**Abstract**

Advanced Genetics: Mosaic Analysis Tools & Short Conditional Introns

Image-based lineage tracing has allowed the interrogation of adult tissue turnover kinetics and lineage potential of different cell populations. Based on the multicolour reporter systems, several mosaic genetic systems have been developed. Previously, we reported Red2Onco, which ectopically expresses mutated oncogenes together with the RFP, thereby allowing the dissection of expansion kinetics and neighbouring effects of oncogenic clones. In the current study, we report Red2Flpe, a new mosaic knockout system with multicolour reporters for both mutant and wild-type cells. Red2Flpe shows efficient and specific recombination in the RFP+ clones both in vitro and in vivo. To facilitate new conditional knockout (cKO) mouse line generation, we have also developed a Short Conditional intrON (SCON) technology that is suitable for one-step cKO allele generation via zygote injection. SCON is compatible with both Cre/loxP- and Flp/frt-based cKO recombination systems. Utilizing Red2Flpe and Sox2-SCONfrt, we investigated the functions of Sox2 in the adult oesophagus in which Sox2 has been thought to be crucial for stem cell maintenance and tissue turnover. In conclusion, we have constructed a toolkit for in vivo mosaic knockout studies that is suitable for clonal tracing with internal controls and a pipeline for one-step generation of cKO alleles.