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Data in Brief

Association of aging with gene expression profiling in mouse submandibular glands

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ABSTRACT

Aging, also called senescence, is thought to be a physiological phenomenon that commonly occurs in various organs and tissues (Enoki et al., 2007 [1]). Many older adults experience dysfunction in their salivary glands, for example xerostomia, which is defined as dry mouth resulting from reduced or absent saliva flow (Nagler et al., 2004 [2]). In the present study, we investigated gene expression in submandibular glands of young (8 weeks old) and adult (50 weeks old) mice to analyze association of aging with gene expression profiling in mouse submandibular glands. Whole-genome gene expression profiles were analyzed using an Illumina Sentrix system with Mouse-WG-6 v.2 Expression BeadChips (Illumina). Of the genes screened, 284 showed detection values at a significance level of $P < 0.01$. Among those, the expression of 94 genes (33%) showed a greater decrease in adult mice as compared to young mice. On the other hand, that of 190 genes (77%) was increased in the adults more than in young mice. The data obtained in this study are publicly available in the Gene Expression Omnibus (GEO) database (accession number GSE66857).

Specifications	
Organism/cell line/tissue	*Mus musculus*/submandibular glands of young (8 weeks old) and adult (50 weeks old) mice
Sex	Male
Sequencer or array type	Illumina Mouse-WG-6 v.2 expression bead chip
Data format	Raw and analyzed
Experimental factors	Expression patterns in submandibular glands of young (8 weeks old) and adult (50 weeks old) mice were analyzed to determine aging-dependent gene expression.
Experimental features	Microarray analysis of gene expression associated with aging in mouse submandibular glands.
Consent	N/A
Sample source location	1-5-8 Hatanodai, Shinagawa, Tokyo 142-8555, Japan

1. Direct link to deposited data

http://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE66857.

2. Experimental design, materials, and methods

2.1. Introduction

The submandibular glands (SMGs) participate as major salivary secreting organs to secrete fluids rich in proteins that are critical for maintenance of oral health [1-3]. The SMGs have been reported to increase in proportional volume of fat and connective tissues with a reduction in that of acini with aging, though without any remarkable change in the volume of the duct system [4]. For investigation of age-dependent changes in the expression of genes in SMGs, a gene expression array can provide a comprehensive view of the expression pattern. The Illumina Sentrix system using Mouse-WG-6 v.2 Expression BeadChips (Illumina) reflects the latest advancements in mouse genomics and provides biologically relevant information for gene expression studies, while GenomeStudio Data Analysis Software is useful for visualizing and analyzing data obtained with the

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Table 1
Sequences of primers used for quantitative PCR.

Gene	Primer	Sequence
Pdcd4	Forward	5′-GGATGAGACCGCATTTGAGAA-3′
	Reverse	5′-AGGCTAAGGACACTGCCAACAC-3′
Ttr	Forward	5′-GGTCAAAGTCCTGGATGCTGTC-3′
	Reverse	5′-CCAGTACGATTTGGTGTCCAGTTC-3′
Pdk4	Forward	5′-CAGGTTATGGGACAGACGCTATCA-3′
	Reverse	5′-TGCTTGGGATACACCAGTCATCA-3′
Ly6d	Forward	5′-CCAGCAGGGCCATGTCA-3′
	Reverse	5′-AGGTCAGTCTGGCAGCATTGT-3′
Kik1	Forward	5′-TGACAGATGACATGTTGTGTGCAG-3′
	Reverse	5′-GATACCCGGCACATTGGGTTTA-3′
Creld2	Forward	5′-GCGATGGCCAGTACTGTGAGAA-3′
	Reverse	5′-CTGTACAGCCCACGCAGGTAGA-3′
Igfbp2	Forward	5′-GGCCGGTACAACCTTAAGCA-3′
	Reverse	5′-GGGTTCACACACCAGCACTC-3′
Sdf2l1	Forward	5′-GCTGCACTCACACGACATCAA-3′
	Reverse	5′-CGCGAATCCGCCAGTAACTA-3′
Tgm2	Forward	5′-CAACCTGACCCTGGATCCCTA-3′
	Reverse	5′-TCAGGCACCCGCTGTACTTC-3′

Illumina array platform. In the present study, we used cDNA microarray analysis to detect age-associated changes in gene expression of mouse SMGs.

2.2. Animal treatment

All animal experiments were conducted in accordance with the guidelines of Showa University. C57BL/6J mice were obtained from Sankyo Laboratory and housed in the Animal Facility at Showa University. We used 8- and 50-week-old mice as the young and adult, respectively, groups.

2.3. Tissue preparation

For general histopathological examinations, all samples were fixed in 4% paraformaldehyde and processed into frozen sections using routine procedures, then stained with hematoxylin–eosin (H–E).

2.4. Whole-genome expression assay and microarray data analysis

Whole-genome gene expression profiles in the SMGs of each mouse were analyzed using an Illumina Sentrix system with Mouse-WG-6 v.2 Expression BeadChips (Illumina), which includes 45,281 Illumina probes to detect transcriptants covering 30,854 genes, using a previously reported method [5]. First, total RNA was extracted with TRIzol reagent (Life Technologies) from whole SMGs, then 500 ng was subjected to RNA amplification, which was performed with an Illumina TotalPrep RNA Amplification Kit (Ambion), according to the manufacturer's instructions. Biotinylated cRNA was then hybridized to Mouse-WG-6 v.2 Expression BeadChips and reacted with streptavidin-cy3 (GE Healthcare). Finally, the expression intensity of the transcripts on the BeadChips was detected using an Illumina iScan reader. Raw BeadChip image data were subjected to expression analyses using the manufacturer's software (GenomeStudio v.2011.1, Gene Expression Module v.1.9.0). A heat map was generated by hierarchical clustering of the selected transcripts based on the gene expression profiles (expression ratio: AVG_signal of adult mice/AVG_signal of young mice).

2.5. Quantitative real-time PCR

Total RNA was extracted using TRIzol reagent (Life Technologies), then reverse transcribed using ReverTra Ace® qPCR RT Master Mix (TOYOBO). Quantitative real-time PCR was performed using a SYBR green Fast PCR system (GE Healthcare), with the following primer sequences shown in Table 1.

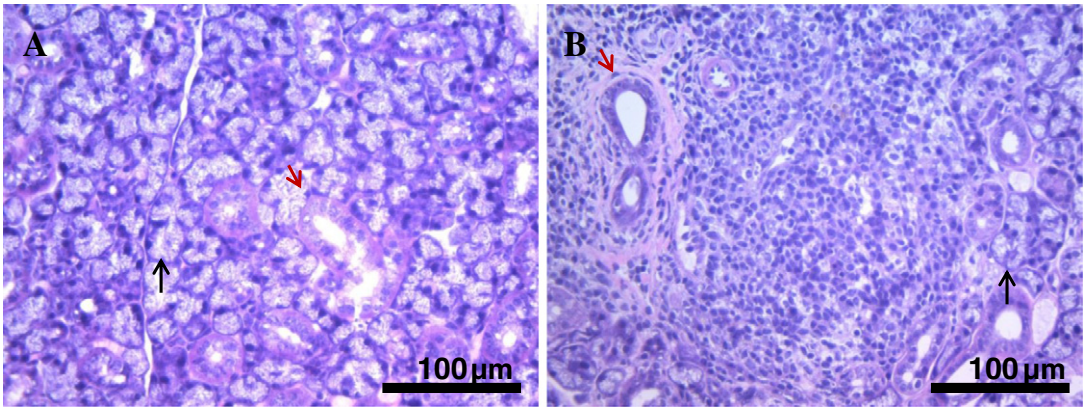

Fig. 1. Representative sections from submandibular glands of young and adult mice (H & E, scale bars: 100 μm). Black arrows, acinar cell; red arrows, duct.