

Plant Gene Register

Nucleotide Sequence of a cDNA Encoding Starch-Branching Enzyme, or Q-Enzyme I, from Rice Endosperm¹

Yasunori Nakamura* and Hiroaki Yamanouchi

National Institute of Agrobiological Resources and National Institute of Sericultural and Entomological Science,
Tsukuba, Ibaraki 305, Japan

The programmed regulation of carbohydrate-metabolizing enzymes is associated with an increase in the production of starch in rice endosperm (4, 5). The increase in starch-branching enzyme, or Q-enzyme, plays a critical role in the initiation and regulation of the biochemical process of starch accumulation in rice endosperm (4) (Table I). Q-enzyme catalyzes the synthesis of amylopectin by introducing the α -1,6-glycosylic linkage from α -1,4-glucan. QEI² is the predominant form of the two isozymes, which are distinguishable immunologically from each other in developing rice endosperm (3).

Figure 1 shows the nucleotide sequence of the rice endosperm QEI cloned from a λ gt11 cDNA library and the deduced amino acid sequence. The site of translation initiation is presumed to be at nucleotide sequence 30 of the sequence because (a) the rice endosperm QEI-40 cDNA is a full-length cDNA, or very close to it, and (b) the size (about 90 kD) of the *in vitro* translated product from QEI mRNA, as estimated from SDS-PAGE, is similar to a *M_r* of 93,258 (open reading frame of 820 amino acids) for the QEI protein predicted from its cDNA sequence. The nucleotide and predicted protein sequences of QEI of rice endosperm exhibit little similarity with those of Q-enzyme from bacteria, *Escherichia coli* (2) and *Synechococcus* (1). A leader sequence for amyloplast import remains to be determined. The amino terminal sequence of the native QEI could not be determined by microsequence analysis because the N-terminal amino acid of the native enzyme is modified.

We observed that Q-enzyme, as well as ADP-glucose pyrophosphorylase, increases in activity at about the same time when active starch synthesis is occurring in rice endosperm, whereas the bulk of the other enzymes involved in carbohydrate metabolism accumulates during the earlier phase of the endosperm development (4). Therefore, it is postulated that QEI plays a key role in the efficient production of starch in developing rice endosperm.

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² Abbreviation: QEI, Q-enzyme I.

Table I. Characteristics of the QEI cDNA from Rice Endosperm

Organism:	<i>Oryza sativa</i> L. (rice), cv Fujihikari (<i>japonica</i> subspecies).
Location on Chromosome:	Nuclear genome; chromosome location not known.
Function:	Encodes starch-branching enzyme or Q-enzyme (α -1,4 glucan: α -1,4 glucan 6-glucosyltransferase (EC 2.4.1.18).
Subcellular Location:	Amyloplasts.
Techniques:	cDNA library in λ gt11, restriction fragment subcloning into Bluescript (Stratagene), and dideoxy sequencing of both strands.
Method of Isolation; Subsequent Identification:	cDNA library was screened with antiserum to QEI from rice endosperm. Full-length rice endosperm QEI cDNA clones were identified by rescreening the isolated clones with the cDNA insert produced by the polymerase chain reaction method by using oligonucleotides identical with segments of the amino acid sequences of QEI purified from rice endosperm (3).
(G + C) Content:	45.1%.
Expression Characteristics:	QEI is involved in developmental and tissue-specific expression (5).
Antibodies:	Raised in rabbit against rice endosperm QEI (3).
EMBL Accession No.:	D10752.

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1	CG CCT CCA CGG CCA CCG ACA TCC GCC GCA ATG CTG TGT CTC ACC TCC TCT TCC TCC TCC GCG CCG CCT CCG CTC CTT CCC TCT	83
1		18
84	CTC GCT GAT CGA CCG AGC CCG GGA ATC GCG GGC GGG GGT GGC AAT GTT CGC CTG AGC GTG GTT TCT TCG CCG CGC CGG TCG TGG	167
19	L A D R P S P G I A G G G G G N V R L S V V S S P R R S W	46
188	CCT GGA AAG GTC AAG ACC AAT TTC TCA GTT CCT GCG ACT GCG CGA AAA AAC AAA ACC ATG GTG ACT GTT GTG GAG GAG GTC GAC	251
47	P G K V K T N F S V T A R K N K T M V T V V E N S K	74
252	CAC CTT CCT ATA TAT GAT CTG GAC CCT AAG TTG GAG GAA TTC AAG GAT CAC TTC AAC TAT AGG ATA AAA AGA TAC CTC GAC CAG	335
75	H L P I Y D L D P K L E E P K D H F N Y R I K R Y L D Q	102
338	AAA TGC CTG ATT GAA AAA CAT GAG GGG GGC CTT GAA GAA TTT TCT AAA GGC TAT TTG AAG TTT GGG ATT AAT ACA GTT GAT GGT	419
103	K C L I E K H E G G I S K G Y L K P G I N T V D G	130
420	GCC ACA ATA TAT CGT GAA TGG GCG CCT GCT GCA CAA GAA GCA CAG CTC ATT GGT GAG TTC AAT AAC TGG AAT GGT GCA AAA CAC	503
131	A T I Y R E W A P A Q E A Q L I G E P N N W N G A K H	158
504	AAG ATG GAG AAG GAT AAA TTT GGC ATT TGG TCA ATC AAG ATT TCA CAT GTC AAT GGG AAG CCT GCC ACT CCT CAC AAT TCC AAG	587
159	K H E K D K P G I W S I K I S H V N K T M V I P H N S K	186
588	GTT AAA TTT CGC TTT AGG CAT GGG GGT GGA GCA TGG GTT GAT CGT ATT CCC GCA TGG ATT CGT TAT GCA ACT TTT GAT GCC TCT	671
187	V K F R F R H G G G A W V D R I P A W I R Y A T F D A S	214
872	AAA TTT GGA GCT CCA TAT GAT GGT GTA CAC TGG GAT CCT CCA GCC TGT GAA AGG TAC GTG TTT AAG CAT CCT CGA CCT CCA AAA	755
215	K F G A P Y D G V H W D P A C E R Y V P K H P R P D K	242
756	CCT GAT GCT CGA CGC ATC TAT GAG GCT CAT GTG GGG ATG AGT GGT GAA GAG CCA GAA GTA AGC ACA TAC AGA GAA TTT GCA GAC	839
243	P D A P R I Y E A H V G N S G E E P E V S T Y R E F A D	270
840	AAT GTG TTA CCA CGC ATA CCG GCA AAT AAC TAC AAC ACA GTT CAG TTA ATG GCA ATC ATG GAA CAT TCG TAC TCT TTT	923
271	N V L P R I R A N N Y N T V Q L H A I M E H S Y Y A S F	298
924	GGG TAT CAC GTG ACA AAT TTT TTC GCA GTC AGC AGC AGA TCA GGA ACA CCA GAG GAT CTG AAA TAT CTT GTT GAC AAG GCA CAT	1007
289	G Y H V T N P P A V S S R S G T P E D L K Y L V D A H	326
1008	AGT TTA GGA TTA CGA GTT CTG ATG GAT GTT GTC CAT AGC CAT GCG AGT AAT AAT GTG ACC GAT GGT CTA AAT GGC TAT GAC GTT	1091
327	S L G L R V L M D V V H S H A S N N V T D G L N G Y D V	354
1092	GGA CAA AAC ACT CAT GAG TCT TAT TTT CAT ACA GGA GAT AGG GGC TAC CAT AAA CTC TGG GAT AGT CGT TTC TTG AAC TAT GCC	1175
355	Q N T H E S Y P H T G Y K L W D S R L F Y A	382
1176	AAT TGG GAG GTC TTA AGA TTT CTT TCT AAT TTG AGA TAT TGG ATG GAC GAA TTC ATG TTT GAT GGC TTC CGA TTT GAT GGG	1259
383	N W E V L R P L L S N L R Y W M D E F M F D G F R P D G	410
1280	GTT ACA TCA ATG CTA TAC CAT CAC CAT GGT ATC AAT AAG GGA TTT ACT GGA AAC TAC AAG GAG TAT TTC AGT TTG GAT ACC GAT	1343
411	V T S M L Y H H H G I N K G P T G N Y K E L Y P S L V D T D H L R Y K M A	438
1344	GTG GAT GCA AIT GTT TAC ATG ATG CTC GCA AAC CAT TTA ATG CAT AAA CTC TTG CCG GAA GCA ACT ATT GTT GCT GAA GAT GTT	1427
439	V D A I V Y M M L A N H L M H K L L P E A T I V A E D V	466
1428	TCG GGC ATG CCA GTG CTT TGT CCG CCA GTT GAT GAA GGT GGA GTA GGG TTT GAC TTC CGC CTG GCA ATG GCC ATT CCT GAT AGA	1511
467	S G M P V L C R P V D E G G G P D F R L A M A T G R R Y D	494
1512	TGG ATT GAC TAC GTG AAG AAC AAA GAG GAC CCG AAA TGG TCA ATG AGT GAA ATA GTG CAA ACT TTG ACT AAC AGG AGA TAT ACA	1595
495	W I D Y L R N K E D Q S I V G D K T I A F L L M D K E	522
1596	GAA AAA TGC ATT GCC TAT GCC GAG AGC CAT GAT CAG TCC ATT GTT GGT GAC AAG ACT ATA GCA TTT CTC TTG ATG GAC AAG GAA	1879
523	E K C I A Y A E S H D Q S I V G D K T I A F L L M D K E	550
1880	ATG TAC ACT GGC ATG TCA GAC TTG CAG CCT GCT TCA CCT ACC ATC AAC CGT GGC ATT GCA CTC CAA AAG ATG AIT CAC TTC ATT	1763
551	M Y T G M S D L Q P T I N R G I A L Q K M I H P I	578
1784	ACG ATG GCC CTT GGA GAT GGT GGC TAC TTA AAT TTT ATG GGC AAT GAG TTT GGC CAT CCA GAA TGG ATT GAC TTT CCA AGA GAA	1847
579	T M A L G G D G Y L N P M G N E P G H P E W I D F P R E	606
1848	GGC AAC AAC TGG AGC TAT GAT AAA TGC AGA CGT CAG TGG AGC CTT GTC GAC ACT GAT CAC CTT CGA TAC AAG TAT ATG AAT GCA	1931
607	G N N W S Y D K C R R R W S L V D T D H L R Y K M A	634
1932	TTT GAT CAA GCA ATG AAT GCA CTC GAG GAG GAA TTT TCC TTC CTG TCA TCA TCA AAG CAG ATT GTT AGC GAC ATG AAC GAG AAA	2015
635	F D Q A M N A L E E E P S P L S S S K Q I V S D H N E K	662
2016	GAT AAG GTT AIT GTC TTT GAA CGT GGA GAT TTG GTT TTT GTT TTC AAT TTT CAT CCC AAC AAA ACT TAC AAG GGT TAC AAA GTC	2099
663	D K V I V F E R G D L V F V P N P H P N K T Y K G Y K V	690
2100	GGA TGT GAC TTG CCC GGG AAG TAC AGA GTA GCT CTG GAC TCT GAT GCT TTG GTC TTT GGT GGC CAT GGA AGA GTT GGC CAT GAT	2183
691	G C D L P G K Y R V A L D S D A L V P G G H G R V G H D	718
2184	GTG GAT CAC TTC ACG TCT CCC GAG GGA ATG CCA GGA GTA CCA GAA ACA AAT TTC AAC AAC CGC CCT AAC TCA TTC AAA GTC CTT	2267
719	V D H P T S P E G M P G V P E T N P N N R P N S P K V L	746
2268	TCC CCG CCC CGT ACC TGT GTG GCT TAC TAT CGC GTT GAT GAA GAT CGT GAA GAG CTC AGG AGG GGT GGA GCA GTT GCT TCT GGA	2351
747	S P P R T C V A E D R E L R R R G G A V A S G	774
2352	AAG ATT GTT ACA GAG TAT ATC GAT GTT GAA GCA ACA AGT GGG GAG ACT ATC TCT GGT GGC TGG AAG GGC TCC GAG AAG GAC GAT	2435
775	K I V T E Y I D V E A T S G E T I S G G W K G S E K D D	802
2436	TGT GGC AAG AAA GGG ATG AAG TTT GTG TTT CGG TCT TCT GAC GAA GAC TGC AAA TGA AGC ATC AGA TTT CTT GAT CAG GAG CAA	2519
803	C G K K G M K P V P R S S D E D C K	820
2520	CTG TTG GTG CCC TTG TAA TCT GGA GAT CCT GGC TTG CCT TGG ACT TGG TTG TGG TTC TTT AGC AGT TGC TAT GTA CCT ATC TAT	2603
2804	GAT ATG AAC TTT ATG TAT AGT TCG CCT TAA AGA AAG AAT AAG CAG TGA TGA TGT GGC CTT AAA CCT GAG CTG CAC AAG CCT AAT	2687
2688	GTA AAA ATA AAG TTT CAG GCT TTC ATC CAG AAT AAA ACA GCT GTT CAT TTA CCA AAA AAA AAA AAA AA	2758

Figure 1. Nucleotide sequence and its amino acid sequence of the rice endosperm QEI-40 cDNA encoding rice endosperm starch-branching enzyme. The open reading frame starts at nucleotide 30 and ends at 2489. The short black lines under the sequence indicate the amino acid sequences determined by actual peptide sequencing. The location of probe used as cDNA insert for identification of rice endosperm QEI clones is underlined.