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

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

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

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

Using split-Gal4 Drivers:

$$\frac{+}{+};\ \underset{(near\ \{attP40\})}{SPARC2\text{-}effector};\ + \quad X \quad \frac{\underset{\{attp18\}}{promoter^{*}\text{-}PhiC31}}{Y};\ \underset{\{attP40\}}{enhancer\text{-}GAL4AD^{**}};\ \underset{\{attP2\}}{enhancer\text{-}GAL4DBD} \qquad OR$$

$$\frac{+}{+};\ \underset{(near\ \{attP40\})}{SPARC2\text{-}LexA::p65};\ \underset{\{variable\}}{lexAop\text{-}effector} \quad X \quad \frac{\underset{\{attp18\}}{promoter^{*}\text{-}PhiC31}}{Y};\ \underset{\{attP40\}}{enhancer\text{-}GAL4AD};\ \underset{\{attP2\}}{enhancer\text{-}GAL4DBD}$$

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

** Note: Janelia split-Gal4 drivers use the p65ADZp activating domain[21].

Isaacman-Beck et al., 2020. Extended Data 4.