showed that the correlations might have been considerably higher had it not been for three subjects. Interviews revealed that one of them had not eaten breakfast at the time of testing, whereas the other two came direct from afternoon naps. Table 2 shows the results of the second study. Only one of the subjects was tested in both groups.

TABLE 2

CORRELATIONS BETWEEN VISUAL FUSION AND A.C.E. RAW SCORES OF 21 MALE STUDENTS AT UNIVERSITY OF FLORIDA

| Light flash duration for which fusion measured | Quantitative score | Language score | Total score |
|---|---|---|---|
| 38 ms | −.405 | −.515 | −.513 |
| 84 " | −.478 | −.532 | −.485 |
| 135 " | −.114 | −.161 | −.147 |
| 250 " | −.121 | −.172 | −.147 |

Here there are five correlations significant at the 5% level of confidence, and again all the coefficients are negative. It is very interesting to note that light flashes longer than 84 ms do not yield increasingly higher correlations between A.C.E. scores and measures of visual fusion. In this second study, however, the correlations for the 38-ms interval are of the same general magnitude as those for flashes of 84 ms. The L-Scores and Q-Scores do not show the differences which occurred in the first study.

Because time often appears as an exponent in equations describing certain energy relationships,[2] there is the possibility that the relationships under consideration may not be linear. Logarithmic transformations of the data were made, and coefficients of correlations were calculated between (1) A.C.E. scores and logarithms of visual fusion measures, (2) logarithms of A.C.E. scores and visual fusion measures, and (3) logarithms of A.C.E. scores and logarithms of visual fusion measures. In general, resulting correlations were higher than the linear relationships, particularly for the Q-Scores and T-Scores, but the differences were not great.

The correlations between the visual fusion measures for the 84-ms light flash and the A.C.E. scores are strikingly high in view of the homogeneity of the groups studied—namely, college students representing a highly selective group in range of intelligence. It is probable that results for a more heterogeneous group would show even better correlation. In view of the order of determinations of the measures of visual fusion, the results indicate that the relationship was not a function of learning.

The writer is considerably puzzled, however, by the way the coefficients increase with the increase in the length of light flash up to 84 ms and then decrease with a further increase in length of light flash. The shortest noticeable dark intervals averaged 2.75 ms for the 84-ms light flash in the first group, and 6.44 ms in the second study, the difference being attributed to the change in intensity. The total time for a cycle is such that the frequencies represented are within the range of the alpha frequencies of brain waves.

From the results of this experiment, it may be concluded that the shortest noticeable dark period for a light flash of some critical length promises to be a significant physiological correlate of intelligence. It must now be studied for a larger and more heterogeneous group, and be compared to performance on standard intelligence tests and other physiological variables.

[2] The charge of a condenser at any time is expressed by the equation $V = E_{bb}(1 - e^{-t/RC})$.

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## Studies on the Mechanism of Nitrate Assimilation in *Neurospora*


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Although studies concerned with the assimilation of nitrate by plants have indicated the complex nature of this reduction process, in no case has the chemical mechanism involved been clearly demonstrated (see reviews by Burström [*3*], Street [*12*], and McKee [*9*]). Available evidence indicates that the process is enzymatically catalyzed, but there is no general agreement as to the nature of the products of this reduction (*4*, *13*, *15*). Granick and Gilder (*5*) demonstrated the importance of a porphyrin in the reduction of nitrate to nitrite by *Hemophilus influenzae*, and other studies have implicated molybdenum (*10*) and manganese (*2*). (See, however, Arnon [*1*]). The actual site and mechanism of action of these metals are at present unknown.

In an effort to gain greater insight into the process of nitrate assimilation it appeared that the technique of genetically blocking specific chemical steps would prove fruitful, not only in identifying the intermediates in this chain of reactions, but also in demonstrating catalytic components of the system. Over 100 mutant strains of *Neurospora crassa* (microconidial—Tatum), obtained in this laboratory, were unable to grow on nitrate as a sole nitrogen source. These were tested for their ability to utilize nitrite and ammonia. It was found that several mutants which fail to utilize nitrate can grow normally when supplied with nitrite. By mixing these mutants, two at a time, in a liquid basal medium containing only nitrate nitrogen, it was possible to show that there are

at least three distinct mutant types which affect nitrate reduction (mutants A16, 2006, and A361). Growth on nitrate can be obtained by inoculating a nitrate medium with any two of these cultures, an observation indicating that under conditions of heterokaryosis what one mutant lacks another can furnish. Unfortunately it has not been possible to do genetic studies on these strains. Similar heterokaryon tests on mutants which fail to utilize nitrite indicate that there also exist at least three distinct mutant types affecting this step, namely 1896, 2003, and UV392. Heterokaryon formation between any of these two allows growth on the nitrite medium.

*Physiological studies on the individual mutants*

A nitrate basal medium supplemented with a synthetic vitamin mixture, with yeast extract, or with a concentrated dialyzate of the wild type *Neurospora,* would not support growth of mutants A16, A361, and 2006. There was likewise no growth of either mutant A16 or 2006 on a nitrate medium to which was added either a filter-sterilized extract of wild type mycelium grown on nitrate, or a concentrate of an ammonia plus nitrate medium upon which A16 or 2006 had been grown for 3–4 days. However, when these two mutants are grown on a nitrate basal medium (supplemented with a small amount of nitrite) in the same flask but separated by a dialyzing membrane, it is apparent from the enhanced growth of A16 that some diffusible growth factor lacking in A16 is produced by strain 2006. The converse, however, has not been observed. Further studies along these lines are in progress, as well as studies on the reduction *in vitro* of nitrate with mixtures of extracts from these three mutants.

In studies made with mutants 1896, 2003, and UV392, it was observed that UV392 would grow on a nitrite or nitrate basal medium when supplemented with yeast extract (5–10 mg/20 ml of basal medium). This growth-promoting factor in yeast extract is also present in the wild type *Neurospora* extract, from which it can be removed by dialysis. Since it permits the growth of UV392 on a nitrite medium, the factor acts catalytically. Extensive tests indicate that it is not one of the known vitamins or an intermediate of the Krebs-Szent-Györgyi cycle. The nitrite medium supplemented with various concentrations of *Neurospora* trace element solution likewise fails to promote growth. The new growth factor is at present being isolated and purified from yeast extract.

*Mutants affecting the utilization of ammonia*

If the mechanism by which nitrate is assimilated does not involve ammonia as a necessary intermediate, such as has been postulated by Burström (*3*) and others, it should be possible to obtain genetic blocks which inhibit the utilization of ammonia but which do not affect the utilization of nitrate. It has been possible to obtain mutants (Nos. 41, 43, 17) which are unable to grow on Fries ammonia minimal medium, but can still utilize nitrate or nitrite. This evidence strongly suggests that in the reduction of nitrate or nitrite, the intermediates are coupled and reduced as an organic compound, the product of whose reduction is an amino compound, R-$NH_2$, which then transfers the amino group by transamination, or some similar process, thus bringing about the synthesis of amino acids. These observations are also supported by the work of Marthaler (*8*), who found that *Epilobium angustifolium* can use nitrate, but not ammonia, as a source of nitrogen for growth.

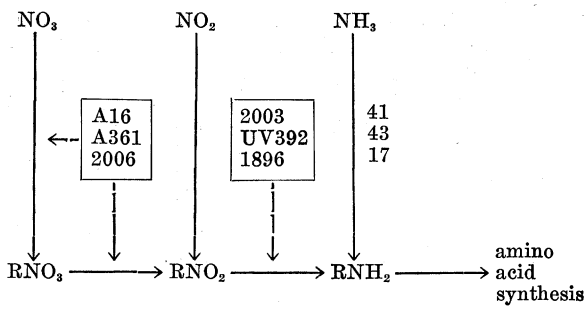


FIG. 1.

The results of the present investigation, along with the observations of others, would suggest tentatively (Fig. 1) the pathway of nitrate, nitrite, and ammonia utilization in *Neurospora.* The sites of the various blocks are indicated.

The identification of the postulated organic intermediates must await additional experimental evidence. As the work of Lemoigne *et al.* (*6, 7*), Virtanen and Csáky (*14*), and Wood *et al.* (*16, 17*) suggests, it is possible that an oxime of oxaloacetate or α-ketoglutarate is an intermediate in the assimilation of nitrite. Tests of the above-mentioned mutants with these oximes are under way at the present time. Suggestive evidence relating to problems of transamination and nitrate and ammonia utilization has come from the work of Stokes *et al* (*11*) on the pH mutant of *Neurospora* requiring pyridoxine. These workers demonstrated that a block in the synthesis of pyridoxine can in some way affect the utilization of nitrate and ammonia differentially. It is possible that further work along this line would implicate pyridoxine or its phosphate derivative in the scheme of nitrate utilization.

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