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THE INFLUENCE OF CARBON DIOXIDE ON SPORULATION AND CITRIC ACID SYNTHESIS IN DARK-GROWN PENICILLIUM ISARIIFORME

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SUMMARY

In higher carbon dioxide concentrations sporulation in dark-grown *Penicillium isariiforme* is inhibited, whereas citric acid production is increased.

1. INTRODUCTION

Light and atmospheric conditions exert an important influence on morphogenesis of fungi of the *Penicillium clavigerum* section. It was established that for *P. isariiforme* light is indispensable for the formation of coremia, bundles of spore-bearing hyphae that react strongly phototropic (CARLILE et al. 1962; PISKORZ 1967; BENNINK 1972b). Carbon dioxide, however, may affect the morphology of the coremia in these fungi (PISKORZ 1972). In a previous paper we reported the inhibitory effect of high concentrations of carbon dioxide on the formation of conidiospores on coremia of *P. isariiforme* (GRAAFMANS 1973).

According to BENNINK (1972a), sporulation in *P. isariiforme* would be induced by low intensity illumination. We have now observed, however, that dependent on the carbon dioxide concentration in the atmosphere, sporulation can occur in darkness.

2. MATERIALS AND METHODS

Methods, materials and experimental conditions were, if not otherwise mentioned, as described previously (GRAAFMANS 1973).

3. RESULTS AND DISCUSSION

In the following experiment cultures were used after a cultivation period of five days. Air-carbon dioxide mixtures (0.03, 5 and 20% carbon dioxide) were humidified and passed over the mycelia at a rate of 30 l/h. It was observed that in normal air (0.03% CO_2), after 72 hours in darkness, the mycelia were covered with a layer of conidiospores. At 5 and 20% carbon dioxide, however, sporulation was inhibited, in the last treatment even completely. These results are fully in agreement with those found for light-exposured cultures (GRAAFMANS 1973).

Owing perhaps to different experimental conditions, Piskorz, studying the influence of carbon dioxide up to a concentration of 6% in the same strain, did not find an effect on sporulation (PISKORZ 1972).

In the study with illuminated cultures we demonstrated that inhibition of conidiospore formation on coremia under the influence of high carbon dioxide concentrations was coupled with a stimulation of citric acid synthesis (GRAAFMANS 1973). Moreover, we found from experiments with $^{14}CO_2$ that active incorporation of this compound in citric acid occurred.

Evidence is now presented that also in dark-grown cultures carbon dioxide had influence on citric acid synthesis, as is shown in *table 1*. Taking into account the amount present on the fifth day after inoculation, the citric acid content in medium and mycelium appeared to increase, depending on the concentration of carbon dioxide in the atmosphere.

Table 1. Citric acid content of cultures of *Penicillium isariiforme* kept at different CO_2 concentrations in darkness

		Citric acid content (μg/mg dry wght. of mycelium)[b]		
	Time[a]	0.03% CO_2	5% CO_2	20% CO_2
Mycelium	0	14.9 (±0.1)		
	24	30.8 (±0.9)	37.9 (±1.0)	36.1 (±0.6)
	48	33.3 (±0.1)	48.9 (±0.2)	57.4 (±0.1)
Medium[c]	0	92.4 (±2.2)		
	24	114.4 (±13.3)	165.8 (±15.7)	186.0 (±22.0)
	48	148.5 (± 0.8)	288.3 (±15.2)	293.4 (± 2.0)

[a] Time expressed in hours after a 5-day cultivation period.
[b] Figures are averages of duplicates; range is indicated between brackets.
[c] Citric acid, expressed in μg, excreted in the medium/mg dr.wght. of mycelium.

In several fungi accumulation of metabolites has been associated with a specific state of differentiation. High ethanol production, for instance, occurred in vegetative mycelium of *Neurospora crassa*, decreasing under sporulating conditions, however, by 96% (WEISS & TURIAN 1966). In *Achlya ambisexualis* and *A. heterosexualis* a water-soluble beta-1,3-glucan was synthesised and stored during vegetative growth. But during sex organ morphogenesis the rate of consumption exceeded the rate of synthesis (FARO 1972).

It would be of interest to undertake further studies on the significance of a relationship between differentiation and citric acid synthesis in *P. isariiforme*.

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