

JOINT EVENT

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**Anterograde monosynaptic transneuronal tracers derived from herpes simplex virus 1 strain H129****Min Hua Luo, Wen Bo Zeng, Hai Fei Jiang, Yousheng Shu, Shao Qun Zeng, Fuqiang Xu and Fei Zhao**

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**H**erpes simplex virus type 1 strain 129 (H129) has represented a promising anterograde neuronal circuit tracing tool, which complements the existing retrograde tracers. However, the current H129 derived tracers are multi synaptic, neither bright enough to label the details of neurons nor capable of determining direct projection targets as monosynaptic tracer. The present study is to introduce an improved multi synaptic anterograde tracer H129-G4 with strong labeling intensity, and a novel mono-synaptic anterograde tracer H129- $\Delta$ TK-tdT. Based on the bacterial artificial chromosome of H129, H129-G4 was obtained by inserting binary tandemly connected GFP cassettes into the H129 genome, and H129- $\Delta$ TK-tdT was obtained by deleting the thymidine kinase (TK) gene and adding tdTomato coding gene to the H129 genome. H129-G4 is capable of transmitting through multiple synapses, labeling the neurons by green fluorescent protein, and visualizing the morphological details of the labeled neurons. H129- $\Delta$ TK-tdT neither replicates nor spreads in neurons alone, but transmits to and labels the postsynaptic neurons with tdTomato in the presence of complementary expressed TK from a helper virus. H129- $\Delta$ TK-tdT is also capable to map the direct projectome of the specific neuron type in the given brain regions in Cre transgenic mice. In the tested brain regions where circuits are well known, the H129- $\Delta$ TK-tdT tracing patterns are consistent with the previous results. With the assistance of the helper virus complementarily expressing TK, H129- $\Delta$ TK-tdT replicates in the initially infected neuron, transmits anterogradely through one synapse, and labeled the postsynaptic neurons with tdTomato. The H129- $\Delta$ TK-tdT anterograde monosynaptic tracing system offers a useful tool for mapping the direct output in neuronal circuitry. H129-G4 is an anterograde multisynaptic tracer with a labeling signal strong enough to display the details of neuron morphology.

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