Improvement and Application of Gene Engineering Technique in the Silkworm *Bombyx mori*

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Transgenic silkworm is utilized for various research objects such as gene functional analysis as well as practical objects. Improvement of gene engineering technique is very critical for the further facilitation of the silkworm study.

1) Identification of novel promoters

The versatile promoters are essential for the induction of transgene expression. We found that the 1.3 kb upstream fragment of *lp44* gene could induce gene expression specifically in the hemocyte oenocytoid cells. We further analyzed the silkworm enhancer trap train and found that the *hsp90* 2.9 kb upstream fragment could induce strong gene expression in the silkworm cell culture as well as in various silkworm developmental stages and/or tissues [1].

2) Development of novel knock-in technique

Genome editing is a very powerful technique for the modification of the targeted genes. In the silkworm TALEN can disrupt the target gene very efficiently. However, the knock-in has been difficult according to the low activity of homologous recombination in this species. We applied the novel knock-in technique termed PITCh (Precise Integration into Target Chromosome) to the silkworm and found that this technique could mediate the precise and efficient integration of a donor vector harboring *hsp90* promoter and GFP into *BLOS2* gene [2].

3) Analysis of gene regulation mechanism in the silk gland

Silk gland is a tissue that is responsible for the massive production of the protein. We analyzed the mechanism of gene expression in the silk gland and found that a *Hox* gene *Antennapedia* regulates expression of multiple major silk protein genes such as *sericin-1*, *sericin-3*, *fhxh4* and *fhxh5* in the middle silk gland [3], whereas in the posterior silk gland a homeobox transcriptional factor *Arrowhead* functions for the regulation of *h-fibroin*, *l-fibroin* and *fibrohexamerin* expression.

References

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