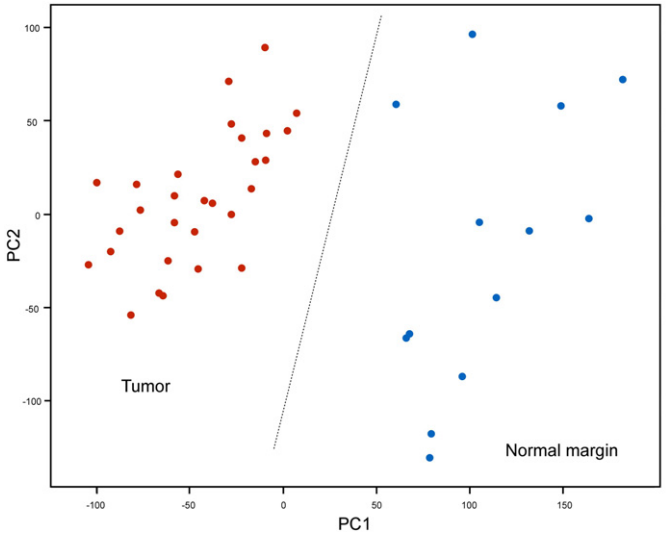

Fig. 2. Scatter plot representing the principal component analysis from expression arrays. The dots are colored by tumor (red) and adjacent non-neoplasic tissue samples (blue).