1 **AUTOPHAGY AS AN EMERGING ARENA FOR PLANT-PATHOGEN**

2 **INTERACTIONS**

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Abstract

Autophagy is a highly conserved degradation and recycling process that controls cellular homeostasis, stress adaptation, and programmed cell death in eukaryotes. Emerging evidence indicates that autophagy is a key regulator of plant innate immunity and contributes with both pro-death and pro-survival functions to antimicrobial defences, depending on the pathogenic lifestyle. In turn, several pathogens have co-opted and evolved strategies to manipulate host autophagy pathways to the benefit of infection, while some eukaryotic microbes require their own autophagy machinery for successful pathogenesis. In this review, we present and discuss recent advances that exemplify the important role of pro- and antimicrobial autophagy in plant-pathogen interactions.

Highlights

- Autophagy is an integral part of plant-pathogen interactions.
- A large variety of microbial pathogens target or are targeted by plant autophagy.
- Autophagy in eukaryotic microbial pathogens is essential for pathogenesis.
- Plant autophagy participates in defense responses against invading microbes.
- Successful pathogens have evolved strategies to manipulate plant autophagy.

Introduction

Autophagy is an evolutionary conserved process in eukaryotes that employs double-membrane vesicular structures, termed autophagosomes, to enclose and deliver cytoplasmic material for vacuolar/lysosomal degradation and recycling [1]. Depending on how the cellular cargo is recruited to the developing autophagosomes, autophagy can act as an unspecific (bulk) catabolic pathway for nutrient remobilization and energy supply, or as selective mechanism to eliminate superfluous and harmful compounds including aggregated proteins and damaged organelles [2]. While basal levels of autophagy serves mainly cellular homeostasis and quality control, increased autophagy activity allows adaptation to stressful conditions caused by a large variety of developmental and environmental cues [3]. Besides the significant contribution to cellular and organismal

56 survival, autophagy has been implicated in the regulation and execution of programmed

57 cell death (PCD) in various eukaryotic organisms [4]. In plants, autophagy is increasingly

58 recognized for its central importance in development, reproduction, metabolism,

59 senescence and tolerance to abiotic and biotic stresses [5,6]. In this review, we focus on

60 the role of autophagy during plant–pathogen interactions. In particular, we discuss the

61 most recent evidence showing that plant autophagy may benefit either the host by

62 participating in immune responses, or the invading agent, by contributing to infection.

63

64 The plant immune system has evolved several layers to fend off pathogenic organisms [7].

65 Perception of conserved microbial-associated molecular patterns (MAMPs) by surface

66 receptors leads to activation of basal defenses known as MAMP-triggered immunity (MTI).

67 Adapted pathogens interfere with MTI by secreting effectors that, in turn, can be

68 recognized by resistance (*R*) genes to initiate effector-triggered immunity (ETI). ETI often

69 culminates in a local PCD reaction at the site of pathogen attack, termed the

70 hypersensitive response (HR) [8]. During the last years, it has become evident that

71 autophagy is engaged in various aspects of plant immunity [9]. Most notably, autophagy

72 was shown to regulate basal resistance as well as immunity- and disease-related cell

73 death responses to microbial pathogens with different infection strategies. However, due to

74 the concomitant involvement of plant autophagy in homeostatic, metabolic and

75 developmental processes, the dissection of autophagic mechanisms underlying host

76 immunity and microbial pathogenesis is still in its infancy.

77

78 Most plant pathogens except viruses do not enter the cytoplasmic space, and there is

79 limited evidence for direct autophagic targeting of pathogens or their individual

80 components in in a process resembling xenophagy in metazoans Interestingly, similar to

81 microbes in other host organisms [10,11], an increasing number of examples indicate that

82 phytopathogens are able to manipulate plant autophagy to their own advantage. As

83 detailed below, these include inhibition of autophagy mechanisms contributing to immunity

84 [12-14] and the activation of autophagy pathways to target defense compounds or to

85 potentially enhance nutrient acquisition [15-17].

86

87 **The role of autophagy in eukaryotic plant pathogens**

88

89   It is well established that autophagy components and pathways in eukaryotic microbes are
90   important for pathogenesis and plant invasion. Several studies published in the last
91   decade and summarized in [18] showed that microbial autophagy mediates the
92   development of appressoria, specialized infection structures used by fungi and oomycetes
93   to enter the plant tissue. More recently, new components mediating autophagy-dependent
94   plant infection by fungi have been discovered (Figure 1). The conserved retromer complex
95   is involved in protein trafficking from endosomes to the trans-Golgi network, and was
96   shown to be essential for autophagy-dependent host penetration by the rice blast fungus
97   *Magnaporthe oryzae* [19]. Interestingly, retromer also contributes to the regulation of
98   autophagy-dependent immune cell death in plants [20]. Furthermore, the *M. oryzae* Rab
99   GTPase MoYpt7 is required for fungal autophagy, appressoria development and
100   pathogenicity [21]. Autophagy is also involved in hyphal fusion and positively regulates the
101   virulence of *Fusarium oxysporum* [22]. In *Botrytis cinerea* the autophagy gene BcATG1 is
102   essential for pathogenesis, besides playing a critical role in numerous developmental
103   processes [23]. In several other phytopathogenic fungi, autophagic regulation of organelle
104   quantity has been shown to play a major role in the metabolic switch responsible for the
105   transition to virulence [24].
106
107   **The role of autophagy in plant immunity**
108
109   Despite some remaining controversy, both pro-death and pro-survival functions of
110   autophagy are now generally recognized to contribute to anti-microbial defenses and
111   disease resistance, depending on the pathosystem and pathogenic lifestyle.
112
113   Autophagy can have a positive regulatory role during HR [25] (Figure 1). Several
114   Arabidopsis mutants disrupted in core autophagy (*ATG*) genes or related pathway
115   components displayed significantly reduced HR upon infection with avirulent strains of the
116   bacterium *Pseudomonas syringae* pathovar (pv) *tomato* (*Pst* DC3000) harboring the
117   effector proteins AvrRps4 or AvrRpm1 [20,26-28]. However, autophagy defects seemed to
118   compromise *R* gene-mediated disease resistance only in case of *Pst* DC3000 AvrRps4
119   [20,29], supporting the earlier observed decoupling of HR from growth restriction for
120   AvrRpm1-containing bacteria [30]. Knock-down of *ATG6* homologs in wheat further
121   revealed the engagement of autophagy in broad-spectrum immunity conditioned by the
122   *Pm21 R* gene towards the powdery mildew fungus *Blumeria graminis* f. sp. *tritici* (*Bgt*) [31].

123   Intriguingly, constitutive activation of autophagy in *Nicotiana benthamiana* due to silencing

124   of the ATG3-interacting cytosolic glyceraldehyde-3-phosphate dehydrogenase (GAPC)

125   enhanced *N* gene-mediated HR and resistance against *Tobacco mosaic virus* (TMV) [32].

126   This finding substantiates the death-promoting effect of enhanced autophagy during ETI

127   [33], and explains the increased TMV accumulation previously noted in HR lesions of

128   autophagy-deficient *N. benthamiana* leaves [34]. Furthermore, it adds to the emerging

129   picture that the positive role of autophagy in immunity-related PCD is opposite to its

130   function in preventing premature senescence and runaway cell death outside of the

131   primary infection sites [28,35].

132

133   How autophagy exerts the dual roles during HR activation and containment is not well

134   understood. The influence of autophagy on cellular survival is likely linked to homeostatic

135   functions required to counterbalance infection-induced systemic responses such as ROS

136   production, salicylic acid (SA) signaling, accumulation of misfolded/aggregated proteins,

137   and endoplasmic reticulum stress [26,28,36]. In contrast, the pro-death mechanism of

138   autophagy remains largely undefined, but may also involve the regulation of SA

139   homeostasis and/or the level of NON-EXPRESSOR OF PATHOGENESIS-RELATED

140   GENES 1 (NPR1), that negatively impacts HR [26,28,37]. Future work could further

141   address the potential engagement of selective autophagic processes, e.g. in the removal

142   of negative HR regulators [35].

143

144   There is compelling evidence and a broad consensus that autophagy positively controls

145   plant resistance to necrotrophic pathogens (Figure 1). Autophagy deficiency in Arabidopsis

146   mutants resulted in spreading necrotic lesions and enhanced fungal growth upon infection

147   with *B. cinerea, Alternaria brassicicola, and Plectosphaerella cucumerina* [38-40], and

148   restored susceptibility to a non-pathogenic mutant strain of *Sclerotinia sclerotiorum* [13].

149   Notably, autophagy-mediated disease resistance to *B. cinerea* engages the upstream

150   regulator BAG6 (BCL2-ASSOCIATED ATHANOGENE FAMILY PROTEIN 6) [41]. While

151   Arabidopsis *bag6* mutants were defective in autophagy induction and hypersusceptible to

152   *B. cinerea*, ectopic expression of BAG6 in *N. benthamiana* leaves activated autophagy and

153   cell death, which prevented fungal infection [41]. Hence, pathogen-induced necrotic cell

154   death and disease development is restricted by autophagy and/or immunity-related

155   (autophagic) PCD. This mechanism agrees with the inhibition of necrosis by autophagy

156   during execution of vacuolar cell death in development [42]. The molecular basis of the

157 crosstalk remains largely unknown, although it is evident that protection from *B. cinerea*
158 infection occurs independently of selective autophagy mediated by the cargo receptor
159 NEXT TO BRCA1 GENE 1 (NBR1) [29]. Resistance to necrotrophs may be also mediated
160 by autophagy via modulation of hormone homeostasis, e.g. to stimulate jasmonic acid (JA)
161 defence signaling removal of plant- and removal of pathogen-derived toxic cellular
162 constituents [39].
163
164 In animals, autophagy is a key mechanism in the fight against invading intracellular
165 bacterial and viral pathogens. In contrast, there is surprisingly little knowledge about the
166 contribution of autophagy to basal resistance against viruses, the major intracellular
167 pathogens in plants. Autophagy has been associated with plant antiviral RNA silencing by
168 mediating the targeted degradation of viral silencing suppressors including the
169 cucumovirus protein 2b and potyvirus protein HCpro [43]. Interestingly, potyviral challenge
170 of Arabidopsis lines with reduced expression of the negative autophagy regulator TARGET
171 OF RAPAMYCIN (TOR) revealed strongly decreased levels of Watermelon mosaic virus,
172 whereas Turnip mosaic virus accumulation was only slightly affected [44]. Although the
173 significance of these findings has yet to be verified under autophagy-deficient conditions,
174 they imply an antiviral role of autophagy against some potyviruses, and potentially other
175 unrelated viral species. In this context, it remains to be determined whether autophagy can
176 directly eliminate viruses in a process similar to mammalian xenophagy [45].
177
178 Finally, the role of autophagy in basal resistance to (hemi)biotrophic pathogens is a matter
179 of ongoing debate. So far, there is no evidence that autophagy is directly involved in the
180 regulation of MTI. In addition, despite some conflicting results, autophagy deficiency
181 seems to rather enhance resistance to the virulent bacterial strain *Pst* DC3000 and some
182 powdery mildew fungal species [9]. These findings could be partly linked to the impact of
183 autophagy on SA levels and signaling, which might be further tested in plant systems with
184 enhanced autophagy levels.
185
186 **Pathogen manipulation and pro-microbial role of autophagy**
187
188 Considering the long-lasting co-evolutionary battle between plants and their pathogens, it
189 is not surprising that successful microbes have evolved sophisticated strategies to
190 modulate autophagy to their benefit (Figure 1).

191

192 The necrotroph *S. sclerotiorum* requires the phytotoxin oxalic acid (OA) to trigger

193 unrestricted host cell death and establish successful infection. OA-deficient mutants are

194 non-pathogenic and activate autophagy leading to restrictive HR-like cell death and

195 resistance [13]. Autophagy deficiency restored pathogenicity, indicating that *S.*

196 *sclerotiorum* secretes OA to suppress antimicrobial autophagy. A similar autophagy-

197 mediated mechanism operates in the non-host *Ustilago maydis*-barley interacton. The

198 biotrophic smut fungus *U. maydis* is recognized by barley, triggering a defense response

199 that neutralizes the pathogen and prevents disease, but results in large necrotic areas and

200 stunted leaf growth. In contrast, *U. maydis* mutants lacking the *Pep1* effector show

201 hallmarks of autophagy at the attempted penetration site and remain restricted to the

202 infected area, which might indicate that *Pep1* is an autophagy inhibitor [12]. These findings

203 suggest that autophagy suppression might be a virulence strategy shared by pathogens

204 with completely different lifestyles.

205

206 In line with this notion, binding and activation of TOR by the Cauliflower mosaic virus

207 (CaMV) P6 protein has recently been proposed to inhibit autophagy and impact resistance

208 responses to bacterial pathogens [14]. CaMV infection and transgenic expression of P6

209 increased the susceptibility to *Pst* DC3000 infection and facilitated growth of the effector-

210 delivery deficient *Pst* mutant *hrc*⁻. This effect appears to be in agreement with P6-induced

211 impairment of MTI responses including oxidative burst and SA accumulation. However, it

212 would be surprising if P6 suppression of autophagy is causally linked to the observed

213 phenotype, as *atg* mutants have been shown to display enhanced rather than reduced SA

214 levels and bacterial resistance [38]. Hence, future efforts need to clarify the involvement of

215 autophagy during CaMV infection and to reveal the potential role of TOR-binding of P6 to

216 modulate this pathway for enhanced pathogenicity.

217

218 Other pathogens induce autophagy as part of their infection strategy. For example, the

219 secreted effector AWR5 from the bacterium *Ralstonia solanacearum* inhibits TOR, which

220 results in the activation of autophagy [17]. Although the mechanistic details of this host-

221 pathogen interaction remain to be elucidated, a tantalizing scenario would be that

222 autophagy induction in the host stimulates plant cell dismissal and metabolic re-routing.

223 This would be beneficial for *R. solanacearum* during its transition to the necrotrophic

224 phase by facilitating nutrient acquisition. Viral pathogens might also promote and hijack

225 autophagy pathways to invade host cells. For instance, the viral silencing suppressor P0
226 was shown to trigger autophagic degradation of ARGONAUTE1, an essential component
227 of antiviral RNA-induced silencing complexes [16]. Given the frequent connections
228 between viruses and autophagy in animals [46], future research will most likely provide
229 more cases of virus-induced autophagic degradation of antiviral defense components in
230 plants, perhaps even including small RNAs.

231

232 Another interesting example for the manipulation of the host autophagy machinery by a
233 plant pathogen comes from the hemibiotrophic oomycete *Phytophthora infestans.* The
234 RXLR effector protein PexRD54 was shown to bind to a specific host ATG8 protein, which
235 prevented interaction of ATG8 with the autophagy cargo receptor Joka2/NBR1 [15]. Joka2-
236 mediated selective autophagy was further reported to positively influence plant resistance
237 to *P. infestans*; hence, depletion of Joka2 by PexRD54 enhances susceptibility of the host.
238 Interestingly, both Joka2 and PexRD54 trigger the formation of autophagosomes and
239 activate autophagy. This led the authors to speculate that Joka2 facilitates removal of plant
240 or pathogen proteins that negatively impact immunity, whereas PexRD54 might co-opt the
241 autophagy pathway to selectively eliminate defense-related compounds or to recycle and
242 redistribute nutrients in favor of the pathogen.

243

244 **Conclusions / Future directions**

245

246 This review highlights the importance of autophagy in the field of plant-pathogen
247 interactions. Autophagy has emerged as a central part of the plant weaponry against
248 invading microbial pathogens. Its significance for plant defense is supported by the
249 evolution of microbial strategies to manipulate the host autophagy machinery for enhanced
250 virulence and disease establishment. In addition, autophagy in eukaryotic phytopathogens
251 has evolved as an essential process in the development of functional infection structures.
252 However, the examples illustrating the key roles of autophagy in plant-biotic interactions
253 are still limited both in number and mechanistic detail. Current efforts in several
254 laboratories around the world will certainly help to revert this situation in the coming years
255 and further reveal the highly complex and multifaceted integration of autophagy into the
256 plant immune system.

257

258 A key direction of future research will be the identification and characterization of selective
259 autophagy receptors that drive plant defense responses and are still hidden in the gray
260 shades of "bulk" autophagy. In a more refined interaction, we envisage that plants employ
261 and pathogens manipulate particular selective autophagy pathways to benefit defense and
262 disease, respectively. So far, very few autophagy cargo receptors and their substrates
263 have been identified in plants, but the generally very complex outcome of disease in
264 autophagy deficient plants may indicate that selective processes with distinct functions
265 operate in parallel within the full autophagy response. To dissect these mechanisms in
266 greater detail, we need to establish plant lines with increased "bulk" autophagy to support
267 conclusions from knock-out mutants, and complement these general systems by targeting
268 specifically the different selective autophagy pathways. In addition, due to concomitant,
269 often overlapping roles of autophagy in cellular homeostasis and various developmental
270 and environmental stress responses, it is essential to more precisely inhibit or activate
271 autophagy by inducible and cell type-specific approaches.

273 Another important area of research relates to the largely unexplored crosstalk between
274 autophagy and other cellular pathways that govern proteostasis, hormone signaling, and
275 programmed cell death in plant-microbe interaction. Notably, the plant ubiquitin-
276 proteasome system was recently found to be degraded by autophagy in response to
277 nutrient starvation or chemical and genetic proteasome inhibition [47]. Whether a similar
278 interplay occurs during immunity and disease is not known; however, recent evidence
279 indicates that the 26S proteasome is central to plant immunity and targeted by multiple
280 pathogen effectors to suppress SA-mediated host defenses [48].

282 Overall, there are still only very few pathogens identified that directly modulate the plant
283 autophagy machinery to the benefit of infection. Among these, suppression of autophagy
284 seems to be most common strategy, whereas the potential subversion of bulk and
285 selective pathways still remains merely speculative. However, the fundamental role of
286 autophagy in host immunity and microbial pathogenesis anticipates that phytopathogens
287 have evolved sophisticated capacities to evade and exploit autophagy as demonstrated for
288 a multitude of metazoan pathogens, thus adding further complexity to this emerging arena
289 of plant-microbe interactions.

291 **Acknowledgements**

301

302

Figure Legend

304

Figure 1: Anti- and pro-microbial roles of autophagy during plant-pathogen interactions.

Autophagy is an integral part of plant immunity. Arabidopsis infection with avirulent *Pseudomonas syringae* pv. tomato DC3000 (*avrRps4*) induces autophagy, which contributes to the hypersensitive response (HR) and disease resistance. Infection of Arabidopsis with the necrotrophic fungus *Botrytis cinerea* triggers cleavage of the BAG6 protein, which results in autophagy activation and reduced disease development. Plant autophagy also participates in antiviral defense by targeted degradation of viral silencing suppressors such as the potyvirus protein HCpro and the cucumovirus protein 2b.

Plant pathogens manipulate the host autophagy machinery to counteract host defense and promote virulence. *Phytophthora infestans* effector PexRD54 binds ATG8 and outcompetes the plant selective autophagy receptor Joka2 from autophagosome association, thereby enhancing disease susceptibility of the host. The AWR5 effector from *Ralstonia solanacearum* inhibits TOR to activate autophagy, which is presumed to be beneficial for nutrient acquisition and successful infection. *Sclerotinia sclerotiorum* secretes the toxin oxalic acid to suppress autophagy and HR-like autophagic cell death as part of the host defense response against necrotrophic infection.

323

Autophagy in eukaryotic microbial pathogens contributes to pathogenesis. In *Magnaporthe oryzae*, the retromer complex and Rab GTPase MoYpt7 regulate autophagy mechanisms required for appressoria development and function during infection.

References and recommended reading

Papers of particular interest, published within the period of review, have been highlighted as:

• of special interest

•• of outstanding interest

1. He C, Klionsky DJ: **Regulation mechanisms and signaling pathways of autophagy**. *Annu Rev Genet* 2009, **43**:67-93.

2. Farre JC, Subramani S: **Mechanistic insights into selective autophagy pathways: lessons from yeast**. *Nat Rev Mol Cell Biol* 2016, **17**:537-552.

3. Boya P, Reggiori F, Codogno P: **Emerging regulation and functions of autophagy**. *Nat Cell Biol* 2013, **15**:1017.

4. Anding AL, Baehrecke EH: **Autophagy in Cell Life and Cell Death**. *Curr Top Dev Biol* 2015, **114**:67-91.

5. Yang X, Bassham DC: **New Insight into the Mechanism and Function of Autophagy in Plant Cells**. *Int Rev Cell Mol Biol* 2015, **320**:1-40.

6. Michaeli S, Galili G, Genschik P, Fernie AR, Avin-Wittenberg T: **Autophagy in Plants-- What's New on the Menu?** *Trends Plant Sci* 2016, **21**:134-144.

7. Jones JD, Dangl JL: **The plant immune system**. *Nature* 2006, **444**:323-329.

8. Cui H, Tsuda K, Parker JE: **Effector-triggered immunity: from pathogen perception to robust defense**. *Annu Rev Plant Biol* 2015, **66**:487-511.

9. Zhou J, Yu JQ, Chen ZX: **The perplexing role of autophagy in plant innate immune responses**. *Molecular Plant Pathology* 2014, **15**:637-645.

10. Gomes LC, Dikic I: **Autophagy in antimicrobial immunity**. *Mol Cell* 2014, **54**:224-233.

11. Winchell CG, Steele S, Kawula T, Voth DE: **Dining in: intracellular bacterial pathogen interplay with autophagy**. *Curr Opin Microbiol* 2016, **29**:9-14.

356   12. Hof A, Zechmann B, Schwammbach D, Huckelhoven R, Doehlemann G: **Alternative**
357   **cell death mechanisms determine epidermal resistance in incompatible**
358   **barley-*Ustilago* interactions**. *Mol Plant Microbe Interact* 2014, **27**:403-414.

359   13. Kabbage M, Williams B, Dickman MB: **Cell death control: the interplay of apoptosis**
360   **and autophagy in the pathogenicity of *Sclerotinia sclerotiorum***. *PLoS Pathog*
361   2013, **9**:e1003287.

362   • 14. Zvereva AS, Golyaev V, Turco S, Gubaeva EG, Rajeswaran R, Schepetilnikov MV,
363   Srour O, Ryabova LA, Boller T, Pooggin MM: **Viral protein suppresses oxidative**
364   **burst and salicylic acid-dependent autophagy and facilitates bacterial growth**
365   **on virus-infected plants**. *New Phytol* 2016, **211**:1020-1034.
366   This study proposes that binding and activation of the TOR kinase by the
367   Cauliflower mosaic virus P6 protein prevents SA-mediated induction of autophagy
368   and renders plants more susceptible to virulent bacterial infection.

369   •• 15. Dagdas YF, Belhaj K, Maqbool A, Chaparro-Garcia A, Pandey P, Petre B,
370   Tabassum N, Cruz-Mireles N, Hughes RK, Sklenar J, et al.: **An effector of the**
371   **Irish potato famine pathogen antagonizes a host autophagy cargo receptor**.
372   *Elife* 2016, **5**.
373   This article presents a primary example of how a plant pathogen modulates the
374   autophagy pathway to counteract host defenses and enhance virulence. The
375   *Pytophthora infestans* RXLR effector protein PexRD54 interacts with ATG8 to
376   stimulate autophagosome formation and outcompetes the ATG8-binding cargo
377   receptor Joka2 required for antimicrobial selective autophagy.

378   16. Derrien B, Baumberger N, Schepetilnikov M, Viotti C, De Cillia J, Ziegler-Graff V, Isono
379   E, Schumacher K, Genschik P: **Degradation of the antiviral component**
380   **ARGONAUTE1 by the autophagy pathway**. *Proc Natl Acad Sci U S A* 2012,
381   **109**:15942-15946.

382   • 17. Popa C, Li L, Gil S, Tatjer L, Hashii K, Tabuchi M, Coll NS, Arino J, Valls M: **The**
383   **effector AWR5 from the plant pathogen *Ralstonia solanacearum* is an**
384   **inhibitor of the TOR signalling pathway**. *Sci Rep* 2016, **6**:27058.
385   The authors show that TOR inactivation by the secreted effector AWR5 from
386   *Ralstonia solanacearum* constitutively activates autophagy, which leads to the
387   speculation that autophagy induction might contribute to pathogenicity by
388   facilitating nutrient acquisition during the necrotrophic phase of bacterial infection.

389 18. Talbot NJ, Kershaw MJ: **The emerging role of autophagy in plant pathogen attack**
390 **and host defence**. *Curr Opin Plant Biol* 2009, **12**:444-450.

391 19. Zheng W, Zhou J, He Y, Xie Q, Chen A, Zheng H, Shi L, Zhao X, Zhang C, Huang Q,
392 et al.: **Retromer Is Essential for Autophagy-Dependent Plant Infection by the**
393 **Rice Blast Fungus**. *PLoS Genet* 2015, **11**:e1005704.

394 20. Munch D, Teh OK, Malinovsky FG, Liu Q, Vetukuri RR, El Kasmi F, Brodersen P,
395 Hara-Nishimura I, Dangl JL, Petersen M, et al.: **Retromer contributes to**
396 **immunity-associated cell death in Arabidopsis**. *Plant Cell* 2015, **27**:463-479.

397 21. Liu XH, Chen SM, Gao HM, Ning GA, Shi HB, Wang Y, Dong B, Qi YY, Zhang DM, Lu
398 GD, et al.: **The small GTPase MoYpt7 is required for membrane fusion in**
399 **autophagy and pathogenicity of** *Magnaporthe oryzae*. *Environ Microbiol* 2015,
400 **17**:4495-4510.

401 22. Corral-Ramos C, Roca MG, Di Pietro A, Roncero MI, Ruiz-Roldan C: **Autophagy**
402 **contributes to regulation of nuclear dynamics during vegetative growth and**
403 **hyphal fusion in** *Fusarium oxysporum*. *Autophagy* 2015, **11**:131-144.

404 23. Ren W, Zhang Z, Shao W, Yang Y, Zhou M, Chen C: **The autophagy-related gene**
405 **BcATG1 is involved in fungal development and pathogenesis in Botrytis**
406 **cinerea**. *Mol Plant Pathol* 2017, **18**:238-248.

407 24. Oku M, Takano Y, Sakai Y: **The emerging role of autophagy in peroxisome**
408 **dynamics and lipid metabolism of phyllosphere microorganisms**. *Front Plant*
409 *Sci* 2014, **5**:81.

410 25. Hofius D, Schultz-Larsen T, Joensen J, Tsitsigiannis DI, Petersen NH, Mattsson O,
411 Jorgensen LB, Jones JD, Mundy J, Petersen M: **Autophagic components**
412 **contribute to hypersensitive cell death in Arabidopsis**. *Cell* 2009, **137**:773-783.

413 26. Coll NS, Smidler A, Puigvert M, Popa C, Valls M, Dangl JL: **The plant metacaspase**
414 **AtMC1 in pathogen-triggered programmed cell death and aging: functional**
415 **linkage with autophagy**. *Cell Death Differ* 2014.

416 27. Hackenberg T, Juul T, Auzina A, Gwizdz S, Malolepszy A, Van Der Kelen K, Dam S,
417 Bressendorff S, Lorentzen A, Roepstorff P, et al.: **Catalase and NO CATALASE**
418 **ACTIVITY1 Promote Autophagy-Dependent Cell Death in Arabidopsis**. *Plant*
419 *Cell* 2013, **25**:4616-4626.

420 • 28. Munch D, Rodriguez E, Bressendorff S, Park OK, Hofius D, Petersen M: **Autophagy**
421 **deficiency leads to accumulation of ubiquitinated proteins, ER stress, and**
422 **cell death in Arabidopsis**. *Autophagy* 2014, **10**:1579-1587.

This study provides evidence that the plant retromer complex is involved in pathogen-triggered HR and plays an important role in the regulation of the autophagic processes, similar to other eukaryotic organisms.

29. Dong J, Chen W: **The role of autophagy in chloroplast degradation and chlorophagy in immune defenses during *Pst* DC3000 (*AvrRps4*) infection**. *PLoS ONE* 2013, **8**:e73091.

30. Coll NS, Vercammen D, Smidler A, Clover C, Van Breusegem F, Dangl JL, Epple P: **Arabidopsis type I metacaspases control cell death**. *Science* 2010, **330**:1393-1397.

31. Yue J, Sun H, Zhang W, Pei D, He Y, Wang H: **Wheat homologs of yeast ATG6 function in autophagy and are implicated in powdery mildew immunity**. *BMC Plant Biol* 2015, **15**:95.

•• 32. Han S, Wang Y, Zheng X, Jia Q, Zhao J, Bai F, Hong Y, Liu Y: **Cytoplastic Glyceraldehyde-3-Phosphate Dehydrogenases Interact with ATG3 to Negatively Regulate Autophagy and Immunity in *Nicotiana benthamiana***. *Plant Cell* 2015, **27**:1316-1331.

This study reveals a direct link between the cytosolic glyceraldehyde-3-phosphate dehydrogenases (GAPC) and the autophagy pathway, and shows that stimulated autophagy activity in GAPC-silenced tobacco leaves promotes HR and disease resistance upon avirulent virus infection.

33. Kwon SI, Cho HJ, Kim SR, Park OK: **The Rab GTPase RabG3b positively regulates autophagy and immunity-associated hypersensitive cell death in Arabidopsis**. *Plant Physiol* 2013, **161**:1722-1736.

34. Liu Y, Schiff M, Czymmek K, Talloczy Z, Levine B, Dinesh-Kumar SP: **Autophagy regulates programmed cell death during the plant innate immune response**. *Cell* 2005, **121**:567-577.

35. Minina EA, Bozhkov PV, Hofius D: **Autophagy as initiator or executioner of cell death**. *Trends Plant Sci* 2014, **19**:692-697.

36. Yoshimoto K, Jikumaru Y, Kamiya Y, Kusano M, Consonni C, Panstruga R, Ohsumi Y, Shirasu K: **Autophagy negatively regulates cell death by controlling NPR1-dependent salicylic acid signaling during senescence and the innate immune response in Arabidopsis**. *Plant Cell* 2009, **21**:2914-2927.

455 37. Fu ZQ, Yan S, Saleh A, Wang W, Ruble J, Oka N, Mohan R, Spoel SH, Tada Y, Zheng
456   N, et al.: **NPR3 and NPR4 are receptors for the immune signal salicylic acid in**
457   **plants**. *Nature* 2012, **486**:228-232.

458 38. Lenz HD, Haller E, Melzer E, Gust AA, Nurnberger T: **Autophagy controls plant**
459   **basal immunity in a pathogenic lifestyle-dependent manner**. *Autophagy* 2011,
460   **7**:773-774.

461 39. Lai Z, Wang F, Zheng Z, Fan B, Chen Z: **A critical role of autophagy in plant**
462   **resistance to necrotrophic fungal pathogens**. *Plant J* 2011, **66**:953-968.

463 40. Katsiarimpa A, Kalinowska K, Anzenberger F, Weis C, Ostertag M, Tsutsumi C,
464   Schwechheimer C, Brunner F, Huckelhoven R, Isono E: **The deubiquitinating**
465   **enzyme AMSH1 and the ESCRT-III subunit VPS2.1 are required for**
466   **autophagic degradation in Arabidopsis**. *Plant Cell* 2013, **25**:2236-2252.

467 •• 41. Li Y, Kabbage M, Liu W, Dickman MB: **Aspartyl Protease-Mediated Cleavage of**
468   **BAG6 Is Necessary for Autophagy and Fungal Resistance in Plants**. *Plant Cell*
469   2016, **28**:233-247.
470   The authors show that infection with *B. cinerea* triggers cleavage of the co-
471   chaperone BAG6 in a caspase-1-like dependent manner and activates autophagy
472   in the host. Autophagy induction results in disease resistance, coupling fungal
473   recognition with defense activation.

474 42. Minina EA, Filonova LH, Fukada K, Savenkov EI, Gogvadze V, Clapham D, Sanchez-
475   Vera V, Suarez MF, Zhivotovsky B, Daniel G, et al.: **Autophagy and metacaspase**
476   **determine the mode of cell death in plants**. *J Cell Biol* 2013, **203**:917-927.

477 43. Nakahara KS, Masuta C, Yamada S, Shimura H, Kashihara Y, Wada TS, Meguro A,
478   Goto K, Tadamura K, Sueda K, et al.: **Tobacco calmodulin-like protein provides**
479   **secondary defense by binding to and directing degradation of virus RNA**
480   **silencing suppressors**. *Proc Natl Acad Sci U S A* 2012, **109**:10113-10118.

481 44. Ouibrahim L, Rubio AG, Moretti A, Montane MH, Menand B, Meyer C, Robaglia C,
482   Caranta C: **Potyviruses differ in their requirement for TOR signalling**. *J Gen*
483   *Virol* 2015, **96**:2898-2903.
484   This study reports enhanced resistance of TOR-silenced Arabidopsis lines to
485   potyvirus infection, which implies an important role of activated autophagy in
486   antiviral immunity.

487 45. Paulus GL, Xavier RJ: **Autophagy and checkpoints for intracellular pathogen**
488   **defense**. *Curr Opin Gastroenterol* 2015, **31**:14-23.

489   46. Dong X, Levine B: **Autophagy and viruses: adversaries or allies?** *J Innate Immun*
490   2013, **5**:480-493.
491   47. Marshall RS, Li F, Gemperline DC, Book AJ, Vierstra RD: **Autophagic Degradation**
492   **of the 26S Proteasome Is Mediated by the Dual ATG8/Ubiquitin Receptor**
493   **RPN10 in Arabidopsis**. *Mol Cell* 2015, **58**:1053-1066.
494   48. Ustun S, Sheikh A, Gimenez-Ibanez S, Jones A, Ntoukakis V, Bornke F: **The**
495   **Proteasome Acts as a Hub for Plant Immunity and Is Targeted by**
496   *Pseudomonas* **Type III Effectors**. *Plant Physiol* 2016, **172**:1941-1958.
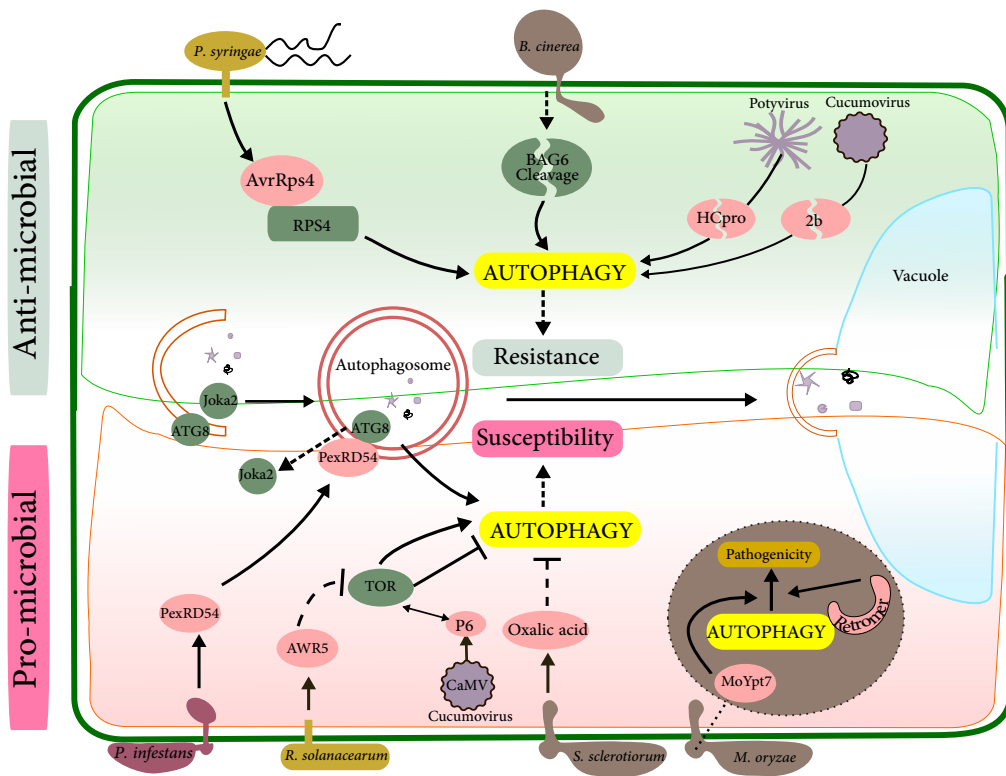
Highlights (3-5 bullet points 85 characters each)

- Autophagy is an integral part of plant-pathogen interactions.
- A large variety of microbial pathogens target or are targeted by plant autophagy.
- Autophagy in eukaryotic microbial pathogens is essential for pathogenesis.
- Plant autophagy participates in defense responses against invading microbes.
- Successful pathogens have evolved strategies to manipulate plant autophagy.