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## Abstract

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*Physiological examination of a strain of Neurospora* isolated from forest soil from Bruner, Borneo, by J. H. Warcup showed it to be a naturally occurring nitrate non-utilizer. Nutritional experiments in liquid modified Fries' minimal media using a variety of nitrogen sources show that the Borneo isolate is unable to utilize either nitrate or nitrite at temperatures ranging from 18°C to 35°C and at pH's ranging from 5.5 to 7.5. The ammonium ion, amides, amino acids and adenine are good nitrogen sources. The requirement for reduced nitrogen is not alleviated by the addition of vitamin supplements or pyruvate.

The characteristic has been examined genetically by means of a backcrossing program into N. crassa 74 A using an intermediate strain for the initial cross, and it is apparently determined by a single gene. Well over 1000 backcross isolates were tested for utilization or non-utilization of both nitrate and nitrite. The response was always the same to both. Crosses to markers on all chromosome of N. crassa have shown that this gene, designated as nit-4, is in linkage group IV, about 15 map units to the right of cot. Thus nit-4 is linked to nit-3 which is to the left of cot. Strains bearing the mutant gene nit-3 cannot utilize nitrate but respond well to nitrite. Crosses of nit-4 to standard markers 33 nit-2 and nit (2003) show independent assortment of these genes.

Revertants to nitrate utilization were obtained in low frequency when conidia of a colonial mutant of the Borneo isolate were plated on nitrate medium. The frequency of revertants could be increased by exposing conidia to B-propiolactone. These revertants have not yet been analyzed genetically. ---
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| | pH 5.5 | | | pH 6.5 | | | pH 7.5 | | |
|---|---|---|---|---|---|---|---|---|---|
| | 18 C | 25 C | 35 C | 18 C | 25 C | 35 C | 18 C | 25 C | 35 C |
| A. Zero Nitrogen | | | | | | | | | |
| Borneo | 5.3 | 4.5 | 6.3 | 8.9 | 7.3 | 6.8 | 8.1 | 10.0 | 0.7 |
| 74 A | 6.7 | 5.6 | 4.7 | 8.3 | 8.3 | 11.3 | 6.4 | 8.4 | 4.0 |
| B. $NaNO_2$ | | | | | | | | | |
| Borneo | 3.9 | 4.2 | 4.3 | 7.0 | 7.5 | 7.2 | 7.5 | 7.5 | 3.1 |
| 74 A | 32.9 | 43.4 | 25.7 | 37.9 | 51.9 | 31.9 | 30.9 | 34.3 | 27.3 |
| C. $NaNO_3$ | | | | | | | | | |
| Borneo | 7.4 | 8.5 | 5.9 | 8.1 | 9.9 | 8.5 | 9.3 | 9.0 | 9.3 |
| 74 A | 44.7 | 87.3 | 67.5 | 61.0 | 94.2 | 68.7 | 59.3 | 88.3 | 64.4 |
| D. $NH_4Cl$ | | | | | | | | | |
| Borneo | 29.4 | 45.7 | 46.5 | 31.9 | 64.3 | 53.6 | 13.7 | 36.0 | 6.5 |
| 74 A | 74.6 | 97.1 | 74.5 | 98.0 | 112.9 | 86.3 | 40.7 | 88.4 | 73.5 |

Mean dry weights in mg. of 3 replicate mycelial pads of Borneo strain and N. crassa 74 A after 7 days growth in modified Fries' liquid medium with different nitrogen sources, buffered at 3 pH levels at 3 temperatures. A. No added nitrogen. B. $NaNO_2$ at 0.0185 g nitrogen per liter. C. $NaNO_3$ at 1.111 g nitrogen per liter. D. $NH_4Cl$ at 1.111 g nitrogen per liter.