

# Supporting Information

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## SI Experimental Procedures

**Plant Materials.** A total of 70 rice (*Oryza sativa* L.) varieties were selected for association analysis, most of which used to be or currently are widely grown as major cultivars in China. Thirty-three *indica* and thirty-seven *japonica* cultivars were selected for study: 02428, 2070, **9308**, **93-11**, 98110, IAPAR9, C418, Cbao, Ce64-7, Changyou94, Chunjiang025, **Chunjiang06**, Gang46B, Guanglingxiangnuo, Guangluai4, Gui99, **Guichao2**, **Guixiangsinuo**, Huangjinqing, IR24, IR36, IR64, IRBB13, IRBB5, IRBB50, Ji86-11, Jiahua1, **Jiangzhouxiangnuo**, Jiazao935, Jingxi17, Juanguang, **Kasalath**, Kinmaze, Liantangzao, **LongtefuB**, Lunhui422, **Minghui63**, Miyang46, Nanfengnuo, Nanjing6, **Nipponbare**, Nongken58, Qiufengnuo, Sengkeu, Shaonuo, **Suyunuo**, **Taihunuo**, Taiwannuo, **TaichungNative1**, Taichungsen46, Teqing, **Wyunjing7**, XieqingzaoB, Xiushui04, Xiushui11, Xiushui110, Xiushui63, Yangguang, Yongjing04, Yongyou1, Koshihikari, Zhefu802, Zhaiyeqing8, Zhendao5125, **Zhenshan97B**, Zhong9B, Zhongchao123, **Zhonghan3**, Zhongjian100, Zhongxiangnuo. Of these, 16 varieties (indicated in boldface) were further chosen as core germplasms because of their grain ECQs, and the entire genomic sequences, including 1.5- to 2.0-kb 5' upstream and 1.0- to 1.5-kb 3' downstream regions, of the 18 SSRGs in each core variety were fully sequenced. Rice plants were grown in the experimental fields of the China National Rice Research Institute at Hangzhou or at Hainan during the natural growing seasons in 2005 and 2006.

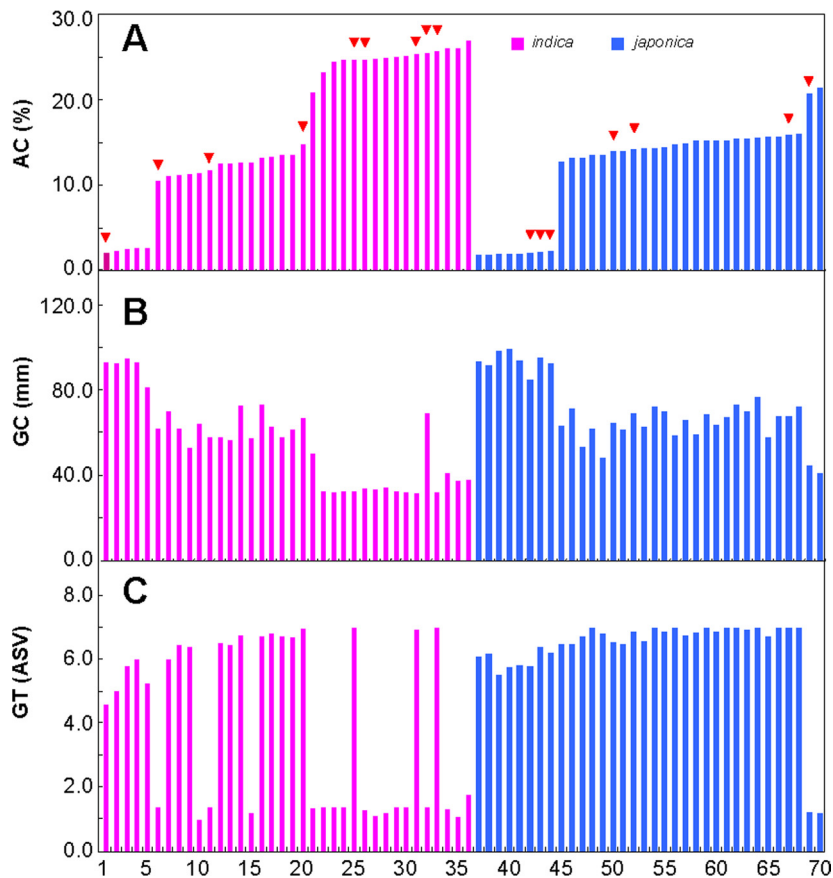
**DNA Preparation and Sequencing.** Rice genomic DNA was prepared as described in ref. 1 and each SSRG was amplified by PCR with primers as given in [Table S7](#). Sequencing was performed on an ABI 3730 DNA Analyzer.

**SSR Primers for Population Structure Evaluation.** RM428, RM259, RM5, RM128, RM14, RM211, RM475, RM263, RM525, RM489, RM251, RM16, RM520, RM514, RM335, RM471, RM252, RM255, RM122, RM289, RM334, RM587, RM585, RM528, RM412, RM180, RM11, RM336, RM234, RM248, RM38, RM223, RM308, RM264, RM316, RM566, RM242, OSR28, RM205, RM474, RM216, RM258, RM333, RM286, RM332, RM441, RM101, RM519, and RM17.

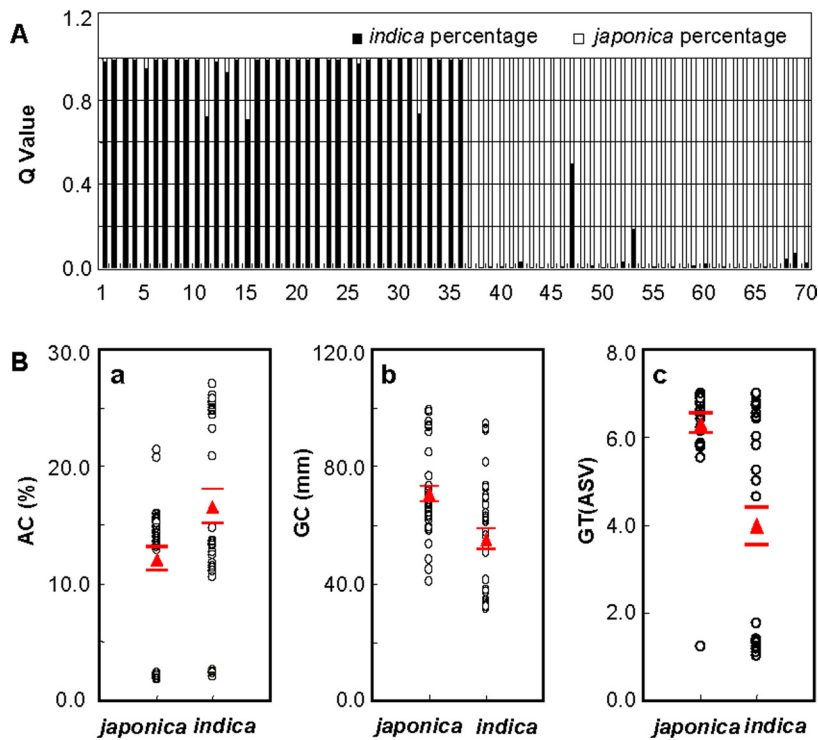
**Protein Blotting Analysis.** The Wx proteins were extracted and purified from same amount of mature grains of different rice lines, as described by Sano (2, 3), separated by SDS/PAGE, and visualized by staining with Coomassie brilliant blue. The total seed protein extracts containing SBEs were prepared as described previously, separated by SDS/PAGE, and electrophoretically transferred onto a nitrocellulose membrane for probing with rabbit polyclonal antibodies against SBE3 (2, 3). A goat anti-rabbit IgG conjugated to alkaline phosphatase was used in the protein blotting to detect the specific SBE3.

1. Hu Y, Bao F, Li J (2000) Promotive effect of brassinosteroids on cell division involves a distinct CycD3-induction pathway in *Arabidopsis*. *Plant J* 24:693-701.
2. Sano Y (1985) Gene regulation at the *Waxy* locus in rice. *Gamma Field Symposia* 24:63-79.

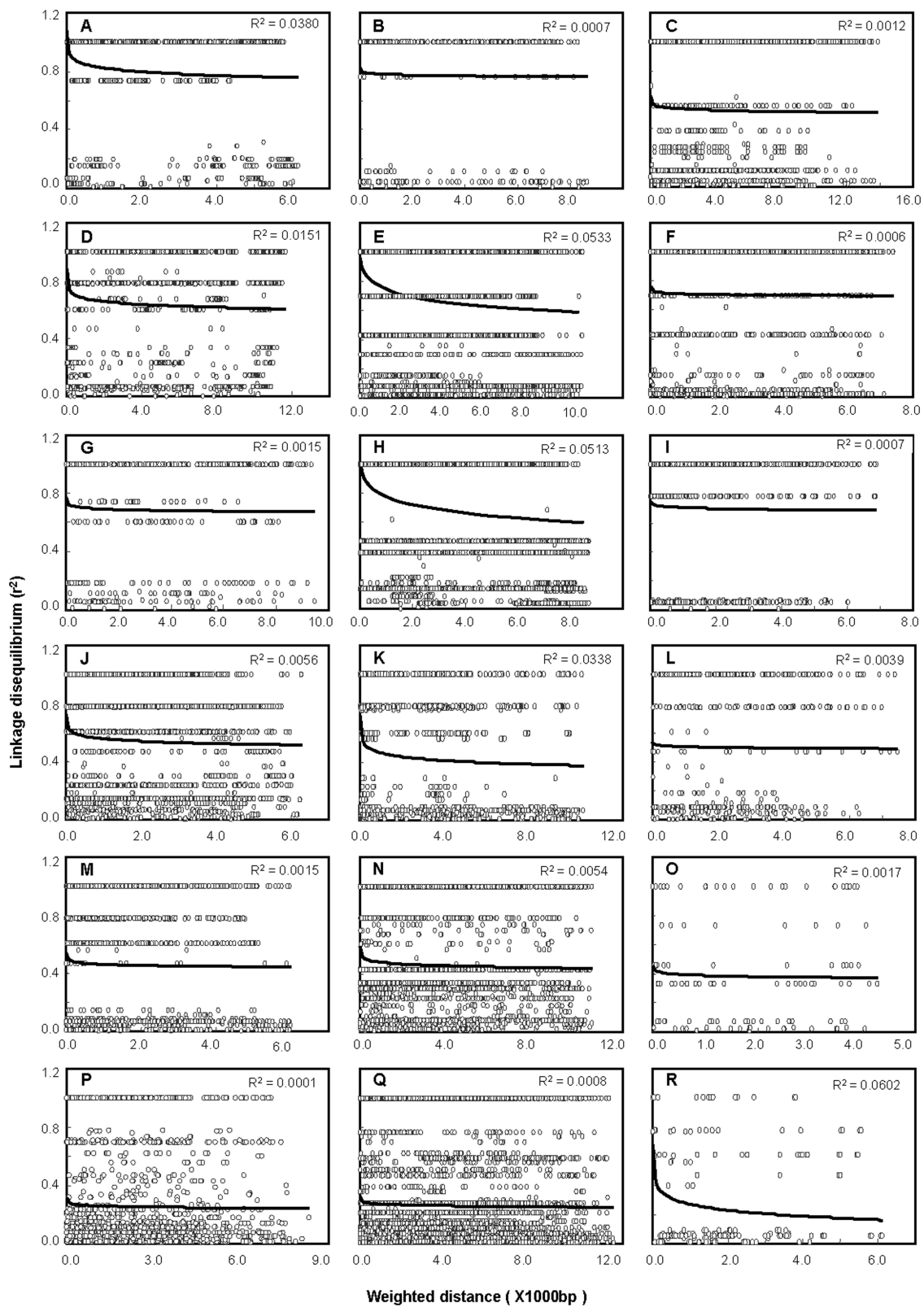
3. Yamagata H, Sugimoto T, Tanaka K, Kasai Z (1982) Biosynthesis of storage proteins in developing rice seeds. *Plant Physiol* 70:1094-1100.



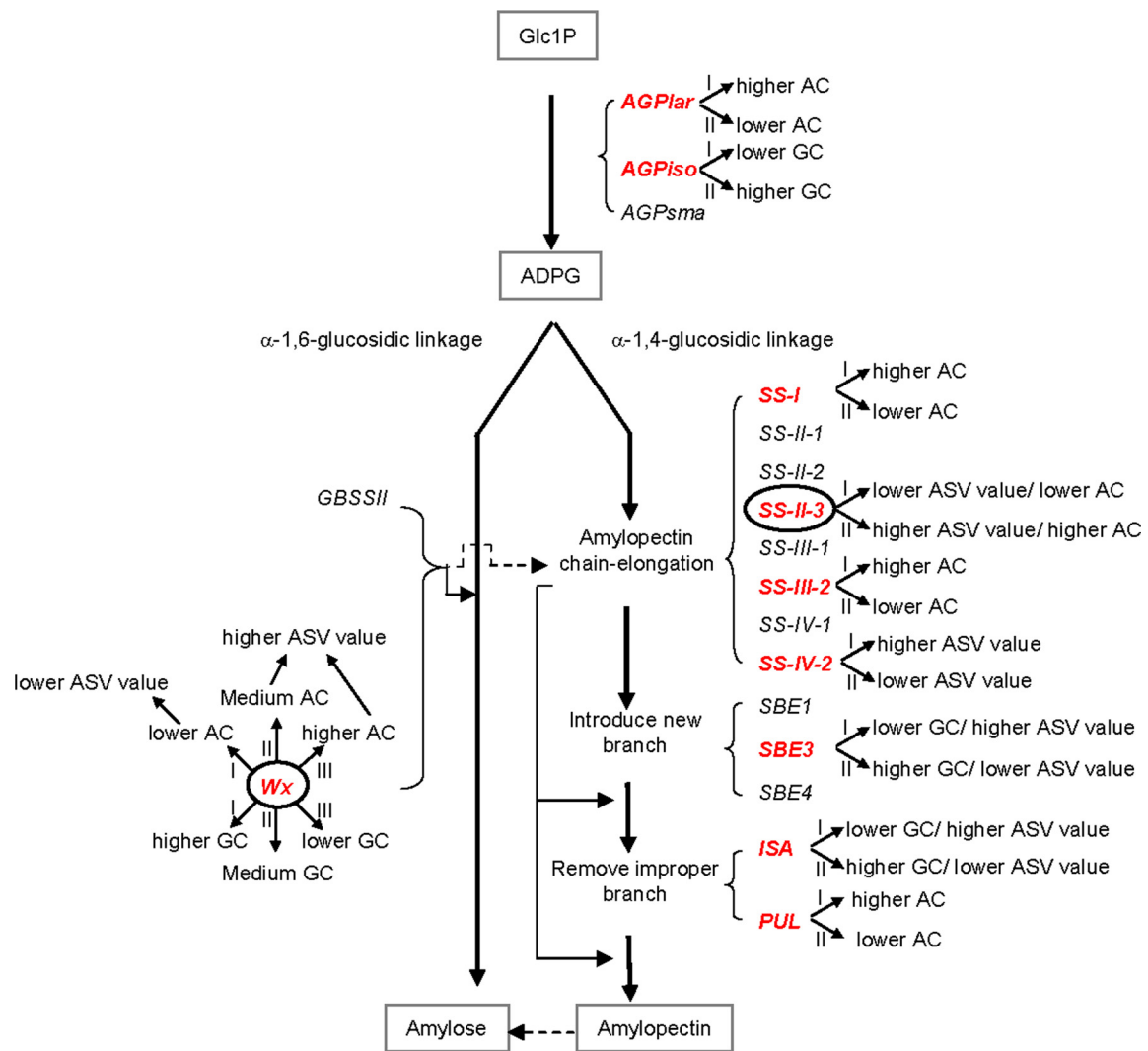
**Fig. S1.** Grain quality of varieties. Grain quality was independently evaluated in 2005 and 2006 for (A) amylose content (AC), (B) gel consistency (GC), and (C) gelatinization temperature (GT) (evaluated as the alkali spreading value, ASV). The surveyed varieties from 1–70 are Guixiangsinuo, Nanfengnuo, Taiwannuo, Ce64-7, Zhongxiangnuo, **Minghui63**, Gui99, Jiazao935, 2070, IRBB50, **9308**, Miyang46, Lunhui422, Zhongjian100, 02428, IR24, IRBB5, IRBB13, C418, **93-11**, IR64, Zhaiyeqing8, Liantangzao, Gang46B, **Taichung Native1**, **Zhenshan97B**, IR36, Guangluai4, Teqing, Zhong9B, **LongtepuB**, **Kasalath**, **Guichao2**, XieqingzaoB, Zhefu802, Nanjing6, Guanglingxiangnuo, Taichungsen46, Qiufengnuo, Shaonuo, Zhendao5125, **Suyunuo**, **Taihunuo**, **Jiangzhouxiangnuo**, Yangguang, Huangjinqing, Cbao, Koshihikari, Zhongchao123, **Nipponbare**, Juanguang, **Chunjiang06**, Changyou94, Xiushui04, Kinmaze, Jiahua1, Jingxi17, Xiushui110, Yongjing04, Ji86-11, Xiushui11, 98110, Xiushui63, Chunjiang025, Sengkeu, Nongken58, **Wuyunjing7**, Yongyou1, **Zhonghan3**, IAPAR9. Variety names in boldface are core varieties, which are marked with a red triangle in A.



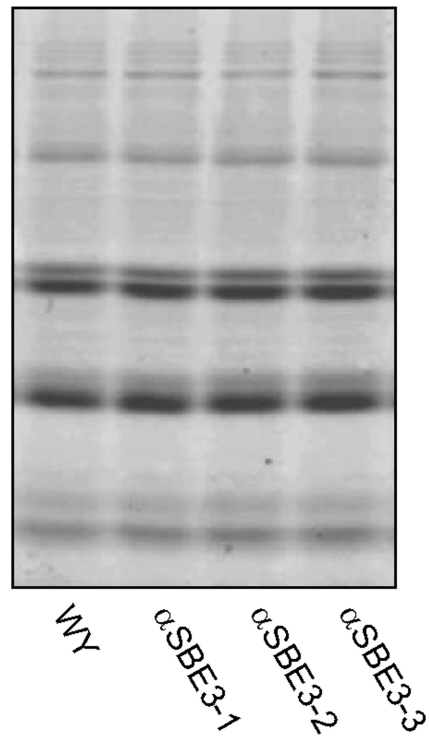
**Fig. S2.** Population structure and effect on rice ECQs. (A) Population structure of the surveyed varieties. (B) Effect of population structure on ECQs. (a) The AC difference between *indica* and *japonica*. The AC mean of *indica* was significantly higher than that of *japonica*. (b) The GC difference between *indica* and *japonica*. The GC mean of *indica* was significantly lower than that of *japonica*. (c) The GT difference between *indica* and *japonica*. The GT mean of *indica* was significantly lower than that of *japonica*. The red triangles and lines are means ± SE. The varieties in (A) from 1–70 are Guixiangsinuo, Nanfengnuo, Taiwanuo, Ce64-7, Zhongxiangnuo, **Minghui63**, Gui99, Jiazao935, 2070, IRBB50, **9308**, Miyang46, Lunhui422, Zhongjian100, 02428, IR24, IRBB5, IRBB13, C418, **93-11**, IR64, Zhaiyeqing8, Liantangzao, Gang46B, **TaichungNative1**, **Zhenshan97B**, IR36, Guangluai4, Teqing, Zhong9B, **LongtefuB**, **Kasalath**, **Guichao2**, XieqingzaoB, Zhefu802, Nanjing6, Guanglingxiangnuo, Taichungsen46, Qiufengnuo, Shaonuo, Zhendao5125, **Suyunuo**, **Taihunuo**, **Jiangzhouxiangnuo**, Yangguang, Huangjinjing, Cbao, Koshihikari, Zhongchao123, **Nipponbare**, Juanguang, **Chunjiang06**, Changyou94, Xiushui04, Kinmaze, Jiahua1, Jingxi17, Xiushui110, Yongjing04, Ji86-11, Xiushui11, 98110, Xiushui63, Chunjiang025, Sengkeu, Nongken58, **Wuyujing7**, Yongyou1, **Zhonghan3**, IAPAR9. Boldface indicates core varieties.



**Fig. S3.** Linkage disequilibrium (LD) decay in the 18 starch synthesis-related genes. Each LD, which is indicated by squared correlations of allele frequencies ( $r^2$ ), shows a nonlinear regression of  $r^2$  against weighted distance between polymorphic sites. (A) *Wx*, (B) *SBE1*, (C) *SBE3*, (D) *SBE4*, (E) *SSI*, (F) *SSII-3*, (G) *ISA*, (H) *PUL*, (I) *SSII-2*, (J) *GBSSII*, (K) *SSIV-1*, (L) *SSIV-2*, (M) *AGP<sub>sma</sub>*, (N) *SSII-1*, (O) *AGPI<sub>lar</sub>*, (P) *SSIII-1*, (Q) *SSIII-2*, and (R) *AGPI<sub>so</sub>*.



**Fig. S4.** Associated genes affecting eating and cooking quality in the rice starch biosynthesis pathway. The genes marked in red were significant. Two major genes, *Wx* and *SSI-3*, were circled.



**Fig. S5.** Loading controls of protein blotting analyses. Total protein was extracted from the wild-type (lane WY), *SBE3* RNAi transgenic plants (lanes  $\alpha$ SBE3-1 to  $\alpha$ SBE3-3), separated by SDS/PAGE, and stained with Coomassie brilliant blue.

## Other Supporting Information Files

- [Table S1 \(PDF\)](#)
- [Table S2 \(PDF\)](#)
- [Table S3 \(PDF\)](#)
- [Table S4 \(PDF\)](#)
- [Table S5 \(PDF\)](#)
- [Table S6 \(PDF\)](#)
- [Table S7 \(PDF\)](#)