# SUPPLEMENTARY MATERIAL 

## corresponding to:

## Galectin-1 enhances the generation of neural crest cells

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## PCR primers

Sox10: 5'-TTCAGGCTCACTACAAGAGTG-3'(forward) and 5'-TCAGAGATGGCAGTGTAGAGG-3'(reverse);
Venus: 5'-ATGGTGAGCAAGGGCGAGGA-3'(forward) and 5'-TTCTGCTGGTAGTGGTCGGCGA-3'(reverse);
P75: 5'-ATACGGTGACCACTGTGATG-3'(forward) and 5'-TCCACAATGTCAGCTCTCTG-3'(reverse);
Pax3: 5'-GTTGCGTCTCTAAGATCCTG-3'(forward) and 5'-GCGTCCTTGAGCAATTTGTC-3'(reverse);
Snail: 5'-ATAGCGAGCTGCAGGACGCGTGTGT-3'(forward), 5'-AGGCCGAGGTGGACGAGAAGGACGA-3'(reverse);
Gal-1: 5'-TTCAATCCTCGCTTCAATGCCC-3'(forward) and 5'-TCACTCAAAGGCCACGCACTTA-3'(reverse);
Gapdh: 5'-CTTCACCACCATGGAGAAGGC-3'(forward) and 5'-GGCATGGACTGTGGTCATGAG-3'(reverse).

## Primers for quantitative-PCR

Nestin: 5'-AAAGGAAAGGCAGGAGTCCCTGAA-3'(forward) and 5 '-TGGTCCTCTGCGTCTTCAAACCTT -3'(reverse);
Hoxb9: 5'-AAAAAGCGCTGTCCCTACACC-3'(forward) and 5 '-AGGAGTCTGGCCACTTCATG-3'(reverse);
Ncam: 5'-GGATGCCTCCATCCACCTC-3'(forward) and 5 '-GGCCGTCTGATTCTCTACATAGG-3'(reverse);
Snail: 5'-TTGTGTCTGCACGACCTGTG-3'(forward) and 5 '-CACTGGTATCTCTTCACATCC-3'(reverse);
Sox10: 5'-CAGTCCGGCAAGGCAGACCC-3'(forward) and 5 '-GCAGGTATTGGTCCAGCTCAGTCAC-3'(reverse);
Sox9: 5'-AGTACCCGCATCTGCACAAC-3'(forward) and 5 '-TACTTGTAATCGGGGTGGTCT -3'(reverse);
Foxd3: 5'-ATCCTGGTCCATCTGTCCTG-3'(forward) and 5 '-GCAGAGTCCAGGATTGGGTA-3'(reverse);
$\beta$-Actin: 5'-CATCCGTAAAGACCTCTATGCCAAC-3'(forward) and 5 '-ATGGAGCCACCGATCCACA-3'(reverse).


Supplementary Fig. S1. SOX10+ neural crest (NC)-like cells differentiate into neurons, glial cells, and melanocytes in the presence of GAL-1. (A) Sox10-IRES-Venus embryonic stem (ES) cells were differentiated into NC-like cells in the presence of GAL-1 at the indicated concentration. After 12 days, Sox10+ NC-like cells were inoculated at 200 cells/well onto ST2 monolayers. After 21 days in culture, the colonies were immunostained with TuJ-1 for neuron and anti-GFAP for glial cells. Melanocytes were detected as pigmented cells. The number of different types of colonies was counted. The experiment was performed 3 times, and each result is indicated as the average for all 3 experiments. (B) The presence of GAL-1, rhGAL-1/Ox, or CSGAL-1 did not change the expression of NC cell-related genes in NC-like cells. (C) Typical image of neurons, glial cells, and melanocytes generated from SOX10+ NC-like cells in the presence of GAL-1 at the indicated concentration. Scale bar, $100 \mu \mathrm{~m}$.


Supplementary Fig. S2. The presence of GAL-1, rhGAL-1/Ox, or CSGAL-1 did not change the differentiation potency the neural crest (NC)-like cells. Typical image of neurons, glial cells and melanocytes generated from Sox10+ NC-like cells in the presence of GAL-1, rhGAL-1/Ox, or CSGAL-1. Sox10-IRES-Venus embryonic stem (ES) cells were differentiated into NC-like cells in the presence of GAL-1, rhGAL-1/Ox, or CSGAL-1 at $1.0 \mathrm{ng} / \mathrm{ml}$. After 12 days, Sox10+ NC-like cells were inoculated at 200 cells/well onto ST2 monolayers. After 21 days in culture, the colonies were immunostained with the antibodies indicated in Supp. Fig. S1. Scale bar, $200 \mu \mathrm{~m}$.


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