Manipulation of Gene Expression in Zebrafish Using Caged Circular Morpholino Oligomers

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Scheme S1. Synthesis of photocleavable linker (**PL**)

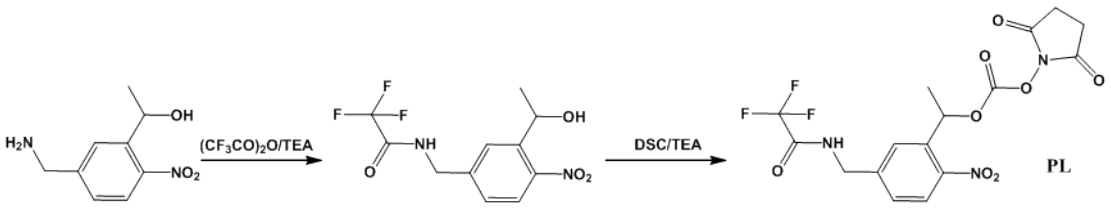
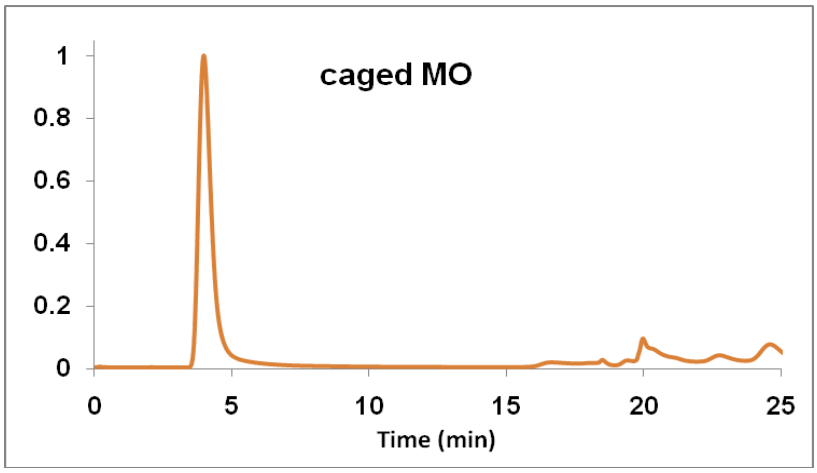
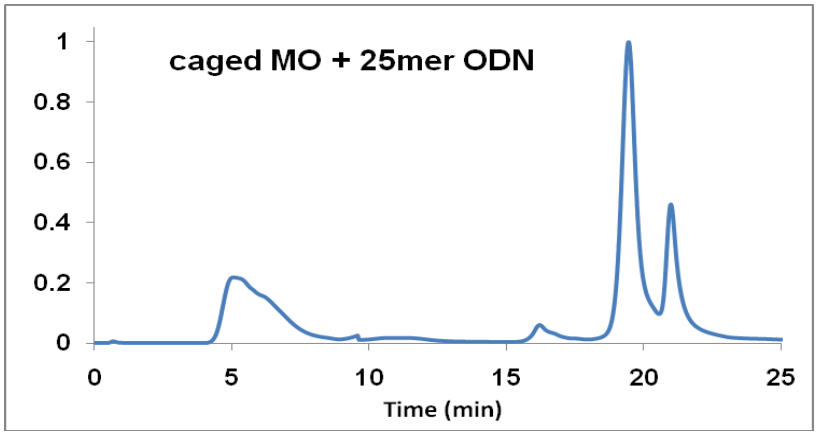


Figure S1. Ion exchange HPLC traces for caged circular MO-cat2 with the addition of different ODNs The caged circular MO-cat2 passed through ion exchange column quickly due to non-charged backbone. A 25mer sense ODN (ODN-25) can still bind the caged circular MO-cat2 and the retention time for the duplex is much longer, while a 30mer ODN hairpin (ODN-30) is immune to the binding of the caged circular MO-cat2.





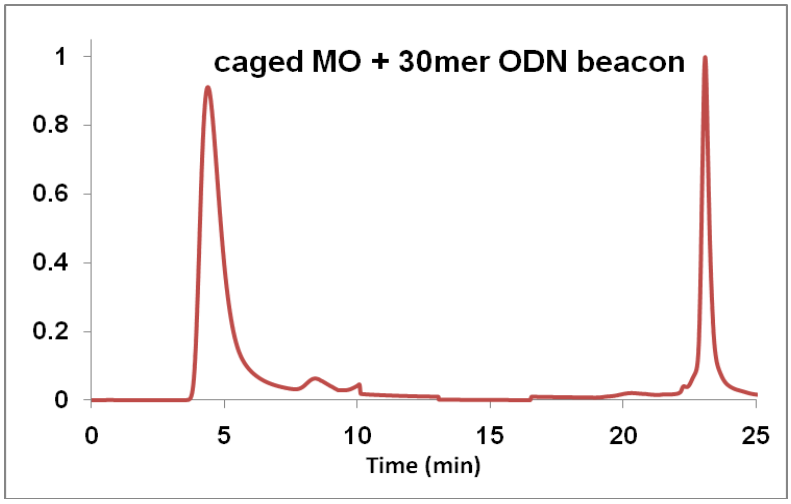


Figure S2 Gel mobility shift assays of caged circular MO-ntl and its complementary oligonucleotide sequence (ODN-38). L1, ODN-38 only; L2, ODN-38+caged cMO-ntl; L3, ODN-38+caged cMO-ntl+ 10 min light irradiation.

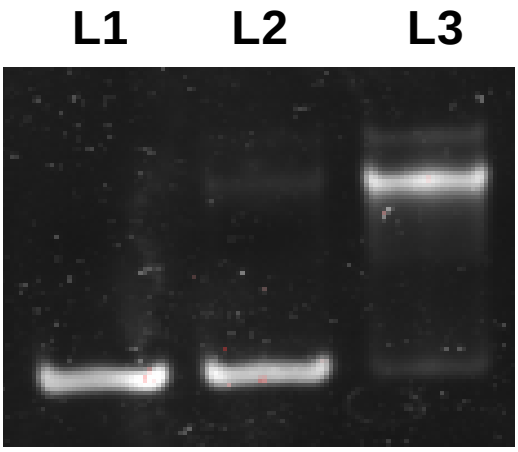
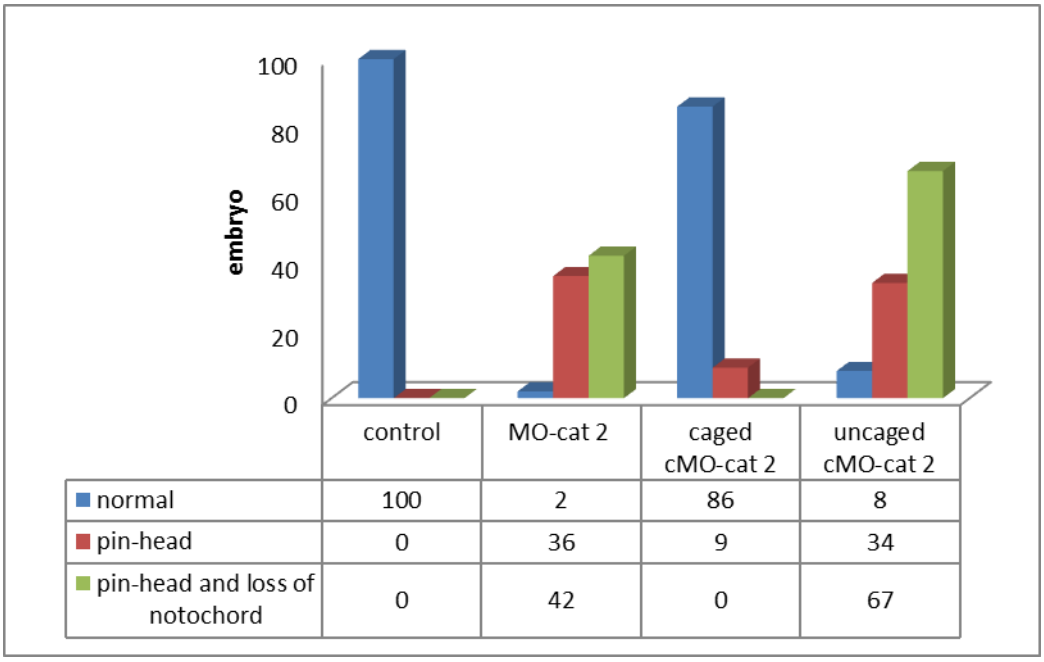


Figure S3. Phenotype observation of zebrafish embryos injected with linear *β-catenin-2* MO (MO-cat 2), caged circular MO-cat 2 (caged cMO-cat 2), and uncaged circular MO-cat 2 (uncaged cMO-cat 2). Zebrafish with pin-head and clear loss of notochord or zebrafish with pin head and less clear loss of notochord are all counted as no "*β-catenin-2*" phenotypes.



	control	MO-cat 2	caged cMO-cat 2	uncaged cMO-cat 2
normal	100	2	86	8
pin-head	0	36	9	34
pin-head and loss of notochord	0	42	0	67

Figure S4. Representative transmitted light images of 24-hpf zebrafish embryo phenotypes. A, uninjected embryos irradiated for 10 min developed normally. B, Embryos with linear MO-ntl injection exhibited clear *no tail* phenotype. C. Injection with caged circular MO-ntl (caged cMO-ntl) had no effect in the dark. D, Injection with caged cMO-ntl and 10-min light irradiation (uncaged cMO-ntl) at 3 hpf gave *no tail* phenotype.

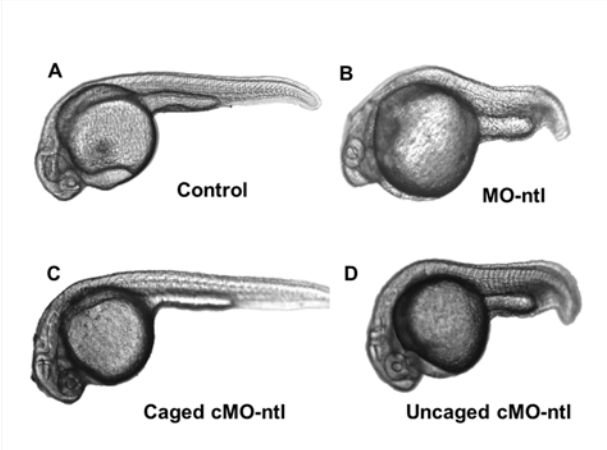
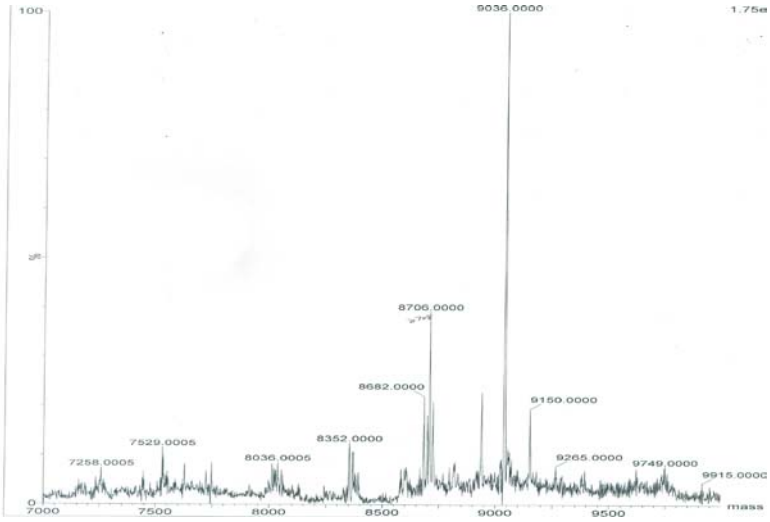
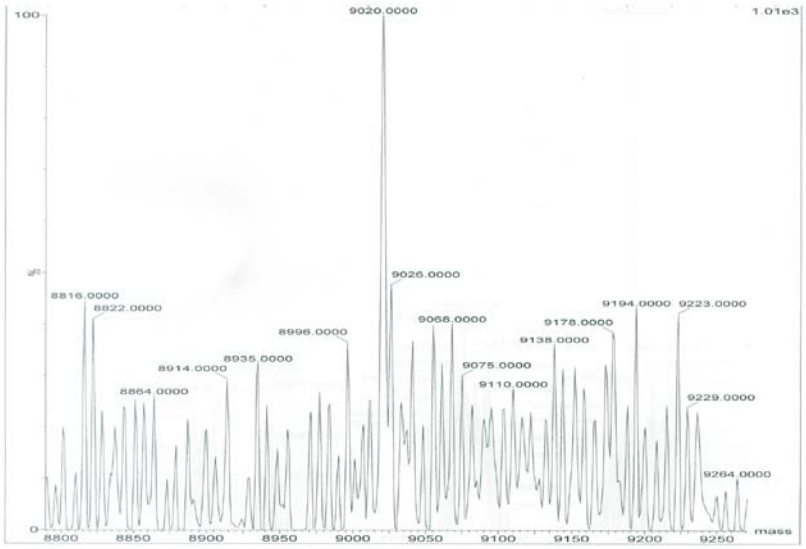


Figure S5 MS spectra of MO-cat2 and MO-ntl before and after circularization.

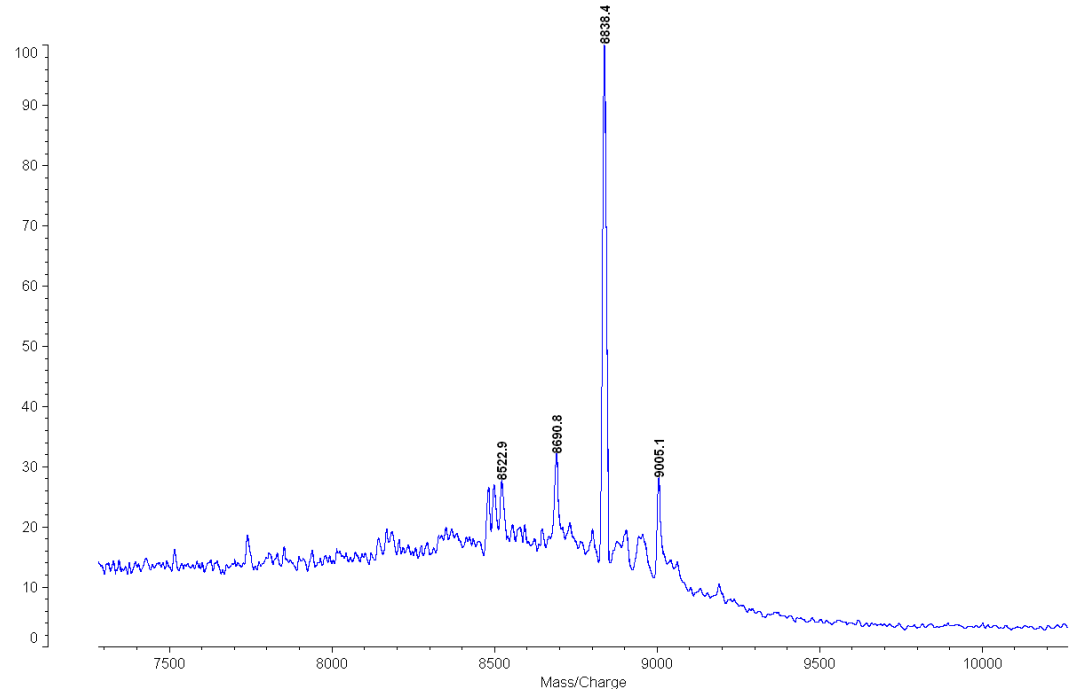
MO-cat2 before cyclization



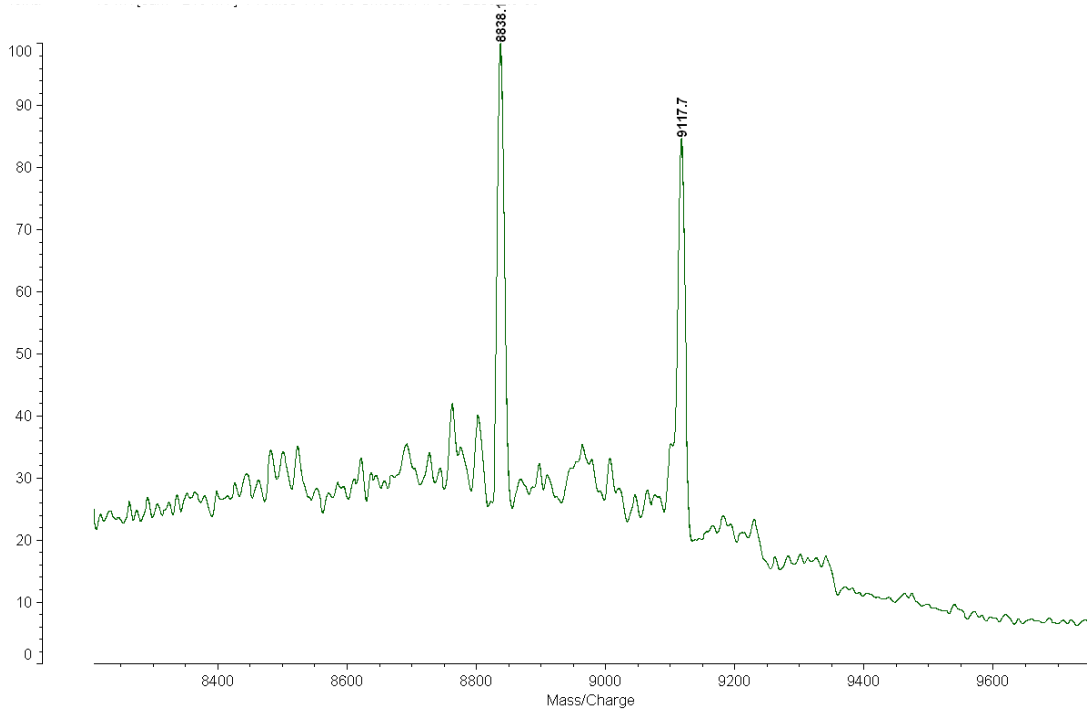
MO-cat2 after cyclization



linear MO-ntl directly cleaved from resin



linear MO-ntl after the addition of **PL** and acid moiety. The peak of 8838 may be due to the uncaging when laser was used to ionize the sample for MALDI measurement.



MO-ntl after cyclization