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Better grains for feed; translational miscoding may lead to amino acid enrichment in cereal crops.

Zachary Stone and William Folk

The twenty aminoacyl tRNA synthetases have evolved from either of two distinct progenitors and are distinguishable by their structures and tRNA binding domain. The extant lysyl synthetases (KRS) are an exception, in that most archaeobacteria and some bacteria express a KRS related to one progenitor but in all known eukaryotes, including plants, the KRS has evolved from another progenitor. We propose to exploit these differential requirements for charging so as to cause targeted and regulated recoding by altered tRNA^{Lys} in plants. We have demonstrated that translational recoding by altered tRNA^{Lys} will cause nutritional enrichment of seed storage proteins of rice and maize (Wu et al, 2003; Wu et al, 2007). However, the amount of recoding required to achieve sufficient enhancement to satisfy human nutritional requirements may exceed the tolerance for translational recoding in vegetative and reproductive tissues, there by reducing agronomic traits. Consequently, the utilization of the altered (Bb)tRNA^{Lys} must be targeted to endosperm, using endosperm specific promoters to express the BbKRS. The BbKRS gene has been modified to contain preferred plant codons and to remove many processing signals important in eukaryotes; it must be placed under the control of CaMV35S promoter and endosperm specific promoters and sequences confirmed. A Flag tag was place at the C-terminal of all constructs for detection of expression. Concomitantly, the (Bb)tRNA^{Lys} placed surrounded by plant tRNA flanking sequences has been constructed so as to permit expression in plants (Ulmasov and Folk, 1995) and expression in plant protoplasts will be examined by Northern blotting.