



N-vironment

**The role of the N-end rule pathway in controlling
plant response to the environment**

Francesco Licausi

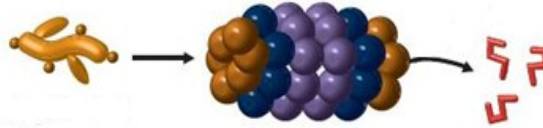
Members of the consortium



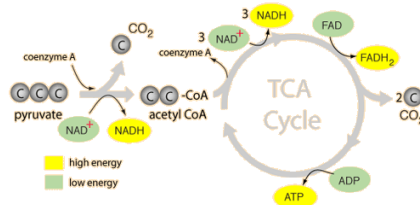
Michael Holdsworth – University of Nottingham



Andreas Bachmair – University of Vienna



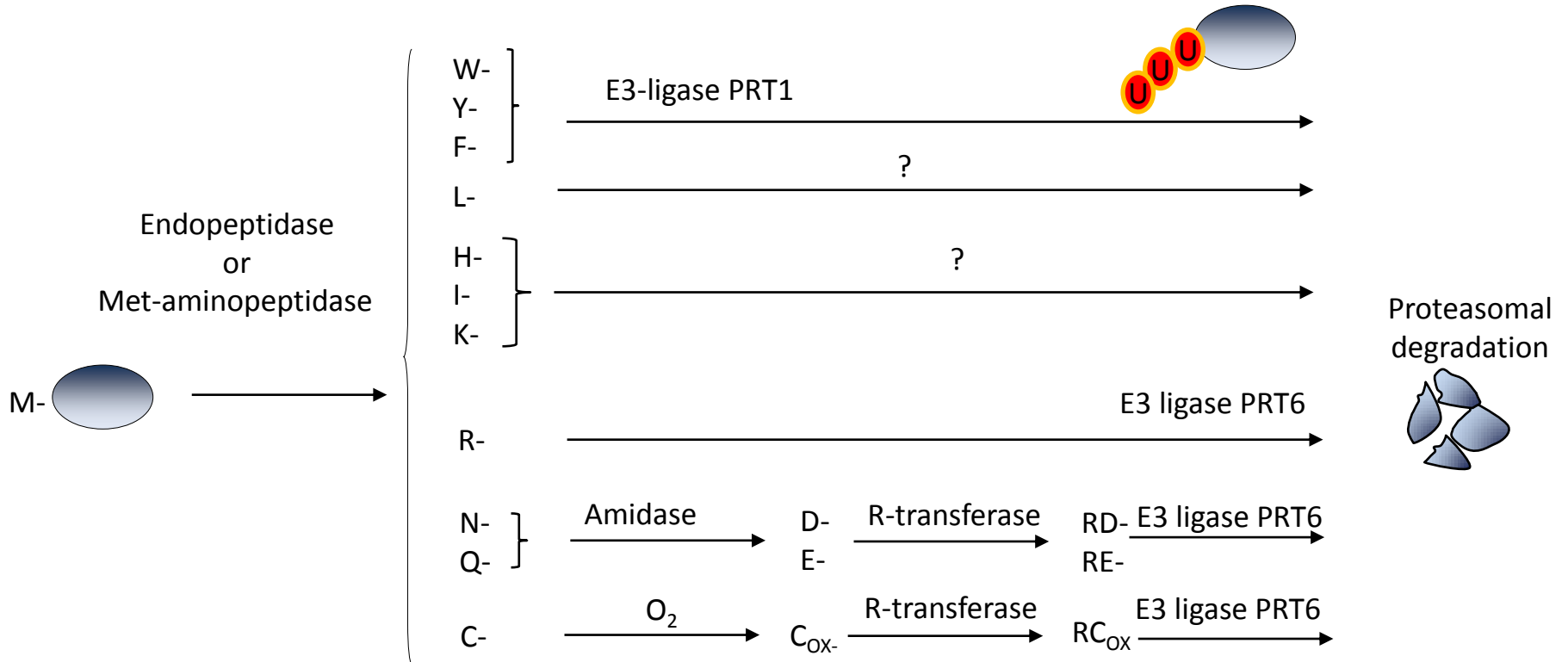
Joost van Dongen – RWTH University Aachen



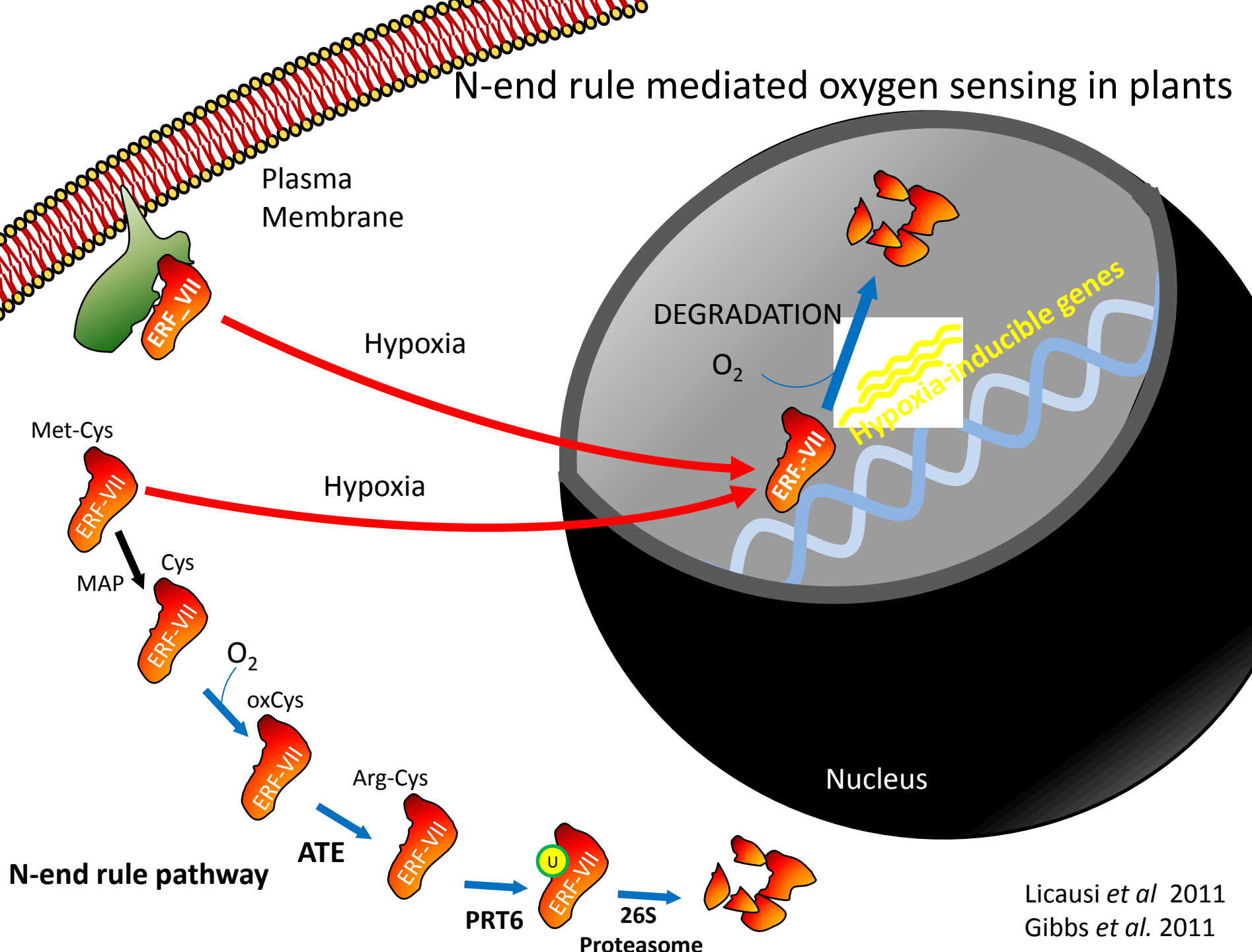
Francesco Licausi – Scuola Superiore Sant'Anna (Pisa)



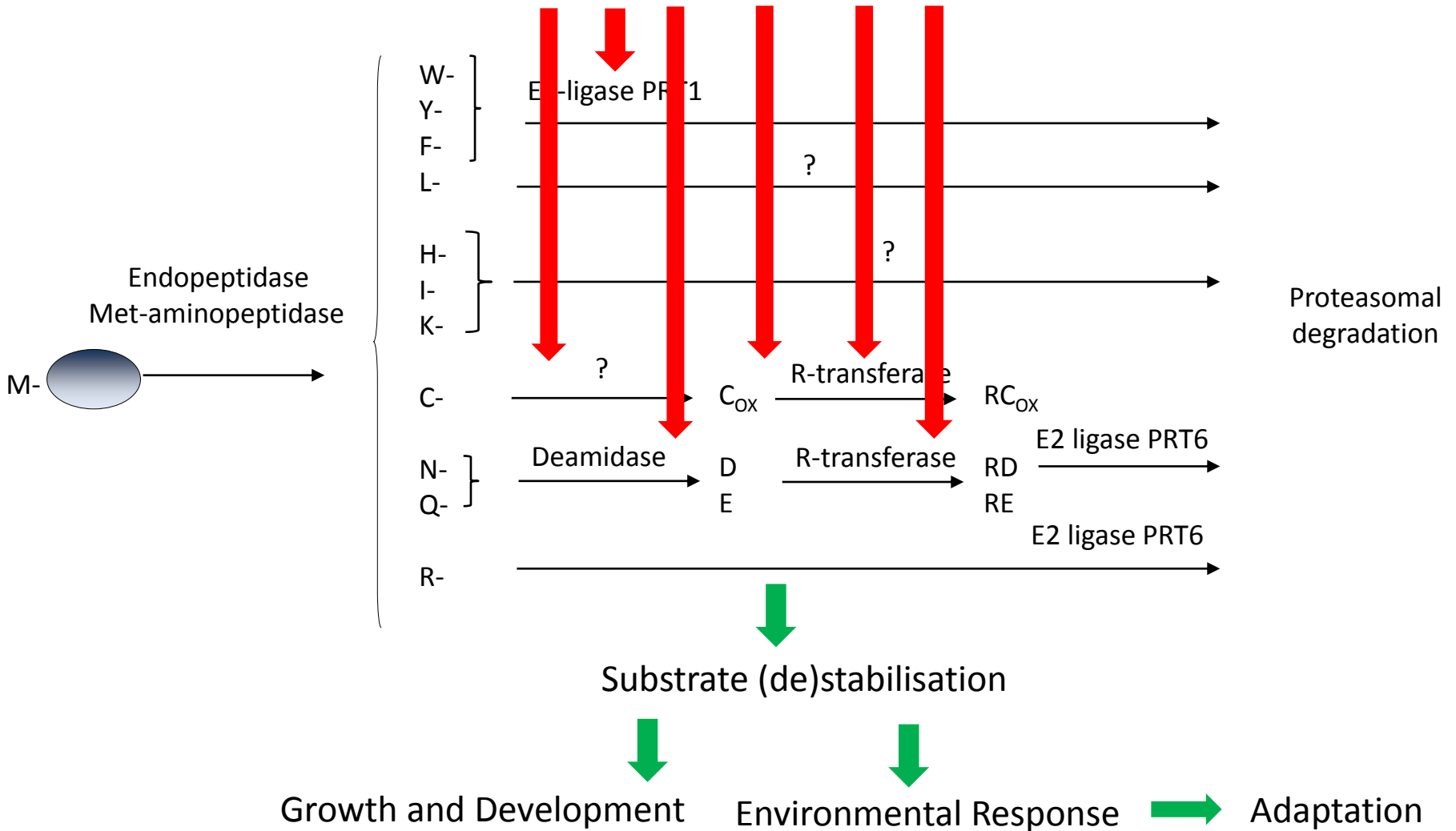
The N-end rule pathway



N-end rule mediated oxygen sensing in plants



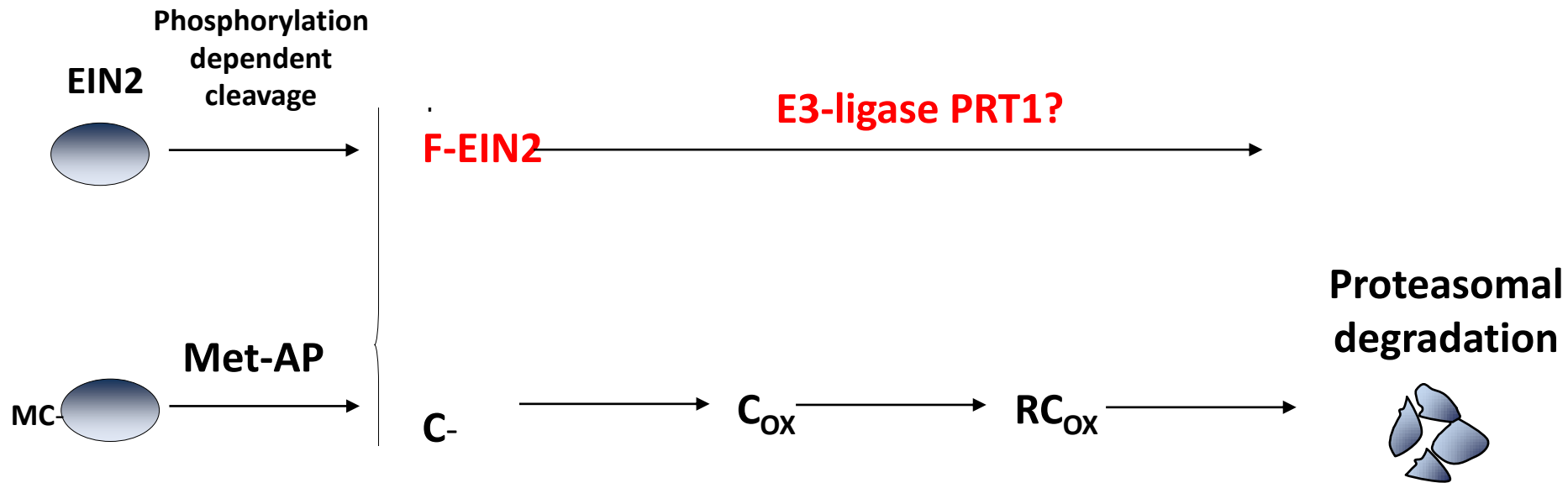
The N-vironment project



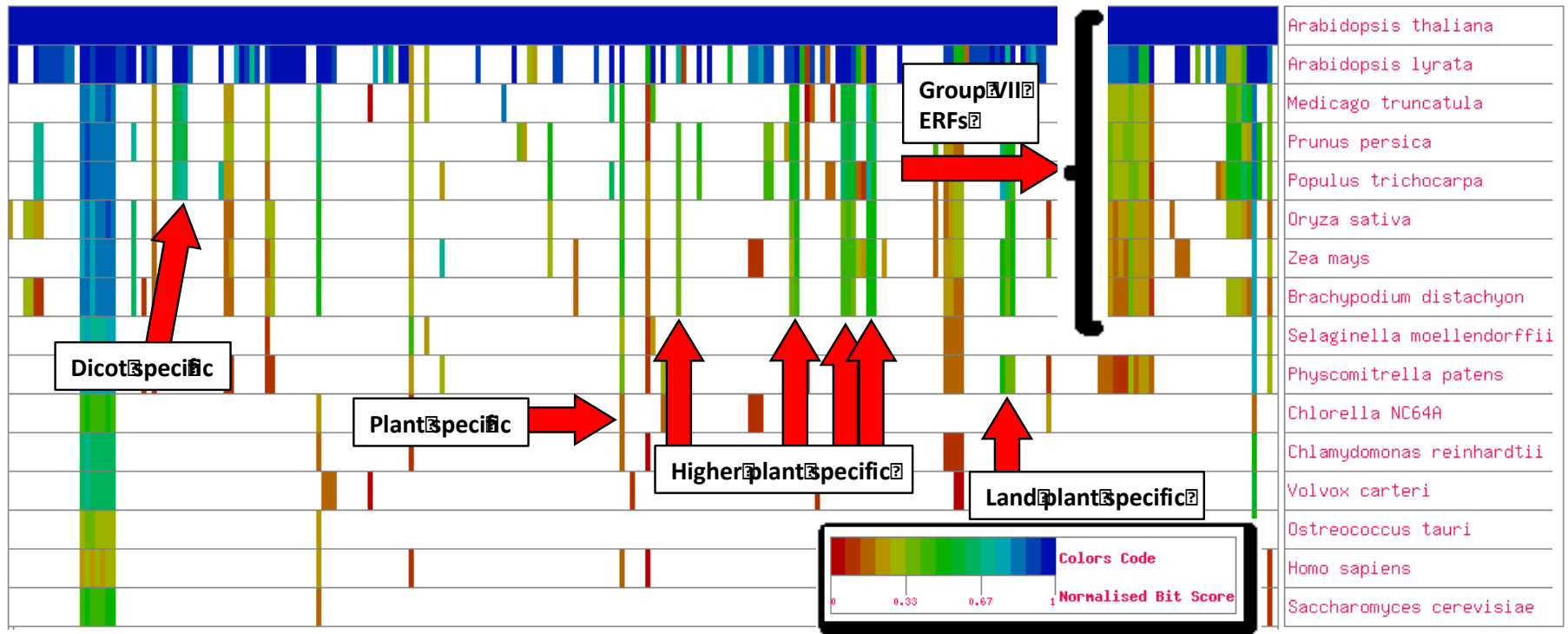
Main aims of the N-vironment project

1. Novel **substrates** and **enzymes** of the N-end rule pathway? (WP1 and 2)
2. Integration of the N-end rule pathway into **signaling pathways** (WP3 and 4)
3. Extent of the N-end rule pathway in controlling **environmental adaptation** (WP5 and 6)

WP1. New substrates of the N-end rule pathway?



WP1 Identification of new substrates of the MC- branch

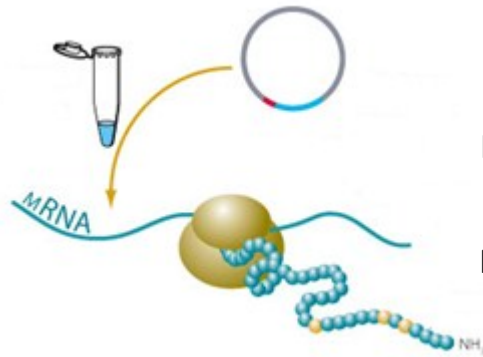


~25% of these MC-proteins will be true substrates, including:

- transcription factors
- kinases
- F-box proteins
- proteins of unknown function

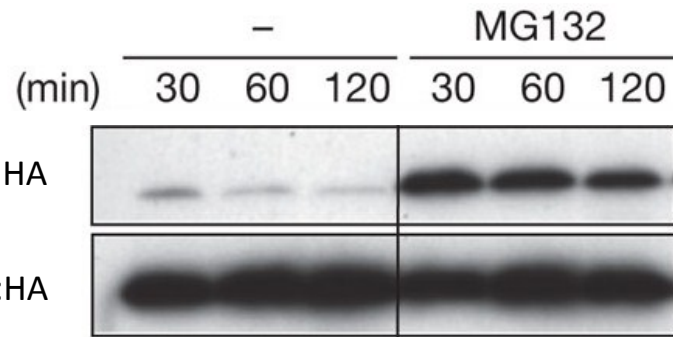
WP1 Validation of new substrates of the MC- branch

In vitro



MC-PROTEIN:HA

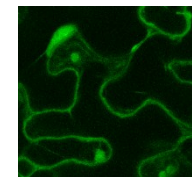
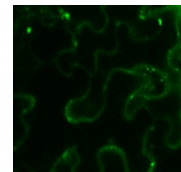
MA-PROTEIN:HA



Gibbs et al. 2011

MC-PROTEIN:GFP

MA-PROTEIN:GFP

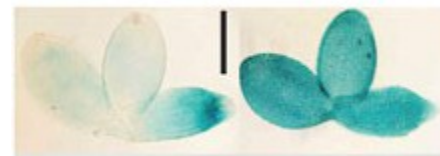


Col-0

prt6

Weits et al. unpub

MC-PROTEIN:GUS

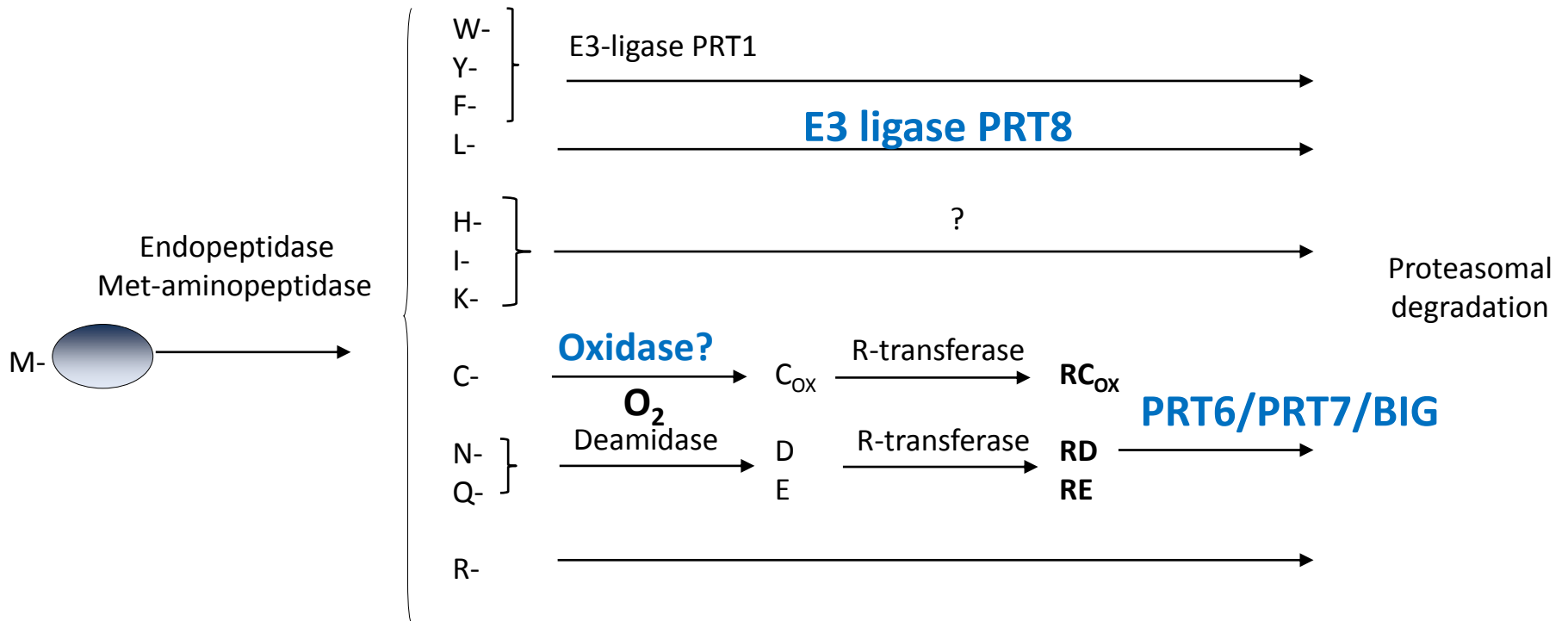


Gibbs et al. 2011

In vivo



WP2. Characterisation of new enzymatic components



WP2 Identification of additional R-branch E3-ligases

Arabidopsis



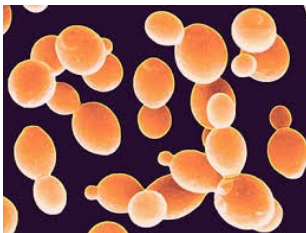
WT
prt7
prt6/prt7
prt7/BIG
prt6/prt7/BIG



Destabilizing residue



Yeast

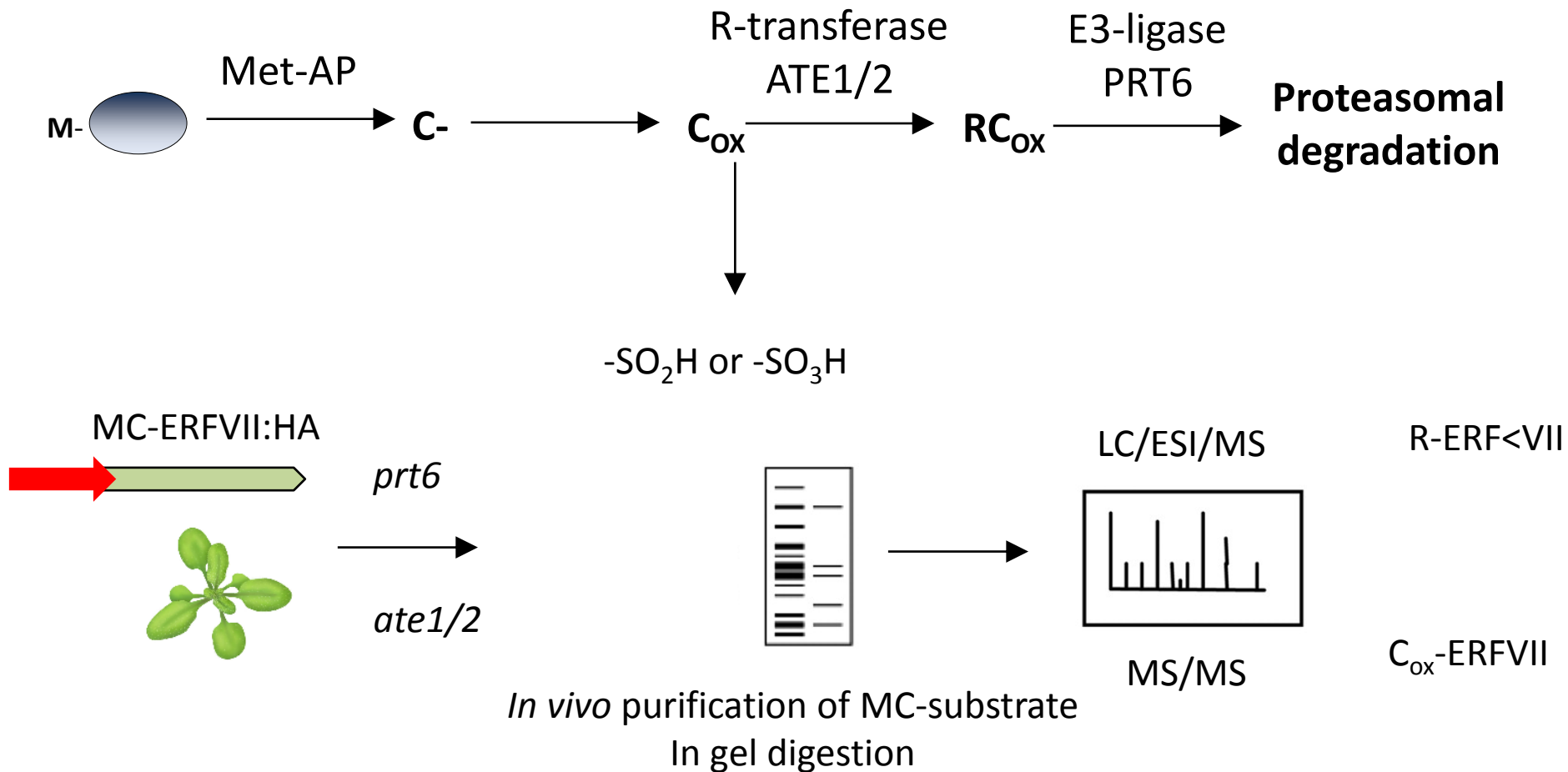


WT
ubr1p

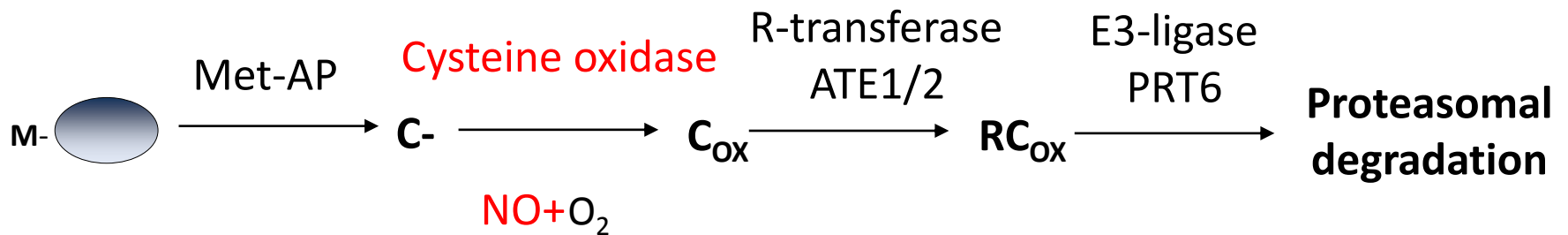
Complementation



WP3 Identification of in vivo intermediates



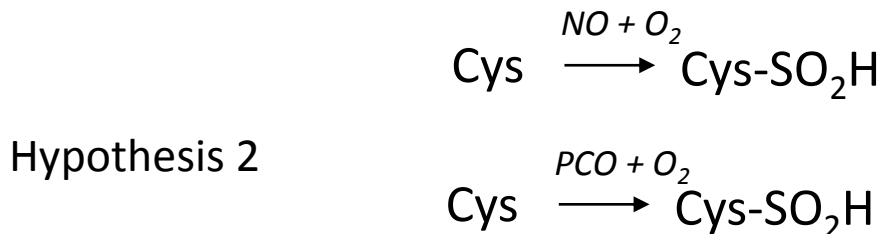
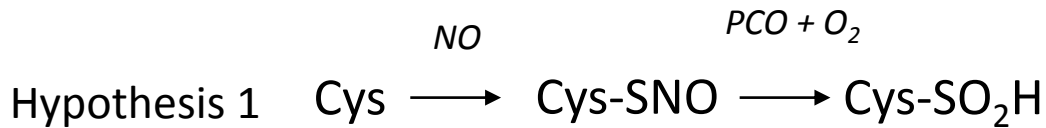
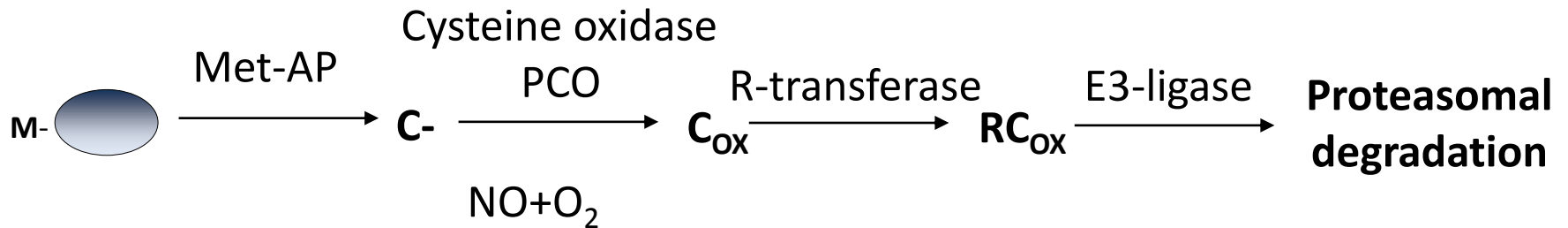
WP3 Regulation of Group-VII ERF stabilisation and destabilisation



Plant cysteine oxidases control the oxygen-dependent branch of the N-end-rule pathway
Weits et al, 2014 Nat Comm

Nitric oxide sensing in plants is mediated by proteolytic control of group VII ERF transcription factors
Gibbs et al, 2014 Mol Cell

WP3 Regulation of Group-VII ERF stabilisation and destabilisation

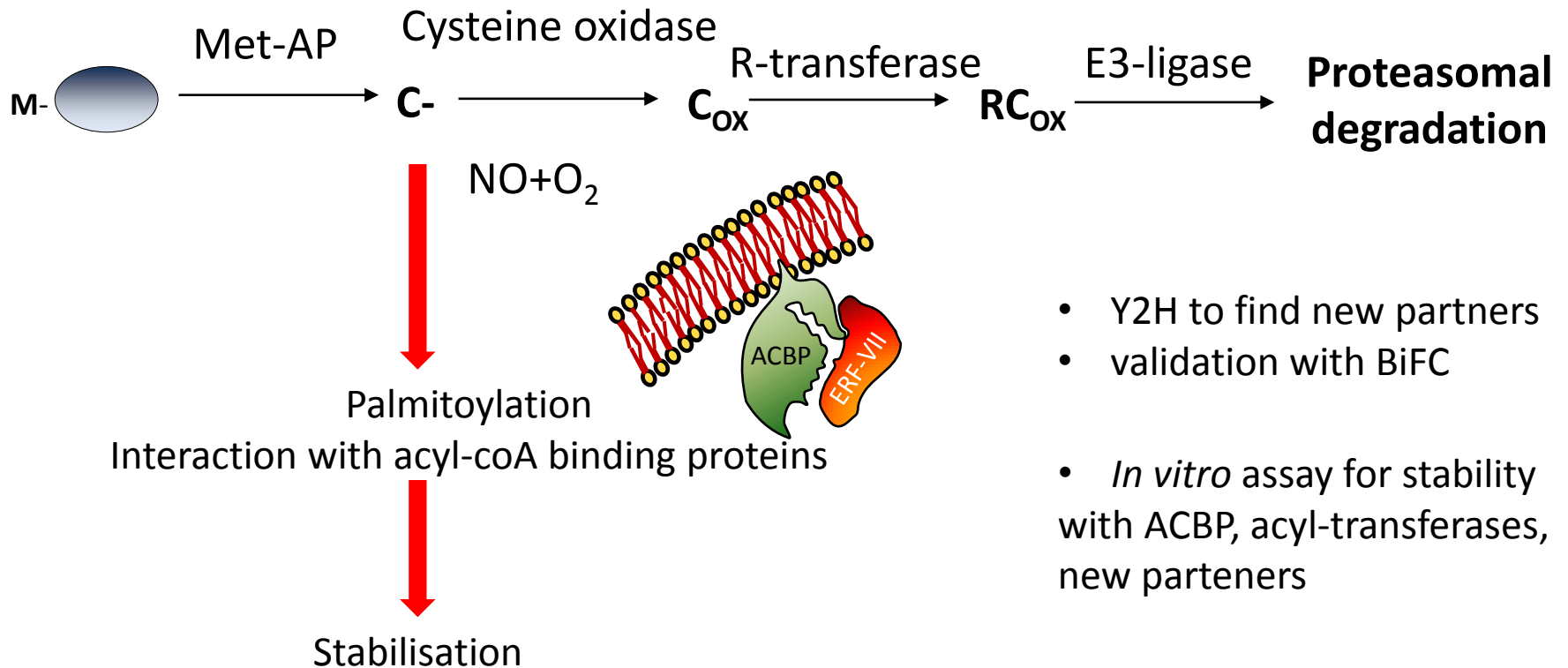


35S:PCO 1in nia1nia2

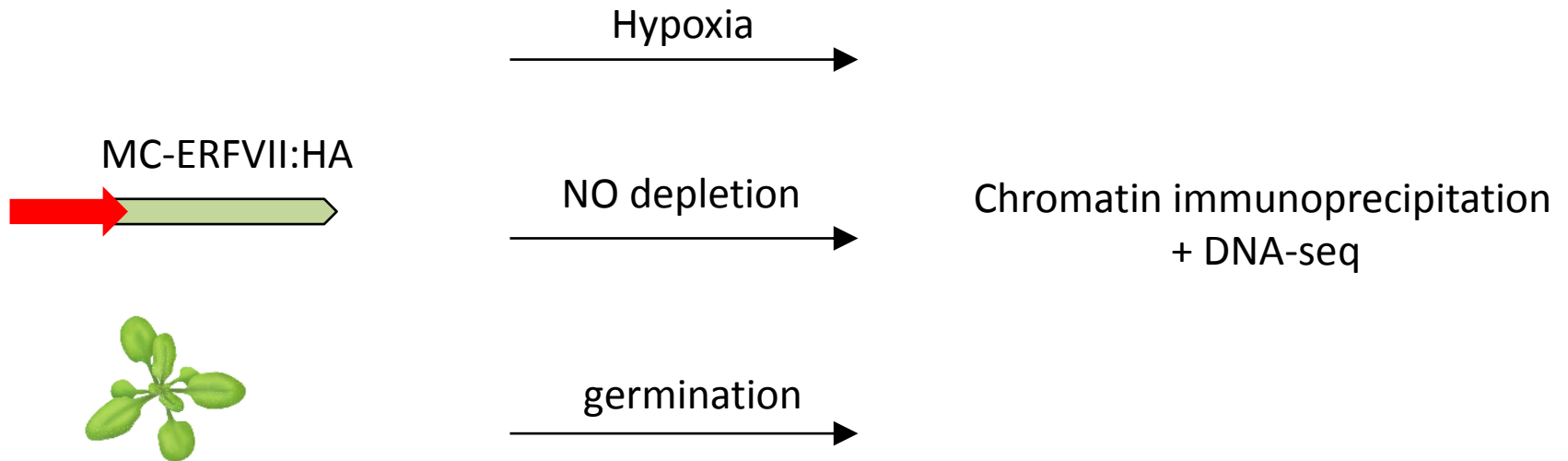
SNAP (NO donor) to *pco1pco2*

CPTIO (NO scavenger) to *35S:PCO1*

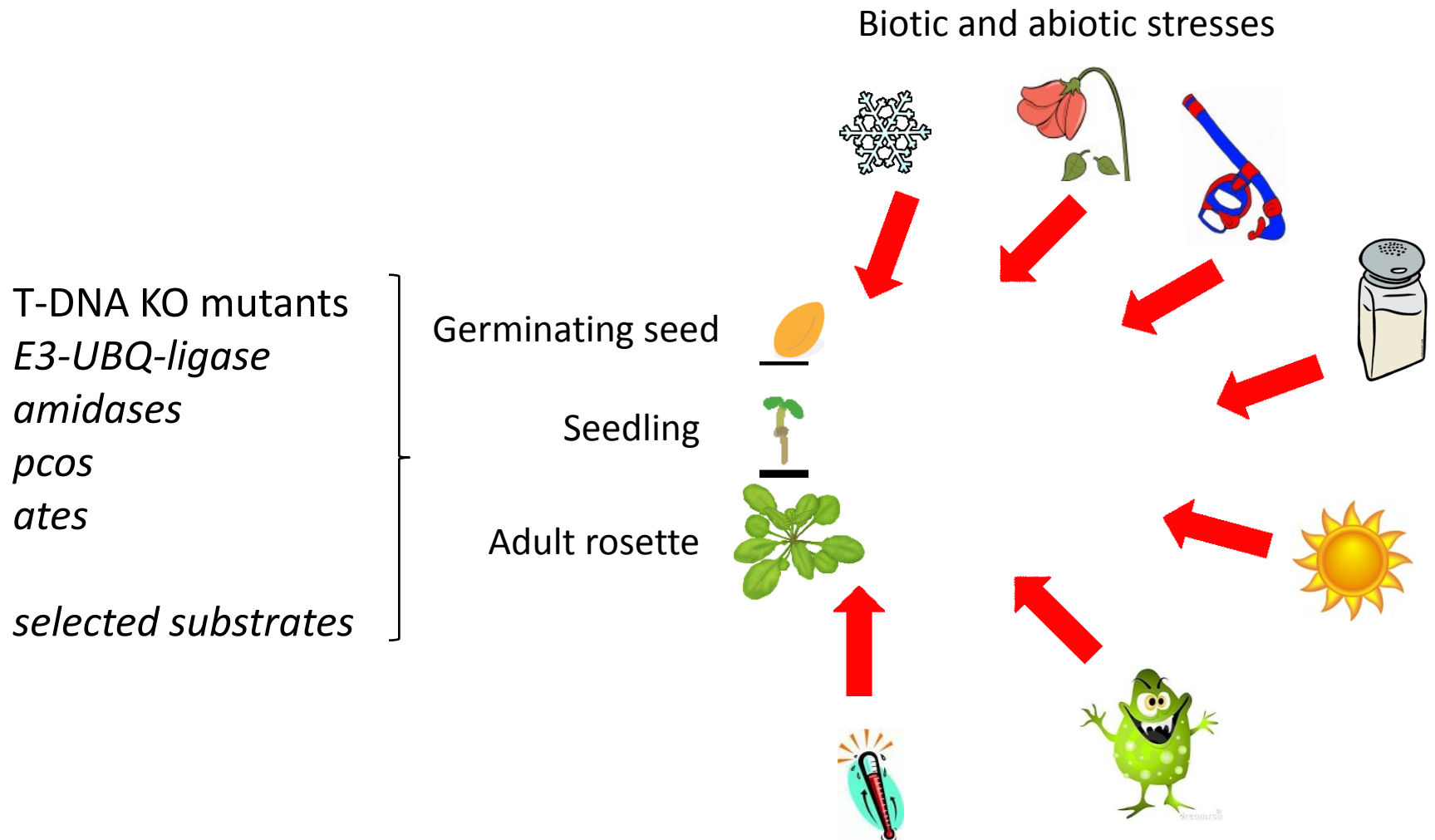
WP4 Physical protection of ERF-VII transcription factors



WP4 Deciphering the downstream cascade after stabilisation of substrates



WP5 Extent of the role of the N-end rule pathway in plant response to the environment



WP 6 Investigating the role of the N-end rule pathway in tomato

Solanum lycopersicum

Wild-type



amiATE



Used to evaluate:

- Environmental stresses
- organoleptic quality
- nutritional properties

Key deliverables

- a searchable platform to access and link the large datasets (open after 6months)
- *open access* publication will be preferred
- Knowledge to improve crop performance in a changing environment