

Crystal structures of eukaryote glycosyltransferases reveal biologically relevant enzyme homooligomers

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ABSTRACT

Glycosyltransferases (GTases) transfer sugar moieties to proteins, lipids or existing glycan or polysaccharide molecules. GTases form an important group of enzymes in the Golgi, where the synthesis and modification of glycoproteins and glycolipids take place. Golgi GTases are almost invariably type II integral membrane proteins with the C-terminal globular catalytic domain residing in the Golgi lumen. The enzymes themselves are divided into 103 families based on their sequence homology. There is an abundance of published crystal structures of GTase catalytic domains deposited in the Protein Data Bank (PDB). All of these represent either of the two main characteristic structural folds, GT-A or GT-B, or present a variation thereof. Since GTases can function as homomeric or heteromeric complexes *in vivo*, we have summarized structural features of the dimerization interfaces in crystal structures of GTases, as well as considered the biochemical data available for these enzymes. For this review, we have considered all 898 GTase crystal structures in the Protein Data Bank and highlight the dimer formation characteristics of various GTases based on 24 selected structures.

Keywords:

Protein Structure; Dimerization; biologically relevant dimer; crystallographic dimer; protein-protein interfaces; GTase fold

Eukaryotic cells are coated with glycans of variable composition and structure. These glycans are covalently attached to membrane proteins and lipids as a result of glycosylation, and form the basis of various cellular recognition events needed for cell-cell contacts or in differentiating between the own and the foreign by the immune system. Glycosylation, therefore, must be a very precise process, and improper glycosylation is in many cases manifested in diseases due to impaired cellular recognition. Such diseases include congenital disorders of glycosylation, inflammation, diabetes and cancers (for recent reviews, see Hennet & Cabalzar (2015) [1], Chang & Yang (2016) [2] and Vajaria *et al.* (2016) [3]).

Glycan synthesis takes place in the endoplasmic reticulum and the Golgi apparatus and involves a complex interplay between a number of carbohydrate-acting enzymes, donor and acceptor substrates, nucleotide-activated sugars and their transporters. Therefore, and to ensure fidelity in glycan synthesis, there is a specific requirement for the presence of distinct sets of glycosidases and glycosyltransferases (GTases) in the cell. The latter form a huge ensemble of enzymes currently divided into 103 sequence-based families, according to the CAZy database [4] (www.cazy.org). They catalyse the addition of specific sugar moieties in specific sequence and chemical configuration (i.e. the linkages between sugar units and the stereochemistry of the product and the substrate - inverting or retaining) to specific acceptor molecules, which can be carbohydrates, proteins or lipids. Given the huge variety of glycan structures needed for normal cellular recognition events, it is therefore not a surprise that the total amount of different GTases in the CAZy database approaches 250.

Glycosyltransferases - the topic of this review - are almost invariably type II integral membrane proteins with a short cytoplasmic tail, a single transmembrane domain, a stem region and a globular catalytic domain located in the Golgi lumen. Due to the difficulties in both producing and crystallizing full-length type II membrane proteins, all the crystal structures of GTases thus far solved represent their soluble, globular catalytic domains.

Glycosyltransferases form homomers

GTases have been shown to form enzyme dimers, tetramers and oligomers in live cells mainly via interactions between their catalytic domains [5-7], and it has been suggested that ordered protein arrays in the trans-Golgi might contain GTases [8]. Considering that these enzymes do not use a template, a question of considerable interest is whether enzyme complex formation is part of the cellular mechanism to ensure the fidelity of glycan synthesis.

How to analyze dimerization?

For homomeric complexes, it has been shown that dimerization is the most common transition occurring during the assembly of protein complexes [9], cyclization being the next most common, while fractional transitions are the rarest. We therefore focused on dimerization interfaces, acknowledging that even if the GTases may form higher order oligomers, dimerization would still be a biologically relevant step in the homomer formation. Even with the abundance of structural information, analysis of protein dimerization (or formation of higher-order oligomers) with the help of crystal structures is not straightforward. Protein crystals may contain more than one protein molecule in the asymmetric unit (the smallest repetitive unit of the crystal). In such cases, these two or more molecules are typically symmetrically arranged. This so called non-crystallographic symmetry is a feature separate of the crystallographic symmetry and would not necessarily exist, if the interaction observed in the crystallized species was not due to functional reason. Instead, the crystal unit cell and the crystal symmetry would then simply form differently. Crystal formation necessarily involves molecular contacts; therefore, the problem is to separate functionally relevant, or “physiological”, protein-protein contacts from interactions that merely bring about and maintain the crystal packing. Consequently, other data including biochemical characterization of the complexes by using e.g. gel filtration, analytical ultracentrifugation or dynamic light scattering must be taken into account.

In favourable cases there is a well justified logical reason for the protein to form dimers, for example in the case when a ligand binding site is formed from residues located in different monomers, or when a prediction of protein-protein interactions on the basis of analysing interaction site properties can be made with high confidence. The latter approach is a very active field of research, and a great many server-based analysis tools are now freely available [10, 11]. For this review we have reanalysed all the 898 GTase crystal structures in the Protein Data Bank (PDB, www.rcsb.org) [12] using the above criteria, and present our view on various GTase dimers that are likely to also form functionally relevant complexes *in vivo*.

Selection of GTase structures to study and their structural characteristics

At the time we started this work, the contents of the CAZy data base and the PDB included a total of 898 crystal structures of GTases. After thorough analysis of all GTase families, we chose structures of 172 unique proteins such that 44 of the 103 GTase families were represented by at least one crystal structure. 61% of all GTase crystal structures are eukaryotic, of which 40% represent human proteins. A fair number of these structures are complexes with donor nucleotide-activated sugars and/or acceptor glycans, or molecules representing only parts of them.

Based on literature a major motivation to obtain high quality GTase structures seems to be to get atomic resolution details of the catalytic mechanisms and ligand binding modes in order to use this data for drug design. GTase structures from a wide range of species are often usable for functional analysis due to the structural conservation between enzymes across species. Each coordinate entry of the PDB is filed as a separate structure, although many of the entries are redundant. This is due to structure-function studies requiring structures of proteins in several different states, including apo- and multiple holo structures with different ligands bound. An additional reason for structural redundancy is that most GTases fall into two similar fold types: GT-A and GT-B, and variants thereof, with only a limited degree of structural difference. The structural conservation is not reflected in the sequence similarity: the average sequence identity was found to be only 12% and 11% for GT-A and GT-B folds, respectively, in a set of 67 non-redundant GTase structures representing 28 families [13]. A small portion of the GTases possess neither the GT-A nor the GT-B fold, but display slightly

different topological properties [14]. GTases within a given family usually share the same fold type [15].

GT-A and GT-B folds have similar spatial arrangements consisting of α/β alternations, with variable N- and C-termini. Although the size of the α and β parts vary, the overall structure is always held together by a continuous central twisted β -sheet called the Rossmann fold, which is flanked by α -helices on both sides [16]. The GT-A fold contains one six-stranded β -sheet showing a 321465 topology, in which $\beta 6$ is antiparallel to the other strands (Fig. 1; Fig. 2A). Insertions breaking the α/β alternation are often found between $\beta 5$ and $\beta 6$, and more rarely between other strands. A smaller antiparallel two-stranded β -sheet that consists of $\beta 4'$ (a short strand flanking $\beta 4$) and βC (a short strand in the variable C-terminus), is usually present in eukaryotic GT-A folds (Fig. 2A). This two-stranded β -sheet is sometimes accompanied by parallel or antiparallel short β strands from the variable C-terminal part. Other common features of the GT-A fold are the Asp-X-Asp (also known as DxD) motif, and a divalent cation binding motif, usually flanking $\beta 4$ [15, 17-19], that is needed for activity. Some GTases may occasionally lack these features and still be considered as part of the GT-A fold family.

The GT-B fold consists of two separate Rossmann fold motifs, each of them consisting of a six-stranded parallel β -sheet with a 321456 topology, and connected by a linker region [20] (Fig. 1; Fig. 2A). The two domains face each other, with the active site located within the resulting cleft. Some variant GTases possess a fold closely resembling the canonical GT-A or GT-B topology, but with a different order of β -strands. These variants have sometimes been regarded as new fold types, increasing the confusion in the classification. The classification we describe above is based on a common structural core shared within the dataset of the GTase structures used in this study.

The GTase structures in the CAZy database were imported, family by family, into Excel for analysis. Out of 898 crystal structures, 338 contained more than a single protein molecule in the asymmetric unit, and were selected for further investigation. These 338 structures were then sorted by kingdom, species, and unique protein name. Of these, 164 were from eukaryote species, among which 82 were of human origin, representing 15 different GTases. We then set out to analyze all these human

GTases in detail, including also homologues from other species when appropriate. The PDB codes of the 164 selected eukaryote GTase structures as well as the associated PDB files were gathered using a custom python script. In the case where more than one structure was available for a given protein, structural alignments were made in order to choose the most representative one, typically the example with the highest resolution. We did not discriminate between apo- and holoenzymes, since the local conformational changes brought about by substrate binding generally did not affect overall fold or dimerization properties.

Our final selection contains 24 structures from 18 different GTases, representing both the main GT-A and GT-B folds and their variants (Fig. 1, Table 1). Each structure was evaluated for the likelihood of a physiological dimer being present in the asymmetric unit of the crystals using various criteria/tools (Table 1). The nature of the interface and thermodynamic properties were assessed employing the jsPISA macromolecular surface and interface calculation tool [21], Voronoi tessellation, i.e. the DiMoVO server [22], and the EPPIC [23] server. Evolutionary conservation of the interface was assessed using the InterEvol [24] server.

In the following paragraphs, we will first review various GTase dimers as they are described in the literature, and also refer to the existing biochemical evidence of their dimerization, if such data is available. We then summarize, with help of bioinformatic tools, their likelihood of representing physiologically relevant enzyme dimers.

GT-A folds

β-Glucuronyltransferases (PDB codes 3CU0, 1V84, 2D0J)

β -Glucuronyltransferases (EC 2.4.1.135) belong to family 43 inverting GTases, which use UDP-glucuronate as the donor substrate. They add the glucuronic acid moiety to an existing galactosyl-galactosyl-xylosyl- or galactosyl-xylosylprotein acceptor depending on the specific enzyme. Crystal

structures have been solved for three of the human enzymes: glucuronyltransferase-I (GlcAT-I; PDB 3CU0) [25], glucuronyltransferase-P (GlcAT-P; PDB 1V84) [26], and glucuronyltransferase-S (GlcAT-S; PDB 2D0J) [27].

The GlcAT-I structure appears as a functional dimer (Fig. 1). Both monomers are required for binding to the acceptor molecule. More specifically, the oxygen and nitrogen atoms of the side chain of residue Gln318 of one monomer are at a hydrogen bonding distance from the O-6 atom of the Gal-1 moiety of the acceptor bound to the active site of the other monomer [28]. Furthermore, if the O-6 position is sulphated, the NE2 atom of Gln318 from the other monomer undergoes a conformational change and positions itself at a 3.0 Å distance from the O-4 oxygen atom of the sulphate [25]. Enzyme kinetic studies provide additional evidence in favour of a functionally relevant GlcAT-I dimer: a sulphated or a phosphorylated acceptor enhances GlcAT activity, but only if the enzyme is dimeric [25].

GlcAT-P structure [26] is highly similar with GlcAT-I. This holds true also for the dimer interface area. For example, the last β -strand, containing the Gln318 residue, extends to the active site of the other monomer, exactly as in GlcAT-I. GlcAT-P has also been shown to exist as a dimer by gel filtration under non-denaturing conditions [29], as well as by analytical ultracentrifugation, even when the N-terminal part containing the transmembrane domain is deleted [30].

GlcAT-S structure [27] was solved by using the GlcAT-P structure as the search model in molecular replacement, and the same conclusions regarding GlcAT-S dimerization could be drawn.

Glycogenins (PDB codes 1LL0, 3U2U, 4UEG)

Glycogenins (GTase family 8; EC 2.4.1.186) are autocatalytic proteins serving not only as the core of the glycogen structure, but also as enzymes catalyzing the addition of the first UDP-glucose

molecules in the initial phase of glycogen synthesis. In the catalysis, the stereochemistry of the added glucose is retained as α .

Several crystal structures of glycogenins have been solved: glycogenin-1 from rabbit (rGYG1; PDB 1LL0) [32] and human (PDB 3U2U) [33], as well as human glycogenin-2 (PDB 4UEG) [34, 35] serve as representative examples.

Rabbit glycogenin (rGYG1) was crystallized in two crystal forms - one containing 10 molecules (five dimers) per asymmetric unit, while the other holding only one molecule per asymmetric unit. In the former crystal form (tetragonal), the monomers of the dimers are related to each other by a non-crystallographic 2-fold axis creating identical dimers compared to the crystallographic dimers of the latter crystal form (orthorhombic) [32]. The decameric variant of rGYG1 is likely to be an artefact of concentrating the protein for crystallization for three reasons: (i) the purified rGYG1 was suggested to be a dimer by density gradient centrifugation [31]; (ii) the active sites of glycogenin monomers in the complex would in this form be placed unfavourably with regard to the glycogen biosynthesis by the glycogen synthase; (iii) the interface areas between the dimers (that form the decamer) cover only 7% of the total surface area. Thus, the decamer likely connects dimers to support crystal packing. In the orthorhombic crystal form of rGYG1, 20% of the total surface area is involved in dimer contacts, likely representing a physiologically relevant dimer as this value is typical for proteins that possess high affinity binding with each other [36].

The ensemble of rGYG1 structures [33] with different intermediates of glycogen synthesis has revealed a “lid” domain, which guides the substrates in the narrow dimer interface. The substrates are then subjected to either intra- or intersubunit catalysis, depending on the chain length of the nascent glycan chain and steric factors in the channel. The term “intrasubunit mechanism” refers to an activity of the glycogenin monomer, while the “intersubunit mechanism” involves catalytic residues from both monomers in a glycogenin dimer. The findings by Issoglio and co-workers [37], who studied the mechanisms of monomeric and dimeric rabbit muscle glycogenin, fully support the above view. They found that, while a glycogenin monomer is sufficient for priming glycogen

biosynthesis *in vivo* via the intrasubunit mechanism, the intersubunit mechanism mediated by the glycogenin dimer is needed for full polymerization capacity of glycogenin.

Human glycogenins have been shown to form non-covalent dimers with shared enzymatic activity between monomers. All crystal forms of the human glycogenin [33] contain dimers. One of the glycogenin monomers acts as the glucose-introducing transferase, while the other serves for glucose branching in the growing glycogen chain [34, 35]. Glycogenin-1 is also co-purified with glycogenin-2, and *vice versa*, suggesting that the two glycogenins may also form heterodimers.

Xylosyltransferases (PDB code 4WLM)

Xyloside xylosyltransferase-1 (XXYLT1; GTase family 8; a retaining α -1,3-xylosyltransferase; EC 2.4.2.n3) catalyzes the addition of an α -D-xylose to an existing xylose-glucose disaccharide to complete the synthesis of the trisaccharide O-linked to EGF-like repeats in Notch proteins [38]. XXYLT1 possesses the typical GT-A fold signature of DxD motif to coordinate a catalytic Mn^{2+} ion. Human XXYLT1 has been expressed in Sf9 cells as a full-length type II membrane protein and purified [38]. It was found that XXYLT1 forms SDS-resistant homodimers linked together by a disulfide bond between the transmembrane domains. The crystal structure of the luminal catalytic domain of XXYLT1 [39] is also a dimer, with an interface area between monomers well in the range typical for functionally relevant protein-protein interactions, although the ΔG of -12.7 kcal/mol is rather low (Table 1). It was assumed that the catalytic domains provide additional dimerization contacts in XXYLT1 [39]. The active sites of the catalytic domains do not overlap with the dimer interface area, and the active sites appear to be positioned in such a way that it is consistent with the orientation of the Notch acceptor proteins.

N-acetylglucosaminyl- and N-acetylgalactosaminyltransferases (PDB codes 2GAK, 1OMZ, 5FV9)

Crystal structures of three different N-acetylglucosaminyltransferases have been published. These are (i) core 2 β -1,6-N-acetylglucosaminyltransferase (C2GnT; GTase family 14; EC 2.4.1.102) [40], (ii) α -1,4-N-acetylglucosaminyltransferase (Extl2; GTase family 64; EC 2.4.1.223) [41] from mouse, and (iii) human polypeptide N-acetylgalactosaminyltransferase (GalNT2; GTase family 27; EC 2.4.1.41) [42]. Both of the two glucosaminyltransferases use UDP-N-acetylglucosamine as the substrate, but they act on different acceptor glycans in different biosynthetic pathways: C2GnT adds N-acetylglucosamine to an N-acetylgalactosamine with a 1,6-linkage making the core 2 structure of mucin type O-glycans, while Extl2 produces 1,4-linked glucuronic acid and N-acetylglucosamine repeats found in heparin sulfate chains. The human galactosaminyltransferase GalNT2 uses UDP-N-acetyl- α -D-galactosamine as a substrate to add the first sugar in mucin biosynthesis.

C2GnT was found to exist both as monomers and dimers in cells [43], while the predominant form in solution (secreted in culture media) was monomeric [40, 43]. Surprisingly, in the crystal structure the two C2GnT monomers form a disulfide-bonded dimer via Cys235 residues. However, this dimer may not reflect the physiological situation, since the Cys235 is unique to the murine enzyme. The DiMoVo score for C2GnT (2GAK) is also low (Table 1), supporting the view that the observed dimer is probably a result of crystal packing. On the other hand, the jsPISA analysis suggests that the C2GnT dimer could well be a biologically relevant dimer, even without the disulfide bridge (Table 1). Of the two molecular forms, only the dimer could be crystallized. The fact that C2GnT crystal structure contains the stem domain (in addition to the catalytic domain) makes it a rare exception among the purified and crystallized GTases. Two disulfide bridges connect the stem domain to the catalytic domain, but due to high temperature factors of the stem domain and the lack of extensive contacts between the two domains, it may not represent the conformation present in the full-length protein [40].

Extl2 does not form a disulfide-bonded dimer, but the dimeric nature of the enzyme could be assigned with more confidence than for C2GnT due to the dimer interface area, the ΔG of binding and other characteristics of jsPISA interaction radar analysis (Table 1). However, no direct experimental evidence on the protein behaviour in solution exists to support this view.

GalNT2 was crystallized with three independent dimers in the asymmetric unit. Our analysis with the EPPIC server indicates that the interactions between the monomers are only crystal contacts, despite the other parameters favouring the existence of biologically relevant dimers (Table 1). Structural studies by others on the same enzyme revealed a crystallographic dimer [44] or a dimer with an interface not likely to be biologically significant [45].

The three structures described above do not superimpose well, with an r.m.s. deviation of atomic positions in pairwise comparisons ranging from 6.7 to 16.4 Å, as estimated with PyMOL (The PyMOL Molecular Graphics System, Version 1.8 Schrödinger, LLC.).

ABO blood group antigen glycosyltransferases (PDB codes 3U0X, 3U0Y)

ABO blood group antigens attached to membrane proteins or lipids contain a common N-acetylgalactosamine-galactose-fucose trisaccharide core, which is non-antigenic and defines the type O blood. This core structure is then modified to blood type A and B antigens upon addition of an N-acetylgalactosamine or a galactose, respectively, as a terminal sugar by a relevant glycosyltransferase (GT family 6). Several high resolution apo- and holostructures of both blood group A specifying α -1,3-N-acetylgalactosaminyltransferase (GTA; EC 2.4.1.40) and blood group B specifying α -1,3-galactosyltransferase (GTB; EC 2.4.1.37) from human have been solved. In addition, a chimeric enzyme (AAGlyB) capable of transferring either of the terminal sugars has been constructed and its structure solved [46]. All of these structures are highly similar, as expected given that the GTA and GTB enzymes differ only by four amino acid residues.

The GTA (PDB 3U0Y) and GTB (PDB 3U0X) structures were solved to 1.6 Å and 1.85 Å resolution, respectively, in complex with a GTB-specific inhibitor compound [47], and present as dimers. The respective monomers are related by 2-fold symmetry, which may indicate biological relevance [48]. The stem regions of the two monomers extend to form a large dimer interface dominated by random coil, and mediating the physical interaction between the two type II membrane proteins. Dimer formation of the crystallizable species of GTA in solution has been experimentally verified by

SDS-PAGE [49]. This type of dimer contact - formed through the stem regions - appears to be a rather unique feature of only some glycosyltransferases.

GT-A variants

Sialyltransferases (PDB code 5BO7)

ST8 α -N-acetyl-neuraminide α -2,8-sialyltransferase 3 (ST8SialIII; EC 2.4.99.-) is an oligo/poly-sialylating sialyltransferase, which uses a CMP-activated sialic acid unit as a donor to add a sialic acid to a terminal position with an α -2,8 linkage on different acceptors [50]. The enzyme belongs to GTase family 29 and its crystal structure revealed a variant of the common GT-A fold [51, 52]. ST8SialIII structure displays a 612345 topology where all the strands are parallel (instead of 321465 with β 6 antiparallel). Being active on oligo- and polysialylation, a positively charged binding pocket is needed to accommodate the negatively-charged donor and acceptor molecules. The ST8SialIII crystal structure [51] revealed that such a groove is indeed formed by patches of the surface forming the dimer interface, emphasizing that the active enzyme is by necessity a dimer. In contrast, monosialylating enzymes such as ST3GalI and ST6GalI operate on uncharged acceptor molecules, and therefore do not need - and do not have - large positive binding areas [51, 53-54]. ST8SialIII's dimer interface contains symmetrical pairs of hydrogen bonds created by residues which are not conserved in monomeric ST8SialII and ST8SialIV enzymes. Static light scattering experiments carried out by Volkers *et al.* (2015) [51] confirmed that ST8SialIII is a dimer also in solution.

In the ST8SialIII dimer, the two monomers are linked to each other in a manner placing the two active sites on the same side of the dimer, but about 20 Å away from the dimer interface to opposite directions. This enables both monomers to simultaneously bind a dimeric target molecule, or possibly utilize allostery in their function [51].

Galactosyltransferases (PDB code 4IRP)

β -1,4-galactosyltransferase 7 (β 4GalT7; EC 2.4.1.133) is a proteoglycan synthesizing enzyme that adds a galactose to the second position of a growing saccharide core structure of a glycoprotein acceptor (GlcA β 1–3Gal β 1–3Gal β 1–4Xyl β 1-O-[serine]), which already contains the initiating xylose residue. It is also a drug development target for glycosaminoglycan synthesis [55]. It belongs to GTase family 7 and its crystal structure [56] revealed a variant of the GT-A fold in which the β 3 strand is replaced by a strand (β 7) present in the C-terminal domain. Thus, the topology is 721465 (Fig. 2A). The monoclinic crystal had four β 4GalT7 molecules in the asymmetric unit, forming two copies of a dimer. The dimeric nature of the protein is supported by the finding that the stoichiometry of UDP binding by β 4GalT7 was between 0.4 and 0.6 [57]. Subsequent gel filtration analysis under native conditions provided evidence for dimer formation, suggesting that only one of the monomers in the dimer is able to bind UDP-galactose.

GT-B folds

Glycogen phosphorylases (PDB codes 1YGP, 5IKO, 4BQE, 2IEG, 3DDS)

We also included glycogen phosphorylase (GP; EC 2.4.1.1) in the list of selected enzymes, together with some others (see below), because it is classified as a member of GT family 35. Yet, its catalytic activity differs from “classical” GTases due to the role of the enzyme in storage energy mobilization. It produces glucose-1-phosphate from linear stretches of glycogen chains by cleaving the α -1,4 glycosidic bonds. Glycogen phosphorylase is a well-known prototypic allosteric enzyme that can exist in a monomeric inactive state as well as in dimeric or tetrameric active states. It is well established that phosphorylation of a specific serine residue and binding of AMP increase the activity of the enzyme by triggering the conformational change of an unstructured loop into an α -helix, and by a shift in allosteric state, respectively. The sites of both of these activation events reside near the dimer interface, as deduced from the human liver GP structure [58]. A wealth of crystallographic and biochemical evidence shows that the active unit of GPs is a dimer. The change

of the oligomeric state from monomer to dimer upon activation has also recently been shown by dynamic light scattering [59].

Brain, liver and muscle isoenzyme structures of GP have been determined from human and various other organisms. The structures are highly homologous, exemplified by the 83.3% sequence identity between the isoenzymes in rabbit muscle (PDB 2IEG) [60] and human brain [59]. Despite this apparent structural identity, the dimer interface has some flexibility without affecting the activity of the enzyme. The liver isoenzyme [58] is structurally the most rigid: the dimer interface area is 3350 Å² (PDB entry 1FA9). The corresponding values for muscle (2240 Å²) [61] and brain (1400 Å²) [59] GP dimer interfaces reflect the extent of conformational changes taking place during activation of the enzymes. The same phenomenon is also seen in the yeast GP structures [62, 63].

Inhibition of glycogen phosphorylase activity is a potential strategy for drug development e.g. for diabetes treatment. Not surprisingly, structural studies with various ligands are gradually increasing our understanding of the dynamics and allostery of oligomeric structures of glycogen phosphorylases, like e.g. rabbit muscle [64] and human liver [65, 66] variants.

Instead of glycogen phosphorylases, plants have glucan phosphorylases that belong to the same GT family 35. The *Arabidopsis thaliana* glucan phosphorylase PHS2 crystal structure at 1.7 Å resolution [67] revealed a dimer, in which the active site of each monomer is buried in a cavity away from the dimer interface area. The structure is also well superimposable with the glycogen phosphorylase GT-B fold enzyme structures, and can therefore be regarded with confidence as a physiologically relevant dimer.

Glycogen synthases (PDB codes 3NB0)

Glycogen synthases (EC 2.4.1.11; GT family 3) catalyse the addition of glucose units from UDP-glucose to a growing glycogen chain. Crystal structures of the yeast isoenzyme Gsy2p have been

solved both in the apo state and in the glucose-6-phosphate activated state [68]. The amino acid sequence of Gsy2p is 51.7% identical (78.5% similar) to the corresponding human enzyme.

The structure of Gsy2p is an A/B/C/D tetramer, which is formed from different structurally or functionally relevant dimers: the interfaces between each monomer accommodate binding sites for either the allosteric activator glucose-6-phosphate or the donor and acceptor molecules. Each of the four monomers have a long α -helix extending from the core enzymatic domain, such that these four helices form a coiled coil arrangement in the centre of the tetramer (as seen for the B/D dimer in Fig. 1). These helices form the extensive monomer-monomer interaction surfaces seen in Table 1.

Sucrose synthase (PDB code 3S28)

Sucrose is synthesized from NDP-glucose and D-fructose by sucrose synthase (EC 2.4.1.13). Sucrose synthases are retaining GTases belonging to the GT family 4. Structural and biochemical studies of the *Arabidopsis thaliana* enzyme AtSus1 have shown that the oligomeric state of the enzyme is linked to the regulation of its activity [69]. AtSus1 was shown to exist solely as a tetramer by analytical gel filtration. The analysis of the crystal structure using the jsPISA server revealed two types of monomer-monomer interactions responsible for the oligomerization of AtSus1: A/B (C/D), and A/D (B/C), with interface areas of 1280 Å² and 1076 Å², respectively. Interestingly, the GT-B domains themselves do not play any major role in forming these interactions. Instead, sucrose synthase contains separate cellular targeting and peptide binding domains, which mediate the oligomerization contacts. It appears that the transition of AtSus1 tetramers to dimers precedes the phosphorylation of Ser 167, and it has been suggested that the change in oligomerization state regulates this phosphorylation step [69]. Hardin *et al.* (2006) [70] have also reported that the maize enzyme exists as a dimer rather than a tetramer.

GT-B variants

Fucosyltransferases (PDB code 4AP5, 3ZY5)

Fucose is one of the sugars found either directly linked to proteins via *O*-linkage to a serine or threonine residue, or added as a terminal sugar on branched glycan chains. Structures of fucosyltransferases catalysing both of these types of additions have been solved.

Protein *O*-fucosyltransferases 1 and 2 (POFUT1 and POFUT2; EC 2.4.1.221) are inverting enzymes of GT families 65 and 68, respectively. They transfer an α -L-fucosyl residue from GDP- β -L-fucose to the hydroxyl group of serine residues in acceptor proteins.

Human POFUT2 crystal structure is known both in apo form (PDB 4AP5) and in complex with the donor substrate (PDB 4AP6) [71]. The two molecules in the asymmetric unit of the apoprotein form a non-crystallographic dimer with an extensive monomer-monomer interface of 1670 Å². The substrate-binding cavity is formed between the two monomers such that a loop from one molecule partially covers the cavity of the other molecule. In the substrate-bound state, however, the dimer interface is reduced to 1315 Å² due to the accommodation of the substrate. Interestingly, the structure of the enzyme-substrate complex indicated that the physiologically relevant form of POFUT2 is dimeric, since in this holoenzyme structure the dimer is formed in the same way despite holding only one molecule per asymmetric unit. Thus, a crystallographic dimer in this case seems to be identical to the biologically relevant non-crystallographic dimer simply out of necessity. POFUT2 possesses a two-domain topology, representing a variant of the GT-B fold. The first domain shows a 3217465 topology, β 5 being antiparallel to the others. The second domain shows an all-parallel 3214 topology when an α -helix replaces β 5 next to β 4 in an interesting deviation from the majority of structures.

The only known crystal structure for a POFUT1 is the one of *Caenorhabditis elegans* enzyme (PDB 3ZY5; a complex with GDP-fucose). There is only one chain (A) in the asymmetric unit of the monoclinic unit cell, but there is a significantly large interface area (1297 Å²) with the

crystallographic symmetry mate molecule (A'). Therefore we included this putative A/A' dimer structure in our study. The first domain in each monomer shows a 321756 topology with an antiparallel β 3 strand, while the second domain shows a 32145 topology with all strands aligned in a parallel fashion. The EPPIC analysis (Table 1) indicates that the structure of POFUT1 is a crystallographic dimer, although other metrics suggest it to be a biological dimer. Interestingly, the same protein - but with a bound GDP instead of GDP-fucose - crystallizes with two molecules per asymmetric unit (PDB 3ZY3). Despite a sufficiently large interaction surface (1096 Å²), jsPISA analysis renders the structure a probable crystallographic dimer. It seems likely that POFUT1 does not form biological dimers, as also both the gel filtration chromatography and analytical ultracentrifugation data of Lira-Navarrete *et al.* (2011) [72] indicated that *C. elegans* POFUT1 is a monomeric protein.

C. elegans POFUT1 (424 residues in POFUT1 isoform 1) and human POFUT2 do not share considerable sequence similarity despite catalyzing the same reaction: based on ExPASy homology analysis, they share 26.8% identity (49.7% similarity) over a 179 amino acid overlap. In contrast, human POFUT1 (for which no crystal structure is available yet) is identical in sequence with the human POFUT2 over the common 383 amino acid residue part.

N-acetylglucosaminyltransferases (PDB code 4GYW)

N-acetylglucosaminyltransferase (OGT; EC 2.4.1.255) belongs to family GT41 of inverting GTases. It transfers N-acetylglucosamine from the sugar donor UDP-GlcNAc onto specific serine or threonine residues of nucleocytoplasmic proteins. It is a different GT-B variant compared to the fucosyltransferase POFUT1 described above: in addition to its GTase domain topology, it is also a considerably larger protein (1046 residues) due to its 13 tetratricopeptide repeats (TPR) containing domain. The GT-B domain topology of OGT is 3214567 for the first subdomain and 32145 for the second subdomain, with all elements parallel to each other. In the crystal structure (PDB 4GYW) [73] there is only one molecule per asymmetric unit, but molecules A and A', which are related by crystallographic symmetry, form a dimer. In fact, the TPR domains are responsible for this dimerization. This has been shown by using the TRP domain alone in crystallization [74]. N-

acetylglucosaminyltransferase therefore seems to represent an interesting and novel variant of the GT-B fold, in addition to its unique dimerization properties.

DIMER INTERFACE ANALYSES

The dimerization interface for each of the selected structures was analyzed in order to review whether any similarities exist between them. We considered six different criteria: interaction surface area and energy-related metrics, amino acid composition, secondary structure composition, topology, evolutionary conservation, and active site position in the dimer structure.

Interface area and energy-related metrics

All the selected structures show an interface area larger than 900 Å². This is commonly accepted as the minimum area for biologically relevant dimers [23, 75]. The areas vary from 941 Å² (C2GnT) to 3355 Å² (Gph1) (Table 1). The solvation free energy ΔG and the total binding energy vary from -7 to -32 kcal/mol and -14 to -48 kcal/mol, respectively. These three parameters are part of the jsPISA interaction radar score [21], and are as such reliable measures to assess dimerization in crystal structures. In table 1, we also list the jsPISA score, which is a weighted average of each of the radar metrics. A value higher than 50% depicts a good probability for the interface to be biologically relevant [21].

The DiMoVo method [22] also uses the interface area as the main criterion in assessing whether the dimers are crystallographic or biologically relevant, but it also considers other criteria such as frequencies and pairwise distances of amino acids. In this way, the predictive value compared to the interface area alone is improved from 78% to even 97%. The boundary value of the DiMoVo score is 0.5; values below 0.5 quite accurately predict crystallographic dimers, while values above 0.5 predict biological dimers. Interestingly, a low DiMoVo score was obtained for hGyg1, PHS2 and GPb (Table 1) despite their good energy metrics.

The EPPIC method [23] considers evolutionary conservation as a criterion for interaction sites. In our study, all the structures with a very low DiMoVo score also scored congruently in the EPPIC assessment (Table 1).

Amino acid composition

To analyse the amino acid composition at the dimer interfaces, we calculated the ratios between the frequency of amino acids observed at the interface and the frequency of amino acids within the full-length sequence of the crystallized proteins. Alanine residues were statistically significantly absent from the interfaces, whereas Arginine and Proline residues were statistically over-represented (Supp. Fig. 1). This finding is in line with Hashimoto *et al.* (2010) [13], whose study material consisted of 73 nonredundant GTase structures representing 31 families, but were not restricted to necessarily having non-crystallographic symmetry mates in the asymmetric unit.

Secondary structure composition

All types of secondary structures were observed in the dimerization interfaces: α -helices, β -strands, loops and disordered regions (Fig. 3A). We analyzed the secondary structure compositions of each of the topological elements responsible for dimerization contacts (Fig. 3B), and found that loops and helices are invariably the major feature. Hashimoto *et al.* (2010) [13] also found in their data set that β -strands are under-represented in the dimer interfaces.

Topology

Topological elements responsible for dimerization were analysed by examining their position with regard to the core β -strands of GT-A and GT-B folds (Fig. 2A-B and 2B). We found features that were

shared between different topological elements, as well as features that distinguish the two folds from each other (Fig. 2C).

Structures belonging to the GT-A fold were found to display a conserved dimerization interface topology, with two core dimerization elements making contacts with each other. The first element resides in the region between $\beta 5$ and $\beta 6$ (Fig. 2A and 2C, magenta); the second element is in the region after $\beta 6$ (Fig. 2A and 2C, blue). In addition to these two core elements, some families use additional elements for dimerization (Fig. 2C). For example, glucuronyltransferases use $\alpha 1$ (Fig. 2A and 2C, red), as well as the surface created by the $\beta 4'$ - βC (Fig. 2A and 2C, green). The region between $\beta 4$ and $\beta 5$ is also used by N-acetylglucosaminyltransferases, galactosyltransferases and xylosyltransferases (Fig. 2A and 2C, green). Galactosyltransferases use amino acids located in N-terminal of the core fold (before $\beta 1$) (Fig. 2A and 2C, brown).

GTase structures with the GT-B fold also display similarities in the dimerization interface topology, with the nuance that the topological elements may lie on the domain “a” or domain “b” (first and second Rossmann fold domains, respectively). Glycogen phosphorylases and sucrose synthases use almost always domain “a” for dimerization, whereas glycogen synthases, fucosyltransferases and N-acetylglucosaminyltransferases use elements from both “a” and “b” domains. The first core dimerization element of GT-B fold enzymes is the N-terminal region of the core fold, either before $\beta 1a$ or $\beta 1b$ (Fig. 2B and 2C, brown, blue); the second element is the region between either $\beta 2a$ and $\beta 3a$ or $\beta 2b$ and $\beta 3b$ (Fig. 2B and 2C, purple). The sole exception is the sucrose synthase family, which employs only the first core element and the region between $\beta 4a$ and $\beta 5a$ as an additional element (Fig. 2B and 2C, green). In glycogen phosphorylases and sucrose synthases, the region between $\beta 3a$ and $\beta 4a$ participates as an additional element (Fig. 2B and 2C, orange).

Interestingly, the structures of ST8SialIII, B4GalT7, PoFUT1 and PoFUT2, as well as OGT, which are GT-A or GT-B fold variants, display mixed dimerization elements from both folds. PoFUT1 and PoFUT2 (GT-B variants) use the region between $\beta 5$ and $\beta 6$, specific to the GT-A fold dimerization interface, as well as the regions between $\beta 2$ and $\beta 3$, specific to the GT-B fold dimerization interface. ST8SialIII (a GT-A variant) employs the N-terminal region before $\beta 1$ and the region between $\beta 3$ and

$\beta 4$, common to GT-B fold dimerization interface, and the region between $\beta 4$ and $\beta 5$ specific to GT-A fold. In B4GalT7 the N-terminal region before $\beta 1$ and the region between $\beta 2$ and $\beta 3$ specific to GT-B fold, as well as the C-terminal region after $\beta 6$, act as core element of GT-A fold dimerization.

These data emphasize the high variability existing between the identified dimer interfaces, a phenomenon in line with the existence of multiple distinct enzyme dimers. In this regard, the lack of any consensus motifs for dimerization, and the use of various topological arrangements, suggest that any individual enzyme uses a specific interaction surface only for binding itself and not any non-relevant enzyme. If the latter would be the case, the end result would be a mix of all kinds of enzyme dimers and also “mixed” glycans these enzyme complexes might make. This outcome is not desirable, and seems to be prevented by highly distinct interfaces allowing only specific interactions. A similar situation must also exist between sequentially acting enzymes that are known to form heteromeric complexes with each other [7]. Whether the interfaces in the latter case are similar to those used for the formation of enzyme homodimers remains to be clarified.

Evolutionary conservation

We also evaluated the amino acid sequence conservation in the dimerization interfaces. Briefly, multiple sequence alignments were generated by querying the sequence of each studied GTase against the OMA orthology database [76], using the InterEvolAlign server. We found various types of conservation profiles (Fig. 4), from strict conservation (red), high conservation (orange) to more diverse (yellow). The multiple sequence alignments are detailed in Suppl. Fig. 2.

Active site positioning

From the functional point of view, a feature of particular interest is how the active sites of the monomers relate to the dimer interface. In general, at least three possibilities exist: (i) the active sites are far away from each other, suggesting either an independent catalytic activity for both of

them or that dimerization is a stabilizing factor; (ii) the active sites are located close to each other to facilitate cooperative substrate binding and catalysis; or (iii) the active sites overlap with the dimerization interface in order to provide a mechanism to regulate the enzymatic activity via dimerization.

Since not all the structures contained a substrate or any other bound ligand, we inspected donor and acceptor substrate binding sites and the metal binding site (for GT-A folds) as a guide to locate the active sites. In most of the GT-A folds the active site is near $\beta 4$ and $\beta 4'$ (Fig. 2A), while in GT-B folds it seems to be predominantly located in the linker region between the two Rossmann fold domains. In most homodimers, however, the active sites are located far away from the dimerization interface, in some cases near the opposite ends of the dimer. In contrast, even though the active sites in the glucuronyltransferase dimer reside very close to each other (20 Å away), they both are still easily accessible.

DISCUSSION

In this review, we analysed various GTases using the available crystal structures of their globular catalytic domains to determine whether any of them represent biologically relevant dimers. Likely candidates were identified by choosing crystals with more than one molecule per asymmetric unit. Only the crystal structures of the globular catalytic domains of GTases are available, but there are good grounds to assume that these domains are responsible for, or at least contribute to, dimerization of the full-length GTases. This assumption is consistent with dimerization being a regulator of the enzymatic activity of the GTases. The fact that none of the GTases contain the dimerization signature sequence LxxGVxxGVxxT of single-spanning transmembrane helices [77], and that their ca. 40-80 residues long stem domains appear to lack regular secondary structure, provide strong support for the view that the catalytic domains have an important role in linking GTases to homodimers.

Phylogenetic analysis of GTases by Hashimoto *et al.* (2010) indicated that certain GTase families could be classified either as “monomer families” or “dimer families”. Structures belonging to families GT44, GT7 and GT27 (GT-A fold) and GT5, GT9 and GT80 (GT-B fold) are monomers, while GT81 and GT43 (GT-A fold) and GT35 and GT23 (GT-B fold) represent homodimers. Only a few families seem to contain a mixed population of GTase oligomers. Accordingly, structures from families 35 and 43 were over-represented in our analysis (Table 1 and Fig. 1), while none of the “monomer family” structures passed the criteria used in our study. Hashimoto *et al.* (2010) also found that, especially for the GT-B fold, homo-oligomer interfaces are more typically formed from helices and terminal regions or loop structures than from β -strands. A typical example for a GT-A fold enzyme is glucuronyltransferase GlcAT-I (family 43) [25], where the homodimer interface is formed from C-terminal ends including a long loop and the last α -helix: the substrate binding sites are near the interface and acceptor substrates are in contact with both GlcAT-I monomers. Furthermore, glycogen phosphorylase (family 35) structures form homodimers via α -helices, which are missing from family 5 monomeric glycogen glucosyltransferases [13].

As discussed by Krissinel & Henrick [78], the challenge of dividing up dimers into physiological and non-physiological ones continues to exist. It is not trivial to judge a crystallized protein as a biological dimer with confidence. The main problem here is that it is still hard to define absolute values or even reliable characteristics for a biological interface; otherwise the problem could be tackled by a bioinformatics approach. Nevertheless, the most common characteristics to assess the relevance of a dimer are the interface area (in Å²), the solvation free energy gain (kcal/mol) between the transition of isolated and interfaced structures, and the number of salt bridges or hydrogen bonds at the interface. As an example, a maximum free energy of dissociation (ΔG_0) of 15 - 20 kcal/mol should represent a biological dimer, and usually 10 or more hydrogen bonds are found in a relevant interface. However, many dimers or higher oligomers may be transient and thus possess “weak” interactions *in vivo*, which may not prevail under crystallization conditions. Transient complexes with dissociation constants higher than 100 µM ($\Delta G_0 \leq 5$ kcal/mol) may have only a 10% probability to form crystals [79], while stable complexes can be expected to crystallize without undergoing a change in the oligomerization state. The properties of the interface itself do not completely determine the binding energy, but also depends on other factors, like the size and shape of the complex and the entropy change. Therefore the function of the protein should always be taken into account along with the analysis of its crystal structure. However, it is estimated that the values obtained by calculating the binding energy and the entropy of dissociation are 80% accurate for the identification of macromolecular assemblies in crystals [78].

GTases have been shown to be able not only to function as homo-oligomers but also as hetero-oligomers [5-7]. The hetero-oligomers can also involve more than two GTases, forming functional multienzyme complexes [80]. To this day, however, no heteromeric complexes between two GTases have been crystallized, making analyses of their interactions impossible. Nevertheless, a few examples where a glycosyltransferase is forming a complex with a non-glycosyltransferase need to be addressed here briefly. β -1,4-galactosyltransferase 1 (β 4GalT1) has been crystallized in complex with α -lactalbumin (LA) and various substrates [81]. The binding site of LA partially overlaps with the substrate binding site, consistent with a regulatory role of the ligand in the complex: instead of an N-acetylglucosamine, a glucose is accepted for binding. A large conformational change of a critical loop region takes place upon LA binding. The other known example is the hetero-complex between EryCIII (3- α -mycarosylerythronolide B desosaminyl transferase), a GTase from family

1, and its partner EryCII, a cytochrome P450 family protein. The crystal structure of the EryCIII-EryCII complex has been determined [82] and it reveals a heterotetramer with an elongated quaternary organization. A homodimer of EryCIII forms the center of the complex, while EryCII molecules reside on the periphery. It is evident in this case that the interaction surfaces for homomer and heteromer formation are located in distinct surface areas of the GTase, which is a valid observation to keep in mind for possible analogy with other heterocomplexes to be solved in the future. Conversely, as indicated earlier, glycogenins 1 and 2 (Gyg1 and Gyg2) co-purify [35], indicating that the two glycogenins may also form heterodimers. Since the crystal structures of Gyg1 and Gyg2 homodimers superimpose very well (with r.m.s. deviation of 0.865Å), we hypothesize that the same interaction surface might be used both for homomers and for heteromers of these two GTases, which may be competing with each-other.

It is also worth noting that highly specific dimerization – whether homo- or heteromeric – is more likely to employ interfaces that further increase the strength of interaction. In contrast, transient interactions, with possibly a choice of interaction partners, call for interfaces that may not be clearly distinguishable from crystal contacts. This could indicate that hetero-oligomers, as well as some homo-oligomers, could be so transient that their isolation for crystallization is not favorable enough.

Lastly, it is inevitable that the data we chose - 898 crystal structures of glycosyltransferases deposited to the Protein Data Bank - contain some which are physiological enzyme dimers, but happen to have crystallized with one molecule per asymmetric unit, and therefore escaped our analysis. Equally well, as discussed above, it could be questioned whether some of our chosen cases are true dimers, or instead crystal artefacts - depending on the subjective weighting of criteria. However, it is neither possible nor meaningful to carefully review all the 898 available structures. We believe that the way we selected the structures, and the data we obtained, provides further support for the conclusion that glycosyltransferases can form - and do form - physiological dimers not only in crystals but *in vivo*.

CONCLUDING REMARKS

The main outcomes of this review are as follows. First of all, each GTase fold type uses different topological elements for constructing their dimerization interfaces. These elements serve as fingerprints within a group of a particular fold. An interesting observation is also that variant folds can use mixed topological elements from the basic GT-A and GT-B folds. Additionally, it is typical that homodimerization does not bring the active sites of the GTase monomers close to each other. Moreover, our survey revealed that different glycosyltransferases form biologically relevant homodimeric complexes. This conclusion is supported by both biochemical and structural evidence. No hetero-oligomers between different glycosyltransferases have been structurally characterized, and this poses a future challenge for understanding glycosyltransferase function.

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FIGURE LEGENDS

Figure 1.

Ribbon drawings of the 24 GTase homodimeric structures comprising the research material of this study. All structures are presented in orientations which easily show the secondary structural elements in the dimer interface, with the location of interacting residues in those structural elements colour coded as follows: before $\beta 1$ (brown), between $\beta 1$ and $\beta 2$ (red), between $\beta 2$ and $\beta 3$ (purple), between $\beta 3$ and $\beta 4$ (orange), $\beta 4'$ - βc / between $\beta 4$ and $\beta 5$ (green), between $\beta 5$ and $\beta 6$ (magenta), after $\beta 6$ (blue). Each structure can be identified with the enzyme acronym; the same identification is used in Table 1 and in the text. GT-A fold and GT-A variant structures are on the left, while GT-B fold and GT-B variant structures on the right.

Figure 2.

Topological elements responsible for dimerization are presented separately for GT-A and GT-B folds as topology diagrams (A and B respectively) and as a table indicating the use of each topological element by the studied GTases (C). A. B. Topology of the GT-A and GT-B folds. The common structural core β -sheet is in grey with the strands numbered. The topological elements connecting the core β -strands are shown with α -helices as circles, β -strands as arrows and loops/random structure as plain lines, and colour coded as follow: before $\beta 1$ (brown), between $\beta 1$ and $\beta 2$ (red), between $\beta 2$ and $\beta 3$ (purple), between $\beta 3$ and $\beta 4$ (orange), $\beta 4'$ - βc / between $\beta 4$ and $\beta 5$ (green), between $\beta 5$ and $\beta 6$ (magenta), after $\beta 6$ (blue). In (C) the same elements are tabulated to clarify the use of each element in dimer formation by each fold type. Colour coding is the same as in (A,B). As discussed in the text, certain topological elements are used for dimerization mainly or exclusively by GT-A enzymes, while a different set of elements is utilized by GT-Bs. Additionally, the mixed nature of the variant folds is evident.

Figure 3.

Analysis of the frequency of occurrence of secondary structure elements (α -helices, β -strands, loops and disordered regions) in the dimer interfaces of the 24 GTase homodimers of this study in the overall dataset (A) and in each topological element (B).

Figure 4.

Evolutionary conservation of the amino acid sequence of the dimerization interface, visualized on each monomer (the interface facing the reader) of the 24 GTases as a color gradient: from red (strictly conserved) through orange (high conservation) to yellow (more diverse). The residues not involved in the dimerization interface are displayed in grey. The placement of the monomers in the figure is the same as for the dimers in figure 1.

Supplement Figure 1.

Log-ratio of the frequency of amino acids observed at the interface and within the full-length sequence of the crystallized domains of the 24 GTase homodimers of this study. Stars indicate the statistical significance according to critical values of χ^2 .

Supplement Figure 2.

Multiple sequence alignments of the dimerization interface for each of the 24 GTases. As the residue numbers are not sequential, they are detailed below each alignment.

TABLE LEGEND

Table 1.

Summary of the analysis of the dimer interface of the 24 GTase structures. Numerical values or assessment given by each tool is discussed in the text. Organism codes: *hsa* stands for *Homo sapiens*, *ocu* for *Oryctolagus cuniculus*, *mmu* for *Mus musculus*, *sce* for *Saccharomyces cerevisiae*, *ath* for *Arabidopsis thaliana*, *cel* for *Caenorhabditis elegans*. Chains indicates the name of the chains in the crystal structure used to analyse the dimer interface: monomers related by a crystallographic symmetry are highlighted (grey background) in contrast to monomers related by a non-crystallographic symmetry (white background). IA, interface area; DG, solvation energy; BE, total binding energy. jsPISA score is a weighted average of each of the jsPISA radar metrics, for which a value higher than 50% depicts good possibility of the interface being biologically relevant. DiMoVo score values below 0.5 predict crystal dimers, while values above 0.5 predict biological dimers. EPPIC server assessment predicts biologically relevant dimers (Bio) or crystal dimers (Xtal). For each metric, grey background indicates off-limits values suggesting a crystallographic dimer rather than a dimer of physiological relevance: IA < 1200 Å², DG > -10 kcal/mol, BE > -16 kcal/mol, jsPISA score < 50%, DiMoVo score < 0.5, EPPIC assessment for a crystal dimer.

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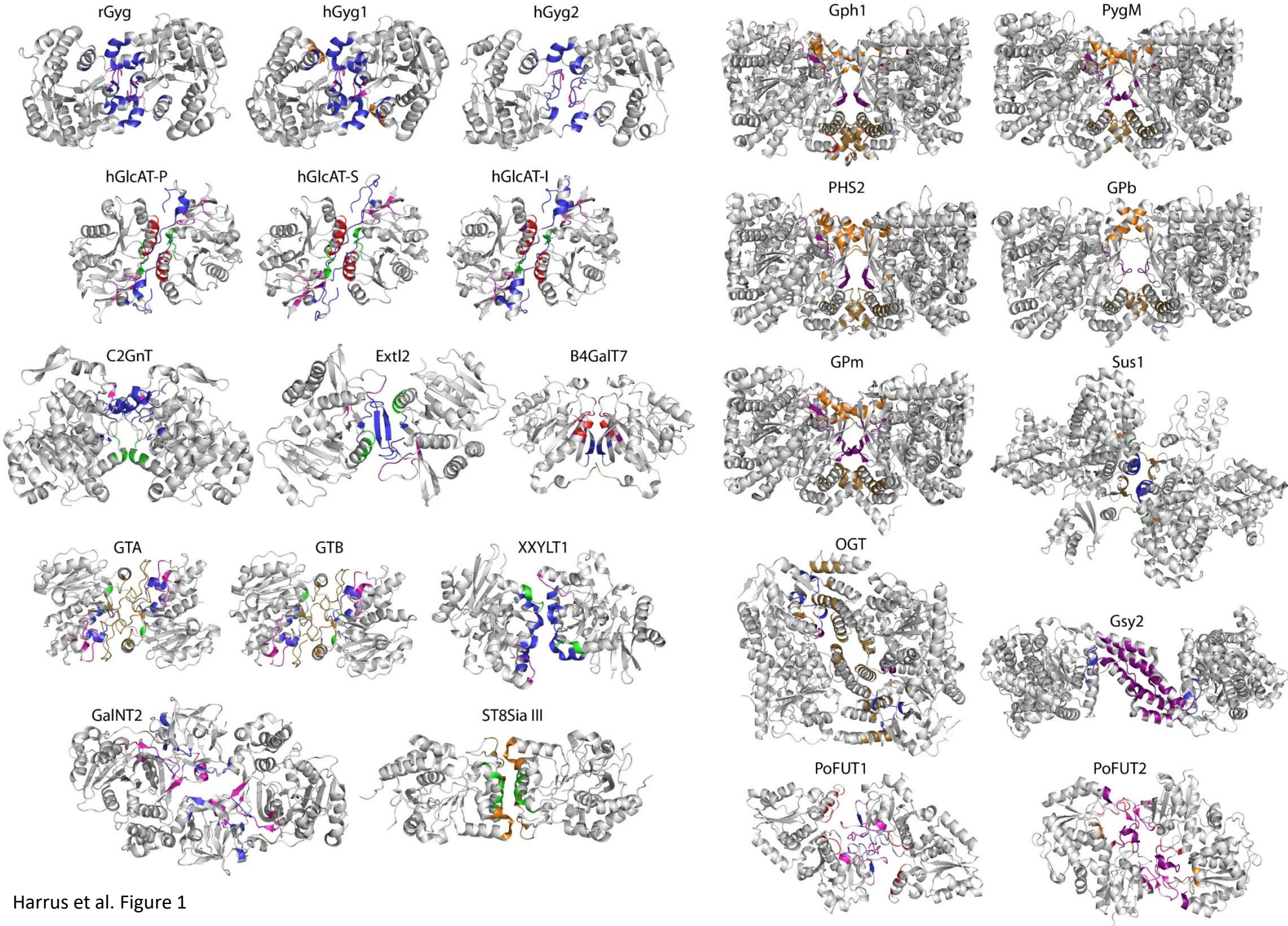
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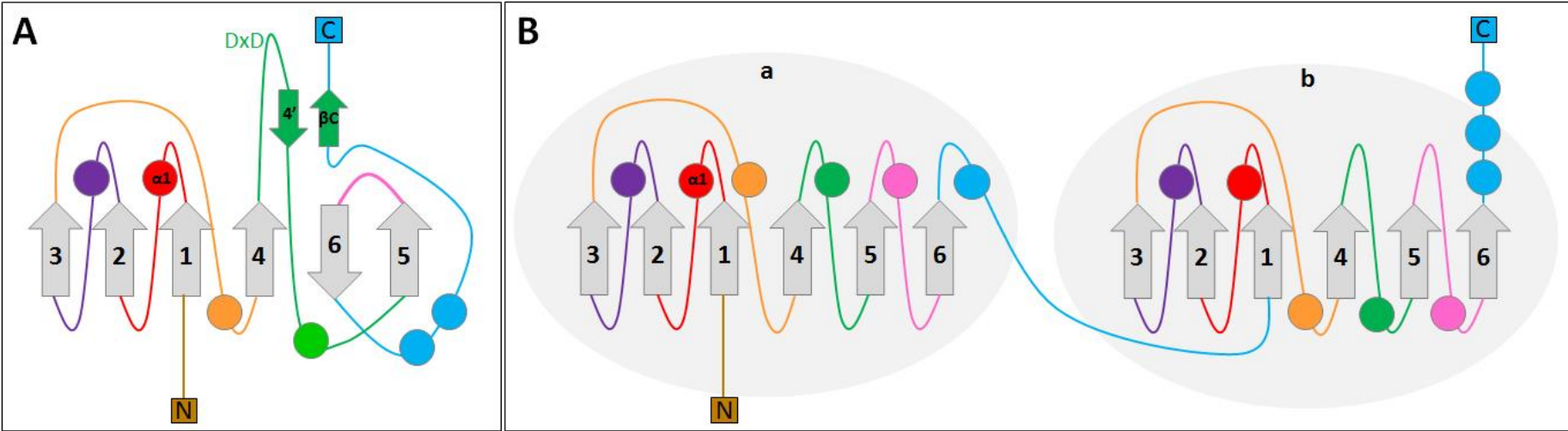
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brown: before $\beta 1$

red: between $\beta 1$ and $\beta 2$

purple: between $\beta 2$ and $\beta 3$

orange: between $\beta 3$ and $\beta 4$

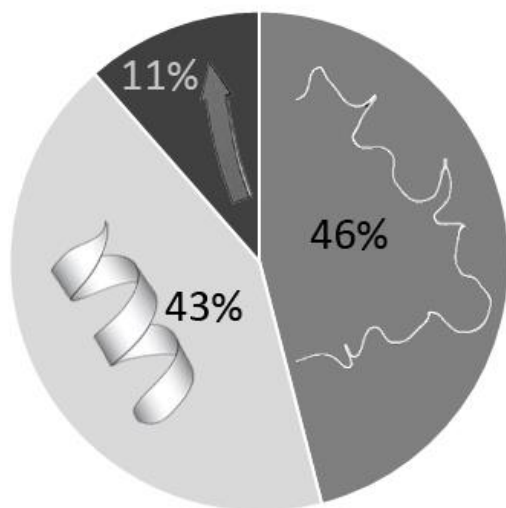
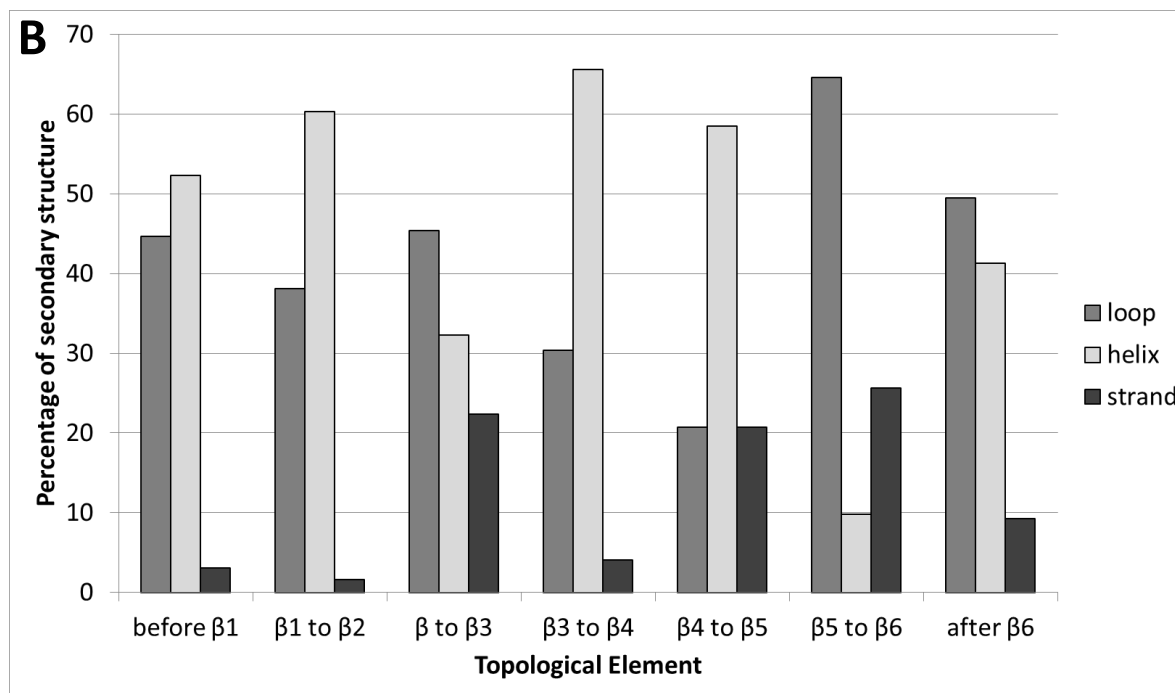
green: $\beta 4'$ - βc , or between $\beta 4$ and $\beta 5$

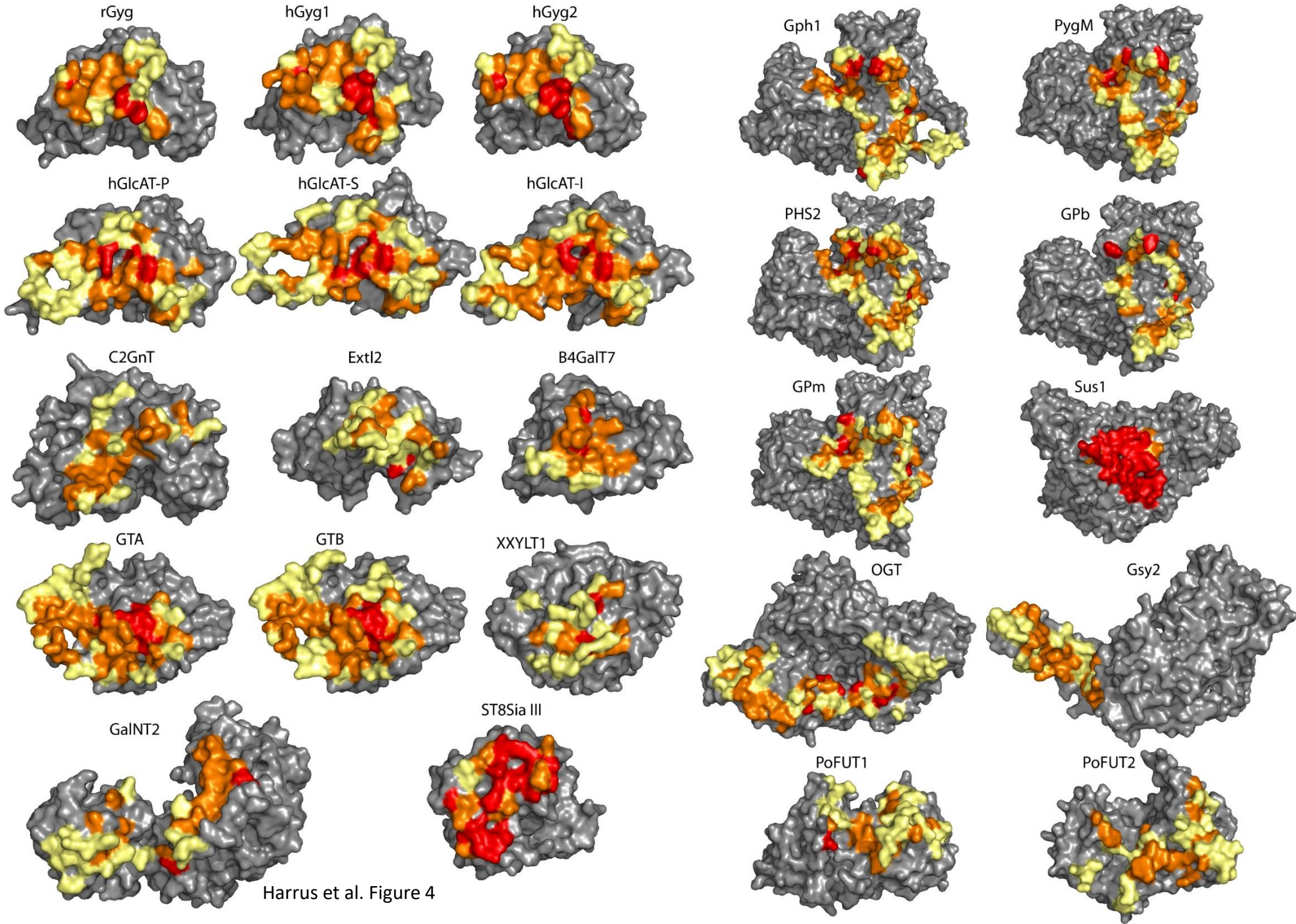
magenta: between $\beta 5$ and $\beta 6$

blue: after $\beta 6$

C

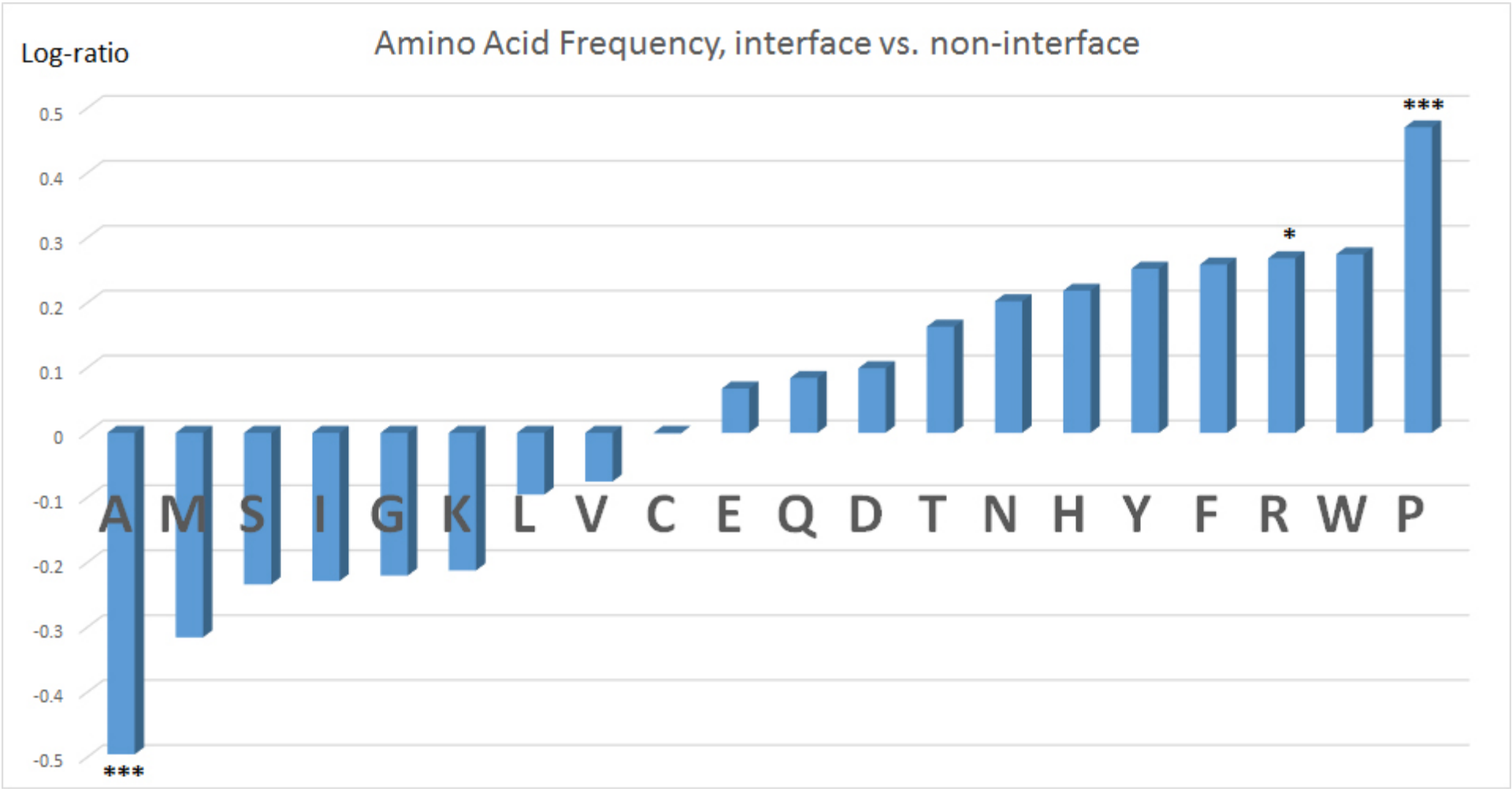
Fold	Acronym	Topological Elements Responsible For Dimerization						
GT-A	rGyg							
	hGyg1							
	hGyg2							
	GlcAT-P							
	GlcAT-S							
	GlcAT-I							
	C2GnT							
	ExtI2							
	GalNT2							
	GTB							
	GTA							
	XXYLT1							
GT-A-variants	ST8Sia III							
	B4GalT7							
GT-B	Gph1							
	PygM							
	GPm							
	PHS2							
	GPb							
	Gsy2							
	Sus1							
GT-B-variants	PoFUT2							
	PoFUT1							
	OGT							

A**B**



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Family	Organism	Acronym	PDB code	Chains	IA (Å ²)	DG (kcal/mol)	BE (kcal/mol)	jsPISA	DiMoVo	EPPIC
Glycogenins	<i>ocu</i>	rGyg	1LL0	EF	1205	-23.37	-26.92	64	0.65	Bio
	<i>hsa</i>	hGyg1	3U2U	AB	1420	-29.41	-31.63	63	0.45	Bio
	<i>hsa</i>	hGyg2	4UEG	AB'	1067	-23.43	-24.76	58	0.51	Bio
Glucuronyltransferases	<i>hsa</i>	GlcAT-P	1V84	AB	2049	-21.61	-36.12	72	0.68	Bio
	<i>hsa</i>	GlcAT-S	2D0J	AB	2225	-11.36	-31.08	73	0.57	Bio
	<i>hsa</i>	GlcAT-I	3CU0	AB	2029	-28.52	-41.11	72	0.87	Bio
N-acetylglucosaminyltransferases	<i>mmu</i>	C2GnT	2GAK	AB	941	-10.03	-14.19	56	0.10	Xtal
	<i>mmu</i>	ExtI2	1OMZ	AB	1294	-16.4	-23.06	60	0.73	Bio
	<i>hsa</i>	GalNT2	5FV9	AB	1348	-12.23	-18.01	66	0.66	Xtal
Galactosyltransferases	<i>hsa</i>	GTB	3U0X	AB	2696	-32.01	-48.09	79	1.32	Bio
	<i>hsa</i>	GTA	3U0Y	AB	2498	-27.12	-41.35	78	1.27	Bio
Xylosyltransferases	<i>mmu</i>	XXYLT1	4WLM	AB	1214	-12.7	-19.8	63	0.73	Bio
Sialyltransferases	<i>hsa</i>	ST8Sia III	5BO7	AB	974.7	-8.99	-14.62	59	0.33	Xtal
Galactosyltransferases	<i>hsa</i>	B4GalT7	4IRP	AB	757.8	-12.18	-14.84	54	0.67	Bio
Glycogen phosphorylases	<i>sce</i>	Gph1	1YGP	AB	3355	-28.04	-44.12	71	0.83	error
	<i>ocu</i>	PygM	2IEG	AB	2067	-18.99	-25.52	70	0.51	Bio
	<i>hsa</i>	GPm	3DDS	AB	2507	-26.71	-37	75	0.69	Bio
	<i>ath</i>	PHS2	4BQE	AB	2586	-23.45	-34.11	67	0.43	Bio
	<i>hsa</i>	GPb	5IKO	AC	1471	-7.446	-15.29	57	0.40	Bio
Glycogen synthases	<i>sce</i>	Gsy2	3NB0	BD	1594	-32.15	-35.26	66	0.87	Bio
Sucrose synthases	<i>ath</i>	Sus1	3S28	BC	1284	-18.23	-22.17	62	0.78	Bio
Fucosyltransferases	<i>hsa</i>	PoFUT2	4AP5	AB	1717	-15.52	-20.71	62	0.15	Xtal
	<i>cel</i>	PoFUT1	3ZY5	AA'	1297	-12.54	-18.18	62	0.27	Xtal
N-acetylglucosaminyltransferases	<i>hsa</i>	OGT	4GYW	AA'	2058	-12.14	-18.07	60	0.60	Bio



rGyg	PD	P	G	W	P	D	C	F	D	A	T	T	D	I	H	L	P	F	I	L	I	S	S	Y	P	A	F	A	F	G	
Homo_sapiens	PD	P	G	W	P	D	C	F	D	A	T	T	D	I	H	L	P	F	I	L	I	S	S	Y	P	A	F	A	F	G	
Bos_taurus	PD	P	G	W	P	D	C	F	D	A	T	T	D	I	H	L	P	F	I	L	I	S	S	Y	P	A	F	A	F	G	
Mus_musculus	PD	P	G	W	P	D	C	F	D	A	T	T	D	I	H	L	P	F	V	L	I	S	S	Y	P	A	F	A	F	G	
Monodelphis_domestica	PD	P	G	W	P	D	C	F	D	A	T	T	D	I	H	L	P	F	I	L	I	S	S	Y	P	A	F	A	F	G	
Anolis_carolinensis	PD	P	G	W	P	D	C	F	D	A	T	A	D	I	H	L	P	F	I	L	I	S	S	Y	P	A	F	A	F	G	
Taeniopygia_guttata	PD	P	G	W	P	D	C	F	D	A	T	T	D	M	H	L	P	F	I	L	T	S	S	Y	P	A	F	A	F	G	
Gallus_gallus	PD	P	G	W	P	D	C	F	D	A	T	T	D	M	H	L	P	F	I	L	T	S	S	Y	P	A	F	A	F	G	
Xenopus_(Silurana)	PD	P	G	W	P	D	C	F	D	A	T	K	D	I	H	L	P	F	V	L	V	S	S	Y	P	A	F	A	F	G	
Danio_rerio	PD	P	G	W	P	D	C	F	D	A	T	A	D	I	H	L	P	F	I	M	I	A	T	Y	P	A	F	Q	Y	G	
Tetraodon_nigroviridis	PD	P	G	W	P	D	C	F	D	A	T	A	D	I	H	L	P	F	I	L	V	A	T	Y	P	A	F	Q	F	G	
Ornithorhynchus_anatinus	PD	S	G	W	P	D	C	F	D	A	T	A	D	I	H	L	P	F	I	L	S	A	T	Y	P	A	F	Q	F	G	
Ixodes_scapularis	PD	V	G	W	P	D	C	F	D	Q	S	T	K	D	I	H	L	S	F	I	M	N	V	T	Y	P	A	Y	Q	F	S
Branchiostoma_floridae	PD	I	G	W	P	D	C	F	D	Q	G	T	K	D	I	H	L	S	F	L	M	T	I	S	Y	P	A	F	R	F	G
Nematostella_vectensis	PD	I	G	W	P	D	C	F	D	Q	S	H	E	D	I	H	L	S	F	I	M	N	A	T	Y	P	A	Y	E	F	G
Daphnia_pulex	AD	A	G	W	P	D	C	F	D	Q	A	T	K	D	I	R	L	P	F	I	M	S	G	S	Y	P	A	Y	Q	F	G
Drosophila_pseudoobscura	PD	V	S	W	P	D	C	F	D	Q	A	T	A	D	I	H	L	P	F	V	V	Y	A	C	Y	P	A	F	Q	F	R
Drosophila_willistoni	PD	V	S	W	P	D	C	F	D	Q	A	T	A	D	I	H	L	P	F	V	V	Y	A	C	Y	P	A	F	Q	F	R
Drosophila_melanogaster	PD	V	S	W	P	D	C	F	D	Q	S	T	A	D	I	H	L	P	F	V	V	Y	A	C	Y	P	A	F	Q	F	R
Aedes_aegypti	PD	V	G	W	P	D	C	F	D	Q	A	H	K	D	I	H	L	P	F	I	T	V	A	S	Y	P	A	F	Q	F	G
Anopheles_gambiae	PD	I	G	W	P	D	C	F	D	Q	A	H	K	D	I	H	L	P	F	I	T	V	A	S	Y	P	A	F	Q	F	G
Ciona_intestinalis	PD	A	G	W	P	D	M	F	D	Q	S	T	S	D	T	R	L	P	F	L	M	T	A	T	Y	P	A	F	Q	Y	G
Monosiga_brevicollis	PD	I	G	W	P	D	C	F	D	Q	A	T	Q	G	G	R	L	P	F	A	M	N	A	G	Y	P	A	F	R	F	K
Trichoplax_adhaerens	PD	V	G	W	P	D	C	F	D	Q	A	T	S	D	I	H	L	P	F	I	M	T	S	W	Y	P	A	L	R	F	S
Caenorhabditis_elegans	SD	I	G	W	P	D	S	Y	D	Q	R	D	L	P	S	R	L	P	F	I	M	G	A	T	Y	A	A	Y	R	Y	G
Ustilago_maydis	PD	T	G	W	P	D	A	W	D	Q	G	S	D	A	R	L	S	F	R	V	H	G	T	F	P	A	Y	R	Y	G	
Laccaria_bicolor	PD	V	G	W	P	D	I	W	D	Q	R	G	.	G	D	R	L	S	F	T	T	T	A	T	Y	P	A	Y	R	Y	G
Cryptococcus_neoformans	PD	T	G	W	P	D	C	F	D	Q	G	G	G	G	D	R	L	S	F	T	V	S	A	T	W	P	A	Y	R	F	G
Phaeosphaeria_nodorum	PD	I	G	W	P	D	A	F	D	Q	Q	R	L	K	F	I	C	N	A	Q	W	P	A	Y	Y	Y	K
Yarrowia_lipolytica	PD	V	G	W	P	D	V	F	D	Q	S	T	S	S	E	R	A	P	F	T	V	N	G	G	Y	P	A	Y	R	F	K
Neurospora_crassa	PD	I	G	W	P	D	L	F	D	Q	R	N	.	.	T	R	L	S	F	T	V	S	A	Q	Y	P	A	Y	H	F	Q
Kluyveromyces_lactis	PD	C	G	W	P	D	L	I	D	Q	C	H	D	G	D	R	L	P	F	F	V	N	A	Q	Y	P	A	I	F	F	A
Aspergillus_fumigatus	PD	V	G	W	P	D	I	F	D	Q	H	R	L	S	F	T	C	S	A	Q	Y	P	A	Y	H	F	Q
Lodderomyces_elongisporus	PD	S	G	W	P	D	I	F	D	Q	H	R	L	P	Y	L	V	N	Q	Q	Y	P	A	F	R	F	F
Candida_albicans	SD	S	G	W	P	D	I	F	D	Q	I	R	L	P	Y	L	V	N	Q	Q	Y	P	A	F	R	F	F
Scheffersomyces_stipitis	PD	A	G	W	P	D	I	F	D	Q	S	A	G	K	N	R	L	P	Y	V	V	N	G	Q	Y	P	A	L	R	F	F
Debaryomyces_hansenii	PD	S	G	W	P	D	I	F	D	Q	S	K	G	L	N	R	L	P	F	L	V	S	Q	Q	Y	P	A	F	R	F	F
Saccharomyces_cerevisiae	PD	I	G	W	P	D	M	I	D	Q	C	H	K	V	S	R	L	P	F	T	V	N	Y	Q	S	P	A	M	F	F	Q
Eremothecium_gossypii	PD	C	G	W	P	D	L	I	D	Q	C	H	R	G	T	T	L	P	F	L	V	N	A	Q	A	P	A	L	Y	F	R
Candida_glabrata	PD	I	G	W	P	D	I	I	D	Q	M	I	A	N	Y	R	L	P	Y	L	V	N	Y	E	C	P	A	M	F	F	G

P123 D124 P125 G126 W127 P128 D129 C130 F158 D159 Q163 A174 T175 T176 D177 I178 H181 L182 P183 F184 I185 L188 I191 S192 S195 Y196 P198 A199 F200 A202 F203 G204

hGygl	H	L	M	P	D	P	G	W	P	D	C	F	D	Q	T	A	T	T	D	I	H	L	P	F	I	L	I	S	S	Y	L	P	A	F	K	V	F	G
Bos taurus	H	L	M	P	D	P	G	W	P	D	C	F	D	Q	T	A	T	T	D	I	H	L	P	F	I	L	I	S	S	Y	L	P	A	F	K	A	F	G
Equus caballus	H	L	M	P	D	P	G	W	P	D	C	F	D	Q	T	A	T	T	D	I	H	L	P	F	I	L	I	S	S	Y	L	P	A	F	K	A	F	G
Oryctolagus cuniculus	H	L	M	P	D	P	G	W	P	D	C	F	D	Q	T	A	T	T	D	I	H	L	P	F	I	L	I	S	S	Y	L	P	A	F	K	A	F	G
Mus musculus	H	L	M	P	D	P	G	W	P	D	C	F	D	Q	T	A	T	T	D	I	H	L	P	F	V	L	I	S	S	Y	L	P	A	F	K	A	F	G
Monodelphis domestica	H	L	M	P	D	P	G	W	P	D	C	F	D	Q	T	A	T	T	D	I	H	L	P	F	I	L	I	S	S	Y	L	P	A	F	K	A	F	G
Anolis carolinensis	H	L	L	P	D	P	G	W	P	D	C	F	D	Q	T	A	T	A	D	I	H	L	P	F	I	L	I	S	S	Y	L	P	A	F	K	A	F	G
Taeniopygia guttata	H	L	M	P	D	P	G	W	P	D	C	F	D	Q	T	A	T	T	D	M	H	L	P	F	I	L	T	S	S	Y	L	P	A	F	K	A	F	G
Gallus gallus	H	L	L	P	D	P	G	W	P	D	C	F	D	Q	T	A	T	T	D	M	H	L	P	F	I	L	T	S	S	Y	L	P	A	F	K	A	F	G
Xenopus_(Silurana)	H	L	M	P	D	P	G	W	P	D	C	F	D	Q	T	A	T	K	D	I	H	L	P	F	V	L	V	S	S	Y	L	P	A	F	K	A	F	G
Danio rerio	H	M	M	P	D	P	G	W	P	D	C	F	D	Q	S	A	T	A	D	I	H	L	P	F	I	L	I	A	T	Y	L	P	A	F	K	Q	Y	G
Tetraodon nigroviridis	H	L	M	P	D	P	G	W	P	D	C	F	D	Q	S	A	T	A	D	I	H	L	P	F	I	L	V	S	S	Y	L	P	A	F	K	Q	Y	G
Ornithorhynchus anatinus	S	L	L	P	D	S	G	W	P	D	C	F	D	Q	S	A	T	A	D	I	H	L	P	F	I	L	S	A	T	Y	L	P	A	F	K	Q	F	G
Caenorhabditis briggsae	N	L	I	A	D	I	G	W	P	D	S	Y	D	Q	D	S	T	L	P	A	R	L	P	F	I	M	G	A	T	Y	A	A	Y	K	R	Y	G	
Caenorhabditis elegans	N	L	I	S	D	I	G	W	P	D	S	Y	D	Q	D	R	D	L	P	S	R	L	P	F	I	M	G	A	T	Y	A	A	Y	K	R	Y	G	
Ixodes scapularis	N	L	L	P	D	V	G	W	P	D	C	F	D	Q	Q	S	T	K	D	I	H	L	S	F	I	M	N	V	T	Y	L	P	A	Y	R	Q	F	S
Drosophila pseudoobscura	N	L	L	P	D	V	S	W	P	D	C	F	D	Q	Q	A	T	A	D	I	H	L	P	F	V	V	Y	A	C	Y	L	P	A	F	K	Q	F	R
Drosophila willistoni	N	L	L	P	D	V	S	W	P	D	C	F	D	Q	Q	A	T	A	D	I	H	L	P	F	V	V	Y	A	C	Y	L	P	A	F	K	Q	F	R
Drosophila melanogaster	N	L	L	P	D	V	S	W	P	D	C	F	D	Q	Q	S	T	A	D	I	H	L	P	F	V	V	Y	A	C	Y	L	P	A	F	K	Q	F	R
Aedes aegypti	N	L	L	P	D	V	G	W	P	D	C	F	D	Q	S	A	H	K	D	I	H	L	P	F	I	T	V	A	S	Y	L	P	A	F	K	Q	F	G
Anopheles gambiae	N	L	L	P	D	I	G	W	P	D	C	F	D	Q	S	A	H	K	D	I	H	L	P	F	I	T	V	A	S	Y	L	P	A	F	K	Q	F	G
Daphnia pulex	H	L	L	A	D	A	G	W	P	D	C	F	D	Q	S	A	T	K	D	I	R	L	P	F	I	M	S	G	S	Y	R	P	A	Y	K	Q	F	G
Branchiostoma floridae	K	L	L	P	D	I	G	W	P	D	C	F	D	Q	T	G	T	K	D	I	H	L	S	F	L	M	T	I	S	Y	L	P	A	F	N	R	F	G
Nematostella vectensis	N	L	L	P	D	I	G	W	P	D	C	F	D	Q	S	S	H	E	D	I	H	L	S	F	I	M	N	A	T	Y	A	P	A	Y	K	E	F	G
Trichoplax adhaerens	H	L	L	P	D	V	G	W	P	D	C	F	D	Q	E	A	T	S	D	I	H	L	P	F	I	M	T	S	W	Y	A	P	A	L	N	R	F	S
Monosiga brevicollis	H	L	L	P	D	I	G	W	P	D	C	F	D	Q	E	A	T	Q	G	G	R	L	P	F	A	M	N	A	G	Y	A	P	A	F	E	R	F	K
Ciona intestinalis	H	L	L	P	D	A	G	W	P	D	M	F	D	Q	S	S	T	S	D	T	R	L	P	F	L	M	T	A	T	Y	S	P	A	F	A	Q	Y	G
Ustilago maydis	N	L	L	P	D	T	G	W	P	D	A	W	D	Q	D	A	A	D	S	Q	R	L	S	F	R	V	H	G	T	F	A	P	A	Y	Q	R	Y	G
Laccaria bicolor	G	L	L	P	D	V	G	W	P	D	I	W	D	Q	R	G	.	.	G	D	R	L	S	F	T	T	T	A	T	Y	A	P	A	Y	E	R	Y	G
Cryptococcus neoformans	G	L	M	P	D	T	G	W	P	D	C	F	D	Q	E	G	G	G	D	R	L	S	F	T	V	S	A	T	W	A	P	A	Y	K	R	F	G	
Phaeosphaeria nodorum	N	L	M	P	D	I	G	W	P	D	A	F	D	Q	Q	R	L	K	F	T	C	N	A	Q	W	E	P	A	Y	R	Y	Y	K	
Yarrowia lipolytica	A	L	L	P	D	V	G	W	P	D	V	F	D	Q	E	S	T	S	S	E	R	A	P	F	T	V	N	G	G	Y	E	P	A	Y	T	R	F	K
Neurospora crassa	N	L	M	P	D	I	G	W	P	D	L	F	D	Q	M	R	.	.	N	T	R	L	S	F	T	V	S	A	Q	Y	I	P	A	Y	K	H	F	Q
Aspergillus fumigatus	N	L	M	P	D	V	G	W	P	D	I	F	D	Q	M	H	R	L	S	F	T	C	S	A	Q	Y	I	P	A	Y	K	H	F	Q
Kluyveromyces lactis	N	M	L	P	D	C	G	W	P	D	L	I	D	Q	C	H	D	G	D	R	L	P	F	F	V	P	A	Q	Y	S	P	A	I	K	F	F	A	
Lodderomyces elongisporus	K	E	T	P	D	S	G	W	P	D	I	F	D	Q	E	H	R	L	P	Y	L	V	N	Q	Q	Y	L	P	A	F	H	R	F	F
Candida albicans	K	E	Q	S	D	S	G	W	P	D	I	F	D	Q	E	I	R	L	P	Y	L	V	N	Q	Q	Y	L	P	A	F	N	R	F	F
Scheffersomyces stipitis	K	Q	R	P	D	A	G	W	P	D	I	F	D	Q	E	S	A	G	K	N	R	L	P	Y	V	V	N	G	Q	Y	L	P	A	L	H	R	F	F
Debaryomyces hansenii	K	S	Q	P	D	S	G	W	P	D	I	F	D	Q	E	S	K	G	L	N	R	L	P	F	L	V	S	Q	Q	Y	L	P	A	F	D	R	F	F
Eremothecium gossypii	N	A	L	P	D	C	G	W	P	D	L	I	D	Q	L	C	H	R	G	T	T	L	P	F	L	V	P	A	Q	A	T	P	A	L	D	Y	F	R
Saccharomyces cerevisiae	N	L	L	A	D	I	G	W	P	D	M	I	D	Q	Q	C	T	D	E	Q	L	S	F	T	V	P	L	Q	S	S	P	A	M	N	Y	F	K	
Candida glabrata	N	M	L	P	D	I	G	W	P	D	I	I	D	Q	Q	S	T	K	.	E	R	L	P	Y	L	V	P	Y	E	C	P	P	A	M	K	F	E	G

H71 L74 M75 P124 D125 P126 G127 W128 P129 D130 C131 F159 D160 Q164 T169 A175 T176 T177 D178 I179 H182 L183 P184 F185 I186 L189 I192 S193 S196 Y197 L198 P199
A200 F201 K202 V203 F204 G205

hGyg2	P	P	G	W	P	D	C	F	D	N	S	T	T	D	I	H	H	L	P	F	I	L	S	P	A	F	Q	F	G
Callithrix_jacchus	P	P	G	W	P	D	C	F	D	N	S	T	A	D	I	R	H	L	P	F	I	L	N	P	A	F	Q	F	G
Canis_lupus	P	P	G	W	P	D	C	F	D	N	S	T	A	D	I	H	H	L	P	F	I	L	N	P	A	F	R	F	G
Equus_caballus	P	P	G	W	P	D	C	F	D	N	S	T	A	D	I	H	H	L	P	F	I	L	N	P	A	F	Q	F	G
Pan_troglodytes	P	P	G	W	P	D	C	F	D	N	S	T	T	D	I	H	H	L	P	F	I	L	N	P	A	F	Q	F	G
Bos_taurus	P	P	G	W	P	D	C	F	D	N	S	T	A	D	I	Q	H	L	P	F	I	L	N	P	A	F	Q	F	G
Ornithorhynchus_anatinus	P	S	G	W	P	D	C	F	D	N	A	T	A	D	I	R	H	L	P	F	I	L	S	P	A	F	Q	F	G
Gallus_gallus	P	S	G	W	P	D	C	F	D	N	A	T	A	D	I	G	H	L	P	F	I	L	S	P	A	F	H	F	G
Anolis_carolinensis	P	S	G	W	P	D	C	F	D	N	A	T	K	D	I	S	H	L	P	F	I	L	S	P	A	F	H	F	G
Xenopus_(Silurana)	P	S	G	W	P	D	C	F	D	N	A	T	A	D	I	S	H	L	P	F	I	L	S	P	A	F	Q	F	G
Mus_musculus	P	P	G	W	P	D	C	F	D	N	A	T	T	D	I	T	H	L	P	F	V	L	I	P	A	F	A	F	G
Oryctolagus_cuniculus	P	P	G	W	P	D	C	F	D	N	A	T	T	D	I	R	H	L	P	F	I	L	I	P	A	F	A	F	G
Danio_rerio	P	P	G	W	P	D	C	F	D	N	A	T	A	D	I	T	H	L	P	F	I	M	I	P	A	F	Q	Y	G
Tetraodon_nigroviridis	P	P	G	W	P	D	C	F	D	N	A	T	A	D	I	S	H	L	P	F	I	L	V	P	A	F	Q	Y	G
Ciona_intestinalis	P	A	G	W	P	D	M	F	D	N	S	T	S	D	T	S	R	L	P	F	L	M	T	P	A	F	Q	Y	G
Ixodes_scapularis	P	V	G	W	P	D	C	F	D	N	S	T	K	D	I	N	H	L	S	F	I	M	N	P	A	Y	Q	F	S
Monosiga_brevicollis	P	I	G	W	P	D	C	F	D	N	A	T	Q	G	G	E	R	L	P	F	A	M	N	P	A	F	R	F	K
Anopheles_gambiae	P	I	G	W	P	D	C	F	D	N	A	H	K	D	I	Q	H	L	P	F	I	T	V	P	A	F	Q	F	G
Aedes_aegypti	P	V	G	W	P	D	C	F	D	N	A	H	K	D	I	A	H	L	P	F	I	T	V	P	A	F	Q	F	G
Drosophila_melanogaster	P	V	S	W	P	D	C	F	D	N	S	T	A	D	I	K	H	L	P	F	V	V	Y	P	A	F	Q	F	R
Drosophila_willistoni	P	V	S	W	P	D	C	F	D	N	A	T	A	D	I	K	H	L	P	F	V	V	Y	P	A	F	Q	F	R
Drosophila_pseudoobscura	P	V	S	W	P	D	C	F	D	N	A	T	A	D	I	K	H	L	P	F	V	V	Y	P	A	F	Q	F	R
Daphnia_pulex	A	A	G	W	P	D	C	F	D	N	A	T	K	D	I	S	R	L	P	F	I	M	S	P	A	Y	Q	F	G
Nematostella_vectensis	P	I	G	W	P	D	C	F	D	N	S	H	E	D	I	S	H	L	S	F	I	M	N	P	A	Y	E	F	G
Branchiostoma_floridae	P	I	G	W	P	D	C	F	D	N	G	T	K	D	I	S	H	L	S	F	L	M	T	P	A	F	R	F	G
Trichoplax_adhaerens	P	V	G	W	P	D	C	F	D	N	A	T	S	D	I	N	H	L	P	F	T	M	T	P	A	L	R	F	S
Caenorhabditis_elegans	S	I	G	W	P	D	S	Y	D	N	R	D	L	P	S	E	R	L	P	F	I	M	G	A	A	Y	R	Y	G
Ustilago_maydis	P	T	G	W	P	D	A	W	D	N	A	A	S	R	G	W	R	L	S	F	R	V	H	P	A	Y	R	Y	G
Laccaria_bicolor	P	V	G	W	P	D	I	W	D	R	G	.	.	G	D	W	R	L	S	F	T	T	T	P	A	Y	R	Y	G
Yarrowia_lipolytica	P	V	G	W	P	D	V	F	D	N	S	T	S	G	S	W	R	A	P	F	T	V	N	P	A	Y	R	F	K
Cryptococcus_neoformans	P	T	G	W	P	D	C	F	D	N	G	G	G	G	D	W	R	L	S	F	T	V	S	P	A	Y	R	F	G
Phaeosphaeria_nodorum	P	I	G	W	P	D	A	F	D	N	Q	R	L	K	F	T	C	N	P	A	Y	Y	Y	K
Neurospora_crassa	P	I	G	W	P	D	L	F	D	N	N	.	.	.	T	Y	R	L	S	F	T	V	S	P	A	Y	H	F	Q
Aspergillus_fumigatus	P	V	G	W	P	D	I	F	D	N	H	R	L	S	F	T	C	S	P	A	Y	H	F	Q
Eremothecium_gossypii	P	C	G	W	P	D	L	I	D	N	C	H	R	N	E	W	T	L	P	F	L	V	N	P	A	L	Y	F	R
Kluyveromyces_lactis	P	C	G	W	P	D	L	I	D	N	C	H	D	T	E	W	R	L	P	F	F	V	N	P	A	I	F	F	A
Lodderomyces_elongisporus	P	S	G	W	P	D	I	F	D	N	H	R	L	P	Y	L	V	N	P	A	F	R	F	F
Candida_albicans	S	S	G	W	P	D	I	F	D	N	I	R	L	P	Y	L	V	N	P	A	F	R	F	F
Scheffersomyces_stipitis	P	A	G	W	P	D	I	F	D	N	S	A	G	K	N	W	R	L	P	Y	V	V	N	P	A	L	R	F	F
Debaryomyces_hansenii	P	S	G	W	P	D	I	F	D	N	S	K	G	L	N	W	R	L	P	F	L	V	S	P	A	F	R	F	F
Candida_glabrata	P	I	G	W	P	D	I	I	D	N	S	T	.	K	E	W	R	L	P	Y	L	V	N	P	A	M	F	F	G
Saccharomyces_cerevisiae	P	I	G	W	P	D	M	I	D	N	S	K	E	M	E	W	R	L	P	F	T	V	N	P	A	M	F	F	Q

P126 P128 G129 W130 P131 D132 C133 F161 D162 N170 S177 T178 T179 D180 I181 H182 H184 L185 P186 F187 I188 L191 S200 P201 A202 F203 Q205 F206 G207

GlcAT-P	PV	KATR	N	T	L	H	D	N	T	Y	S	L	A	F	G	G	L	R	Y	K	T	V	F	T	I	W	T	R	T	E	P	V	L	V	N	E	F	T	D	P	V	E	I
Danio rerio	PV	KATR	N	T	L	H	D	N	T	Y	S	L	A	F	G	G	L	R	Y	K	T	V	F	T	I	W	T	R	T	E	P	V	L	V	N	E	F	T	D	P	V	E	I
Xenopus (Silurana)	PV	KATR	N	T	L	H	D	N	T	Y	S	L	A	F	G	G	L	R	Y	K	T	V	F	T	I	W	T	R	T	E	P	V	L	V	N	E	F	T	D	P	V	E	I
Branchiostoma floridae	HV	KATR	Q	T	L	H	D	N	T	Y	S	L	G	L	G	G	M	R	F	Y	T	Y	W	T	V	W	T	R	T	E	P	F	L	I	Q	A	.	S	D	P	I	E	V
Oryctolagus cuniculus	PV	KATR	N	T	R	Q	D	N	T	Y	S	L	G	L	G	G	R	R	Y	T	G	W	T	V	W	T	R	T	E	P	V	K	Y	R	L	D	V	I	E	V	.	.	.
Ixodes scapularis	HV	EATR	H	T	R	L	D	N	T	Y	D	L	G	L	G	G	L	V	V	N	A	V	W	S	V	W	T	R	T	E	P	G	P	R	R	K	A	D	E	K	G	S	L
Daphnia pulex	PL	KATR	Q	T	L	L	D	N	T	Y	S	L	G	L	G	S	V	R	F	S	T	G	W	T	V	W	T	R	T	E	P	D	L	K	G	K	I	V	D	W	V	E	V
Nematostella vectensis	LT	KATR	Q	T	L	H	D	N	T	Y	D	I	G	I	G	G	L	I	W	H	T	D	W	T	V	W	T	Q	I	A	P	R	L	Q	R	L	P	M	E	V	.	.	.
Ciona intestinalis	WT	KATR	Q	T	L	H	D	N	T	Y	T	L	G	L	G	G	L	K	F	Y	T	A	W	S	V	W	T	R	T	E	P	A	L	I	K	Q	.	S	N	S	M	E	V
Ciona savignyi	WT	KATR	Q	T	L	H	D	N	T	Y	S	L	G	L	G	G	L	K	F	Y	T	A	W	S	V	W	T	R	T	E	P	A	L	I	R	Q	.	S	N	S	M	E	V
Aedes aegypti	PV	KATR	H	V	R	L	D	N	T	Y	S	T	G	L	G	G	L	M	V	N	S	A	W	T	V	W	T	R	T	E	P	Q	L	V	K	S	.	S	D	T	L	E	V
Anopheles gambiae	PV	KATR	Q	V	R	L	D	N	T	Y	S	T	G	L	G	G	L	M	V	N	S	A	W	K	V	W	T	R	T	E	P	A	L	Q	K	S	.	S	N	D	M	E	V
Drosophila melanogaster	PA	KATR	H	L	M	L	D	N	S	Y	S	T	G	L	G	G	L	M	V	N	A	A	W	T	V	W	T	R	T	E	T	A	L	L	K	.	S	D	G	M	E	V	
Drosophila willistoni	PA	KATR	H	L	M	L	D	N	S	Y	S	V	G	L	G	G	L	M	V	N	A	A	W	R	V	W	T	R	T	E	T	A	L	Q	R	Q	.	S	D	G	M	E	V
Apis mellifera	PV	KATR	Q	T	L	H	D	N	T	Y	S	I	G	L	G	G	L	M	V	N	A	A	W	T	V	W	T	R	T	E	P	M	L	I	K	K	.	S	N	I	I	E	V
Trichoplax adhaerens	AT	KATR	Q	T	Q	H	D	N	T	Y	D	K	G	L	G	G	L	R	F	R	V	V	F	R	I	W	T	T	T	A	A	Y	V	K	A	A	.	E	D	D	V	E	V
Caenorhabditis remanei	AA	KATR	Y	T	S	H	D	N	T	Y	D	L	G	I	G	G	M	F	V	N	A	V	W	T	V	W	T	R	T	E	S	I	E	K	L	T	F	N	E	L	V	K	I
Caenorhabditis briggsae	AA	KATR	Y	T	S	H	D	N	T	Y	D	L	G	I	G	G	M	F	V	N	S	I	W	T	V	W	T	R	T	E	S	V	E	K	L	T	F	N	E	L	V	A	Y
Caenorhabditis elegans	AA	RATR	Y	T	S	H	D	N	T	Y	D	L	G	I	G	G	M	F	V	N	A	V	W	T	V	W	T	R	T	E	P	I	D	R	L	T	F	N	S	L	V	D	N
Monosiga brevicollis	SS	HVTR	Y	T	R	Q	D	N	T	Y	S	L	A	F	G	G	L	S	Y	H	V	A	W	D	V	W	T	R	T	E	P	K	L	P	P	D	I	I
Drosophila pseudoobscura	REIP	TR	H	T	L	H	D	N	T	Y	D	L	G	F	A	D	Y	G	V	L	D	S	W	S	I	W	T	Q	I	K	K	S	E	Y	L	D	L	G	A	K	M	G	V
Oryza sativa	PQAY	NR	H	V	K	N	E	R	S	Y	M	S	A	I	T	G	I	K	L	N	T	K	K	N	V	W	F	N	L	E	P	G	W	S	L	H	D	A	I	P	V	.	.
Sorghum bicolor	PHAY	NR	H	V	K	N	E	R	V	Y	S	V	A	T	V	G	A	R	L	H	T	R	R	T	V	W	F	D	L	E	P	G	W	L	L	Q	D	I	V	P	I	.	.
Populus trichocarpa	ALAY	NR	Q	V	R	L	D	N	V	Y	S	L	A	M	A	Q	S	K	V	H	T	K	S	S	I	W	L	H	L	D	H	G	W	L	L	Q	E	V	Q	P	I	.	.
Vitis vinifera	ALAF	NR	Q	V	R	L	D	N	I	Y	S	L	A	M	A	Q	S	K	L	H	T	K	S	S	I	W	L	H	L	E	R	G	W	L	L	Q	D	V	L	P	I	.	.
Arabidopsis thaliana	AMAY	NR	Q	T	R	L	D	N	I	Y	S	L	A	M	A	Q	S	K	L	H	T	K	S	S	I	W	L	H	L	D	L	G	W	A	I	Q	A	I	T	M	.	.	

P96 V97 K99 A100 T103 R104 N107 T108 L110 H111 D197 N198 T199 Y200 S201 L202 A220 F221 G223 G224 L225 R226 Y227 K242 T243 V244 F245 T305 I307 W310 T312 R313
T314 E315 P317 V318 L319 V320 N321 E322 F327 T328 D329 P330 V332 E333 I334

GlcAT-S	P	V	K	A	T	R	N	T	R	Q	R	D	N	T	Y	S	L	E	L	G	G	R	R	Y	R	L	Y	G	W	T	K	V	W	H	T	R	T	E	K	V	N	L	N	E	P	H	D	T	V	K	I	E	V
Xenopus_(Silurana)	P	V	K	A	T	R	N	T	R	Q	R	D	N	T	Y	S	L	E	L	G	G	R	R	Y	R	V	Y	G	W	T	K	V	W	H	T	R	T	E	K	V	N	L	N	E	P	P	D	T	I	K	I	E	V
Danio_reio	A	V	K	A	T	R	N	T	R	Q	R	D	N	T	Y	S	L	E	L	G	G	R	R	Y	R	L	Y	G	W	T	Q	V	W	H	T	R	T	E	K	V	N	L	N	E	P	Q	D	S	V	F	I	E	V
Takifugu_rubripes	P	V	K	A	T	R	H	A	R	Q	R	D	N	T	Y	S	L	E	F	G	G	R	S	Y	R	L	Y	G	W	T	Q	V	W	H	T	R	T	E	K	P	H	L	N	E	P	R	D	T	V	V	I	E	V
Branchiostoma_floridae	H	V	K	A	T	R	Q	T	L	H	S	D	N	T	Y	S	L	Q	L	G	G	M	R	F	R	V	Y	Y	W	T	K	V	W	H	T	R	T	E	K	P	K	M	Q	E	Q	R	S	D	P	R	I	E	V
Oryzias_latipes	P	V	K	A	T	R	N	T	L	H	E	D	N	T	Y	S	L	E	F	G	G	L	R	Y	S	V	K	V	F	T	K	I	W	H	T	R	T	E	K	P	V	L	N	E	G	G	T	D	P	N	V	E	I
Meleagris_gallopavo	P	V	K	A	T	R	N	T	L	H	D	D	N	T	Y	S	L	E	F	G	G	L	R	Y	S	V	K	V	F	T	K	I	W	H	T	R	T	E	K	P	V	L	N	E	G	G	T	D	P	N	V	E	I
Ixodes_scapularis	H	V	E	A	T	R	H	T	R	L	R	D	N	T	Y	D	L	R	L	G	G	L	V	V	K	L	N	V	W	S	Q	V	W	H	T	R	T	E	A	P	K	L	M	E	P	K	N	S	V	A	D	E	L
Daphnia_pulex	P	L	K	A	T	R	Q	T	L	L	Q	D	N	T	Y	S	L	E	L	G	S	V	R	F	R	L	S	G	W	T	K	V	W	H	T	R	T	E	N	P	K	L	D	L	V	M	V	D	W	T	V	E	V
Apis_mellifera	P	V	K	A	T	R	Q	T	L	H	N	D	N	T	Y	S	I	K	L	G	G	L	M	V	K	I	N	A	W	T	K	V	W	H	T	R	T	E	P	P	Q	L	V	E	K	K	S	N	I	G	I	E	V
Anopheles_gambiae	P	V	K	A	T	R	Q	V	R	L	R	D	N	T	Y	S	T	E	L	G	G	L	M	V	K	L	N	A	W	K	D	V	W	H	T	R	T	E	T	P	K	L	A	E	K	K	S	N	D	G	M	E	V
Aedes_aegypti	P	V	K	A	T	R	H	V	R	L	R	D	N	T	Y	S	T	E	L	G	G	L	M	V	K	L	N	A	W	T	E	V	W	H	T	R	T	E	A	P	K	L	A	E	K	K	S	D	T	G	L	E	V
Drosophila_willistoni	P	A	K	A	T	R	H	L	M	L	R	D	N	S	Y	S	V	E	L	G	G	L	M	V	K	L	N	A	W	R	D	V	W	H	T	R	T	E	K	T	K	L	S	E	R	K	S	D	G	G	M	E	V
Drosophila_melanogaster	P	A	K	A	T	R	H	L	M	L	R	D	N	S	Y	S	T	E	L	G	G	L	M	V	R	L	N	A	W	T	D	V	W	H	T	R	T	E	K	T	K	L	A	E	K	Q	S	D	G	G	M	E	V
Trichoplax_adhaerens	A	T	K	A	I	R	Q	T	Q	H	R	D	N	T	Y	D	K	D	L	G	G	L	R	F	G	R	R	V	F	R	E	I	W	H	T	T	A	R	A	H	L	R	E	A	T	E	D	D	K	V	E	V	
Ciona_intestinalis	W	T	K	A	T	R	Q	T	L	H	F	D	N	T	Y	T	L	Q	L	G	G	L	K	F	G	K	Y	A	W	S	E	V	W	H	T	R	T	E	K	P	K	M	H	E	K	K	S	N	S	K	M	E	V
Ciona_savignyi	W	T	K	A	T	R	Q	T	L	H	F	D	N	T	Y	S	L	R	L	G	G	L	K	F	G	K	Y	A	W	S	K	V	W	H	T	R	T	E	K	P	K	M	Q	E	R	K	S	N	S	E	M	E	V
Caenorhabditis_remanei	A	A	K	A	T	R	Y	T	S	H	R	D	N	T	Y	D	L	K	I	G	G	M	F	V	T	L	N	V	W	T	K	V	W	H	T	R	T	E	K	S	K	L	K	E	K	G	N	E	L	G	V	K	I
Caenorhabditis_briggsae	A	A	K	A	T	R	Y	T	S	H	R	D	N	T	Y	D	L	K	I	G	G	M	F	V	T	L	N	V	W	T	K	V	W	H	T	R	T	E	K	P	K	L	K	E	K	G	N	S	L	G	V	D	N
Caenorhabditis_elegans	A	A	R	A	T	R	Y	T	S	H	R	D	N	T	Y	D	L	K	I	G	G	M	F	V	T	L	N	V	W	T	K	V	W	H	T	R	T	E	K	P	K	L	K	E	K	G	N	S	L	G	V	D	N
Nematostella_vectensis	F	V	K	A	T	Q	N	A	K	G	R	D	N	T	Y	D	S	E	F	G	A	R	W	G	V	H	N	W	K	K	V	W	H	T	R	T	E	T	P	R	I	G	E	K	H	S	D	P	D	I	E	T	
Monosiga_brevicollis	S	S	H	V	T	R	Y	T	R	Q	D	D	N	T	Y	S	L	E	F	G	G	L	S	Y	G	V	H	A	W	D	R	V	W	H	T	R	T	E	L	P	N	L	Q	E	G	P	D	R	R	P	I	I	I
Drosophila_pseudoobscura	P	E	L	A	T	R	Y	T	K	H	R	D	N	T	Y	D	I	S	L	T	K	T	G	V	S	I	Y	G	W	R	D	I	W	H	T	Q	T	K	K	N	P	P	Q	A	K	Y	N	T	D	K	L	L	V

P92 V93 K95 A96 T99 R100 N103 T104 R106 Q107 R132 D187 N188 T189 Y190 S191 L192 E193 L211 G213 G214 R215 R216 Y217 R219 L221 Y231 G233 W234 T294 K295 V296
W299 H300 T301 R302 T303 E304 K305 V306 N307 L308 N310 E311 P312 H315 D317 T318 V319 K320 I321 E322 V323

GlcAT-I	L	V	K	A	E	V	R	Q	T	S	L	T	Y	S	R	E	P	G	L	G	G	L	R	F	H	T	A	W	C	T	V	W	T	R	T	E	P	K	M	K	Q	E	L	R	G	S	D	P	I	E	V
Monodelphis domestica	L	V	K	A	E	I	R	Q	T	S	L	T	Y	S	R	E	P	G	L	G	G	L	R	F	H	T	A	W	C	T	V	W	T	R	T	E	P	K	M	K	Q	E	L	R	G	S	D	P	I	E	V
Anolis carolinensis	L	V	K	A	E	V	R	Q	T	M	H	T	Y	S	L	R	P	G	L	G	G	L	R	F	Y	T	A	W	C	T	V	W	T	R	T	E	P	K	M	K	Q	E	L	R	G	S	D	P	I	E	V
Xenopus (Silurana)	P	H	R	A	E	T	R	Q	T	L	L	T	Y	S	V	R	P	G	L	G	G	L	R	Y	H	T	A	W	C	T	V	W	T	R	T	E	P	K	L	K	Q	E	L	R	G	S	D	L	I	Q	V
Takifugu rubripes	L	V	K	A	E	T	R	Q	T	L	H	T	Y	S	L	Q	P	G	L	G	G	M	K	Y	H	T	G	W	C	S	V	W	T	R	T	E	P	K	M	K	R	E	L	R	G	S	D	P	V	E	V
Oryzias latipes	L	V	K	A	E	T	R	Q	T	L	H	T	Y	S	L	Q	P	G	L	G	G	M	K	Y	H	T	G	W	C	T	V	W	T	R	T	E	P	K	M	K	R	E	L	R	G	S	D	P	L	E	V
Danio rerio	L	V	K	A	E	T	R	H	T	L	H	T	Y	S	L	Q	P	G	L	G	G	M	K	F	H	T	G	W	C	T	V	W	T	R	T	E	P	K	M	K	R	E	L	M	G	S	D	P	V	E	V
Branchiostoma floridae	H	V	K	A	E	T	R	Q	T	L	H	T	Y	S	L	Q	P	G	L	G	G	M	R	F	Y	T	Y	W	C	T	V	W	T	R	T	E	P	K	M	K	Q	E	L	R	P	S	D	P	I	E	V
Ixodes scapularis	H	V	E	A	E	T	R	H	T	R	L	T	Y	D	L	R	P	G	L	G	G	L	V	V	N	A	V	W	C	S	V	W	T	R	T	E	P	K	L	R	K	K	V	R	N	P	S	K	G	S	L
Daphnia pulex	P	L	K	A	E	T	R	Q	T	L	L	T	Y	S	L	E	P	G	L	G	S	V	R	F	S	T	G	W	C	T	V	W	T	R	T	E	P	K	L	N	D	G	M	I	V	.	D	W	V	E	V
Nematostella vectensis	L	T	K	A	D	T	R	Q	T	L	H	T	Y	D	I	R	P	G	I	G	G	L	I	W	H	T	D	W	C	T	V	W	T	Q	T	A	P	K	I	K	N	E	L	K	G	S	P	M	E	V	.
Gallus gallus	P	V	K	A	E	T	R	N	T	L	H	T	Y	S	L	E	P	A	F	G	G	L	R	Y	K	T	V	F	C	T	I	W	T	R	T	E	P	V	L	K	K	.	.	.	T	D	P	V	E	I	
Ciona intestinalis	W	T	K	A	D	T	R	Q	T	L	H	T	Y	T	L	Q	P	G	L	G	G	L	K	F	Y	T	A	W	C	S	V	W	T	R	T	E	P	K	M	K	H	E	L	K	S	S	N	S	M	E	V
Ciona savignyi	W	T	K	A	D	T	R	Q	T	L	H	T	Y	S	L	R	P	G	L	G	G	L	K	F	Y	T	A	W	C	S	V	W	T	R	T	E	P	K	M	K	Q	E	L	K	S	S	N	S	M	E	V
Trichoplax adhaerens	A	T	K	A	D	I	R	Q	T	Q	H	T	Y	D	K	D	P	G	L	G	G	L	R	F	R	V	V	F	C	R	I	W	T	T	A	A	H	L	N	R	E	V	T	L	E	D	D	V	E	V	
Anopheles gambiae	P	V	K	A	E	T	R	Q	V	R	L	T	Y	S	T	E	P	G	L	G	G	L	M	V	N	S	A	W	C	K	V	W	T	R	T	E	P	K	L	D	A	E	L	K	K	S	N	D	M	E	V
Aedes aegypti	P	V	K	A	E	T	R	H	V	R	L	T	Y	S	T	E	P	G	L	G	G	L	M	V	N	S	A	W	C	T	V	W	T	R	T	E	P	K	L	D	A	E	L	K	H	S	D	T	L	E	V
Drosophila willistoni	P	A	K	A	E	T	R	H	L	M	L	S	Y	S	V	E	P	G	L	G	G	L	M	V	N	A	A	W	C	R	V	W	T	R	T	E	T	K	L	T	S	E	L	K	R	S	D	G	M	E	V
Drosophila melanogaster	P	A	K	A	E	T	R	H	L	M	L	S	Y	S	T	E	P	G	L	G	G	L	M	V	N	A	A	W	C	T	V	W	T	R	T	E	T	K	L	A	A	E	L	Q	R	S	D	G	M	E	V
Apis mellifera	P	V	K	A	E	T	R	Q	T	L	H	T	Y	S	I	K	P	G	L	G	G	L	M	V	N	A	A	W	C	T	V	W	T	R	T	E	P	Q	L	N	V	E	L	K	H	S	N	I	I	E	V
Monosiga brevicollis	S	S	H	V	D	T	R	Y	T	R	Q	T	Y	S	L	E	R	A	F	G	G	L	S	Y	H	V	A	W	L	D	V	W	T	R	T	E	P	N	L	R	Q	E	L	R	P	I	I	I	.	.	.
Caenorhabditis remanei	A	A	K	A	D	T	R	Y	T	S	H	T	Y	D	L	K	P	G	I	G	G	M	F	V	N	A	V	W	C	T	V	W	T	R	T	E	S	K	L	M	I	E	T	F	N	E	A	H	L	G	V
Caenorhabditis briggsae	A	A	K	A	D	T	R	Y	T	S	H	T	Y	D	L	K	P	G	I	G	G	M	F	V	N	S	I	W	C	T	V	W	T	R	T	E	S	K	L	M	V	E	T	F	N	E	S	H	L	G	V
Caenorhabditis elegans	A	A	R	A	D	T	R	Y	T	S	H	T	Y	D	L	K	P	G	I	G	G	M	F	V	N	A	V	W	C	T	V	W	T	R	T	E	P	K	L	S	I	D	T	F	N	E	A	H	L	G	V
Drosophila pseudoobscura	R	E	I	P	E	T	R	H	T	L	H	T	Y	D	L	G	P	G	F	A	D	Y	G	V	L	D	S	W	C	S	I	W	T	Q	T	K	K	K	F	G	P	K	I	K	A	S	D	H	T	I	L
Vitis vinifera	A	L	A	F	Y	N	R	Q	V	R	L	I	Y	S	L	E	P	A	M	A	Q	S	K	N	H	T	N	E	C	S	I	W	L	H	L	E	R	N	L	Y	P	R	L	D	V	V	L	I	K	.	
Arabidopsis thaliana	A	M	A	Y	Y	N	R	Q	T	R	L	I	Y	S	L	E	P	A	M	A	Q	S	K	N	H	T	N	E	C	S	I	W	L	H	L	D	L	D	V	Y	P	Q	A	L	Q	A	L	I	M	K	.
Oryza sativa	P	H	A	Y	Y	N	R	H	V	K	D	A	Y	S	A	D	P	A	I	V	G	T	K	Y	H	T	N	Q	C	T	V	W	F	E	L	E	P	Q	V	Y	P	I	L	D	A	V	I	T	.		

L87 V88 K90 A91 E92 V94 R95 Q98 T99 S101 L102 T198 Y199 S200 R201 E202 P217 G219 L220 G222 G223 L224 R225 F226 H240 T241 A242 W243 C301 T302 V304 W307 T309
 R310 T311 E312 P314 K315 M316 K317 Q318 E319 L322 R327 G328 S329 D330 P331 I333 E334 V335

C2GnT	KCTK	TPPS	SKYDLSDM	NF	DVS	NGAPYW	QH
Rattus_norvegicus	KSTK	TPPS	SKYDLSDM	NF	DVS	NGAPYWK	H
Oryctolagus_cuniculus	KSMQ	TPSL	SKYDLSDM	RF	DVS	KGAPYWK	H
Bos_taurus	KLME	TPSL	SKYDLSDM	QF	DVS	KGAPYV	H
Homo_sapiens	KLME	TPPA	SKYDLSDM	QF	DVS	KGAPYWK	H
Sus_scrofa	MLKE	TPSL	SKYDMSDM	HF	DVS	KGAPYWT	H
Canis_lupus	KSME	TPSL	SKYDMSDM	HF	DVS	KGAPYWK	H
Equus_caballus	KSME	TPSL	SKYDMSDM	HF	DIS	KGAPYWM	H
Ornithorhynchus_anatinus	KSRE	TPSS	SKYDVSDM	HF	DVS	KGAPYWK	H
Anolis_carolinensis	KAKN	TPSA	SKYDVSDM	NF	DVS	KGAPYWK	H
Taeniopygia_guttata	KAKS	TPSS	SKYDVSDM	NF	DVS	KGAPYWN	H
Meleagris_gallopavo	KAKN	TPSS	NKYDVSDM	NF	DVS	KGAPYWN	H
Monodelphis_domestica	KTME	SPSS	SKYDISDM	QL	DI	FKGAPYW	QP
Xenopus_(Silurana)	MGKE	TPPA	NKYDVSDM	NL	DVA	KGAPYF	KH
Tetraodon_nigroviridis	MLRN	LPRP	NKFDMTDL	NH	DGS	PD	AVYQ
Danio_rerio	SHNN	TPSP	NKYEQSDM	NH	DLN	S	GAPYQ
Ciona_intestinalis	SNYD	I	PPPH	KYDQNEL	QL	...	LVYQ
Branchiostoma_floridae	QVGN	V	PSK	...	PWSF	...	KQYCH
Trichoplax_adhaerens	YSHG	IRDN	TRNKRYN	MKS	EE
Nematostella_vectensis	ATNN	I	PV	GDP	PFSQS

K232 C235 T237 K279 T280 P330 P337 S338 S339 K341 Y342 D343 L344 S345 D346 M347 N348 F359 D362 V363 S364 N365 G366 A367 P368 Y369 W390 Q394 H395

Ext12	Q	F	S	I	Q	F	P	R	F	N	G	D	Q	Y	P	F	K	P	S	N	I	M	I	S	Q	F	G	F	Y	A	N	H	K			
Rattus_norvegicus	Q	F	S	I	Q	F	P	R	F	N	G	D	Q	Y	S	F	K	P	S	N	I	M	I	S	Q	F	G	F	Y	A	N	H	K			
Homo_sapiens	P	F	S	V	Q	F	P	R	F	N	G	D	Q	Y	T	F	K	P	S	N	I	M	I	S	Q	F	G	F	Y	A	N	H	K			
Bos_taurus	Q	F	S	V	Q	F	P	R	F	N	G	D	H	Y	T	F	K	P	S	N	I	M	I	S	Q	F	G	F	Y	A	N	H	K			
Sus_scrofa	Q	F	S	V	Q	F	P	R	F	N	G	D	H	Y	T	F	K	P	S	N	I	M	I	S	Q	F	G	F	Y	A	N	H	K			
Equus_caballus	Q	F	S	V	Q	F	P	R	F	N	G	D	Q	Y	T	F	K	P	S	N	I	M	I	S	Q	F	G	F	Y	A	N	H	K			
Monodelphis_domestica	H	F	S	V	Q	F	P	R	F	N	G	D	Q	Y	T	F	K	P	S	N	I	M	I	S	Q	F	G	F	Y	A	N	H	K			
Ornithorhynchus_anatinus	H	F	S	V	Q	F	P	R	F	S	G	D	Q	Y	T	F	K	P	S	N	I	M	I	S	Q	F	G	F	Y	A	N	H	K			
Anolis_carolinensis	Y	F	S	I	Q	F	P	R	F	N	G	D	Q	Y	P	F	K	P	S	N	I	M	I	S	Q	F	G	F	Y	A	N	H	K			
Meleagris_gallopavo	H	F	S	V	Q	F	P	R	F	N	G	D	Q	Y	P	F	K	P	S	N	I	M	I	S	Q	F	G	F	Y	A	N	H	K			
Taeniopygia_guttata	H	F	S	V	Q	F	P	R	F	N	G	D	Q	Y	P	F	K	P	S	N	I	M	I	S	Q	F	G	F	Y	A	N	H	K			
Xenopus_(Silurana)	Y	F	S	V	Q	F	P	R	F	T	G	D	M	Y	A	L	K	P	S	S	I	M	I	S	Q	F	G	F	Y	A	N	H	K			
Danio_rerio	P	F	S	V	Q	F	P	R	F	G	G	D	R	Y	P	F	K	P	S	N	I	M	I	T	Q	L	G	F	Y	A	N	H	K			
Tetraodon_nigroviridis	P	F	S	V	Q	F	P	R	F	G	G	D	S	N	L	M	V	S	Q	F	G	F	Y	A	N	H	M			
Nematostella_vectensis	D	F	E	A	E	F	G	R	W	T	N	D	V				
Ixodes_scapularis	D	F	E	V	E	F	S	R	W	T	N	D	V	A	K	V	I	E	F	R	L	F	K	D	N	F	K	L	K	R	F
Branchiostoma_floridae	D	F	E	V	E	F	G	R	W	T	N	D	V	A	K	V	V	E	H	R	L	Y	K	D	N	F	K	L	K	R	F
Apis_mellifera	D	F	E	I	E	F	S	R	W	T	N	S	I	A	K	V	V	E	F	R	L	F	K	D	I	F	K	L	K	R	F
Drosophila_melanogaster	D	F	E	V	E	F	S	R	W	T	N	Q	I	P	K	V	V	E	F	R	L	F	R	D	N	F	K	L	K	R	Y
Aedes_aegypti	D	F	E	V	E	F	S	R	W	T	N	Q	I	P	K	V	V	E	F	R	L	F	K	D	N	F	K	L	K	R	F
Anopheles_gambiae	D	F	E	V	E	Y	S	R	W	T	N	Q	I	P	K	V	V	E	F	R	L	F	K	D	N	F	K	L	K	R	F
Caenorhabditis_remanei	Q	L	R	V	E	N	A	R	H	T	C	Q	M	P	K	T	K	N	V	T	S	A	Y	D	C	S	F	M	G	I	T
Caenorhabditis_elegans	Q	L	R	V	E	N	A	R	H	T	C	Q	M	P	K	T	S	Q	F	R	L	F	K	T	R	L	N	H	Q	K	C
Caenorhabditis_briggsae	Q	L	R	V	E	N	A	R	H	T	C	Q	M	P	K	T	S	Q	F	R	L	F	K	T	R	L	N	H	Q	K	C
Trichoplax_adhaerens	D	F	R	V	E	S	A	R	K	T	C	Q	L	P	K	V	S	Q	F	R	L	Y	R	T	R	V	G	E	Q	K	C
Ciona_intestinalis	D	F	R	V	E	S	G	R	H	T	C	E	L	P	K	V	T	Q	F	R	L	F	K	T	R	L	D	K	Q	K	C
Vitis_vinifera	S	F	N	V	S	A	P	R	W	W	T	G	T	Y	P	T	S	M	K	A	V	D	S	R	S	F	W		
Populus_trichocarpa	S	F	K	V	S	A	P	R	W	W	T	G	T	Y	P	T	T	V	K	A	V	D	S	R	I	F	W		
Arabidopsis_thaliana	H	F	N	V	S	A	P	R	W	W	S	G	T	Y	P	T	S	M	K	A	V	D	S	R	N	F	W		
Monosiga_brevicollis	Q	M	R	V	G	H	A	R	V	A	C	H	P	Q	P	L	D	S	T	T	L	T	L	N	I	A	R	L	F	G	C	V	R			

Q159 F163 S166 I167 Q170 F171 P180 R181 F196 N205 G206 D207 Q208 Y209 P260 F264 K266 P267 S312 N313 I314 M315 I316 S317 Q318 F319 G320 F321 Y323 A324 N325 H326 K327

GalNT2	PD	T	V	N	N	Q	Y	V	G	A	S	A	D	L	W	D	Y	P	E	R	R	R	P	E	R	T	H	F	A	D	Y	E	N	H	M	D	L	V	K	Q	G	R	R
Danio rerio	PD	T	V	N	N	Q	Y	V	G	A	S	A	D	L	W	D	Y	L	E	R	R	R	P	E	R	T	H	F	A	D	Y	E	N	H	M	D	L	V	K	Q	G	R	R
Tetraodon nigroviridis	PD	T	V	N	N	Q	Y	V	G	A	S	A	D	L	W	D	Y	L	D	R	R	R	P	E	R	T	H	F	A	D	Y	E	N	H	M	D	L	V	K	Q	G	R	R
Xenopus (Silurana)	PD	T	V	N	N	Q	Y	V	G	A	S	A	D	L	W	D	Y	P	E	R	R	R	P	E	R	T	H	F	A	D	Y	E	N	H	M	D	L	V	K	Q	G	R	R
Branchiostoma floridae	PD	T	V	N	N	Q	Y	V	G	A	S	A	D	L	W	D	Y	A	N	R	N	R	P	E	R	T	H	F	A	D	Y	E	N	H	S	D	L	V	K	H	G	Q	K
Ixodes scapularis	PD	T	V	N	S	K	Y	F	G	A	S	S	D	L	W	E	F	N	K	R	E	R	P	E	K	T	G	S	E	G	F	T	N	H	G	G	L
Apis mellifera	PD	T	V	S	T	Q	Y	I	G	A	S	A	D	L	W	E	Y	Q	T	R	Q	R	P	E	V	S	H	L	L	D	Y	P	N	H	H	G	L	P	L	Q	I	D	
Drosophila willistoni	PD	T	V	S	N	Q	Y	I	G	A	S	A	D	L	W	E	Y	P	A	R	S	R	P	D	Q	T	H	L	I	D	F	P	N	H	D	D	L	V	V	K	S	D	
Drosophila melanogaster	PD	T	V	S	N	Q	Y	I	G	A	S	A	D	L	W	E	Y	P	S	R	A	R	P	D	Q	T	H	L	I	D	F	P	N	H	D	D	L	V	V	K	A	D	
Aedes aegypti	PD	T	V	S	T	Q	Y	I	G	A	S	A	D	L	W	E	Y	T	A	R	H	R	P	Q	T	H	L	V	D	Y	Q	N	H	L	D	L	V	K	Y	D			
Anopheles gambiae	PD	T	V	S	T	Q	Y	I	G	A	S	A	D	L	W	E	Y	N	A	R	K	R	P	Q	G	S	N	V	A	G	Y	S	N	H	H	D	L	V	L	K	Y	D	
Nematostella vectensis	PD	T	V	N	D	S	Y	I	G	A	S	A	D	I	W	D	N	P	E	K	Q	R	P	E	Q	T	S	Q	A	G	F	D	N	H	L	D	L	T	E	G	R	K	
Daphnia pulex	PD	Y	V	A	S	Q	Y	I	A	A	S	T	E	L	W	E	L	A	E	K	A	R	P	E	T	T	R	G	A	G	Y	A	N	H	G	S	W	P	I	L	P	S	
Trichoplax adhaerens	PD	T	V	H	T	N	Y	I	G	S	A	D	L	W	D	S	S	E	Q	S	R	P	E	N	S	G	K	P	G	F	P	N	N	I	Q	R	V	D	P	E	K		
Pongo abelii	RD	T	V	S	N	A	Y	L	A	A	S	A	D	L	W	E	Q	L	E	K	M	R	P	E	T	S	Q	N	T	A	G	I	N	Q	Q	G	K	T	I	Q	M	N	K
Caenorhabditis briggsae	PD	S	V	N	N	N	Y	V	G	A	S	A	D	L	W	E	F	E	E	R	K	R	P	Q	N	S	R	K	E	N	F	A	N	N	S	Q	M	S	V	V	K	E	
Caenorhabditis elegans	PD	S	V	N	N	N	Y	V	G	A	S	A	D	L	W	E	F	E	Q	R	K	R	P	Q	E	S	R	K	E	S	F	G	N	N	S	Q	L	S	T	V	K	E	
Monosiga brevicollis	PD	V	V	S	N	R	Y	.	S	A	S	P	V	V	W	K	S	.	.	R	S	R	P	E	R	S	K	S	V	G	Y	R	N	H	E	D	A	H	T	R	S	E	P
Caenorhabditis remanei	PD	Y	V	S	T	E	Y	V	T	A	S	E	T	T	W	Y	A	K	R	L	N	R	P	E	P	T	K	K	D	G	Q	G	N	S	D	D	L	G	K	E	R	S	N
Ciona intestinalis	PD	V	V	S	T	E	F	M	V	G	S	D	M	T	W	Y	P	Q	R	M	D	H	P	D	Q	T	R	K	E	N	F	N	C	D	D	I	S	M	V	K	H	N	
Cryptosporidium parvum	SD	Y	S	D	T	E	F	V	N	G	G	I	G	C	F	P	Q	S	P	P	R	R	P	E	E	T	N	N	P	G	W	S	T	H	W	N	K	D	W	N	.	.	.

P119 D120 T121 V255 N257 N260 Q262 Y263 V264 G265 A266 S267 A268 D269 L270 W282 D283 Y284 P287 E288 R290 R291 R356 P435 E436 R438 T459 H462 F463 A464 D465
Y471 E472 N479 H492 M493 D494 L495 V500 K509 Q511 G512 R514 R519

GTB	E	F	M	V	S	L	P	R	M	V	Y	P	Q	P	K	V	L	T	P	C	R	K	D	V	L	V	V	T	P	W	L	P	I	V	W	T	F	N	I	L	Q	E	P	L	L	Y	G	S	S	R	F	P	D	E	G	D	F	Q	E	R	L	R	H	K	T	K	V
Oryctolagus_cuniculus	.	.	.	A	V	S	C	R	M	L	Y	P	Q	P	K	V	L	T	P	S	R	S	D	V	L	V	L	T	P	W	L	P	I	V	W	T	F	S	I	L	Q	E	P	L	L	Y	S	A	D	R	F	P	D	E	G	D	F	A	E	R	L	Y	H	K	T	K	V
Rattus_norvegicus	Q	K	V	V	S	V	P	R	M	A	Y	P	Q	P	N	V	L	T	P	I	R	N	D	V	L	V	F	T	P	W	L	P	I	I	W	T	F	N	I	L	Q	E	A	L	L	Y	R	S	R	R	F	P	D	E	G	D	F	V	E	H	L	Y	H	K	T	K	I
Mus_musculus	A	V	T	R	N	A	Y	L	Q	P	R	V	L	K	P	T	R	K	D	V	L	V	L	T	P	W	L	P	I	I	W	T	F	N	I	L	Q	E	T	F	L	Y	S	S	S	R	F	P	D	R	G	D	F	L	E	H	L	Y	H	K	T	K	I
Bos_taurus	C	R	I	P	E	V	P	R	L	L	Y	P	K	A	Q	L	L	K	P	L	R	V	D	V	L	V	M	T	P	W	F	P	V	V	W	T	F	D	L	L	Q	E	P	L	L	Y	A	A	D	R	F	P	D	E	G	D	F	P	E	R	L	S	H	K	S	K	L
Xenopus_(Silurana)	L	T	E	V	K	L	E	R	M	L	Y	P	K	P	E	T	L	K	P	P	R	T	D	V	L	T	I	T	P	W	L	P	I	V	W	T	Y	N	V	L	Q	E	D	V	L	Y	G	A	G	R	F	P	D	E	G	D	F	E	E	K	L	Y	H	K	T	K	I
Pongo_abelii	K	P	L	Q	P	V	V	W	S	Q	Y	P	Q	P	K	L	L	E	H	R	P	T	Q	L	L	T	L	T	P	W	L	P	I	V	S	T	F	N	L	L	I	E	D	L	I	Y	A	V	P	H	F	A	S	E	G	D	F	A	R	E	I	S	N	K	S	K	X
Taeniopygia_guttata	K	L	I	Q	L	F	P	Q	L	F	Y	Q	Q	P	R	V	L	A	P	K	R	Q	D	V	L	T	V	T	P	W	L	P	I	I	W	T	F	D	I	L	A	E	D	T	I	F	D	V	P	R	F	P	G	E	G	D	F	R	K	E	L	S	H	K	S	K	E
Meleagris_gallopavo	K	P	V	Q	L	F	P	Q	L	F	Y	Q	Q	P	R	V	L	A	P	K	R	Q	D	V	L	T	V	T	P	W	L	P	I	I	W	T	F	S	I	L	V	E	D	I	M	F	N	V	P	R	F	P	G	E	G	D	F	K	K	E	L	S	H	K	S	K	E
Gallus_gallus	K	P	V	Q	L	F	P	Q	L	F	Y	Q	Q	P	R	V	L	A	P	K	R	Q	D	V	L	T	V	T	P	W	L	P	I	V	W	T	F	S	I	L	A	E	D	M	I	F	N	V	P	R	F	P	G	E	G	D	F	K	K	E	L	T	H	K	S	K	E
Danio_rerio	R	S	L	K	T	S	P	G	F	Q	Y	K	Q	P	S	L	L	A	.	G	R	A	D	V	V	S	L	S	P	W	L	P	I	I	W	T	F	N	L	I	I	E	N	L	I	F	D	Y	P	R	F	P	G	E	G	D	Y	E	E	R	L	Y	N	K	T	K	Q
Sus_scrofa	D	N	R	G	E	L	P	L	V	D	W	F	N	P	E	K	R	P	E	V	V	T	I	T	R	W	K	P	V	V	W	T	Y	N	V	L	Y	E	Q	S	L	Y	K	A	H	P	F	P	G	Q	G	D	F	T	Q	N	L	L	N	K	T	K	D
Tetraodon_nigroviridis	K	S	G	A	D	F	G	G	A	F	F	P	T	R	S	R	G	D	V	Q	T	C	T	P	W	K	P	I	I	W	M	F	D	L	Y	T	E	E	S	L	Y	R	L	P	K	Y	E	.	.	G	D	F	R	W	A	L	L	H	K	S	R	D	

E61 F62 M63 V64 S65 L66 P67 R68 M69 V70 Y71 P72 Q73 P74 K75 V76 L77 T78 P79 C80 R81 K82 D83 V84 L85 V86 V87 T88 P89 W90 L91 P93 I94 V95 W96 T99 F100 N101 I104 L105 Q108 E223 P227 L228 L232 Y237 G238 S239 S240 R241 F244 P257 D259 E260 G261 D262 F263 Q275 E276 R279 L311 R312 H313 K314 T316 K317 V335

GTA	E F M V S L P R M V Y P Q P K V L P C K D V L V V T P W L P I V W T F N I L Q E L Y G S S R F P D E G D F L R H K K V
Oryctolagus_cuniculus C R M L Y P Q P K V L P S S D V L V L T P W L P I V W T F S I L Q E L Y S A D R F P D E G D F L Y H K K V
Rattus_norvegicus	V L V V S V P R M A Y P Q P N V L P I N D V L V F T P W L P I I W T F N I L Q E L Y R S R R F P D E G D F L Y H K K I
Mus_musculus	. V L V A V T R N A Y L Q P R V L P T K D V L V L T P W L P I I W T F N I L Q E L Y S S S R F P D R G D F L Y H K K I
Bos_taurus	L G F P E V P R L L Y P K A Q L L P L V D V L V M T P W F P V V W T F D L L Q E L Y A A D R F P D E G D F L S H K K L
Xenopus_(Silurana)	C G V V K L E R M L Y P K P E T L P P T D V L T I T P W L P I V W T Y N V L Q E L Y G A G R F P D E G D F L Y H K K I
Pongo_abelii	. . . Q P V V W S Q Y P Q P K L L H R T Q L L T L T P W L P I V S T F N L I I E I Y A V P H F A S E G D F I S N K K X
Taeniopygia_guttata	S G K Q L F P Q L F Y Q Q P R V L P K Q D V L T V T P W L P I I W T F D I L A E I F D V P R F P G E G D F L S H K K E
Meleagris_gallopavo	N W K Q L F P Q L F Y Q Q P R V L P K Q D V L T V T P W L P I I W T F S I L V E M F N V P R F P G E G D F L S H K K E
Gallus_gallus	N W K Q L F P Q L F Y Q Q P R V L P K Q D V L T V T P W L P I V W T F S I L A E I F N V P R F P G E G D F L T H K K E
Danio_rerio	L S G K T S P G F Q Y K Q P S L L G . A D V S L S P W L P I I W T F N L I I E I F D Y P R F P G E G D Y L Y N K K Q
Sus_scrofa	F W I G E L P L V D W F N P E . . . K P E V V T I T R W K P V V W T Y N V L Y E L Y K A H P F P G Q G D F L L N K K D
Tetraodon_nigroviridis G G A F F P T R . . . S G D V Q T C T P W K P I I W M F D L Y T E L Y R L P K Y E . R G D F L L H K R D

E61 F62 M63 V64 S65 L66 P67 R68 M69 V70 Y71 P72 Q73 P74 K75 V76 L77 P79 C80 K82 D83 V84 L85 V86 V87 T88 P89 W90 L91 P93 I94 V95 W96 T99 F100 N101 I104 L105 Q108
E223 L232 Y237 G238 S239 S240 R241 F244 P257 D259 E260 G261 D262 F263 L311 R312 H313 K314 K317 V335

XXYLT1	D	N	F	L	P	G	R	E	A	R	Q	S	P	L	S	H	W	Q	E	H	E	L
Equus_caballus	D	N	F	L	P	G	R	E	A	R	Q	S	P	L	S	R	Q	Q	E	H	E	L
Xenopus_(Silurana)	D	N	F	M	H	M	K	E	A	R	Q	S	R	L	N	E	A	Q	E	H	E	L
Meleagris_gallopavo	D	N	F	Q	E	G	R	E	A	R	R	S	K	L	N	Q	M	K	E	H	E	L
Taeniopygia_guttata	D	N	F	P	E	G	K	E	A	R	Q	S	K	L	N	Q	M	K	E	H	E	L
Monodelphis_domestica	D	R	F	P	P	G	R	E	A	R	Q	S	A	L	R	R	A	R	E	H	E	L
Anolis_carolinensis	D	R	F	P	E	G	R	P	A	R	A	S	G	L	N	G	P	A	E	H	G	L
Danio_rerio	N	H	F	P	S	D	R	D	S	R	R	S	T	V	N	Q	R	R	E	H	E	L
Trichoplax_adhaerens	H	E	F	Q	P	N	R	E	R	K	S	K	L	G	Q	L	N	E	Y	D	M	
Branchiostoma_floridae	D	H	F	E	S	D	K	D	K	R	G	S	R	L	N	S	V	A	R	Y	Q	L
Nematostella_vectensis	K	K	F	E	S	S	M	E	N	R	R	S	K	T	N	Q	A	D	E	H	D	L
Drosophila_melanogaster	D	N	F	L	P	H	S	N	R	R	N	S	K	S	L	E	E	T	E	Y	N	L
Drosophila_pseudoobscura	D	H	F	L	P	H	S	N	R	R	N	S	K	S	L	E	E	T	E	Y	N	L
Drosophila_willistoni	D	N	F	L	P	H	S	N	R	R	E	S	K	T	V	E	E	R	E	Y	N	L
Anopheles_gambiae	D	Q	F	A	P	D	S	D	R	R	R	S	R	I	E	E	T	N	E	F	G	L
Aedes_aegypti	N	R	F	S	S	D	L	D	R	Q	S	R	L	E	E	S	N	E	F	G	L	
Daphnia_pulex	E	Y	F	D	D	Q	L	D	K	R	Q	S	V	I	Q	S	S	N	E	Y	Q	L
Ixodes_scapularis	A	L	F	P	E	T	R	H	R	A	L	S	A	R	N	G	Y	E	R	E	L	

D242 N243 F244 L245 P246 G247 R275 E297 A298 R300 Q301 S302 P303 L304 S306 H307 W313 Q316 E339 H340 E342 L343

ST8Sia-III	GLHHKYVS	NNNNLL	TI	RHAT	RTDVE		
Anolis_carolinensis	GLHHKYVS	SNSNNLL	TI	RHAT	RTDIE		
Xenopus_(Silurana)	GLHHKYVS	SNNNNLL	TI	RHAS	RTDVE		
Takifugu_rubripes	GLHHKYVS	GENNNLL	TV	RHAT	RTDVE		
Gasterosteus_aculeatus	GLHHKYVS	GENNNLL	TV	RHAT	RTDVE		
Danio_rerio	GLHHKYVS	GENNNLL	TI	RHAT	RTDVE		
Oryzias_latipes	GLHHKYVS	SNNNNLL	TI	RHAT	RTDVE		
Tetraodon_nigroviridis	GLHHKYVS	SNNNNLL	TI	RHAT	RTDVE		
Branchiostoma_floridae	GVH	QKYP	SA	SNNLQ	TI	RHNM	RTDLE

G124 L126 H128 H134 K135 Y136 V137 S139 S141 N142 N143 N220 N221 L222 L223 T224 I225 R228 H251 A254 T255 R258 T259 D262 V265 E266

B4GalT7	GS	FFFFEELLVNVH	TA	LS	VG	VN
Monodelphis domestica	PR	FFFFEELLVNVH	TL	VS	VA	VN
Taeniopygia guttata	PG	FFFFEELLVNVH	TD	LS	VA	VN
Anolis carolinensis	PR	FFFFEELLVNVH	TS	LS	VA	VN
Xenopus (Silurana)	PR	FFFFEELMINVH	SE	VT	IS	VN
Takifugu rubripes	ER	FFFFEELLVNVH	QE	VV	IG	VN
Tetraodon nigroviridis	ER	FFFFEELLVNVH	QE	VL	VG	VN
Danio rerio	SR	FFFFEELLVNVH	KE	VS	IS	VN
Nematostella vectensis	..	YNFEELLVNVH	RQ	LM	ID	IN
Branchiostoma floridae	RR	FDFEELLVNIH	HNM	KE	VE	IN
Aedes aegypti	IQ	FDFEELLVNNH	REL	T	ID	VN
Drosophila melanogaster	VN	FDFEELLVNVH	HEM	L	ID	IN
Drosophila pseudoobscura	VN	FDFEELLVNVH	QGL	T	IE	VN
Drosophila willistoni	VS	FDFEELLVNVH	HEL	Q	MD	VN
Daphnia pulex	..	FDFEELLVNVH	SS	LH	ID	LN
Ixodes scapularis	TR	FDFEELLVNVH	HRL	V	ID	VD
Caenorhabditis remanei	KAY	DL	EE	E	INT	HRL
Caenorhabditis elegans	KT	YDL	EE	L	RINT	HQL
Caenorhabditis briggsae	KAY	DF	EE	L	RINT	HRL
Ciona savignyi	SR	YG	FDE	LLI	HVV	VAME
Trichoplax adhaerens	MR	FDF	DEL	MVNVH	HQ	LTV
Monosiga brevicollis	EQ	FGEA	EL	KVNV	R	T

G77 S78 F101 E103 F105 E106 E107 L108 L109 V130 N132 V134 H259 T298 A299 L300 S301 V302 G303 V309 N311

Gph1	IFSLRIRWTHQ	VEQDRFID	HET	TLAR	SLYNCDMM	RDVI	WNQDEL	LCFYIFE	T	PYNS	GIERN	EPRT	EF	DLN	FNN	GYVQ	VYPN	FQ	GLPGK		
Candida_glabrata	EDIKRAL.WHQ	VDQDRFID	HET	TLGR	SLYNCDLM	RDVI	WNQDEL	LCFYIFE	T	PYNF	GIERN	EPRT	EF	DFA	FNN	GYVQ	VYPN	FQ	GLPGK		
Kluyveromyces_lactis	ENI.QREIWHG	VDQSKFIQ	HET	TLAR	SLYNCDLQ	RDVI	WNQDEL	LCFYIFE	T	PYNF	AIERS	EPRT	EF	DFA	FNN	GYVQ	VYPN	FQ	GLPGK		
Eremothecium_gossypii	EDI.QRELWHE	VEQGKFQI	HET	TLAR	SLYNCDFE	RDVI	WNQEQ	LCFYIFE	T	PYNF	FRIERS	EPKT	EF	DFS	FNS	GYVQ	VYPN	FQ	GLPGK		
Candida_albicans	QDISKHIWYSQ	LDEEEFVK	HET	SLGR	SMYNCDLN	RDVI	WAQSD	LCFYIFE	T	PYNY	SIDRN	EPKT	EF	DFS	FNA	GYVQ	VYPN	FQ	GLPGK		
Lodderomyces_elongisporus	QDISKHIWFSQ	LQEEEFVR	HET	NLGR	SMYNCDLK	RDII	WAQNEL	LCFYIFE	T	PYKY	TLDL	RH	EPKN	EF	DFT	FNA	GYVQ	VYPN	FQ	GLPGK	
Scheffersomyces_stipitis	QDISKHIWYSQ	LDEESFVK	HET	TLAR	NMYNCDLN	RDII	WAQTEL	LCFYIFE	T	PYKY	SIMRS	EPKE	EF	DFT	FNA	GYVQ	VYPN	FQ	GLPGK		
Debaryomyces_hansenii	QDISKKIWKTH	LEEEEFIK	HEI	TLAR	NMYNCDLN	RDII	WAQREL	LCFYIFE	T	PYKY	TINRS	EPKN	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Yarrowia_lipolytica	QDIERQIWFS	LQQQEFVR	HET	SLAR	SLYNCDLQ	RDVI	WNQKDF	LCFYIFE	T	PYTY	GIPRN	EPKQ	EF	DFA	FNA	GYVQ	VYPN	FQ	GLPGK		
Neurospora_crassa	EEIPRKAWLHQ	TDETEVVR	HET	TLAR	SMYNCDDL	RDII	WNQDEL	LCFYIFE	T	PYDF	F.PRH	DPKS	EF	DFQ	FNS	GYVQ	VYPN	FQ	GLPGK		
Aspergillus_fumigatus	EEIPREAWH.S	ADEKELVR	HET	TLAR	SLYNCDLL	RDII	WNQKDL	LCFYIFE	T	PYDF	F.PRH	DPKS	EF	DFQ	FNS	GYVQ	VYPN	FQ	GLPGK		
Cryptococcus_neoformans	SPWGEKTWRKK	AQANTIVR	HNT	SLGR	QVYNVDVL	RDII	WNARK	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Laccaria_bicolor	TKWGEKAWMR	SSSTRSVVN	HQT	SLAR	QPYNLDLF	RDII	WNMKN	LCFYIFE	T	PYTY	QPLRL	DMKR	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPAK		
Dictyostelium_discoideum	ITVTKKKGSFA	LDQKDILD	HEY	TLAR	TKYNDFY	RDII	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Populus_trichocarpaFMRFLK	SSQKDILD	HEY	TVAR	SRFSFDFH	RDII	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Vitis_viniferaFMRFLS	SNVQKDILD	HEY	TVAR	SRFSFDFH	RDII	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Ostreococcus_'lucimarinus'MRYLRN	NMQESIVN	HEY	TMAR	NRQYDFD	RDII	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Cryptosporidium_parvum	GDAREKLWYES	YEQRSIVN	HEY	TLAR	TRFNDFY	RDII	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Oryza_sativa	.EGCAAEEKVAA	SAAGNISF	HQY	SPHF	SPLAFGEE	RDVI	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLATK		
Sorghum_bicolor	..ECAAAEKVAA	SAAGNISY	HQY	NPHF	SPLAFGEE	RDII	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLATK		
Arabidopsis_thaliana	.AKTLPEKIKAN	TAGNIVY	HKY	SPHF	SPLKFGEE	RDII	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLATK		
Ornithorhynchus_anatinus	RDII	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Homo_sapiens	.ALQKRKQIRG	IAAKSFFNR	HFF	TLVK	DRNVATRH	RDVI	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Gallus_gallus	.SLQRRKQIRG	IAAKGFFNR	HFF	TLVK	DRNVATRH	RDVI	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Xenopus_(Silurana)	.ALQKRKQIRG	IAAKGFFNR	HFF	TLVK	DRNVATRH	RDVI	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Anolis_carolinensis	RDII	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Meleagris_gallopavo	.SMVKGRATIS	LMCHILIS	NGG	ALGE	KPGSALNS	RQA	YQLGL	LFIFE	EADRY	GKARP	EPKN	DF	NLQ	FNV	GYVQ	VYPN	FQ	GLPGK		
Danio_rerio	.SLHRRKQIRG	IAAKSFFNR	HFF	TLVK	DRNVATRH	RDVI	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Rattus_norvegicus	.SLQKRKQIRG	LSKKNFNR	HFF	TLVK	DRNVATRH	RDVI	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Ciona_intestinalis	TSVQKRKQIRG	ISKKSFFNR	HHY	TLVK	DRNVATRH	RDVI	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Anopheles_gambiae	.SQVKRKQIRG	INKKDFNR	HHY	TLVK	DRNVATRH	KDV	WI	QYQL	GLYIFE	EP	DRY	GKARP	EPKI	DF	NLKF	FND	GYVQ	VYPN	FQ	GLPGK	
Aedes_aegypti	.SQEKRKQIRG	INKRGFFNR	HHY	TLVK	DRNVATRH	KDV	WI	QYQL	GLYIFE	EP	DRY	GKARP	EPKI	DF	NLKF	FND	GYVQ	VYPN	FQ	GLPGK	
Drosophila_melanogaster	.SQARRKQIRG	ITKKNFNR	HHY	TLVK	DRNVATRH	KDV	WI	QYQL	GLYIFE	EP	DRY	GKARP	EPKI	DF	NLKF	FND	GYVQ	VYPN	FQ	GLPGK	
Ixodes_scapularis	.AAHKKRKQIRG	IVKKAFFNR	HHY	TLVK	DRNVATRH	KDV	WI	QYQL	GLYIFE	EP	DRY	GKARP	EPKI	DF	NLKF	FND	GYVQ	VYPN	FQ	GLPGK	
Branchiostoma_floridae	.SQHKKRKQIRG	ITKKSFFNR	HFF	TLVK	DRNVATRH	RDVI	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Caenorhabditis_elegans	TSTHRRKQIRG	ISKKAFFNR	HFF	SIK	DRNVATRH	RDVI	WNQKDL	LCFYIFE	T	PYTY	REIARL	DPKQ	EF	DFN	FNA	GYVQ	VYPN	FQ	GLPGK		
Nematostella_vectensis	.ELAKKRQIRR	LNKEAFFNR	HHY	SLVK	DRNVATRH	KDV	WI	QYQL	GLYIFE	EP	DRY	GKARP	EPKI	DF	NLKF	FND	GYVQ	VYPN	FQ	GLPGK	
Trichoplax_adhaerens	ASFVRRKQIRE	IQKKSFFNR	HHY	TELK	DRNNATRH	REAS	WI	QYEL	GLYIFE	EP	DRY	GKARP	EPKI	DF	NLKF	FND	GYVQ	VYPN	FQ	GLPGK	
Monosiga_brevicollis	SRDLFN	HHC	TLAK	DVSVA TRY	RDM	WI	HRYN	MGLYIFE	EP	DRY	GKARP	EPKI	DF	NLKF	FND	GYVQ	VYPN	FQ	GLPGK	
Giardia_intestinalis	IQGISLQFHFO	VDQSNII	SEKY	HLGR	DSTTIDFI	RNI	DI	WRQ	KDL	LCFYIFE	EP	DRY	GKARP	EPKI	DF	NLKF	FND	GYVQ	VYPN	FQ	GLPGK

I3 F8 S9 L11 R12 L12 R13 W13 T14 H16 Q17 V18 E25 Q28 D29 R30 F31 I32 D33 H34 E36 T37 T38 L39 A40 R41 S42 L43 Y44 N45 C46 D47 M49 M57 R60 D61 V64 I65 W67 N68 Q72 D113
E114 L115 G116 F117 Y163 I165 F166 E177 T178 P179 Y181 N184 S185 G186 I191 E192 R193 N194 E195 P229 R247 T250 E251 F252 D253 L254 N255 F257 N258 N259 G260 Y262 V266
Q269 V278 Y280 P281 N282 F285 Q287 G288 L291 P611 G612 K617

Pygm	KHF	TLVK	DNVTRDF	RDV	GWI	QEF	GFNEE	ADRY	YKRP	EMKN	YI	VL	RN	EN	RV	LY	PLP																																	
Danio_rerio	THF	TLVK	DNVTRDF	RDV	GWI	QEF	GFNEE	ADRY	YKRP	ELKC	YI	VL	RN	EN	RV	LY	PLP																																	
Gallus_gallus	KHF	TLVK	DNVTRDF	RDV	GWI	QEF	GFNEE	ADRY	YKRP	EMKN	YI	VL	RN	EN	RV	LY	PLP																																	
Meleagris_gallopavo	HGGALGEK	GS	L	NPR	QEF	GFNEE	ADRY	YKRP	EMKN	YI	VL	RN	EN	RV	LY	PLP																																
Anolis_carolinensis	E	GFNEE	ADRY	YKRP	EMKN	YI	VL	RN	EN	RV	LY	PLP																																
Ornithorhynchus_anatinus	M	NSAGEY	GFNEE	ADRH	KRP	ELRN	YI	VL	RN	EN	RV	LY	PLP																																
Xenopus_(Silurana)	KHF	TLVK	DNVTRDF	RDV	GWI	QEY	GFNEE	ADRH	KRP	EMRN	YI	VL	RN	EN	RV	LY	PLP																																	
Ixodes_scapularis	KHY	TLVK	DNVTRDF	KDV	GWI	QEY	GF	SKEP	DRY	YKRP	EMKV	YI	VL	RN	EN	RV	LY	PLP																																
Drosophila_melanogaster	KHY	TLVK	DNVTRDF	KDV	GWI	QEY	GF	AEEP	DRY	YKRP	EMKI	YI	VL	RN	EN	RV	LY	PLP																																
Aedes_aegypti	RHY	TLVK	DNVTRDF	KDV	SWI	QEY	GF	AEEP	DRY	YKRP	EMKI	YI	VL	RN	EN	RV	LY	PLP																																
Anopheles_gambiae	KHY	TLVK	DNVTRDF	KDV	SWI	QEY	GF	AEEP	DRY	YKRP	EMKI	YI	VL	RN	EN	RV	LY	PLP																																
Ciona_intestinalis	KHY	TLVK	DNVTRDF	RDV	GWI	QEY	GFNEE	ADRY	YKRP	EMKN	YI	V	C	R	N	EN	RV	LY	PLP																															
Branchiostoma_floridae	KHF	TLVK	DNVTRDF	RDV	GWI	QDY	GFTEEP	DQY	YKRP	EMKK	YI	V	C	R	N	EN	RV	LY	PLP																															
Caenorhabditis_elegans	KHF	SI	IK	DNVTRDF	RDV	SWI	QEY	GF	KEEP	DRF	FKRP	EMKN	YV	V	M	R	N	EN	RV	LY	PLP																													
Nematostella_vectensis	EHY	SLVK	DNVTQDF	KDV	GWI	QEY	GF	KEH	PER	F	KRP	EMKK	YI	V	C	R	N	EN	RV	LY	PLP																													
Trichoplax_adhaerens	KHY	TE	LK	DNNTDRF	REAS	WI	QEY	GFTEEP	DRY	YKRP	EMKK	YV	V	C	R	N	EN	RV	LY	PLP																														
Monosiga_brevicollis	DHC	TLAK	DSVTRDL	RDMS	WH	REY	GF	EELP	DK	F	V	R	P	E	V	R	N	E	R	M	C	L	Y	PLP																										
Arabidopsis_thaliana	GKY	SP	HF	SLKGEQY	RD	I	Q	NNVRH	GF	KEI	P	DE	KI	R	H	D	L	K	A	Y	E	A	Q	H	S	Q	T	V	L	Y	P	L	A																	
Sorghum_bicolor	GQY	N	P	H	F	S	L	A	G	E	Q	Y	RD	I	Q	NNLR	Y	GF	K	E	F	A	D	D	K	I	R	H	D	L	K	A	Y	E	A	Q	H	A	Q	A	V	L	Y	P	L	A				
Oryza_sativa	GQY	SP	HF	S	L	A	G	E	Q	Y	RD	V	Q	NNLR	Y	GF	K	E	I	A	D	E	K	I	R	H	D	L	K	A	Y	E	A	Q	H	A	Q	A	V	L	Y	P	L	A						
Cryptosporidium_parvum	REY	TL	AR	T	F	N	D	N	A	R	RD	I	E	L	N	E	T	Y	GF	E	H	P	Y	V	Q	I	R	Q	D	V	T	R	Y	V	V	C	R	E	Y	S	V	L	Y	P	L	P				
Debaryomyces_hansenii	EEI	TL	AR	N	Y	N	D	L	G	Q	RD	I	I	W	A	Q	Q	Y	GF	E	Q	P	Y	E	Y	I	R	S	E	V	K	N	Y	Q	V	A	Q	E	S	S	V	L	Y	P	L	P				
Scheffersomyces_stipitis	EET	TL	AR	N	Y	N	D	L	A	Q	RD	I	I	W	A	Q	Q	Y	GF	K	E	T	P	Y	K	Y	I	R	S	E	V	K	E	Y	Q	V	G	Q	E	S	S	V	L	Y	P	L	P			
Lodderomyces_elongisporus	EET	N	L	G	R	S	Y	N	D	L	A	Q	RD	I	I	W	A	Q	Q	Y	GF	K	E	T	P	Y	K	Y	L	R	H	E	V	K	N	Y	Q	V	A	Q	E	S	A	V	L	Y	P	L	P	
Candida_albicans	EET	S	L	G	R	S	Y	N	D	L	A	Q	RD	V	I	W	A	Q	Q	Y	GF	K	E	T	P	Y	N	Y	I	R	N	E	V	K	T	Y	Q	V	A	Q	E	S	S	V	L	Y	P	L	P	
Eremothecium_gossypii	GET	TL	AR	S	Y	N	D	F	A	Q	RD	V	I	W	N	Q	Q	Y	GF	S	E	T	P	Y	N	F	I	R	S	E	V	K	T	Y	S	V	A	Q	E	S	A	V	L	Y	P	L	P			
Kluyveromyces_lactis	SET	TL	AR	S	Y	N	D	L	A	Q	RD	V	I	W	N	Q	Q	Y	GF	A	E	T	P	Y	N	F	I	R	S	E	V	R	T	Y	K	V	E	Q	E	S	A	V	L	Y	P	L	P			
Candida_glabrata	DET	TL	G	R	S	Y	N	D	L	A	E	RD	I	I	W	N	Q	Q	Y	GF	A	E	T	P	Y	N	F	I	R	N	E	V	R	T	Y	K	V	G	Q	E	S	A	V	L	Y	P	L	P		
Saccharomyces_cerevisiae	DET	TL	AR	S	Y	N	D	M	A	E	RD	V	I	W	N	Q	E	Y	GF	A	E	T	P	Y	N	S	I	R	N	E	V	R	T	Y	K	V	A	Q	E	S	A	V	L	Y	P	L	P			
Yarrowia_lipolytica	QET	S	L	A	R	S	Y	N	D	L	A	Q	RD	V	V	N	Q	E	Y	GF	K	E	Q	P	Y	T	Y	I	R	N	E	V	K	Q	Y	I	V	S	Q	E	T	A	V	L	Y	P	L	P		
Neurospora_crassa	TET	TL	AR	S	Y	N	D	Q	A	S	RD	I	L	W	N	Q	R	Y	GF	K	E	V	P	Y	D	F	F	R	H	D	V	K	S	Y	E	V	A	Q	E	T	A	V	L	Y	P	L	P			
Aspergillus_fumigatus	KET	TL	AR	S	Y	N	D	L	A	S	RD	I	I	W	N	Q	R	Y	GF	K	E	I	P	Y	D	F	F	R	H	D	V	K	S	Y	E	V	A	Q	E	T	A	V	L	Y	P	L	P			
Cryptococcus_neoformans	NNT	S	L	G	R	Q	Y	N	D	V	A	Q	RD	L	D	W	N	A	N	Y	GF	K	E	A	P	D	R	I	R	L	D	V	K	Q	Y	E	V	A	S	E	N	R	V	L	Y	P	L	P		
Laccaria_bicolor	RQT	S	L	A	R	Q	Y	N	D	F	G	Q	RD	L	V	W	N	M	K	Y	GF	Q	E	A	P	P	E	N	L	R	L	D	V	K	R	Y	E	V	E	S	N	D	A	S	V	L	Y	P	L	P
Dictyostelium_discoideum	KEY	TL	AR	T	Y	N	D	F	S	Q	RD	I	E	W	N	Q	N	Y	GF	E	E	V	P	Y	V	A	I	R	L	D	I	K	K	Y	L	V	E	K	Q	E	N	S	V	L	Y	P	L	P		
Ostreococcus 'lucimarinus'	EY	T	M	A	R	N	Y	Q	D	F	E	N	RD	I	E	W	N	Q	Q	Y	GF	R	E	H	P	Y	N	F	I	R	P	Y	V	K	R	Y	V	I	L	K	Q	E	T	S	V	L	Y	P	L	P
Populus_trichocarpa	KEY	T	V	A	R	S	F	S	D	F	E	Q	RD	I	E	W	H	L	Q	Y	GF	R	E	Q	P	Y	N	F	I	R	V	H	V	K	D	Y	I	V	V	R	Q	E	T	S	V	L	F	P	L	P
Vitis_vinifera	KEY	T	V	A	R	S	F	S	D	F	E	Q	RD	I	E	W	H	Q	Q	Y	GF	R	E	Q	P	Y	N	F	I	R	V	H	V	K	G	Y	I	V	V	R	Q	E	T	C	V	L	Y	P	L	P
Giardia_intestinalis	SKY	H	L	G	R	D	T	T	D	F	G	Q	R	N	I	D	W	R	Q	S	Y	GF	K	E	F	P	Y	T	H	I	R	L	D	I	Q	C	Y	I	R	R	M	E	N	F	V	L	Y	P	L	S

K29 H36 F37 T38 L39 V40 K41 D42 N44 V45 T47 R49 D50 F53 R60 D61 V64 G65 W67 I68 Q72 E162 F163 G164 F166 N167 E177 E178 A179 D181 R184 Y185 K191 R193 P194
E195 M224 K247 N250 Y262 I263 V266 L267 R269 N270 E273 N274 R277 V278 L279 Y280 P281 L291 P611

GPM	HF	TLVK	DNVTRD	RDV	GW	IQ	QE	YGF	KR	VE	EAD	RYKS	RP	E	FML	LAN	DDY	I	VLR	NEN	R	V	L	P	E	K																											
Monodelphis domestica	HF	TLVK	DNVTRD	RDV	GW	IQ	QE	YGF	KQ	VE	EAD	RHKA	RP	E	FML	LAN	DDY	I	VLR	NEN	R	V	L	P	E	K																											
Onchithorhynchus anatinus	NS	AGE	YGF	KR	VE	EAD	RHKA	RP	E	FML	LAN	DDY	I	VLR	NEN	R	V	L	P	E	K																												
Gallus gallus	HF	TLVK	DNVTRD	RDV	GW	IQ	QE	YGF	KR	VE	EAD	RHKA	RP	E	YML	LAN	DDY	I	VLR	NEN	R	V	L	P	E	K																											
Xenopus (Silurana)	HF	TLVK	DNVTRD	RDV	GW	IQ	QE	YGF	KK	VE	EAD	RHKA	RP	E	FML	MAN	DDY	I	VLR	NEN	R	V	L	P	E	K																											
Danio rerio	HF	TLVK	DNVTRD	RDV	GW	IQ	QE	YGF	KA	IE	EAD	RYKA	RP	E	YML	MAN	DDY	I	VLR	NEN	R	V	L	P	E	K																											
Meleagris gallopavo	GGAL	GKGS	LN	PR	Q	E	YGF	KV	VE	EAD	RYKA	RP	E	YML	MAN	DDY	I	VLR	NEN	R	V	L	P	E	K																										
Anolis carolinensis	E	YGF	KV	VE	EAD	RYKA	RP	E	YML	MAN	DDY	I	VLR	NEN	R	V	L	P	E	K																										
Ciona intestinalis	HY	TLVK	DNVTRD	RDV	GW	IQ	QE	YGF	KR	VE	EAD	RYKA	RP	E	YMF	MSNS	DDY	I	VCR	NEN	R	V	L	P	E	K																											
Branchiostoma floridae	HF	TLVK	DNVTRD	RDV	GW	IQ	QD	YGF	KQ	TE	E	P	DQYKS	RP	E	F	T	Y	MAK	NEY	I	VCR	NEN	R	V	L	P	E	K																								
Anopheles gambiae	HY	TLVK	DNVTRD	KDV	SW	IQ	QE	YGF	KR	VE	EAD	RYKA	RP	E	YMF	MSI	DDY	I	VLR	NEN	R	V	L	P	E	K																											
Aedes aegypti	HY	TLVK	DNVTRD	KDV	SW	IQ	QE	YGF	KK	IE	E	P	DRYKA	RP	E	YMF	MSI	DDY	I	VLR	NEN	R	V	L	P	E	K																										
Drosophila melanogaster	HY	TLVK	DNVTRD	KDV	GW	IQ	QE	YGF	KK	VE	EAD	RYKA	RP	E	F	M	F	MSI	DDY	I	VLR	NEN	R	V	L	P	E	K																									
Ixodes scapularis	HY	TLVK	DNVTRD	KDV	GW	IQ	QE	YGF	KV	A	E	P	DRYKA	RP	E	YMF	MSV	NDY	I	VLR	NEN	R	V	L	P	E	K																										
Nematostella vectensis	HY	SLVK	DNVTQD	KDV	GW	IQ	QE	YGF	ED	VE	H	P	ERFKA	RP	E	Y	L	Y	MAK	DDY	I	VCR	NEN	R	V	L	P	E	K																								
Caenorhabditis elegans	HF	SI	IK	DNVTRD	RDV	SW	IQ	QE	YGF	LR	IE	E	P	DRFKA	RP	E	YMF	MAN	H	DY	V	VMR	NEN	R	V	L	P	L	K																								
Trichoplax adhaerens	HY	TELK	DNNTRD	REAS	SW	IQ	QE	YGF	KV	IE	E	P	DRYKP	RP	E	Y	I	M	MSK	DDY	V	VMR	NEN	R	V	L	P	I	K																								
Monosiga brevicollis	HCT	TLAK	DSVTRD	RDMS	WH	RE	YGF	A	REEL	P	D	K	FVP	RP	E	Y	I	L	V	S	N	S	N	I	VLR	N	ERM	CL	P	E	K																						
Arabidopsis thaliana	EF	T	P	L	F	SE	KEPK	RD	I	M	WNEK	YGF	RTE	E	A	A	D	E	L	I	V	RN	D	V	S	V	V	A	S	D	K	H	T	A	E	L	F	E	K	F	V	L	P	E	K								
Sorghum bicolor	QY	N	P	H	F	S	LAGEQ	RD	I	Q	WNL	R	YGF	HAE	E	F	A	D	D	KIP	RH	D	V	V	K	L	A	A	D	Q	Y	E	A	Q	H	A	Q	Q	A	V	L	P	E	K									
Oryza sativa	QY	S	P	H	F	S	LAGEQ	RDV	Q	WNL	R	YGF	C	T	E	E	I	A	D	E	K	I	V	RH	D	I	V	N	L	A	A	D	Q	Y	E	A	Q	H	A	Q	Q	A	V	L	P	E	K						
Cryptosporidium parvum	EY	T	L	A	R	T	FNDNA	RD	I	E	L	N	E	T	YGF	K	V	F	E	H	P	Y	V	Q	I	E	RQ	D	V	T	Q	V	P	R	E	K	Y	V	CR	Q	E	Y	S	V	L	P	Q	K					
Debaryomyces hansenii	EI	T	L	A	R	N	YNDLG	RD	L	I	W	A	Q	Q	YGF	K	I	V	E	Q	P	Y	E	Y	I	N	RS	E	I	Q	L	V	P	N	E	D	Y	Q	V	A	Q	Q	E	S	S	V	L	P	H	K			
Scheffersomyces stipitis	ET	T	L	A	R	N	YNDLA	RD	L	I	W	A	Q	Q	YGF	K	V	I	E	T	P	Y	K	Y	I	M	RS	E	I	Q	L	V	P	E	D	Y	Q	V	G	Q	Q	E	S	S	V	L	P	S	K				
Lodderomyces elongisporus	ET	N	L	G	R	S	YNDLA	RD	I	I	W	A	Q	Q	YGF	K	I	I	E	T	P	Y	K	Y	L	D	RH	E	I	Q	L	V	P	N	E	D	Y	Q	V	A	Q	Q	E	S	A	V	L	P	E	K			
Candida albicans	ET	S	L	G	R	S	YNDLA	RDV	I	W	A	Q	Q	YGF	K	I	I	E	T	P	Y	N	Y	I	D	RNE	E	I	Q	L	V	P	T	E	D	Y	Q	V	A	Q	Q	E	S	S	V	L	P	K	K				
Eremothecium gossypii	ET	T	L	A	R	S	YNDFA	RDV	I	W	N	Q	Q	YGF	K	I	V	E	T	P	Y	N	F	I	E	RS	E	I	Q	L	V	P	T	E	D	Y	S	V	A	Q	Q	E	S	A	V	L	P	S	K				
Kluyveromyces lactis	ET	T	L	A	R	S	YNDLA	RDV	I	W	N	Q	Q	YGF	K	I	V	E	T	P	Y	N	F	I	E	RS	E	I	Q	I	V	P	T	E	D	Y	K	V	E	Q	Q	E	S	A	V	L	P	E	K				
Candida glabrata	ET	T	L	G	R	S	YNDLA	RD	I	I	W	N	Q	Q	YGF	K	I	V	E	T	P	Y	N	F	I	E	RNE	E	V	Q	L	V	P	T	E	D	Y	K	V	G	Q	Q	E	S	A	V	L	P	Q	K			
Saccharomyces cerevisiae	ET	T	L	A	R	S	YNDMA	RDV	I	W	N	Q	Q	YGF	K	I	V	E	T	P	Y	N	S	I	E	RNE	E	V	Q	L	V	P	T	E	D	Y	K	V	A	Q	Q	E	S	A	V	L	P	Q	K				
Yarrowia lipolytica	ET	S	L	A	R	S	YNDLA	RDV	V	W	N	Q	Q	YGF	K	I	V	E	Q	P	Y	T	Y	I	P	RNE	E	I	S	R	V	P	Q	E	D	Y	I	V	S	Q	Q	E	T	A	V	L	P	S	K				
Neurospora crassa	ET	T	L	A	R	S	YNDQA	RD	I	L	W	N	Q	R	YGF	E	I	V	E	V	P	Y	D	F	F	RH	D	V	T	K	V	A	S	E	D	Y	E	V	A	Q	Q	E	T	A	V	L	P	R	K				
Aspergillus fumigatus	ET	T	L	A	R	S	YNDLA	RD	I	I	W	N	Q	R	YGF	E	V	E	I	P	Y	D	F	F	RH	D	I	T	Q	V	A	S	E	D	Y	E	V	A	Q	Q	E	T	A	V	L	P	R	K					
Cryptococcus neoformans	NT	S	L	G	R	Q	YNDVA	RD	L	D	W	N	A	N	YGF	L	S	L	E	A	P	P	D	R	I	A	RL	D	V	T	L	V	P	Q	G	N	Y	E	V	A	S	E	N	R	V	L	P	A	K				
Laccaria bicolor	QT	S	L	A	R	Q	YNDFG	RD	L	V	W	N	M	K	YGF	L	S	L	E	A	P	P	E	N	L	P	RL	D	V	T	L	V	P	R	G	N	Y	E	V	E	S	N	D	A	S	V	L	P	F	K			
Dictyostelium discoideum	EY	T	L	A	R	T	YNDFS	RD	I	E	W	N	Q	N	YGF	G	Y	T	E	V	P	Y	V	A	I	E	RL	D	V	Q	Q	I	P	K	E	N	Y	L	V	E	K	Q	E	N	S	V	L	P	S	K			
Ostreococcus 'lucimarinus'	EY	T	M	A	R	N	YQDFE	RD	I	E	W	N	Q	YGF	T	N	H	E	P	Y	N	F	I	E	R	P	Y	I	S	S	V	P	R	E	D	Y	V	I	L	K	Q	E	T	S	V	L	P	Q	K				
Populus trichocarpa	EY	T	V	A	R	S	FSDFE	RD	I	E	W	H	L	Q	YGF	V	L	H	E	Q	P	Y	N	F	I	E	R	V	H	V	T	E	V	P	D	Q	D	Y	I	V	V	R	Q	E	T	S	V	L	P	Q	K		
Vitis vinifera	EY	T	V	A	R	S	FSDFE	RD	I	E	W	H	Q	Q	YGF	V	L	H	E	Q	P	Y	N	F	I	E	R	V	H	V	S	E	V	P	G	Q	D	Y	I	V	V	R	Q	E	T	C	V	L	P	Q	K		
Giardia intestinalis	KY	H	L	G	R	D	T	T	D	F	G	R	N	I	D	W	R	Q	S	YGF	T	R	E	E	F	P	Y	T	H	I	E	R	L	D	K	D	R	I	A	C	DDY	I	I	R	M	E	N	F	V	L	P	S	K

H36 F37 T38 L39 V40 K41 D42 N44 V45 T47 R49 D50 R60 D61 V64 G65 W67 I68 Q72 E162 Y163 G164 F166 K169 R171 V176 E177 E178 A179 D181 R184 Y185 K191 S192
R193 P194 E195 F196 M197 L222 L224 A248 N250 D251 D261 Y262 I263 V266 L267 R269 N270 E273 N274 R277 V278 L279 Y280 P281 E287 K289

PHS2	APENY	HKY	SPHF	SPL	RDI	QWN	YVHL	F	EIP	E	D	E	K	F	I	V	RH	D	V	RKAAE	FLF	FNE	GYE	AQH	SQQ	TVP	PGATN	GLAT		
Populus_trichocarpa	APLNY	HQF	SPHF	SPF	RDI	QWN	YVYL	F	EIA	E	D	E	K	F	I	V	RHD	V	RRASD	FLF	FND	GYES	QHS	SQQ	AVP	PGATN	GLAT			
Vitis_vinifera	APLNY	HQY	SPHF	SPF	RDI	QWN	YVYL	F	EIA	E	D	E	K	F	I	V	RHD	V	RRAAE	FLF	FND	GYES	AQH	SQQ	AVP	PGATS	GLAT			
Oryza_sativa	APANF	HQY	SPHF	SPL	RDV	QWNY	L	YLF	EFA	E	D	E	K	F	I	V	RHD	I	RKAAE	FLF	FND	GYE	AQH	AAQ	AVP	PGATE	GLAT			
Sorghum_bicolor	APSNY	HQY	NPHF	SPL	RDI	QWNY	L	YLF	EFA	E	D	D	K	F	I	P	RHD	V	RKAAE	FLF	FND	GYE	AQH	AAQ	AVP	PGATE	GLAT			
Ostreococcus_lucimarinus'	REV	EWNY	A	YLF	EYA	D	D	E	V	G	V	A	R	Q	P	Q	P	A	F	N	A	S	YEMGTNSMA	YPGGTE	GLAT	
Monosiga_brevicollis	...LR	HHCH	TLAK	DVS	RDMS	WHRY	I	FEL	PDD	D	K	F	G	V	P	R	EY	RRS	N	FLS	NHG	YIV	LRN	ERM	C	YPNNFE	GLPG			
Trichoplax_adhaerens	EPQSR	HHY	TEL	KDRN	REAS	WI	QQY	I	FEE	PDD	D	R	Y	G	K	P	R	EY	RKS	K	FLS	N	FNA	G	YVV	CRNEN	R	YPNNFI	GLPG	
Caenorhabditis_elegans	GAQAR	HHF	SI	I	KDRN	RDVSWI	QQY	I	FEE	PDD	D	R	F	G	K	A	R	EY	RKA	N	FLK	F	N	DGY	V	VMR	NEN	R	YPNNML	GLPG
Nematostella_vectensis	RPVAR	HHY	SLV	KDRN	KDVGI	QQY	I	FEE	HPD	D	E	R	F	G	K	A	R	EY	RKA	K	FLS	F	N	DGY	I	V	CRNEN	R	YPNNFE	GLPG
Branchiostoma_floridae	GQASR	HHF	TLV	KDRN	RDVGWI	QQY	I	FEE	ADD	D	Q	Y	G	K	S	R	EY	RKA	K	FLK	F	N	DGY	I	V	CRNEN	R	YPNNME	GLPG	
Ciona_intestinalis	GASSR	HHY	TLV	KDRN	RDVGWI	QQY	I	FEE	EAD	D	D	R	Y	G	K	A	R	EY	RKS	N	FLG	F	N	TGY	I	V	CRNEN	R	YPNNFE	GLPG
Xenopus_(Silurana)	GVGGR	HHF	TLV	KDRN	RDVGWI	QQY	I	FEE	EAD	D	D	R	H	G	K	A	R	EY	RRR	N	FLR	F	N	VGY	I	V	LRNEN	R	YPNNFE	GLPG
Gallus_gallus	GVGGR	HHF	TLV	KDRN	RDVGWI	QQY	I	FEE	EAD	D	D	R	H	G	K	A	R	EY	RRR	N	FLR	F	N	VGY	I	V	LRNEN	R	YPNNFE	GLPG
Ornithorhynchus_anatinus	MALY	I	FEE	EAD	D	D	R	H	G	K	A	R	EY	RRR	N	FLR	F	N	VGY	I	V	LRNEN	R	YPNNFE	GLPG
Sus_scrofa	GVGGR	HHF	TLV	KDRN	RDVGWI	QQY	I	FEE	EAD	D	D	R	H	G	K	A	R	EY	RRR	N	FLR	F	N	VGY	I	V	LRNEN	R	YPNNFE	GLPG
Callithrix_jacchus	GVGGR	HHF	TLV	KDRN	RDVGWI	QQY	I	FEE	EAD	D	D	R	H	G	K	A	R	EY	RRR	N	FLR	F	N	VGY	I	V	LRNEN	R	YPNNFE	GLPG
Meleagris_gallopavo	GYKIS	NN	GGAL	GEKPG	RQFI	F	E	EAD	D	D	R	Y	G	K	A	R	EY	RKA	N	FLQ	F	N	VGY	I	V	LRNEN	R	YPNNFE	GLPG
Anolis_carolinensis	FIFE	EAD	D	D	R	Y	G	K	A	R	EY	RKA	N	FLF	F	N	VGY	I	V	LRNEN	R	YPNNFE	GLPG		
Danio_rerio	GAGNR	HHF	TLV	KDRN	RDVGWI	QQFI	F	E	EAD	D	D	R	Y	G	K	A	R	EY	RKA	C	FLK	F	N	VGY	I	V	LRNEN	R	YPNNFE	GLPG
Homo_sapiens	GAGNR	HHF	TLV	KDRN	RDVGWI	QQFI	F	E	EAD	D	D	R	Y	G	K	A	R	EY	RKA	N	FLK	F	N	VGY	I	V	LRNEN	R	YPNNFE	GLPG
Ixodes_scapularis	GAQAR	HHY	TLV	KDRN	KDVGI	QQY	I	FKE	PD	D	D	R	Y	G	K	A	R	E												

A20 P22 E23 N33 Y36 H37 K39 Y40 S41 P42 H43 F44 S45 P46 L47 R63 D64 I67 Q68 W70 N71 Y74 V75 H166 L168 F169 E180 I181 P182 E183 D184 E187 K188 F189 I194 V195
R196 H197 D198 V199 R246 K251 A252 A254 E255 L257 L259 F260 F262 N263 E264 G265 Y267 E268 A271 Q272 H274 S275 Q278 Q279 T282 V283 Y285 P286 G287 A289
T290 N292 G293 L296 A618 T619

GPb	RHF	TLVK	DNVD	RDVG	WI	QF	ADRY	KRP	EYMN	DY	I	VL	RNL	N	R	D	N	I																							
Monodelphis domestica	RHF	TLVK	DNVD	RDVG	WI	QF	ADRY	KRP	EYMN	DY	I	VL	RNL	N	R	D	N	E																							
Equus caballus	DVIC	LVGR	CL	ADRY	KRP	EYMN	DY	I	VL	RNL	N	R	D	N	L																							
Gallus gallus	RHF	TLVK	DNVD	RDVG	WI	QF	ADRY	KRP	EYMN	DY	I	VL	RNL	N	R	D	N	L																							
Meleagris gallopavo	SG	GAL	GE	KGS	SPR	QF	ADRY	KRP	EYMN	DY	I	VL	RNL	N	R	D	N	L																				
Anolis carolinensis	F	ADRY	KRP	EYMN	DY	I	VL	RNL	N	R	D	N	I																				
Xenopus (Silurana)	RHF	TLVK	DNVD	RDVG	WI	QF	ADRY	KRP	EYMN	EY	I	VL	RNL	N	R	D	N	I																							
Danio rerio	RHF	TLVK	DNVD	RDVG	WI	QF	ADRY	KRP	EYMN	DY	I	VL	RNL	N	R	D	N	E																							
Bos taurus	RHF	TLVK	DNVD	RDVG	WI	QF	ADRY	KRP	EFTN	DY	I	VL	RNL	N	R	D	D	E																							
Ornithorhynchus anatinus	NS	AGY	ADRH	KRP	EYMN	DY	I	VL	RNL	N	R	D	N	E																					
Sus scrofa	RHF	TLVK	DNVD	RDVG	WI	QY	ADRY	KRP	EYMN	DY	I	VL	RNL	N	R	D	S	E																							
Ciona intestinalis	RHY	TLVK	DNVD	RDVG	WI	QY	ADRY	KRP	EYMN	S	Y	I	V	C	R	N	L	N	R	D	H	E																			
Branchiostoma floridae	RHF	TLVK	DNVD	RDVG	WI	QY	P	D	Q	Y	K	R	P	E	F	T	K	N	Y	I	V	C	R	N	L	N	R	D	Y	E											
Anopheles gambiae	RHY	TLVK	DNVD	KDVS	WI	QY	P	D	R	Y	K	R	P	E	Y	M	I	D	Y	I	V	L	R	N	L	N	R	D	G	V											
Aedes aegypti	RHY	TLVK	DNVD	KDVS	WI	QY	P	D	R	Y	K	R	P	E	Y	M	I	D	Y	I	V	L	R	N	L	N	R	D	S	E											
Drosophila melanogaster	RHY	TLVK	DNVD	KDVG	WI	QY	P	D	R	Y	K	R	P	E	F	M	I	D	Y	I	V	L	R	N	L	N	R	D	E	D											
Ixodes scapularis	RHY	TLVK	DNVD	KDVG	WI	QY	P	D	R	Y	K	R	P	E	Y	M	V	N	Y	I	V	L	R	N	L	N	R	D	H	E											
Nematostella vectensis	RHY	SLVK	DNVD	KDVG	WI	QY	P	E	R	F	K	R	P	E	Y	L	K	D	Y	I	V	C	R	N	A	N	R	D	C	M											
Caenorhabditis elegans	RHF	SI	I	K	DNVD	RDVS	WI	QY	P	D	R	F	K	R	P	E	Y	M	N	H	Y	V	M	R	N	L	N	R	D	Y	E										
Trichoplax adhaerens	RHY	TELK	DNND	REAS	WI	QY	P	D	R	Y	K	R	P	E	Y	I	K	D	Y	V	V	C	R	N	L	N	R	D	M	A											
Monosiga brevicollis	RHC	TLAK	DSVD	RDMS	WH	R	Y	P	D	K	F	V	R	P	E	Y	I	N	S	Y	I	V	L	R	N	L	R	M	D	D	E										
Arabidopsis thaliana	YKY	SPHF	SLKQ	RD	I	Q	W	N	V	H	P	D	E	K	I	R	H	D	V	V	A	D	Y	E	A	Q	H	S	R	Q	T	..									
Sorghum bicolor	YQY	NPHF	SLAQ	RD	I	Q	W	N	L	Y	A	D	D	K	I	R	H	D	V	V	A	D	Y	E	A	Q	H	A	K	Q	A	D	..								
Oryza sativa	FQY	SPHF	SLAQ	RD	V	Q	W	N	L	Y	A	D	E	K	I	R	H	D	I	V	A	D	Y	E	A	Q	H	A	R	Q	A	D	..								
Cryptosporidium parvum	NEY	TLAR	T	F	N	A	R	D	I	E	L	N	E	Y	P	Y	V	Q	I	R	Q	D	V	T	R	E	Y	V	V	C	R	Q	R	Y	S	D	A	P			
Debaryomyces hansenii	KEI	TLAR	N	Y	N	G	R	D	L	I	W	A	Q	Y	P	Y	E	Y	I	R	S	E	I	Q	N	E	Y	Q	V	A	Q	K	S	S	D	..					
Scheffersomyces stipitis	KET	TLAR	N	Y	N	A	R	D	L	I	W	A	Q	Y	P	Y	K	Y	I	R	S	E	I	Q	E	E	Y	Q	V	G	Q	R	S	S	D	..					
Lodderomyces elongisporus	RET	N	L	G	R	S	Y	N	A	R	D	I	I	W	A	Q	Y	P	Y	K	Y	L	R	H	E	I	Q	N	E	Y	Q	V	A	Q	K	S	A	D	..		
Candida albicans	KET	S	L	G	R	S	Y	N	A	R	D	V	I	W	A	Q	Y	P	Y	N	Y	I	R	N	E	I	Q	T	E	Y	Q	V	A	Q	R	S	S	D	..		
Eremothecium gossypii	QET	T	L	A	R	S	Y	N	A	R	D	V	I	W	N	Q	Y	P	Y	N	F	I	R	S	E	I	Q	T	E	Y	S	V	A	Q	R	S	A	D	..		
Kluyveromyces lactis	QET	T	L	A	R	S	Y	N	A	R	D	V	I	W	N	Q	Y	P	Y	N	F	I	R	S	E	I	Q	T	E	Y	K	V	E	Q	R	S	A	D	..		
Candida glabrata	DET	T	L	G	R	S	Y	N	A	R	D	I	I	W	N	Q	Y	P	Y	N	F	I	R	N	E	V	Q	T	E	Y	K	V	G	Q	R	S	A	D	..		
Saccharomyces cerevisiae	DET	T	L	A	R	S	Y	N	A	R	D	V	I	W	N	Q	Y	P	Y	N	S	I	R	N	E	V	Q	T	E	Y	K	V	A	Q	R	S	A	D	..		
Yarrowia lipolytica	RET	S	L	A	R	S	Y	N	A	R	D	V	V	W	N	Q	Y	P	Y	T	Y	I	R	N	E	I	S	Q	E	Y	I	V	S	Q	R	T	A	D	E	.	
Neurospora crassa	RET	T	L	A	R	S	Y	N	A	R	D	I	L	W	N	Q	Y	P	Y	D	F	E	R	H	D	V	T	S	E	Y	E	V	A	Q	R	T	A	D	D	Q	
Aspergillus fumigatus	RET	T	L	A	R	S	Y	N	A	R	D	I	I	W	N	Q	Y	P	Y	D	F	E	R	H	D	I	T	S	E	Y	E	V	A	Q	R	T	A	D	D	D	
Cryptococcus neoformans	RNT	S	L	G	R	Q	Y	N	A	R	D	L	D	W	N	A	Y	P	P	D	R	I	R	L	D	V	T	Q	G	Y	E	V	A	S	E	N	R	D	E	D	
Laccaria bicolor	NQT	S	L	A	R	Q	Y	N	G	R	D	L	V	W	N	M	Y	P	P	E	N	L	R	L	D	V	T	R	G	Y	E	V	E	S	N	S	A	S	D	A	E
Dictyostelium discoideum	DEY	T	L	A	R	T	Y	N	S	R	D	I	E	W	N	Q	Y	P	Y	V	A	I	R	L	D	V	Q	K	E	Y	L	V	E	K	Q	R	N	S	D	T	T
Ostreococcus 'lucimarinus'	NEY	T	M	A	R	N	Y	Q	E	R	D	I	E	W	N	Q	Y	P	Y	N	F	I	R	P	Y	I	S	R	E	Y	V	I	L	K	Q	R	T	S	D	E	P
Populus trichocarpa	DEY	T	V	A	R	S	F	S	E	R	D	I	E	W	H	L	Y	P	Y	N	F	I	R	V	H	V	T	D	Q	Y	I	V	V	R	Q	R	T	S	D	F	.
Vitis vinifera	DEY	T	V	A	R	S	F	S	E	R	D	I	E	W	H	Q	Y	P	Y	N	F	I	R	V	H	V	S	G	Q	Y	I	V	V	R	Q	R	T	C	D	S	.
Giardia intestinalis	SKY	H	L	G	R	D	T	T	G	R	N	I	D	W	R	Q	Y	P	Y	T	H	I	R	L	D	K	D	C	D	Y	Y	I	R	R	M	E	N	F	D	S	S

R33 H36 F37 T38 L39 V40 K41 D42 N44 V45 D50 R60 D61 V64 G65 W67 I68 Q72 F163 A179 D181 R184 Y185 K191 R193 P194 E195 Y196 M197 N250 D251 Y262 I263 V266
L267 R269 N270 L271 N274 R277 D283 N838 I839

Gsy2	RHGC	F	DFD	V	LE	VH	V	T	I	G	I	F	D	A	T	R	Y	P	H	N	E	L	P	T	D	L	L	K	L	K	I	L					
Candida glabrata	RHGS	Y	DFD	V	LE	V	D	V	T	I	G	I	F	D	A	M	K	F	P	H	N	E	L	P	T	D	L	L	K	L	K	V	L				
Eremothecium gossypii	RHWN	Y	DFD	V	LE	V	K	V	T	I	G	L	F	E	A	M	R	Y	P	R	S	E	I	P	T	D	L	L	L	T	L	K	V	L			
Kluyveromyces lactis	RHGQ	F	DFD	V	LE	V	K	V	T	I	G	I	F	E	A	M	R	Y	P	H	N	E	I	P	T	S	L	L	F	K	L	K	V	L			
Phaeosphaeria nodorum	RYGH	N	DFD	I	L	H	V	N	I	A	V	A	L	F	E	S	L	T	W	T	E	G	D	L	P	E	D	L	.	I	K	L	R	L	M		
Aspergillus fumigatus	RYGH	N	DFD	V	L	R	I	E	I	E	I	G	M	Y	E	C	L	A	W	K	E	G	N	M	P	D	E	L	.	I	R	L	R	L	M		
Neurospora crassa	RYGH	Y	DFE	I	L	R	V	D	I	E	I	G	I	F	E	S	V	K	W	H	E	G	P	L	P	E	E	L	.	I	R	L	R	L	M		
Botryotinia fuckeliana	RYGH	N	DFD	I	L	R	V	D	I	E	V	G	I	F	E	A	L	K	W	H	E	G	V	M	P	D	E	L	.	I	R	L	R	L	M		
Yarrowia lipolytica	KYGH	L	DFD	M	L	E	V	N	I	Q	I	G	M	L	D	C	A	R	H	N	S	H	E	I	P	G	L	L	.	L	R	L	K	V	L		
Candida albicans	KYGN	Y	DFD	V	L	E	I	G	V	Q	V	G	L	F	E	C	A	R	Y	P	N	T	E	V	P	T	I	L	.	I	R	L	K	I	L		
Lodderomyces elongisporus	KYGN	Y	DFD	I	L	E	I	E	V	Q	V	G	L	F	E	C	A	R	Y	P	N	I	E	V	P	S	I	L	.	I	R	L	K	I	L		
Debaryomyces hansenii	KYGN	Y	DFD	I	L	E	I	E	V	Q	V	G	L	F	E	C	A	R	F	P	N	N	E	V	P	S	I	L	.	I	R	L	K	I	L		
Scheffersomyces stipitis	KYGN	Y	DFD	I	L	E	I	E	V	Q	V	G	L	F	E	C	A	R	F	P	N	A	E	V	P	S	I	L	.	I	R	L	K	I	L		
Ustilago maydis	KYGH	Y	DFD	T	L	R	V	E	I	Q	V	G	L	F	E	M	A	R	Y	Q	G	E	D	V	I	D	P	L	.	L	K	L	K	I	L		
Laccaria bicolor	RYGH	Y	DFD	T	L	R	V	T	I	Q	V	G	L	F	D	A	A	R	F	H	G	S	T	I	P	T	P	L	.	L	Q	L	K	I	L		
Cryptococcus neoformans	RYGH	Y	DFD	T	L	K	V	E	V	T	I	S	I	F	E	A	C	R	Y	S	G	E	E	V	P	N	P	L	.	L	R	L	K	V	L		
Nematostella vectensis	RYGH	F	DFD	T	L	K	V	N	I	Q	I	G	I	Y	E	C	V	K	N	L	P	S	G	L	.	L	I	L	K	V	A		
Anolis carolinensis	RYGH	L	DFN	R	L	W	A	N	V	K	F	G	L	Y	E	L	L	V	N	L	P	D	M	M	.	L	F	M	K	I	T		
Xenopus (Silurana)	RYGH	L	DFN	R	L	C	A	N	I	K	F	G	L	Y	E	L	L	V	N	L	P	D	M	M	.	L	F	L	K	I	T		
Homo sapiens	RYGH	L	DFN	R	L	W	A	N	V	K	F	G	L	Y	E	L	L	V	S	L	P	D	M	M	.	L	F	M	K	I	T		
Ornithorhynchus anatinus	RYGH	L	DF	S	R	L	W	A	N	V	K	F	G	L	Y	E	L	L	V	N	L	P	D	M	M	.	L	F	M	K	L	T	
Danio rerio	RYGH	L	DFN	R	L	W	A	Q	V	K	F	G	L	Y	E	L	L	R	E	I	P	D	M	I	.	L	F	M	K	I	T		
Meleagris gallopavo	RYGH	L	DF	S	R	L	W	A	Q	V	K	F	G	L	Y	N	L	L	K	E	I	P	D	L	I	.	L	I	M	K	I	T	
Taeniopygia guttata	RYGH	L	DFD	R	L	W	A	Q	V	K	F	G	L	Y	N	L	L	K	E	I	P	D	L	I	.	L	I	M	K	I	T		
Monodelphis domestica	RYGH	L	DFD	R	L	W	A	N	V	K	F	G	L	Y	D	L	L	K	E	I	P	D	L	I	.	L	V	M	K	I	T		
Pan troglodytes	RYGH	L	DFD	R	L	W	A	H	V	K	F	G	L	Y	D	L	L	R	E	I	P	D	L	I	.	L	L	M	K	I	T		
Trichoplax adhaerens	RHGH	F	N	F	K	T	L	R	V	N	I	Q	M	G	L	F	E	A	L	M	E	I	I	D	G	L	.	L	F	L	K	I	A
Branchiostoma floridae	RYGH	Y	DFD	V	L	R	V	A	V	Q	I	G	I	F	E	C	L	G	V	W	R	Q	Y	L	S	A	N	S	.	L	R	I	A	I	A		
Ixodes scapularis	RYGH	Y	DFD	S	L	R	V	Q	M	Q	I	G	M	F	E	C	L	S	K	I	P	K	G	L	.	I	L	L	K	I	S		
Drosophila melanogaster	RYGH	I	DFD	I	L	R	I	N	V	Q	V	G	M	F	D	C	L	Q	N	I	P	N	A	L	.	L	L	I	K	M	M		
Drosophila pseudoobscura	RYGH	M	DFD	I	L	R	I	N	V	Q	V	G	M	F	D	C	V	K	R	L	P	E	V	L	.	L	M	I	K	M	M		
Apis mellifera	RYGH	Y	DFD	T	L	R	I	N	I	Q	I	G	M	Y	E	C	L	S	R	M	P	D	V	L	.	L	T	I	K	L	L		
Anopheles gambiae	RYGH	F	N	F	N	T	L	R	I	N	I	Q	I	G	M	Y	E	C	L	Q	Q	L	P	E	G	I	.	L	I	I	K	L	L
Aedes aegypti	RYGH	F	N	F	D	T	L	R	L	N	I	Q	I	G	M	Y	D	C	L	K	H	L	P	D	G	I	.	L	M	I	K	L	L
Caenorhabditis elegans	RHGH	L	DFD	T	L	K	V	D	I	K	V	G	I	F	D	C	L	Q	H	L	P	E	P	L	.	M	N	L	K	I	L		
Ciona intestinalis	RYGH	F	D	L	N	V	L	K	V	D	I	K	I	G	L	F	D	C	L	R	K	L	P	N	E	L	.	L	V	L	K	M	N
Monosiga brevicollis	KYGH	I	E	W	D	V	L	T	V	K	I	Q	V	G	M	L	E	V	M	R	E	L	P	Q	S	L	.	L	V	L	K	L	A
Dictyostelium discoideum	RYGH	Y	DFD	L	M	R	C	N	I	V	M	G	L	F	E	T	S	R	K	M	I	S	P	L	.	L	L	L	K	I	L		
Giardia intestinalis	RYGT	.	T	F	N	M	I	K	T	R	L	A	I	S	L	V	D	L	T	N	T	A	L	S	N	L	A	S	T	.	L	I	I	M	K	S	S

R298 H302 G303 C304 F305 D306 F307 D308 V375 L378 E379 V382 H383 V385 T386 I389 G390 I393 F394 D395 A397 I398 R399 Y400 P401 H402 N403 E408 L409 P410 T411
D412 L413 L416 L417 K422 L425 K426 I429 L432

Sus1	NFTLELDPFETELHKVDLERYEMFYALKR
Populus_trichocarpa	NFVLELDPFETELHKVDRERYEMFYALKR
Vitis_vinifera	NFVLELDPFETELHKVDRERYEMFYALKR
Oryza_sativa	NFVLELDPFETELHKVERERYEMFYALKR

N132 F133 T134 L135 E136 L137 D138 P141 F142 E390 T393 E394 L424 H427 K428 V511 D779 L781 E782 R785 Y786 E788 M789 F790 Y791 A792 L793 K794 R796

PoFUT1	CMGN	KW	TPRQAIY	AEP	CH	EGNPF	V	DE	GDP	GF	DQEY	S	FDV	H	T	R	L	F
Caenorhabditis_remanei	CMGN	KW	TPRRAIY	DKE	CH	EGNPF	V	DE	GDP	GF	DQEY	S	FDV	H	T	R	L	F
Caenorhabditis_briggsae	CMGN	.	WNPRKAIY	DTD	CH	EGNPF	V	DE	GEP	GF	EQEY	S	FDV	H	T	R	L	F
Ciona_intestinalis	CMGN	Y	HSTKRSE	DKK	CP	SGNPF	P	SE	.	P	T	F	H	K	S	H	G	Y
Trichoplax_adhaerens	CMGN	I	FYYR...	ENK	CA	EGNPF	K	Y	V	G	N	G	GF	N	V	K	Y	G
Ixodes_scapularis		Y	CH									G	G	R	W	G	F
Daphnia_pulex	CMGN	T	YMARQG..	TD	CN	EGNPF	V	SE	G	.	P	Y	D	H	K	S	G	F
Drosophila_willistoni	CMGN	V	YMERSLQQ	EKN	CH	DGNPF	V	SE	G	.	P	F	D	H	S	W	G	F
Drosophila_pseudoobscura	CMGN	V	YMERKNNP	DQP	CH	DGNPF	V	SE	G	.	P	F	D	H	S	W	G	F
Drosophila_melanogaster	CMGN	V	YKERKNDP	DKP	CH	DGNPF	V	SE	A	.	P	F	D	H	S	Y	G	F
Aedes_aegypti	CMGN	I	YTERGLDGS	TG	CN	SGNPF	V	SE	G	.	P	Y	D	H	K	W	G	F
Anopheles_gambiae	CMGN	I	YTERGLDGS	TG	CN	SGNPF	A	SE	G	.	P	Y	D	H	K	W	G	F
Nematostella_vectensis	CMGN	V	WLPPSS..	TK	CQ	DGNPF	D	Y	E	.	H	Y	H	D	L	H	G	F
Branchiostoma_floridae	CMGN	K	FETMAQRSV	DKK	CP	EGNPF	V	SE	.	E	G	Y	N	D	Q	F	G	F
Danio_rerio	CMGN	I	FESAHRSD	DKK	CP	DGNPF	D	SV	.	G	G	F	S	Y	E	H	G	F
Oryzias_latipes	CMGN	R	FETAQRSAD	DKK	CP	DGNPF	D	SV	.	G	G	F	S	Y	E	H	G	F
Tetraodon_nigroviridis	CMGN	R	FEEAAQRSE	DKK	CP	EGNPF	D	SV	.	G	G	F	S	Y	Q	H	G	F
Takifugu_rubripes	CMGN	R	FETAQRTAD	DKK	CP	DGNPF	D	SV	.	G	G	F	S	Y	Q	H	G	F
Xenopus_(Silurana)	CMGN	V	FETAAQRSP	DKK	CP	DGNPF	I	SE	.	D	G	F	S	Y	Q	H	G	F
Ornithorhynchus_anatinus	.	MGN	VFEQAAERSS	DGK	CP	DGNPF	S	SE	.	S	G	F	S	Y	E	H	G	F
Mus_musculus	CMGN	V	FEEVAQRSP	DKK	CP	EGNPF	N	SE	.	T	G	F	S	Y	E	H	G	F
Homo_sapiens	CMGN	V	FEEVAQRSP	DKK	CP	EGNPF	N	SE	.	T	G	F	S	Y	E	H	G	F
Canis_lupus	CMGN	V	FEEVAQRSP	DKK	CP	EGNPF	N	SK	.	S	R	F	S	Y	E	H	G	F
Bos_taurus	CMGN	V	FEEVAQRSP	DKK	CP	EGNPF	N	SE	.	A	G	F	S	Y	E	H	G	F
Anolis_carolinensis	CMGN	V	FESAQRTV	DKK	CP	DGNPF	D	SE	.	G	G	F	S	Y	E	H	G	F
Meleagris_gallopavo	P	AGN	VFEAAQRSA	DKN	CP	DGNPF	D	SE	.	K	G	F	S	Y	E	H	G	F
Gallus_gallus	CMGN	V	FEEAAQRSA	DKS	CP	DGNPF	D	SE	.	K	G	F	S	Y	E	H	G	F
Monosiga_brevicollis	CMGN	H	SRYDAQP.	VD	CR	Q	GNPF	V	DR	.	G	Y	L	H	N	H	G	F

C37 M38 G39 N43 K116 W120 T121 P122 R123 Q124 A125 I126 Y127 A131 E132 P133 C135 H136 E139 G140 N141 P142 F143 V155 D157 E158 G161 D162 P164 G165
F167 D168 Q171 E186 Y187 S194 F199 D244 V248 H251 T256 R258 L260 F261

OGT	RRK	LEPEHQ	EMHK	EIRPYQI	QT	RQLH	DSGNP	EENRGQINLATQNSQ	YGRPAVEPQ	IFPV
Danio rerio	QRK	LEPEHQ	EMHK	EIRPYQL	QT	RQLH	DSGNP	ENNQGQINLATQNSQ	YGRPAVEPA	IFPV
Branchiostoma floridae	KCK	LEPEHQ	EMHK	EIRPYQM	QT	RQLH	DSGQP	ETTQGVNLASQNVQ	YGRPAVEQP	IFHV
Anopheles gambiae	RLK	LEPEHN	ELHK	EIRPYAL	QT	RQLH	DSGNP	EATASGQVNLATQNQQ	YGRPAVEAA	IFNV
Aedes aegypti	RLK	LEPEHN	ELHK	EIRPYAL	QT	RQLH	DSGNP	DTASGQINLATQNQQ	YGRPAVETP	IFNV
Apis mellifera	RLK	LEPEHN	EMHK	EIRPYQL	QT	RQLH	DSGNP	EATASGQCNMATQNQQ	YGRPAVEPP	LFNV
Drosophila melanogaster	RLK	LEPDHK	EMHK	EIRPYSI	QT	RQLH	DSGNP	ESATGQVNLATQNQR	YMRPAVEQP	IFNV
Ciona intestinalis	ASK	LEPEHQ	ELHK	EIRPYQI	QT	RQLH	DSGSP	ASPKNSQLNLAVQAS	YRRPAVEAA	IFPV
Nematostella vectensis	RCK	LEPEHQ	ELHK	EIRPFQI	QS	RQLH	DSGNP	ETLNGESVEKEEGSQ	YGKPAVESQ	IFPV
Trichoplax adhaerens	KRK	LEPEHQ	FAHK	EIRPYQI	QT	RQLH	DSGSP	ENSNGTLLNNGSV.AQ	YGRPAAEAA	VFNV
Caenorhabditis remanei	RLK	LEPEHQ	DLHK	EIRPYSI	AN	RQLH	DAGNA	EQMTGQMNLSGSHAQ	YQRPYQEEP	VFNV
Caenorhabditis elegans	RLK	LEPEHN	DLHK	EIRPYSI	AN	RQLH	DAGNA	EQMTGQMNLSGSHAQ	YQRPYQEEP	VFNV
Caenorhabditis briggsae	RLK	LEPEHQ	DLHK	EIRPYSI	AN	RQLH	DAGNA	EQMTGQMNLSGSHAQ	YQRPYQEEP	VFNV
Vitis vinifera	TKA	LATGFA	DS	CNELRPLSI	QIN	HTLY	DSGHEADNCSD	YGRPAEAEMP	IFDV
Populus trichocarpa	SKA	LATGFS	DS	CNELRPLSI	QIN	HTLY	DSGHEADTCSD	YGRPAEAEMP	IFDV
Arabidopsis thaliana	SKA	LATGFS	DS	CNELRPLTI	QM	HNL	LYDSGHEADNSSD	YGRPAEAEMP	IFDV
Sorghum bicolor	SKA	ISSGLA	DT	CTELRPLNI	QV	QTL	LYDSGHETTVCSD	YGRPAEETS	IFDV
Ustilago maydis	.PY	LISASAL	QY	TQLQPYNI	KEL	I	DQGRQDSQGAQ	...SEQLFEN	RPAEEA	IFDV
Yarrowia lipolytica	.SL	LSPSSLS	YQY	LSPYK	IDLN	KLI	DNGQSLSDVGYSWDSEES	IFPDQ	PPAESN	LFPV
Neurospora crassa	.AY	LLSESAL	AY	QYLTPHDI	SE	KALV	DRGRNDRMLFEN	RPELENA	IFDV
Aspergillus fumigatus	.AY	LLSPSSL	AY	NYLHSYQI	IRE	QQLV	DAGRNDWLFEN	RPDLEQA	IFDV
Botryotinia fuckeliana	.AY	LLSPSAL	AY	NYLLSYPI	KE	KHLV	DQGRSDWLFPS	RPDLESAW	IFDV
Phaeosphaeria nodorum	.PP	MVSPHAL	AY	YLLQKYD	IQE	QTLV	DKGRSDWIFPD	RPDLETA	IFDV
Monosiga brevicollis	HKR	LGPSHP	ERHQ	EIKPYNRV	Q	RLC	DLGVD	EQVSALTNMANQLRF	YGRPPAEQL	IFPV

R338 R341 K342 L344 E345 P348 E349 H354 Q368 E369 M372 H373 K375 E376 I378 R379 P382 Y388 Q402 L405 Q406 T409 R410 Q413 L425 H429 D431 S432 G433 N434
P436 E437 E778 N781 R782 G783 Q784 I785 N796 L798 A799 T801 Q802 N805 S823 Q824 Y825 G826 R867 P869 A870 V871 E873 P874 Q887 I891 F892 P894 V895