# Nucleotide Sequence of the *Rhodobacter capsulatus fruK* Gene, Which Encodes Fructose-1-Phosphate Kinase: Evidence for a Kinase Superfamily Including Both Phosphofructokinases of *Escherichia coli*

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The fruK gene encoding fructose-1-phosphate kinase (FruK), located within the fructose (fru)-catabolic operon of Rhodobacter capsulatus, was sequenced. FruK of R. capsulatus (316 amino acids; molecular weight = 31,232) is the same size as and is homologous to FruK of Escherichia coli, phosphofructokinase B (PfkB) of E. coli, phosphotagatokinase of Staphylococcus aureus, and ribokinase of E. coli. These proteins therefore make up a family of homologous proteins, termed the PfkB family. A phylogenetic tree for this new family was constructed. Sequence comparisons plus chemical inactivation studies suggested the lack of involvement of specific residues in catalysis. Although the Rhodobacter FruK differed markedly from the other enzymes within the PfkB family with respect to amino acid composition, these enzymes exhibited similar predicted secondary structural features. A large internal segment of the Rhodobacter FruK was found to be similar in sequence to the domain bearing the sugar bisphosphate-binding region of the large subunit of ribulose 1,5-bisphosphate carboxylase/oxygenase of plants and bacteria. Proteins of the PfkB family did not exhibit statistically significant sequence identity with PfkA of E. coli. PfkA, however, is homologous to other prokaryotic and eukaryotic ATP- and PP<sub>1</sub>-dependent Pfks (the PfkA family). These eukaryotic, ATP-dependent enzymes each consist of a homotetramer (mammalian) or a heterooctamer (yeasts), with each subunit containing an internal duplication of the size of the entire PfkA protein of E. coli. In some of these enzymes, additional domains are present. A phylogenetic tree was constructed for the PfkA family and revealed that the bacterial enzymes closely resemble the N-terminal domains of the eukarvotic enzyme subunits whereas the C-terminal domains have diverged more extensively. The PP<sub>1</sub>-dependent Pfk of potato is only distantly related to the ATP-dependent enzymes. On the basis of their similar functions, sizes, predicted secondary structures, and sequences, we suggest that the PfkA and PfkB families share a common evolutionary origin.

In earlier papers we presented the nucleotide sequences of two of the three structural genes within the Rhodobacter capsulatus fructose (fru) operon. These genes include fruB(HI), encoding a multiphosphoryl transfer protein of the phosphoenolpyruvate:fructose phosphotransferase system (PTS) (46, 50, 61), and fruA, encoding the fructose permease, i.e., the fructose-specific enzyme II of the PTS (60). These studies revealed some unusual features of the Rhodobacter PTS proteins, which together catalyze the concomitant uptake and phosphorylation of fructose to yield fructose-1-phosphate (fructose-1-P). The further metabolism of fructose requires the ATP-dependent phosphorylation of fructose-1-P catalyzed by fructose-1-P kinase (FruK; ATP: D-fructose-1-phosphate-6-phosphotransferase, EC 2.7.1.56). This enzyme has been purified from *Escherichia coli* (7), and after this paper was submitted for publication, the nucleotide sequence of its structural gene in E. coli was published and the homology of the encoded protein with other carbohydrate kinases was noted (43). In this paper we report the complete sequence of the fruK gene encoding FruK of R. capsulatus.

Two phosphofructokinases (Pfks) have been identified in *E. coli*. The major Pfk (PfkA) is a tetramer consisting of four

identical subunits (subunit molecular weight = 35,000) (58). The enzyme shows cooperative kinetics with respect to its substrate, fructose-6-phosphate, and is subject to allosteric control by phosphoenolpyruvate (6). The crystal structures of PfkA and of the Pfk from Bacillus stearothermophilus have been determined (19, 26). These two enzymes, very similar in sequence and structure, are homologous to two internally repeated domains in both the  $\alpha$ - and  $\beta$ -subunits of the ATP-dependent octameric yeast Pfk, the two internally repeated domains of the single type of subunit of the mammalian ATP-dependent Pfks, and the two dissimilar subunits ( $\alpha$  and  $\beta$ ) of the potato PP<sub>i</sub>-dependent Pfk (8, 25, 45). The family of these homologous proteins is here designated the PfkA family. The minor Pfk of E. coli (PfkB) is a homodimer (subunit molecular weight of about 33,000) which can associate to a less active homotetramer. It does not show cooperative kinetics or inhibition by phosphoenolpyruvate (4, 31). It lacks immunological cross-reactivity with PfkA (30) and has not been noted to show sequence similarity with members of the PfkA family (4, 25).

In this report, FruK of *R. capsulatus* is shown to be homologous to FruK and PfkB of *E. coli* as well as other bacterial kinases (the PfkB family). The predicted secondary structures of FruK and PfkB are shown to be similar to the known secondary structure of PfkA. Phylogenetic trees for

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both the PfkA and PfkB families are constructed. The reported results lead us to suggest that the PfkA and the PfkB families share a common origin but diverged from each other long before the genes encoding the individual members of either of these families diverged.

# MATERIALS AND METHODS

Materials. Restriction endonucleases, T4 DNA ligase, and T4 DNA polymerase were obtained from Bethesda Research Laboratories, Gaithersburg, Md., or Boehringer Mannheim Biochemicals, Inc. The T7 polymerase sequencing kit was from Pharmacia. The deoxyadenosine 5'-( $[\alpha^{-35}S]$ thio)triphosphate (1,335 Ci/mmol) was purchased from the New England Nuclear Corp., Boston, Mass. Isopropyl-B-D-thiogalactopyranoside, 5-bromo-4-chloro-3-indolyl-B-D-galactopyranoside (X-Gal), and lysozyme were purchased from Boehringer Mannheim Biochemicals. Acrylamide, bisacrylamide, and N, N, N', N'-tetramethylethylenediamine were from Bio-Rad Laboratories, and agarose was from Bethesda Research Laboratories. Deoxynucleotides and dideoxynucleotides were obtained from Pharmacia and P-L Biochemicals, Inc. All other chemicals and enzymes used were of the highest quality available commercially.

**Bacterial strains, plasmids, and phage.** E. coli TG1 was provided by T. Gibson of the Laboratory of Molecular Biology, Medical Research Council, Cambridge, United Kingdom (9), and strain XL1-Blue was obtained from Stratagene. R. capsulatus 37b4 was as described previously (13). The pBluescript SK(+) and KS(+) plasmids as well as helper phage VCS-M13 were obtained from Stratagene.

Synthesis of oligodeoxynucleotides. Oligodeoxynucleotides were synthesized from  $\beta$ -cyanoethyl phosphoramidite precursors on a model 380B Applied Biosystems DNA synthesizer. The oligonucleotides were synthesized as trityl-on derivatives which were deblocked at 58°C for 12 h in concentrated ammonium hydroxide. These solutions were then applied to Applied Biosystems OPC oligonucleotide purification columns. The oligonucleotides were eluted as described by the manufacturer. These solutions were dried under vacuum.

Growth media and selection conditions. Transformants were selected on Luria-Bertani (LB) plates (Difco Laboratories) (38) containing ampicillin (50  $\mu$ g/ml). For the subcloning of fragments into pBluescript SK(+) or KS(+), LB plates containing 1 mM isopropylthio- $\beta$ -galactoside and 30  $\mu$ g of X-Gal per ml were used.

**DNA procedures.** *E. coli* XL1-Blue and TG1 carrying the pBluescript plasmids were grown at  $37^{\circ}$ C in LB medium containing ampicillin (50 µg/ml). Single-stranded DNA from the Bluescript plasmids was prepared with helper phage VCS-M13 as described by the Stratagene manual. Small- and large-scale preparation of plasmid DNAs was accomplished by the method of Birnboim (5). Competent cells of *E. coli* TG1 and XL1-Blue were prepared by the CaCl<sub>2</sub> method (37).

Nucleotide sequencing was conducted with the T7 sequencing kit of Pharmacia with Deaza <sup>35</sup>Sequencing Mixes as described previously in order to overcome GC compression due to the high GC content of *Rhodobacter* DNA (66% overall; 70% for the *fru* operon). We reported previously the location of the *fruK* gene on an *Eco*RI-*Pst*I fragment of 2.0 kb (61). This fragment was subcloned into plasmids Bluescript SK(+) and KS(+). Universal and synthesized primers were used for the sequencing. The entire fragment was sequenced in both directions at least twice, and all sequences were overlapping in each direction.

Computer analyses. The Staden programs were used for statistical analyses and structural prediction studies (57). For homology screening, the Fasta program (44) was used with the combined NBRF (PIR protein sequence data base release 25.0 plus preliminary entries new release 43.0), a translation of GenBank DNA libraries (release 65.0), and EMBL sequence data base (release 19.0). Multialignment was conducted by employing the Newat program (20). The significance of homology was calculated by using the ALIGN program (28) as well as the Los Alamos program (28). The Genetics Computer Group Sequence Analysis Software Package (GCG Package) was used for calculation of codon usage and determination of open reading frames. These programs are available through the University of California, San Diego, Computer Center. Phylogenetic tree construction was described by Doolittle and Feng (17, 20).

Extract preparation and enzyme inactivation studies. Cells of R. capsulatus were grown in complex medium, and extracts were prepared as described previously (13). FruK was assayed by using a spectrophotometric assay (13). Treatment with N-ethylmaleimide, phenylmethylsulfonyl fluoride (PMSF), and diethylpyrocarbonate (DEPC) was conducted in 20 mM Tris-HCl buffer, pH 7.5, with the reagent concentrations at 30 mM for N-ethylmaleimide and at 2, 5, and 10 mM for PMSF and DEPC. Reagent solutions were prepared fresh immediately before use. Exposure of the enzyme of interest (R. capsulatus PTS or FruK) was carried out at 37°C for 1 h (59). Subsequently, excess dithiothreitol (for N-ethylmaleimide inactivation), serine (for PMSF inactivation), or histidine (for DEPC inactivation) was added, and incubation was continued for 30 min. The fructose PTS of R. capsulatus was assayed as described previously (13).

## RESULTS

Nucleotide sequence of the *fruK* gene encoding FruK of R. capsulatus. Figure 1 presents the nucleotide sequence of the R. capsulatus fruK gene and flanking regions. Preceding the start codon of the fruK gene is a potential Shine-Dalgarno sequence, GGGGG, located 5 bp upstream of the ATG initiation codon (+1), within the end of the *fruB(HI)* gene which overlaps the fruK gene by 4 bp. Interestingly, in E. coli and Salmonella typhimurium there is a 1-bp overlap between the fruB(MH) and fruK genes. Such overlapping genes suggest translational coupling between fruB and fruK. The fruK open reading frame in R. capsulatus is 951 bp long, corresponding to 317 codons (316 amino acids in the protein; molecular weight = 31,232). The molecular weight is substantially smaller than that determined experimentally for FruK of R. capsulatus (39 kDa [61]) but is essentially the same as that estimated for the E. coli FruK by sodium dodecyl sulfate (SDS) gel electrophoresis (molecular weight  $\approx$  30,000 [7]). The *Rhodobacter fruK* open reading frame is present within the 1,020-bp chromosomal fragment, the sequence of which is shown in Fig. 1.

Alignment of FruK from *R. capsulatus* with other bacterial kinases: the PfkB family. Figure 2 shows alignment of the *R. capsulatus* FruK protein with four bacterial kinases. These enzymes include FruK of *E. coli* (43), PfkB of *E. coli* (EC 2.7.1.11) (12), phosphotagatokinase of *Staphylococcus aureus* (the *lacC* gene product) (48, 57a), and ribokinase of *E. coli* (EC 2.7.1.15) (27). The consensus sequence is presented below the aligned sequences, and residues conserved in four or five of the proteins are indicated by asterisks or exclamation marks, respectively, above the aligned sequences.

R	A	L	A	с	т	т	A	A	E	v	R	G	L	к	*			_	_		_
CGC	GCC	GCT	GGC	CTG	CAC	GAC	CGCC	GCC	GA	AGTO	GCG	GGG	GCT	GAA	M ATG	T ACG	L CTG	RCGC	I ATC	A G	6 2880
CCF	T	V STT	S TCG	L CTC	N AAT	s TCC	A GCC0	V STC	D	Q CAG	T	V STG	T	V	P	G GGC	F	T	A	۵ ۵	26 2940
	A	v	N	R	v	λ	A	s	R	т	D	A	G	G	ĸ	с. С	 v	N N	v	2	46
ATC	CGG	GTG	AAC	CGG	GTG	GCC	SCC	rČG	CGC	ATC	GAT	GCG	GGC	GGC	AAG	GGG	GTC	AAT	GTC	G	3000
cci	s	F TTC	L CTG	A GCC	H CAT	V GTC	G GGC(	H	G GGG(	V GTC	A GCG	V GTG	ACCO	G GGG	L FTG	L CTC	G GGC	A GCC	Ė GAG	N A	66 3060
ATC	A SCG	A GCG	L CTG	F	A GCG	R CGC	H CAT	F FTC	A GCG(	A GCG	T ACG	G GGG	L CTG	V GTC	D GAT	A GCC	C TGT	Q CAG	R CGT	c	86 3120
	P	G	A	т	R	т	N	v	к	I	v	D	P	L	Q	D	Q	v	т	D	106
TGC	:00	GGC	GCG	ACG	CGG.	ACG	AAT	GTG	AAG	ATC	GTC	GAT	CCG	CTG	CAA	GAT	CAG	GTC	ACC	G	3180
ATC	L TG	N AAC	F TTT	P CCG	G GGC	I ATC	A GCC	A GCC	G GGG	P	A GCC	D GAT	L	D GAC	A GCC	V GTG	A GCC	A GCG	ACC	c	126 3240
TG	T	E GAG	L CTT	L CTG	A GCG	Q CAG	G GGC(	L CTG	D GAT	W TGG	V GTT	A GCG	L CTG	C IGC	G GGC	S AGC	L	P	A GCG	G G	146 3300
GGJ	I	G GGC	A GCC	E GAG	A GCC	Y TAT	A GCC	E GAA	L CTG	A GCG	A GCC	L CTT	A GCC	R CGC	K AAG	G GGC	G GGC	A GCG	R	v G	166 3360
TGO	A SCG	L CTG	D GAC	т АСТ	S TCG	G GGG	P	A GCG	L	G GGT	L CTG	A GCG	L CTG	A GCG	A GCA	R .CGG	P	D	I	v G	186 3420
	к	P	N	v	A	Е	L	G	A	H	L	G	R	т	L	т	G	L	Е	s	206
TC	AG	CCC.	AAT	GTC	GCG	GAA	CTG	GGC	GCG	CAT	CTG	GGC	CGC:	ACC	CTG	ACC	GGC	CTI	GAG	A	3480
GCC	V STG	R CGC	E GAG	A GCC	A GCG	R CGC	D GAT	L	A GCG	A GCC	S TCG	G GGC	V GTC	G GGG	L CTG	V GTC	A GCT	V GTC	S TCG	M A	226 3540
TGC	G GGG	A GCG	G GGG	G GGC	A GCG	V GTA	L CTG	V STG	R CGC	G GGC	A GCC	E GAG	A GCG	V GTG	L CTG	A	I	P	P	A G	246 3600
CAI	T ACC	P CCC	I ATC	A GCC	S TCG	T ACC	V GTG	G GGG	A GCG	G GGC	D GAC	A GCG	M ATG	V GTG	A GCC	G	L	I	H CAT	A G	266 3660
cco	A SCC	TACC	L CTT	G GGT	ь стс	D GAC	L CTG	A GCC	E GAG	TACC	A GCC	R CGT	L CTG	A GCC.	T ACC	AGCC	F	S TCG	L CTG	G	286 3720
GCC	A. SCG	L CTG	G GGC	E GAG	I ATC	G GGC	P	H CAT	L CTG	P CCG	P CCG	P CCG	E GAG	R CGC	L CTT	A GCC	A GCG	L	A GCC	R	306 3780
GCZ		V GTC	T	V GTC	K AAA	T	L CTG	P	P	V GTC	* TGA	GCC	GCA		GAG	GAA	ccc	M	S	K	316 3840

FIG. 1. Nucleotide sequence of a 1,020-bp segment encompassing the *fruK* gene of *R. capsulatus* and the deduced aminoacyl sequence of the 316 residues of FruK. The nucleotide sequence starts at bp 2821, numbered as described previously (61). Aminoacyl numbering starts at the ATG codon (+1) which is believed to serve as the initiation codon. Note the 4-bp overlap between the *fruK* gene and the preceding *fruB(HI)* gene. The intercistronic region between *fruK* and the following *fruA* gene (60) is 18 bp. The doubly underlined residue (corresponding to bp 3725) represents the start of the segment published previously (60).

The statistical analyses of the aligned sequences are summarized in Table 1. The percent identities and the comparison scores (expressed in standard deviations) are sufficient to establish that these five proteins are homologous (16). A phylogenetic tree for these proteins is shown in Fig. 3. We refer to these proteins as the PfkB family. It can be seen that among these proteins, the two FruKs are most similar, that the phosphotagatokinase and PfkB are more distant from the two FruKs, and that ribokinase is more distant from the other proteins than the latter proteins are from each other.

Comparative secondary structural analyses, hydropathy, and amphiphilicity of *R. capsulatus* FruK and *E. coli* PfkB. Comparisons of secondary structural predictions for FruK of *R. capsulatus* and PfkB of *E. coli* are shown in Fig. 4. As can be seen, the plots for these two proteins are very similar for predicted random coil, turns,  $\beta$ -structure, and  $\alpha$ -helix. Hydropathy plots (32) of these proteins reveal that both are strongly hydrophilic. They may both consist of alternating stretches of  $\alpha$ - and  $\beta$ -structure as does PfkA, as determined from the X-ray crystallographic structure. Three extended regions of striking amphiphilicity (18) are also revealed (Fig. 4). Corresponding plots of the other homologous sequences

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FruKRc	1	MTL	RIAT	VSLN	IS.AV	DOTV	TVP	GF.	TAD	AVN	RVA	ASRI	DAGO	KG	NV	ASFI	LAH	GHC	VAV
FrukEc		MSR	RVAT	TTLN	P.AY	DLVG	FCP	ET.	ERG	EVN	LVK	TTGI	HAAG	KGI	NV	KVI	KD	GTE	VTV
PfkBFc		MV	PTVT	TTL	P ST	DSAT	TTP	OT .	VDF	FN	CAV	DHD	SNDC	CCI	NU	DA	TAHI	000	ATA .
PtaKSa			MTT	TTLN	ID SV	DISV	PLT	AT.	KLD	DVN	PVO	FVSI	TACC	KCT	NU	PDVI	104	CFI	DVT A
PibVFc		MON	ACST	WIC	CTMA	DUTT	NIO	CFD.	TDC	ETW.	TON		A FCC	VCI	NO	178		CAL	TAF
KIDKEC		righ.	1031		31NA	Unit	1100	511	110	L1 V	100	niçı	A1 00	NOr			101.	50A	1111
		w .				n				EVA				ve	MU			~	
Jonsens		m .	KT 1	11.0		U	F			E v Ia	•		AGO	, NG	In v I	•	L.A.	0	v
Frukke	59	TGL	LGAL	N . AA	LFAR	HFAA	TGL	VDA	COR	LPG	ATR	TNV	TADE	LQL	QV.	IDL	NFPO	31A/	IGPA
Frukec		GGF	LGKL	N.QL	GFQQ	LFSE	LGI	ANK	FQV	VQG	RIK	INVE	LTEP		EV.	DFI	NVS	SFE.	TPA
PIKBEC		IFP.	AGG/	T.GE	HLVS	LLAD	ENV	PVA	TVE	AKD	WTR	QNL	IVHVE	EASC	EQ	RF	VMP	JAAI	LNED
PtaKSa		SGF	IGGE	CL.GC	<b>QFIAK</b>	KLDH	ADI	кна	FYN	IKG	ETR	NCI	ILH.	• EC	SQQ:	FEI	LEQ	SPE:	IDNQ
RibKEC		IVC	TGDD	SIGE	SVRQ	QLAT	DNI	DIT	PVS	VIK	GES	TGV	LIF	/NGI	EGE	IVI	SIH	AGAI	NAAL
consens		G	G	G		LA	I	A		G	TR	NV		0	5 1	г		3 A	
								**	1*	*									1
FruKRc	118	DLD	AVA	TLT	ELLAC	GLDW	WAL	CGS	LPA	GIG	AEA	YAEI	AAL	ARKO	GGA	RVA	LDT	SGP	ALGL
FruKEc		DWE	RFV	DSL	SWLGO	. FDM	IVCV	SGS	LPS	GVS	PEA	FTD	MTRI	RS	CP	CII	FDS	SRE/	ALVA
PfkBEc		EFR	OLE	OVL	EIESG	AI	LVI	SGS	LPP	GVK	LEK	LTO	ISL	RKNI	KGS.	AAS	STV	LGO	GLSA
PtaKSa		EAA	GFI	HFE	DLLEK	.VEA	VAI	SGS	LPK	GLN	ODY	YAO	IERO	CONI	KGV	PVI	LDC	SGA	TLOT
RibKEc		SPA	LVE	ORE	RTANA	SALI	MOI	ESP	LES	VMA	AAK	TAH		ONI	KTT.	VAL	NPA	PAR	ELPD
ALDROO									220										
consens					T.		v	SGS	I.P	G	Е	A		N	KG		D	SG	τ.
compens					-		•	000		•	-								2
Trave VID o	170					1.		CDG			cup					-			
FFUKRC	1/0	ALA	A	(PD1)	VKPNV	ALLA		ARI			SVR.	LAAI		136		VAV	SMG.	AGG	AVLV
Frukec		GLK	A	APWL	VKPNF	GRELI	.1 ₩/	GR	LPE		DVI	LAAI	IALR	EQG.	IAH	~~1	SLG	AEG	ALWV
PIKBEC		ALA	1G.I	NIEL	VKPN	KELS	SAL\	NRE	LIC	PDD	VRK	AAQ	LIVN	SGR	AKK	~~~	SLG	PQG.	ALGV
Ptaksa		VLE	NPY	KPTV.	IKPNI	SEL	QLL	NQI	LDF	SLE	SLK	QAV	SQPL	FEG	IEW	110	SLG.	AQG.	AFAK
RIDKEC		ELL	A	LVDI	ITPNE	TEAL	CKLI	GIF	IVEN	IDED	AAK	. AA	2VLH	EKG	IRT	VLI	TLG	SRG	VWAS
consens		L	Y V	P	VKPN	EL	L	GR	L		ĸ	AA	L	G	I	v v	SLG	A G	a v
				•	*	**	***	1	*					*					
FruKRc	235	RGA	EAV	LAIP	PATPI	[AST]	/GAC	DAN	<b>IVAC</b>	SLIH	IAAT	LGL	DLAE	TAR:	LAT	AFS	LGA	LGE	IGPH
FruKEc		NAS	GEW	IAKP	PSVD\	/VST	/GAC	DSN	(VGC	LIY	GLL	MRE	SSEH	<b>TLR</b>	LAT	Α	VAA	LAV	SQSN
PfkBEc		DSE	INCI	OVVP	PALKS	SQST	/GAC	GDRI	LVG	MTI	KLA	ENA	SLEE	MVR			FGV	AAG	SAAT
PtaKSa		HNH	ITFY)	RVNI	PTIS	LNP	GSC	DST	rvac	ITS	SAIL	NHE	NDHD	LLK	KAN	т	LGM	LNA	OEAO
RibKEC		VNG	EGO	RVPG	FRVO	VDT	LAAC	DT	NG/	LIT	TALL	EEK	PLPE	AIR	FAH		. AA	AAI	ÄVTŘ
consens				V P	P	ST	/GAC	GD	VGC	LI	ALL		LE	R	A		GA	LA	
FruKRC	295	I.PI	PEP		ARTW	רעאיי	.PP	,											
FrukEc		VCI	TTNP	POL	AAMM	DVD	( OPI	- FN											
PfkBFc		LNC	CTP	L C S H	DDTO	TVA	VICI												
DESVC-		- mil	(31 K /1/10	TNNY	20101	NOTE	100	;											
FLANDA		101			PETE	ADI DI	A PGL	*											
RIDKEC		KG/	NUPS	vrwR	LEIO	AL PO	ĸQŔ												
		~	-																
consens		ف	- K																

FIG. 2. Multiple alignment of FruK of *R. capsulatus* (FruKRc) with the same enzyme of *E. coli* (FruKEc), PfkB of *E. coli* (PfkBEc), phosphotagatokinase of *S. aureus* (PtaKSa), and ribokinase of *E. coli* (RibKEc). An exclamation mark above the aligned sequences indicates a residue conserved in all five sequences. An asterisk indicates a residue conserved in four of the five sequences. The consensus sequence (consens) is provided below the aligned sequences. The statistical analyses of the alignments are summarized in Table 1. The Fasta (44) and Bestfit (39) programs were used to give optimal alignment.

depicted in Fig. 2 were similar (data not shown). The degrees of sequence identity observed for members of this family support the conclusion that they share essential structural features throughout their lengths.

Comparison of the amino acid compositions of FruK of R. capsulatus and homologous bacterial kinases. Table 2 presents the amino acid composition of FruK of R. capsulatus and compares it with those of FruK of E. coli, phosphotagatokinase of S. aureus, ribokinase of E. coli, and the two Pfks of E. coli (PfkB and PfkA). As described above, all of these proteins except PfkA of E. coli are sufficiently similar in sequence to establish that they are homologous, and as noted in Table 2, they are all of a very similar size. Despite these facts, the amino acid composition of FruK of R. capsulatus differs drastically from those of the other proteins. The amino acids are listed in Table 2 in the order of their frequency in FruK of R. capsulatus. All of the predominant amino acids in FruK of R. capsulatus (above the first space in Table 2) show increased frequencies relative to the other proteins, whereas almost all of the amino acids present in FruK of R. capsulatus in smaller amounts (below the second space in Table 2) show decreased frequencies rela-

 
 TABLE 1. Statistical analyses<sup>a</sup> of the aligned sequences which compose the PfkB family

Ductoin	FruKE	c	PfkBE	с	PtaKS	a	RibKEc		
Protein	%I	CS	%I	CS	%I	CS	%I	CS	
FruKRc FruKEc PfkBEc PtaKSa	36 (316)	51	27 (299) 29 (283)	18 29	30 (233) 25 (307) 24 (303)	34 33 25	53 (15) 33 (127) 21 (292) 20 (243)	0.3 14 11 6	

<sup>a</sup> Abbreviations used for the different proteins are the same as those described in the legend to Fig. 2. %I, percent identity; CS, comparison score given in SD determined by using the RDF2 program and 50 shuffles (44). Values in parentheses with the percent identities give the numbers of residues in the segment compared.

tive to those in the homologous proteins. Thus, for example, alanine, a predominant amino acid in all of these proteins, is about twice as abundant in FruK of *R. capsulatus* as in the other proteins. L, G, V, T, and P also show increased frequencies in FruK of *R. capsulatus* relative to the other proteins. By contrast, E, S, N, Q, K, C, and Y generally show increased frequencies of occurrence in the homologous proteins relative to FruK of *R. capsulatus*.

Nonessential residues in FruK. R. capsulatus FruK contains only 2 cysteyl, 1 tyrosyl, and 1 tryptophanyl residues, and none of these residues are conserved in E. coli FruK or PfkB (Fig. 2). A high concentration of N-ethylmaleimide did not inhibit R. capsulatus FruK activity, suggesting that no essential cysteyl residue is present in this enzyme. By contrast, the PTS activity of the same extract was completely abolished by this treatment. The PfkB family proteins contain four to eight histidyl residues, but none of them are conserved in R. capsulatus FruK and in E. coli FruK and PfkB. FruK of R. capsulatus was treated with DEPC as described in Materials and Methods. The enzyme was inactivated to the extent of only 25% by 2 mM DEPC, although it was largely inactivated by 10 mM DEPC.

FruK of E. coli has been shown to be inhibited by PMSF, a reagent specific for activated seryl residues (7). Employing the procedure outlined by Buschmeier et al. (7) (see Materials and Methods), we found that FruK in extracts of R. capsulatus was only weakly inactivated by 2 mM PMSF but was largely inactivated by 5 mM PMSF. FruK of R. capsu-



FIG. 3. Phylogenetic tree of homologous members of the PfkB family. Relative evolutionary distances are provided next to the branches. Abbreviations for the five enzymes are as indicated in the legend to Fig. 2. The tree was constructed as described previously (17, 20).



FIG. 4. Comparison of secondary structural predictions for FruK of *R. capsulatus* and PfkB of *E. coli*. C, coil; T, turn; E, extended  $\beta$ -structure; H,  $\alpha$ -helix; HY, hydropathy; AM, amphiphilicity. The Analysep program from the Staden package was used to estimate secondary structure (57). Aminoacyl residue number is plotted on the x axis.

*latus* contains 11 seryl residues. Four of these are observed in the consensus sequence (Fig. 2), but none are conserved in all of the homologous kinases.

Multiple alignment of FruK with Rubisco large subunits from plants and bacteria. Figure 5 shows the alignment of a 127-aminoacyl residue segment of FruK from *R. capsulatus* with five different ribulose 1,5-bisphosphate carboxylase/ oxygenase (Rubisco) large subunits. Among the 21 totally

 
 TABLE 2. Comparison of sizes and amino acid compositions of proteins in this study<sup>a</sup>

Amino		No. of each amino acid composing:												
acid	FruKRc	FruKEc	PfkBEc	PfkAEc	PtaKSa	RibKEc								
A	66	30	37	27	22	49								
L	42	30	34	24	37	26								
G	36	29	27	38	24	27								
V	32	33	28	25	23	25								
Т	23	16	16	15	16	17								
Р	18	13	15	9	13	14								
R	16	18	14	21	5	15								
D	15	17	10	23	14	13								
E	11	19	22	22	23	23								
S	11	21	24	14	16	15								
I	10	15	15	28	25	24								
N	7	11	14	8	22	17								
F	6	12	4	9	9	8								
н	6	4	6	7	8	6								
Q	5	11	15	5	20	14								
К	5	12	13	13	17	9								
М	3	8	4	13	2	3								
С	2	4	4	7	3	1								
Y	1	2	5	11	9	1								
w	1	7	1	1	1	2								
Total	316	312	308	320	310	309								
MW	31,232	33,708	33,853	32,290	32,388	34,758								

<sup>a</sup> Abbreviations for the proteins are described in the legend to Fig. 2. PfkAEc, PfkA of *E. coli*.

					-	-						:			•		-		* !	:	
FRUK	85	RLPG	ATRT	NV KI	VDP	1.00	ovt	נזמי	NFP	GTA	AGP		DAV		гі.т	EL		31.0	wva	LCO	351.
ALEC	319	RMSG	CDHT	HAGTU	VGH	TEG	E P	DT	TLG	FVD	T.T.B	חח	TER	DR	SRG	TEI	FTO	<u>م</u>	wvs	T.P	SVI.
PEHC	319	RMSG	CDHT	HACTU	VGR	TEG	ER	FM	TLG	FVD	T.T.S	nne	TER	DR	ARC	TFI	FTO	ň	WVS	MD	201
PFAC	326	RLSG	GDHT	HAGTU	VGN	T.FG	FR	ET	TLG	FVD	T.T.B	וחח	TKK	DP	SRG	TVI	FTO	ň	WUS	T.P	271
TORC	319	RMSG	CDHT	HSGTI	NGR	(LEG	5 8	n T	TLG	FVD	LLE		TUP		SPC	TVI	ETO.	ň	SUC	T.D	201.
AFUT	321	PLAC	UDHM	HTCT	VGB	(LEC		LT	voc	~~~	VCE		TOT		TPC	LEI	FDO	5	WBC	T.PI	KVM
ALU I	321	NDAG				(DDG			100					0.0	110	DL I			- AAC	Divi	
		*	-				*	•	*					:			**				-
FDIIK	144	PACT	CAFA	VART		DVC			1.01	60D				חסו	T 1/12	DM					
NEC	376	DUNC	CCIU			PETE	CDD		LOI	205	71/	100			21/2	110		LGN	mbe		
DEAC	376	DUNC	0010	UNITIM		PETE		20			71.00	ne :	JON7	LPG.			~~				
PEAC	303	DUNC	CCTH	VWRITIE		PETE		20		666	110	onr.	JON		****	NR.	V A			1	
TOPC	376	DENC	COTH	VULLA			CDD	.cv			TIC	UD	JCN7	E G	<u></u>	MD	22			:	1 5 3
AFUT	379	DUNC	CCTH	ACOM	101			131	TOF		710	ישחי			. • • • . • •	MD	UN				
ALU1	570	-	9914	AGQUI	цор.	LUPE	-		LQF			SULE	2010	ING	~	IN PA	VA.				LEA
		-			-		-			-	-				-						-
			*	**!!	*																
PD/1/2	204																				
FRUK	204	LESV	RLAA	RULA	156	218															
ALFC	427	CVQA	RNEG	RDLAN	KEG	441															
PEHC	427	CVQA	RNEG	KULAI	ŒС	441															
PEAC	434	CVQA	RNEG	RDLAI	REG	448															
TOBC	427	CVKA	RNEG	RDLA	<b>DEG</b>	441															
AEUT	429	MVLA	RNEG	RDIL	٩EG	443															
			-		-																

FIG. 5. Multiple alignment of FruK of *R. capsulatus* (FruK residues 85 to 318) with corresponding regions of five sequenced Rubisco large subunits. The five sources of Rubisco are alfalfa chloroplast (ALFC) (1), petunia hybrida chloroplast (PEHC) (2), pea chloroplast (PEAC) (62), tobacco chloroplast (TOBC) (56), and *Alcaligenes eutrophus* (AEUT) (3). Residue numbers within each protein are indicated at the beginning of each line and at the end of the segments. The following symbols indicate identity of the residue in FruK with the five Rubisco proteins: \*, all five depicted proteins; !, four of the proteins; ^, three of the proteins; ", two of the proteins; and ', just one of the proteins. Double underlines indicate residues in FruK of *R. capsulatus* which align with all residues in the Rubisco large subunits as well as with those in PfkB of *E. coli* (Fig. 2).

conserved residues, L335 and G464 of alfalfa Rubisco aligned with L100 and G172 of FruK. In the former enzyme, these residues are involved in ribulose-1,5-bisphosphate binding (29). As revealed by the statistical analyses reported in Table 3, the degrees of sequence similarity suggest that these proteins may be homologous. For example, FruK is 27% identical with Rubisco from alfalfa throughout a segment encompassing residues 319 to 441 of the latter enzyme. The Los Alamos comparison score (28) for this segment was 8.5 standard deviations (SD) higher than that obtained with 100 comparisons of randomized sequences of these proteins. The probability of obtaining such a score by chance is less than  $10^{-16}$ . The corresponding value with the ALIGN program was 7.4 SD higher than that obtained with 1,000 comparisons of these randomized sequences. Thus, the results suggest but do not prove that a domain in FruK is homologous to a domain in alfalfa Rubisco. Other Rubisco proteins were also similar to FruK, although the comparison scores were somewhat lower. The sequence of Rubisco from Rhodospirillum rubrum was most divergent from that of FruK, but sequence comparisons among the different Rubisco large subunits revealed that all of these proteins are homologous (Table 3). The residues doubly underlined in Fig. 5 are those which are also aligned with PfkB in Fig. 2. The results show that FruK, PfkB, and the various Rubisco large subunits all possess domains which exhibit sequence similarity. These domains might be involved in sugar bisphosphate binding, as demonstrated in Rubisco by X-ray crystallography (29).

Codon preference for the *fru* operon of *R. capsulatus*. Figure 6 shows the codon preference plots for the three reading frames within the *fru* operon, based on the codon usage for *Rhodobacter* spp. (60a). It can be seen that the first gene, *fruB*(*HI*), is in the A reading frame, whereas both *fruK* 

 TABLE 3. Statistical analyses of comparisons between the aligned sequence FruK of R. capsulatus and those of Rubisco large subunits

First enzyme and region compared	Second enzyme and region compared	% Identity	SD <sup>a</sup>	No. of gaps
FruK, 85–218	AlfC, 319-441	27.1	8.5	4
FruK, 85–218	PehC, 319-441	23.8	6.7	4
FruK, 85–218	PeaC, 326-448	26.2	7.5	4
FruK, 85-218	TobC, 319-441	27.1	6.6	4
FruK, 85–218	AeuT, 321-443	18.7	5.0	4
FruK, 85-218	RspR, 313-430	13.8	1.5	9
AlfC, 319-441	PehC, 319-441	95.1	63.5	0
AlfC, 319-441	PeaC, 326-441	95.1	56.6	0
AlfC, 319-441	TobC, 319-441	94.3	65.8	0
AlfC, 319-441	AeuT, 321-443	61.0	37.7	0
PehC, 319-441	PeaC, 326-441	93.5	53.1	0
PehC, 319-441	TobC, 319-441	89.4	59.2	0
PehC, 319-441	AeuT, 321-443	60.2	39.0	0
PeaC, 326-448	TobC, 319-441	91.1	60.2	0
PeaC, 326-448	AeuT, 321-443	61.0	41.2	0
TobC, 319-441	AeuT, 321-443	60.2	37.3	0
RspR, 313-430	AlfC, 319-441	31.7	14.8	4
RspR, 313-430	PehC, 319-441	31.7	11.4	4
RspR, 313-430	PeaC, 326-448	32.5	11.7	4
RspR, 313-430	TobC, 319-441	31.7	11.7	4
RspR, 313-430	AeuT, 321–443	32.5	11.9	4

<sup>*a*</sup> Los Alamos comparison scores are reported. ALIGN comparison scores were similar. For example, values for the ALIGN comparison of FruK with AlfC, PehC, and PeaC were 7.4, 6.8, and 7.2 SD, respectively, higher than those obtained with 1,000 comparisons of these randomized sequences. Abbreviations are the same as those described in the legend to Fig. 5. RspR, Rubisco from *R. rubrum*.

and fruA are in the C reading frame. Rare codons are utilized very infrequently in all three genes, suggesting (i) that the open reading frames fruB(HI), fruK, and fruA are correctly determined and (ii) that the entire operon can be expressed at a high level. Preceding the fruB(HI) gene, a partial open reading frame (orf X) which shows a diminished frequency of rare codon usage relative to extragenic DNA, but an increased frequency of rare codon usage relative to the three recognized open reading frames of the fru operon is revealed. This 387-bp stretch encodes the C-terminal 128 amino acids of a putative protein which terminates within the fruB(HI) gene with an overlap of 4 bp. This is the same degree of overlap observed between the fruB(HI) and fruKgenes (Fig. 1). Since no predominant protein bands were observed following expression of the 7.4-kb fragment, which starts 1 kb upstream from the beginning of the sequenced part of orfX in the T7 promoter/RNA polymerase system (61), we presume, in agreement with the increased frequency of rare codons within this open reading frame, that the product of this gene either is not expressed at a high level or is not expressed at all. A computer search of the data base for homology with the encoded C-terminal 128 residues of the putative protein did not yield significant positive results.

Alignment of Pfks of prokaryotes and eukaryotes: the PfkA family. Figure 7 shows alignment of three bacterial Pfks (from *E. coli* [25], *B. stearothermophilus* [21], and *Spiroplasma citri* [10]) with homologous domains of a number of eukaryotic Pfks. The statistical analyses of the sequences aligned in Fig. 7 are summarized in Table 4. The percent identities and the comparison scores (in SD) are sufficient to establish that all of these protein segments are homologous. We refer to these proteins and homologous protein segments as the PfkA family. The four sequenced mammalian en-



FIG. 6. Codon preference plots of the *fru* operon of *R*. *capsulatus* generated employing the program of Gribskov et al. (23) but based on *Rhodobacter* codon usage. The program identifies efficiently translated genes as peaks above open reading frames (open boxes below peaks) that contain few rare codons (vertical lines below the open reading frames). The window size and rare codon thresholds were set at 50 and 0.1, respectively. The number of the base pairs in the DNA fragment analyzed is indicated on the x axis.

zymes (from human muscle [55], human liver [34], rabbit muscle [33], and mouse liver [22]) are homotetramers of polypeptide chains about twice the size of the bacterial enzymes (each 780 residues). They consist of two adjacent segments which are homologous to each other and the bacterial Pfks, and they probably arose by tandem intragenic duplication (Table 5). The larger yeast enzyme is a heterooctamer consisting of two large, dissimilar polypeptide chains,  $\alpha$  and  $\beta$  (987 and 959 residues, respectively), each of which, like the mammalian enzyme, contains an internal repeat of the bacterial Pfk equivalent and is present in four copies per enzyme (Fig. 7 and Table 5) (24). Examination of the aligned sequences (Fig. 7) reveals that the N-terminal segments are much more similar to the bacterial enzymes than are the homologous C-terminal segments, which, however, are closely related to the other C-terminal segments. This observation suggests that the duplication event, giving rise to the eukaryotic Pfk subunit from a prokaryotic Pfk-like protein, occurred before the divergence of the yeasts from mammals.

A phylogenetic tree for these ATP-dependent bacterial Pfks and the eukaryotic Pfk segments as well as the potato PP, dependent Pfk is shown in Fig. 8. It can be seen that all of the N-terminal sequences of the eukaryotic, ATP-dependent Pfks cluster together with the bacterial proteins, whereas the C-terminal sequences form a cluster which is quite distant (in relative, apparent, evolutionary time) from the N-terminal segments. Even more distant from the bacterial sequences are homologous segments in each of the two subunits of the potato PP<sub>i</sub>-dependent Pfk (Fig. 8). These two subunits ( $\alpha$  and  $\beta$ ) exhibited 42% overall identity with each other throughout most of their regions of overlap (8). The percent identities observed between the  $\alpha$ - and  $\beta$ -subunits of the PP<sub>i</sub>-dependent plant enzyme and E. coli PfkA were 25% in a 148-residue overlap and 29% in a 222-residue overlap, respectively (comparison scores of 11 and 12 SD, respectively). Corresponding values when these two subunits ( $\alpha$  and  $\beta$ ) were compared with the *B. stearothermophilus* Pfk were 27% identity in 202 overlapping residues (comparison score of 14 SD) and 30% identity in 208 overlapping residues (comparison score of 16 SD), respectively. Thus, the plant enzyme is clearly homologous to members of the PfkA family. It more closely resembled the gram-positive bacterial enzyme than the gram-negative bacterial enzyme. The potato PP<sub>i</sub>-dependent Pfk exhibited greater sequence similarity to the bacterial enzymes than to the other eukaryotic enzymes (data not shown).

Sequence similarity between the R. capsulatus FruK and the two yeast Pfk subunits. Members of the PfkA family exhibited regions of sequence similarity with members of the PfkB family. For example, FruK of R. capsulatus exhibited 17.2% identity with the yeast Pfk  $\beta$ -subunit. The regions of overlap included residues 4 to 302 in FruK of R. capsulatus and residues 668 to 959 in the yeast Pfk  $\beta$ -subunit. Three gaps were introduced to achieve this degree of identity. The percent similarity for this alignment, taking into account both identities and semiconservative substitutions, was 37.1%. The comparison score for this alignment was 3.9 SD. The probability of getting this comparison score by chance is less than 5  $\times$  10<sup>-5</sup>. These values, while insufficient to establish homology, demonstrate a degree of sequence similarity which is not likely to have arisen by chance. Divergent evolution from a common ancestor would provide a reasonable explanation for this sequence similarity, but the possibility of convergent evolution cannot be ruled out.

# DISCUSSION

In early reports (49, 54), a fructose-specific PTS was detected and characterized in the photosynthetic bacteria *Rhodospirillum rubrum* and *Rhodobacter spheroides*. The system was shown to consist of two membrane proteins, one

		* * * **! * * * ! * ! * * ! * * ! * * ! * * ! * * ! * * ! *
E. coli	1	MIKKIGVLTSGGDAPGHNAAIRGVVRSALTEGLEVMGIYDGYLGLYEDRHVOLDRYSVSDMINRGGTFLGSARCPEFRDENIRAVAIENLKKRGIDALVVIGDGGSYMGAMRL
B. Stear.	1	MKRIGVLTSGGDSPGMNAAIRSVVRKAIYHGVEVYGVYHGYAGLIAGNIKKLEVGDVGDIIHRGGTILYTARCPEFKTEEGOKKGIEOLKKHGIEGLVVIGGDGSYOGAKKL
S citri	ī	NI KKIGII TSCCDSOCMNAATACU IKTANAKCI FTYI IDDCYIGI INN WIFUUDNNFADSIMI ICCTVICSADI PFEKDEFUKKAUDI I KKOFIAATUVICCDCSVOCAODI
Human M-N	14	IGKA I AVI. TSGGDAGGMNAAVRAVVRVGI FTGARVFFVHEGYOGI VDGG, DHIVFATWESVSMMI OLGGTVI GSARCKDERERERI RAAVNI VKRGI TNI CVI GGDGSI TGADTERSEW
Dabbit M-N	14	VICK TAVI TSCEDAGEMNAAVDAVDVCI FECADVESVEEDGE DE
Neuro I-N	14	TORATAVITSOCIACIANA NORVATIVETTER TORATET THEORY OF THE STATE STATE STATE STATE STATE AND A STATE STATE STATE AND A STATE STATE STATE AND A STATE AND
House L-N	14	AUXAIGUTISUUDAQGAMAVAVIRAGIIVGAVUTIIEGIEUVEGO. ENIKFAMULSISHIIQLAGIIIGSAKKAATIIREGALAAIMULQAGIIAAUXATOODOOLAANT
Human L-N	13	GANATGVLISGGDRQGARAAVRAVIRAGIIVGARVFLIIEGIEGUVEGG.ENINGANWESVSNIIGLGAIIIGIASSAAFIIREGRAAVRAVIRAGIIVGGDEGLIGANI
Ieast B-N	195	PORTAVALI SUBDAPEANSAVALI VRSALI RUCKALI VIJEGI ELEVROPELI LEFANEUVROVSALUGI NI GIARCHEFANCEURLEGADALI ELEGVALUSELI GALLE RISEN
Ieast A-N	204	KKKI AVHISGGDSFOHNAVKAVVRIGI HFGCDVFAVIEG FEGLERGG. KI LKMAWEUVRGWLSEGGTLI GTARSMEFRKREGRROAAGNLI SOG IDALVVEGDGSLIGADLFRED
Human M-C	399	GSHTVAVNNVGAPAGGNNAVNSTVRIGLIQGNRVLVVHDGFEGLAK.GQIEEAGWSYVGGWTGQGGSKLGTKRTLPKK.SFEQISANITKFNIQGLVIIGGFEAYTGGLELMEGR
Rabbit M-C	399	GSYTVAVMNVGAPAAGMNAAVRSTVRIGLIQGNRVLVVHDGFEGPAK.GQIEEAGWSYVGGWTGQGGSKLGSKRTLPKK.SFEQISANITKFNIQGLVIIGGFEAYTGGLELMEGR
Mouse L-C	398	SNFSLAILNVGAPAAGMNAAVPSAVRTGISEGHTVYIVHDGFEGLAK.GQVQEVGWHDVAGWLGRGGSMLGTKRTLPKP.HLEAIVENLRTYNIHALLVIGGFEAYEGVLQLVEAR
Human L-C	402	SNFSLAILNVGAPAAGMNAAVRSAVRTGISHGHTVYVVHDGFEGLAK.GQVQEVGWHDVAGWLGRGGSMLGTKRTLPKG.QLESIVENIRIYGIHALLVVGGFEAYEGVLQLVEAR
Yeast B-C	585	KRLKIAIVNVGAPAGGINSAVYSMATYCMSQGHRPYAIYNGWSGLARHESVRSLNWKDMLGWQSRGGSEIGTNRVTPEEADLGMIAYYFQKYEFDGLIIVGGFEAFESLHQLERAR
Yeast A-C	592	DRLNIGIVHVGAPSAALNAATRAATLYCLSHGHKPYAIMNGFSGLIQTGEVKELSWIDVENWHNLGGSEIGTNRSVASE.DLGTIAYYFQKNKLDGLIILGGFEGFRSLKQLRDGR
Consensus		K IAVLTSGEDA GANAAVR VVR GI G V V GYEGL G I E W V W GGT IGTAR F E A NLK GI L VIGEDES TGA L
E. COIL B. Coor	114	IEA
D. Stear.	113	IER
S. CIUTI	114	COLOR DE CARDES DE CONTRECENTIONS DE L'ANTIVER DE LE MARTINE SUPERIOR DE LA CONTRE DE CONTRE CONTRE DE LA CON
numan M-N	133	SULLSULVAGATI UELATASSI LATVGLVGSI UNDEGETMATI GEDSALHATMEL VDATT. TRASHQKIFYLEVMGKAGI LALVTSLSCGADWVFT PECPPDDWEEHLCKKL
RADDIT M-N	193	SULLSULVAGAITALEATKSSTLNIVGLVGSIUNUPCGTUMTIGTUSSLIKNITELVUAIT.TTAQSHQRTYLLEVMGRHGGYLALVTSLSGGADWVFIPECPPDDNWEDHLCRRL
Mouse L-N	133	GSLLEELVREGRISESTAQNYAHLTIAGLVGSIDNDFCGTDMTIGTDSALHRIMEVIDAIT.TTAQSHQRTFVLEVMGRHCGYLALVSALASGADWLFIPEAPPEDGWENFMCERL
Human L-N	133	GSLLEELVAEGKISETTARTYSHLNIAGLVGSIDNDFCGTDMTIGTDSALHRIMEVIDAIT.TTAQSHQRTFVLEVMGRHCGYLALVSALASGADWLFIPEAPPEDGWENFMCERL
Yeast B-N	315	PSLIEELLKTNRISNEQYERMIKHINIGGTVGSIDNDMSTTDATIGAYSALDRICKAIDYVE.ATANSHSRAFVVEVMGRNCGWLALLAGIATSADYIFIPEKPATSSEWQDQMCDIV
Yeast A-N	323	PSLVDELVAEGRFTKEEVAPYKNLSIVGLVGSIDNDMSGTDSTIGAYSALERICEMVDYID.ATAKSHSRAFVVEVMGRHCGWLALMAGIATGADYIFIPERAVPHGKWQDELKEVC
Human M-C	513	KQF.DELCIPFVVIPATVSNNVPGSDFSVGADTALNTICTTCDRIKQSAAGTKRRVFIIETMGGYCGYLATMAGLAAGADAAYIFEEPFTIRDLQANVEHLVQKM
Rabbit M-C	513	KQF.DELCIPFVVIPATVSNNVPGSDFSVGADTALNTICTTCDRIKQSAAGTKRRVFIIETMGGYCGYLATMAGLAAGADAAYIFEEPFTIRDLQANVEHLVQKM
Mouse L-C	512	GRY.EELCIVMCVIPATISNNVPGTDFSLGSDTAVNAAMESCDRIKQSASGTKRRVFIVETMGGYCGYLATVTGIAVGADAAYVFEDPFNIHDLKANVEHMTEKM
Human L-C	516	GRY.EELCIVMCVIPATISNNVPGTDFSLGSDTAVNAAMESCDRIKQSASGTKRRVFIVETMGGYCGYLATVTGIAVGADAAYVFEDPFNIHDLKVNVEHMTEKM
Yeast B-C	701	ESY.PAFRIPHVLIPATLSNNVPGTEYSLGSDTALNALMEYCDVVKQSASSTRGRAFVVDCQGGNSGYLATYASLAVGAQVSYVPEEGISLEQLSEDIEYLAQSF
Yeast A-C	706	TQH.PIFNIPMCLIPATVSNNVPGTEYSLGVDTCLNALVNYTDDIKQSASATRRRVFVCEVQGGHSGYIASFTGLITGAVSVYTPEKKIDLASIREDITLLKENF
		• •••• • • • • • • • • • • • • • • • •
Consensus		EL G PGTIDND GTD TIG DTALN I E D I TA SH R FV EVMGR CGYLAL GLA GAD IPE P L
E. coli B. Stear. S. citri Human M-N Rabbit M-N House L-N	207 206 207 248 248 248	KAGIAKGKKHAIVAITEHMCDV.DELAHFIEKETGRETRATVLGHIQRGGSPYPYDRILASRMGAYAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHSIIIVAEGVGSG.VDFGRQIQEATGFETRYTVLGHVQRGGSPYAFDRVLASRLGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVSEMIYPDVHK.LALKVESSGYITRATVLGHVQRGGTPSAFDRILGSRMGVEAVMALLEGTPDTPACVSLSGNQAVKLPLMECVQVTKDV SETRTRGSRLNIIIVAEGAIDNRGKPITSEDINNLVVRLGYDTRVTVLGHVQRGGTPSAFDRILGSRMGVEAVMALLEGTPDTPACVSLSGNQAVKLPLMECVQVTKDV GETRSRGSRLNIIIVAEGAIDNRGKPITSSGVKDLVVRLGYDTRVTVLGHVQRGGTPSAFDRILGSRMGVEAVMALLEGTPDTPACVSLSGNQAVKLPLMECVQVTKDV
E. coli B. Stear. S. citri Human M-N Rabbit M-N Mouse L-N Human L-N	207 206 207 248 248 248 248 252	KAGIAKGKKHAIVAITEHHCDV.DELAHFIEKETGRETRATVLGHIQRGGSPTAFDRVLASRMGAYAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHSIIIVAEGVGSG.VDFGRQIQEATGFETRVTVLGHVQRGGSPTAFDRVLASRLGARAVELLEGKGGRCVGIQNNQLVDHDIAEALAN AMLHQAQKRSVIVVVSEHIYPDVHK.LAKLVESKSGYITRATVLGHTQRGGNFJAHDRVKAFQMAQFAVGQIIAGVGGLAIGNQGDQIIARPINEALSIPRS SETRTRGSRLNIIVAEGAIDRNGKPITSEDIKUVVKRLGYDTRVTVLGHVQRGGTPSAFDRILGSRMGVEAVMALLEGTPOTPACVVSLSGNQAVKLPLMECVQVTKDV GETRSRGSRLNIIIIAEGAIDRNGKPISSSYVKDLVVRLGFDTRVTVLGHVQRGGTPSAFDRILGSRMGVEAVMALLEATPDTPACVVSLSGNQSVRLPLMECVQVTKDV GETRSRGSRLNIIIIAEGAIDRNGKPISSSYVKDLVQRLGFDTRVTVLGHVQRGGTPSAFDRILSSKMGHEAVMALLEATPDTPACVVSLSGNQSVRLPLMECVQVTKDV
E. Coli B. Stear. S. citri Human M-N Rabbi M-N Mouse L-N Human L-N Yeast B-N	207 206 207 248 248 248 252 431	KAGIAKGKKHAIVAITEHMCDV.DELAHFIEKETGRETRATVLGHIQRGGSPVPYDRILASRMGAYAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHSIIVAEGVGG.VDFGRQIQEARGFETRATVLGHVQRGGSPYAFDRVLASRLGARAVELLEGKGGRCVGIQNNOLVPHDIAEALAN. AMLHQAQKESVIVVSEHIYPOVHK.LAKLVSEKSGVITRATVLGHTQRGGPTAHDRVHASRLGARAVELLEGKGGLAIGNGGDQIIARPIHAELSIPRSS SETKIRGSRLNIIIVAEGAIDKNGKPITSEDIKNLVVKRLGYDTRVTVLGHVQRGGTPSAFDRILGSRMGVEAVMALLEGTPDTPACVVSLSGNQAVRLPLMECVQVTKDV GETKSRGSRLNIIIVAEGAIDKNGKPITSEDIKNLVVKRLGYDTRVTVLGHVQRGGTPSAFDRILGSRMGVEAVMALLEGTPDTPACVVSLSGNQAVRLPLMECVQVTKDV GETKSRGSRLNIIIIVAEGAIDNNGKPISSSYVKDLVVQRLGYDTRVTVLGHVQRGGTPSAFDRILSSKMGHEAVMALLEATPDTPACVVSLSGNQSVKLPLMECVQVTKDV GETKSRGSRLNIIIIAEGAIDNNGKPISSSYVKDLVVQRLGYDTRVTVLGHVQRGGTPSAFDRILSSKMGHEAVMALLEATPDTPACVVSLSGNGSVKLPLMECVQVTKDV GETKSRGSRLNIIIIMEGAIDNNGKPISSSYVKDLVVQRLGFDTRVTVLGHVQRGGTPSAFDRILSSKMGHEAVMALLEATPDTPACVVTLSGNGSVKLPLMECVQVTKDV GETKSRGSRLNIIIIMEGAIDNNGKPISSSYVKDLVVQRLGFDTRVTVLGHVQRGGTPSAFDRILSSKMGHEAVMALLEATPDTPACVVTLSGNGSVKLPLMECVQVTKDV GETKSRGSRLNIIIIMEGAIDNNGKPISSSYVKDLVVRLFDTRVTUGHVQRGGTPSAFDRILSSKMGHEAVMALEATPDTPACVVTLSGNGSVKLPLMECVQVTKDV GETKSRGSRLNIIIIMEGAIDNNGKPISSSYVKDLVTRHEFTNITUGHVQRGGTPSAFDRILSSKMGHEAVMALEATPDTPACVVTLSGNGSVKLPLMECVQVTKDV GETKSRGSRLNIIIIMEGAIDNNGKPISSSYVKDLVBNGKEFTNITTGHVGRGTAFAFDRILSSKMGHEAVMALEATPDTPACVVTLSGNGSVKLPLMECVQTKEV
E. coli B. Stear. S. citri Human M-N Rabbit M-N Mouse L-N Human L-N Yeast B-N Yeast A-N	207 206 207 248 248 248 248 252 431 439	KAGIAKGKKHAIVAITEHNCDV.DELAHFIEKETGRETRATVLGHIQRGGSPVPYDRILASRMGAYAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHSIIIVAEGVGSG.VDFGRQIQEATGFETRVTVLGHVQRGGSPTAFDRVLASRLGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN AMLHQAQKRSVIVVVSEHIYPDVHK.LAKLVESKSGYITRATVLGHVQRGGTPAADRYRAPQMAQFAVGQIIAGVGGLAIGNQGDQIIARPINEALSIPRS SETTRRGSRLNIIVAEGAIDRNGKPITSSDINLVVKRLGYDTRVTVLGHVQRGGTPSAFDRILGSRMGVEAVMALLEGTPDTPACVSLSGNQAVKLPLMECVQVTKDV GETRSRGSRLNIIIVAEGAIDRNGKPITSSDINLUVVRLGYDTRVTVLGHVQRGGTPSAFDRILGSRMGVEAVMALLEATPDTPACVSLSGNQAVKLPLMECVQVTKDV GETRSRGSRLNIIIIAEGAIDRNGKPISSSYVKDLVVQRLGPDTRVTVLGHVQRGGTPSAFDRILSSKMGMEAVMALLEATPDTPACVSLSGNQSVKLPLMECVQVTKDV GETRSRGSRLNIIIIAEGAIDRNGKPISSSYVKDLVVQRLGPDTRVTVLGHVQRGGTPSAFDRILGSRMGVEAVMALLEATPDTPACVVTLSGNGSVKLPLMECVQVTKDV GETRSRGSRLNIIIIAEGAIDRNGKPISSSYVKDLVVQRLGPDTRVTVLGHVQRGGTPSAFDRILGSRMGVEAVMALLEATPDTPACVVTLSGNGSVKLPLMECVQVTKDV GETRSRGSRLNIIIIAEGAIDRNGKPISSSYVKDLVVQRLGFDTRVTVLGHVQRGGTPSAFDRILSSKMGMEAVMALLEATPDTPACVVTLSGNGSVKLPLMECVQVTKDV SKHRSRGKRTNTIVVAEGAIDADLTPISPSDVKKVLVDRLGLDTRITLGHVQRGGTAVAHDRUKATQGEANAVDRLATLGEVGVTAVAVLESTPTPFSPLIAVNENKIVKRLMESVKLTKAV QRHRSKGRRNTIIVVAEGALDDQDVFTANDVARLIE.GTRVTVILGHVQRGGTAVAHDRUKATLQGVDAVNAVLESTPTFFSPLIAVNENKINKPLMESVKLTKAV
E. coli B. Stear. S. citri Human M-N Rabbit M-N Mouse L-N Human L-N Yeast B-N Yeast B-N Yeast A-N	207 206 207 248 248 248 252 431 439 617	KAGIAKGKKHAIVAITATIVAEGUDA.LAHFIEKETGRETRATVLGHUQRGGSPYPDRILASRMGAYAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHSIIIVAEGUGSG.VDFGRQIQEATGFETRVTVLGHUQRGGSPTAFDRVLASRLGARAVELLEGKGGRCVGIQNNQLVDHDIAEALAN AMLMQAQKRSVIVVVSEHIYPDVHK.LAKLVESKSGYITRATVLGHUQRGGFSAFDRILGSRMGVEAVMALLEGTPDTPACVSLSGNQAVRLPLMECVQVTKDV SETRTRGSRLNIIVAEGAIDKNGKPITSEDIKNLVVKRLGYDTRVTVLGHUQRGGTPSAFDRILGSRMGVEAVMALLEGTPDTPACVSLSGNQAVRLPLMECVQVTKDV GETRSRGSRLNIIIVAEGAIDRHGKPISSSYVKDLVVRLGYDTRVTVLGHUQRGGTPSAFDRILGSRMGVEAVMALLEATPDTPACVVSLSGNQAVRLPLMECVQVTKDV GETRSRGSRLNIIIVAEGAIDRHGKPISSSYVKDLVVRLGYDTRVTVLGHUQRGGTPSAFDRILGSRMGVEAVMALLEATPDTPACVVSLSGNQAVRLPLMECVQVTKDV SKHRSRGSRLNIIIIVAEGAIDRHGKPISSSYVKDLVVRLGJDTRVTVLGHUQRGGTPSAFDRILGSRMGVEAVMALLEATPDTPACVVSLSGNQSVRLPLMECVQVTKDV GETRSRGSRLNIIIVAEGAIDRHGKPISSSYVKDLVVRLGLDTRVTVLGHUQRGGTPSAFDRILSSKMGHEAVMALLESTPDTPSPLIAVNENIVKRLMECVQVTKDV SKHRSRGRRTIIVVAEGAIDADLTPISPSDVIKVLVDLGLDTRITVLGHUQRGGTPSAFDRILSSKMGHEAVNALLESTPDTPSPLIAVNENIVKRPLMECVQVTKDV SKHRSRGRRTNIIVAEGALDDQLNPVTANDVGALIE.LGLDTKVTLGHUQRGGTPSAFDRILSSKMGHEAVNALESTPDTPSPLIAVNENIVKRPLMESVKLTKAV QRHRSKGRRNNIIVAEGALDDQLNPVTANDVKDALIE.LGLDTKVTLGHUQRGGTPSAFDRILLSKMGHEAVNALESTPDTPSPLIGILENKIIRMPLVESVKLTKAV
E. coli B. Stear. S. citri Human M-N Rabbit M-N House L-N Human L-N Yeast B-N Yeast B-N Human M-C Rabbit M-C	207 206 207 248 248 248 252 431 439 617 619	KAGIAKGKKHAIVAITEHNCDV.DELAHFIEKETGRETRATVLGHIQRGGSPTAFDRILASRMGAYAIDLLLA.GYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHSIIIVAEGVGSG.VD.FGRQIQEATGFETRATVLGHUQRGGSPTAFDRVLASRLGARAVELLLE.GKGGRCVGIQNNQLVDHDIAEALAN AMLHQAQKRSVIVVSEMIYPDVNK.LAKLVESKSGYITRATVLGHUQRGGTPSAFDRILGSRMVCAVAKALLE.GTPDTPACVS.LS.GNQAVELPLMECVQVTKV SETTRRGSRLNIIIVAEGAIDKNGKPITSEDINNLVVKRLGYDTRVTVLGHUQRGGTPSAFDRILGSRMVCAVNALLE.ATPDTPACVS.LS.GNQAVELPLMECVQVTKV GETRSRGSRLNIIIKEGAIDRNGKPITSSSVKDLVVRRLGYDTRVTVLGHUQRGGTPSAFDRILSSKMOHEAVNALLE.ATPDTPACVS.LS.GNQAVELPLMECVQVTKV GETRSRGSRLNIIIKEGAIDRNGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTPSAFDRILSSKMOHEAVNALLE.ATPDTPACVS.LS.GNQSVKLPLMECVQVTKV GETRSRGSRLNIIIKEGAIDRNGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTPSAFDRILSSKMOHEAVNALLE.ATPDTPACVVS.LS.GNGSVKLPLMECVQVTKV GETRSRGSRLNIIIKEGAIDRNGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTASAFDRILSSKMOHEAVNALLE.ATPDTPACVVS.LS.GNGSVKLPLMECVQVTKV QRHRSKGRRNNTIIVAEGAIADDLVDSLJELGLDTKVTLGHUQRGGTAVADRILATLQGUAVALE.STDPTSPLIGLS.GNGSVKLPLMESVKLTKAV QRHRSKGRRNNTIIVAEGAIDDQLNPVTANDVKDALIE.LGLDTKVTLGHUQRGGTAVAHDRMLATLQGVDAVKAVLE.FTPETPSPLIGIL.ENKIIRHPLVESVKLTKAV KTTVKRGLVLRNEKCNENYTTDFIFHLYSE.ECKGIFDSRKVLGHMQGGSTPFDRNFATKMGAKAMMMMGG.KIKESVRNGRI.FA.NTPDGSCVLGARKRALV
E. coli B. Stear. S. citri Human M-N Rabbit M-N Mouse L-N Yeast B-N Yeast A-N Human M-C Rabbit M-C Rabbit M-C	207 206 207 248 248 248 248 248 248 252 431 439 617 619 616	KAGIAKGKKHAIVAITEHHCDV.DELAHFIEKETGRETRATVLGHIQRGGSPTAPDRILASRMGAYAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHSIIIVAEGVGSG.VDFGRQIQEATGFETRVTVLGHVQRGGSPTAPDRVLASRLGARAVELLEGKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVVSEMIYPDVHK.LAKLVESKSGYITHATVLGHVQRGGTPSAPDRILGSRMGVEAVMALLEGTPDTPACVSLSGNQAVELPLMECVQVTKDV SETRTRGSRLNIIVAEGAIDRNGKPITSEDIKUVVRLGYDTRVTVLGHVQRGGTPSAPDRILGSRMGVEAVMALLEGTPDTPACVSLSGNQAVELPLMECVQVTKDV GETRSRGSRLNIIIIAEGAIDRNGKPITSEDIKUVVRLGYDTRVTVLGHVQRGGTPSAPDRILSSRMGHEAVMALLEATPDTPACVVSLSGNQAVELPLMECVQVTKDV GETRSRGSRLNIIIIAEGAIDRNGKPISSSYVKDLVVRLGYDTRVTUGHVQRGGTPSAPDRILSSRMGHEAVMALLEATPDTPACVVSLSGNQSVRLPLMECVQVTKDV GETRSRGSRLNIIIIKEGAIDRNGKPISSSYVKDLVVRLGYDTRVTUGHVQRGGTPSAPDRILSSRMGHEAVMALLEATPDTPACVVSLSGNQSVRLPLMECVQVTKDV GETRSRGSRLNIIIIKEGAIDRNGKPISSSYVKDLVQRLGFDTRVTUGHVQRGGTPSAPDRILSSRMGHEAVMALLEATPDTPACVVTLSGNQSVRLPLMECVQVTKDV SKHRSRGKRTNTIVVAEGAIAADLTPISPSDVHKVLVDRLGLDTRITVIGHVQRGGTPSAPDRILSSKMGHEAVMALLESTPDTPSPLIAVNENKIVKRPLMEEVQHTKEV SKHRSRGRNTTIVVAEGALDDLPVTANDVKALIE.LGLGTVTIGHVQRGGTPAVADRILATLQGLEAVNAVLESTPDTPSPLIAVNENKIINKPLMESVKLTKAV KTVKRGLVLRMEKCHENYTTDFIFNLSEEGKGIFDSRKNVLGHMQQGSPTPFDRMFATKMGAKAMNMAGKIKESYRNGRIFANTPDSGCVLGARKRALVF KTTVKRGLVLRMEKCHENYTTDFIFLISS.EGKGIFDSRKNVLGHMQQGSPTPFDRNFATKMGAKAMNMAGKIKESYRNGRIFANTPDSGCVLGARKRALVF
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Human L-N Yeast B-N Yeast B-N Yeast A-N Human M-C Rabbit M-C House L-C Human L-C	207 206 207 248 248 248 252 431 439 617 619 616 620	KAGIAKGKKHAIVAITEHMCDV.DELAHFIEKETGRETRATVLGHIQRGGSPTAFDRVLASRMGAYAIDLLLA.GYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHSIIIVAEGVGSG.VD.FGRQIQEATGFETRATVLGHUQRGGSPTAFDRVLASRLGARAVELLLE.GKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVSEMIYPDVHK.LAKLVESNGYITRATVLGHUQRGGFSAFDRILGSRMVCAVNALLE.GTPDTPACVVS.LLS.GNQAVKLPLHELSIPRS SETTRRGSRLNIIIVAEGAIDNNGKPITSEDINNLVVRLGYDTRVTVLGHUQRGGTFSAFDRILGSRMVCAVNALLE.GTPDTPACVVS.LLS.GNQAVKLPLHECVQVTKDV GETRSRGSRLNIIIVAEGAIDNNGKPITSSGVKDLVVRLGYDTRVTVLGHUQRGGTFSAFDRILGSRMVCAVNALLE.GTPDTPACVVS.LLS.GNQAVKLPLHECVQVTKDV GETRSRGSRLNIIIKEGAIDRNGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTFSAFDRILSSKMOHEAVNALLE.ATPDTPACVVS.LLS.GNQSVKLPLHECVQVTKDV GETRSRGSRLNIIIKEGAIDNNGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTFSAFDRILSSKMOHEAVNALLE.ATPDTPACVVS.LLS.GNGSVKLPLHECVQVTKDV GETRSRGSRLNIIIKEGAIDNNGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTFSAFDRILSSKMOHEAVNALLE.ATPDTPACVVS.LS.GNGSVKLPLHECVQVTKDV GETRSRGSRLNIIIKEGAIDADNAKPISSSYVKDLVQRLGFDTRVTVLGHUQRGGTSAFDRILSSKMOHEAVNALLE.ATPDTPACVVS.LS.GNGSVKLPLHECVQVTKDV GETRSRGSRLTIIVNEGAIADDUNDNALIE.LGLDTNVTILGHUQRGGTSAFDRILSSKMOHEAVNALE.ATPDTPSPLIG.VI.LS.GNGSVKLPLHECVQVTKEV KTVKRG.LULRMEKCHENTTDFISPSDVKLVLDVRLGFDTRVTULGHUQRGGTSAFDRIKSKNALTUGUDAVKAVLE.STPDTSPSLIG.VIL.ENKIIKRFVLGKAKANLV RTHVRG.LULRMEKCHENTTDFISSSVKDLVGRGGTAVADRGGGSPTPFDRNFATKMGAKAMNMAG.KIKESVKNGRI.FA.NTPDSGCVLGARKRALVF KTTVKRG.LULRMEKCHENTTDFIFNLSS.EGKGIFDSRKNULGHQGGSPTPFDRNFATKMGAKAMNMAG.KIKESVKNGRI.FA.NTPDSGCVLGARKRALVF KTDLQRGLVLRMEKCHENTTFELVNLYSS.EGKGIFDSRKNULGHUGGGGSPTPFDRNFATKMGAKAMNMAG.KIKESVKNGRI.FA.NTPDSGCVLGARKRALVF
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast A-N Human N-C Rabbit M-C Mouse L-C Human L-C Yeast B-C	207 206 207 248 248 248 252 431 439 617 619 616 620 805	KAGIAKGKKHAIVAITEHNCDV.DELAHFIEKETGRETRATVLGHIQRGGSPVPYDRILASRMCAYAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHSIIIVAEGVGSG.VDFGRQIQEATGFETRVTVLGHUQRGGSPTAFDRVLASRLGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN AMLHQAQKRSVIVVVSEHIYPDVHK.LAKLVESKSGYITRATVLGHUQRGGSPTAFDRVLASRLGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN AMLHQAQKRSVIVVSEHIYPDVHK.LAKLVESLSGYITRATVLGHUQRGGTPSAFDRILGSRMCVEAVMALLEGTPDTPACVSLSGNQAVELPLMECVQVTKOV SETTRGSRLNIIIVAEGAIDRNGKPITSEDIKUVKRLGYDTRVTVLGHUQRGGTPSAFDRILGSRMCVEAVMALLEGTPDTPACVSLSGNQAVELPLMECVQVTKOV GETRSRGSRLNIIIIAEGAIDRNGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTPSAFDRILSSRMCHEAVMALLEATPDTPACVSLSGNQAVELPLMECVQVTKOV GETRSRGSRLNIIIIAEGAIDRNGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTPSAFDRILSSRMCHEAVMALLEATPDTPACVVSLSGNQSVRLPLMECVQVTKOV GETRSRGSRLNIIIAEGAIDRNGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTPAAVDRILSSRMCHEAVMALLEATPDTPACVVTLSGNQSVRLPLMECVQVTKOV KTTVKRGLVLRMEGALDDRUNGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTAVADRILSSRMCHEAVMALLESTPDTPSPLIAVNENKIVKRPLMESVKLTKAV GRHRSKGRRNTTIVVAEGALDDRUNPTANDVKDALIE.LGGIFDSRKVLGHUQGGSTPTPFDRFFTKMGAKAMNMAGKIKESYRNGRIFANTPDSGCVLGKRKRALVF KTTVKRGLVLRMEKCHENYTTDIFNLYSE.EGKGIFDSRKVUCHMQGGSPTPFDRMFATKMGAKAMNMAGKKIKESYRNGRIFANTPDSGCVLGKRKRALVF KTTVRGLVLRMEKCHENYTTEJINLYSE.EGKGIFDSRKVUCHQQGGAPTPFDRNYGTKLGVKALLEVSEKLRSYRNGRIFANTPDSGCVLGKRKALVF KTDIQGGLVLRMEKCHENYTTEJINLYSS.EGKGVFDCRTNVLGHLQQGGAPTPFDRNYGTKLGVKALLEVSEKLRSYRNGRIFANAPDSACVIGLRKKAVAF KTDIQGGLVLRMEKCHENYTTEJINLYSS.EGKGVFDCRTNVLGHLQQGGAPTPFDRNYGTKLGVKALLEVSEKLREVYRKGRVFANAPDSACVIGLRKKAVAF
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-N Yeast A-N Rubbit M-C Rabbit M-C Human L-C Yeast B-C Yeast A-C	207 206 207 248 248 248 252 431 439 617 619 616 620 805 811	KAGIAKGKKHAIVAITEHHCDV.DELAHFIEKETGRETRATVLGHIQRGGSPVPTDRILASRMGAYAIDLLLA.GYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHSIIVAEGVGSG.VD.FGRQIQEARGFETRATVLGHVQRGGSPYAFDRVLASRLGARAVELLE.GKGGRCVGIQNNQLVDHDIAEALA.N. AMLHQAQKRSVIVVSEMIYPDVHK.LAKLVESKGGYITRATVLGHVQRGGTPSAFDRILGSRMGVEAVNALLE.GTPDTPACVS.LE.GNQAVNLPLHECVQVTKDV SETRTRGSRLNIIIVAEGAIDNNGKPITSBDINNLVVRLGYDTRVTVLGHVQRGGTPSAFDRILGSRMGVEAVNALLE.GTPDTPACVS.LE.GNQAVNLPLHECVQVTKDV GETRSRGSRLNIIINAEGAIDNNGKPITSBJVKDLVVQRLGYDTRVTVLGHVQRGGTPSAFDRILGSRMGVEAVNALLE.GTPDTPACVS.LE.GNQAVNLPLHECVQVTKDV GETRSRGSRLNIIINEGAIDNNGKPISSSYVKDLVVQRLGYDTRVTVLGHVQRGGTPSAFDRILSSKMGHEAVNALLE.ATPDTPACVS.LS.GNQSVNLPLHECVQVTKDV GETRSRGSRLNIIINEGAIDNNGKPISSSYVKDLVVQRLGYDTRVTVLGHVQRGGTPSAFDRILSSKMGHEAVNALLE.ATPDTPACVS.LS.GNQSVNLPLHECVQVTKDV GETRSRGSRLNIIINEGAIDNNGKPISSSYVKDLVVQRLGYDTRVTVLGHVQRGGTPSAFDRILSSKMGHEAVNALLE.ATPDTPACVVS.LS.GNQSVNLPLHECVQVTKDV GETRSRGSRLNIIINEGAIDNNGKPISSSYVKDLVVQRLGFDTRVTVLGHVQRGGTPSAFDRILSSKMGHEAVNALLE.ATPDTPACVVS.LS.GNQSVNLPLHECVQVTKDV GETRSRGSRLNIIINEGAINDNNGKPISSSYVKDLVQRLGFDTRVTUGHVQRGGTPSAFDRILSSKMGHEAVNALLE.ATPDTPSPLIG.VI.S.MSGSVLPLHECVQVTKDV GETRSRGSRLNIIINEGAINDNNGKPISSSYVKDLVGRLGFDTRVTUGHVQRGGTPSAFDRILSSKMGHEAVNALE.ATPDTPSPLVGV.LS.GNGSVNLPLHECVQVTKDV GRINGGGRTNTIVVEGAIADDUNDTANDVKDALIE.GLGDTKVTLGHVQRGGTAVADVILSSKMGHEAVNALE.TRDTPSPLIG.VI.LS.MNGSVLPLHECVQKTKEV QRHNSKGRRNNTIIVAEGAINADTPISPSDVKLVDRLGGGTPFDRNFFTKMGKAMNNVLS.KIKSSVKNGRI.FA.NNTPSSCVLGKRKRALVF KTTVKRGLVLRNEKCHENYTTDIFHVSS.EGKGIFDSRKNVLGHNQGGSPTPFDRNFFTKMGKAMNHSG.KIKSSVKNGRI.FA.NNTPDSGCVLGKRKAAVF KTDIGGG.LVLNREKCHENYTTELVHLYSS.EGKGFDGSRKNVLGHNQGGSPTPFDRNFFATKMGKAMNHSG.KIKSSVKNGRI.FA.NNTPDSGCVLGKRKKAVF KTDIGGG.LVLNREKCHENYTTELVHLYSS.EGKGVFDCRTNVLGHLQGGAPTPFDRNFGTKLGVKAMLHVSS.KLREVSKRGRV.FA.NNAPDSGCVLGKRKAVF KTDIGGG.LVLNREKCHENYTTELVHLYSS.EGKGVFDCRTNVLGHLQGGAPTFPDRNFGTKLGVKAMLHVSS.KLREVSRNGRI.FA.NNAPDSGCVLGKKKAVF KTDIGGG.LVLNREKCHENYTTELVHLYSS.EGKGVFDCRTNVLGHLQGGAPTFPDRNFGTKLGVKAMLHVSS.KLREVSRNGRI.FA.NNAPDSGCVLGKKKAVF KTDIGGGLVVNLGKASSVSTQLLADISS.EGKGVFDCRTNVLGHLQGGAPTFPDRNFGTKLGV
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast A-N Human N-C Mouse L-C Human L-C Yeast B-C Yeast A-C	207 206 207 248 248 248 252 431 439 617 619 616 620 805 811	KAGIAKGKKHAIVAITEHNCDV.DELAHFIEKETGRETRATVLGHIQRGGSPVPYDRILASRMGAYAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENM KRCHERGKKHSIIIVAEGVGSG.VDFGRQIQEATGFETRVTVLGHUQRGGSPTAFDRVLASRLGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVVSEMIYPDVHK.LAKLVESKSGYITRATVLGHUQRGGSPTAFDRVLASRLGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVSEMIYPDVHK.LAKLVESKSGYITRATVLGHUQRGGTPSAFDRILGSRMCVEAVMALLEGTPDTPACVVSLSGNQAVKLPLMECVQVTKDV SETTRGSRLNIIIVAEGAIDRNGKPITSSDIVNLVVRLGYDTRVTVLGHUQRGGTPSAFDRILGSRMCVEAVMALLEGTPDTPACVVSLSGNQAVKLPLMECVQVTKDV GETKSRGSRLNIIIKEGAIDRNGKPITSSSIVKDLVVQRLGYDTRVTVLGHUQRGGTPSAFDRILGSRMCVEAVMALLEATPDTPACVVSLSGNQAVKLPLMECVQVTKDV GETKSRGSRLNIIIIAEGAIDRNGKPISSSYVKDLVVQRLGPDTRVTVLGHUQRGGTPSAFDRILSSKMCHEAVMALLEATPDTPACVVSLSGNQSVKLPLMECVQVTKDV GETKSRGSRLNIIIIAEGAIDRNGKPISSSYVKDLVQRLGPDTRVTVLGHUQRGGTAAVADRILATLQGLGAVNALLEATPDTPACVVSLSGNQSVKLPLMECVQVTKDV GKHRSGKRTTIVVVAEGAIAADLTPISPSDVHKVLVDRLGLDTRITITLGHUQRGGTAVANDRILATLQGLGAVNALLESTPDTPSPLIAVNENKIVKRLMESVKLTKAV GRHSKGRRNNTIIVAEGALDDQLNPVTANDVKDALIE.LGLDTKVTILGHUQRGGTAVANDRILATLQGVDXKAVLEFTPETPSPLIGIL.ENKIINKPLMESVKLTKAV KTTVKRGLVLRNEKCHENYTTDIFNIJSS.EGKGFDCRTNVLGHUQGGSSTPFPRNFTKNGKAMNMMAG.KIKESYRNGRI.FANTPDSGCVLGKRKALVF KTTVKRGLVLRNEKCHENYTTEFIYHLYSS.EGKGFDCRTNVLGHUQGGSSTPFPRNFTKNGKAMNMMAG.KIKESYRNGRI.FANTPDSGCVLGKRKALVF KTDLQRGLVLRNEKCHENYTTEFIYHLYSS.EGKGFDCRTNVLGHUGGGGSPTPFDRNFTKNGKAMMMMAG.KIKESYRNGRI.FANTPDSGCVLGKRKALVF KTDLQRGLVLRNEKKENYTTFIFIYHLYSS.EGKGFDCRTNVLGHUGGGSPTPFDRNFTKNGKAMMMAG.KIKESYRNGRI.FANTPDSGCVLGKRKALVF KTDLQRGLVLRNEKKENYTTEFIYHLYSS.EGKGFDCRTNVLGHUGGGSPTPFDRNFTKNGKAKMVMAG.KIKESYRNGRI.FANAPDSACVIGLKKKAVAF EXAEGRGRFGKLLLKSTNAKALSATKLAEVITAEAGGFDAKPAYPCHVQQGGLPSSKDVTXASFAVKCKFVISEN.NQAAIAERAAEENFNADDKTISDTAVVGVKGKHVY RHDKGENROKLLVRNEQASSVSTQLLADIISE.ASKGFGVRATAIPGHVQQGGUPSSKDVTXASRFAVKCKRFIEQWNKKNEASPNTDAKVLRFKFDTHGEKVPTVEHEDDSAAVIC
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-N Yeast B-C Rabbit M-C Human L-C Yeast B-C Yeast B-C Yeast A-C	207 206 207 248 248 252 431 439 617 619 616 620 805 811	KAGIAKGKKHAIVAITEHHCDV.DELAHFIEKETGRETRATVLGHIQRGGSPVPTDRILASRHGAYAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENH KRGHERGKKHSIIIVAEGGSG.VDPGRQIQEARGFETRATVLGHVQRGGSPYADRVLASRLGARAVELLLEGKGGRCVGIQNNOLVDHDIAEALAN. AMLHQAQKESVIYVVSEHIYPDVHK.LAKLVESKSGVITATVLGHVQRGGTPSADRVLASRLGARAVELLLEGKGGRCVGIQNNOLVDHDIAEALAN. SETKIRGSRLNIIIVAEGAIDNGKPITSEDINNLVVKRLGYDTRVTVLGHVQRGGTPSADRVLASRLGARAVELLLEGKDGTACVGSLSGNQAVRLPLMECVQVTKDV SETKIRGSRLNIIIVAEGAIDNGKPITSEDINNLVVKRLGYDTRVTVLGHVQRGGTPSADRVLASRLGSRMGVEAVMALLEGTPDTPACVVSLSGNQAVRLPLMECVQVTKDV GETKSRGSRLNIIIVAEGAIDNGKPITSSGVKDLVVRRLGYDTRVTVLGHVQRGGTPSAPRVLISSKMCHEAVMALLEGTPDTPACVVSLSGNQAVRLPLMECVQVTKDV GETKSRGSRLNIIIVAEGAIDNNGKPISSSYVKDLVVQRLGFDTRVTVLGHVQRGGTPSAPRVLISSKMCHEAVMALLEATPDTPACVVSLSGNQSVRLPLMECVQVTKDV GETKSRGSRLNIIIVAEGAIDDNNGKPISSSYVKDLVVQRLGFDTRVTVLGHVQRGGTPSAPRVLISSKMCHEAVMALLEATPDTPACVVSLSGNQSVRLPLMECVQVTKDV GETKSRGSRLNIIIVAEGAIDDNNGKPISSSYVKDLVVQRLGFDTRVTVLGHVQRGGTPSAPRVLISSKMCHEAVMALLEATPDTPACVVSLSGNQSVRLPLMECVQVTKDV GETKSRGSRLNIIVINEGAIDDNNGKPISSSYVKDLVVGRLGFDTRVTVLGHVQRGGTVAVDRILTLQUCVDAVKAVLESTPDTPSPLIGVVBNIIVKRVLPLMECVQVTKDV GKTHSKGRRNNTIVVEGAIADDQLNPVTANDVKDALIE.LGLDTVXILGHVQRGGTVAVDRILATLQUCVDAVKAVLEFTPETSPLIGIVWNIVESVKLFKAV QRHRSKGRRNNTIVVEGALIDDQLNPVTHNDVKDALIE.LGLDTVXILGHVQRGGSPTPFDRNFATKKGAKANNMAGKIKESYRNGRIFANTPDSGCVLGKRKALVF KTTVKRGLVLRNEKCHENYTTDFIFHLYSS.EGKGIFDSRKNVLGHMQGGSPTPFDRNFATKKGAKANNMAGKIKESYRNGRIFANTPDSGCVLGKRKALVF KTDIQRGLVLRNEKCHENYTTEFLYNLYSS.EGKGVFDCRTNVLGHLQGGAPTPFDRNYGTKLGVKANLEVSEKLRSVYKGRVFANAPDSACVIGLKKKVAVF KTDIQRGLVLRNEKCHENYTTEFLYNLYSS.EGKGVFDCRTNVLGHLQGGAPTPFDRNYGTKLGVKANLEVSEKLRSVYKGRVFANAPDSACVIGLKKKVAVF KTDIQRGLVLRNEKCHENYTTEFLYNLYSS.EGKGVFDCRTNVLGHLQGGAPTPFDRNYGTKLGVKANLEVSEKLRSVYKGRVFANAPDSACVIGLKKKVAVF KTDIQRGLVLRNEKCHENYTTEFLYNLYSS.EGKGVFDCRTNVLGHLQGGAPTPFDRNYGTKLGVKANLEVSEKLRSVYKGRVFANAPDSACVIGLKKKVAVF KTDIQRGLVLRNEKCHENYTTEFLYNLYSS.EGKGVFDCRTNVCGHVSSKSRVTAS
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-N Kabbit M-C Human L-C Yeast B-C Yeast B-C Yeast A-C	207 206 207 248 248 248 248 248 248 248 248 248 248	KAGIAKGKKHAIVAITEHUKOV.DELAHFIEKETGRETRATVLGHIQRGGSPTYDTALASRMGAVAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENM         KAGIAKGKKHAIVAITEHUKOV.DELAHFIEKETGRETRATVLGHIQRGGSPTAFDRVLASRLGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN.         AMLHQAQKRSVIVVVSEMIYDVIK.LAKLVESKSGYITRATVLGHUQRGGSPTAFDRVLASRLGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN.         AMLHQAQKRSVIVVVSEMIYDVIK.LAKLVESKSGYITRATVLGHUQRGGTPSAFDRILGSRMVEVAVALLEGTPDTPACVVSLSGNQAVELPLMECVQVTKDV         SETTRRGSRLNIIVAGGAIDRNGKPITSEDIKNLVVKRLGYDTRVTVLGHUQRGGTPSAFDRILGSRMVEVAVALLEGTPDTPACVVSLSGNQAVELPLMECVQVTKDV         GETRSRGSRLNIIIVAGGAIDRNGKPITSSSVKDLVVQRLGYDTRVTVLGHUQRGGTPSAFDRILGSRMVEVAVALLEATPDTPACVVSLSGNQAVELPLMECVQVTKDV         GETRSRGSRLNIIIKGGAIDRNGKPISSSYVKDLVVQRLGYDTRVTVLGHUQRGGTPSAFDRILSSKMOHEAVMALLEATPDTPACVVSLSGNGSVKLPLMECVQVTKDV         GETRSRGSRLNIIIKGGAIDRNGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTAVAFDRISSKMVLANALLEATPDTPACVVSLSGNGSVKLPLMECVQVTKDV         SKHRSKGRRNNTIIVAEGAIADDUNDKDALIE.LGLDTKVTLIGHUQRGGTAVAFDRISSKMVCLANAVLESTPDTPSPLIGILENKIIVKRPLHESVKLTKSV         SKHRSKGRRNNTIIVAEGAIADDUNDVKDALIE.LGLDTKVTLGHUQRGGSTPFPRHYTKHGKAANNMMSG.KIKESYNNGRI.FANTPDSGCVLGRKKRALVF         KTTVKRGLVLRNEKCHENYTTEFLYNLYSS.EGKGVFDCRTNVLGHUQGGSFTPFDRHFATNGAKANNMMG.KIKESYNNGRI.FANTPDSGCVLGRKKRALVF
E. coli B. Stear. S. citri Human M-N Rabbit M-N Mouse L-N Human L-N Yeast B-N Yeast A-N Human N-C Rabbit M-C Mouse L-C Human L-C Yeast B-C Yeast B-C Yeast A-C	207 206 207 248 248 248 252 431 617 619 616 620 805 811	<pre>KAGIAKGKKHAIVALDE.LAHFIEKETGRETRATVLGHIQRGGSPYPDRILASRMGAVAIDLLLA.GYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHSIIIVAEGVGSG.VD.FGRQIQEATGFETRVTVLGHUQRGGSPTAFDRVLASRLGARAVELLLE.GKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVVSEHIYPDVHK.LAKLVESKSGYITRATVLGHUQRGGSPTAFDRVLASRLGARAVELLLE.GKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVVSEHIYPDVHK.LAKLVESKSGYITRATVLGHUQRGGSPTAFDRVLASRLGARAVELLLE.GKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVVSEHIYPDVHK.JCALVESKSGYITRATVLGHUQRGGTPSAFDRILGSRMCVEAVMALLE.GTPDTPACVSLSGNQAVKLPLMECVQVTKDV SETTRRGSRLNIIIVAEGAIDRNGKPITSEDIVNLUVVRLGVDTRVTVLGHVQRGGTPSAFDRILGSRMCVEAVMALLE.GTPDTPACVSLSGNQAVKLPLMECVQVTKDV GETRSRGSRLNIIIIAEGAIDRNGKPISSSYVKDLVVQRLGPDTRVTVLGHVQRGGTPSAFDRILGSRMCVEAVMALLE.ATPDTPACVSLSGNQSVRLPLMECVQVTKDV GETRSRGSRLNIIIIAEGAIDRNGKPISSSYVKDLVVQRLGFDTRVTVLGHVQRGGTPSAFDRILSSKMCHAVMALE.ATPDTPACVVSLSGNQSVRLPLMECVQVTKDV GETRSRGSRLNIIIIAEGAIDRNGKPISSSYVKDLVVQRLGFDTRVTVLGHVQRGGTPSAFDRILSSKMCHAVMALE.ATPDTPACVVTLS.GNQSVRLPLMECVQVTKDV KTTVKRGLVLRMEGALDDQINPTIANDVRLAULELGTVTILGHVQRGGTAVAHDRKATLQVDAVMALE.STPDTPSPLIAVNENKIVKRPLMESVKLIKAV QRHRSKGRRNNTIIVDFGALDQDJNPTIANDVRLAULE.EGKGIFDSRKNVLGHNQGGSPTPFDRHFATKMGAKANNMAGS.KIKESYRNGRIFANTPDSGCVLGKRKALVF KTTVKRGLVLRMEKCHENYTTDFIFHLYSE.EGKGIFDSRKNVLGHQQGGSPTPFDRHFATKMGAKANNMAGS.KIKESYRNGRIFANTPDSGCVLGKRKALVF KTTVKRGLVLRMEKCHENYTTEFLYNLYSS.EGKGVPDCTNVLGHLQQGGAPTPFDRNYGTKLGVKAALWVSE.KLRDVYRKGRVFANAPDSACVIGLKKKAVAF KTDIQRGLVLRMEKCHENYTTEFLYNLYSS.EGKGVPDCTNVLGHLQQGGAPTPFDRNYGTKLGVKAALWVSE.KLRDVYRKGRVFANAPDSACVIGLKKKAVAF KTDIQRGLVLRMEKCHENYTTEFLYNLYSS.EGKGVPDCTNVLGHLQQGGAPTPFDRNYGTKLGVKAALWVSE.KLRDVYRKGRVFANAPDSACVIGLKKKAVAF KTDIQRGLVLRMEKCHENYTTEFLYNLYSS.EGKGVPDCTNVLGHLQQGGAPTPFDRNYGTKLGVKAALWSE.KLRDVYRKGRVFANAPDSACVIGLKKKAVAF KAG I EA GFDTR TVLGHVQQGCVPSSRVRTASRRAVKCIKFIEQWNKKREASPNTDAKVLRFKRDTHGEKVFTVEHEDDSAAVIC ************************************</pre>
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-N Yeast B-C Human L-C Yeast A-C Consensus	207 206 207 248 248 248 248 248 248 248 248 248 252 431 439 617 619 616 620 805 811	KAGIAKGKKHAIVAITEHINCOV.DELAHFIEKFEGETRATVLGHIQRGGSPTAFDRVLASRMGAVAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENM         KRGHERGKKHSIIIVAEGVGSG.VDFGRQIQEATGFETRATVLGHUQRGGSPTAFDRVLASRLGARAVELLLEGKGGRCVGIQNDQLVDHDIAEALAN.         ANLHQAQKRSVIVVVSEMIYPDVIK.LAKLVESKSGYITRATVLGHUQRGGSPTAFDRVLASRLGARAVELLLEGKGGRCVGIQNDQLVDHDIAEALAN.         ANLHQAQKRSVIVVVSEMIYPDVIK.LAKLVESSGYITRATVLGHUQRGGTPSAFDRILGSRMOVEAVMALLEGTPDTPACVVSLSGNQAVKLPLMECVQVTKDV         SETTRGSRLNIIIVAEGAIDNRKPITSEGVKDLVVRLGYDTRVTVLGHUQRGGTPSAFDRILGSRMOVEAVMALLEGTPDTPACVVSLSGNQAVKLPLMECVQVTKDV         GETRSRGSRLNIIIKEGAIDNRKPITSSSYVKDLVVQRLGYDTRVTVLGHUQRGGTPSAFDRILGSRMOVEAVMALLEATPDTPACVVSLSGNQSVKLPLMECVQVTKDV         GETRSRGSRLNIIIKEGAIDNRKPISSSYVKDLVVQRLGYDTRVTVLGHUQRGGTPSAFDRILSSKMOHEAVMALLEATPDTPACVVSLSGNQSVKLPLMECVQVTKDV         GETRSRGSRLNIIIKEGAIDNRKPISSSYVKDLVVQRLGYDTRVTVLGHUQRGGTPSAFDRILSSKMOHEAVMALLEATPDTPACVVSLSGNQSVKLPLMECVQVTKDV         YKHSKGRRNITIIVAEGAIDADLNPTANDVKDALIE.LGLDTKVTLGHUQRGGTASAFDRILSSKMOHEAVMALEATPDTPSLVVLSGNQSVKLPLMEEVQUTKDV         YKHSKGRRNITIIVAEGAIADDQLNPTANDVKDALIE.LGLDTKVTLGHUQRGGSPTPFDRFATKMGAKAMNHMAG.KIKESYNNGRIFANTPDSGCVLGRKRALVF         KTTVKRGLVLRMEKCHENYTTDFIFHLYSE.EGKGIFDSRKNVLGHUQGGSPTPFDRNFATKMGAKAMNHMAG.KIKESYNNGRIFANTPDSGCVLGRKRALVF         KTTVKRGLVLRMEKCHENYTTDFIFHLVSLSS.EGKGVFDCRTNVLGHUQGGSPTPFDRNFATKMGAKAMNHMAG.KIKESYNNGRIFANTPDSGCVLGRKRALVF
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Human N-C Rabbit M-C Mouse L-C Human L-C Yeast B-C Yeast B-C Yeast A-C Consensus E. coli	207 206 207 248 248 248 252 431 439 617 616 620 805 811 305	KAGIAKGKKHAIVALDU, DE. LAMFIEK. ETGRETRATVLGHIQRGGSPVPTORILASRMGAVAIDLLLA. GYGGRCVGIQN. EQLVHHDIIDAIENM KRGHERGKKHSIIIVAEGVGSG.VD., FGRQIQEATGFETRVTVLGHUQRGGSPTAFDRVLASRLGARAVELLLE. GKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVVSEHIYPDVHK.LAKLVESKSGYITRATVLGHUQRGGSPTAFDRVLASRLGARAVELLLE. GKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVSEHIYPDVHK.LAKLVESKSGYITRATVLGHUQRGGTPSAFDRILGSRMGVEAVMALLE. GTPDTPACVSLSGNQAVKLPLMECVQVTKDV SETTRGSRLNIIIVAEGAIDRNGKPITSSDIVLUVURLGYDTRVTVLGHUQRGGTPSAFDRILGSRMGVEAVMALLE. GTPDTPACVSLSGNQAVKLPLMECVQVTKDV GETRSRGSRLNIIIAEGAIDRNGKPITSSSVKDLVVQRLGYDTRVTVLGHUQRGGTPSAFDRILGSRMGVEAVMALLE. ATPDTPACVSLSGNQAVKLPLMECVQVTKDV GETRSRGSRLNIIIIAEGAIDRNGKPISSSVVKDLVQRLGYDTRVTVLGHUQRGGTPSAFDRILSSKMGHEAVMALLE. ATPDTPACVSLSGNQSVKLPLMECVQVTKDV GETRSRGSRLNIIIIAEGAIDRNGKPISSSVVKDLVQRLGFDTRVTVLGHUQRGGTAANDRILATLQGLGAVNALLEATPDTPACVSLSGNQSVKLPLMECVQVTKDV GETRSRGSRLNIIIIAEGAIDRNGKPISSSVKDLVQRLGFDTRVTVLGHUQRGGTAANDRILATLQGUAVNALLE. ATPDTPACVSLSGNQSVKLPLMECVQVTKDV GETRSRGSRLNIIIIAEGAIDRNGKPISSSVENDUVQLGFDTRVTVLGHUQRGGTAVANDRILATLQGUAVNALE. STPDTPSPLIAVN. ENKIVRKPLMESVKLTKAV QRHRSKGRRNNTIVVAEGAIAADLTPISPDVHKVLVDRLGLDTRITITLGHUQRGGTAVANDRILATLQGVDAVKAVLE. STPDTPSPLIAVN. ENKIVRKPLMESVKLTKAV KTTVKRGLVLRMEKCHENYTTDIFHLYSE. EGKGIFDSRKNVLGHWQGGSSPTPFDRHFATKHGAKAMMMAGKIKESYRNGRIFANTPDSGCVLGARKRALVF KTDIQRGLVLRMEKCHENYTTDIFHLYSE. EGKGIFDSRKNVLGHWQGGSSPTPFDRHFATKHGAKAMMMAGKIKESYRNGRIFANTPDSGCVLGARKRALVF KTDIQRGLVLRMEKCHENYTTEFLYNLSS. EGKGVFDCRTNVLGHUGGGGAPTPFDRNYGTKLGVKAMLHVSEKLRSVRNGRIFANTPDSGCVLGARKRALVF KTDIQRGLVLRMEKCHENYTTEFLYNLSS. EGKGVFDCRTNVLGHUGGGGAPTPFDRNYGTKLGVKAMLHVSEKLRSVRNGRUFANAPDSACVIGLKKKAVAF KTDIQRGLVLRMEKCHENYTTEFLYNLSS. EGKGVFDCRTNVLGHUGGGGAPTFPDRNYGTKLGVKAMLHVSEKLRSVRNGRUFANAPDSACVIGLKKKAVAF KTDIQRGLVLRMEKCHENYTTEFLYNLSS. EGKGVFDCRTNVLGHUGGGGAPTFPDRNYGTKLGVKAMLHSE.N.KLRSVRNGRUFANAPDSACVIGLKKKAVAF KTDIQRGCLVLRMEKCHENYTTEFLYNLSS. EGKGVFDCRTNVLGH
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-N Yeast B-C Rabbit M-C Mouse L-C Human L-C Yeast A-C Consensus E. coli B. Stear.	207 206 207 248 248 252 439 617 619 619 620 805 811 305 303	KAGIAKGKKHAIVAITEHNCDV.DELAHFIEKFTGETEATVLGHIQRGGSPTAFDRVLASRMGAVAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENM         KRGHERGKKHSIJIVAEGVGSG.VD.FGRQIQEATGFETRATVLGHUQRGGSPTAFDRVLASRLGARAVELLEE.GKGGRCVGIQNNQLVHHDIAEALAN.         ANLHQAQKRSVIVVSEMIYPDVIK.LALVESKSGYITRATVLGHUQRGGSPTAFDRVLASRLGARAVELLEE.GKGGRCVGIQNNQLVHHDIAEALAN.         ANLHQAQKRSVIVVSEMIYPDVIK.LALVESKSGYITRATVLGHUQRGGTPSAFDRILGSRMOVEAVWALLE.GTPDTPACVVSLSGNQAVKLPLMECVQVTKDV         SETTKRGSRLNIIIVAEGAIDNRKPITSEDVINLVVRLGYDTRVTVLGHUQRGGTPSAFDRILGSRMOVEAVWALLE.GTPDTPACVVSLSGNQAVKLPLMECVQVTKDV         GETKSRGSRLNIIIKEGAIDNRKPITSSSYNDLVVQRLGFDTRVTVLGHVQRGGTPSAFDRILGSRMOVEAVMALLE.ATPDTPACVVSLSGNQSVRLPLMECVQVTKDV         GETKSRGSRLNIIIKEGAIDNRKPISSSYNDLVVQRLGFDTRVTVLGHVQRGGTPSAFDRILSSKMOHEAVMALLE.ATPDTPACVVSLSGNQSVRLPLMECVQVTKDV         GETKSRGSRLNIIIKEGAIDNRKPISSSYNDLVVQRLGFDTRVTVLGHVQRGGTPSAFDRILSSKMOHEAVMALLE.ATPDTPACVVSLSGNQSVRLPLMECVQVTKDV         GETKSRGSRLTIIVAEGAIADDQLMPVTANDVLONGLGFDTRVTUGHVQRGGTAVAFDRATATLQCUDAVALE.STDPTSPLIGULENKIIVKRPLHEEVVQTKKDV         YKHNSKGRRNNTIIVAEGAIADDQLMPVTANDVLONGLGFDTRVTUGHVQRGGTAVAFDRATATLQCUDAVALE.STDPTSPLIGULENKIIVKRUHESVKLTKSV         YKHNSKGRRNTIIVVAEGAIADDQLMPVTANDVLONGLGFDTRVUGHQGGGAPTPFDRMFATKMCKANNMHSG.KIKESYNNGRIFANTPDSGCUCARKRALVF         KTTVKRGLVLRMEKCHENYTTDFIFHLYSE.EGKGIFDSRKNULGHNQGGSPTPFDRMFATKMCKANNMHSG.KIKESYNNGRIFANTPDSGCUCARKRALVF
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Human L-N Human M-C Rabbit M-C Mouse L-C Human L-C Yeast A-C Consensus E. coli B. Stear. S. citri	207 206 207 248 248 248 252 431 439 617 616 620 805 811 305 303 303	KAGIAKGKKHAIVALTEHKOUV.DELAMFIEKETGRETRATVLGHIQRGGSPYPYDRILASRMGAYAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENM         KRGHERGKKHSIIIVAEGVGSG.VDFGRQIQEATGFETRVTVLGHUQRGGSPTAFDRVLASRLGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN.         AMLHQAQKRSVIVVVSEMIYPDVIK.LAKLVESKSGYITHATVLGHUQRGGSPTAFDRVLASRLGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN.         AMLHQAQKRSVIVVVSEMIYPDVIK.LAKLVESKSGYITHATVLGHUQRGGSPTAFDRVLASRLGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN.         AMLHQAQKRSVIVVVSEMIYPDVIK.LAKLVESKSGYITHATVLGHUQRGGTPSAFDRILGSRHOVEAVMALLEGTPDTPACVVSLSGNQAVKLPLMECVQVTKOV         SETTRRGSRLNIIIVAEGAIDRNGKPITSSEVKDLVVRLGYDTRVTVLGHUQRGGTPSAFDRILGSRHOVEAVMALLEATPDTPACVVSLSGNQAVKLPLMECVQVTKOV         GETRSRGSRLNIIITAEGAIDRNGKPISSSYVKDLVVQLGFDTRVTVLGHUQRGGTPSAFDRILSSKMCHEAVMALLEATPDTPACVVSLSGNQSVKLPLMECVQVTKOV         GETRSRGSRLNIIITAEGAIDRNGKPISSSYVKDLVQQLGFDTRVTVLGHUQRGGTAAVADRILATLQGLAVNALLEATPDTPACVVSLSGNQSVKLPLMECVQVTKOV         GETRSRGSRLNIIIVAEGAIDADLTPISSSYVKDLVQRLGPDTRVTVLGHUQRGGTAVADRILATLQGUAVKAVLEATPDTPACVVSLSGNQSVKLPLMECVQVTKOV         GETRSRGSRLNIIIVAEGALDDQULMPVTAMDVKDALIE.LGLDTKVTILGHUQRGGTAVADRILATLQGUAVKAVLETPETPSPLIAVNENKIVKRHALEVKKRKXVX         KTTVKRGLVLRNEKCNENYTTDFIFHLYLSS.EGKGIFDSRKWULGHUQGGSTPTPDRHFATKHGAKAMMMAG.KIKESYRNGRIFANTPDSGCVLGARKRALVF         KTTVKRGLVLRNEKCNENYTTDFIFHLYLSS.EGKGIFDSRKWULGHUGGGSTPTPDRHFATKHGAKAMMMAG.KIKESYRNGRIFANTPDSGCVLGARKRALVF          KTDLQRGLVLRNEKCNENYTTDFIFHLYLSS.EGKGFDCRTNVCHLGUGGGSPTPFDRHFATKHGAKAMMMAG.KIKESYRNGRIFANTPDSGCVLGARKRALVF
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-N Yeast B-C Rabbit M-C Human L-C Yeast A-C Consensus E. coli B. Stear. S. citri Human M-N	207 206 207 248 248 252 439 617 619 616 620 805 811 305 303 309 359	<pre>KAGIAKGKKHAIVAITEHNCDV.DELAHFIEKFTGRETRATVLGHIQRGGSPVPYDRILASRMCAVAIDLLLA.GYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHSIIIVAEGGSG.VDGRQIQEATGFETRATVLGHUQRGSPTAFDRVLASRLGARAVELLE.GKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVSEHIYPDVHK.LAKLVESKSGYITRATVLGHUQRGGPTAFDRVLASRLGARAVELLE.GKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVSEHIYPDVHK.LAKLVESKSGYITRATVLGHUQRGGTPSAFDRILGSRMCVEAVNALLE.GTPDTPACVVSLSGNQAVKLPLMECVQVTKDV SETKTRGSRLNIIIVAEGAIDNNGKPITSBOVKDLVVRLGYDTRVTVLGHUQRGGTPSAFDRILGSRMCVEAVNALLE.GTPDTPACVVSLSGNQAVKLPLMECVQVTKDV GETKSRGSRLNIIIKEGAIDNNGKPITSSSYVKDLVVQRLGFDTRVTVLGHUQRGGTPSAFDRILGSRMCVEAVNALLE.ATPDTPACVVSLSGNQSVKLPLMECVQVTKDV GETKSRGSRLNIIIKEGAIDNNGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTPSAFDRILSSKMCHEAVNALLE.ATPDTPACVVSLSGNQSVKLPLMECVQVTKDV GETKSRGSRLNIIIKEGAIDNNGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTPSAFDRILSSKMCHEAVNALLE.ATPDTPACVVSLSGNQSVKLPLMECVQVTKDV GETKSRGSRLNIIIKEGAIDNNGKPISSSYVKDLVQRLGFDTRVTUGHUQRGGTPSAFDRILSSKMCHEAVNALLE.ATPDTPACVVSLSGNQSVKLPLMECVQVTKDV GETKSRGSRLNIIIKEGAIADNEKEGISSYVKDLVVQRLGFDTRVTUGHUQRGGTPSAFDRILSSKMCHEAVNALE.ATPDTPACVVSLSGNQSVKLPLMECVQVTKDV QRHRSKGRTNITIVAEGAIADDQLNPVTANDVKDALIE.LGLDTVXTILGHUQRGGTPSTPFDRFFTKMCAKANNHSC.KIKSSYKNGRIFANTPDSGCUCARKRALVF KTTVKRGLVLRMEKCHENYTDFIFHLSS.EGKGIFDSRKNVLGHNQGGSPTPFDRFFTKMCAKANNHSG.KIKSSYKNGRIFANTPDSGCUCARKRALVF KTDIQRGLVLRMEKCHENYTDFIFHLSS.EGKGIFDSRKNVLGHUQGGAPTPFDRMFTKMCAKANNHSG.KILSSSTNGRIFANAPDSACVIGLKKVANF KTDIQRGLVLRMEKCHENYTTEFLYNHJSS.EGKGVPCRTNVLGHUQGGAPTPFDRMFTKMCAKANNHSG.KILSSTNNGRIFANAPDSACVIGLKKVANF KTDIQRGLVLRMEKCHENYTTEFLYNHJSS.EGKGVPCRTNVLGHUQGGAPTPFDRMFTKMCKAKANNHSG.KILSSTNNGRIFANAPDSACVIGLKKVANF KTDIQRGLVLRMEKCHENYTTEFLYNHJSS.EGKGVPCRTNVLGHUQGGAPTPFDRMFTKMCKLAVRHENSSSTNGRIFANAPDSACVIGLKKVANF KTDIQRGLVLRMEKCHENYTHEFLYNHUSS.EGKGVPCRTNVLGHUQGGVPSSDRVTARTHGTKLGVKAKLESS.KLREVYRKGRVFANAPDSACVIGLKKVANF KTTVKRCALLKKEASSYSTGULADIISE.ASKGKPCHTAVGHUQGGVPSSDRVTARTHATKAKANNHSG.KILSESTNANGTIFANAPDSACVIGLKKVANF RETMAFGAUNGKLUKARAKLESSSTS</pre>
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-N Human M-C Rabbit M-C Human L-C Yeast B-C Yeast B-C Yeast B-C Yeast B-C Yeast A-C Consensus E. coli B. Stear. S. citri Human M-N Rabbit M-N	207 206 207 248 248 252 439 617 619 616 620 805 811 305 303 309 359 359	KAGIAKGKKHAIVAITEHKCUV.DELANFIEKETGRETRATVLGHIQRGGSPVPYDRILASRMGAVAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENM         KRGHERGKKHSIIIVAEGVGSG.VDFGRQIQEATGFETRATVLGHIQRGGSPTAPDRVLASRLGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN.         AMLHQAQKRSVIVVVSEMIYPDVIK.LAKLVESKSGYITRATVLGHUQRGGSPTAPDRVLASRLGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN.         AMLHQAQKRSVIVVVSEMIYPDVIK.LAKLVESKSGYITRATVLGHUQRGGSPTAPDRVLASRLGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN.         AMLHQAQKRSVIVVVSEMIYPDVIK.LAKLVESKSGYITRATVLGHUQRGGTPSAPDRILGSRMOVEAVMALLEGTPDTPACVSLSGNQAVKLPLMECVQVTKDV         SETTRRGSRLNIIIVAEGAIDNNGKPITSEDVKDLVVRLGYDTRVTVLGHUQRGGTPSAPDRILGSRMOVEAVMALLEGTPDTPACVSLSGNQAVKLPLMECVQVTKDV         GETRSRGSRLNIIIKEGAIDNNGKPITSSSVKDLVVQRLGPDTRVTVLGHUQRGGTPSAPDRILSSKMOHEAVMALLEATPDTPACVSLSGNQSVKLPLMECVQVTKDV         GETRSRGSRLNIIIKEGAIDNNGKPISSSVKDLVVQRLGPDTRVTVLGHUQRGGTAVANDRILSSKMOHEAVMALLEATPDTPACVSLSGNQSVKLPLMECVQVTKDV         GETRSRGSRLTIIVAEGAIAADLPISSSVKDLVVQRLGPDTRVTVLGHUQRGGTAVANDRILSSKMOHEAVMALLEATPDTPACVSLSGNQSVKLPLMEEVQUTKDV         SKHRSKGRRNNTIIVAEGAIAADLPISSSVKDLVVDRLLGLDTKVTILGHUQRGGTAVANDRILATLQGVDAVKAVLEFTPDTPSPLIGLVN.ENKIVRPHAESVLTKAV         SKHRSKGRRNNTIIVAEGAIADDQLNPVTANDVKDALIE.LGLDTKVTILGHUQRGGSTPTPDRNFPTRAKAKAMMMAG.KIKESYNNGRI.FANTPDSGCVLCHRKRALVF         KTVKRGLVLRNEKCNENYTTDIFINLISSEGKGFDGRKNVLGHUQGGSTPTPDRNFPTRAKAKAMMMAG.KIKESYNGRI.FANTPDSGCVLCHRKRALVF
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-N Yeast A-N Human N-C Rabbit M-C Human L-C Yeast A-C Consensus E. coli B. Stear. S. citri Human M-N Rabbit M-N Rabbit M-N	207 206 207 248 248 252 439 617 619 616 620 805 811 305 303 309 359 359	<pre>KAGIAKGKKHAIVAITEHHKCDV.DELAHFIEKFTGRETRATVLGHIQRGGSPVPYDRILASRMGAVAIDLLLA.GYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHSIJIVAEGGSG.VDFGRQIQEARTFETRYTVLGHVQRGGSPYADRVLASRLGARAVELLE.GKGGRCVGIQNNQLVDHDIAEALAN. ANLHQAQKKSVIVVSEMIYPDVHK.LAKLVESKGYITRATVLGHTQRGGNPYADRVLASRLGARAVELLE.GKGGRCVGIQNNQLVDHDIAEALAN. ANLHQAQKKSVIVVSEMIYPDVHK.LAKLVESKGYITRATVLGHTQRGGNPAADDRYARQMAQFAVGQIIA.GVGGLAIGNGGDQIIARPIHEALSIPRSS SETKTRGSRLNIIIVAEGAIDNNGKPITSBCVKDLVVRLGYDTRVTVLGHVQRGGTPSAFDRILGSRMCVEAVNALLE.GTPDTPACVVSLSGNQAVKLPLMECVQVTKDV GETKSRGSRLNIIIVAEGAIDNNGKPITSSSVKDLVVRLGYDTRVTVLGHVQRGGTPSAFDRILGSRMCVEAVNALLE.ATPDTPACVSLSGNQSVKLPLMECVQVTKDV GETKSRGSRLNIIIAEGAIDNNGKPITSSSVKDLVVQRLGFDTRVTVLGHVQRGGTPSAFDRILSSKMCHEAVNALLE.ATPDTPACVVSLSGNQSVKLPLMECVQVTKDV GETKSRGSRLNIIIVAEGAIDDNNGKPITSSSVKDLVVQRLGFDTRVTVLGHVQRGGTPSAFDRILSSKMCHEAVNALLE.ATPDTPACVVSLSGNQSVKLPLMECVQVTKDV GETKSRGSRLNIIIVAEGAIDDNNGKPITSSSVKDLVVQRLGFDTRVTUGHVQRGGTPSAFDRILSSKMCHEAVNALLE.ATPDTPACVVSLSGNQSVKLPLMECVQVTKDV GETKSRGSRLNIIVAEGAIAADTPISSSVKDLVVQRLGFDTRVTUGHVQRGGTPSAFDRILSSKMCHEAVNALE.ATPDTPACVVSLSGNQSVKLPLMECVQVTKDV GETKSRGSRLTIIVAEGAIADDUNPTANDVKDALIE.LGLDTKVTILGHVQRGGTSAFDRILSSKMCHEAVNALE.ATPDTPSPLIGTVL.SVNLPBSCVLGKRKALVF KTTVKRGLVLRNEKCHSKYTDFIFHLSSS.EGKGIFDSRKNVLGHNQGGSPTPFDRHFATKMCAKANNHSG.KIKSSYKNGRIFANTPDSGCVLGKRKALVF KTTVKRGLVLRNEKCHSKYTDFIFHLSS.SKGFDCTNVJCHLQGGAPTPFDRHFATKMCAKANNHSG.KIKSSYKNGRIFANPDSGCVLGKRKALVF KTTVKRGLVLRNEKCHSKYTTEFLYNHLSS.EGKGIFDSRKNVLGHNQGGSVFTPFDRHFATKMCAKANNHSG.KIKSSYKNGRIFANAPDSGCVLGKRKAVAF KTTVKRGLVLRNEKCHSKYTHEFLYNHLSS.EGKGIFDSRKNVLGHNQGGSVFTPFDRHFATKMCAKANNHSG.KIKSSYNGRIFANAPDSGCVLGKRKAVAF KTTVKRGLVLRNEKCHSKYTHEFLYNHLSS.EGKGIFDSRKNVLGHNQGGSVFSTDRTARMAKALKVSE.KLRSVYKGRVFANAPDSGCVLGKKKAVAF KTTVKRGLVLRNEKCHSKYTHEFLYNHLSS.EGKGIFDSRKNVLGHNQGGVFSSKDRVTASRFAVKCIKFIEQMNKKBASSNTDAKVLFRKDTHAVSKLKKAVAF RG I E A GFDTR TVVLGHVQRGG PAFDR LA HG AV LE NV K KTTVLGHVGKS TRANGEKEPDEALUKRGSSFNNNMFYY.KLLAH TKMDEKRFDDAKKLGKSS</pre>
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-C Human L-C Yeast A-C Yeast A-C Yeast A-C Consensus E. coli B. Stear. S. citri Human M-N Rabbit M-N Rabbit M-N	207 206 207 248 248 252 431 439 619 619 619 620 805 811 305 303 309 359 359 359 359 359	<pre>KAGIAKGKKHAIVAITEHKCUV.DELAHFIEKETGRETRATVLGHIQRGGSPTAPDRVLASRUGAVAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHSIIIVAEGVGSG.VDFGRQIQEATGFETRATVLGHIQRGGSPTAPDRVLASRUGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVVSEMIYPDVHK.LAKLVESKSGYITRATVLGHVQRGGSPTAPDRVLASRUGARAVELLLEGKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVVSEMIYPDVHK.LAKLVESSGYITRATVLGHVQRGGTPSAPORLGSRWCPAVNALLEGTPDTPACVVSLSGNQAVKLPLMECVQVTKDV SETTRGSRLNIIIVAEGAIDNAKPITSEGVKDLVVRLGYDTRVTVLGHVQRGGTPSAPORLGSRWCPAVNALLEGTPDTPACVSLSGNQAVKLPLMECVQVTKDV GETRSRGSRLNIIIKEGAIDNAKPITSSSVKDLVVQRLGPDTRVTVLGHVQRGGTPSAPORLGSRWCPAVNALLEATPDTPACVSLSGNQAVKLPLMECVQVTKDV GETRSRGSRLNIIIKEGAIDNAKPITSSSYVKDLVVQRLGPDTRVTVLGHVQRGGTPSAPORLISSKMCHEAVNALLEATPDTPACVSLSGNGSVKLPLMECVQVTKDV GETRSRGSRLNIIIKEGAIDNAKPITSSSYVKDLVVQRLGPDTRVTUGHVQRGGTPSAPORLISSKMCHEAVNALLEATPDTPACVYSLSGNGSVKLPLMECVQVTKDV GETRSRGSRLNIIIKEGAIDNAKPITSSSYVKDLVVQRLGPDTRVTUGHVQRGGTAVATORLISSKMCHEAVNALLEATPDTPACVYSLSGNGSVKLPLMECVQVTKEV SKHRSRGRRTNTIVVAEGAIAADIPISSSYVKDLVDVRLGFDTRVTUGHVQRGGTAVATORLISSKMCHEAVNALEATPDTPSPLGILENKIIKRFLHESVKLTKAV GRHRSKGRRNNTIIVAEGAIADDQLNPVTANOVKDALIE.JGLDTKVTLGHVQRGGTAVATORLISSKMCHEAVNALETPTETPSPLGILENKIIKRFLHESVKLTKAV KTTVKRGLVLRNEKCHENYTTEIISPSDVKLUDHGLGGIFDSKNVLGHMQQGGSPTPFDRNFATKHGAKANNMMG.KIKESYNNGRI.FANTPDSGCVLGRKRALVF KTDIQRGLVLRNEKCHENYTTEIINISS.EGKGVFDCRTNVLHLQGGGSPTPFDRNFATKHGAKANNMMG.KIKESYNNGRI.FANTPDSGCVLGRKRALVF KTDIQRGLVLRNEKCHENYTTEIINISS.EGKGVFDCRTNVLHLQGGGAPTPFDRNFATKHGAKANNMMG.KIKESYNNGRI.FANAPDSACVIGLKKXAVAF KTDIQRGLVLRNEKCHENYTTEIINISS.EGKGVFDCRTNVLHLGQGGAPTPFDRNFATKHGAKANNMMG.KIKESYNNGRI.FANAPDSACVIGLKKKAVAF KTDIQRGLVLRNEKCHENYTTEIINISS.EGKGVFDCRTNVLHLGQGGAPTPFDRNFATKHGAKANNMMG.KINSES.KLRSYNRGRI.FANAPDSACVIGLKKKAVAF KTDIQRG.LLVRAKGKSSSYSTGLLADIISE.ASGGFDAKPAYPGHVQGGCPSSDRVTASFEKLOKANNESSE.KLRSYNRGRI.FANAPDSACVIGLKKKAVAF RDKGENFDEAKLGRSSFNNMEYY.KLLAH TANDEKKFPOZAKLKRSSFNNMEYY.KLLAH TANDEKKFPOZAKLKRSSFNNMEYY.KLLAH TANDEKK</pre>
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-N Yeast A-N Human M-C Human L-C Yeast A-C Consensus E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Human L-N Human L-N Human L-N	207 206 248 248 248 252 431 439 617 619 616 620 805 811 305 303 309 359 359 359 359 359 359 359 359	<pre>KAGIAKGKKHAIVAITEHHKCDV.DELAHFIEKFTGRETRATVLGHIQRGGSPVPYDRILASRMGAVAIDLLLA.GYGGRCVGIQNEQLVHHDIIDAIENM KAGHERGKKHSIJIVAEGGSG.VDFGRQIQEARTFETRVTVLGHVQRGGSPYAPDRVLASRLGARAVELLE.GKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVSEMIYPDVHK.LAKLVESKGYITRATVLGHTQRGGNPYAPDRVLASRLGARAVELLE.GKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVSEMIYPDVHK.LAKLVESKGYITRATVLGHTQRGGNPSAFDRILGSRMGVEAVHALLE.GTPDTPACVVSLEGNQAVKLPLAECVQVTKDV SETTRGSRLNIIIVAEGAIDDNGKPITSBDINLVVRLGYDTNVTVLGHVQRGGTPSAFDRILGSRMGVEAVHALLE.GTPDTPACVVSLEGNQAVKLPLAECVQVTKDV GETTSRGSRLNIIIVAEGAIDDNGKPITSSSYNDLVVQRLGFDTNVTVLGHVQRGGTPSAFDRILGSRMGVEAVHALLE.ATPDTPACVVSLEGNQAVKLPLAECVQVTKDV GETTSRGSRLNIIIAEGAIDDNGKPITSSSYNDLVVQRLGFDTNVTVLGHVQRGGTPSAFDRILSSKMGHEAVHALLE.ATPDTPACVVSLSGNQSVKLPLAECVQVTKDV GETTSRGSRLNIIIVAEGAIDDNGKPITSSSYNDLVVQRLGFDTNVTUGHVQRGGTPSAFDRILSSKMGHEAVHALLE.ATPDTPACVVSLSGNQSVKLPLAECVQVTKDV GETTSRGSRLNIIIVAEGAIDDNGKPITSSSYNDLVVQRLGFDTNVTUGHVQRGGTPSAFDRILSSKMGHEAVHALLE.ATPDTPACVVSLSGNQSVKLPLAECVQTKEV YSKHRSKGRTNIVVEGAIAADIPTISSSYNDLVVQRLGFDTNVTUGHVQRGGTPSAFDRILSSKMGHEAVHALLE.ATPDTPACVVSLSGNQSVKLPLAECVQTKEV YKTTVKGGLVLRMEGAIAADIPTISPSDVKLVUDRLGLGTNTTILGHVQRGGTAVADRILATLQQUDAVKAVLE.STPDTPSPLIGUN. FNIVKRPLHESVKLTKAV QRHNSKGRTNIVIVAEGAIADDQLNPVTANDVKDALIE.LGLGTVXTUGHVQRGGTPFPDRFFTKMGAKANNHSG.KIKESYNNGRIFANTPDSGCVLGKRKAAVF KTTVKGGLVLRMEKCHENYTTPIFHVLYSS.EGKGIFDSRKNVLGHNQGGSPTPFDRFFTKMGAKANNHSG.KIKESYNNGRIFANTPDSGCVLGKRKAAVF KTTVKGGLVLRMEKCHENYTTELVHVSS.EKGKGIFDSRKNVLGHNQGGSVFTPFDRFFTKMGAKANNHSG.KIKESYNNGRIFANAPDSACVIGLKKKAVAF KTTVKGGLVLRMEKCHENYTTELVHVSS.EKGKGVCATNICHLQGGAPTFFDRNYGTKLGVKAMLHVSS.KLKENVSG.N.PANAPDSACVIGLKKKAVAF KTTVKGGLVLRMEKCHENYTTELVHVSS.EKGKVCATAIPHONVQGGVVSSKARVISTRAFANAKALHVSS.KLKSVNGRGVVFANAPDSACVIGLKKKAVAF KTTVKGGLVLRMEKCHENYTKELLANGTSSEGKGFDGARAPAYPGHVQGGVVSSKARVISTRAFANAKALHVSS.KLKSVNGRGVVFANAPDSACVIGLKKKAVAF GG I E A GFDTR TVLGHVQRGG PAPDR LA NG AV LE NV V E NV E KREPKGENKNGLLKRGSSFNNNMYYY.KLLAH TANDEKKFPOLANUTYKS TANDEKK</pre>
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-N Yeast B-C Human L-C Yeast A-C Consensus E. coli B. Stear. S. citri Human M-N Rabbit M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-N Yeast A-N	207 206 207 248 248 252 431 439 617 619 616 620 805 811 305 303 303 359 359 359 363 542 549	<pre>KAGIAKGKKHAIVAITEHHCDV.DE.LAHFIEKETGRETRATVLGHIQRGGSPUPVDRILASRMGAYAIDLLLA.GYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHSIIIVAEGVGSG.VD.FGRQIQEATGFETRVTVLGHUQRGGSPTAFDRVLASRLGARAVELLLE.GKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVSENIFPDVHK.LAKLVESSKGVITATVLGHUQRGGTPAFDRVLASRLGARAVELLLE.GKGGRCVGIQNNQLVDHDIAEALAN. AMLHQAQKRSVIVVSENIFPDVHK.LAKLVESSKGVITATVLGHUQRGGTPAFDRILGSRGVEAVMALLE.GTPDTPACVVSLS.GKQVKLPLHECVQVTKDV SETTKRGSRLNIIIVAEGAIDKKGKPITSEDIKNLVVKRLGVDTRVTVLGHUQRGGTPAFDRILGSRGVEAVMALLE.GTPDTPACVVSLS.GKQVKLPLHECVQVTKDV GETTSRGSRLNIIIIAEGAIDRKGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTPAFDRILGSRGVEAVMALLE.ATPDTPACVVSLS.GKQVKLPLHECVQVTKDV GETTSRGSRLNIIIIAEGAIDRKGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTPAFDRILSSKMGHEAVMALLE.ATPDTPACVVSLS.GKQVKLPLHECVQVTKDV GETTSRGSRLNIIIIAEGAIDRKGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTPAFDRILSSKMGHEAVMALLE.ATPDTPACVVSLS.GKQVKLPLHECVQVTKDV GETTSRGSRLNIIIIAEGAIDRKGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTAVATDRILATLQGLAVMALLE.ATPDTPACVVSLS.GKQVKLPLHECVQVTKDV GETTSRGSRLNIIIAEGAIDAKGKPISSSYVKDLVVQRLGFDTRVTVLGHUQRGGTAVATDRILATLQGLAVMALLE.STPDFACVSLS.GKQVKLPLHECVQVTKDV QRHRSKGRRMNTIIVAEGAIDAUNPVTANDVKDALIE.GLGDTKVTLGHUQGGASPTPFDRNFATKMGAKANNMKG.KIKESYRMGRIFA.NTPDSGCVLGHRKRALVF KTTVKRGLVLANEKCNENTTDFIFNLYSE.EGKGIFDSRKNVLGHMQGGGSPTPFDRNFATKMGAKANNMKG.KIKESYRMGRIFA.NTPDSGCVLGHRKRALVF KTDIQRGLVLANEKCNENTTDFIFNLYSE.SGKGVFDCRTNVLGHLQQGGAFTPFDRNFATKMGAKANNMKG.KIKESYRMGRIFA.NTPDSGCVLGHRKRALVF KTDIQRGLVLANEKCNENTTDFIFNLYSE.SGKGVFDCRTNVLGHLQQGGAFTPFDRNFATKMGAKANNMKG.KIKESYRMGRIFA.NAPDSACVIGLKKAVAF KTDIQRGLVLANEKCHENTTDFIFNLYSE.SGKGVFDCRTNVLGHLQQGGAFTPFDRNFATKMGAKANNMKG.KIKESYRMGRIFA.NAPDSACVIGLKKAVAF KTDIQRG.LVLANEKCHENTTDFIFNLYSE.SGKGVFDCRTNVLGHLQQGGAFTPFDRNFATKMGAKANNMKG.KIKESFNKGKV.FA.NAPDSACVIGLKKAVAF KTDIQRGN.LVLANEKCHENTTDFIFNLYSE.SGKGVFDCRTNVLGHLQQGGFFDFDRNFATKMGAKANNMKG.KIKESFNTAGKVLKVFRKEGVFASANVGKKGSHUY RHDKGENRKGLLVANEGASVYSTGLLADIISE.ASKGKVQGGFPAFDPIDRYATKLGVKANLKNEASPNTDAKVLFRKEVYRGKV.FA.NAPDSACVIGKKKAVAF KTDIQRGNCLLKANGGASSYTSTGLAUHSENKLA ASKGKKK</pre>
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-N Yeast B-C Human M-C Human L-C Yeast B-C Yeast B-C Yeast A-C Consensus E. coli B. Stear. S. citri Human M-N Mouse L-N Human L-N Yeast B-M Yeast A-N	207 206 248 248 248 252 431 439 617 619 616 620 805 811 305 303 309 359 359 359 359 359 359 359 359 359 35	<pre>KAGIAKGKKHAIVAITEHNCDV.DE.LAHFIEKETGRETATVLGHIQRGGSPVPYDRILASRMCAYAIDLLLA.GYGGRCVGIQNEQLVHHDIIDAIENM KRGHERGKKHAIVAITEHNCDV.DE.LAHFIEKETGRETATVLGHUQRGGSPTAPDRVLASRGARAVELLLE.GKGGRCVGIQNNQLVDHDIAEALAN AMLHQAQKRSVIVVSEHIYPDVHK.LAKLVESKSGYITRATVLGHUQRGGTSAFDRVLASRGARAVELLLE.GKGGRCVGIQNNQLVDHDIAEALAN AMLHQAQKRSVIVVSEHIYPDVHK.LAKLVESKSGYITRATVLGHUQRGGTSAFDRILASRMCARAVGAIA.GVGGAVGQIA.GVGGAD.DQIIAPPIMELASIPRSS SETTRGSRLNIIIVAEGAIDDKGKPITSEDIKNLVVKRLGIDTNVTVLGHUQRGGTSAFDRILGSRMCVEAVMALLE.GTPDTPACVVSLS.GRQAVRLPLMECVQVTKDV GETTSRGSRLNIIIVAEGAIDDKGKPITSESVKDLVVQRLGFDTAVTVLGHUQRGTSAFDRILGSRMCVEAVMALLE.ATPDTPACVVSLS.GRQAVRLPLMECVQVTKDV GETTSRGSRLNIIIVAEGAIDDKGKPITSSSVKDLVVQRLGFDTAVTVLGHUQRGTSAFDRILSSKMCHEAVMALLE.ATPDTPACVVSLS.GRQAVRLPLMECVQVTKDV GETTSRGSRLNIIVAEGAIDDKGKPISSSVKDLVVQRLGFDTAVTVLGHUQRGTSAFDRILSSKMCHEAVMALLE.ATPDTPACVVSLS.GRQSVRLPLMECVQVTKDV GETTSRGSRLNTIIVAEGAIDDGARVTSSSVKDLVVQRLGFDTAVTVLGHUQRGTSAFDRILSSKMCHEAVMALLE.ATPDTPACVVS.GRGSVRLPLMECVQVTKDV GETTSRGSRLNIVIIAEGAIDDGARVTSSSVKDLVVQRLGFDTAVTVLGHUQGGTSAFDRILSSKMCHEAVMALLE.ATPDTPACVVS.GRGSVRLPLMECVQVTKDV GHRSKGRRNNTIVAEGAIADDLTPISSDVKNVLVDRLGLGTATTTITGKUQRGGTAVATDRILATLQGLAVMAVLE.STPTPFSPLIGL.S.GRGSVRLPLMECVQVTKDV QRHRSKGRRNNTIVAEGAIADDLTPISSDVKNVLVDRLGLGTATTTITGKUQRGGTAVATDRILATLGQLAVAVLE.STPTSPLFSLIGL.S.N.N.NERCHENTTFPISSDSVVKLTKAV QRHRSKGRRNNGLKLEVTANDVKDALE .GGGIFDSRNVLGHUQGGSSPTPFDRNFATKMGAKANNMMAG.KIKESYRNGGIFANTPDSGCVLGKRKALVF KTTVKRGLVLNRECHENTTTPIYNJSS.GRGVFOCATNVLGHQQGGAPTFPDRNFGTKLGVKANLWSE.KLREVYRGRVFANPDSGACVIGLKKKALVF KTDIQRGLVLRNECHENTTFFLVNIJSS.GRGVFOCATNVLGHQQGGAPTPFDRNFGTKLGVKANLMSE.KLREVYRGRVFANAPDSACVIGLKKKALVF KTDIQRGLVLRNECHENTTFFLVNIJSS.GRGVFOCATNVLGHQQGGLPSPIDRTFRANJKAKANNMAG.KIKESYRNGGIFANPDSGACVIGLKKKALVF KTDIQRGLVLRNECHENTTFFLVNIJSS.GRGVFOCATNVLGHQQGGLPSPIDRNGTKLGVKANLMSE.KLREVYRGRVFANAPDSACVIGLKKGAVG KDRDERFDEALKLRGRSSFNNWEYY.KLLAH GFDTT TVLGHVQRGG PAFDR LA MG AV LE NV E RG I E A GFDTT TVLGHVQRGGS.FFFDRNJGKGSS.FFFDRNGKGLGS</pre>
E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-N Yeast B-C Human L-C Yeast A-C Consensus E. coli B. Stear. S. citri Human M-N Rabbit M-N Rabbit M-N Human L-N Human L-N Human L-N Human L-N Human M-C	207 206 207 248 248 252 431 439 617 619 616 620 805 811 305 303 303 359 359 363 359 363 359 363 542 542 721 721	KAGIAKGKKHAIVAITEHHCDV.DE.LAHFIEKETGRETRATVLGHIQRGGSPVPTDRILASRHGAYAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENH KRGHERGKKHAIVAITEHHCDV.DE.LAHFIEKETGRETRATVLGHIQRGGSPVPTDRILASRHGAYAIDLLLAGYGGRCVGIQNBCUVHHDIIDAIENH KRGHERGKKHSIIVVSENIYPDVK.LALKLYESKSGYITATVLGHVQRGGSPTAPDRUASRLGARAVELLLE.GCKGGRCGDQIIAPHTHEALSIPRSS SETTRGSRLMIIIVAEGAIDHGKPITSEDVILVVVRLGYDTRVTVLGHVQRGCTPSAFDRILGSRHGVEAVMALLEGTPDTPACVVSLSGRQAVRLPLMECVQVTKDV GETRSRGSRLMIIIIAEGAIDHGKPITSSEVKDLVVQRLGPTRVTVLGHVQRGCTPSAFDRILGSRHGVEAVMALLEGTPDTPACVVSLSGRQSVRLPLMECVQVTKDV GETRSRGSRLMIIIIAEGAIDHGKPISSSYVKDLVVQRLGPTRVTVLGHVQRGCTPSAFDRILGSRHGVEAVMALLE.ATPDTPACVVSLSGRQSVRLPLMECVQVTKDV GETRSRGSRLMIIIIAEGAIDHGKPISSSYVKDLVVQRLGPTRVTVLGHVQRGCTPSAFDRILSSKHGMEAVMALLE.ATPDTPACVVSLSGRQSVRLPLMECVQVTKDV GETRSRGSRLMIIIIAEGAIDHGKPISSSYVKDLVVQRLGPTRVTVLGHVQRGCTPSAFDRILSSKHGMEAVMALLE.ATPDTPACVVSLSGRQSVRLPLMECVQVTKDV GETRSRGSRLMIIIIAEGAIDHGKPISSSYVKDLVVQRLGDTRXTVLGHVQRGCTPSAFDRILSSKHGMEAVMALLE.ATPDTPACVVSLSGRQSVRLPLMECVQVTKVV GETRSRGSRLMIIIIAEGAIDHGKPISSSYVKDLVVQRLGDTRXTVLGHVQRGCTPSAFDRILSSKHGMEAVMALLE.ATPDTPACVVTLSGRQSVRLPLMECVQVTKVV GETRSRGSRLMIIIIAEGAIDHGKPISSSYKDLVVQRLGDTRXTVLGHVQRGCTAVAIDRILSKHGEAVMALLE.ATPDTPACVVTLSGRQSVRLPLMECVQVTKVV GETRSRGSRLMIIIIAEGAIDHGKPISSSYKDLVVQRLGDTRXTVLGHVQRGCTAVAIDRILSKKGEAVMALLE.ATPDTPACVVTLSGRQSVRLPLMECVQVTKKV QRHRSKGRNNTIVVREGAIADUTPTIMLYSE.ECKGIFDSRKNVLGHVQRGGSPTPFDRNFTKKGAKANNMAG.KIKESYKNRGTIPAMTPDSCCVLGHKRALVF KTTVKRGLVLRNEKCHENYTTEIYLIXISS.ECKGFUPCRTNVLGHLQQGAPTPFDRNYGKKGAXANNMAG.KIKESYKNRGTFAMTPDSCCVLGHKRALVF KTDIQRGLVLRNEKCHENYTTEIYLIXISS.ECKGFUPCRTNVLGHLQQGAPTPFDRNYGKKGAXANNMAG.KIKESYKNRGTFAMAPDSACVIGLKKKAVAF KTDIQRGLVLRNEKCHENYTTEIYLIXISS.ECKGFUPCRTNVLGHLQGGAPTPFDRNYGKGAXANNMAG.KIKESYKNRGTFAMAPDSACVIGKKKAVYAF KTDIQRGLVLRNEKCHENYTTEIYLIXISS.ECKGFUPCRTNVLGHLQGGAPTPFDRNYGKGAXANNMAG.KIKESYKNRGTFAMAPDSACVIGKKKAVYAF RKDRGRHNGKLLVRAEQASSVYSTQLLADIISE.ASKGRFUPCRATAIGGYGGSKGYKASKATASRAVGKIKFFE RKDGRGGRGKLLVRREQASSVYSTQLLADISE.SEK
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E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-N Yeast B-C Human L-C Yeast A-C Consensus E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Human L-N Human L-N Human L-N Human M-C Rabbit M-C Human L-C Human L-C Human L-C Human L-C Human L-C	207 206 207 248 248 252 431 439 617 619 616 620 805 811 305 303 303 359 359 359 363 359 363 254 721 721 720	<pre>KAGIAKGKKHAIVAITEHKCDV.DE.LAHPIEKETGRETRATVLGHIQRGGSVPYDRILASRGAVAIDLLLA.GYGGRCVGIQNEQLVHHDIIDAIENK KRGHERGKKHSIIVAEGVGSG.VD.FGRQIQEATGFETRATVLGHIQRGCSPYPTDRILASRGATATDLLLA.GYGGRCVGIQNQULVHHDIIDAIENK KRGHERGKKHSIIVAEGVGSG.VD.FGRQIQEATGFETRATVLGHIQRGCTPATADDRYRAFQANGPAUGDIA.GVGGLGAGGAG SETTIGSRLAIIVAEGAIDRWGKPITSEDIKKLVVKELGYDTRVTVLGHVQRGCTPSAFDRILGSRWGVEAVMALLE.GYEDTPACVYSLSGNQAVELPLHECVQVTKDV SETTIGSRLAIIVAEGAIDRWGKPITSEDIKKLVVKELGYDTRVTVLGHVQRGCTPSAFDRILGSRWGVEAVMALLE.GTPDTPACVYSLSGNQAVELPLHECVQVTKDV GETRSRGSRLMIIIAEGAIDRHGKPISSSYVKDLVVQRLGPDTRVTVLGHVQRGCTPSAFDRILSSRWGHEAVMALLE.ATPDTPACVYSLSGNQAVELPLHECVQVTKDV GETRSRGSRLMIIIAEGAIDRHGKPISSSYVKDLVVQRLGPDTRVTVLGHVQRGCTPSAFDRILSSRWGHEAVMALLE.ATPDTPACVYSLSGNQAVELPLHECVQVTKDV GETRSRGSRLMIIVAEGAIDRHGKPISSSYVKDLVVQRLGPDTRVTVLGHVQRGCTPSAFDRILSSRWGHEAVMALLE.ATPDTPACVYSLSGNQAVELPLHECVQVTKEV SKHRSRGRANTIVVAEGAIADLTFISSDVIKUVDRLGDTRTTIGHVQRGTAVAIDRILATLGVQNAVAXUE.STPDTPSLLA.VN.ENNIVREHEEVQUTKEV SKHRSRGRNNTIVVAEGAIADLTFISSDVIKUVDRLGDTRTTUGHVQRGTAVAIDRILATLGVQNAVAXUE.TPTPTPSLLG.IL.ENKIINPLVESVKLTKN VKTVKRGLVLREKCNENTTOFINLYSS.EGGGFDSGRNVLGHNQGGGSTPFDNRFATKKGAANNMHGG.KIKESYRGRI.PA.NTPDSGCVGARKRALVF KTTVKRGLVLREKCNENTTOFINLYSS.EGGGFDSGRNVLGHNQGGGSTPFDNRFATKKGAANNMHGG.KIKESYRGRI.PA.NTPDSGCVGARKRALVF KTDIQRGLVLREKCHENTTEFINLYSS.EGGGVPDCTNVGHLGQGGGFTPFDNRYGTKLGVAALWSE.KLBUYRRGVV.PA.NAPDSACVIGLAKKAVAF KTDIQRGLVLREKCHENTTEFINLSS.EGGGVPDCTNVGHLGQGGGATPFDNRYGTKLGVAALWSE.KLBUYRRGVV.PA.NAPDSACVIGLAKKAVAF KTDIQRGLVLREKCHENTTEFINLSS.EGGGVPDCTNVGHLGVQGGVSSKDRVTASRFAVCIKFIEQWNKKREASPNTDAKVLREKDTHGEKVPTVEHEDDSAVIC KNRDGRANGKLLVREGASSVSTGLLADIISE.ASKGKRGRTAFIGHVQGGVSSKDRVTASRFAVCIKFIEQWNKKREASPNTDAKVLREKTGHKKVVGKKSKY RHDKGENNRGKLLVRHEGASSVSTGLLADIISE.ASKGKFGKGRTAFIGHVGGGVPSSKDRVTASRFAVCIKFIEQWNKKREASPNTDAKVLREKTTHVEHEDDSAVIC RAGGRAFTDEAULGARSFNNNHEYY.KLLAH GKANDERFDEAULGARSFNNNHEYY.KLLAH GKANDERFDEAULGARSFNNNHEYY.KLLAH GKANDERFDEAULGARSFNNNHEYY.KLLAH GKANDERFDEAULGARSFNNNHEYY.KLLAH GKANDERFDEAULGARSFNNN</pre>
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E. coli B. Stear. S. citri Human M-N Rabbit M-N Human L-N Yeast B-N Yeast B-N Yeast B-N Yeast B-C Human L-C Yeast A-C Consensus E. coli B. Stear. S. citri Human M-N Rabbit M-N Human M-N Human M-C Rabbit M-C Ra	207 206 207 248 248 252 439 617 619 616 620 805 811 305 303 309 359 359 359 359 359 359 359 359 359 35	KAGIAKGKKHAIVAITBEHNCOV.DELAHFIEKETGRETRATVIGHIQAGGSPVPYDRILASRHGAYAIDLLLAGYGGRCVGIQNEQLVHHDIIDAIENM KROHERGKKHSIIVAEQVGSG.VDFGRQIQEATGFETRATVIGHUQAGGSPTAPDRULASRHGAYAIDLLLAGYGGRCVGIQNNQLVDHDIAEALAN ANLHQQKRKSVIVVSENITPDVH.LALALVESKSGVITATVIGHUQAGGSPTAPDRULASRHGAYANDLLE.GTPOTACVUSLSGNQAVELPLAECVQVTKOV SETRITGSRLNIIVAEGAIDANGKPITSEDIKNLVVKALGVDTRVTVLGHUQAGGTPSAPDRILOSRHGVAVALLE.GTPOTACVUSLSGNQAVELPLAECVQVTKOV GETRSRGSRLNIIIAEGAIDANGKPITSEDIKNLVVKALGVDTRVTVLGHUQAGGTPSAPDRILOSRHGVAVALLE.ATPOTPACVUSLSGNQAVELPLAECVQVTKOV GETRSRGSRLNIIIAEGAIDANGKPISSGVKOLVVQALGPDTRVTVLGHUQAGGTPSAPDRILOSRHGVAVALLE.ATPOTPACVUSLSGNQAVELPLAECVQVTKOV GETRSRGSRLNIIIAEGAIDANGKPISSGVKOLVVQALGPDTRVTVLGHUQAGGTPSAPDRILSSKHGHEAVNALLE.ATPOTPACVUSLSGNQSVELPLAECVQVTKOV SKHRSRGKTTIVVAEGAIDANGKPISSSVKOLVVQALGPDTRVTVLGHUQAGGTAVADRLAILGGLEAVNALLE.STPOTPSPLIAVNENKIVKRIJLESVCUTKAV GETRSRGSRLNIIIAEGAIDANGKPISSSVKOLVVQALGPDTRVTVLGHUQAGGTAVADRLAILGGLEAVNALLE.STPOTPSPLIAVNENKIVKRIJLESVCUTKAV SKHRSRGKTTIVVAEGAIDANGKPISSSVKOLVVGLGPDTRVTVLGHUQAGGSPTPFDNPATKIGAKANNAHGA.KKESVNAGTIANTPDSGCVGANKKUL SKHRSRGKTTIVVAEGAIDANGKPISSSVKOLVVGLGPTAVGULGAVGAVADULASSKKAVANALS.SKKKEVANALLE.STPOTPSPLIAVNENKIVKRVKTKSV KTTVKRGLVLANEKCHENYTTDFIFHLSS.EGKOVPCATNVLGHUQAGGSPTPFDNNPATKIGAKANNAMAG.KKIKSSVNAGTIANTPDSGCVGANKALVS KTTYKRGLVLANEKCHENYTTFIFHLSS.EGKOVPCATNVLGHUQGGAPTPFDNNYATKIGAKANNAHGA.KKESSNNGTIANTPDSGCVGANKRALVF KTDIQRGLVLANEKCHENYTTFIFHLSS.EGKOVPCATNVLGHUQGGAPTPFDNNYATKIGAKANNAMAG.KKIKSSTNGRTANPDSACVIGLKKAVAF KTDIQRGLVLANEKCHENYTTEJINISS.EGKOVPCATNVLGHUQGGAPTPFDNNYATKIGAKANNAHGA.KKESSNNGTANAPDSACVIGLKKAVAF KTDIQRGA.LLSSTNASKALSATKLAEVITA.EADGAPDAKPAYGCHUQGGGSPSDRATASHATKAVGFIEDNQAAIAERAAEENFNADDKTSDTAAVGVKGSNVY RHOKGENNGKLUKAREGASSVTSQLADISSSKOKGVGATHVEKSSKKGVGGAFANTARAIKASGKIKKESSKOKTAFTARAIKAVGFIED.NQAAIAERAAEENFNADDKTSDTAAVGVKGSNVY RHOKGENNGKLUKAREGASSVTSQLADISSSKOKGVGATHVEKSK KANDDEKKFPDEAKLARGSPHINMEYY.KLLAH GKANDEKFPDEAKLGRGSPHINMIYI.KLAHAGASSKEK GKANDGKFPDEAKG

FIG. 7. Multiple alignment of the PfkA family of ATP-dependent Pfks of bacteria, yeasts, and mammals. The species are as indicated. M and L, muscle and liver enzymes of mammals, respectively; B and A,  $\beta$ - and  $\alpha$ -subunit of the yeast enzyme, respectively; N and C, N-terminal and the C-terminal domains, respectively, of the eukaryotic protein subunits. An exclamation mark above and below the aligned sequences indicates a residue conserved in all sequences. An asterisk above the aligned sequences indicates a residue conserved in the bacterial enzymes and the eukaryotic N-terminal segments. An asterisk below the aligned sequences indicates a residue conserved among the eukaryotic C-terminal segments. The consensus sequence (Consensus) is provided below the aligned sequences. The statistical analyses of the alignments are summarized in Table 4. The Fasta (44) and Bestfit (39) programs were used to give the optimal alignment.

which could be released from the membrane as a highmolecular-weight soluble protein, the other which was an integral constituent of the membrane. The biochemistry of the former protein has been examined (35, 36). As shown in our previous reports (60, 61), these two proteins are a multiphosphoryl transfer protein and a fructose-specific enzyme II of the PTS. They act in concert to produce cytoplasmic fructose-1-P from extracellular fructose. This sugar phosphate ester is then further phosphorylated in the reaction catalyzed by FruK to give the common intermediate of glycolysis, fructose-1,6-bisphosphate. In species of *Rhodo*- spirillum, Rhodobacter, Thiocapsa, Thiocystis, Pseudomonas, Alcaligenes, and Fusobacterium, all of which possess a fructose-specific PTS (15), fructose is probably the only sugar metabolized via the PTS and glycolysis, because 6-Pfk is lacking.

Biochemical experiments have shown that in *R. rubrum*, *R. spheroides*, and *R. capsulatus*, a multiphosphoryl transfer protein, a fructose-specific enzyme II, and FruK are coordinately induced 3-, 10-, and >100-fold, respectively, by inclusion of fructose in the growth medium (13, 49, 54). These observations led to the prediction that the three

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	-N B-C	CS %I C	18)         19         27         (184)         1           08)         33         29         (172)         1           02)         27         26         (178)         2	72) 82 29 (171) 2 10) 19 35 (364) 8	72) 87 28 (171) 2 10) 20 35 (364) 5	84) 87 22 (266) 2 03) 19 38 (366) 7	75) 107 26 (184) 2 03) 21 38 (368) 6	75) 132 26 (179) 2 4) 10 48 (368) 14 22 (256) 1	in SD calculated by
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		SS %	26 25 () 31 29 ()	25 25 ( 18 37 (	20 24 () 19 36 ()	39 30 () 26 38 ()	54 30 () 14 39 ()	24 ()	
	A-N	) I%	11 (236) 13 (202) 15 (233)	3 (371) 1 37 (108)	54 (371) 1 55 (108)	52 (371) 1 11 (101)	52 (374) 1 11 (101)		•
		S	31 54 33	21 <u>5</u> 133 3	15 156	11 5	19		
an	L L	Ι%	30 (274) 31 (281) 33 (292)	43 (72) 66 (353)	43 (72) 65 (355)	32 (124) 94 (357)	23 (177)		•
Hum		S	32 82	156 19	157 26	227 13			:
	L-N	1%	31 (208) 40 (172) 43 (196)	74 (376) 32 (178)	75 (376) 32 (178)	92 (377) 24 (177)			
	с U	CS	(1) 31 (1) 31 (2) 27	1 17 2) 204	) 11 5) 110	3) 11			
use	1	I%	30 (27/ 32 (28] 33 (285	42 (72) 66 (352	42 (72) 65 (355	32 (12:			
Moi		ទ	32 23	290 26	131 30				
	L L	I%	33 (226) 37 (197) 44 (203)	77 (376) 26 (244)	77 (376) 25 (244)				
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Hun	7	S	() 19 32						
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aro-	-ST	S	9) 89 2) 75						
B. ster	phila	1%	46 (319 57 (302						
19	10	S	4) 57						
u	4	I%	48 (31						
	Enzyme		S. citri E. coli B. stearother- mophilus	Human M-N M-C	M-N M-C			east A-N B-N B-N	

 TABLE 5. Size, subunit, and domain compositions of Pfks and selected related enzymes<sup>a</sup>

Enzyme	Approximate no. of aminoacyl residues/chain	No. of polypeptide chains/ enzyme	No. of identified homologous domains/ enzyme
R. capsulatus, FruK	316	1	1
E. coli PfkB	308	2	2
E. coli PfkA	320	4	4
B. stearothermophilus	319	4	4
Pfk			
S. citri Pfk	327	4	4
Mammalian Pfks	780 (N and C)	4	8
Yeast Pfk, a	987 (N and C)	41	
	. ,	8	16
Yeast Pfk, β	959 (N and C)	4	
Potato $P_2$ Pfk, $\alpha$	616	$2_{4}$	4
Potato $P_2$ Pfk, $\beta$	513	2	•

<sup>a</sup> Mammalian Pfk, the four mammalian Pfks for which sequence data are available (human muscle, human liver, rabbit muscle, and mouse liver); Yeast Pfk, Saccharomyces cerevisiae Pfk; Potato P<sub>2</sub> Pfk, potato PP<sub>1</sub>-dependent Pfk;  $\alpha$  and  $\beta$ ,  $\alpha$ - and  $\beta$ -subunits, respectively, of the yeast and potato enzymes; N and C, N-terminal and C-terminal homologous domains, respectively, of the mammalian enzymes and of the two subunits of the yeast enzyme.

fructose enzymes are encoded within a single operon in each of these organisms, a conclusion established for *R. capsulatus* by the sequence analyses reported here. The *fru* operon of *R. capsulatus* may possess a compact structure with a 4-nucleotide overlap between the *fruB(HI)* and *fruK* genes and an 18-nucleotide intercistronic region between *fruK* and *fruA*. These structural features resemble those in the *fru* operons of *E. coli* in which a 1-nucleotide overlap characterizes the chain terminating codon of *fruB(MH)* and the initiation codon of *fruK* and a 16-nucleotide intercistronic region separates *fruK* from *fruA* (43).

Determination of the *R. capsulatus fruK* nucleotide sequence, deduction of the aminoacyl sequence of FruK, and sequence comparisons with proteins in the data base revealed that FruK is homologous to several bacterial kinases, including PfkB of *E. coli*. A phylogenetic tree relating these proteins is shown in Fig. 3. Surprisingly, we also found that an extended segment of the large subunit of Rubisco exhibited a significant degree of sequence similarity (27.1% identity over a 122-residue stretch with four gaps and a comparison score of 8.5 SD) with FruK of *R. capsulatus* (Fig. 5; Table 3). Since this region encompasses the sugar bisphosphate-binding site of Rubisco, it is possible, although unlikely, that this sequence similarity arose by convergent evolution.

The sequence similarities between FruK of *R. capsulatus* and the homologous kinases from other bacteria were not reflected in the amino acid compositions of these proteins. The *R. capsulatus* protein showed a markedly different composition, with the predominant amino acids (Table 2, shown above the spaces; particularly A, L, and G) showing increased occurrence in the *R. capsulatus* FruK relative to the other proteins and the minor amino acids (Table 2, below the spaces; particularly E, S, N, Q, K, C, and Y) showing decreased occurrence in *R. capsulatus* FruK relative to that of the other kinases. Surprisingly, none of the C, S, Y, W, or H residues were fully conserved among the homologous proteins, suggesting that none of these residues play an essential catalytic role in the mechanism of action of these



FIG. 8. Phylogenetic tree of homologous members of the PfkA family. Relative evolutionary distances are provided adjacent to the branches. Abbreviations are the same as indicated in the legend to Fig. 7, except that the PP<sub>i</sub>-dependent Pfk subunits  $\alpha$  and  $\beta$  of the potato are designated Potato, P<sub>2</sub>-A, and P<sub>2</sub>-B, respectively. The tree was constructed as described by Doolittle and Feng (17, 20).

functionally related enzymes. In fact, FruK was not sensitive to inhibition by the cysteyl-specific reagent, *N*-ethylmaleimide, and was inhibited only at high concentrations of the histidyl-specific reagent, DEPC, and the activated serine reagent, PMSF. Since the monomeric *Rhodobacter* protein (60a) contains just one W and one Y, this protein may be good material for fluorescence studies.

Secondary structural predictions (Fig. 4) led to the suggestion that members of the PfkB family of homologous proteins are all structurally similar. Predicted secondary structural elements (coil,  $\beta$ -turns,  $\beta$ -strands, and  $\alpha$ -helix) as well as hydropathy and amphiphilicity analyses were nearly superimposable for FruK of *R. capsulatus* and PfkB of *E. coli* despite their considerable compositional differences. Both proteins appear to consist of alternate  $\beta$ - and  $\alpha$ -structures, as has been demonstrated for PfkA (25). On the basis of their primary structural similarities, the tertiary structures of the homologous PfkB family proteins are probably nearly superimposable (16, 52, 53).

It is interesting to note that PfkA, the predominant Pfk in  $E.\ coli$ , is a nondissociable tetramer, whereas PfkB is a dimer which can associate to a less-active tetramer (see reference 51 for a review). Their subunits are of about the same size, and their genes are located at opposite positions on the circular  $E.\ coli$  chromosome (at 87 and 38 min, respectively). Despite these facts, PfkA does not exhibit significant sequence identity with PfkB, but PfkA is homologous to sequenced Pfks from other bacteria and eukaryotes (8, 25, 45). It has been proposed that the primordial bacterial chromosome was once one-half of its present size, and a chromosomal duplication event occurred, giving rise to a genome with genes of similar structure and function on

opposite sides of the chromosome (47). If this hypothesis is correct, then this chromosomal duplication must have occurred before the divergence of prokaryotes from eukaryotes, and the subunits of the currently recognized eukaryotic Pfks (which presumably arose by intragenic duplication) must have been derived from the precursor of the presentday prokaryotic PfkA. This hypothesis presupposes also that divergence within the PfkB family occurred after this duplication. It is interesting to note that PfkA and PfkB of E. coli differ not only with respect to their sequences and subunit association properties but also with respect to their regulatory properties. Moreover, while the synthesis of PfkA is induced in the presence of glucose, that of PfkB has been reported to be constitutive (11). Thus, PfkA may function primarily in anaerobic glycolysis, whereas PfkB may function under conditions of aerobic metabolism. In view of the structural similarity of PfkB and FruK (Fig. 4), it is interesting to note that the pfkB gene in E. coli and the fru operon in S. typhimurium share the same unusual putative -10 region (CAGACT) and have similar -35 regions (40-42a). The functional and evolutionary significance of these observations has yet to be established.

The PfkA family is large and diverse, with representatives from gram-positive and gram-negative prokaryotes as well as lower and higher eukaryotes. The Pfk subunits of eukaryotes are minimally twice the size of the prokaryotic enzymes, and as documented here, they may have arisen by intragenic duplication. The N-terminal domains of these eukaryotic enzymes exhibit a higher degree of similarity to the prokaryotic enzymes than do the homologous C-terminal domains. It is therefore reasonable to propose that the N-terminal domains retained their original, primary function, namely, catalysis, while the C-terminal domains evolved more rapidly, giving rise to allosteric regulatory domains. In this regard it is interesting to note that substrates and products of the Pfk-catalyzed reactions also serve as allosteric effectors. Thus, the substrate-binding residues have been largely retained in the N- and C-terminal domains of the eukaryotic enzymes (26, 46a). The eukaryotic enzymes have also acquired sensitivity to regulation by other metabolites, such as Krebs cycle intermediates (51). This fact is consistent with their increased degree of structural complexity relative to that of the prokaryotic enzymes. The property of cooperativity, characteristic of all members of the PfkA family, is consistent with their tetrameric structures, and these structures differ from those of the PfkB family members which do not exhibit the property of cooperativity and which can be monomeric or dimeric.

Although PfkA and PfkB of E. coli were not found to exhibit a statistically significant degree of sequence identity, we nevertheless propose that these proteins, and therefore all members of the PfkA and PfkB families, shared a common origin. The evidence is as follows. (i) PfkA and PfkB catalyze the same reaction, and all members of both families catalyze essentially the same reaction with various carbohydrate substrates. (ii) PfkA and PfkB are essentially the same size, a size that is common to all PfkB family members, prokaryotic PfkA family members, and homologous domains of eukaryotic Pfks (Table 2) (46a). (iii) The predicted secondary structures of PfkB and FruK (Fig. 4) correspond to the known secondary structures of PfkA and of the B. stearothermophilus Pfk (19, 26), and the degrees of sequence similarity within either the PfkA or the PfkB family imply conservation of secondary and tertiary structural features (16). (iv) FruK of R. capsulatus, a member of the PfkB family, exhibited regions of sequence similarity both with the PP<sub>i</sub>-dependent Pfk of potatoes (8; unpublished data), which is clearly homologous to members of the PfkA family, and with the yeast Pfk (see Results). While the extent of similarity was insufficient to establish homology, the results are suggestive of a common origin. (v) As noted above, the pfkA and pfkB genes map to opposite sides of the E. coli chromosome, which is consistent with an origin resulting from a chromosomal duplication event (47). These observations, while compelling, must nevertheless be considered preliminary. Substantiation for the proposed common origin for the PfkA and PfkB families may be forthcoming when sequences of additional homologous proteins or the X-ray crystallographic structure of a member of the PfkB family becomes available.

### ACKNOWLEDGMENTS

We thank Michael C. O'Neill for conducting computer analyses of the promoters of the *fru* operons of both R. *capsulatus* and S. *typhimurium*, Michael Baker and DaFei Feng for valuable suggestions concerning the computer analyses, and Mary Beth Hiller for assistance in preparation of the manuscript.

This work was supported by Public Health Service grants 5RO1AI21702 and 2RO1AI14176 from the National Institute of Allergy and Infectious Diseases.

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