Szabályozó *fehérjék* szerepe az öregedési folyamatban

Dr. Vellai Tibor
ELTE Genetikai Tanszék

Terület:
-öregedés szabályozása
-autofágia (sejtes önemésztés)
-sejtes stresszválasz

1, az öregedést okozó faktorok meghatározása

2, a hősokk-választ, endoplazmatikus stressz-választ és az autofágiát összekapcsoló szabályozó fehérjék feltárása

3, az öregedési folyamat szabályozási hálózatának megértése

Terület:
-sejtes stresszválasz
-öregedés szabályozása
-hősokk fehérjék

Dr. Sőti Csaba
SE Orvosi Vegytani Intézet

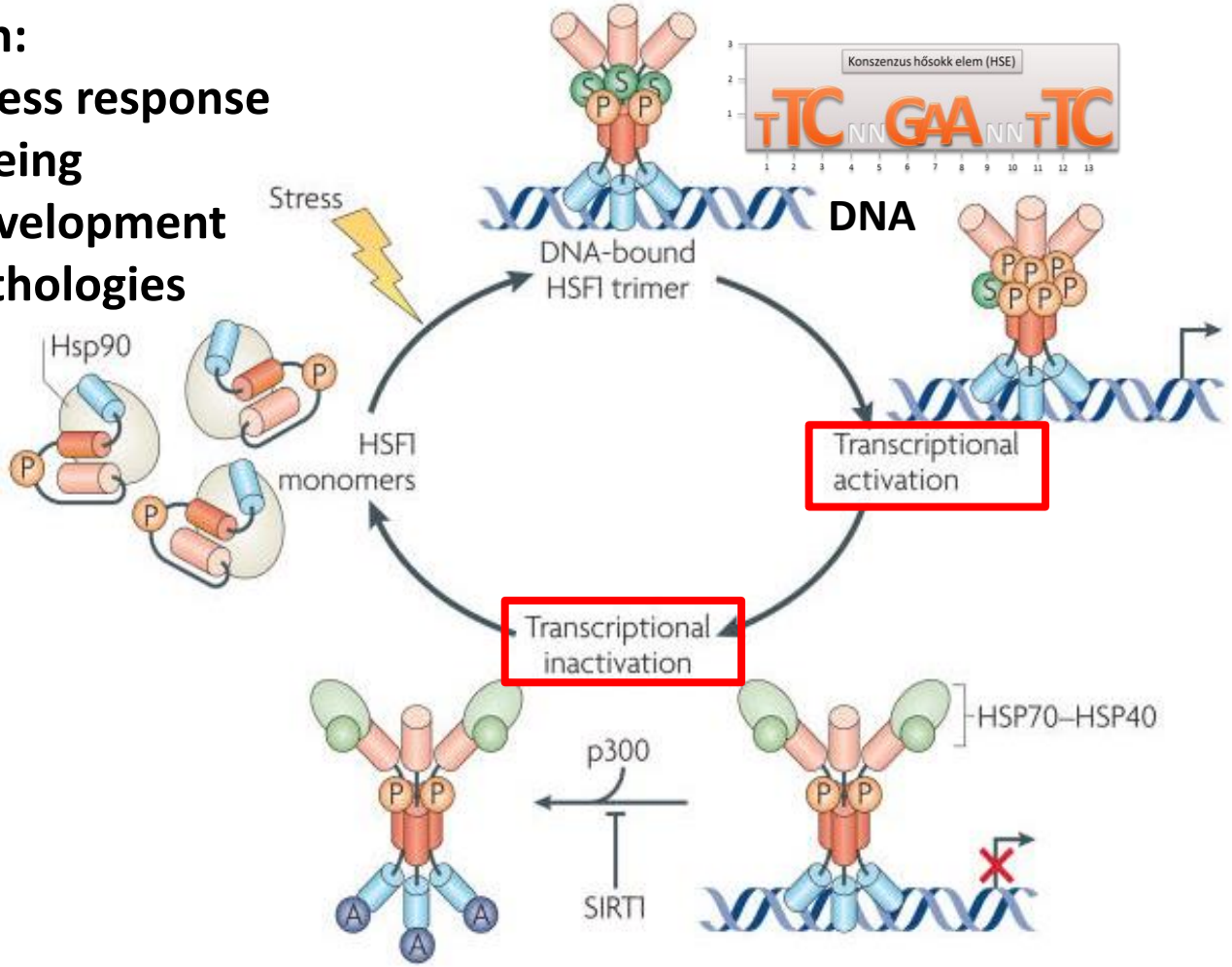
Terület:
-endoplazmatikus stressz
-degeneratív betegségek
-redox homeosztázis

Dr. Bánhegyi Gábor
SE Orvosi Vegytani Intézet

The heat-shock transcription factor HSF-1 directly regulates ER-stress response (*C. elegans* vs. HEK293 cells)

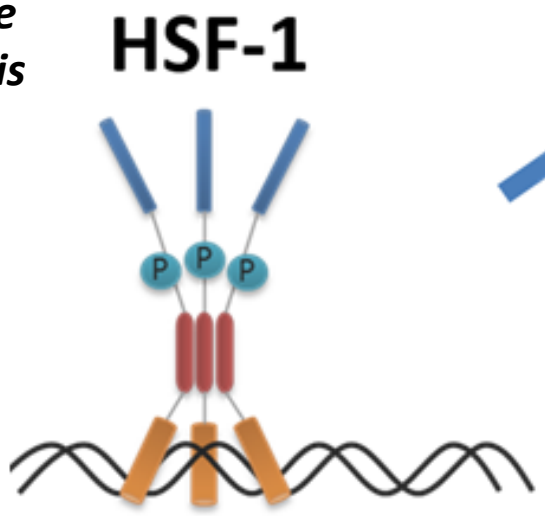
Role in:

- **Stress response**
- **Ageing**
- **Development**
- **Pathologies**



Unfolded Protein Response (UPR) proteins: HSF-1 target genes in *C. elegans*?

The Nematode Caenorhabditis elegans



Consensus sequence:
...**TTC** NN **GAA** NN **TTC**...

?

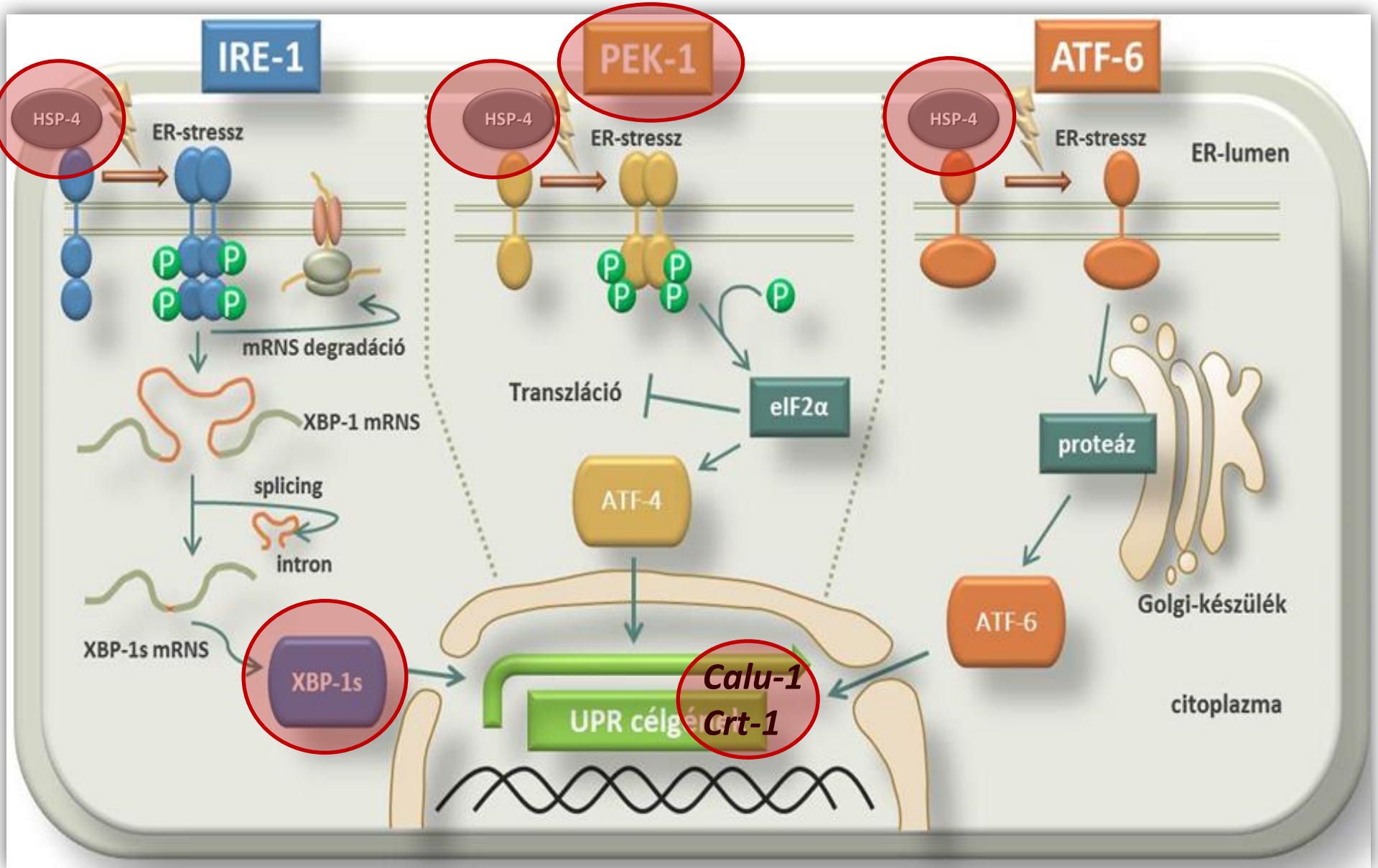
UNFOLDED PROTEIN RESPONSE

Potential HSF-1 target genes

- *hsp-4/BiP*
- *xbp-1/XBP1*
- *pek-1/PERK*
- *calu-1/Calumenin*
- *crt-1/CALR*

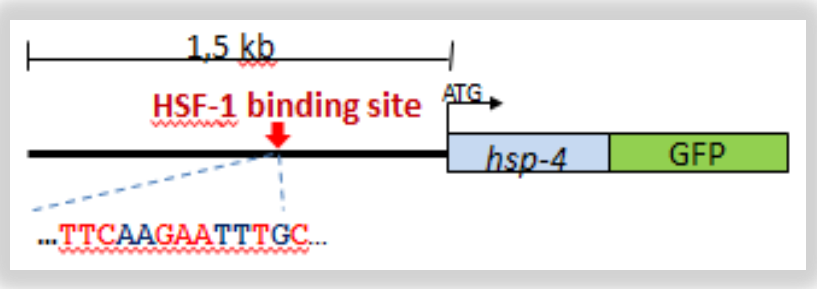
The UPR pathways

HSF-1 targets:



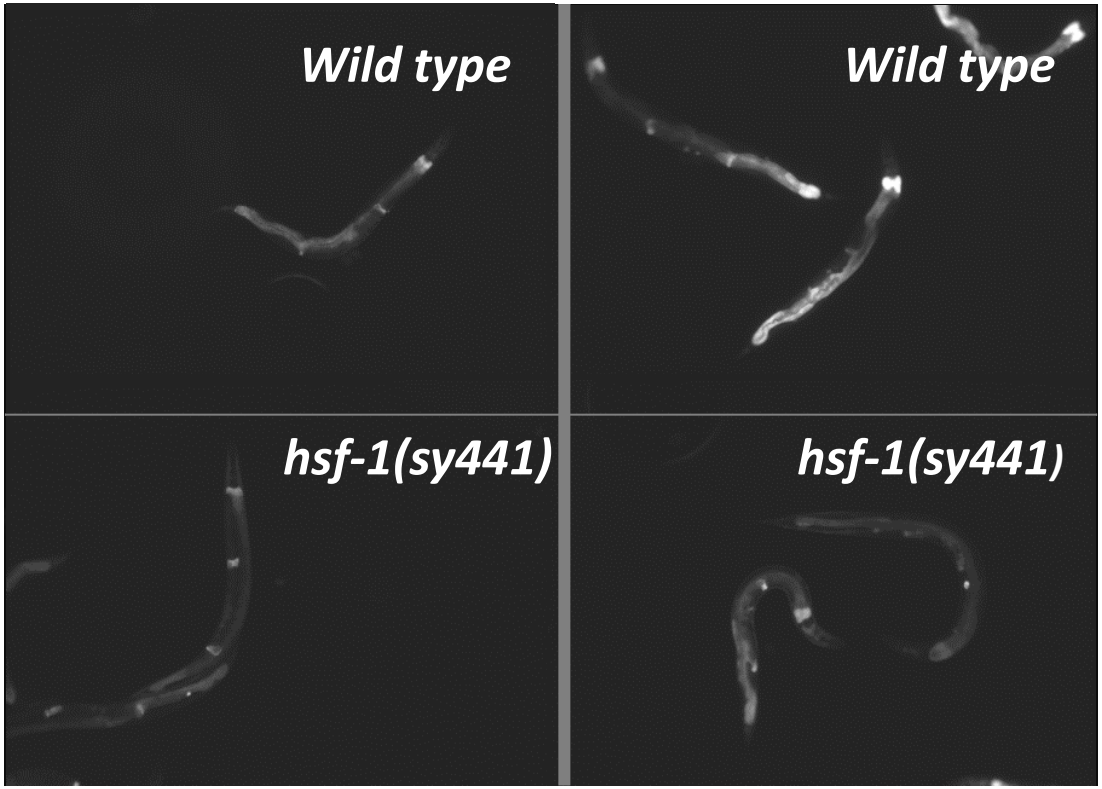
HSF-1 activates components of the UPR

I. HSF-1 upregulates *hsp-4*



hsp-4::gfp

Control (25°C) **Heat shock (35°C)**



HEAT SHOCK

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HSF-1

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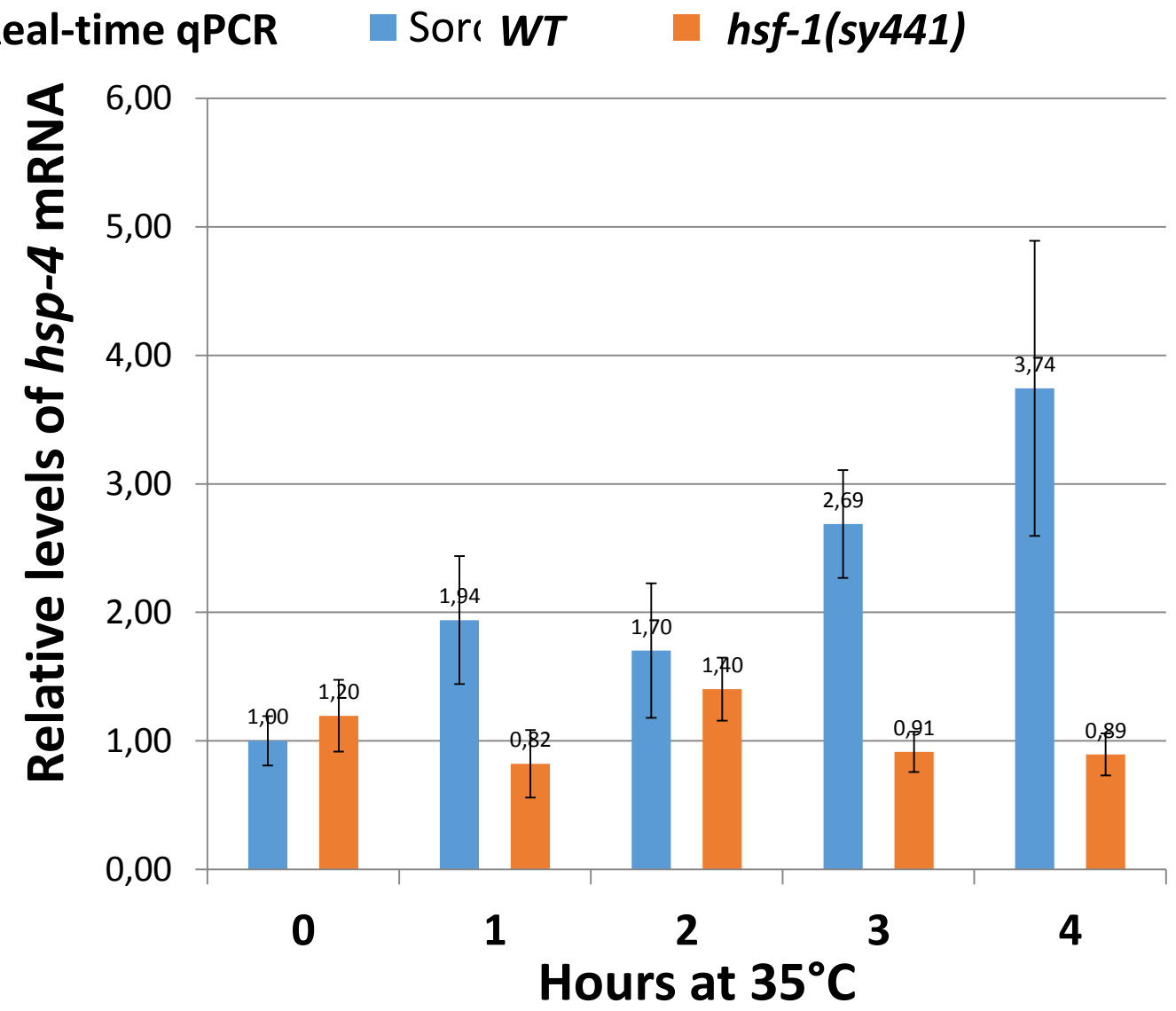
hsp-4/BiP

hsp-4/BIP mRNA levels are increased upon heat stress in a *hfs-1* dependent manner

Heat shock

HSF-1

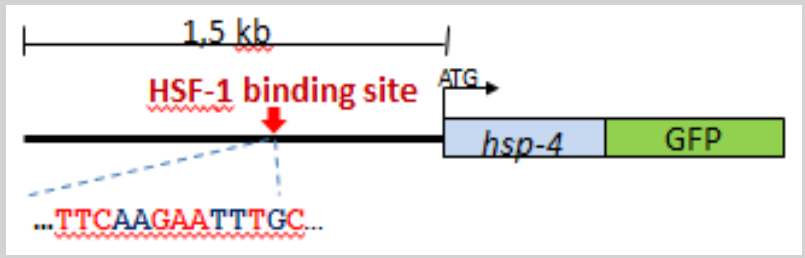
hsp-4



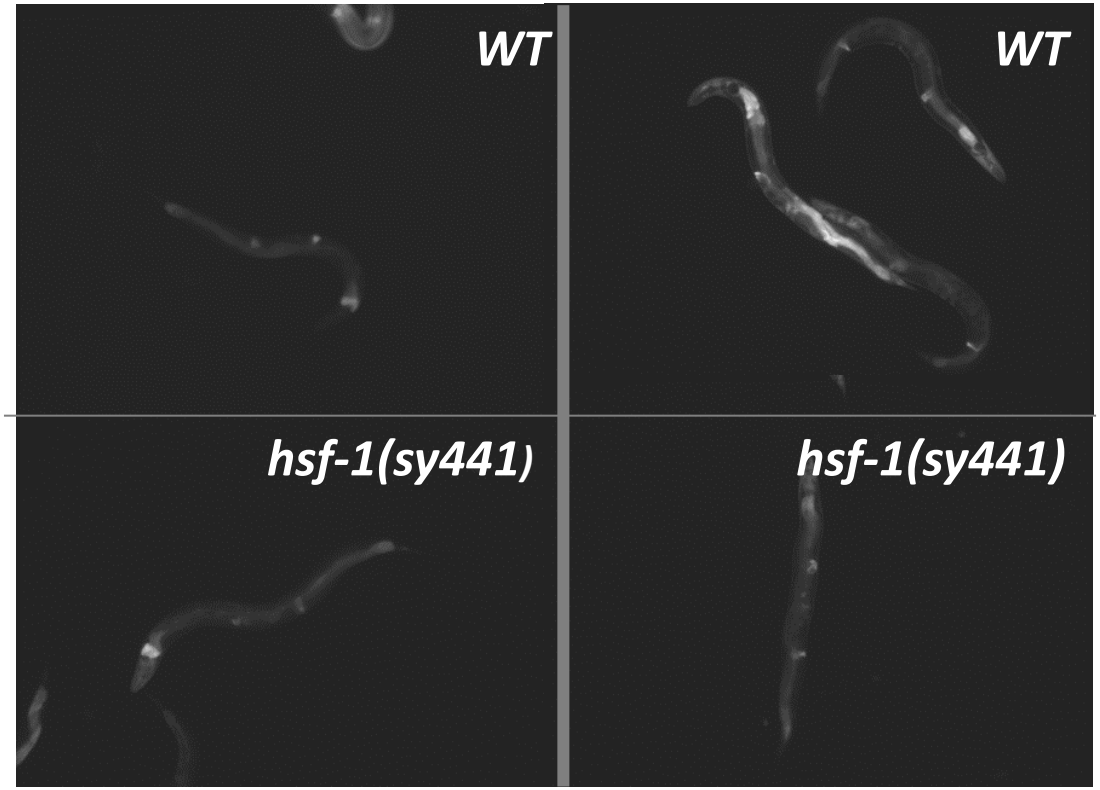
ER stress triggered by DTT induces *hsp-4* expression via HSF-1

Dithiothreitol (DTT)

hsp-4::gfp



Control **8 mM DTT**



ER STRESS

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HSF-1

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hsp-4/BiP

ER stress triggered by DTT induces *hsp-4* expression via HSF-1

I. HSF-1 upregulates *hsp-4*

ER stress

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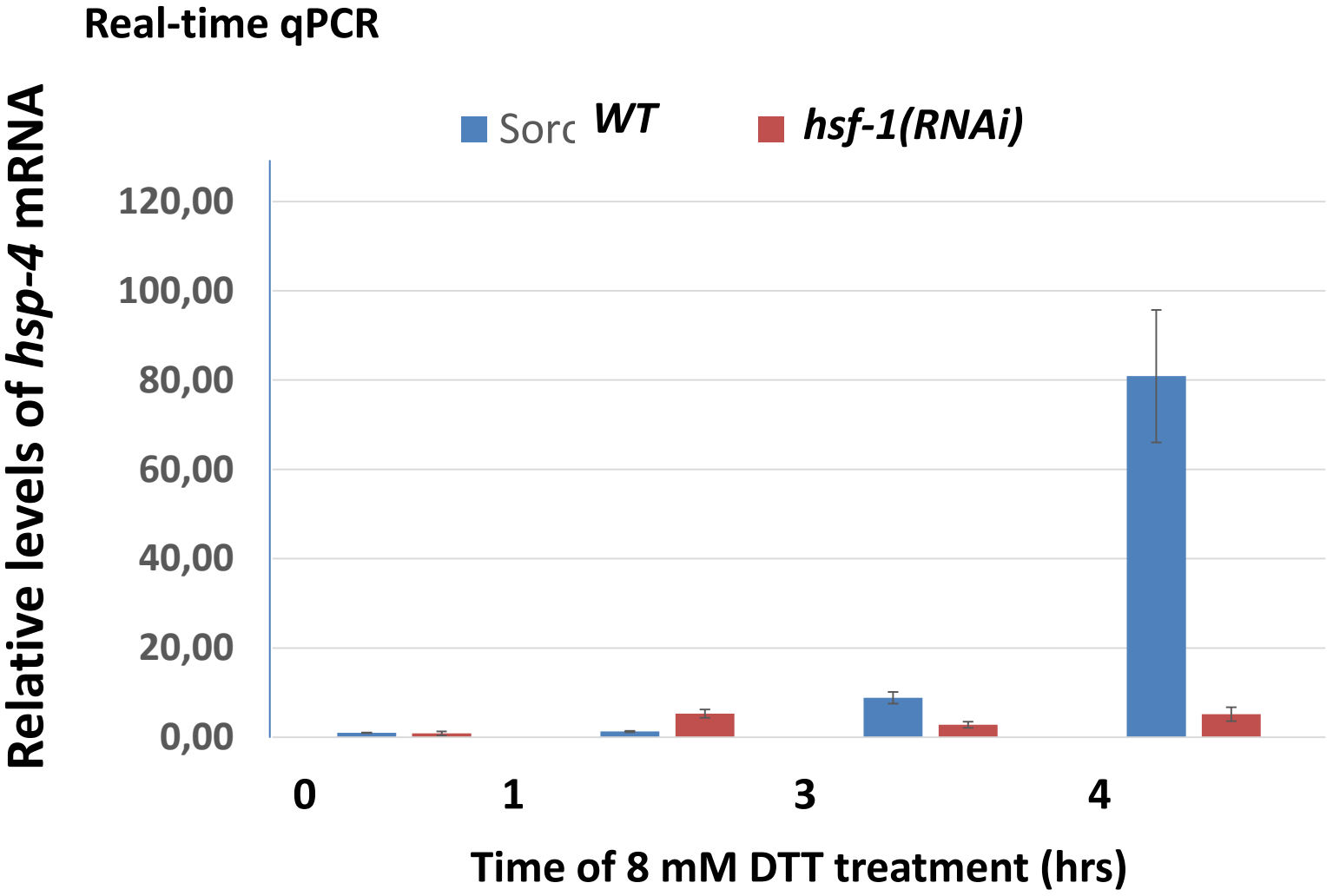
HSF-1

↓

hsp-4



HSF-1 activates components of the UPR

IV. HSF-1 upregulates *xbp-1*

Heat shock

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HSF-1

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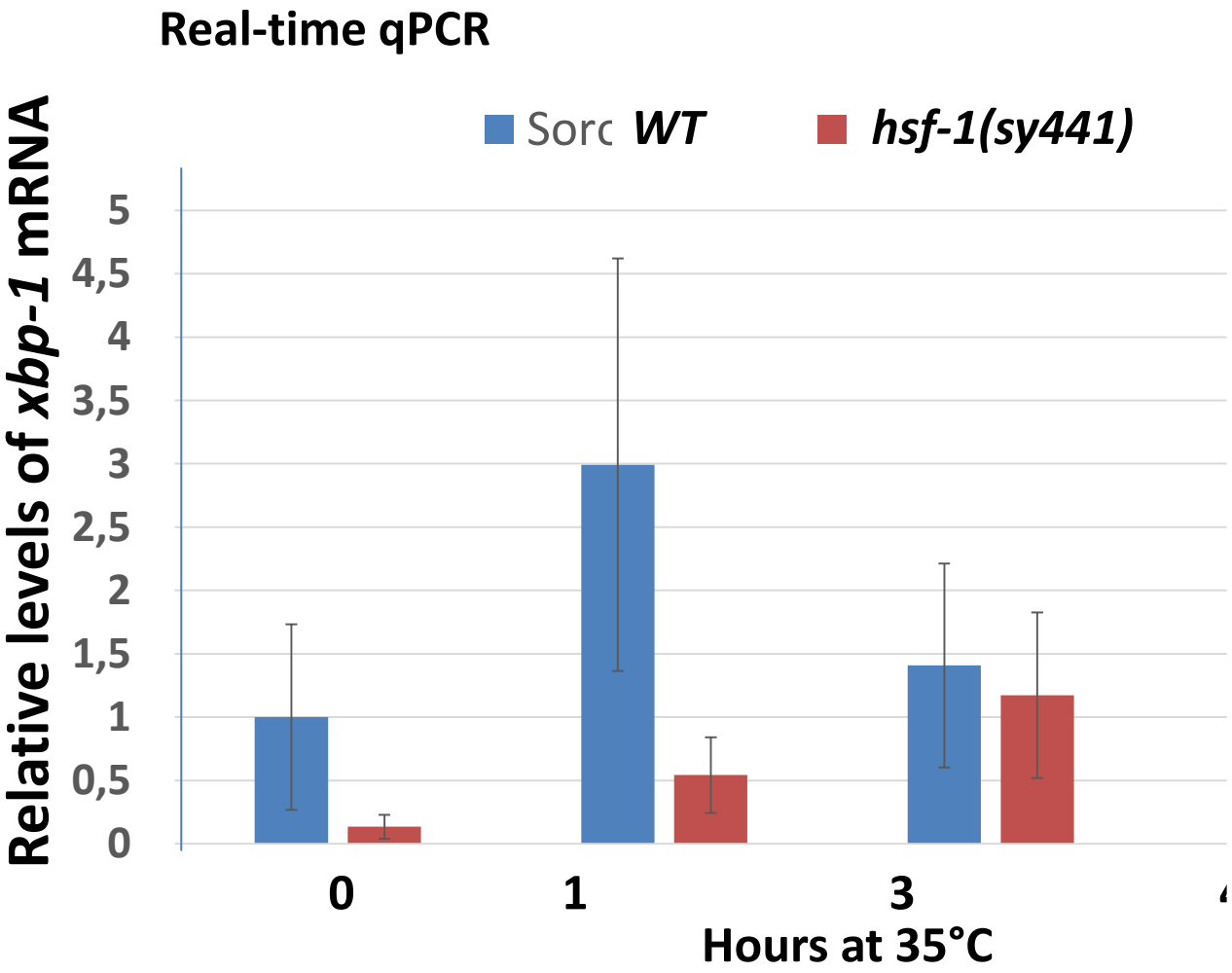
xbp-1

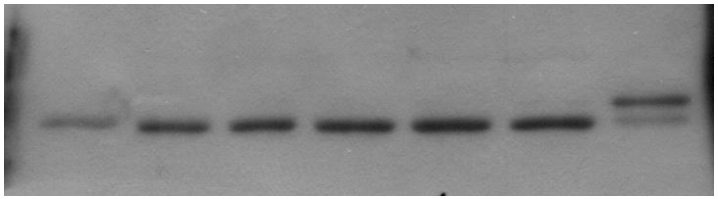


Emlős sejtek (HEK293)

42°C

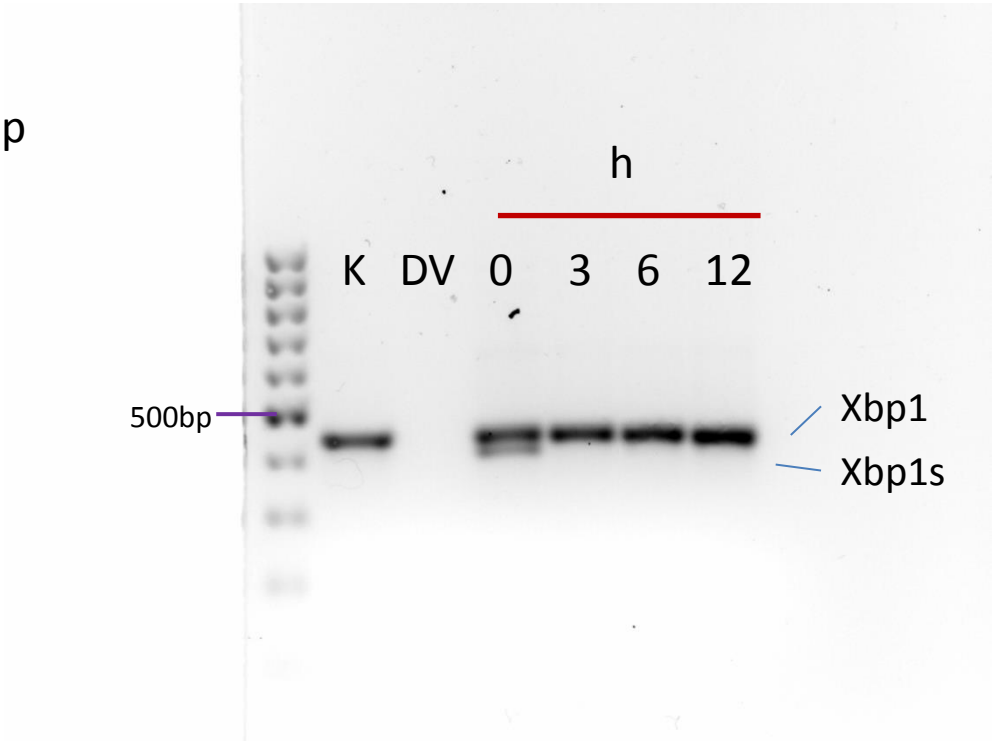
regeneráció 37°C-on

K 0 3 6 12 24 +K Hepg2



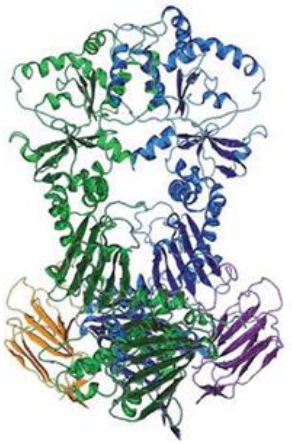
Bip

Xbp1 aktiváció 42°C hősokk hatására



Hsf1 csendesítés: hősokk hatás fennmarad!

A Hsp90 és a HSF1

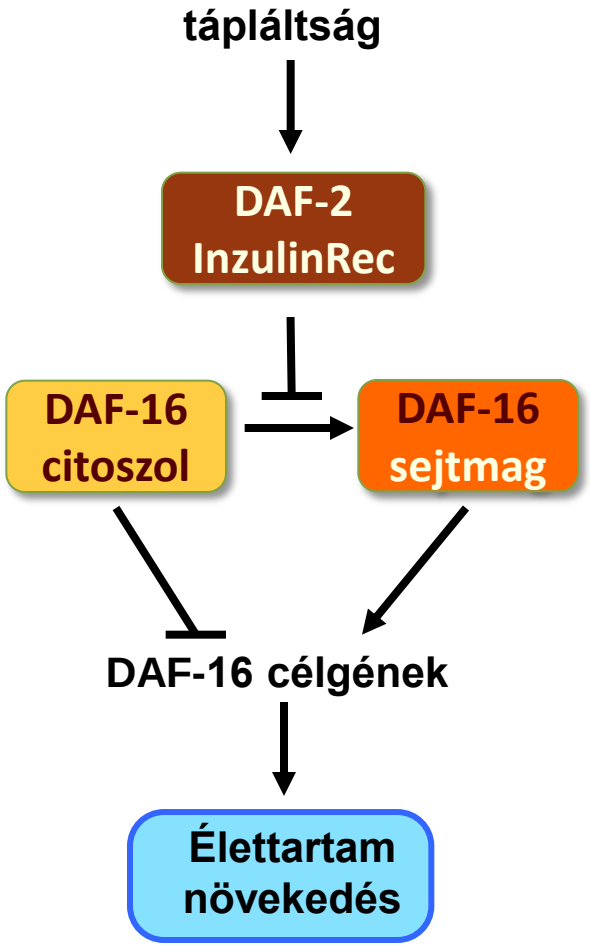


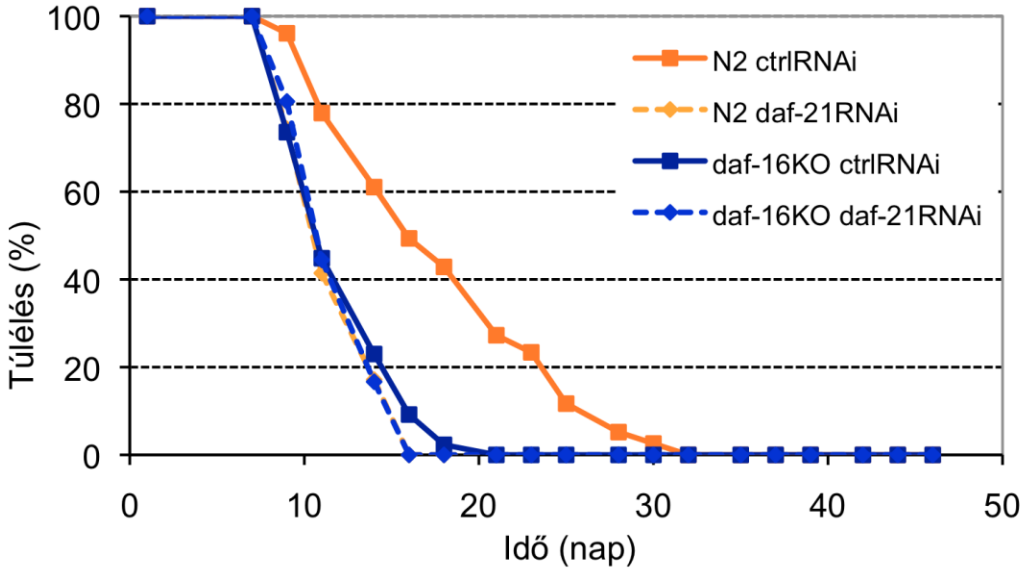
HSF1: hősokk transzkripciós faktor

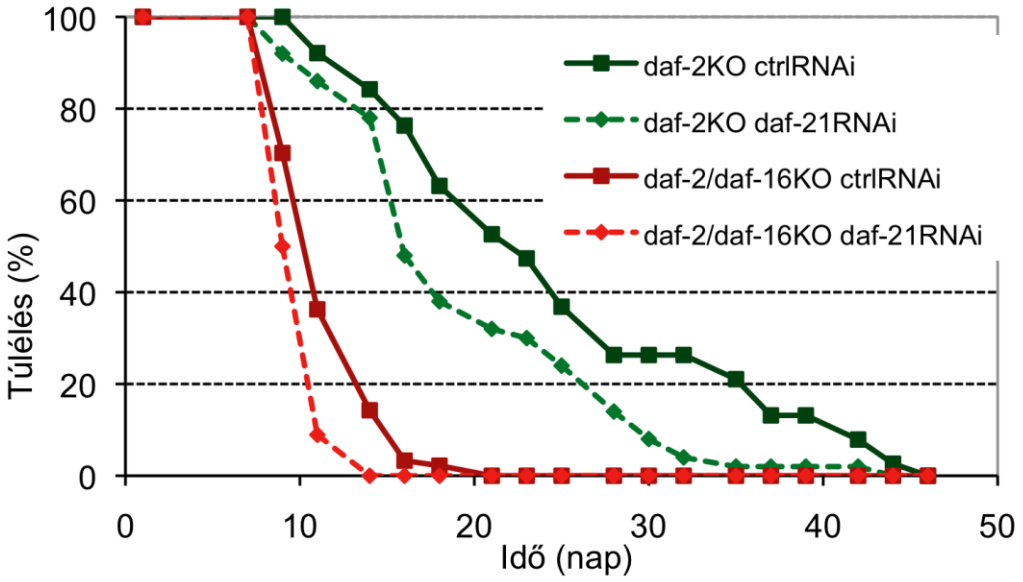
Hsp90: 90 kDa molekulatömegű hősokkfehérje (*C. elegans* DAF-21)

Kliens: többszáz, instabil szerkezetű jelátviteli fehérje, melyeket a Hsp90 stabilizál

Eredmény 1:
a Hsp90/DAF-21 szükséges a *C. elegans* élettartamához

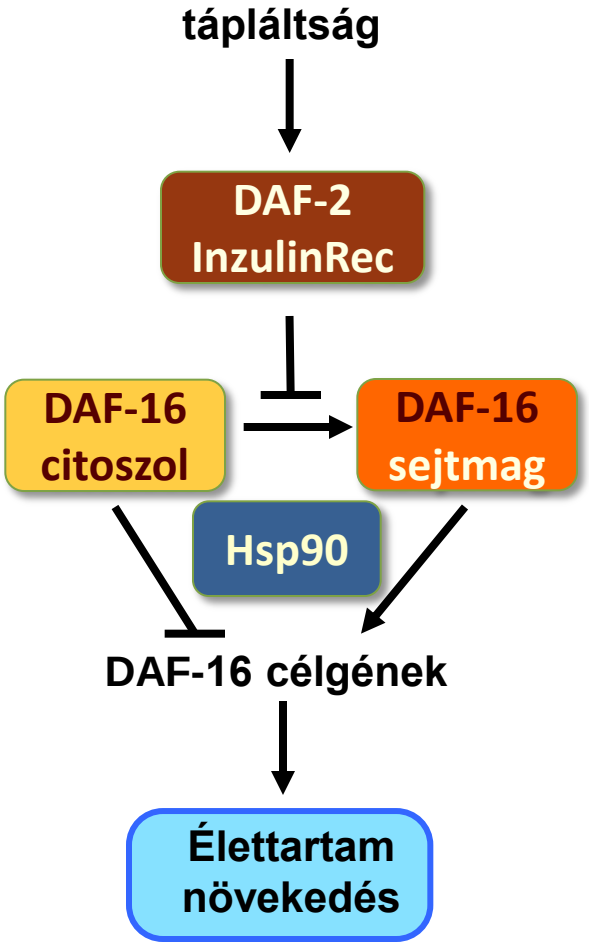






DAF-16: FOXO ortológ

A Hsp90/DAF-21 hozzájárul a DAF-16 sejtmagi transzlokációjához és a DAF-16 függő génexpresszióhoz



Az öregedés hatása a hsp génexpresszióra

Proteotoxikus stressz

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HSF1

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Hsp transzkripció hősokkválasz

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Termo/stressz-tolerancia

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Élettartam növekedés

ÖREGEDÉS

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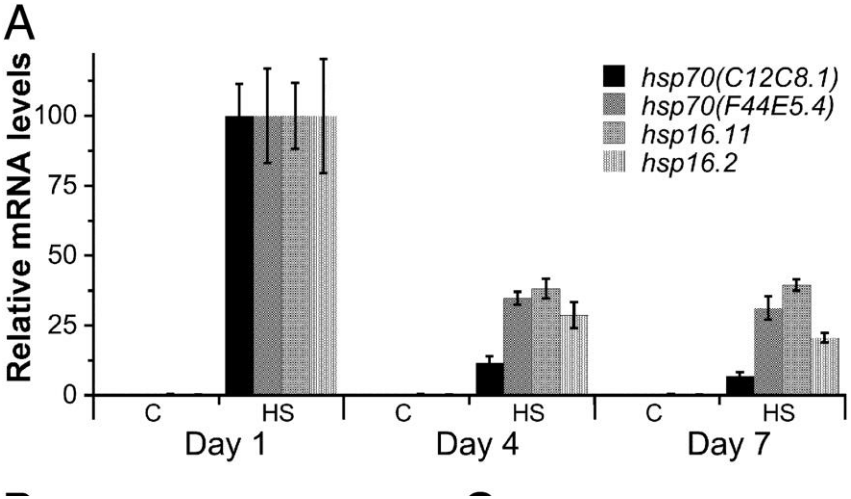
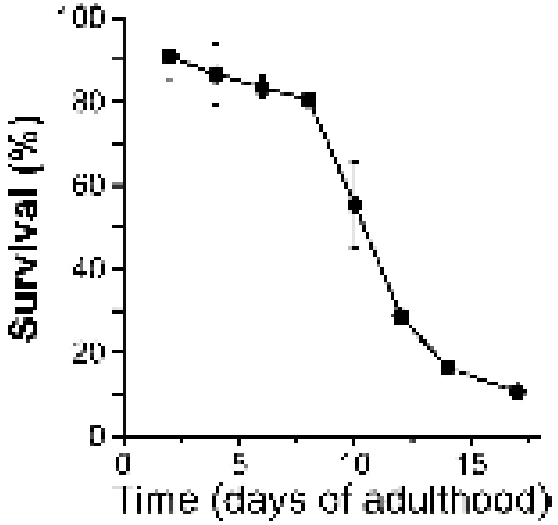
Ben-Zvi és mtsai, PNAS 2009



hsp génexpresszió már 4 napnál hanyatlik
Termotolerancia 8 napig nem változik



3: Az öregedés nem csökkenti, hanem serkenti a hsp génexpressziót?

Somogyvári Milán

Proteotoxikus stressz

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HSF1

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Hsp transzkripció hősokkválasz

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Termo/stressz-tolerancia

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Élettartam növekedés

ÖREGEDÉS

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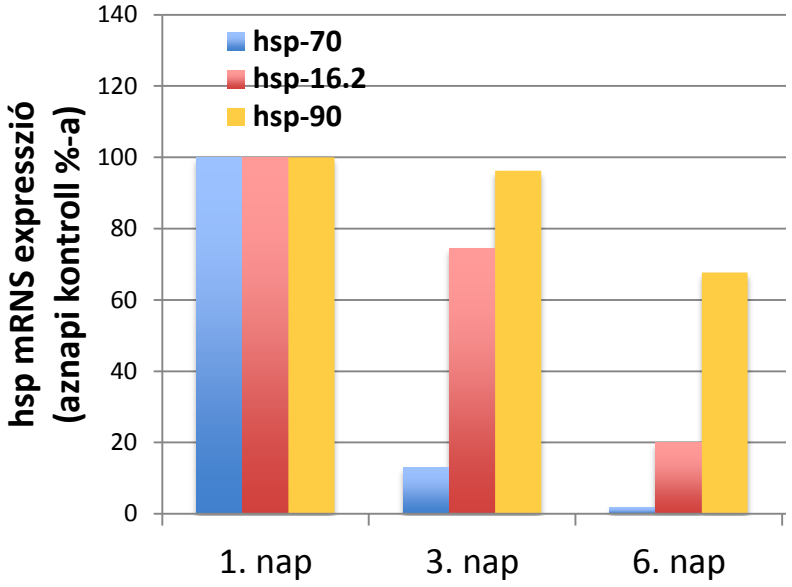
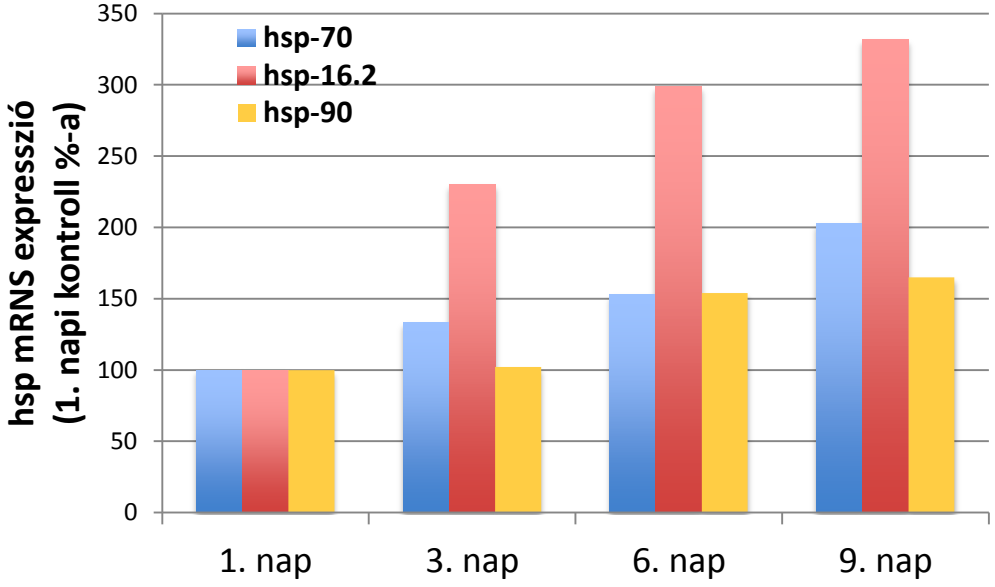
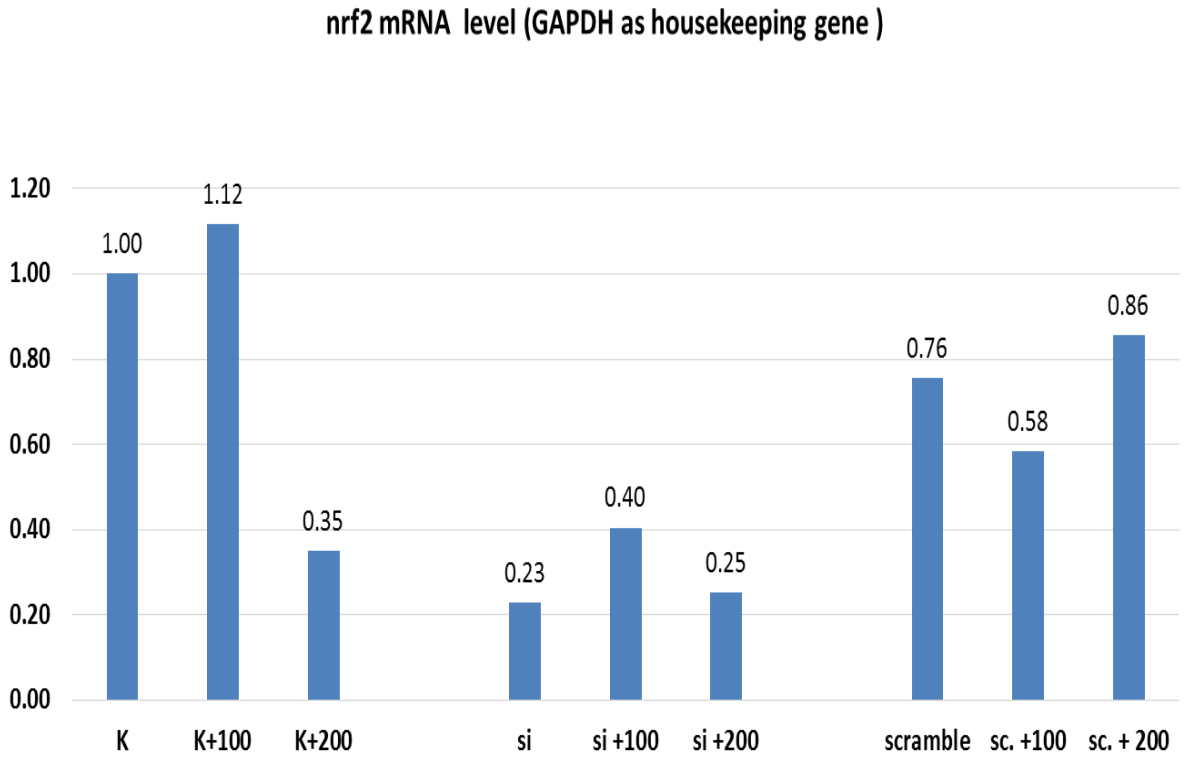




Effect of Nrf2 on AMPK-dependent autophagy

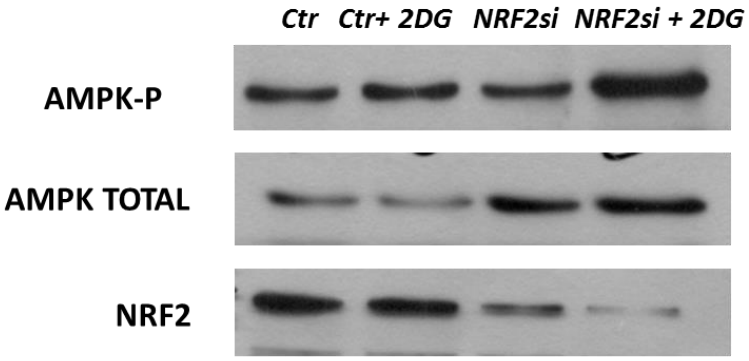
-A negative regulatory loop was observed between skn-1 and aak-2, the orthologs of NRF2 and AMPK *in Caenorhabditis elegans*

-Nrf2 was silenced in HEK293 cells. Oxidative stress was induced by TBHP (*tert*-Butyl hydroperoxide) or starvation was mimicked with 2DG (2-Deoxy-D-glucose)

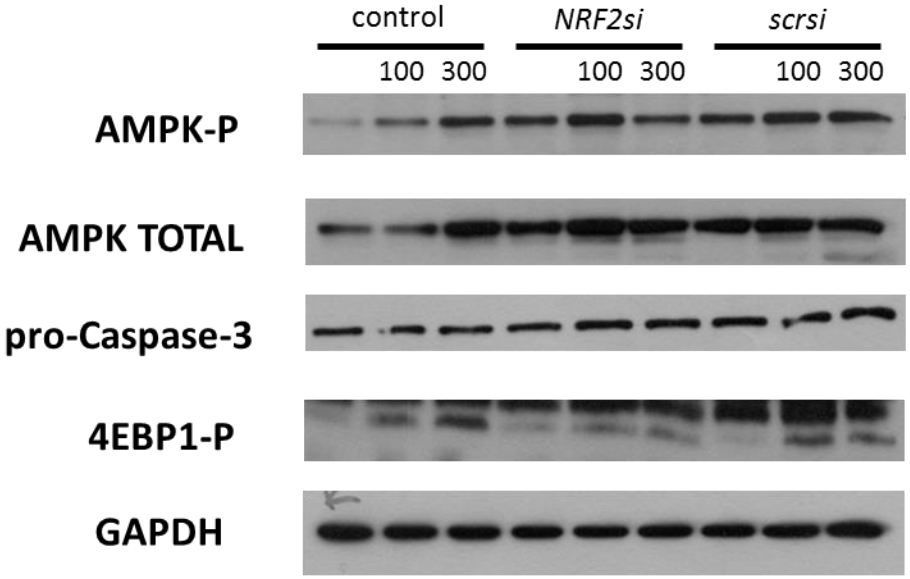


Nrf2 silencing results in increased AMPK activation

2DG treatment (10 mM, 24 h)



TBHP treatment (100 or 300 mM, 1.5 h)



NRF2 negatively regulates autophagy through AMPK inhibition with respect to either oxidative stress or starvation in human cell line as well as their orthologs in *C. elegans*.

Publikációk MEDinPROT támogatással

Csala M, Kardon T, Legeza B, Lizák B, Mandl J, Margittai É, Puskás F, Száraz P, Szelényi P, Bánhegyi G. On the role of 4-hydroxynonenal in health and disease. *Biochim Biophys Acta* 2015 Jan 30;1852(5):826-838

Margittai É, Enyedi B, Csala M, Geiszt M, Bánhegyi G. Composition of the redox environment of the endoplasmic reticulum and sources of hydrogen peroxide. *Free Radic Biol Med.* 2015 Feb 9. pii: S0891-5849(15)00039-8. doi:10.1016/j.freeradbiomed.2015.01.032.

Sturm Á, Ivics Z, Vellai T. The mechanism of ageing: primary role of transposable elements in genome disintegration. *Cell Mol Life Sci*, (revision is submitted)

Holczer M, Márton M, Kurucz A, Bánhegyi G, Kapuy O. A comprehensive systems biological study of autophagy - apoptosis crosstalk during endoplasmic reticulum stress. *BioMed Res Int* (revised version)

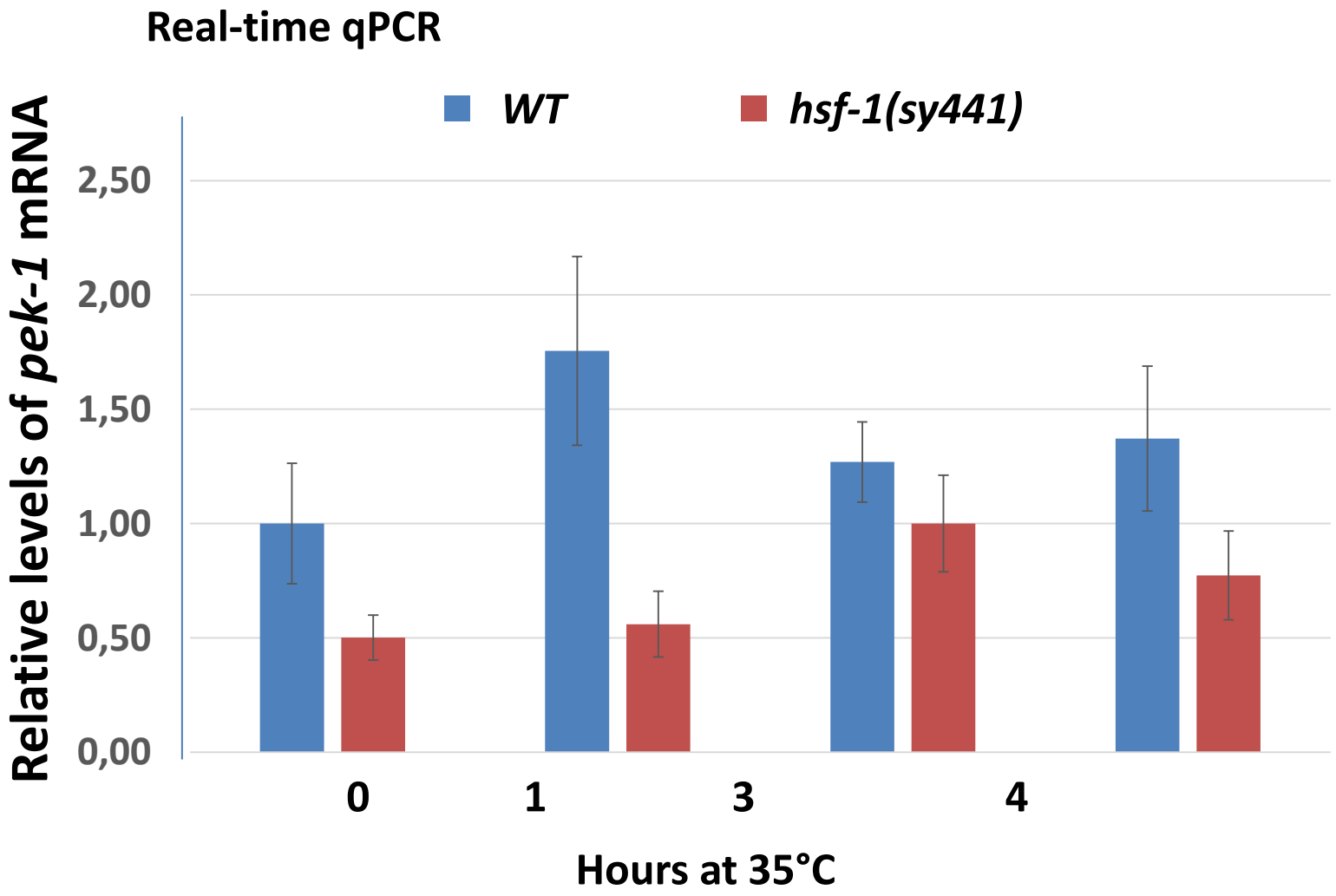
HSF-1 activates components of the UPR

II. HSF-1 upregulates *pek-1*

Heat shock

HSF-1

pek-1



ER stress triggered by DTT induces *pek-1* expression via HSF-1

II. HSF-1 upregulates *pek-1*

ER stress

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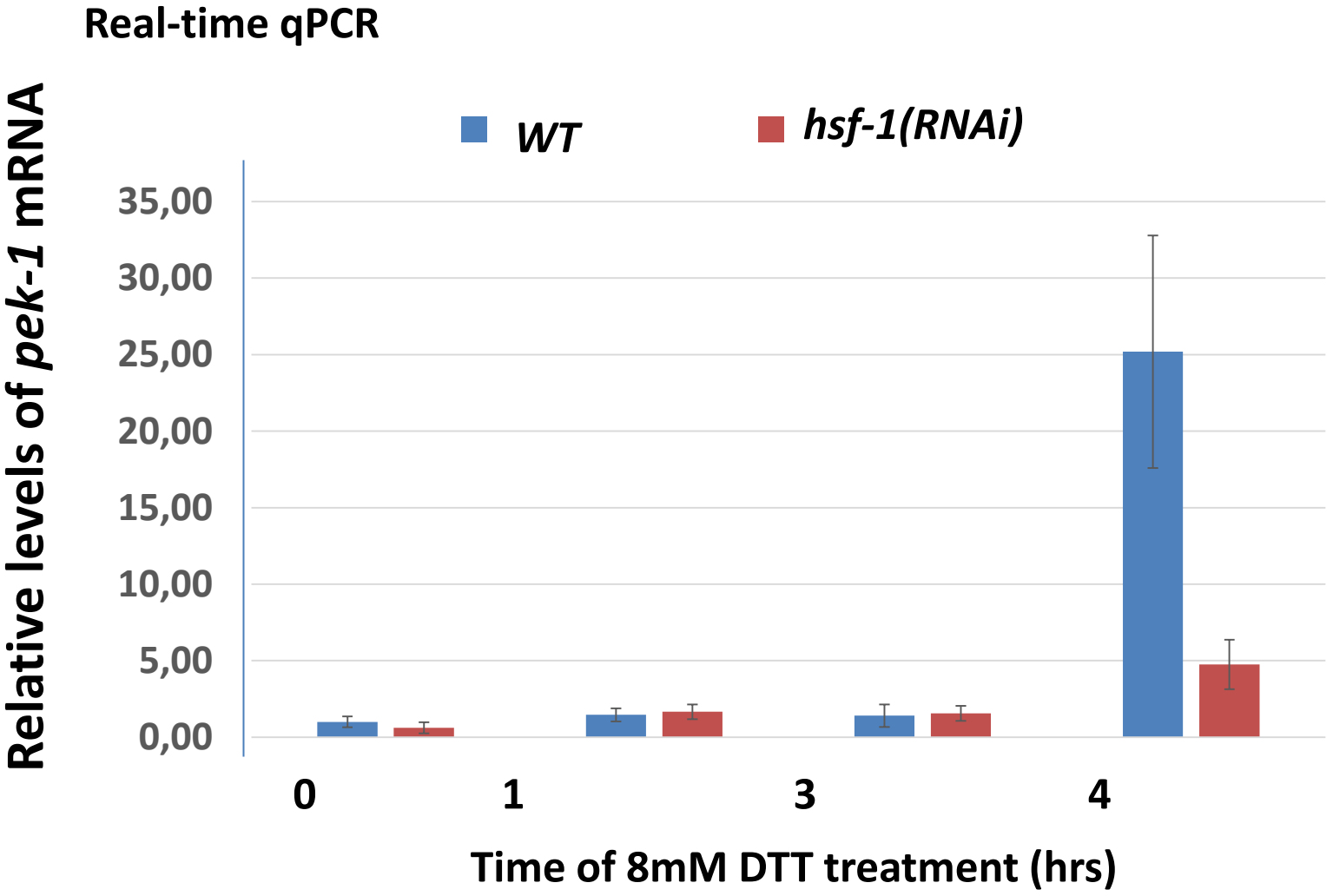
HSF-1

↓

pek-1



HSF-1 activates components of the UPR

V. HSF-1 upregulates *crt-1*

Heat shock

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HSF-1

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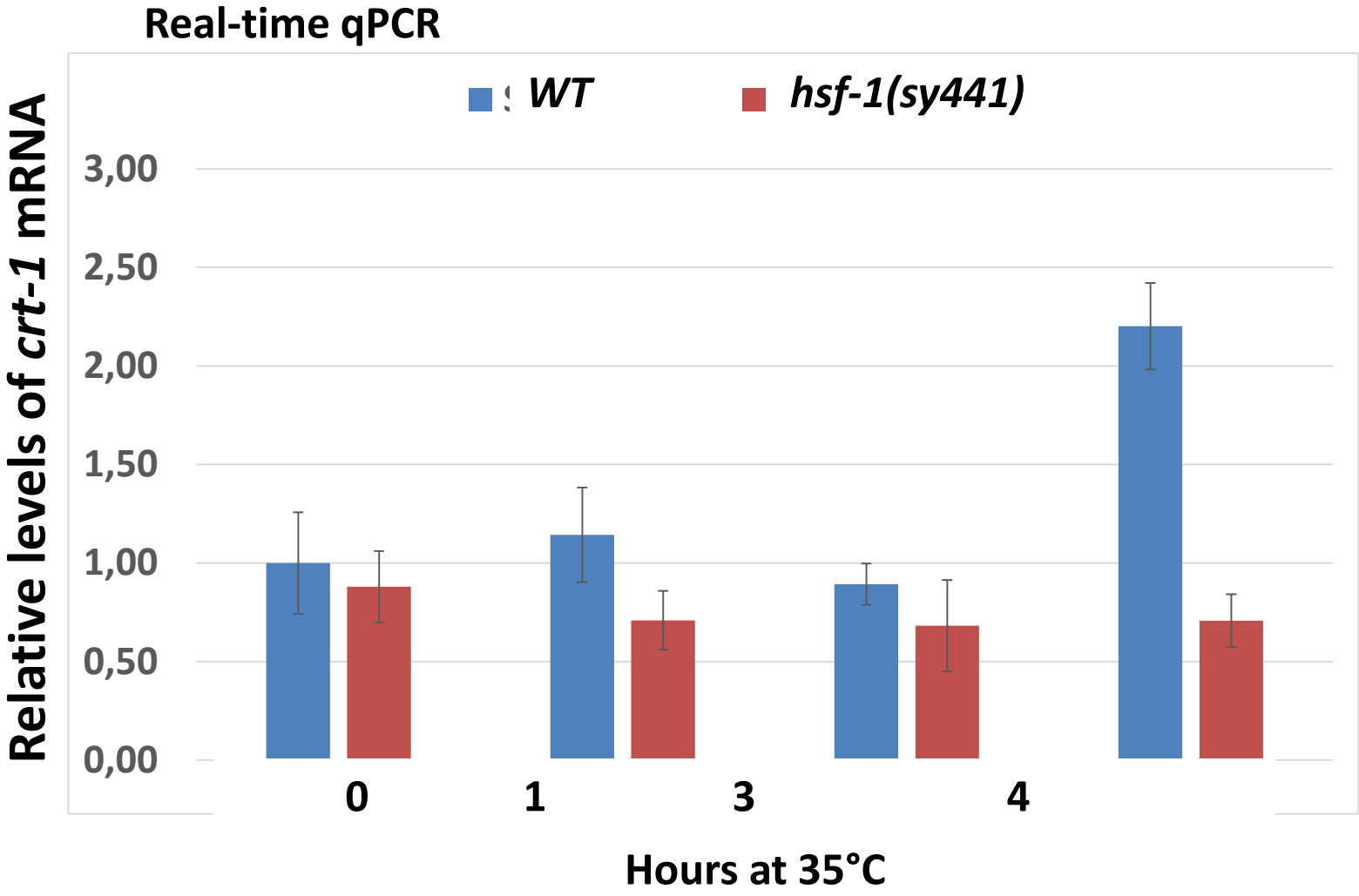
↓

crt-1



HSF-1 activates components of the UPR

III. HSF-1 upregulates *calu-1*

Heat shock

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HSF-1

↓

calu-1