Variable susceptibility of laboratory strains of Aspergillus nidulans to hygromycin B and other ribosomal antibiotics

S. D. Martinelli
A. Zamir

Follow this and additional works at: http://newprairiepress.org/fgr

Recommended Citation

This Regular Paper is brought to you for free and open access by New Prairie Press. It has been accepted for inclusion in Fungal Genetics Reports by an authorized administrator of New Prairie Press. For more information, please contact cads@k-state.edu.
Variable susceptibility of laboratory strains of Aspergillus nidulans to hygromycin B and other ribosomal antibiotics

Abstract
Variable susceptibility of laboratory strains of Aspergillus nidulans to hygromycin and other ribosomal antibiotics.

Creative Commons License
This work is licensed under a Creative Commons Attribution-Share Alike 4.0 License.

This regular paper is available in Fungal Genetics Reports: http://newprairiepress.org/fgr/vol34/iss1/10
Table 2. Effect of Cortisone (20uM) on Ageing mutants of N. crassa

<table>
<thead>
<tr>
<th>strain</th>
<th>Malondialdehyde formation after 72 hrs. (O.D. 535nm-600nm)</th>
<th>UV-fluorescence of culture filtrate after 7 days</th>
<th>Biomass production after up to 7 days (mg/10ml)</th>
<th>Linear growth 14th day (in cm)</th>
<th>Growth potencies in liquid media (No. of subcultures)</th>
<th>Conidial viability after 15 days (Survival percentage)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cont.</td>
<td>1.17* + 0.13 Light green</td>
<td>52.5</td>
<td>79.4</td>
<td>18</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>270</td>
<td>1.01* + 0.02 Light green</td>
<td>55.0</td>
<td>76.9</td>
<td>18</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Cont.</td>
<td>1.09 + 0.24 No fluorescence</td>
<td>67.5</td>
<td>73.2</td>
<td>4</td>
<td>0.52</td>
<td></td>
</tr>
<tr>
<td>345</td>
<td>0.94 + 0.08 No fluorescence</td>
<td>70.0</td>
<td>69.4</td>
<td>4</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>cont.</td>
<td>1.35 + 0.20 Light green</td>
<td>50.0</td>
<td>20.5</td>
<td>5</td>
<td>0.66</td>
<td></td>
</tr>
<tr>
<td>377</td>
<td>1.14 + 0.02 Light green</td>
<td>50.0</td>
<td>20.1</td>
<td>5</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>cont.</td>
<td>1.07 + 0.07 Very lt. green</td>
<td>60.0</td>
<td>27.1</td>
<td>12</td>
<td>15.8</td>
<td></td>
</tr>
<tr>
<td>448</td>
<td>0.99 + 0.06 Very lt. green</td>
<td>62.5</td>
<td>29.8</td>
<td>12</td>
<td>4.0</td>
<td></td>
</tr>
<tr>
<td>cont.</td>
<td>0.07 + 0.01 Very lt. green</td>
<td>65.0</td>
<td>108.3</td>
<td>&gt;19</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>EmA</td>
<td>0.07 + 0.01 Very lt. green</td>
<td>62.5</td>
<td>106.9</td>
<td>&gt;19</td>
<td>100</td>
<td></td>
</tr>
</tbody>
</table>

*: Significant at 5% level

a: Average of two independent experiments

From these results, we conclude that ageing is a stochastic process in which several molecular mechanisms work together, and that the concept of 'free radical' theory cannot be applied in every case as the only phenomenon responsible for ageing. - - - Division of Genetics, Institute of Food and Radiation Biology, Bangladesh Atomic Energy Commission, G.P.O. Box 3787, Dhaka, Bangladesh.

Martinelli, S.D. and A. Zamir

Variable susceptibility of laboratory strains of Aspergillus nidulans to hygromycin B and other ribosomal antibiotics

We have been using the ribosomal antibiotics cycloheximide (CHX), paromomycin (PAR) hygromycin (HYG) and geneticin (GEN) to select mutants with altered ribosomal proteins and have found that a random selection of genetically marked strains of A. nidulans have widely differing responses to these compounds, when added to solid medium. The majority of strains gave the same range of responses as the standard Glasgow strain with the biA1 mutation and from which many Aspergillus strains are derived by mutation and recombination. Particularly notable exceptions are strains SDM12 (FGSC A397), SDM108 (FGSC A64) and SDM10 (isolated from a cross between SDM12 and SDM315 [yA2 phenA2]), which are hypersensitive to 2mM PAR, 200 uM GEN and 300 uM HYG. Strains SDM108 and SDM10 are also very sensitive to 0.3 mg/ml CHX. Master strain D is fairly susceptible to these antibiotics but not to the extent of the aforementioned.

In crosses with these strains the hypersensitive phenotype segregates as follows:

1. Hygromycin resistance

   strain SDM390 (fwA1 pabaA1 sB43 alX4) X strain SDM12 (pantoB100)
   phenotype: normal sensitivity hypersensitive
   segregation: 1:1

2. Hygromycin and cycloheximide resistance

   strain SDM108 (wa3 biA1 riboE6) X strain SDM10 (phenA2)
   phenotype: hypersensitive hygromycin resistance to hypersensitivity: 1:7
cycloheximide resistance to hypersensitivity 1:3
The result from the latter cross was particularly surprising since neither parent exhibited any resistance to cycloheximide and yet the combination could produce resistant progeny presumably resulting from complementary gene action. The resistance was associated with an abnormal morphology, slower growth and poor conidiation. Attempts to back-cross progeny have failed owing to the infertility of these strains. Again because of fertility and reluctance to form diploid strains, it is not known whether the sensitivity of strains SDM12 and SDM108 is allelic. The hygromycin resistance may have been generated by three complementing genes.

The variable sensitivity is important for two reasons. Firstly, our attempts to demonstrate the inheritance of aminoglycoside resistance mutations isolated in strain SDM108 have been hampered by the difficulty of finding other strains with the same resistance genotype as SDM108, for the purpose of outcrossing. Secondly, several workers are using the bacterial hygromycin B resistance as a marker in transformation experiments, but they may have unwittingly picked relatively resistant strains for their experiments. If we can map the hypersensitivity to a linkage group, it should be easy to introduce this allele into the recipient strains used by other laboratories in a controlled way without affecting the fertility of those strains (strain SDM108 and its derivatives are very infertile).

Careful screening of stock strains is recommended before commencing any work with antibiotics. -- Dept. of Biology, Birkbeck College, University of London, Malet Street, London WC1E 7HX, England

McCullough, W.

A conditional morphological (fluffy) mutant of A. nidulans. The mutant was recovered during screening for isocitrate lyase constitutive mutants (McCullough, W. and C. F. Roberts 1980 J. Gen. Microbiol. 120:67-84). The original strain was R21 (pabaA yA2) and mutagenesis by N-methyl-N1-nitro-N-nitrosoguanidine. The mutant (M355) produces a mass of sterile aerial hyphae at 37° C (no conidia, no cleistothecia) which may reach the lid of a Petri dish after 4 days incubation in malt extract agar medium supplemented with sucrose (0.02 M). At 18° C the phenotype is intermediate between mutant (at 37° C) and wild type (R21). Conidia form over the surface of a mutant colony if a plate incubated at 37° C is left for a week at room temperature.

The mutant phenotype is due to a single gene defect and is recessive; a diploid constructed between M355 and FGSC A105 biA1;AcrA1;wa3;phenA2;pyroA4;lysB5;sb3;nicB8;coA1 had a normal morphology. This diploid haploidized (McCully, K.S. and E. Forbes 1965 Genet. Res. 6:352-359) and the mutation localized to chromosome III. A cross between M355 and G335 (pantoC3;cnxH3;sc12) located the gene 3 map units from cnxH3. The gene has been designated fluG1.

Some other properties of the mutant are summarized below:

1. fluG1 strains are invasive in the sense that aerial hyphae overgrow the periphery of other colonies.
2. Aerial hyphae form conidia at the junction between fluG and fluG^+ strains.
3. A mass of cleistothecia form at the junction between fluG and fluG^+ strains, and these are present in approximately the same-proportions (selfed and hybrid) as from a normal cross. In fact, crosses can be set up by inoculating fluG and another strain 2 cm apart on a thick malt extract agar plate. This might be an advantage when forcing "difficult" crosses
4. fluG complements moC in heterokaryons (moC is also located on chromosome III, close to adI).

The original mutant M355 (pabaA1 yA2;fluG1) has been lodged with FGSC. -- Dept. of Biology, Univ. of Ulster at Jordanstown, Newtonabbey BT37 OQB, N. Ireland