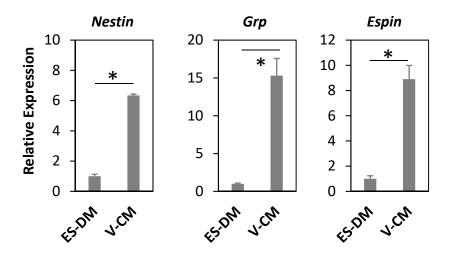
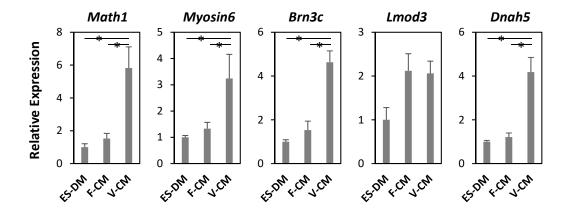
#### **Supplementary Fig. S1.**



Gene expression analysis of both immature and mature HC-related markers in *Math1-GFP* ES cells cultured in ES-DM or V-CM.

Gene expressions of *Nestin* (immature HC-related markers), *Grp* and *Espin* (mature HC-related marker) by *Math1-GFP* ES cells cultured in ES-DM or V-CM for 2 weeks were examined using real-time RT-PCR. Data were normalized to *RPLP0* expression, as an endogenous control.

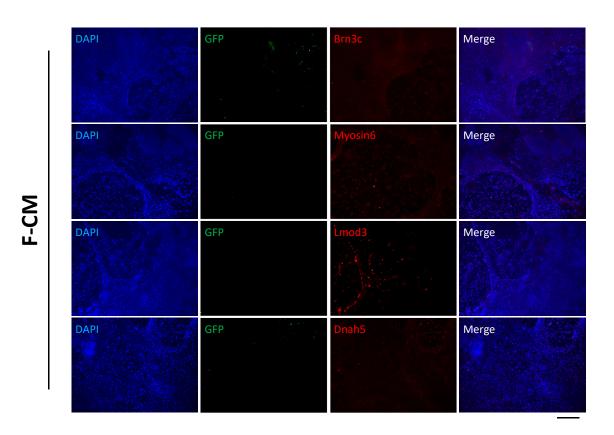
#### Supplementary Fig. S2.



### Gene expression analysis of hair cell-related markers in *Math1-GFP* ES cells cultured in ES-DM, F-CM or V-CM.

Gene expressions of *Math1*, *Myosin6*, and *Brn3c*, (HC-related markers), *Lmod3* (cochlear HC-related marker), and *Dnah5* (vestibular HC-related marker) by *Math1-GFP* ES cells cultured in ES-DM or V-CM for 2 weeks were examined using real-time RT-PCR. Data were normalized to *RPLP0* expression, as an endogenous control.

## **Supplementary Fig. S3.**



Immunocytochemical analysis of hair cell-related markers expressed by *Math1-GFP* ES cells cultured in F-CM.

The expressions of HC-related markers in *Math1-GFP* ES cells cultured in F-CM for 2 weeks were examined with an immunocytochemical method. Scale bar =  $100 \mu m$ 

# **Supplementary Table S1**

Table S1. List of gene-specific primers using in real time RT-PCR analysis

Genes	Primer sequences	Product size	GeneBank
	2 22 4	(bp)	Accession No.
Math1	Forward: 5'- AAATGTCGTATCTCTGCCTCTGGTC	144	NM_007500.4
	Reverse: 5'- AAGTACCCAATGCGGGTCTCAA		
Myosin6	Forward: 5'- ATGGGCTGTGGGAACAGTGATA	134	NM_001039546.2
	Reverse: 5'- CCCAAGAGTGTTGGTTGTCGAG		
Brn3c	Forward: 5'- ATGCGCCGAGTTTGTCTCC	68	NM_138945.2
	Reverse: 5'- AGGCTCTCATCAAAGCTTCCAAATA		
Lmod3	Forward: 5'- TGAATGACATCCGACACAGCAA	123	NM_001081157.1
	Reverse: 5'- GTTCGTGAAATGGCCTCCAG		
Dnah5	Forward: 5'- TGTTGTGTGCAATGAATGAGATGAC	163	NM_133365.3
	Reverse: 5'- TTGATGCCCACAATTCATTAGGAG		
β-actin	Forward: 5'-CATCCGTAAAGACCTCTATGCCAAC	171	NM_007393.5
	Reverse: 5'-ATGGAGCCACCGATCCACA		
RPLP0	Forward: 5'-GTGTTTGACAACGGCAGCATTTA	171	NM_007475.5
	Reverse: 5'-GACGCGCTTGTACCCATTGA		