

The selective antifungal activity of *Drosophila melanogaster* metchnikowin reflects the species-dependent inhibition of succinate–coenzyme Q reductase

Mohammad-Reza Bolouri Moghaddam^{1,2}, Thomas Gross³, Annette Becker³, Andreas Vilcinskas^{1,2,*}, Mohammad Rahnamaeian^{2,*}

¹ Institute for Insect Biotechnology, Justus Liebig University of Giessen, Heinrich-Buff-Ring 26-32, D-35392 Giessen, Germany

² Fraunhofer Institute for Molecular Biology and Applied Ecology, Department of Bioresources, Winchester Strasse 2, D-35394 Giessen, Germany

³ Institute of Botany, Justus Liebig University of Giessen, Heinrich-Buff-Ring 38, D-35392 Giessen, Germany

* Shared senior authorship; correspondence may be addressed to mohammad.rahnamaeian@uni-giessen.de or andreas.vilcinskas@agrار.uni-giessen.de

Email addresses:

Mohammad Reza Bolouri Moghaddam: mohammad.r.bolouri-moghaddam@bio.uni-giessen.de

Thomas Gross: thomas.gross@bio.uni-giessen.de

Annette Becker: annette.becker@bot1.bio.uni-giessen.de

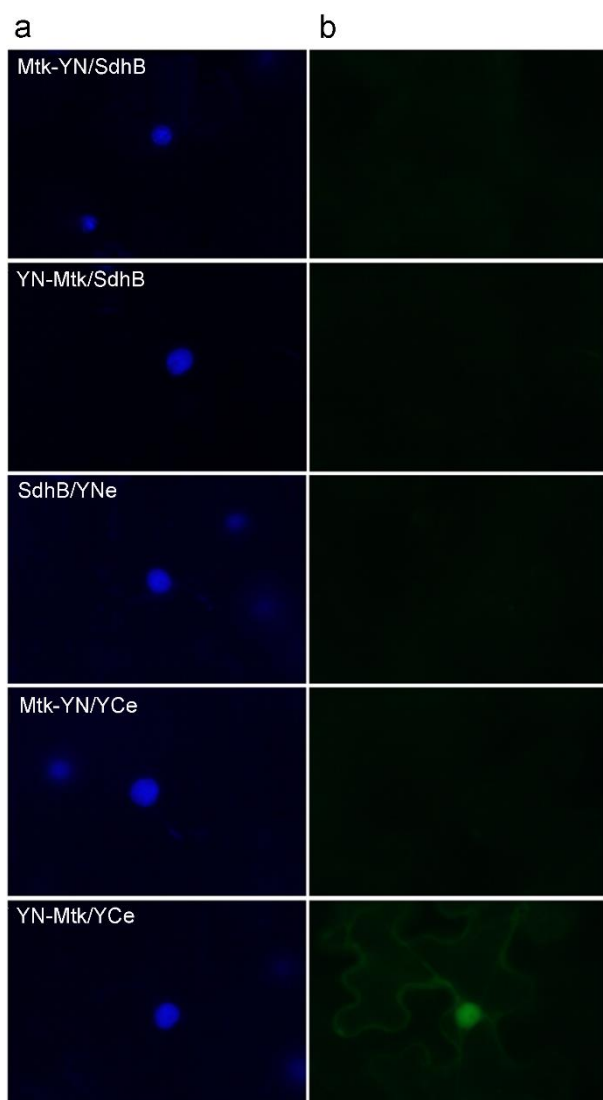
Andreas Vilcinskas: andreas.vilcinskas@agrار.uni-giessen.de

Mohammad Rahnamaeian: mohammad.rahnamaeian@uni-giessen.de

Fig. S1. Testing the interaction between Mtk and SdhB in tobacco cells.

The SdhB fusion constructs showed no YFP fluorescence in the presence of YFP_{Cempty} (YCe) or YFP_{Nempty} (YNe). However, the Mtk constructs exhibited YFP fluorescence in both the cytoplasm and nucleus in the presence of the corresponding YFP construct without a fusion. The co-expression of Mtk and SdhB did not result in detectable YFP fluorescence, indicating that the BiFC assay was not suitable for the verification of interactions between Mtk and SdhB.

(a) DAPI channel; (b) GFP channel.



List of UniProtKB accession numbers of SdhB sequences in fungi for phylogenetic analysis.

Ascomycota:

Q2TWM0 *Aspergillus oryzae*; Q5BAU8 *Emericella nidulans*; Q0UY13 *Phaeosphaeria nodor*;
A3M027 *Scheffersomyces stipites*; I1RNM7 *Gibberella zeae* (*Fusarium graminearum*);
G8ZYS0 *Torulasporea delbrueckii*; A0A0F8BTS8 *Ceratocystis fimbriata* f. sp. *platani*;
A0A0E9NA54 *Saitoella complicate*; J3P9K0 *Gaeumannomyces graminis* var. *tritici*; A5E0Y7
Lodderomyces elongisporus; F7VKJ3 *Sordaria macrospora*; A1DEE7 *Neosartorya fischeri*;
C4JPG2 *Uncinocarpus reesii*; G2Q420 *Myceliophthora thermophile*; G8JXV6 *Eremothecium*
cymbalariae; A5DJ38 *Meyerozyma guilliermondii*; B2AXV1 *Podosporea anserine*; D5G7W1
Tuber melanosporum; G2RFL3 *Thielavia terrestris*; C4Y771 *Clavispora lusitaniae*; K3VJU5
Fusarium pseudograminearum; G2X2B4 *Verticillium dahlia*; K1WHZ3 *Marssonina brunnea*
f. sp. *multigermtubi*; R8BC23 *Togninia minima*; M7T7N1 *Eutypa lata*; G0SBU7 *Chaetomium*
thermophilum; M2MY09 *Baudoinia compniacensis*; R1GBW3 *Botryosphaeria parva*; G1X793
Arthrotritys oligospora; C5FS39 *Arthroderma otae*; E4ZZI8 *Leptosphaeria maculans*; M2SI37
Cochliobolus sativus; F0XF17 *Grosmannia clavigera*; G0V5W8 *Naumovozya castellii*;
M3D0M2 *Sphaerulina musiva*; E3Q6P3 *Colletotrichum graminicola*; S8C423 *Dactylellina*
haptotyla; W6ZHQ3 *Bipolaris oryzae*; R7YXR2 *Coniosporium apollinis*; G0WAJ7
Naumovozya dairenensis; K2SD71 *Macrophomina phaseolina*; R0I5M2 *Setosphaeria*
turcica; A0A084GGK4 *Scedosporium apiospermum*; W3XRT1 *Pestalotiopsis fici*;
A0A0V1Q513 *Debaryomyces fabryi*; M1W191 *Claviceps purpurea*; A0A0D2IAT0
Rhinochrysiella mackenziei; W9XQ08 *Capronia epimyces*; A0A0D2A4H2 *Verruconis*
gallopava; W2RQM1 *Cyphellophora europaea*; I2H497 *Tetrapisispora blattae*; H2B1Y0
Kazachstania Africana; R9XB73 *Ashbya aceri*; A0A0X8HT74 *Eremothecium sinicaudum*;
U4KU36 *Pyronema omphalodes*; V5FBB5 *Byssochlamys spectabilis*; M2WK03 *Dothistroma*

septosporum; S3C661 *Ophiostoma piceae*; A0A084QTY5 *Stachybotrys chlorohalonata*;
A0A0G2DXW8 *Phaeomoniella chlamydospora*; 0A0D1WTD3 *Exophiala Sideris*;
A0A0F7ZS57 *Hirsutella minnesotensis*; A0A0A1T5X1 *Torrubiella hemipterigena*;
A0A0P7BGU0 *Neonectria ditissima*; A0A086T0I5 *Acremonium chrysogenum*; W9C5Z8
Sclerotinia borealis; A0A0C3H6Q6 *Oidiodendron maius*; A0A0F9X6K7 *Trichoderma*
harzianum; A0A063C926 *Ustilaginoidea virens*; A0A136JFM4 *Microdochium bolleyi*;
A0A0G2I336 *Emmonsia crescens*; A0A139HZQ5 *Pseudocercospora musae*; W6MMZ3
Kuraishia capsulate; N1J5Q8 *Blumeria graminis* f. sp. *hordei*; A0A0F4ZJL3 *Thielaviopsis*
punctulata; Q6MVG9 *Neurospora crassa*; A0A0L1HS04 *Stemphylium lycopersici*;
A0A0B1PFX7 *Uncinula necator*; G8EI90 *Zymoseptoria tritici*; A0A1E3Q0M3 *Lipomyces*
starkeyi; A0A168CFB2 *Isaria fumosorosea*; A0A0D9MRU9 *Aspergillus flavus*; F8N0V7
Neurospora tetrasperma; A0A0A2VID1 *Beauveria bassiana*; A0A1J9QJH1 *Emergomyces*
pasteuriana; A0A139GWE5 *Mycosphaerella eumusae*; A0A0A8L5G6 *Kluyveromyces*
dobzhanskii; A0A060T220 *Blastobotrys adenivorans*; A0A0U5B1X1 *Passalora natrassii*;
A0A165K4D7 *Xylona heveae*; W9JVT9 *Fusarium oxysporum*; A0A194VQF5 *Valsa mali*;
A0A0G4KTR3 *Verticillium longisporum*; A7TNK6 *Vanderwaltozyma polyspora*;
A0A1E3PTT9 *Nadsonia fulvescens* var. *elongate*; S3CSY3 *Glarea lozoyensis*; A0A1E3NR25
Pichia membranifaciens; A0A1E3QSX5 *Babjeviella inositovora*; A0A125RKY8 *Pyrenophora*
teres f. *teres* A0A093VHK0 *Talaromyces marneffeii*; A0A1L7WEU8 *Phialocephala subalpina*;
A0A179FS63 *Pochonia chlamydosporia*; A0A0J9XIV1 *Geotrichum candidum*; R4XFE7
Taphrina deformans; L0PD02 *Pneumocystis jirovecii*; A0A1L9SUM9 *Penicillium zonata*;
A0A0J0ADI9 *Gibberella fujikuroi*; A0A177ADU6 *Pseudogymnoascus destructans*;
A0A1E4TJV0 *Tortispora caseinolytica*; A0A1E4RWU9 *Cyberlindnera jadinii*; T5BQ28
Blastomyces dermatitidis; A0A1E3P4I6 *Wickerhamomyces anomalus*; S4SLP3 *Monilinia*
fructicola; B6F0Y3 *Aspergillus oryzae*; A0A1G4JXZ6 *Lachancea mirantina*; A0A178BAK5
Stagonospora sp.; A0A151GIL0 *Drechmeria coniospora*; A0A117E0M6 *Aspergillus niger*;

N1NZC0 *Saccharomyces cerevisiae*; A0A0L0NE48 *Tolyposcladium ophioglossoides*;
A0A177BZZ3 *Paraphaeosphaeria sporulosa*; A0A1C1XRU7 *Diaporthe helianthi*;
A0A061HP54 *Blumeria graminis* f. sp. *tritici*; A0A167XWF0 *Aschersonia aleyrodis*;
A0A0M9VT85 *Escovopsis weberi*; A0A1A0HEA7 *Metschnikowia bicuspidata* var.
bicuspidata; A0A1B8AK78 *Fusarium poae*; A0A022WBT3 *Trichophyton rubrum*;
A0A167H468 *Cordyceps brongniartii*; A0A150V0F8 *Acidomyces richmondensis*;
A0A1J9QWG5 *Diplodia corticola*; A0A0C4DT89 *Magnaportheopsis poae*; A0A175VZ01
Madurella mycetomatis; A0A0H4UQS7 *Venturia inaequalis*; A0A1A7MP27 *Aureobasidium*
pullulans; G4W729 *Stagonosporopsis cucurbitacearum*; A0A0B4UGK3 *Alternaria alternate*;
E0D565 *Podosphaera xanthii*; A0A179I0W7 *Purpureocillium lilacinum*; A0A1D8NFY4
Yarrowia lipolytica; G8AE23 *Millerozyma farinose*; A0A0B7JM52 *Bionectria ochroleuca*;
A0A089WYW9 *Botryotinia fuckeliana*; E0D564 *Podosphaera xanthii*; A0A167F5K0
Sugiyamaella lignohabitans; A0A0B4FV24 *Metarhizium anisopliae*; A0A1E4SKL1 *Candida*
tanzawaensis; N4U420 *Fusarium oxysporum* f. sp. *cubense*; A0A178EBJ2 *Pyrenochaeta* sp.;
A0A0N0NLZ9 *Phialophora attae*; A0A179UJL1 *Ajellomyces dermatitidis*; A0A0F2M4G5
Sporothrix schenckii; A0A0K2QQ30 *Trichophyton mentagrophytes*; A0A1L0AWJ4
Hanseniaspora guilliermondii; A0A167V949 *Ascosphaera apis*; A0A1D2VLH6 *Ascoidea*
rubescens; A0A0J5PVK1 *Aspergillus fumigatus*; A0A1P8VHT0 *Magnaporthe oryzae*;
C0SFU8 *Paracoccidioides brasiliensis*; A0A024S1K3 *Hypocrea jecorina*; A0A178Z551
Fonsecaea erecta; A0A1B7SIZ0 *Ogataea polymorpha*; A0A1J7J773 *Coniochaeta ligniaria*;
A0A1E1MVM0 *Rhynchosporium secalis*; A0A1D5AKD3 *Didymella tanacetii*; A0A166LCX0
Colletotrichum incanum; A0A1Q5UJH8 *Penicillium subrubescens*; S5U054 *Alternaria solani*;
A0A1B2JGC3 *Komagataella pastoris*; A0A084RG07 *Stachybotrys chartarum*; A0A1E5RDC2
Hanseniaspora uvarum; D5MTG0 *Corynespora cassicola*; A0A1E4TQ48 *Pachysolen*
tannophilus; A0A0F4YFV6 *Rasamsonia emersonii*.

Basidiomycota:

P32420 *Ustilago maydis*; Q70KF8 *Uromyces fabae*; G7E3H2 *Mixia osmundae*; A0A067N939 *Pleurotus ostreatus*; A0A0F7S7G2 *Sporisorium scitamineum*; D8PXF2 *Schizophyllum commune*; F4RTE2 *Melampsora larici-populina*; K5WSH2 *Phanerochaete carnosae*; E2RZU1 *Helicobasidium mompa*; R9AD16 *Wallemia ichthyophaga*; S7PV57 *Gloeophyllum trabeum*; W4JQS4 *Heterobasidion irregular*; A0A061HAV5 *Anthracocestis flocculosa*; S8DUS9 *Fomitopsis pinicola*; V5F1L5 *Kalmanozyma brasiliensis*; A0A0P9EXB3 *Rhodotorula graminis*; F8P9Z8 *Serpula lacrymans* var. *lacrymans*; A0A0C3SF13 *Phlebiopsis gigantea*; M2QHB0 *Ceriporiopsis subvermispora*; A0A0J0XIQ7 *Cutaneotrichosporon oleaginosus*; A0A0H2RKC8 *Schizopora paradoxa*; A0A067PGQ6 *Jaapia argillacea*; A0A0C2W5R2 *Amanita muscaria* Koide; A0A067SH62 *Galerina marginata*; A0A067M073 *Botryobasidium botryosum*; A0A0C9VSH8 *Hydnomerulius pinastri*; A0A0C3B1W4 *Piloderma croceum*; A0A0D0AUY5 *Suillus luteus*; A0A0C9XK69 *Laccaria amethystina*; M5FR71 *Dacryopinax primogenitus*; A0A0D7BRQ6 *Cylindrobasidium torrendii*; A0A0L6WZZ0 *Termitomyces* sp.; A0A0C3Q0Z8 *Tulasnella calospora*; A0A0C3B6Q8 *Serendipita vermifera* (*Piriformospora indica*); A0A0D7A300 *Fistulina hepatica*; A0A0F7SFL9 *Phaffia rhodozyma*; A0A0P1BA25 *Ceraceosorus bombacis*; O42714 *Agaricus bisporus*; A0A151VRY3 *Hypsizygus marmoreus*; A0A0B7FBK7 *Thanatephorus cucumeris*; Q76KC3 *Lentinula edodes*; C0MN02 *Hebeloma cylindrosporum*; A0A1I9UVP2 *Agrocybe aegerita*; U5H8G0 *Microbotryum lychnidis-dioicae*; A8Q7S4 *Malassezia globosa*; J6F2W0 *Trichosporon asahii* var. *asahii*; A0A1J8PYA6 *Rhizopogon vesiculosus*; B1AC68 *Coprinopsis cinerea*; A0A173DSZ5 *Ceratobasidium cereal*; A0A0W0FH87 *Moniliophthora roreri*; A0A0C3E185 *Scleroderma citrinum*; A0A1E3K5W9 *Tsuchiyaea wingfieldii*; A0A0C9TL19 *Paxillus involutus*; A0A0D2NNG1 *Hypholoma sublateritium*; X8JUQ9 *Rhizoctonia solani*; M9MBS9 *Pseudozyma Antarctica*; A0A0C3NAI7 *Pisolithus tinctorius*; A0A165SY25 *Neolentinus lepideus*; A0A166E5N6 *Peniophora* sp.; A0A165CXP7 *Laetiporus sulphureus*; A0A137Q2D9 *Leucoagaricus* sp.; A0A165DMJ9 *Exidia*

glandulosa; A0A165REN6 *Daedalea quercina*; A0A165ZW91 *Fibulorhizoctonia* sp.;
A0A180H0E4 *Puccinia triticina*; A0A166DA95 *Sistotremastrum suecicum*; A0A177V4H2
Tilletia walker; A0A165EHJ8 *Calocera cornea*; A0A077R4C7 *Melanopsichium*
pennsylvanicum; G3LGR3 *Ganoderma lucidum*; C0STD8 *Pholiota nameko*; A0A061B326
Rhodospidium toruloides; A0A146H2T9 *Mycena chlorophos*; A0A1B9GRB8 *Kwoniella*
heveanensis; A0A1E3IUN1 *Cryptococcus depauperatus*; A0A1M2VTI8 *Trametes pubescens*;
A0A1C7MPB6 *Grifola frondosa*; X8JV06 *Rhizoctonia solani*.