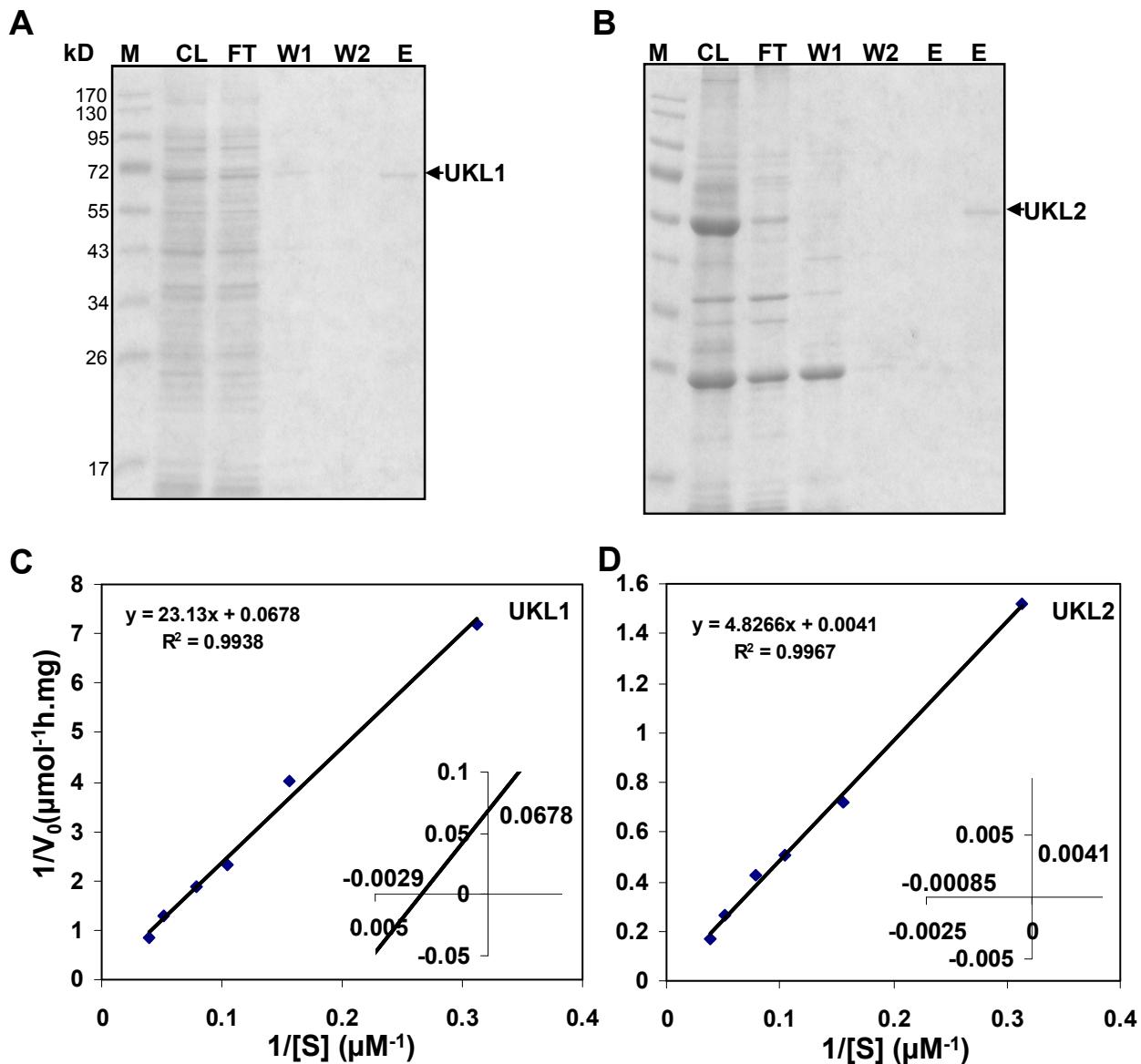
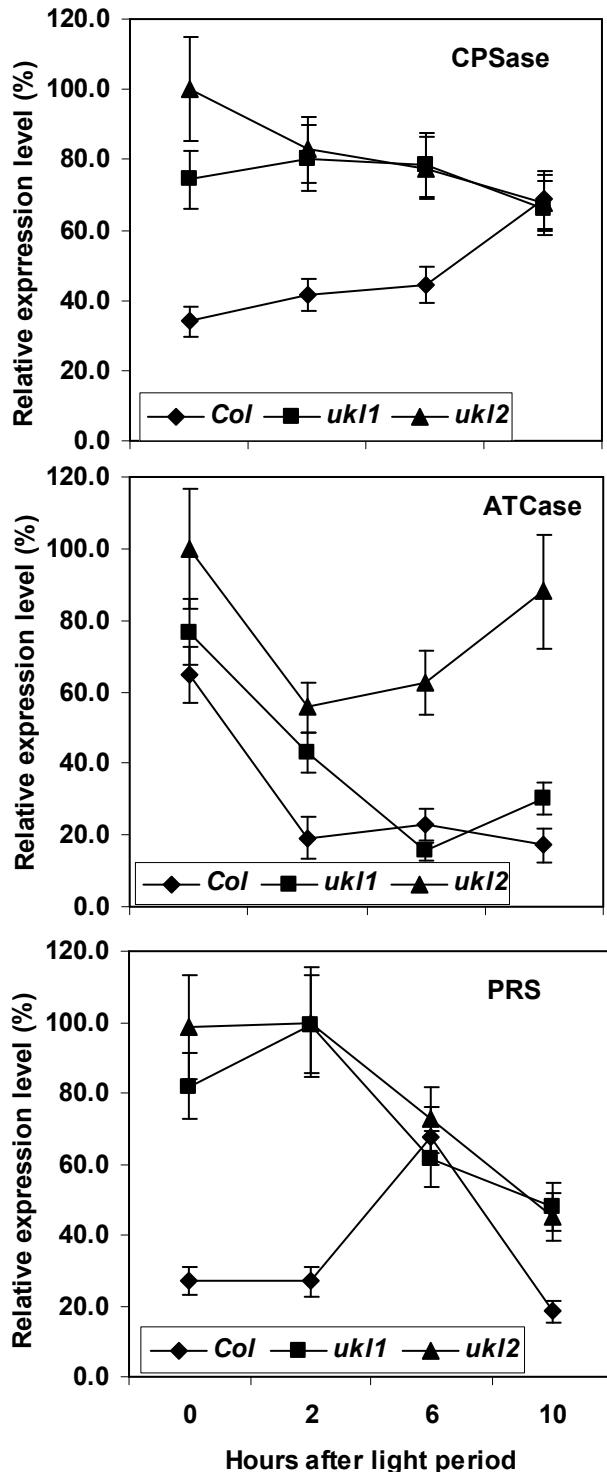


Supplemental Figure 1. *ukl1* and *ukl2* mutant identification. **A**, Schematic representation of T-DNA insertion positions in At5g40870 and At3g27190. LB, T-DNA left border. **B**, *UKL1* gene expression was knocked out in SALK_108468 and *UKL2* expression was highly reduced in SALK_058257 T-DNA insertion lines. RT-PCR was used to detect gene expression and *UBQ* was used as the loading control.

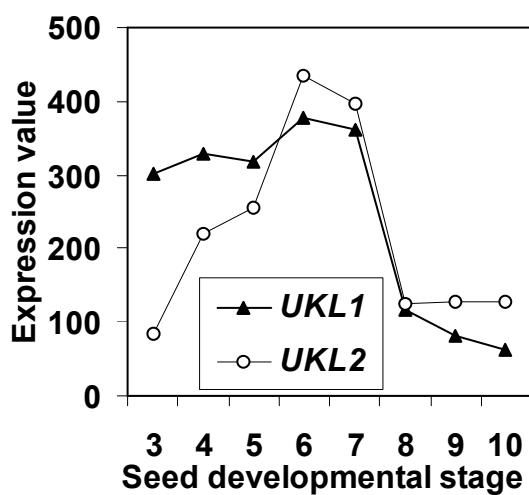


Supplemental Figure 2. Recombinant UKL1 and UKL2 protein kinetic assay. *A, B*, SDS-PAGE Coomassie staining results of Ni-NTA purification of His-tagged recombinant UKL proteins. M, marker bands showing protein size in kilodaltons; CL, crude lysate; FT, flow through; W1 and W2, wash steps 1 and 2; E, eluate. Recombinant proteins are marked with arrows. *C, D*, Lineweaver-Burk plot of UKL1 and UKL2.



Supplemental Figure 3. Transcriptional Expression of Genes in the *de novo* Pyrimidine Biosynthesis Pathway is Altered in the *ukl1* and *ukl2* Mutant.

The signal was normalized to *UBQ* internal standard from three biological replicates, the highest signal is set as 1. CPSase, carbamoylphosphate synthase; ATCase, aspartate transcarbamoylase; PRS, PRPP synthetase.



Supplemental Figure 4. *UKL1* and *UKL2* are Differentially Expressed during Seed Development.
The microarray expression data of *UKL1* and *UKL2* across different seed developmental stages from the public domain is used.

Supplemental Table 1. Primers used in this study

Primer	Sequence (5' to 3')	Target
1	ATGCAAGTGCCTACTGTCG	UKL1
2	TTACCGCCTGCTGCAAAGGA	UKL1
3	GAAGGATCCATGCCCGAAGATTCTTCT	UKL1
4	GGGCGGCCGCCTGGTCTCCTCGTCGGT	UKL1
5	CACCATGCCCGAAGATTCTTCT	UKL1
6	CTACTGGTCTCCTCGTCGGT	UKL1
7	ATGTGGCTTCAGGTACTCCT	UKL2
8	ACACTGAGCAGCTTGCATT	UKL2
9	GAAGGATCCATGCCAGAAGATTCAACG	UKL2
10	GGGCGGCCGCCTCGTCGGTACCAAAGTA	UKL2
11	CACCATGCCAGAAGATTCAACG	UKL2
12	CTACTCGTCGGTACCAAAGTA	UKL2
13	ACGCTGAATGGGATTTCAAC	At3g27740
14	TGAGCACTGATCTCCACCTG	At3a27740
15	CCATTGCTAACGACCCAGAT	At1g29900
16	CAAAGCGATTGCTTGTC	At1g29900
17	ACTCCCTCTCCTGCTCTCC	At1g75330
18	AACACGAGCACATCACGAG	At1g75330
19	ATTGAAGGGTATGCCTCGTG	At3g54470
20	GGGTTTCCGCCTTATGAT	At3g54470
21	TGGTCCAGGAAGTGGAAAAG	At5g26667
22	CCTCATTACGAGGGAAACCA	At5g26667
23	TCCGTCTTCTCTCGCAAT	At5g63310
24	TTCCAAGCCATACACACAA	At5g63310
25	CATCAAGTGCAGTCAGAA	At1g10700
26	CGAAGATCACAGCAGGTGAA	At1g10700
27	AGGTTGCGATGTCTTTGG	At1g32380
28	CCTGGCTACTCCACCAACAT	At1g32380
29	GGACCTGGGAAAATCAAG	At2g44530
30	CGGAGTGAAGATCACAAGCA	At2g44530