14. *wavy leaf 1* causes depressed growth of above- and under-ground organs in rice

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Although a large number of mutants have been identified, almost nothing is known about how growth and development are regulated in rice. From a M_2 population of cv. Taichung 65 chemically mutagenized with N-methyl-N-nitrosourea, we have identified two allelic mutants that showed pleiotropic defects in the development of various organs. Since the characteristics of the mutants differed from those reported to date, we designated them *wavy leaf 1-1* (*waf1-1*) and *waf1-2*. The *waf1* exhibited abnormalities throughout the life cycle. The mature embryo showed underdeveloped coleoptile, malformed leaf primordia and a slightly flattened SAM (Fig. 1A, B). At two weeks after germination, about a half of the seedlings had died. The rest of the seedlings were dwarf and formed aberrant leaves (Fig. 1C, D). The seminal root was rather short, and the number of crown roots was reduced (Fig. 1E). In a rare case, the seminal root stopped growing immediately after germination. Thus, *waf1*is associated with the growth and development of both above- and under-ground organs.

The most remarkable abnormality is observed in leaves. The leaf blade frequently became wavy (Fig. 2A) or infrequently furcated due to unsynchronized growth of lateral two halves (Fig. 2B). In a severe case, a thread-like structure was separated from the middle of the leaf blade (Fig. 2C), being a reminiscent of *sho* leaves (Itoh et al, 2000). In these leaves, the growth of the central region would fail to synchronize with that of lateral region. The cross section revealed that the wavy leaf was defective in cell enlargement and proliferation. The bulliform cells were frequently enlarged and ill-shapen, in addition, normal single cell-file was disturbed locally to form two-cell-layered bulliform cells (Fig. 2D, E). Abnormal differentiation was also observed in the bundle sheath in which cells were increased in number, and arranged rather randomly (Fig.2D, E).

Most of *waf1* plants aborted before heading. Quite rarely, *waf1* formed underdeveloped panicles in which flowers were morphologically abnormal. The above phenotypes indicate that *waf1* has defects in cell proliferation and enlargement of above- and under-ground organs.

To determine the position of *WAF1* locus, we crossed *WAF1/waf1* heterozygote with indica cv. Kasalath. We selected mutant plants from the F_2 population and used them for the mapping. The locus was roughly mapped to the short arm of chromosome 7 between two markers S11633-2 and E12196. Detailed mapping with STS markers confined the gene within 1.8 cM (Fig. 3).

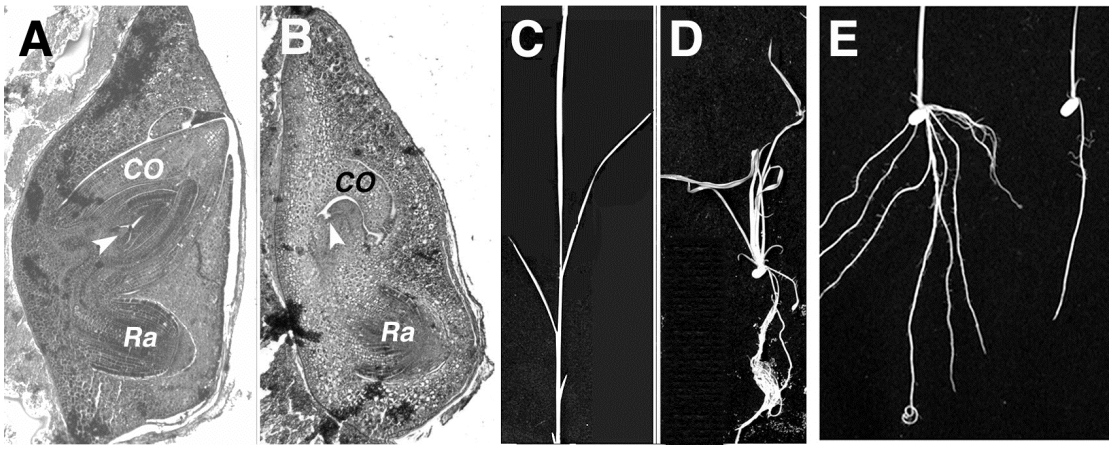

Fig. 1. Phenotype of *waf1-1* mutant. (A) Mature wild-type embryo. (B) Mature *waf1-1* embryo. (C) Two-week-old wild-type seedling. (D) Two-week-old *waf1-1* seedling. (E) Root phenotype of wild type (left) and *waf1-1* (right). Arrowheads indicate SAM. CO: coleoptile, Ra: radicle.

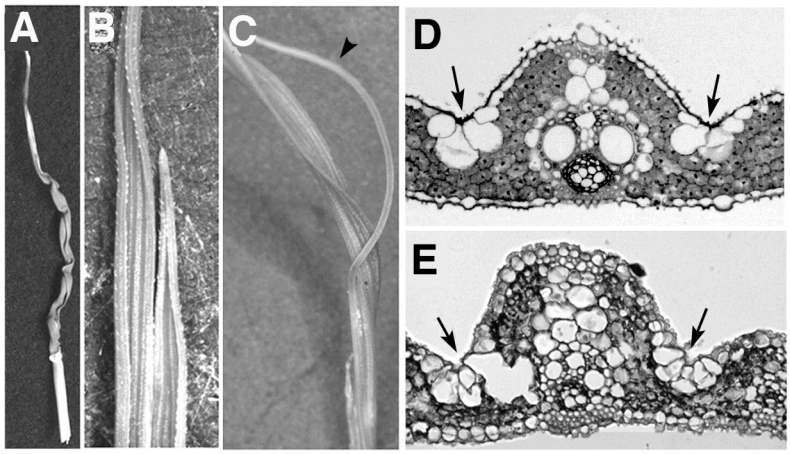

Fig. 2. Leaf phenotypes of *waf1-1*. (A) Wavy leaf. (B) Bifurcated leaf. (C) Separation of thread-like structure from leaf blade. (D) Cross section of wild-type leaf. (E) Cross section of *waf1-1* leaf. Arrowhead indicates thread-like structure, and arrows bulliform cells,

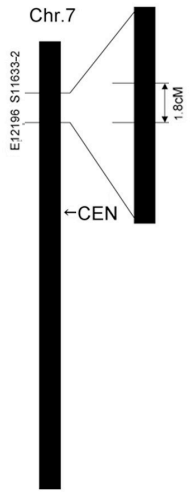

Fig. 3. Map position of *WAF1* locus.

References

Itoh, J.-I., H. Kitano, M. Matsuoka and Y. Nagato, 2000. *SHOOT ORGANIZATION* genes regulate shoot apical meristem organization and the pattern of leaf primordium initiation in rice. Plant Cell 12:2161-2174.