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## 2 **Supplementary Information for**

3 **Sequential *in vivo* labeling of insulin secretory granule pools in INS-SNAP transgenic pigs**

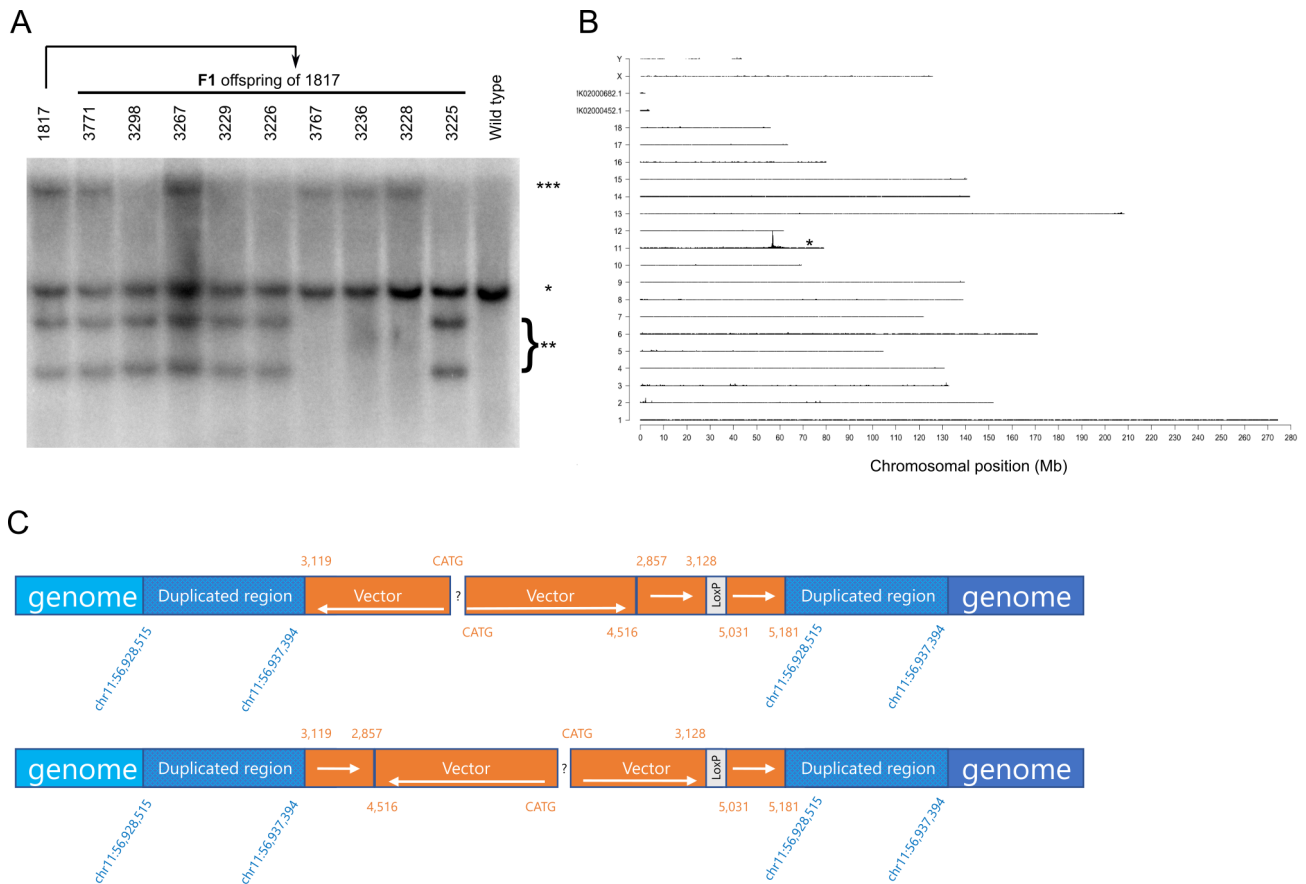
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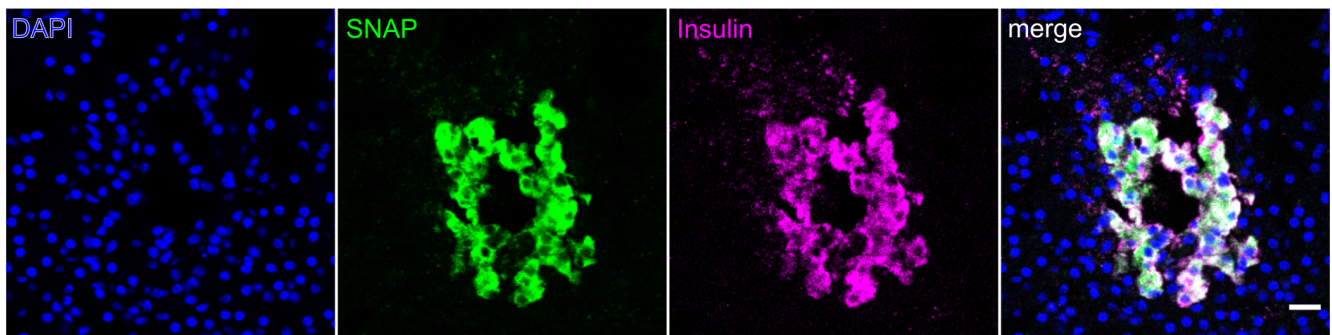
7 **This PDF file includes:**

8 Figs. S1 to S5



**Fig. S1. Southern blot and sequencing results of SOFIA pig offspring.** (A) Southern blot analysis for validation of integration pattern of F1-offspring of founder 1817. DNA was digested with the 'single cutter' restriction endonuclease enzyme *PshAI*. \*, the band representing the endogenous *INS* promoter; \*\*, *neo/δneo* transgene integration site with two integrants where *neo* cassette was deleted only in one *INS-SNAP* integrant; \*\*\*, *δneo* transgene integrant. F1-offspring 3298, 3229, 3226 and 3225 possessed only *neo/δneo* transgene integration site, animals 3767, 3236 and 3228 had only *δneo* transgene integration site, whereas 3771 and 3267 harbored both transgene integration sites. (B) TLA sequencing of an animal from the *INS-SNAP neo/δneo* transgene integrant line revealed a single genomic transgene integration site in chromosome 11. (C) TLA sequencing derived interpretations of construct integration patterns into the identified single genomic transgene integration site.

SOFIA



WT

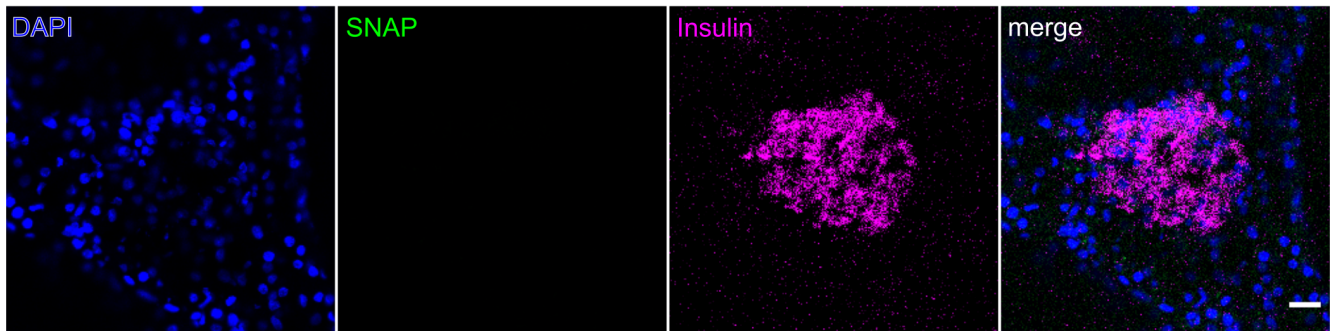
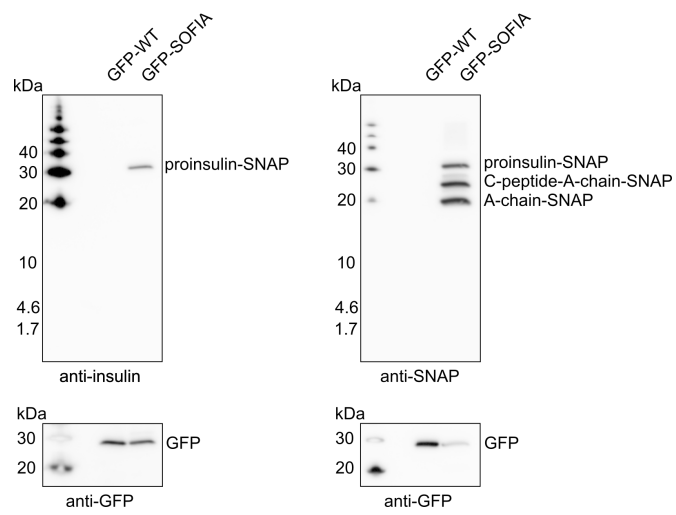
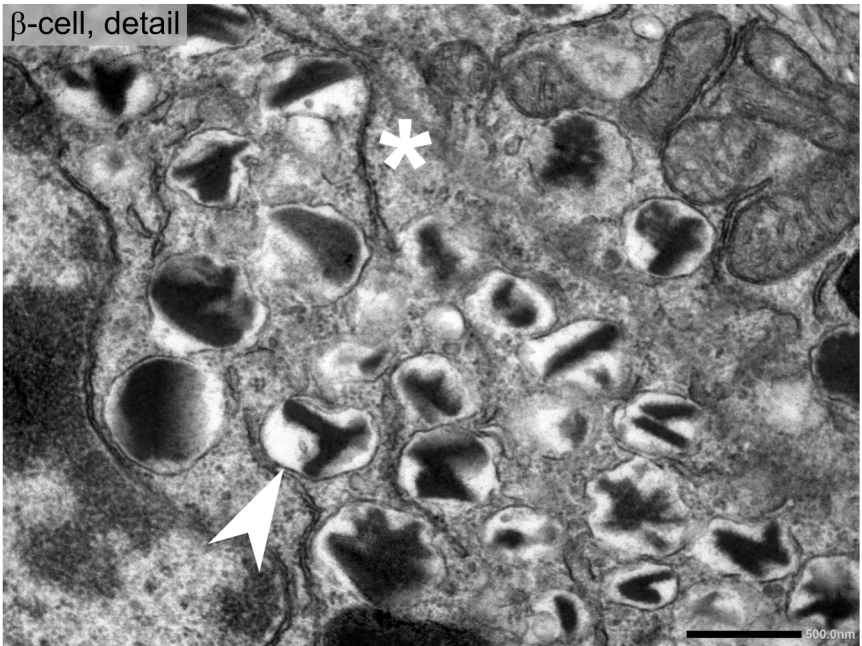
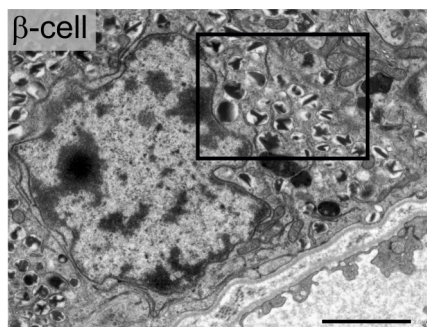
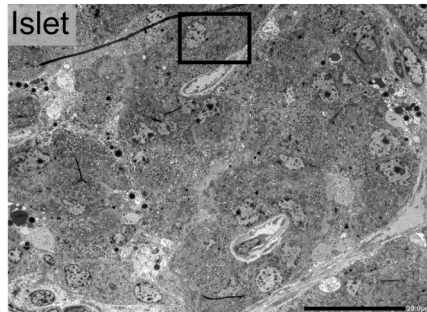


Fig. S2. Immunolabeling against SNAP and insulin in *SOFIA* and WT pancreas. Scale bar: 20  $\mu$ m.



**Fig. S3.** Western blots of reducing tricine gels against insulin, SNAP and GFP on FACS sorted *GFP-SOFIA* or *GFP-WT*  $\beta$ -cells. *GFP-WT*: beta cells from *INS-eGFP* animal; *GFP-SOFIA*:  $\beta$ -cells from *SOFIA/INS-eGFP* animal.

SOFIA



WT

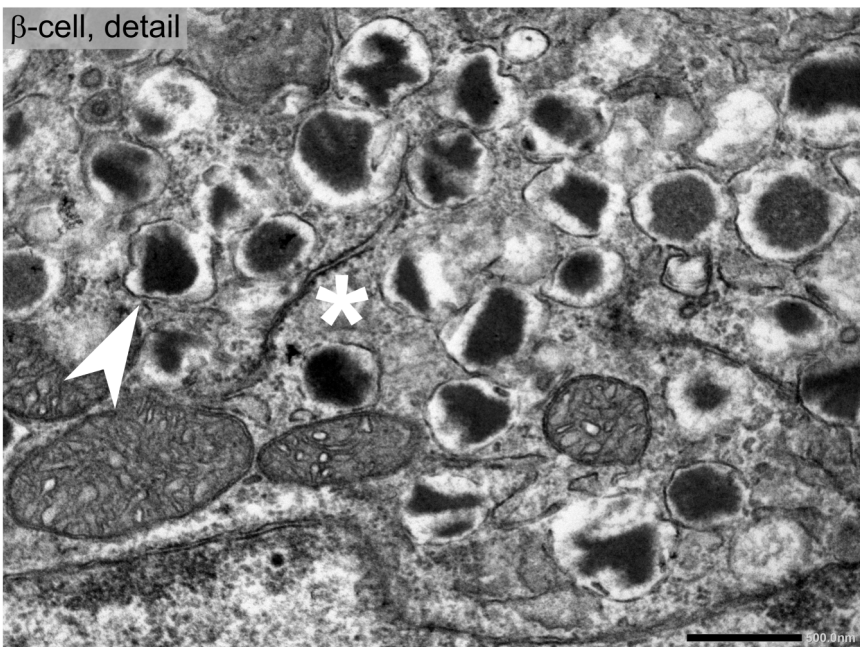
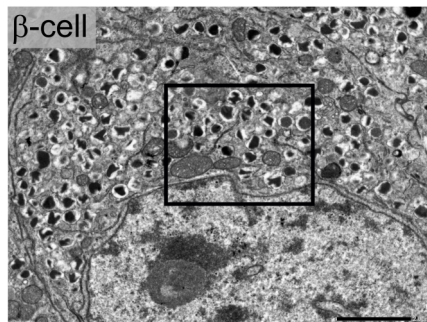
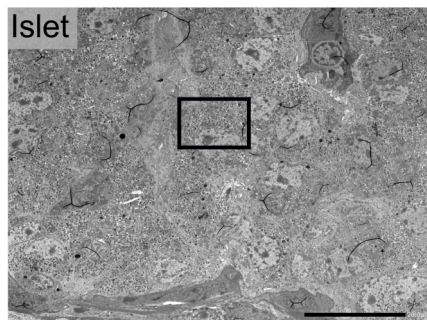


Fig. S4. Comparison of *SOFIA*  $\beta$ -cell ultrastructure with that of WT pigs. TEM of *SOFIA* pig pancreas and WT pig pancreas with overview of islets of Langerhans and zoom into  $\beta$ -cells and subcellular compartments. Arrowheads point to insulin SGs, asterisks point to endoplasmic reticulum. Scale bars: Islet: 20  $\mu$ m,  $\beta$ -cell: 2  $\mu$ m, detail: 0.5  $\mu$ m.

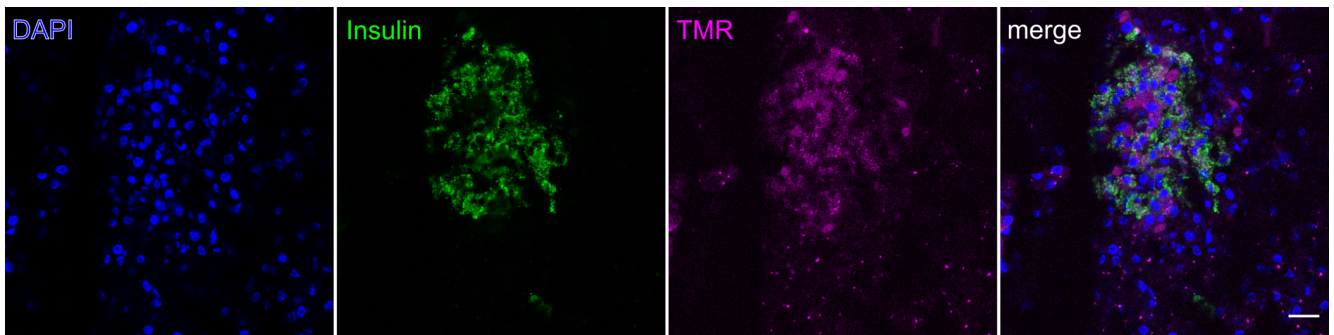


Fig. S5. Immunolabeling against insulin on cryosections of *SOFIA* pig pancreas labeled with TMR. Scale bar: 20  $\mu$ m.