

Resource Summary Report

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eGFP-pro α 2(I)-G610C

RRID:Addgene_119827

Type: Plasmid

Proper Citation

RRID:Addgene_119827

Plasmid Information

URL: <http://www.addgene.org/119827>

Proper Citation: RRID:Addgene_119827

Insert Name: Type I procollagen α 2 chain

Organism: Mus musculus

Bacterial Resistance: Ampicillin

Defining Citation: [PMID:30287488](#)

Vector Backbone Description: Backbone Size:5400; Vector Backbone:pcDNA3.1; Vector Types:Mammalian Expression; Bacterial Resistance:Ampicillin

Comments: These plasmids were constructed after the G610C mouse model of OI in which the Gly610 residue (starting from the N-terminal end of the triple helix) was substituted with Cys. These mouse type I procollagen plasmids were designed for immortalized and primary murine osteoblasts and fibroblasts. Their expression in other cells might cause significant disruption of assembly, folding and trafficking of type I procollagen, resulting in artifacts. Even in osteoblasts and fibroblasts a fraction of transfected chains, particularly pro α 2, might be trafficked/degraded as monomers instead of being integrated into procollagen heterotrimers. In our experience, the best evidence of normal behavior of the transfected chains is the appearance of fluorescent extracellular collagen fibers ~ 12 h after transfection. Fluorescent molecules should be incorporated into fibers even when the transfected chains contain the N-propeptide cleavage site, because the cleavage is highly conformation sensitive and always incomplete. We recommend following these guidelines when using the plasmids 1) Only mouse cells that have high expression of endogenous type I procollagen are suitable for studies of physiologically-relevant processes with these constructs. 2) At least 100 μ M ascorbic acid at and after transfection is required to ensure proper procollagen folding (for some cells, preincubation with ascorbic acid before transfection might be needed). 3) Optimization of transfection efficiency is needed for all cell types. 4) Experiments beyond 24h after

transfection are not recommended, to avoid excessive accumulation of aggregates and cell malfunction caused by increased procollagen synthesis. 5) Lack of extracellular fluorescent fibers 12-24 h after transfection indicates improper procollagen synthesis/trafficking or cellular malfunction. 6) Excessive expression of transfected chains might cause rapid accumulation of large procollagen aggregates in the ER, resulting in cellular malfunction and death. Contact information: Shakib Omari (shakib.omari@nih.gov), Sergey Leikin (leikins@mail.nih.gov).

Plasmid Name: eGFP-pro α 2(I)-G610C

Relevant Mutation: Col1a2 exons 2-5 replaced by fluorescent tag, Glycine 610 mutated to Cysteine

Record Creation Time: 20220422T221626+0000

Record Last Update: 20260815T080316+0000

Ratings and Alerts

No rating or validation information has been found for eGFP-pro α 2(I)-G610C.

No alerts have been found for eGFP-pro α 2(I)-G610C.

Data and Source Information

Source: [Addgene](#)

Usage and Citation Metrics

We found 2 mentions in open access literature.

Listed below are recent publications. The full list is available at [RRID](#) .

Makareeva E, et al. (2025) LC3 and GABARAP independent autophagy of misfolded procollagen in mouse osteoblasts. *Autophagy*, 21(12), 2932.

De Leonibus C, et al. (2024) Sestrin2 drives ER-phagy in response to protein misfolding. *Developmental cell*, 59(16), 2035.