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Review

Functional diversity of the eukaryotic translation initiation factors belonging to eIF4 families

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Abstract

Protein synthesis in eukaryotic cells is fundamental for gene expression. This process involves the binding of an mRNA molecule to the small ribosomal subunit in a group of reactions catalyzed by eukaryotic translation initiation factors (eIF) eIF4. To date, the role of each of the four eIF4, i.e. eIF4E, eIF4G, eIF4A and eIF4B, is well established. However, with the advent of genome-wide sequencing projects of various organisms, families of genes for each translation initiation factor have been identified. Intriguingly, recent studies have now established that certain eIF4 proteins can promote or inhibit translation of specific mRNAs, and also that some of them are active in processes other than translation. In addition, there is evidence of tissue- and developmental-stage-specific expression for some of these proteins. These new findings point to an additional level of complexity in the translation initiation process. In this review, we analyze the latest advances concerning the functionality of members of the eIF4 families in eukaryotic organisms and discuss the implications of this in the context of our current understanding of regulation of the translation initiation process.

Keywords: Initiation of translation; eIF4E; eIF4G; eIF4A; eIF4B

1. Introduction

In eukaryotes, protein synthesis, or translation as it is also called, is fundamental for gene expression and is mainly regulated at the initiation step. Protein synthesis is initiated by the recruitment of the 5′-untranslated region (UTR) of the mRNA to the small subunit of the ribosome. This reaction is catalyzed by the eukaryotic translation initiation factors (eIF) of the eIF4 families. First, eIF4E binds the cap structure (m^7GpppN, where N is any nucleotide) at the 5′ end of the mRNA. Next, eIF4A, an ATPase/RNA helicase, unwinds the secondary structure in the 5′UTR allowing the small ribosomal subunit to scan along the mRNA and to reach the start codon; a process that is stimulated by eIF4B and in mammals by eIF4B and eIF4H. After interacting with both the 18S rRNA and eIF3, eIF4B also mediates the binding of the 40S ribosome to the mRNA. In addition, after interacting with eIF4E, eIF4A, poly A-binding protein (PABP), and the ribosome-associated eIF3, the scaffold eIF4G coordinates the binding of the small ribosomal subunit to the mRNA. Factors eIF1, eIF1A, and eIF5 assist the proper positioning of the small ribosomal subunit to the start codon. For picornaviral mRNAs and some cellular mRNAs, 5′ UTR recognition is mediated independent of the cap structure through an internal ribosome entry site (IRES) that is located in the proximity of the initiation codon. Finally, before protein translation can proceed, other reactions, such as the binding of the initiator methionyl-tRNA to the small ribosomal subunit, and the joining of a large ribosomal subunit with the 40S initiation complex to form an 80S ribosome complex, are required (Berthelot et al., 2004; Browning, 1996; Gingras et al., 1999; Hershey and Merrick, 2000; Kozak, 1978, 2002; Pain, 1996; Pestova and Hellen, 2000; Pestova and Kolupaeva, 2002).

The presence of eIF4E, eIF4A, and eIF4G proteins is essential for cap binding and the subsequent RNA helicase

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activities leading to protein translation (Browning, 1996; Gingras et al., 1999; Hershey and Merrick, 2000; Lüking et al., 1998; Pain, 1996; Pestova and Hellen, 2000; Prevot et al., 2003; Rogers et al., 2002; von der Haar et al., 2004). Genome-wide sequencing projects have revealed the presence of gene families for each eIF4 factor in numerous eukaryotic species. Intriguingly, recent studies have even identified more specialized activities of these factors in the protein synthesis process than previously thought and even that some factors play a role in processes other than translation.

Here, we examine the newly discovered specialized roles of members of each eIF4 family and suggest that this diversity adds a new level of complexity to the regulation of gene expression by enabling protein synthesis to be regulated in specific tissues and/or at different developmental stages. The function of some eIF4 proteins in non-translational processes as well as the evolutionary relationships of the eIF4 gene families are also discussed.

2. Versatile eIF4E

Many different eIF4E-related proteins have been characterized in eukaryotes: Three in mammals, termed eIF4E-1 (Rychlik et al., 1987; Sonenberg et al., 1979), 4EHP (Rom et al., 1998), and eIF4E-3 (Joshi et al., 2004); three in plants, termed eIF4E, eIF(iso)4E (Allen et al., 1992; Browning, 1996; Browning et al., 1992; Metz et al., 1992a; Rodriguez et al., 1998), and novel cap-binding protein nCBP (Ruud et al., 1998); five in *C. elegans* (Jankowska-Anyszka et al., 1998; Keiper et al., 2000); two in zebra fish (Fahrenkrug et al., 1999; Robalino et al., 2004); two in *Xenopus* (Wakiyama et al., 2001); two in *S. pombe* (Ptushkina et al., 1996, 2001); two in *Leishmania* (Yoffe et al., 2004); and eight in *Drosophila* (Hernández et al., 1997, 2005; Hernández and Sierra, 1995; Lavoie et al., 1996; Maroto and Sierra, 1989). In contrast, only one *eIF4E* gene is present in *S. cerevisiae* (Altmann et al., 1987). Interestingly, the first viral *eIF4E* gene was recently discovered in the Mimivirus, the largest eukaryotic virus known to date (Raoult et al., 2004). Some *eIF4E* genes are also restricted to specific phylogenetic groups. For example, eIF(iso)4E is found only in plants (Browning, 1996; Browning et al., 1992), nCBP in metazoans (Ruud et al., 1998), and mouse-related eIF4E-3 in chordates (Joshi et al., 2004). Analysis of the phylogenetic relationships of the eIF4E family members illustrates that they group into eight clades (Fig. 1a): (1) *Drosophila* proteins (except for eIF4E-8/d4EHP) (gray); (2) *Xenopus*, human eIF4E-1, and zebra fish proteins (orange); (3) *C. elegans* (except for IFE-4) proteins (violet); (4) fungi proteins (blue); (5) plant proteins, subdivided in eIF4Es and eIF(iso)4Fs (green); (6) *Drosophila* eIF4E-8/d4EHP, mammalian 4EHP, *C. elegans* IFE-4, *Arabidopsis* nCBP, mouse eIF4E-3, and *Leishmania* eIF4E-1 (brown); (7) *Leishmania* eIF4E-2 (yellow); and (8) eIF4E from Mimivirus (red).

In general, eIF4E is characterized by its cap-binding activity (Browning, 1996; Gingras et al., 1999; Hershey and Merrick, 2000; Pain, 1996; Pestova and Hellen, 2000; Rogers et al., 2002; von der Haar et al., 2004), although some cellular processes require the activity of specific eIF4E proteins. Indeed, the accumulation of one eIF4E isoform appears to be a universal requirement for gametogenesis (Table 1). In *C. elegans*, IFE-1 is required for spermatogenesis (Amiri et al., 2001), and in *Drosophila*, removal of the maternal contribution of eIF4E-1 and eIF4E-2 results in females with no ovary development (Hernández et al., 2004b). The latter observation is in agreement with the finding that the interaction of eIF4E-1 with Cup and Barentsz is required for ovary development (Nakamura et al., 2004; Wilhelm et al., 2003; Zappavigna et al., 2004). During this developmental stage, eIF4E-1 plays a role in the ventral furrow development (Gong et al., 2004), and the interaction of eIF4E-1-Cup-Smaug is crucial for the proper space-regulated *nanos* translation (Nelson et al., 2004). These observations indicate that *Drosophila* eIF4E-1 is essential for ovary and embryo development. In fact, a lack of eIF4E-1 activity in *Drosophila* embryogenesis is lethal (Hernández et al., 2004b). On the other hand, in *S. pombe*, eIF4E-1 supports general cap-dependent translation, whereas eIF4E-2 is involved in translation during stress response (Ptushkina et al., 1996, 2004).

Due to differential cap-binding activities some eIF4Es play a role in the selection of the type of mRNA to be translated. In plants, wheat eIF4E and eIF(iso)4E (Carberry et al., 1991) as well as *A. thaliana* nCBP and eIF(iso)4E (Ruud et al., 1998) bind methylated cap structures with different affinities in vitro. In *C. elegans*, the in vitro activity of the five eIF4Es is related to the differential recognition of mono- and trimethylated mRNA cap structures present in this organism. IFE-3 binds only 7-methylguanosine caps and is essential for cell viability. In contrast, IFE-1, IFE-2, and IFE-5 are non-essential proteins and bind 2,2,7-trimethylguanosine caps, a structure present in 70% of the mRNAs from *C. elegans* (Jankowska-Anyszka et al., 1998; Keiper et al., 2000). Finally, in *C. elegans*, a small subset of mRNAs, most of them related to egg laying, are recognized specifically by IFE-4 in vivo (Dinkova et al., 2005).

The differences in activity between eIF4E proteins of a particular organism have been studied in vivo by complementation experiments of a conditionally lethal yeast mutant deficient in eIF4E. Human eIF4E-1 (Altmann et al., 1989), zebra fish eIF4E-1A (Robalino et al., 2004), *A. thaliana* eIF4E (Rodriguez et al., 1998), and *Drosophila* eIF4E-1, eIF4E-2, eIF4E-3, eIF4E-4, and eIF4E-7 (Hernández et al., 2005) rescue yeast growth in the absence of endogenous eIF4E. This might reflect the finding that different binding affinities for eIF4G and also for eIF4E-BPs (eIF4E-binding proteins) have been found for the eight *Drosophila* (Hernández et al., 2005) and the three mammalian (Joshi et al., 2004) eIF4Es.

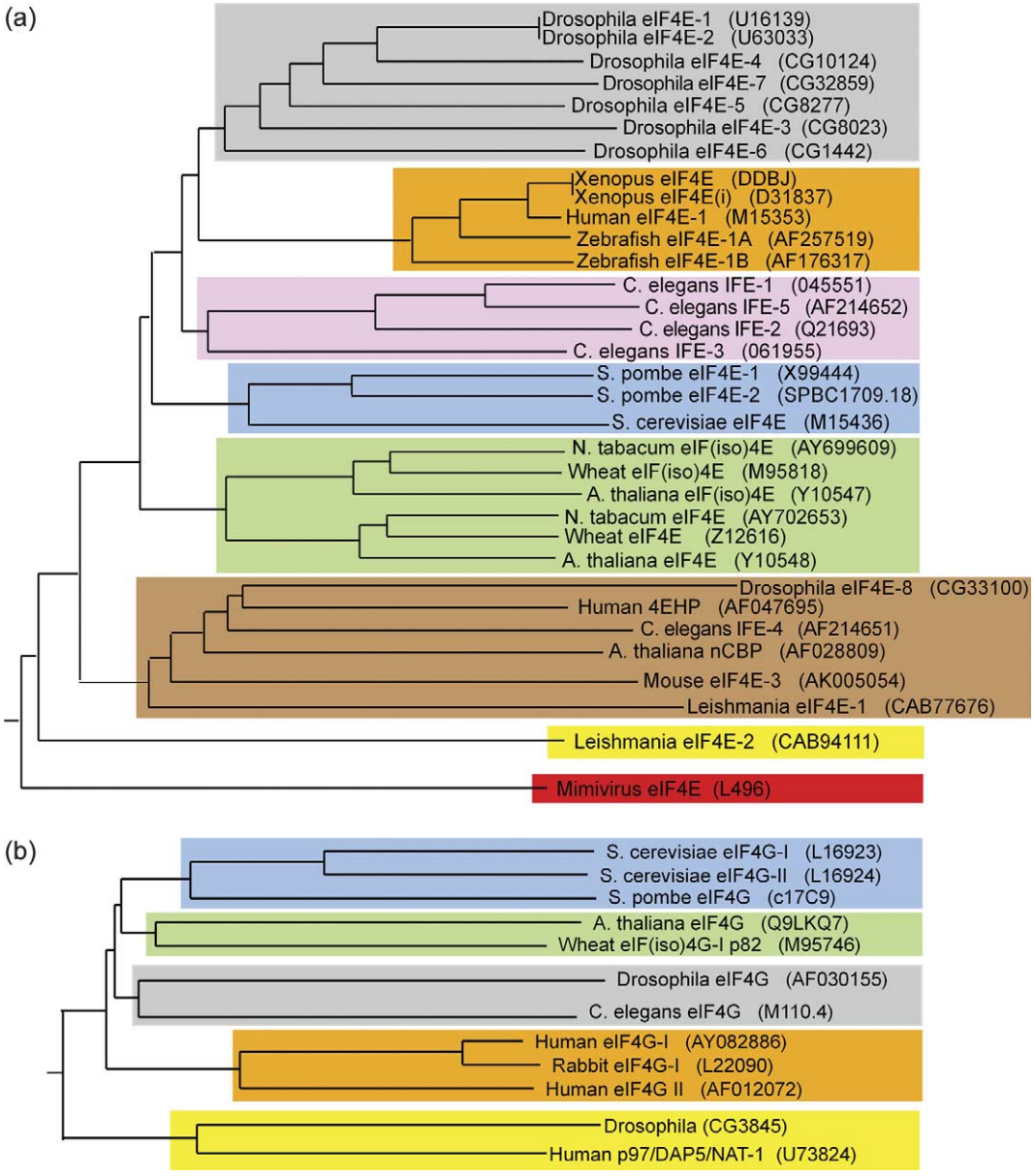

Fig. 1. Evolutionary relationships of (a) eIF4E or (b) eIF4G family members. Cladograms were constructed using the CLUSTAL W algorithm (Thompson et al., 1994) in the Megaline program of the DNA Star software package. Since the carboxy-terminal moiety of eIF4E is highly conserved and contains all the functional residues (Gingras et al., 1999; von der Haar et al., 2004), it was used to construct the eIF4Es tree. For a definition and comparison of the carboxy-terminal moiety of eIF4Es, see Hernández et al. (2005). The accession numbers are in parenthesis.

Interestingly, although eIF4E and most of its cognates are thought to promote initiation of translation, some members of this family have diverged in function. Human 4EHP shares sequence similarity and is structurally related to eIF4E (Rom et al., 1998). However, 4EHP and its closest ortholog *Drosophila* eIF4E-8/d4EHP (Fig. 1a) bind to the cap structure but not to eIF4G (Hernández et al., 2005; Rom et al., 1998), and thus may act as translational repressors. Indeed, Cho et al. (2005) showed that during *Drosophila* embryogenesis, eIF4E-8/d4EHP binds to both to the cap of caudal mRNA and to Bicoid, thereby inhibiting the cap-dependent translation of the caudal mRNA in the anterior of the embryo. Moreover, since *Drosophila* eIF4E-6 binds the cap structure but does not interact with eIF4G and also since neither complements the yeast eIF4E, this may indicate that eIF4E-6 can also act as a translational repressor (Hernández et al., 2005). It was also proven that, although is expressed, zebra fish eIF4E-1B does not bind to the cap and eIF4G (Robalino et al., 2004).

As depicted in Table 1, eIF4E cognates from several species are differentially expressed in the tissues and/or at different developmental stages which suggests a degree of functional diversity. Interestingly, differences in the cellular location of eIF4E cognates (either cytoplasmic or both nuclear and cytoplasmic) have been found between species (Table 1). Overall, for organisms with several eIF4E-related

Table 1
Location of eIF4 mRNA and/or proteins

Protein	Known localization and pattern of expression	Source
eIF4E cognates		
At eIF4E	mRNA expressed in all tissues, except in some root cells	Rodriguez et al. (1998)
At eIF(iso)4E	mRNA enriched in floral and young tissues	Rodriguez et al. (1998)
Ce IFE-1	mRNA detected in all cells in early embryogenesis and enriched in the germ line from larvae 3 on. Protein predominantly present in the adult germ line associated with P granules	Amiri et al. (2001)
Ce IFE-3 and 5	Protein is predominantly present in the adult germ line cells	Amiri et al. (2001)
Ce IFE-2 and 4	Protein is predominantly present in the adult somatic cells	Amiri et al. (2001)
Dm eIF4E-1	mRNA upregulated during autophagic cell death and enriched in gonads, especially ovaries. Cytoplasmic protein. Constitutively expressed throughout development, with the highest mRNA accumulation at the early embryonic stage, especially in the embryo pole cells. Accumulation of the protein in the pole of oocyte	Hernández et al. (2005), Hernández et al. (1997), Hernández and Sierra (1995), Nakamura et al. (2004), Wilhelm et al. (2003), Zappavigna et al. (2004), Gorski et al. (2003), and Parisi et al. (2004)
Dm eIF4E-3, 4, 5, 6 and 7	mRNA expressed from larve 2 stage on, with a peak in pupa	Hernández et al. (2005)
Dm eIF4E-5	mRNA upregulated during autophagic cell death	Gorski et al. (2003)
Dm eIF4E-8/d4EHP	mRNA expressed in early embryogenesis	Hernández et al. (2005)
L eIF4E	Cytoplasmic protein	Yoffe et al. (2004)
M eIF4E-1	mRNA ubiquitous and enriched in testis and skeletal muscle. Nuclear and cytoplasmic protein	Joshi et al. (2004), Miyagi et al. (1995) and Dostie et al. (2000)
M eIF4E-3	mRNA expressed in heart, lung, and skeletal muscle	Joshi et al. (2004)
M 4EHP	mRNA ubiquitous but enriched in testis. Low expression in heart and brain	Joshi et al. (2004)
Sc eIF4E	Nuclear and cytoplasmic protein	Lejbkowicz et al. (1992)
W eIF4E	Protein constitutively expressed during seed development, with the lowest expression during early stages of it. By 5–7 days germination, the protein level declined in leaves but remained high in scutella and roots	Gallie et al. (1998)
W eIF(iso)4E	Constant expression of the protein during seed development up to mid-development. By 5–7 days germination, the protein level declined in leaves but remained high in scutella and roots	Gallie et al. (1998)
Xl eIF4Es	mRNA constitutively expressed during oocyte and embryo development, with the strongest expression in early oocytes	Wakiyama et al. (1995)
Z eIF4E-1A	mRNA ubiquitously and constitutively expressed with the highest accumulation in ovary	Fahrenkrug et al. (1999) and Robalino et al. (2004)
Z eIF4E-1B	mRNA expressed in early embryonic development, muscle, gonads and erythrocytes. Asymetric expression in embryo	Fahrenkrug et al. (1999) and Robalino et al. (2004)
eIF4G cognates		
Dm CG3845	mRNA upregulated during autophagic cell death	Gorski et al. (2003)
M eIF4G-I	mRNA expressed in all tissues. Upregulated in liver and testis. Nuclear and cytoplasmic protein	Gradi et al. (1998) and McKendrick et al. (2001)
M eIF4G-II	mRNA expressed in all tissues, with the lowest amount in lung, heart, liver, and placenta. Upregulated in testis and fetal brain	Gradi et al. (1998)
M p97/NAT-1/DAP5	mRNA ubiquitously expressed in all tissues	Imataka et al. (1997), Levy-Strumpf et al. (1997), and Yamanaka et al. (1997)
W eIF4G	Protein constitutively expressed during seed development. Steady increase of the levels throughout it. By 3 days of germination, the protein is present in high level in the embryo scutello and expanding shoot, but is present at lower level in roots. By 5–7 days of germination, the protein is not detectable in any tissue	Gallie et al. (1998)
W eIF(iso)4G	Protein constitutively expressed during seed development. Steady decrease of the levels throughout it. By 5 days of germination, the protein is present in all tissues but is not detectable in 7-day-old leaves	Gallie et al. (1998)
eIF4A cognates		
Dm eIF4A-I	mRNA and protein ubiquitous in embryogenesis, constitutively expressed during development	Dorn et al. (1993) and Hernández et al. (2004a)
Dm eIF4A-III	Nuclear protein	Palacios et al. (2004)
M eIF4A-I	mRNA ubiquitous with the highest level in thymus. In excess over eIF4A-II in almost all tissues. Cytoplasmic protein	Nielsen and Trachsel (1988), Chan et al. (2004), Ferraiuolo et al. (2004), Palacios et al. (2004), and Shibuya et al. (2004)

(*continued on next page*)

Table 1 (*continued*)

Protein	Known localization and pattern of expression	Source
M eIF4A-II	mRNA expressed in all tissues. Enriched in skeletal muscle, brain, and gonads, with the lower amounts of mRNA in liver, pancreas, thymus, and spleen	Nielsen and Trachsel (1988) and Sudo et al. (1995)
M eIF4A-III	mRNA expressed in all tissues. Enriched in testis and heart and placenta, with lower amounts of mRNA in leukocytes, ovary and brain. Nuclear and cytoplasmic protein	Li et al. (1999), Chan et al. (2004), Ferraiuolo et al. (2004), Palacios et al. (2004), and Shibuya et al. (2004)
Nt eIF4A-2 and 3	mRNA ubiquitous. Lowest expression in fruit, young leaves, and sepal and the highest in root, stem, polen and shoot apex. mRNA of eIF4A-3 is highly enriched in petal	Owttrim et al. (1991, 1994) and Brander and Kuhlemeier (1995)
Nt eIF4A-5, 9, 11, and 15	mRNA ubiquitous. Lowest expression in stem, roots, and fruit. eIF4A-5 is the least expressed in all organs	Owttrim et al. (1994)
Nt eIF4A-8	mRNA expressed in anther and mature pollen	Brander and Kuhlemeier (1995) and op den Camp and Kuhlemeier (1998)
Xl eIF4A-I	mRNA expressed at all embryonic stages and tissues	Morgan and Sargent (1997)
Xl eIF4A-II	mRNA present in low amounts prior to stage 11 and increased sharply thereafter, localized only in dorsal ectoderm. mRNA is 30 times more aboundant than that of eIF4A-I in some embryonic stages	Morgan and Sargent (1997)
Xl eIF4A-III	mRNA constitutively expressed with asymetric spatial expression during embryogenesis. Enriched after stage 9.5 in ventral ectoderm of gastrula	Weinstein et al. (1997)
W eIF4A	Protein constitutively expressed during seed development with highest levels during the early stages. By 5–7 days germination, the protein level declined in leaves but remained high in scutella and roots	Gallie et al. (1998)
eIF4B cognates		
Dm eIF4B-S and -L	mRNA and proteins ubiquitous in embryogenesis, constitutively expressed during development with a peak during early embryonic development. Three times higher amount of eIF4BS than eIF4B-L at all stages. Cytoplasmic protein	Hernández et al. (2004c)
W eIF4B	Level of protein increases to a maximum during mid-development of the seed before declining. By 3 days of germination, the protein is present in high level in the embryo scutello and expanding shoot, but is present at lower level in roots. By 5–7 days of germination, the protein is aboundant in leaves but not detectable in scutella and roots	Gallie et al. (1998)

At, *Arabidopsis thaliana*; Ce, *Caenorhabditis elagans*; Dm, *Drosophila melanogaster*; L, *Leishmania*; M, mammalian; Nt, *N. tabacum*; Sc, *Saccharomyces cerevisiae*; Sp, *Schizosaccharomyces pombe*; W, wheat germ; Xl, *Xenopus laevis*; Z, zebra fish.

proteins, only a single eIF4E isoform that is ubiquitously and constitutively expressed seems to be responsible for routine cap-dependent translation. This isoform is represented by *Drosophila* eIF4E-1 (Hernández et al., 2005; Hernández and Sierra, 1995), mammalian eIF4E-1 (Joshi et al., 2004), *C. elegans* IFE-3 (Keiper et al., 2000), zebra fish eIF4E-1A (Robalino et al., 2004), and plant eIF4E (Rodriguez et al., 1998) (Table 2). The other eIF4E-related proteins may be eIF4E isoforms that are active only in particular tissues or bind to only certain mRNAs. This confers a considerable amount of versatility to the activity of eIF4E or eIF4E-structurally related proteins with a completely different function, as demonstrated for eIF4E-8/d4EHP (Cho et al., 2005) (Tables 1 and 2).

Furthermore, human eIF4E-1 has been recently found to also play an additional role as it is involved in nuclear mRNA transport of a subset of specific mRNAs (Strudwick and Borden, 2002). This observation indicates that eIF4E is versatile enough to utilize the features required for cap-binding activity for participating in other processes in the cell (Strudwick and Borden, 2002).

3. Unknown eIF4G

So far, two eIF4G proteins have been characterized from human, eIF4G-I (Bradley et al., 2002; Byrd et al., 2002; Lloyd et al., 1987; Tahara et al., 1981; Yan et al., 1992) and eIF4G-II (Gradi et al., 1998); one from rabbit, eIF4G-I (Lamphear et al., 1993); two from wheat, eIF4G (Lax et al., 1985, 1986) and eIF(iso)4G (Allen et al., 1992; Browning et al., 1987, 1992; Lax et al., 1985, 1986) with ortholog genes in other plants (Browning, 1996; Browning et al., 1992); two from *S. cerevisiae*, TIF4631 and TIF4632 (Goyer et al., 1993); one from *S. pombe* (Hashemzadeh-Bonehi et al., 2003); and one from *Drosophila*, Dm eIF4G (Hernández et al., 1998; Zapata et al., 1994). Human p97/NAT1/DAP-5 is an eIF4G-related protein that contains

Table 2
Known activities of eIF4 proteins

Protein	Activity	Source
eIF4E cognates		
Dm eIF4E-1, M eIF4E-1, Ce IFE-3, Sp eIF4E-1, Sc eIF4E, Plant eIF4E, Z eIF4E-1A	Supports general cap-dependent initiation of translation by recognizing the cap structure (7-methyl guanosine). Essential gene	Browning (1996), Altmann et al. (1987), Fahrenkrug et al. (1999), Gingras et al. (1999), Hershey and Merrick (2000), Jankowska-Anyszka et al. (1998), Pain (1996), Pestova and Hellen (2000), Rodriguez et al. (1998), Maroto and Sierra (1989) and Hernández et al. (2005)
Dm eIF4E-1	Required for ovary and embryo development	Gong et al. (2004), Hernández et al. (2004b), Nakamura et al. (2004), Nelson et al. (2004), Wilhelm et al. (2003), and Zappavigna et al. (2004)
M eIF4E-1	mRNA nucleo-cytoplam transport	Strudwick and Borden (2002)
Plant eIF(iso)4E	Supports general cap-dependent initiation of translation by recognizing the cap structure	Browning (1996)
Sp eIF4E-2	Supports cap-dependent initiation of translation during stress response	Ptushkina et al. (2004)
Ce IFE-1, IFE-2, IFE-5	Binds to 2,2,7 trimetyl cap structures. Non-essential gene	Jankowska-Anyszka et al. (1998) and Keiper et al. (2000)
Ce IFE-1	Required for spermatogenesis	Amiri et al. (2001)
Ce IFE-4	Involved in expression of specific mRNAs involved in egg lying. Non-essential gene	Dinkova et al. (2005)
Dm eIF4E-8/d4EHP	Negative translation regulator	Cho et al. (2005) and Hernández et al. (2005)
eIF4G cognates		
M eIF4G-I and eIF4G-II, Dm eIF4G, Sc eIF4G-I and eIF4G-II, plant eIF4G	Scaffold protein. Supports general cap- and IRES-dependent initiation of translation	Browning (1996), Gallie and Browning (2001), Gingras et al. (1999), Hentze (1997), Hershey and Merrick (2000), Pain (1996), Pestova and Hellen (2000), Prevot et al. (2003), and Zapata et al. (1994)
p97/NAT-1/DAP5	Translational inhibitor of cap-dependent mRNAs. Supports translation initiation of IRES-driven mRNAs encoding for proteins involved in apoptosis	Imataka et al. (1997), Yamanaka et al. (1997), Levy-Strumpf et al. (1997), Henis-Korenblit et al. (2002), and Warnakulasuriyarachchi et al. (2004)
Plant eIF(iso)4G	Supports general cap-dependent initiation of translation	Browning (1996)
eIF4A cognates		
M eIF4A-I, W eIF4A-I, Sc eIF4A	ATP-dependent RNA helicase. Supports general cap- and IRES-dependent initiation of translation. Essential gene	Browning (1996), Gingras et al. (1999), Hershey and Merrick (2000), Pain (1996), Pestova and Hellen (2000), and Rogers et al. (2002)
M eIF4A-III, Dm eIF4A-III	Involved in splicing, mRNA localization and mRNA decay. Essential gene	Chan et al. (2004), Ferraiuolo et al. (2004), Palacios et al. (2004), and Shibuya et al. (2004)
eIF4B cognates		
M eIF4B and eIF4H, Dm eIF4B, Sc eIF4B, W eIF4B	Supports general initiation of translation. Non-essential gene	Browning (1996), Gingras et al. (1999), Hershey and Merrick (2000), López de Quinto et al. (2001), Pain (1996), Pestova and Hellen (2000) and Richter-Cook et al. (1998), and Hernández (2004c)

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the domains present in eIF4G allowing for interaction with both eIF3 and eIF4A (Imataka et al., 1997; Levy-Strumpf et al., 1997; Yamanaka et al., 1997). In *Drosophila*, the annotated gene CG3845 encodes an ortholog of p97/NAT-1/DAP5. The phylogenetic relationships depicted in Fig. 1b show that eIF4G-related proteins are divided into the following groups: (1) fungi proteins (blue); (2) plants proteins (green); (3) *Drosophila* eIF4G and *C. elegans* eIF4G (gray); (4) mammalian eIF4Gs (orange); and (5) human p97/NAT-1/DAP5 and *Drosophila* CG3845 proteins (yellow). The last group displays the least similarity to all other eIF4G cognates.

The role of eIF4G as a scaffold during the initiation of translation is well documented (Browning, 1996; Gingras et al., 1999; Hentze, 1997; Hershey and Merrick, 2000; Pain, 1996; Pestova and Hellen, 2000; Prevot et al., 2003). Although functional diversity of a few eIF4G-related proteins has also been established, this is not as common as for the eIF4E and eIF4A families (see latter in the text). In yeast, the translation of HSP101 mRNA specifically requires TIF4632 to be translated under certain stress conditions (Wells et al., 2004); whereas in mammals, capped mRNAs are specifically translated by eIF4G-II at the onset of hematopoietic cell differentiation (Caron et al., 2004). In plants, eIF4G is more efficient than eIF(iso)4G in supporting translation of structured mRNAs, and thereby supports internal initiation of translation (Gallie and Browning, 2001). Mammalian eIF4G-1 also associates with the nuclear cap-binding complex (CBC) (McKendrick et al., 2001), which raises the possibility that eIF4G-1 may also mediate mRNA export from the nucleus to cytoplasm (Prevot et al., 2003). p97/NAT-1/DAP5 seems to be a general translational

inhibitor (Imataka et al., 1997; Yamanaka et al., 1997). However, when cleaved by caspases, p97/NAT-1/DAP5 promotes the translation of IRES-dependent mRNAs whose products are involved in apoptosis, like its own mRNA as well as mRNAs encoding for c-Myc, XIAP, Apaf-1 (Henis-Korenblit et al., 2002), and HIAP2 (Warnakulasuriyarachchi et al., 2004).

Temporal and spatial expression has also been observed in tissues for some eIF4G-related proteins (Table 1). However, to date, the biological relevance of this expression is still totally unknown, and more work is needed to understand the physiological relevance of all eIF4G-related proteins. Currently, the eIF4G family remains one of the least characterized of all the eIF4 families.

4. Ancient eIF4A

Three eIF4A proteins have been studied in humans, eIF4A-I, eIF4A-III (Li et al., 1999), and eIF4A-II (Sudo et al., 1995); two in mouse, eIF4A-I (Nielsen et al., 1985) and eIF4A-II (Nielsen and Trachsel, 1988); one in rabbit (Conroy et al., 1990); three in *Xenopus* (Morgan and Sargent, 1997; Weinstein et al., 1997); one in *S. cerevisiae* encoded by the genes *TIF-1* and *TIF-2* (Linder and Slonimski, 1989); two in *Drosophila*, Dm eIF4A (Dorn et al., 1993; Hernández et al., 2004a) and Dm eIF4A-III (Palacios et al., 2004); and one in *E. coli* (Lu et al., 1999). While two genes have been cloned in *A. thaliana* (Metz et al., 1992b) and one in wheat (Metz and Browning, 1993), more than 10 *eIF4A* genes have been identified in tobacco (Brander et al., 1995; Owttrim et al., 1994). Other putative *eIF4A* genes, identified by sequence similarity to those already characterized, have been found in many species including mouse, rice, and *C. albicans* (Fig. 2a). The phylogenetic relationships of eIF4A family members show that they are grouped in six clades (Fig. 2a): (1) plant proteins (green); (2) mammalian, *Xenopus* eIF4A classes I and II, together with *Drosophila* eIF4A (orange); (3) fungi proteins (blue); (4) eIF4A-III (yellow); (5) *E. coli* eIF4A (red); and (6) archaeobacteries (violet).

The universal necessity for RNA helicases in cell metabolism is undisputed (Lüking et al., 1998). The translation initiation factor eIF4A-I, which has been extensively characterized in mammals, yeast and wheat, is an RNA helicase that is active during the initiation of translation (Browning, 1996; Gingras et al., 1999; Hershey and Merrick, 2000; Pain, 1996; Pestova and Hellen, 2000; Rogers et al., 2002). An eIF4A-related protein, eIF4A-III, was recently discovered to play an unrelated role than in translation. Human and *Drosophila* eIF4A-III form part of the exon junction complex which is formed during the splicing process of mRNAs. This complex is essential for nonsense-mediated mRNA decay in mammals (Chan et al., 2004; Ferraiuolo et al., 2004; Palacios et al., 2004; Shibuya et al., 2004). In *Drosophila*, eIF4A-III is also essential for the proper localization of *oskar* mRNA in oocytes (Palacios et al., 2004). Null Dm eIF4A mutants were found to be lethal (Dorn et al., 1993), which further indicates that eIF4A and eIF4A-III play different, non-redundant roles. Hence, whereas eIF4A-I plays a major role in the initiation of translation, eIF4A-III might serve as a link between splicing, mRNA localization, mRNA decay, and cell differentiation. Although eIF4A-II could be an isoform of eIF4A-I, its real function is not known. Moreover, there is so far no plausible explanation for the disparity in the number of *eIF4A* genes identified among plants.

Members of the eIF4A family play a role in a variety of developmental processes. In *Xenopus*, eIF4A-II seems to be involved in the development of neuroectodermus (Morgan and Sargent, 1997), and in tobacco, eIF4A-8 may play a role in the development of gametophyte (op den Camp and Kuhlemeier, 1998). Moreover, the activity of specific eIF4A-related proteins has been directly related to the growth status of the cell. This is the case for human eIF4A-I, whose expression is upregulated in melanomas (Eberle et al., 1997). Also, *Drosophila* eIF4A (but not eIF4A-III) is overexpressed in wing imaginal discs in the tumor-suppressor mutants *ft* and *l(2)gd*, in which excessive cell proliferation occurs (Hernández et al., 2004a). In addition, mutations in *Drosophila eIF4A* gene affect larval growth, cell proliferation, and DNA replication (Galloni and Edgar, 1999). In mouse, eIF4A-I is synthesized preferentially in growing cells, while eIF4A-II synthesis is associated with growth-arrested cells (Nielsen and Trachsel, 1988; Williams-Hill et al., 1997).

Since eIF4A-I and eIF4A-III are present in very divergent taxonomical groups, including mammals, plants, *Xenopus*, fungi, and *Drosophila* (Fig. 2a), it is possible that both eIF4A proteins are paralogous genes that were present in an ancestor of nowadays eukaryotic cells. It is interesting that eIF4A-I is unique among eIF4s in archeobacteries (Dennis, 1997) (also see Fig. 2a). Archaeal mRNAs lack cap structure, and neither eIF4E nor eIF4G homologs appear to be present in these organisms (Dennis, 1997). Since the nucleous-cytoplasm of eukaryotic cells evolved from proto-archaeabacterial cells (Woese, 2000), eIF4A-I could already have been present in the Archaea translational apparatus of an eukaryotic ancestor.

As for the members of other eIF4 families, the tissue- and temporal-specific expression of the eIF4A family members is well documented for several organisms, although the functional significance of these differences is unknown (Table 1).

5. Evasive eIF4B and eIF4H

To date, characterized members of the eIF4B and eIF4H family include human eIF4B (Milburn et al., 1990), wheat eIF4B, *A. thaliana* eIF4B-1 and eIF4B-2 (Metz et al., 1999), *S. cerevisiae TIF3* (Milburn et al., 1990), and *Drosophila*

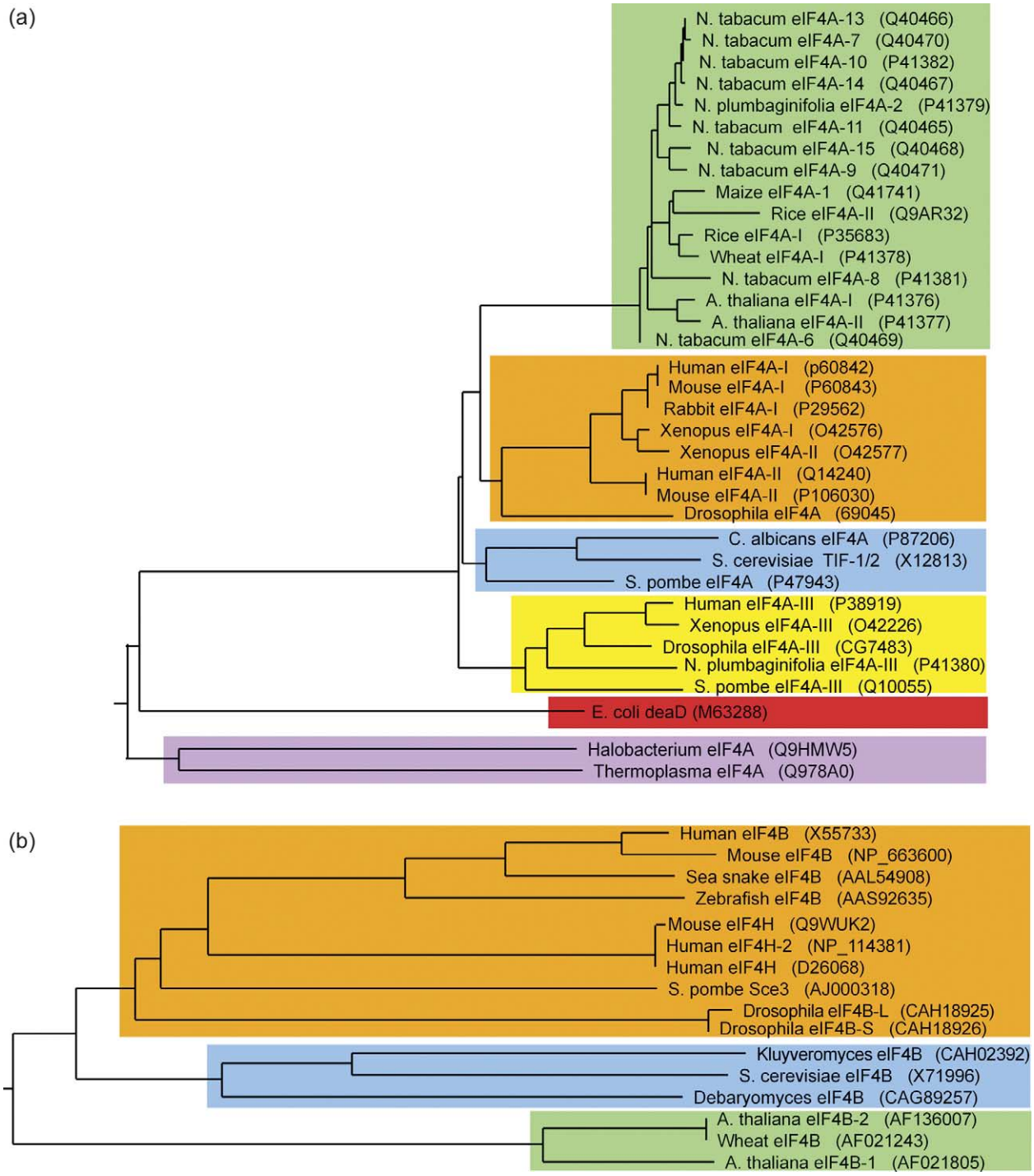

Fig. 2. Evolutionary relationships of (a) eIF4A or (b) eIF4B and eIF4H family members. Cladograms were constructed as described in Fig. 1. The accession numbers are in parenthesis.

eIF4B-L and eIF4B-S (Hernández et al., 2004c). Genes encoding putative eIF4B have also been identified in the genome of other organisms (Fig. 2b). Human eIF4H displays functional similarities with human eIF4B (Richter-Cook et al., 1998). *S. pombe SCE3* encodes an RNA-binding protein involved in cell division (Schmidt et al., 1997) that appears to be a fungi ortholog of eIF4H (Fig. 2b). The evolutionary relationships depicted in Fig. 2b show that eIF4Bs from plants (green), fungi (blue), and those from mammalian, zebra fish, sea snake, and *Drosophila*, together with eIF4Hs and *SCE3* (orange), form three separate groups.

The involvement of eIF4B and eIF4H during initiation of translation is well established (Browning, 1996; Gingras et al., 1999; Hershey and Merrick, 2000; López de Quinto et al., 2001; Pain, 1996; Pestova and Hellen, 2000;

Richter-Cook et al., 1998). However, the study of the expression of the eIF4B isoforms in different tissues or during development has been addressed only in *Drosophila* (Hernández et al., 2004c) and in wheat during seed development and germination (Gallie et al., 1998) (Table 1). Until now, there are no reports concerning the expression of eIF4H. The lack of information about eIF4B and eIF4H may in part be due to the fact that both proteins are the least conserved of all eIF4s, with essentially no sequence similarity between the eIF4Bs from yeast and mammals with those from plants (Metz et al., 1999). This renders their identification in other organisms more difficult.

6. Concluding remarks

All protein members from each eIF4 multigenic family are structurally- and sequence-related proteins, and although their general function has been known for quite a while, their functional diversity has only recently begun to be recognized (Table 2). On the other side, a recent survey of the human genome detected 199 proteins containing potential eIF4E-binding sites, and a model for the regulation of eIF4E activity in a tissue-specific context was proposed (Topisorivic et al., 2003). Here, we extend this model to the members of all eIF4 families that could be regulated in a tissue- and/or temporal-specific manner by different interactors.

6.1. The combinatorial eIF4F complexes formation

In multicellular organisms, many genes are expressed in a developmental- and tissue-specific manner. In many cases, regulation of this expression is performed at the level of initiation of translation. The existence of families of all eIF4s and combination of members thereof would allow for the formation of various eIF4F complexes with different biochemical properties and functions. Although in some cases, redundancy of functionality may also exist, eIF4Fs with different components may confer selectivity for mRNA translation under certain conditions, or in different tissues or at developmental stages. This phenomenon would imply an additional level of regulation of protein synthesis. The involvement of some eIF4 isoforms in developmental- or tissue-specific processes as well as in the translation of certain mRNAs argue for this scenario.

6.2. Perspectives for functional studies on eIF4 families members

The expression and specific activity of most of the newly identified eIF4 factors is largely uninvestigated and new studies are needed in order to understand the physiological meaning of the wide diversity of the eIF4 families. On the other hand, the recent discovery that some eIF4 proteins are translational repressors (e.g. *Drosophila* eIF4E-8/d4EHP) or even have a role in a process other than translation (e.g. eIF4E-1 and eIF4A-III) indicates that the diversity of the eIF4 families has a greater biological significance than previously thought. Whether other members of eIF4 families have a role in a process other than translation remains to be seen. Thus, these observations have opened a new and unsuspected line of investigation in an area of research still in its infancy.

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References

Allen, M.L., Metz, A.M., Timmer, R.T., Rhoads, R.E., Browning, K.S., 1992. Isolation and sequence of the cDNAs encoding the subunits of the isozyme of wheat protein synthesis initiation factor 4F. J. Biol. Chem. 267, 23232–23236.

Altmann, M., Handschin, C., Trachsel, H., 1987. mRNA cap-binding protein: cloning of the gene encoding protein synthesis initiation factor eIF-4E from *Saccharomyces cerevisiae*. Mol. Cell. Biol. 7, 998–1003.

Altmann, M., Müller, P.P., Pelletier, J., Sonenberg, N., Trachsel, H., 1989. A mammalian translation initiation factor can substitute for its yeast homologue in vivo. J. Biol. Chem. 264, 12145–12147.

Amiri, A., Keiper, B.D., Kawasaki, I., Fan, Y., Kohara, Y., Rhoads, R.E., 2001. An isoform of eIF4E is a component of germ granules and is required for spermatogenesis in *C. elegans*. Development 128, 3899–3912.

Berthelot, K., Muldoon, M., Rajkowitsch, L., Hughes, J., McCarthy, J.E., 2004. Dynamics and processivity of 40S ribosome scanning on mRNA in yeast. Mol. Microbiol. 51, 987–1001.

Bradley, C.A., Padovan, J.C., Thompson, J.L., Benoit, C.A., Chait, B.T., Rhoads, R.E., 2002. Mass spectrometric analysis of the N terminus of translational initiation factor eIF4G-1 reveals novel isoforms. J. Biol. Chem. 277, 12559–12571.

Brander, K.A., Kuhlemeier, C., 1995. A pollen-specific DEAD-box protein related to translation initiation factor eIF-4A from tobacco. Plant Mol. Biol. 27, 637–649.

Brander, K.A., Mandel, T., Owttrim, G.W., Kuhlemeier, C., 1995. Highly conserved genes coding for eukaryotic translation initiation factor eIF-4A of tobacco have specific alterations in functional motifs. Biochem. Biophys. Acta 1261, 442–444.

Browning, K.S., 1996. The plant translational apparatus. Plant Mol. Biol. 32, 107–144.

Browning, K.S., Maia, D.M., Lax, S., Ravel, J.M., 1987. Identification of a new protein synthesis initiation factor from wheat germ. J. Biol. Chem. 262, 538–541.

Browning, K.S., Webster, C., Roberts, J.K., Ravel, J.M., 1992. Identification of an isozyme from protein synthesis initiation factor 4F in plants. J. Biol. Chem. 267, 10096–10100.

Byrd, M.P., Zamora, M., Lloyd, R.E., 2002. Generation of multiple isoforms of eukaryotic translation initiation factor 4GI by use of alternate translation initiation codons. Mol. Cell. Biol. 22, 4499–4511.

Carberry, S.E., Darzynkiewicz, E., Goss, D.J., 1991. A comparison of the binding of methylated cap analogues to wheat germ protein synthesis initiation factors 4F and (iso)4F. Biochemistry 30, 1624–1627.

Caron, S., Charon, M., Cramer, E., Sonenberg, N., Isabelle, Z., Dusanter-Fourt, I., 2004. Selective modification of eukaryotic initiation factor 4F (eIF4F) at the onset of cell differentiation: recruitment of eIF4GII and long-lasting phosphorylation of eIF4. Mol. Cell. Biol. 24, 4920–4928.

Chan, C.C., Dostie, J., Diem, M.D., Feng, W., Mann, M., Rappsilber, J., Dreyfuss, G., 2004. eIF4A3 is a novel component of the exon junction complex. RNA 10, 200–209.

Cho, P.F., Poulin, F., Cho-Park, Y.A., Cho-Park, I.B., Chicoine, J.D., Lasko, P., Sonenberg, N., 2005. A new paradigm for translational control: inhibition via 5′–3′ mRNA tethering by Bicoid and the eIF4E cognate 4EHP. Cell 121, 411–423.

Conroy, S.C., Dever, T.E., Owens, C.L., Merrick, W.C., 1990. Characterization of the 46,000-dalton subunit of the eIF-4F. Arch. Biochem. Biophys. 282, 363–371.

Dennis, P.P., 1997. Ancient ciphers: translation in Archaea. Cell 89, 1007–1010.

Dinkova, T.D., Keiper, B., Korneeva, N.L., Aamodt, E.J., Rhoads, R.E., 2005. Translation of a small subset of *Caenorhabditis elegans* mRNAs is dependent on a specific eukaryotic translation initiation factor 4E isoform. Mol. Cell. Biol. 25, 100–113.

Dorn, R., Morawietz, H., Reuter, G., Saumweber, H., 1993. Identification of an essential *Drosophila* gene that is homologous to the translation initiation factor eIF-4A of yeast and mouse. Mol. Gen. Genet. 237, 233–240.

Dostie, J., Lejbkowicz, F., Sonenberg, N., 2000. Nuclear eukaryotic initiation factor 4E (eIF4E) colocalizes with splicing factors in spackles. J. Cell Biol. 148, 239–147.

Eberle, J., Krasagakis, K., Orfanos, C.E., 1997. Translation initiation factor eIF-4A1 mRNA is consistently overexpressed in human melanoma cells in vitro. Int. J. Cancer 71, 396–401.

Fahrenkrug, S.C., Dahlquist, M.O., Clark, K.J., Hackett Jr.., P.B., 1999. Dynamic and tissue-specific expression of eIF4E during zebrafish embryogenesis. Differentiation 65, 191–201.

Ferraiuolo, M.A., Lee, C.-S., Ler, L.W., Hsu, J.L., Costa-mattioli, M., Luo, M.-J., et al., 2004. A nuclear translation-like factor eIF4AIII is recruited to the mRNA during splicing and functions in nonsense-mediated decay. Proc. Natl Acad. Sci. USA 101, 4118–4123.

Gallie, D.R., Browning, K.S., 2001. eIF4G functionally differs from eIFiso4G in promoting internal initiation, cap-independent translation, and translation of structured mRNAs. J. Biol. Chem. 276, 36951–36960.

Gallie, D.R., Le, H., Tanguay, R.L., Browning, K.S., 1998. Translation initiation factors are differentially regulated in cereals during development and following heat shock. Plant J. 14, 715–722.

Galloni, M., Edgar, B., 1999. Cell-autonomous and non-autonomous growth-defective mutants of *Drosophila melanogaster*. Development 126, 2365–2375.

Gingras, A.C., Raught, B., Sonenberg, N., 1999. eIF4 initiation factors: effectors of mRNA recruitment to ribosomes and regulators of translation. Annu. Rev. Biochem. 68, 913–963.

Gong, L., Puri, M., Unlu, M., Young, M., Robertson, K., Viswanathan, S., et al., 2004. *Drosophila* ventral furrow morphogenesis: a proteomic analysis. Development 131, 643–656.

Gorski, S.M., Chittaranjan, S., Pleasance, E., Freeman, J.D., Anderson, C.L., Varhol, R.J., et al., 2003. A SAGE approach to discovery of genes involved in autophagic cell death. Curr. Biol. 13, 358–363.

Goyer, C., Altmann, M., Lee, H.S., Blanc, A., Deshmukh, M., Woolford Jr.., J., et al., 1993. *TIF4631* and *TIF4632*: two yeast genes encoding the high-molecular-weight subunits of the cap-binding protein complex (eukaryotic initiation factor 4F) contain an RNA recognition motif-like sequence and carry out an essential function. Mol. Cell. Biol. 13, 4860–4874.

Gradi, A., Imataka, H., Svitkin, Y.V., Rom, E., Raught, B., Morino, S., Sonenberg, N., 1998. A novel functional human eukaryotic translation initiation factor 4G. Mol. Cell. Biol. 18, 334–342.

Hashemzadeh-Bonehi, L., Curtis, P.S., Morley, S.J., Thorpe, J.R., Pain, V.M., 2003. Overproduction of a conserved domain of fission yeast and mammalian translation initiation factor eIF4FG causes aberrant cell morphology and results in disruption of the localization of F-actin and the organization of microtubules. Genes Cells 8, 163–178.

Henis-Korenblit, S., Shani, G., Sines, T., Marash, L., Shohat, G., Kimchi, A., 2002. The caspase-cleaved DAP5 protein supports internal ribosome entry site-mediated translation of death proteins. Proc. Natl Acad. Sci. USA 99, 5400–5405.

Hentze, M.W., 1997. eIF4G: a multipurpose ribosome adapter? Science 275, 500–501.

Hernández, G., Sierra, J.M., 1995. Translation initiation factor eIF-4E from *Drosophila*: cDNA sequence and expression of the gene. Biochim. Biophys. Acta 1261, 427–431.

Hernández, G., Diez del Corral, R., Santoyo, J., Campuzano, S., Sierra, J.M., 1997. Localization, structure and expression of the gene for translation initiation factor eIF4E from *Drosophila melanogaster*. Mol. Gen. Genet. 253, 624–633.

Hernández, G., Castellano, M.M., Agudo, M., Sierra, J.M., 1998. Isolation and characterization of the cDNA and the gene for eukaryotic translation initiation factor 4G from *Drosophila melanogaster*. Eur. J. Biochem. 253, 27–35.

Hernández, G., Lalioti, V.S., Vandekerckhove, J., Sierra, J.M., Santarén, J.F., 2004a. Identification and characterization of the expression of the translation initiation factor 4A (eIF4A) from *Drosophila*. Proteomics 4, 316–326.

Hernández, G., Vazquez-Pianzola, P., Sierra, J.M., Rivera-Pomar, R., 2004b. Internal ribosome entry site drives cap-independent translation of *reaper* and *heat shock protein* 70 mRNAs in *Drosophila* embryos. RNA 10, 1783–1797.

Hernández, G., Vazquez-Pianzola, P., Zurbriggen, A., Altmann, M., Sierra, J.M., Rivera-Pomar, R., 2004c. Two functionally redundant isoforms of *Drosophila melanogaster* eukaryotic initiation factor 4B are involved in cap-dependent translation, cell survival and proliferation. Eur. J. Biochem. 271, 2923–2936.

Hernández, G., Altmann, M., Sierra, J.M., Urlaub, H., Diez del Corral, R., Schwartz, P., Rivera-Pomar, R., 2005. Functional analysis of seven genes encoding eight translation initiation factor 4E (eIF4E) isoforms in *Drosophila*. Mech. Dev. 122, 529–543.

Hershey, J.W.B., Merrick, W.C., 2000. The pathway and mechanism of initiation of protein synthesis. In: Sonenberg, N. (Ed.), Translational Control of Gene Expression Monographs, vol. 39. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY, pp. 33–38.

Imataka, H., Olsen, H.S., Sonenberg, N., 1997. A new translational regulator with homology to eukaryotic translation initiation factor 4G. Eur. Mol. Biol. Org. J. 16, 817–825.

Jankowska-Anyszka, M., Lamphear, B.J., Aamodt, E.J., Harrington, T., Darzynkiewicz, E., Stolarski, R., Rhoads, R.E., 1998. Multiple isoforms of eukaryotic protein synthesis initiation factor 4E in *Caenorhabditis elegans* can distinguish between mono- and trimethylated mRNA cap structures. J. Biol. Chem. 273, 10538–10542.

Joshi, B., Cameron, A., Jagus, R., 2004. Characterization of mammalian eIF4E-family members. Eur. J. Biochem. 271, 2189–2203.

Keiper, B.D., Lamphear, B.J., Deshpande, A.M., Jankowska-Anyszka, M., Aamodt, E.J., Blumenthal, T., Rhoads, R.E., 2000. Functional characterization of five eIF4E isoforms in *Caenorhabditis elegans*. J. Biol. Chem. 275, 10590–10596.

Kozak, M., 1978. How do eukaryotic ribosomes select initiation regions in messenger mRNA? Cell 22, 459–467.

Kozak, M., 2002. Pushing the limits of the scanning mechanism for initiation of translation. Gene 299, 1–34.

Lamphear, B.J., Yan, R., Yang, F., Waters, D., Liebig, H.D., Klump, H., et al., 1993. Mapping the cleavage site in protein synthesis initiation factor eIF-4 gamma of the 2A proteases from human Coxsackievirus and rhinovirus. J. Biol. Chem. 268, 19200–19203.

Lavoie, C.A., Lachance, P., Sonenberg, N., Lasko, P., 1996. Alternatively spliced transcripts from the *Drosophila* eIF4E gene produce two different cap-binding proteins. J. Biol. Chem. 271, 16393–16398.

Lax, S., Fritz, W., Browning, K., Ravel, J., 1985. Isolation and characterization of factors from wheat germ that exhibit eukaryotic initiation factor 4B activity and overcome 7-methylguanosine 5′-triphosphate inhibition of polypeptide synthesis. Proc. Natl Acad. Sci. USA 82, 330–333.

Lax, S., Lauer, S.J., Browning, K., Ravel, J.M., 1986. Purification and properties of protein synthesis initiation and elongation factors from wheat germ. Methods Enzymol. 118, 109–128.

Lejbkowicz, F., Goyer, C., Darveau, A., Neron, S., Lemieux, R., Sonenberg, N., 1992. A fraction of the mRNA 5′ cap-binding protein, eukaryotic initiation factor 4E, localizes to the nucleus. Proc. Natl Acad. Sci. USA 89, 9612–9616.

Levy-Strumpf, N., Deiss, L.P., Berissi, H., Kimchi, A., 1997. DAP-5, a novel homolog of eukaryotic translation initiation factor 4G isolated as aputative modulator of gamma interferon-induced programmed cell death. Mol. Cell. Biol. 17, 1615–1625.

Li, Q., Imataka, H., Morino, S., Rogers, G.W., Richter, N.J., Merrick, W.C., Sonenberg, N., 1999. Eukaryotic translation initiation factor eIF4AIII is functionally distinct from eIF4AI and eIF4AII. Mol. Cell. Biol. 19, 7336–7346.

Linder, P., Slonimski, P.P., 1989. An essential yeast protein, encoded by duplicated genes *TIF1* and *TIF2* and homologous to the mammalian translation initiation factor eIF-4A, can suppress a mitochondrial missense mutation. Proc. Natl Acad. Sci. USA 86, 2286–2290.

Lloyd, R.E., Jense, H.G., Ehrenfeld, E., 1987. Restriction of translation of capped mRNA in vitro as a model for poliovirus-induced inhibition of host-cell protein synthesis—relationship to p200 cleavage. J. Virol. 61, 2480–2488.

López de Quinto, S., Lafuente, E., Martínez-Salas, E., 2001. IRES interaction with translation initiation factors: functional characterization of novel RNA contacts with eIF3, eIF4B, and eIF4GII. RNA 7, 1213–1226.

Lu, J., Aoki, H., Ganoza, C., 1999. Molecular characterization of a prokaryotic translation factor homologous to the eukaryotic initiation factor eIF4A. Int. J. Biochem. Cell Biol. 31, 215–229.

Lüking, A., Stahl, U., Schmidt, U., 1998. The protein family of RNA helicases. Crit. Rev. Biochem. Mol. Biol. 33, 259–296.

Maroto, F.G., Sierra, J.M., 1989. Purification and characterization of mRNA cap-binding protein from *Drosophila melanogaster* embryos. Mol. Cell. Biol. 9, 2181–2190.

McKendrick, L., Thompson, E., Ferreira, J., Morley, S.J., Lewis, J.D., 2001. Interaction of eukaryotic translation initiation factor 4G with the nuclear cap-binding complex provides a link between nuclear and cytoplamic functions of the m(7) gusnosine cap. Mol. Cell. Biol. 21, 3632–3641.

Metz, A.M., Browning, K.S., 1993. Sequence of a cDNA encoding wheat eukaryotic protein synthesis initiation factor 4A. Gene 131, 299–300.

Metz, A.M., Timmer, R.T., Browning, K.S., 1992a. Isolation and sequence of a cDNA encoding the cap binding proteins of wheat eukaryotic protein synthesis factor 4F. Nucleic Acids Res. 20, 4096.

Metz, A.M., Timmer, R.T., Browning, K.S., 1992b. Sequences for two cDNAs encoding *Arabidopsis* eukaryotic protein synthesis initiation factor 4A. Gene 120, 313–314.

Metz, A.M., Wong, K.C., Malmstrom, S.A., Browning, K.S., 1999. Eukaryotic initiation factor 4B from wheat and *Arabidopsis thaliana* is a member of a multigene family. Biochem. Biophys. Res. Commun. 266, 314–321.

Milburn, S.C., Hershey, J.W.B., Davies, M.V., Kelleher, K., Kaufman, R.J., 1990. Cloning and expression of eukaryotic initiation factor 4B cDNA: sequence determination identifies a common RNA recognition motif. Eur. Mol. Biol. Org. J. 9, 2783–2790.

Miyagi, Y., Kerr, S., Sugiyama, A., Asai, A., Masabumi, S., Fujimoto, H., Kuchino, Y., 1995. Abundant expression of translational initiation factor eIF-4E in post-meiotic germ cells of the rat testis. Lab. Invest. 72, 890–898.

Morgan, R., Sargent, M.G., 1997. The role in neural patterning of translation initiation factor eIF4AII: induction of neural fold genes. Development 124, 2751–2760.

Nakamura, A., Sato, K., Hanyu-Nakamura, K., 2004. *Drosophila* Cup is an eIF4E binding protein that associates with Bruno and regulates oskar mRNA translation in oogenesis. Dev. Cell 6, 69–78.

Nelson, M.R., Leidal, A.M., Smibert, C.A., 2004. *Drosophila* Cup is an eIF4E-binding protein that functions in Smaug-mediated translational repression. Eur. Mol. Biol. Org. J. 23, 150–159.

Nielsen, P.J., Trachsel, H., 1988. The mouse protein synthesis initiation factor 4A gene family includes two related functional genes which are differentially expressed. Eur. Mol. Biol. Org. J. 7, 2097–2105.

Nielsen, P., McMaster, G.K., Trachsel, H., 1985. Cloning of eukaryotic protein synthesis initiation factor genes: isolation and characterization of cDNA clones encoding factor eIF-4A. Nucleic Acids Res. 13, 6867–6880.

op den Camp, R.G.L., Kuhlemeier, C., 1998. Phosphorylation of tobacco eukaryotic translation initiation factor 4A upon pollen tube germination. Nucleic Acids Res. 26, 2058–2062.

Owttrim, G.W., Hoffmann, S., Kuhlemeier, C., 1991. Divergent genes for translation initiation factor eIF-4A are coordinately expressed in tobacco. Nucleic Acids Res. 19, 5491–5496.

Owttrim, G.W., Mandel, T., Trachsel, H., Thomas, A., Kuhlemeier, C., 1994. Characterization of the tobacco eIF-4A gene family. Plant Mol. Biol. 26, 1747–1757.

Pain, V.M., 1996. Initiation of protein synthesis in eukaryotic cells. Eur. J. Biochem. 236, 747–771.

Palacios, I.M., Gatfield, D., Johnston, D.S., Izurralde, E., 2004. An eIF4AIII-containing complex required for mRNA localization and nonsense-mediated mRNA decay. Nature 427, 753–757.

Parisi, M., Nuttall, R., Edwards, P., Minor, J., Naiman, D., Lü, J., et al., 2004. A survey of ovary-, testis-, and soma-biased gene expression in *Drosophila* melanogaster adults. Genome Biol. 5, R40.

Pestova, T.V., Hellen, C.U.T., 2000. The structure and function of initiation factors in eukaryotic protein synthesis. Cell. Mol. Life Sci. 57, 651–674.

Pestova, T.V., Kolupaeva, V.G., 2002. The roles of individual eukayotic translation initiation factors in ribosomal scanning and initiation codon selection. Genes Dev. 16, 2906–2922.

Prevot, D., Darlix, J.L., Ohlmann, T., 2003. Conducting the initiation of protein synthesis: the role of eIF4G. Biol. Cell 9, 141–156.

Ptushkina, M., Fierro-Monti, I., van den Heuvel, J., Vasilescu, S., Birkenhager, R., Mita, K., McCarthy, J.E.G., 1996. *Schizosaccharomyces pombe* has a novel eukaryotic initiation factor 4F complex containing a cap-binding protein with the human eIF4E C-terminal motif KSGST. J. Biol. Chem. 271, 32818–32824.

Ptushkina, M., Berthelot, K., von der Haar, T., Geffers, L., Warwicker, J., McCarthy, J.E., 2001. A second eIF4E protein in *Schizosaccharomyces pombe* has distinct eIF4G-binding properties. Nucleic Acids Res. 29, 4561–4569.

Ptushkina, M., Malys, N., McCarthy, J.E.G., 2004. eIF4E isoform 2 in *Schizosaccharomyces pombe* is a novel stress-response factor. Eur. Mol. Biol. Org. Rep. 5, 311–316.

Raoult, D., Audic, S., Robert, C., Abergel, C., Renesto, P., Ogata, H., et al., 2004. The 1.2 megabase genome sequence of mimivirus. Science 306, 1344–1350.

Richter-Cook, N.J., Dever, T.E., Hensold, J.O., Merrick, W.C., 1998. Purification and characterization of a new eukaryotic protein translation factor. Eukaryotic initiation factor 4H. J. Biol. Chem. 273, 7579–7587.

Robalino, J., Joshi, B., Fahrenkrug, S.C., Jagus, R., 2004. Two zebrafish eIF4E family members are differentially expressed and functionally divergent. J. Biol. Chem. 279, 10532–10541.

Rodriguez, C.M., Freire, M.A., Camilleri, C., Robaglia, C., 1998. The *Arabidosis thaliana* cDNAs encoding for eIF4E and eIF(iso)4E are not functionally equivalent for yeast complementation and are differentially expressed during plant development. Plant J. 13, 465–473.

Rogers, G.W., Komar, A.A., Merrick, W.C., 2002. eIF4A: the goodfather of the DEAD box helicases. Prog. Nucleic Acid Res. Mol. Biol. 72, 307–331.

Rom, E., Kim, H.C., Gingras, A.C., Marcotrigiano, J., Favre, D., Olsen, H., et al., 1998. Cloning and characterization of 4EHP, a novel mammalian eIF4E-related cap-binding protein. J. Biol. Chem. 273, 13104–13109.

Ruud, K.A., Kuhlow, C., Goss, D.J., Browning, K.S., 1998. Identification and characterization of a novel cap-binding protein from *Arabidopsis thaliana*. J. Biol. Chem. 273, 10325–10330.

Rychlik, W., Domier, L.L., Gardner, P.R., Hellmann, G.M., Rhoads, R.E., 1987. Amino acid sequence of the mRNA cap-binding protein from human tissues. Proc. Natl Acad. Sci. USA 84, 945–949.

Schmidt, S., Hofmann, K., Simanis, V., 1997. *Sce3*, a suppressor of the *Schizosaccharomyces pombe* septation mutant *cdc11*, encodes a putative RNA-binding protein. Nucleic Acids Res. 25, 3433–3439.

Shibuya, T., Tange, T., Sonenberg, N., Moore, M.J., 2004. eIF4AIII binds spliced mRNA in the exon junction complex and is essential for nonsense-mediated dacay. Nat. Struct. Mol. Biol. 11, 346–351.

Sonenberg, N., Rupprecht, K.M., Hecht, S.M., Shatkin, A.J., 1979. Eukaryotic mRNA cap binding protein: purification by affinity chromatography on sepharose-coupled m7GDP. Proc. Natl Acad. Sci. USA 76, 4345–4349.

Strudwick, S., Borden, K.L., 2002. The emerging roles of translation factor eIF4E in the nucleus. Differentiation 70, 10–22.

Sudo, K., Takahashi, E., Nakamura, Y., 1995. Isolation and mapping of the human eIF4A2 gene homologous to the murine protein synthesis initiation factor 4A-II gene *Eif4a2*. Cytogenet. Cell Genet. 71, 385–388.

Tahara, S.M., Morgan, M.A., Shatkin, A.J., 1981. Two forms of purified m7G-cap binding protein with different effects on capped mRNA translation extracts of uninfected and polio-virus-infected HeLa cells. J. Biol. Chem. 256, 7681–7694.

Thompson, J.D., Higgins, D.G., Gibson, T.J., 1994. CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choise. Nucleic Acids Res. 22, 4673–4680.

Topisorivic, I., Kuljkovic, B., Cohen, N., Perez, J.M., Skrabanek, L., Borden, K.L.B., 2003. The proline-rich homeodomain protein, PRH, is a tissue-specific inhibitor of eIF4E-dependent cyclin D1 mRNA transport and growth. Eur. Mol. Biol. Org. J. 22, 689–703.

von der Haar, T., Gross, J.D., Wagner, G., McCarthy, J.E.G., 2004. The mRNA cap-binding protein eIF4E in post-transcriptional gene expression. Nat. Struct. Mol. Biol. 11, 503–511.

Wakiyama, M., Saigoh, M., Shiokawa, K., Miura, K., 1995. mRNA encoding the translation initiation factor eIF-4E is expressed early in *Xenopus* embryogenesis. Fed. Eur. Biochem. Soc. Lett. 360, 191–193.

Wakiyama, M., Suzuki, A., Saigoh, M., Sakai, N., Miyoshi, H., Kojima, S., Miura, K., 2001. Analysis of the isoform of *Xenopus* eukaryotic translation initiation factor 4E. Biosci. Biotechnol. Biochem. 65, 232–235.

Warnakulasuriyarachchi, D., Cerquozzi, S., Cheung, H.H., Holcick, M., 2004. Translational induction of HIAP2 during endoplasmic reticulum stress attenuates cell death and is mediated via an inducible IRES. J. Biol. Chem. 279, 17148–17157.

Weinstein, D.C., Honoré, E., Hemmati-Brivanlou, A., 1997. Epidermal induction and inhibition of neural fate by translation initiation factor 4AIII. Development 124, 4235–4242.

Wells, D.R., Tanguay, R.L., Le, H., Gallie, D.R., 2004. HSP101 functions as a specific translational regulatory protein whose activity is regulated by nutrient status. Genes Dev. 12, 3236–3251.

Wilhelm, J.E., Hilton, M., Amos, Q., Henzel, W., 2003. Cup is an eIF4E binding protein required for both the translational repression of *oskar* and the recruitment of Barentsz. J. Cell Biol. 163, 1197–1204.

Williams-Hill, D.M., Duncan, R.F., Nielsen, P.J., Tahara, S.M., 1997. Differenial expression of the murine eukaryotic translation initiation factor isogenes eIF4AI and eIF4AII is dependent upon cellular growth status. Arch. Biochem. Biophys. 338, 111–120.

Woese, C.R., 2000. Interpreting the universal phylogenetic tree. Proc. Natl Acad. Sci. USA 97, 8392–8396.

Yamanaka, S., Poksay, K.S., Arnold, K.S., Innerarity, T.L., 1997. A novel translation repressor mRNA is edited extensively in livers containing tumors caused by the transgene expression of the apoB mRNA-editing enzyme. Genes Dev., 321–333.

Yan, R., Rychlik, W., Etchison, D., Rhoads, R.E., 1992. Amino acid sequence of the human protein synthesis initiation factor eIF4gamma. J. Biol. Chem. 267, 23226–23231.

Yoffe, Y., Zuberek, J., Lewdorowicz, M., Zeira, Z., Keasar, C., Orr-Dahan, I., et al., 2004. Cap-binding activity of an eIF4E homolog from *Leishmania*. RNA 10, 1764–1775.

Zapata, J.M., Martínez, M.A., Sierra, J.M., 1994. Purification and characterization of eukaryotic polypeptide chain initiation factor 4F from *Drosophila* embryos. J. Biol. Chem. 269, 18047–18052.

Zappavigna, V., Piccioni, F., Villaescusa, C., Verrotti, A.C., 2004. Cup is a nucleocytoplasmic shutting protein that interacts with the eukaryotic translation initiation factor 4E to modulate *Drosophila* ovary development. Proc. Natl Acad. Sci. USA 101, 14800–14805.