

University of Dundee

The second International Symposium on Fungal Stress

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Published in:
Fungal Biology

DOI:
[10.1016/j.funbio.2017.10.011](https://doi.org/10.1016/j.funbio.2017.10.011)

Publication date:
2018

Document Version
Peer reviewed version

[Link to publication in Discovery Research Portal](#)

Citation for published version (APA):

Alder-Rangel, A., Bailão, A. M., da Cunha, A. F., Soares, C. M. A., Wang, C., Bonatto, D., Dadachova, E., Hakalehto, E., Eleutherio, E. C. A., Fernandes, É. K. K., Gadd, G. M., Braus, G. H., Braga, G. U. L., Goldman, G. H., Malavazi, I., Hallsworth, J. E., Takemoto, J. Y., Fuller, K. K., Selbmann, L., ... Rangel, D. E. N. (2018). The second International Symposium on Fungal Stress: ISFUS. *Fungal Biology*, 122(6), 386-399. <https://doi.org/10.1016/j.funbio.2017.10.011>

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The Second International Symposium on Fungal Stress: ISFUS

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63

64

65 **Abstract**

66 The topic of ‘fungal stress’ is central to many important disciplines, including medical
67 mycology, chronobiology, plant and insect pathology, industrial microbiology, material sciences,
68 and astrobiology. The International Symposium on Fungal Stress (ISFUS) brought together
69 researchers, who study fungal stress in a variety of fields. The second ISFUS was held in May
70 2017 in Goiania, Goiás, Brazil and hosted by the Instituto de Patologia Tropical e Saúde Pública
71 at the Universidade Federal de Goiás. It was supported by grants from CAPES and FAPEG.
72 Twenty-seven speakers from 15 countries presented their research related to fungal stress
73 biology. The Symposium was divided into seven general topics: 1. Fungal biology in extreme
74 environments; 2. Stress mechanisms and responses in fungi: molecular biology, biochemistry,
75 biophysics, and cellular biology; 3. Fungal photobiology in the context of stress; 4. Role of stress
76 in fungal pathogenesis; 5. Fungal stress and bioremediation; 6. Fungal stress in agriculture and
77 forestry; and 7. Fungal stress in industrial applications. This article provides an overview of the
78 science presented and discussed at ISFUS-2017.

79

80 **Keywords:** agricultural mycology; forest mycology; industrial mycology; medical mycology;
81 fungal stress mechanisms and responses.

82

83 **Introduction**

84 Research into fungal biology and fungal stress can help solve many issues related to
85 human health, climate change, food security, environmental impacts, etc. (Rangel et al., 2015a;
86 Rangel et al., 2015b). Fungi can be used for bioremediation (Gadd, 2016); to replace synthetic
87 pesticides (Li et al., 2010; Rangel and Correia, 2003; Santi et al., 2011); to produce biofuels

88 (Alper et al., 2006; Lam et al., 2014), novel antibiotics (Mygind et al., 2005), enzymes
89 (Maheshwari et al., 2000), and useful chemicals (Hagedorn and Kaphammer, 1994). Soil-
90 dwelling fungi may enhance plant health and crop production of in arid environments (Gal-
91 Hemed et al., 2011; Molina-Montenegro et al., 2016), and fungi can help degrade and valorize
92 organic waste materials (Hultberg and Bodin, 2017). Understanding fungal metabolism at
93 biophysical extremes for life can enable effective preservation of foods, documents, and artefacts
94 and has implications for astrobiology in relation to habitability of hostile environments and
95 preventing contamination of other planetary bodies during space exploration (Stevenson et al.,
96 2015a; Stevenson et al., 2015b; Stevenson et al., 2017). On the other hand, fungal pathogens
97 represent a serious threat to crops, animals, and humans (Rangel et al., 2015a). Fungi are also
98 used as models for basic research on the biology of eukaryotic cells (Rangel et al., 2015a). Thus,
99 understanding how fungi deal with stress during growth or while infecting a host will help
100 optimize the use of fungi in biotechnological applications, improve the environment, and fight
101 fungal diseases.

102 Like all living organisms, fungi must cope with a variety of stresses to survive, including
103 ionizing radiation (Dadachova and Casadevall, 2008; Zhdanova et al., 2000); water activity
104 (Stevenson et al., 2015a); acidic and alkaline environments (Rangel et al., 2015c; Steiman et al.,
105 2004); hypoxic or anoxic stress (Bonaccorsi et al., 2006; Camilo et al., 2008; Hillmann et al.,
106 2015; Rangel et al., 2015c); chaotropicity (Hallsworth et al., 2003); hydrophobicity (Bhaganna et
107 al., 2010); poisons and toxic chemicals (Pennisi, 2004; Pointing, 2001); solar UV radiation
108 (Braga et al., 2001; Braga et al., 2015; Braga et al., 2002); agricultural and industrial pollutants
109 (Pennisi, 2004; Rangel et al., 2010a); biotic stress (Druzhinina et al., 2011; Saxena et al., 2015);
110 nutritive stress (Ferreira et al., 2017; Rangel et al., 2006a); oxidative stress (reactive oxygen

111 species, ROS) (Azevedo et al., 2014; Eleutherio et al., 2015; Huarte-Bonnet et al., 2015; Rangel,
112 2011); heat stress (Rangel et al., 2005; Rangel et al., 2010b; Souza et al., 2014); cold activity
113 (Santos et al., 2011); and extreme cold (Chin et al., 2010; Selbmann et al., 2015).

114 The International Symposium on Fungal Stress (ISFUS) is a unique meeting which brings
115 together mycology community under the common umbrella of "stress", be it environmental or
116 host-related, and promotes unique interaction between researchers and cross-pollination of ideas.
117 Before ISFUS, there had never been a scientific meeting specifically dedicated to the study of
118 fungal stress.

119 The first ISFUS was held in 2014 in São José dos Campos, in the state of São Paulo,
120 Brazil featured presentations from 33 researchers from 10 countries, including students and
121 young, mid-career, and senior researchers. A good number of discussions, collaborations, and
122 new friendships were also initiated. (Rangel et al., 2015a; Rangel et al., 2015b). Researchers,
123 who had been studying fungal stress for years and knew each other through journal articles were
124 given the opportunity to finally meet. Young scientists were able to connect with more
125 experienced researchers. From the first ISFUS, over 33 collaborative articles have been
126 published, which included those published in a special issue of *Current Genetics* that was
127 devoted to ISFUS 2014 (Rangel et al., 2015a; Rangel et al., 2015b). The first ISFUS was
128 supported by a generous grant from FAPESP (São Paulo Research Foundation).

129 The first ISFUS was such a success that the organizers decided to host another ISFUS.
130 The second ISFUS occurred in May 2017 in Goiania, Goiás, Brazil. Drauzio E.N. Rangel
131 conceived and was the primary organizer of both symposia. Alene Alder-Rangel, Gilberto U. L.
132 Braga, John E. Hallsworth, and Luis M. Corrochano helped bring the symposia to fruition.

133 The second ISFUS was supported by grants from CAPES and FAPEG. The Instituto de
134 Patologia Tropical e Saúde Pública at the Universidade Federal de Goiás (UFG) acted as the host
135 institution. Corporate sponsors included Elsevier (Amsterdam, Netherlands - which provided the
136 students awards), Biocontrol (Sertãozinho, SP, Brazil), Koppert Biological Systems (Piracicaba,
137 SP, Brazil), and Alder’s English Services (Goiânia, GO, Brazil). The logo of the 2017
138 symposium (Figure 1) features one of the most-studied ascomycetes, *Aspergillus nidulans*, and
139 illustrates several key stress parameters that fungi cope with to survive. Twenty-seven speakers
140 from 15 countries (Figures 2 and 3) presented talks about their cutting-edge research related to
141 fungal stress. In addition, there were 42 poster presentations. One hundred participants attended
142 ISFUS-2017, with about a third from the UFG, and the rest from other Brazilian and
143 international universities.

144 The ISFUS-2017 Abstracts Book, which feature abstracts from the presentations and
145 posters, is available in the Electronic Supplementary Material 1 of this article. Articles
146 highlighting the contributions to the conference are published in this *Fungal Biology* special
147 issue entitled “Biology of Fungal Systems under Stress”, which focuses on cellular biology,
148 ecology, environment, agriculture, medical mycology, and biotechnology in the context of fungal
149 stress biology (Chen et al., 2017; Ferreira et al., 2017; Huarte-Bonnet et al., 2017b; Keyhani,
150 2017; Malo et al., 2017; Muniz et al., 2017; Tonani et al., 2017).

151 By design, the second ISFUS facilitated interactions between researchers from Brazil and
152 other countries. Researchers were provided platforms to discuss their work in-depth with the
153 diverse yet intimate group. Long lunch breaks and social activities provided many opportunities
154 for all the delegates to interact, share ideas, and promote lasting contacts and collaborations.
155 Assembling active researchers for the purpose of interdisciplinary exchange will inevitably

156 stimulate ideas for new lines of research across international borders. Several such new
157 international collaborations are already underway due to this ISFUS.

158 **ISFUS 2017: a brief synopsis**

159 The ISFUS-2017 began Monday morning with a welcome talk by Drauzio E. N. Rangel.
160 He encouraged everyone to follow their intuition as inspired by Albert Einstein: “The intellect
161 has little to do on the road to discovery. There comes a leap in consciousness, call it Intuition or
162 what you will, the solution comes to you and you don't know how or why.” Drauzio said:
163 “Intuition comes when you have an open and clear heart like a child to follow where your
164 curiosity leads you, in science and in life.”

165 Flávia Aparecida de Oliveira (Figure 4), the Director of the Instituto de Patologia
166 Tropical e Saúde Pública (the host institution), welcomed researchers. The president of the
167 Foundation for Research Support of Goiás (FAPEG), Maria Zaira Turchi (Figure 5), praised the
168 work of the organizers of the Symposium saying, “Times are difficult; despite the little financial
169 incentive in Brazil, the determination of faculty professors to make things happen is very
170 beautiful.” Also participating in the opening ceremony were Dr. João Teodoro Pádua (Figure 5),
171 Chief of Staff of the University presidency; Dr. Luis M. Corrochano, Universidad de Sevilla
172 (Figure 5); Dr. John E. Hallsworth, Queen’s University Belfast (Figure 5); and Alene Alder-
173 Rangel, the co-chair (Figure 5).

174 The Symposium was organized around seven general topics related to fungal stress
175 accordingly to the program found in the Electronic Supplementary Material 2:

176 **1. Fungal biology in extreme environments**

177 The microbial biosphere is limited by thermodynamic and biophysical parameters such as
178 temperature, water activity, chaotropicity, salinity, and ionizing radiation. Fungi survive and, in

179 the case of extremophiles, retain metabolic activity and grow at the thermodynamic fringes for
180 life on Earth. Therefore, elucidating cellular stress mechanisms and responses and adaptations in
181 fungi are imperative to understand life in the context of the various constraints for Earth
182 ecosystems (Runner and Brewster, 2003; Stevenson et al., 2017; Yakimov et al., 2015).

183 Ekaterina Dadachova talked about the resistance of melanized fungi to both sparsely and
184 densely ionizing radiation (Casadevall et al., 2017; Pacelli et al., 2017a; Shuryak et al., 2015).
185 Two melanized fungi - a fast-growing *Cryptococcus neoformans* and a slow-growing *Cryomyces*
186 *antarcticus* - were subjected to densely ionizing deuterons. Melanin protected both fungi;
187 however, *Cryomyces antarcticus* was more resistant to deuterons than *Cryptococcus neoformans*.
188 The irradiated cells were analyzed by a panel of metabolic assays – XTT (2,3-bis(2-methoxy-4-
189 nitro-5-sulfophenyl)-5-[(phenylamino)carbonyl]-2H-tetrazolium hydroxide), MTT (2-(4,5-
190 dimethyl-2-thiazolyl)-3,5-diphenyl-2H-tetrazolium bromide), and ATP (adenosine triphosphate).
191 XTT showed increased activity in melanized strains of both species, while the activity in non-
192 melanized cells either remained stable or decreased. In a follow-up study performed with only *C.*
193 *neoformans* cells, transmission electron microscopy (TEM) demonstrated the removal of
194 polysaccharide capsules by radiation, in both melanized and non-melanized cells, and
195 considerable damage to the cell wall and organelles was observed in the non-melanized cells.

196 Laura Selbmann also discussed the resistance of the Antarctic cryptoendolithic fungus
197 *Cryomyces antarcticus* to radiation. This organism is a perfect model for astrobiological studies
198 because it lives in the closest Mars analogue on Earth, the McMurdo Dry Valleys in Antarctica
199 (Selbmann et al., 2015). This fungus has been selected for a number of astrobiological projects:
200 STARLIFE (Moeller et al., 2017); and two funded by the European Space Agency (ESA) and
201 Italian Space Agency (ASI): LIFE and BIOlogy and Mars Experiment, (Onofri et al., 2012;

202 Onofri et al., 2015); and Lichens and Fungi Experiment, BIOMEX (de Vera et al., 2012). Its
203 resistance was tested in terms of survival and DNA damage in response to different types of
204 space-relevant radiation (i.e. UV) and sparsely (up to 117 kGy) and densely ionizing radiation
205 (up to 1000 Gy). The fungus showed considerable resistance to all the conditions tested,
206 remaining viable up to 55.81 kGy (Pacelli et al., 2017b).

207 John E. Hallsworth's talk, 'A story of glycerol', detailed the interventions that this polyol
208 can make in the cellular biology and ecology of microbes, such as enhancement of biological
209 control and reduction of temperature and water-activity minima for growth (Hallsworth and
210 Magan, 1995); mechanisms by which glycerol exerts these activities including its ability to
211 enhance macromolecular flexibility (entropically increased disorder) at high concentrations; a
212 method to quantify this entropic activity of solutes (chaotropicity) (Cray et al., 2013); and a long-
213 term research endeavor to try to demonstrate that microbes can retain metabolic activity and
214 maintain growth below the limit for life recognized since the 1960s (i.e. 0.605 water activity)
215 (Stevenson et al., 2015a). This culminated in the discovery that glycerol can facilitate
216 differentiation and germination of xerophilic fungi, most notably *Aspergillus penicillioides*,
217 down to 0.585 water activity (Stevenson et al., 2017). He finished the talk by posing a series of
218 intriguing questions e.g.: 1) does glycerol determine the extent of and failure points for the
219 functional biosphere, and 2) does abiotic glycerol influence habitability of hostile environments
220 (both terrestrial & extraterrestrial)?

221 **2. Stress mechanisms and responses in fungi: molecular biology, biochemistry, biophysics,** 222 **and cellular biology**

223 Certain fungal species such as *Saccharomyces cerevisiae*, *A. nidulans*, and *Neurospora*
224 *crassa* have been used by the scientific community for decades as effective eukaryotic models.

225 Many genetic, molecular, cell biology, biochemical, and biophysical research tools and
226 techniques have been developed and perfected using these model systems. The value of the tools
227 and techniques is evidenced by research that addresses questions about the fundamental
228 processes which drive fungal stress and responses including the perception of the stress, signal
229 transduction, and cellular responses to fungal stresses (Brown and Goldman, 2016; de Nadal and
230 Posas, 2015; Ho and Gasch, 2015; Rangel et al., 2015b).

231 Gustavo H. Goldman presented studies about regulation of *Aspergillus nidulans* CreA-
232 mediated catabolite repression by Fbx23 and Fbx47, which are F-box subunits of the Skp, Cullin,
233 F-box containing (SCF) ubiquitin ligase complex. Carbon catabolite repression (CCR) is a
234 process that selects the energetically most-favorable carbon source in an environment, by
235 suppressing the use of less-favorable carbon sources when a better one is available (Brown et al.,
236 2014). Glucose is the preferential carbon source for most microorganisms because it is rapidly
237 metabolized, generating quick energy for growth. In the filamentous fungus *Aspergillus*
238 *nidulans*, CCR is mediated by the transcription factor CreA, a C₂H₂ finger domain DNA-
239 binding protein (Ries et al., 2016). The aim of his work was to investigate the regulation of
240 CreA. CreA depends in part on *de novo* protein synthesis and is regulated in part by
241 ubiquitination. CreC, the scaffold protein in the CreB-CreC deubiquitination (DUB) complex, is
242 essential for CreA function and stability. Goldman's research group screened a collection of null
243 mutations for F-box encoding genes and identified two of them as important for carbon
244 catabolite repression and derepression. Immunoprecipitation of one of them revealed several
245 potential targets involved in CreA regulation.

246 Maria Celia Bertolini focused on the *Neurospora crassa* RUV-1 protein, which is identified
247 as a protein involved in heat stress response (Freitas et al., 2008). This protein, together with its

248 paralogue RUV-2, belongs to the AAA+ ATPase protein family and is annotated as an ATP-
249 dependent DNA helicase. The proteins have been identified as components of several
250 macromolecular complexes, implicated in many cellular processes in different organisms. In *N.*
251 *crassa*, the *ruv-1* transcript and RUV-1 protein are up-regulated under heat stress; however, *ruv-*
252 *2* transcript is not regulated under the same condition. In addition, cellular localization analyses
253 showed that both proteins move to the nucleus under heat stress.

254 **3. Fungal photobiology in the context of stress**

255 The entire second day of the Symposium focused on fungal photobiology. Fungi respond to
256 light as environmental signals that modulate several aspects of their biology, including
257 development and metabolism. However, excess light causes biological stress and most fungi
258 respond by synthesizing protective pigments and enzymes for repairing UV-induced DNA
259 damage (Braga et al., 2015; Braga et al., 2006; Fischer et al., 2016; Idnurm et al., 2010; Rangel
260 et al., 2006b; Rangel et al., 2011).

261 Luis M. Corrochano began the morning explaining how light is the ultimate source of
262 energy for life. However, light is both a signal from the environment and a damaging agent for
263 all organisms. Most fungi respond to light by regulating gene transcription, and a key response to
264 light is the activation of genes for repairing UV-induced DNA damage. In *Phycomyces*, a
265 cryptochrome seems to act as a blue-light regulated DNA repair enzyme (Tagua et al., 2015).

266 Gerhard Braus stated that light represents a stress signal in fungi and induces different
267 reactions. *A. nidulans* develops in the soil in the absence of light, primarily in closed sexual
268 fruiting bodies linked to a specific secondary metabolism as overwintering structures. In contrast,
269 light promotes and accelerates the formation of conidiophores, which release asexual spores into
270 the air. Various control layers coordinate fungal development, virulence, and secondary

271 metabolism. They include the control of transcription and histone modification, signal perception
272 and transduction as well as protein localization and stability (Bayram and Braus, 2012; Sarikaya-
273 Bayram et al., 2014; Sarikaya-Bayram et al., 2015).

274 Monika Schmolz's presentation was about *Trichoderma reesei*, an important producer of
275 plant cell-wall degrading enzymes and heterologous proteins. Therefore, it is of utmost
276 importance to understand the factors influencing the regulation cascade that lead to high
277 efficiency production of enzymes - particularly the previously unconsidered effect of light
278 (Stappler et al., 2016). Regulation of cellulase gene expression is connected with regulation of
279 secondary metabolism in *T. reesei* with differences in light and darkness (Monroy et al., 2017).
280 Screening for cellulose-sensing receptors permitted identification of two G-protein coupled
281 receptors (GPCRs). These GPCRs are essential for chemotropical sensing of the building block
282 glucose and morphological changes on natural substrate surfaces. Additionally, these receptors
283 act as checkpoints for posttranscriptional up-regulation of secreted cellulose degrading enzymes
284 on cellulose and lactose (Stappler et al., 2017). Analysis of the photoreceptor ENV1 revealed an
285 evolutionarily conserved mechanism to integrate stress responses with light response in the
286 Hypocreales (Lokhandwala et al., 2015). This finding highlights the importance of stress
287 responses in diverse interconnected regulatory processes in *T. reesei*, such as light-dependent
288 regulation, enzyme expression, metabolite production, and chemical communication in nature.

289 Luis Larrondo showed how circadian clocks are molecular devices that allow organisms to
290 anticipate daily cyclic challenges by temporally modulating different processes. Using clock-null
291 mutants of *Botrytis cinerea*, Larrondo's group found that interaction between this
292 phytopathogenic fungus and its host varies with the time of day (Canessa et al., 2013; Hevia et
293 al., 2016; Hevia et al., 2015). In *Neurospora*, the FREQUENCY protein (FRQ) is the main

294 component of the circadian oscillator (Ruoff et al., 2005), a role that is also conserved for the
295 *Botrytis* ortholog BcFRQ1. This protein also appears to play a critical function in asexual/sexual
296 decisions. Nevertheless, developmental phenotypes triggered by the absence of FRQ can be
297 reversed by nutritional cues.

298 Kevin Fuller explained that for the saprophyte and opportunistic pathogen *Aspergillus*
299 *fumigatus*, visible light leads to conspicuous effects on colonial growth, e.g. the induction of
300 mycelial pigments or asexual spores, as well as an induction of genes involved in DNA repair
301 (Fuller et al., 2016; Fuller et al., 2013). Transcriptome analysis revealed that in *A. fumigatus*
302 most regulated genes are repressed by light, including those involved in oxidative
303 phosphorylation, ergosterol biosynthesis, and metal ion homeostasis. The biological significance
304 of these light-repressed categories is more difficult to discern, but likely reflects a difference
305 between metabolic conditions the fungus faces at the soil surface and deeper in the soil/compost.
306 Interestingly, there is a correspondence between genes that are induced under hypoxia (Barker et
307 al., 2012) and those repressed by light, and so the current model is proposed: at the sub-surface
308 (dark), the fungus experiences low-oxygen concentrations and genes involved in hypoxia
309 adaptation are up-regulated by the conserved regulator SrbA (Willger et al., 2008); at the soil
310 surface, where oxygen levels are ambient, photoreceptors (LreA, FphA) down-regulate those
311 hypoxia-adaptive pathways, including ergosterol metabolism and iron homeostasis. As the
312 surface is also a site for optimal dispersal (i.e. open air) and exposure to ultraviolet radiation,
313 genes involved in sporulation and resistance to genotoxic damage are induced by light. Fuller
314 and colleagues are currently dissecting the interplay between canonical light and hypoxia-
315 regulatory pathways as well as probing the conservation of ergosterol biosynthesis and drug
316 sensitivity by light in other fungal pathogens.

317 Gilberto U. L. Braga explained that antimicrobial photodynamic treatment (APDT) is a
318 promising alternative to conventional antifungal agents that can be used to kill fungi, which
319 cause diseases in animals or plants (de Menezes et al., 2014; Gonzales et al., 2017). APDT, using
320 phenothiazinium photosensitizers, efficiently kills planktonic cells of *Candida* species and
321 conidia of several pathogenic fungi, damages the fungal plasma membrane increasing its
322 permeability and greatly impacting their proteomes (Brancini et al., 2016).

323 Drauzio E. N. Rangel completed the day by discussing how fungi illuminated during
324 mycelial growth produce conidia with increased stress tolerance. Light is an important stimulus
325 for many fungi and it has been shown to induce production of *Metarhizium robertsii* conidia with
326 increased stress tolerance (Rangel et al., 2015c; Rangel et al., 2011). White light, as well as blue
327 light during mycelial growth, induces higher conidial stress tolerance, higher germination rates,
328 and higher virulence in *Metarhizium robertsii*, but nutritional stress always produces conidia
329 with more intense stress tolerance and virulence than conidia produced under white or blue light
330 (Rangel, 2011; Rangel et al., 2008; Rangel et al., 2006a; Rangel et al., 2012).

331 **4. Role of stress in fungal pathogenesis**

332 Fungal pathogens have evolved numerous mechanisms to escape host defenses such as
333 thermotolerance, toxin production, masking or modulating pathogen-associated molecular
334 patterns (PAMPs) and pattern recognition receptors (PRRs), and overcoming oxidative defenses
335 (Sales-Campos et al., 2013; Stappers and Brown, 2017). Research on fungal pathogenesis is a
336 wide field that encompasses basic research on host-pathogen interactions, cell and molecular
337 biology, and development and aging, as well as applied research in crop protection, food
338 security, public health, and medicine.

339 Jon Y. Takemoto addressed global challenges for crop production and food security and the
340 critical roles for research into fungal stress and biology (Fisher et al., 2012). He described
341 strategies behind the recent discovery of a new generation of aminoglycoside fungicides aimed
342 to help counter the critical shortage of effective, safe, and environmentally friendly fungicides
343 against crop diseases. K20 is a new membrane-targeting amphiphilic aminoglycoside that is not
344 toxic and a broad-spectrum antifungal, which can be produced at scalable, kilogram levels
345 (Chang and Takemoto, 2014). K20 by itself, or in combination with current crop fungicides
346 (employed at lower than recommended rates), shows promise in combating several crop diseases
347 including the devastating wheat disease, Fusarium Head Blight.

348 Célia M. A. Soares discussed metabolic changes in *Paracoccidioides* spp. during human
349 host infection. Members of the *Paracoccidioides* complex, the etiologic agents of
350 paracoccidioidomycosis, cause disease in healthy and immunocompromised patients in Latin
351 America. Her team developed a method to harvest *Paracoccidioides brasiliensis* yeast cells from
352 infected murine lung to facilitate *in vivo* transcriptional and proteomic profiling (Lacerda Pigosso
353 et al., 2017). They compared the *in vivo* to *in vitro* and *ex vivo* responses of *Paracoccidioides*
354 spp., as obtained by proteomic analysis (Lima et al., 2014; Parente-Rocha et al., 2015).

355 Iran Malavazi described the contribution of the cell wall integrity pathway to virulence in
356 *Aspergillus fumigatus*. He showed that besides its role in cell wall reinforcement and remodeling,
357 the cell wall integrity pathway (Rocha et al., 2015) is an important hub for production of fungal
358 secondary metabolites. The fumiquinazoline (Fq) production is regulated by the transcription
359 factor RlmA and the MAP kinase MpkA. In fact, the RlmA transcription factor binds to the
360 promoter region of most of the genes of the Fq cluster genes. The results indicate an

361 unprecedented connection of the CWI pathway with the biosynthesis of a conidia-born secondary
362 metabolite.

363 Marcia R. von Zeska Kress related her research about antimicrobial photodynamic
364 inactivation and photodynamic therapy. *Neoscytalidium* spp. and *Fusarium* spp. are filamentous
365 fungi widely distributed in nature that cause non-dermatophyte onychomycosis that have
366 significant resistance to commercial antifungal therapy. APDT with the phenothiazinium
367 photosensitizers methylene blue, toluidine blue, new methylene blue, and the pentacyclic
368 phenothiazinium S137 were able to kill both quiescent and germinated arthroconidia and
369 microconidia of *Neoscytalidium* spp. and *Fusarium* spp., respectively. The photodynamic
370 therapy with phenothiazinium photosensitizers on *Fusarium moniliforme* infection of *Galleria*
371 *mellonella*, the model for fungal virulence and susceptibility testing, showed that this therapy is a
372 promising alternative to antifungal treatment against this filamentous fungus (de Menezes et al.,
373 2016; Tonani et al., 2017).

374 Alexandre M. Bailão explained that the black fungi *Fonsecaea pedrosoi* and
375 *Cladophialophora carrionii* are the most common agents of Chromoblastomycosis, a
376 subcutaneous mycosis frequently diagnosed in tropical regions. The virulence strategies used by
377 these fungi are poorly understood. As iron is an essential element, pathogenic fungi have
378 developed molecular mechanisms to obtain the metal during infection. *F. pedrosoi* and *C.*
379 *carrionii* have genes encoding for reductive and siderophore-mediated iron-uptake systems, and
380 the transcriptional levels of those genes are induced upon iron limitation. Moreover, these
381 pathogens produce ferricrocin as intra- and extracellular siderophores (Silva-Bailão et al., 2017).

382 **5. Fungal stress and bioremediation**

383 Fungi are ubiquitous in polluted habitats, and they exhibit remarkable tolerance to organic

384 and inorganic contaminants. Some fungi synthesize specific metal-binding peptides or
385 metallothioneins in response to metal pollutants. Fungi also exhibit morphological differentiation
386 in response to toxic stress, such as the formation of hyphal aggregates and cords, melanized cell
387 forms as well as thigmotropism and chemotropism to locate a favorable microenvironment.
388 Metabolic versatility underpins enzyme expression, carbon metabolism, pollutant transport, and
389 production and excretion of metabolites that immobilize oxalates, oxides, phosphates, and
390 carbonates (Gadd, 2007, 2010; Gadd, 2016). The morphological and metabolic versatility of
391 fungi provides several advantages for bioremediation approaches, not the least their capacity to
392 combat and overcome stress in adverse environments.

393 Rosane M. Peralta was the only speaker about using fungi for bioremediation. The
394 capability of white rot fungi to biodegrade several recalcitrant pollutants has generated a
395 considerable research interest in this area of industrial/environmental microbiology. The ability
396 of white rot fungi to degrade pollutants appears to be related to the capability of producing
397 extracellular non-specific lignin-degrading enzymes, especially peroxidases and laccases, as well
398 as to produce intracellular oxidases generating of H₂O₂ and cytochrome P450 (Coelho-Moreira et
399 al., 2013; Maciel et al., 2013). WRF can be an alternative to reduce the ecological problems
400 caused by the accumulation of these products in nature.

401 **6. Fungal stress in agriculture and forestry**

402 Stress conditions, particularly UV radiation and heat from sunlight are important regulators
403 of fungal communities in agriculture and forest (Bidochka et al., 2001; Ferreira et al., 2017;
404 Wang and Wang, 2017). Many fungi have been developed into commercial biological control
405 agents and are being mass produced to be used in agriculture and forestry (Li et al., 2010) to
406 promote plant growth (Vega et al., 2009), promote plant defense responses (Vega et al., 2009);

407 and to control plant diseases (Costa et al., 2013; Druzhinina et al., 2011), plant parasitic
408 nematodes (Siddiqui and Mahmood, 1996), aquatic weeds (Cother and Gilbert, 1994), terrestrial
409 weeds (Moraes et al., 2014), and insects (Alston et al., 2005; Faria and Wraight, 2007; Keyser et
410 al., 2017). Different abiotic environmental factors cause stress and consequently harm these
411 important fungi in agricultural systems, highlighting the need to study stress tolerance of fungi
412 used in agriculture and forestry.

413 Elias Hakalehto's presentation was about competitive interactions between fungi and other
414 microbes in stressed ecosystems. In mixed cultures, fast-growing Gram-positive bacteria play a
415 dominant role. For example, the fungi that pioneer in soil do not compete with bacteria as much
416 for the degradation products as they spread their influence on novel sources of organic raw
417 materials. The bacterial strains, in turn, receive an advantage of the biodegradation by fungi.
418 Therefore, the instances of confrontation between fungi and bacteria are limited to specific
419 conditions only. The effects of the industrially upgraded fungal enzymes have been tested in
420 biorefinery trials for producing carbon and energy sources for undefined mixed cultures fortified
421 with some industrial bacterial strains under controllable gas flow (den Boer et al., 2016; Schwede
422 et al., 2017). The fungi colonize hostile or poor environments with the help of their aerial
423 mycelium; transport nutrients along the growing hyphae; change the surroundings by enzymatic
424 activities; and mobilize nutrients from various sources and niches. Fungi also produce sexual and
425 asexual spores to conquer new areas, and nutrient sources (fungal spores fill the atmosphere).
426 There is evidence about the relatively even distribution of the spores in the layers of the
427 atmosphere detected from samples collected by a jet at altitudes of 300-1000 m (Hakalehto,
428 2015).

429 Roger D. Finlay discussed ways in which root-associated fungi mediate stress responses of

430 plants. Symbiotic mycorrhizal fungi play important roles in reducing abiotic and biotic stress to
431 plants in forestry and agriculture, and can minimize negative effects of soil acidification, Al-
432 toxicity, and base cation leaching (Finlay, 2008). High-throughput DNA sequencing and stable
433 isotope-based studies suggest that ectomycorrhizal fungi may play an important role in
434 fractionation of Mg isotopes and uptake of base cations through weathering of minerals (Fahad et
435 al., 2016). These fungi also influence patterns of stable carbon storage in humified material and
436 secondary minerals. Moreover, these fungi may have global impacts on sequestration or release
437 of atmospheric CO₂ (Clemmensen et al., 2013; Finlay and Clemmensen, 2017).

438 Chengshu Wang elucidated the cause-effect relationships between oxidative stress and
439 fungal culture degeneration. Filamentous fungi undergo frequent culture degeneration during
440 successive maintenance on artificial media by showing fluffy mycelium growth and colony
441 sectorization (Butt et al., 2006). The degenerate fungal cultures show the loss or reduced abilities
442 to sporulate, perform sexual cycle, fruit, and/or produce secondary metabolites. Molecular and
443 biochemical characterizations reveal that fungal culture degeneration is a sign of cell aging
444 (Wang et al., 2005), and the occurrence of spontaneous oxidative stress, i.e., cellular
445 accumulation of reactive oxygen species, is connected with mitochondrial dysfunction and
446 thereby fungal culture degeneration (Li et al., 2014; Li et al., 2008; Xiong et al., 2013).

447 Octavio Loera explained how sublethal oxidant states improve production and quality of
448 conidia in entomopathogenic fungi (EF). EF conidia control insect plagues in crop fields where
449 abiotic factors weaken conidia. During production of conidia by *Metarhizium*, *Beauveria*, and
450 *Isaria*, controlled oxidant stress improves the conidial yields, and production of stronger and
451 more infective conidia is possible (Muñiz-Paredes et al., 2017). This implies metabolic
452 adjustments leading to cross protection mechanisms, which could be feasible in large-scale

453 processes (Miranda-Hernández et al., 2016).

454 Nicolás Pedrini studies molecular interactions between EF and insects. During insect-
455 cuticle degradation as well as during invasion and proliferation throughout their host, EF secrete
456 a suite of enzymes and secondary metabolites that help them cope with the stressful situation
457 they have to endure to finally achieve a successful infection (Huarte-Bonnet et al., 2017a;
458 Pedrini, 2017; Wang and Wang, 2017). Moreover, insects trigger innate immune reactions to
459 prevent microbial proliferation. By analysis of available transcriptomic and metabolomic data,
460 several components and mechanisms involved in this fungi-insect interaction were reviewed.

461 Everton K. K. Fernandes presented a study about stress tolerance of EF conidia and
462 blastospores. The study compared the tolerance to heat (45 °C) and UV-B radiation between
463 conidia and blastospores of *Metarhizium* spp. and *Beauveria bassiana* s.l. He discussed the
464 principles for this comparison concerning their use for biological control of arthropods.

465 **7: Fungal stress in industry**

466 Many industrial processes essential for meeting societal needs use fungi that play central
467 roles in those processes. Examples are industrial ethanol production using yeast strains and the
468 brewery industry utilizing *Saccharomyces pastorianus*. Research into the biology of fungi within
469 industrial systems have catalyzed research into other areas such as eukaryotic responses to
470 oxidative stress and effects on aging (Wei et al., 2007; Zhao and Bai, 2009).

471 Elis C. A. Eleutherio explained how fungi can be used as models to study oxidative stress
472 (da Silva et al., 2012; Fernandes et al., 2007). Evidence shows that oxidative stress is connected
473 to life span (Mannarino et al., 2008). Throughout the world, leading causes of death are age-
474 related diseases, such as cancer and neurodegenerative diseases. Her presentation highlighted the

475 value of the yeast *S. cerevisiae* as a model to investigate the oxidative stress response and its
476 potential impact on aging and age-related diseases (Brasil et al., 2013; França et al., 2017).

477 Diego Bonatto's topic was the delicate balance between hybrid genomes and brewery
478 stresses in lager yeasts. *Saccharomyces pastorianus* has been employed in the brewery industry
479 for the fermentation of lager beers, a product consumed worldwide. Despite its industrial
480 importance, little is known about how *S. pastorianus* deals with brewery stress. His group has
481 evaluated the major stress-associated biological mechanisms using transcriptome and systems
482 biology analyses. [ref](#)

483 Anderson Ferreira da Cunha finished the Symposium with his study of thermotolerance and
484 ethanol-resistance in *Saccharomyces cerevisiae* strains. Several stress factors are involved in the
485 efficiency of production during ethanol fermentation. High ethanol concentrations and high
486 temperatures are the most relevant ones. A good industrial strain must be sufficiently robust to
487 respond well to this environmental stress, without altering its fermentative characteristics during
488 the whole crop season. Since 2010, his group has been sampling different fermentation tanks
489 trying to find yeasts able to grow at temperatures above 40 °C and at concentrations of ethanol
490 above 12%. They have isolated four different thermotolerant and two ethanol-tolerant strains,
491 which produced superior ethanol yield than strains currently used in ethanol plants. These strains
492 showed a high potential of direct application for ethanol production.

493

494 **Elsevier Student Competition Awards**

495 Elsevier sponsored awards to recognize excellent work by graduate students in the area of
496 fungal stress. For the award competitions, students submitted original research articles formatted
497 for journal publication. Two of these articles are featured in this special issue (Huarte-Bonnet et

498 al., 2017b; Muniz et al., 2017). The award winners also gave oral presentations at ISFUS. The
499 articles were read and judged by several of the international speakers following the evaluation
500 criteria found in the website <https://isfus.wordpress.com/>.

501 Four doctoral students received silver Elsevier awards: Mariane Paludetti Zubieta (PhD
502 student in Functional and Molecular Biology at the Universidade Estadual de Campinas). Her
503 article was titled: “Understanding the production of recombinant proteins in *Aspergillus nidulans*
504 by global proteome profiling” (Figure 6). Ronaldo A. Pereira-Junior (PhD student in Tropical
505 Medicine and Public Health, Universidade Federal de Goiás), and his article was titled:
506 “Riboflavin: A supplement that increases the tolerance of *Metarhizium* species against UV-B
507 radiation by over-expressing photolyase, laccase, and polyketide synthase genes” (Figure 7 and
508 10). Carla Huarte-Bonnet (PhD student in Biotechnology and Molecular Biology, Universidad
509 Nacional de La Plata) from Argentina was the only non-Brazilian award winner. Her article was
510 titled: “Alkane-grown *Beauveria bassiana*: mycelial pellets formation, oxidative stress induction,
511 and cell surface alterations” (Figure 8 and 10) (Huarte-Bonnet et al., 2017b). Elen Regozino
512 Muniz (PhD student in Tropical Medicine and Public Health at the Universidade Federal de
513 Goiás). Her article was titled: “Impact of short-term temperature challenges on the larvicidal
514 activities of the entomopathogenic watermold *Leptolegnia chapmanii* against *Aedes aegyptii*, and
515 development on infected dead larvae” (Figure 9 and 10) (Muniz et al., 2017).

516 The weekend after the ISFUS-2017 about half of the international speakers, several
517 Brazilian speakers, and a few international participants took an excursion to Pirenópolis, in the
518 state of Goiás. This historic town, about 130 km from Goiânia, attracts many tourists due to its
519 richly preserved history and surrounding mountains. On a guided tour to Pireneus State Park, we
520 experienced the beautiful waterfalls and then hiked up a hill to view the magnificent sunset over

521 the Pico dos Pireneus. The following day was spent at Vagafogo Wildlife Sanctuary, walking
522 through the riparian forest and tasting a large variety of local foods for brunch. We also enjoyed
523 the history and cuisine of Pirenópolis. The excursion was an excellent opportunity to see more of
524 Brazil and become better acquainted with our colleagues.

525 **Conclusion**

526 This special issue of *Fungal Biology* was inspired by the International Symposium on
527 Fungal Stress. Most of the articles were written by speakers or participants of ISFUS-2017.
528 Individuals who find the subject of fungal stress fascinating and attractive are invited to consider
529 attending the next ISFUS, tentatively planned for 2019.

530 **Acknowledgments**

531 This work was supported by grants from the São Paulo Research Foundation (FAPESP)
532 of Brazil 2010/06374-1, 2013/50518-6, and 2014/01229-4 for D.E.N.R, and to the Brazilian
533 National Council for Scientific and Technological Development (CNPq) PQ2 302312/2011-0,
534 and PQ1D 308436/2014-8 The work was also facilitated by grants in support of the International
535 Symposium on Fungal Stress (ISFUS)-2017 meeting from the Coordenação de Aperfeiçoamento
536 de Pessoal de Nível Superior (CAPES) of Brazil - PAEP 88881.123209/2016-01 and by a grant
537 from the Fundação de Amparo à Pesquisa do Estado de Goiás of Brazil - 201710267000110. We
538 are also thankful to Sheba Agarwal-Jans, Microbiology & Mycology Publisher, at Elsevier by
539 providing the Student Awards, and to Becky Long, Journal Manager, ELSEVIER Global Journal
540 Production for the editorial assistance of this special issue.

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1012 FIGURE LEGENDS

1013 Figure 1. Logo of the second International Symposium on Fungal Stress (ISFUS-2017) that was
1014 hosted at the Universidade Federal de Goiás in Goiânia, GO, Brazil. This figure shows the some
1015 of the stress parameters that fungi are subjected to such as ionizing radiation, acidic and alkaline
1016 environments, hypoxic or anoxic conditions, poisons in general such as genotoxic and oxidative
1017 products, UV radiation from Sun, pollution from industry and agriculture, salt stress, nutritive
1018 stress, and heat from solar radiation and other sources.
1019

1020 Figure 2. Speakers of the second International Symposium on Fungal Stress – ISFUS-2017.
1021 Standing from left to right: Elias Hakalehto (Finland), Jon Y. Takemoto (USA), Maria Celia
1022 Bertolini (Brazil), John E. Hallsworth (UK), Kevin K. Fuller (USA), Rosane M. Peralta (Brazil),
1023 Luis M. Corrochano (Spain), Luis Larrondo (Chile), Roger D. Finlay (Sweden), Laura
1024 Selbmann (Italy), Monika Schmoll (Austria), Célia M. A. Soares (Brazil), Elis Eleutherio
1025 (Brazil), Ekaterina Dadachova (Canada), Gerhard Braus (Germany), Iran Malavazi (Brazil),
1026 Gustavo Goldman (Brazil), Diego Bonatto (Brazil), Anderson F. da Cunha (Brazil), and
1027 Chengshu Wang (China). Standing from left to right: Drauzio E. N. Rangel (Brazil), Amanda
1028 Rangel (daughter of the organizers), Alene Alder-Rangel (Brazil), Gilberto U. L. Braga (Brazil),
1029 Octavio Loera (Mexico), Alexandre M. Bailão (Brazil), and Marcia R. Z. Kress (Brazil).
1030

1031 Figure 3. Speakers of the second International Symposium on Fungal Stress – ISFUS. From left
1032 to right in the top row: Kevin Fuller (USA), Gustavo Goldman (Brazil), Chengshu Wang
1033 (China), Gerhard Braus (Germany), Rosane M. Peralta (Brazil), Iran Malavazi in the back
1034 (Brazil), Elis Eleutherio (Brazil), Célia M. A. Soares in the back (Brazil), Jon Y. Takemoto
1035 (USA), Maria Celia Bertolini (Brazil), Marcia R. Z. Kress in the back, not visible (Brazil).
1036 Monika Schmoll (Austria), John E. Hallsworth in the back (UK), Elias Hakalehto in the back
1037 (Finland), Luis Larrondo (Chile), Luis M. Corrochano (Spain), Roger Finlay (Sweden), Laura
1038 Selbmann (Italy), Anderson F. da Cunha in the back (Brazil), Ekaterina Dadachova (Canada),
1039 Diego Bonatto (Brazil), and Alexandre M. Bailão (Brazil). From left to right in the lower row:
1040 Drauzio E. N. Rangel (Brazil), Gilberto U. L. Braga (Brazil), and Octavio Loera (Mexico).
1041

1042 Figure 4. Opening Ceremony: Flávia Aparecida de Oliveira, the director of IPTSP (the host
1043 institution) welcoming researchers and students.
1044

1045 Figure 5. Opening Ceremony: Alene Alder-Rangel, John E. Hallsworth, Luis M. Corrochano,
1046 Maria Zaira Turchi, João Teodoro Pádua, Flávia Aparecida de Oliveira, and Drauzio E. N.
1047 Rangel.
1048

1049 Figure 6. Elsevier Award given to Mariane Paludetti Zubieta, presented by Drauzio E. N. Rangel
1050 (left) and Luis M. Corrochano (right).
1051

1052 Figure 7. Elsevier Award given to Ronaldo A. Pereira-Junior, presented by Drauzio E. N. Rangel
1053 (left) and Luis M. Corrochano (right).
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1055 Figure 8. Elsevier Award given to Carla Huarte-Bonnet, presented by Drauzio E. N. Rangel (left)
1056 and Luis M. Corrochano (right).

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1058 Figure 9. Elsevier Award given to Elen Regozino Muniz, presented by Drauzio E.N. Rangel
1059 (left) and Luis M. Corrochano (right).

1060
1061 Figure 10. Elsevier Award winners Ronaldo A. Pereira-Junior, Carla Huarte-Bonnet, and Elen
1062 Regozino Muniz. From left to right: Alene Alder-Rangel, Dr. João Teodoro Pádua (from the
1063 office of the University President), Flávia Aparecida de Oliveira (Director of IPTSP
1064 Universidade Federal de Goiás), Elen Regozino Muniz (PhD student), Drauzio E. N. Rangel
1065 (professor Universidade Federal de Goiás), Carla Huarte-Bonnet (PhD student), Luis M.
1066 Corrochano (professor Universidad de Sevilla), and Ronaldo A. Pereira-Junior (PhD student).