

SPARC and SPARC2 user guide

a) Important notes:

1. SPARC and SPARC2 activity requires a minimum of three transgenes: 1) *SPARC-effector*, 2) *promoter-PhiC31*, and 3) *enhancer-GAL4*.
2. *SPARC-effector* or *SPARC2-effector* and *promoter-PhiC31* transgenes MUST be maintained in separate stocks. If both transgenes are present in the same fly, there is a possibility that unanticipated PhiC31 expression in the germline will lead to stable recombination of the *SPARC* or *SPARC2* cassette.
3. For all new *SPARC-effector* combinations, we recommend using SPARC2.

b) Example crossing schemes for SPARC or SPARC2

$$\frac{+}{+}; \textit{SPARC2-Effector}; + \quad \times \quad \frac{+}{Y}; \textit{promoter*} \textit{-PhiC31}; \textit{enhancer-Gal4} \\ \textit{(near attP40)} \quad \quad \quad \textit{\{su(Hw)attP5\}} \quad \quad \quad \textit{\{attP2\}} \quad \quad \quad \text{OR}$$

$$\frac{+}{+}; \textit{SPARC2-LexA::p65}; \textit{lexAop-effector} \quad \times \quad \frac{+}{Y}; \textit{promoter*} \textit{-PhiC31}; \textit{enhancer-GAL4} \\ \textit{(near attP40)} \quad \quad \textit{(variable)} \quad \quad \quad \textit{\{su(Hw)attP5\}} \quad \quad \quad \textit{\{attP2\}}$$

* Available stocks: *20XUAS-PhiC31*, *tub-PhiC31*, and *nSyb-PhiC31*

Using split-Gal4 Drivers:

$$\frac{+}{+}; \textit{SPARC2-effector}; + \quad \times \quad \frac{\textit{promoter*} \textit{-PhiC31}}{Y} \textit{\{attP18\}}; \textit{enhancer-GAL4AD}^{**}; \textit{enhancer-GAL4DBD} \\ \textit{(near attP40)} \quad \quad \quad \textit{\{attP40\}} \quad \quad \quad \textit{\{attP2\}} \quad \quad \quad \text{OR}$$

$$\frac{+}{+}; \textit{SPARC2-LexA::p65}; \textit{lexAop-effector} \quad \times \quad \frac{\textit{promoter*} \textit{-PhiC31}}{Y} \textit{\{attP18\}}; \textit{enhancer-GAL4AD}; \textit{enhancer-GAL4DBD} \\ \textit{(near attP40)} \quad \quad \textit{\{variable\}} \quad \quad \quad \textit{\{attP40\}} \quad \quad \quad \textit{\{attP2\}}$$

* Available stocks: *20XUAS-PhiC31* and *nSyb-PhiC31*.

** Note: *Janelia* split-Gal4 drivers use the p65ADZp activating domain²¹.

a, Important notes regarding SPARC and SPARC2 use and stock maintenance. **b**, Example crossing schemes for SPARC or SPARC2 to allow expression of effectors.