Long noncoding RNAs (IncRNA) MALAT1 in regulating osteogenic and adipogenic differentiation using a novel molecular nanobiosensor

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**Fig. S1.** Stability of ds-GapM-LNA nanobiosensor.

**Fig. S2.** siRNA silencing efficiency.

**Fig. S2.** Dynamic tracking of MALAT1 expression during osteogenic differentiation.

**Fig. S3.** Dynamic tracking of MALAT1 expression during adipogenic differentiation.

**Fig. S4.** Representative bright field and fluorescence images of hMSCs after 15 days under different treatments.

**Fig. S5.** Comparison of cell proliferation of hMSCs under control siRNA and MALAT1 siRNA treatments during osteogenic and adipogenic induction.

**Tab. S1.** ds-GapM-LNA probes and quencher sequences
**Fig. S1. Stability of ds-GapM-LNA nanobiosensor.** Comparison of fluorescence intensity of ds-GapM-LNA nanobiosensor in the presence of target sequence. All the concentrations were set to 100 nM. Data are expressed as mean ±SEM. p-Values were calculated using a two-sample t-test within groups. *ns*, not significant.
**Fig. S2. siRNA silencing efficiency.** The expression of IncRNA MALAT1 was evaluated and analyzed using RT-PCR assay. Experiments were performed at least three times. The relative expression levels of IncRNAs were determined by the equation $2^{-\Delta\Delta CT}$. Data are expressed as mean ± s.e.m. (n = 3). A two-tailed t-test was used to analyze differences between control siRNA and MALAT1 siRNA. *, $p < 0.05$; **, $p < 0.01$; ***, $p < 0.005$. 

![Graph showing relative expression of MALAT1 under control and MALAT1 siRNA conditions]
Fig. S3. Dynamic tracking of MALAT1 expression during osteogenic differentiation.

Representative merged images of hMSCs during osteogenic differentiation. Images were taken every two days until 15 days of differentiation. Green fluorescence indicates MALAT1 expression. Scale bar: 100 μm.
Fig. S4. Dynamic tracking of MALAT1 expression during adipogenic differentiation.

Representative merged images of hMSCs during adipogenic differentiation. Images were taken every two days until 15 days of differentiation. Green fluorescence indicates MALAT1 expression. Scale bar: 100 µm.
Fig. S5. Representative bright field and fluorescence images of hMSCs after 15 days under different treatments. (A) Control group; hMSCs were cultured in the basal medium without treatments. (B) Osteogenic induction group; hMSCs were cultured in osteogenic induction medium. (C) Adipogenic induction group, hMSCs were cultured in adipogenic induction medium. Green: MALAT1; Blue: Nucleus. Scale bar: 100 μm.
Fig. S6. Comparison of cell proliferation of hMSCs under control siRNA and MALAT1 siRNA treatments during osteogenic and adipogenic induction. For the control group, hMSCs were maintained in basal culture medium. hMSCs were seeded in 96-well plates with 2000 cells per well. After cells reached 80% confluency, hMSCS were treated with control siRNA and MALAT1 siRNAs. After 24 hours of silencing, hMSCs were induced to osteogenic or adipogenic differentiation. The cell proliferation was evaluated using a cell counting kit (CCK-8) assay. Absorbance was measured and compared at 450 nm. Experiments were repeated independently at least three times. Data are expressed as mean± s.e.m. (n=3, ***, p<0.001, **, p<0.01)
**Tab. S1.** ds-GapM-LNA probes and quencher sequences

<table>
<thead>
<tr>
<th>Name</th>
<th>Sequence (5’-3’)</th>
<th>Fluorophore</th>
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<tbody>
<tr>
<td>Dll4 mRNA</td>
<td>+T+C+G+C+A TACGT GTGTC TGCTG AGTGT +T+C+C+T+G</td>
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<td>Donor</td>
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<td>/3-Iowa BlackFQ</td>
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<td>/56-FAM</td>
</tr>
<tr>
<td>Donor</td>
<td>+C+T+T+C+G AGATA CTGTA</td>
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<tr>
<td>Quencher</td>
<td>AGGTG CCCTA CTGGT CTTCG AGATA CTGTA</td>
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* + represents LNA monomer