Drug-Inducible, Cell-Specific Manipulation of Intracellular Calcium in Zebrafish Through Mammalian TRPV1 Expression

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CALCIUM IS A WIDELY used cellular signal, making it challenging to study in specific cells within living animals. Beerman *et al.*¹ developed a transgenic system to control calcium flux in specific cell types in zebrafish larvae (Fig. 1). Cell-specific expression of the rat TRPV1 (rTRPV1) calcium channel provides a precise and reversible method of controlling calcium flux by addition of capsaicin, a mammalian TRPV1 agonist with no known activity on the orthologous zebrafish channel. Capsaicin can be added directly to water at micromolar concentrations and acts within seconds to induce rapid calcium influx specifically in the TRPV1-expressing cells. Using genetically encoded calcium reporters, calcium dynamics can be observed in real time. Depending on duration and concentration of capsaicin treatment, the TRPV1-capsaicin system can generate pulses of elevated calcium, produce subcellular calcium gradients, or even induce cell death. These Gateway-compatible constructs provide a pharmacological method for controlling calcium dynamics in diverse cell types.

FIG. 1. Expression of rTRPV1 allows for conditional and reversible control of calcium levels in specific cell types. (A) Cartoon of the Gateway construct used to generate the *rTRPV1* transgenic lines. *rTRPV1*, initially in a Gateway pME vector, can be combined with any cell-specific promoter and fluorophore. (B) Different capsaicin treatments lead to varied cellular outcomes (green indicates calcium). Pulsed capsaicin can reversibly alter intracellular calcium levels; gradient application of capsaicin can change subcellular calcium dynamics; prolonged capsaicin treatment at high doses can induce cell death, ablating specific cell populations. (C) Transgenic zebrafish larvae *Tg(LysC:rTRPV1-tdTomato; LysC:GCaMP3)*. GCaMP3 is a genetically encoded calcium indicator that exhibits increased fluorescence when bound by calcium. Scale bar is 200 μ m. Boxed region is enlarged in (D), showing increased levels of GCaMP signal upon capsaicin administration, but no change in basal red fluorescence. rTRPV1, rat TRPV1. Color images available online at www.liebertpub.com/zeb

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Reference

1. Beerman RW, Matty MA, Au GG, Looger LL, Choudhury KR, Keller PJ, *et al.* Direct *in vivo* manipulation and imaging of calcium transients in neutrophils identify a critical role for leading-edge calcium flux. *Cell Rep* 2015;13:1–11.

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