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NOTES AND BRIEF ARTICLES

AN ELEVATED LEVEL OF NADP-LINKED GLUTAMATE DEHYDROGENASE IS NOT A GENERAL FEATURE OF THE CAPS OF AGARIC SPOROPHORES

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During the development of the sporophore of *Coprinus cinereus* (Schaeff. ex Fr.) S. F. Gray (= *C. lagopus sensu* Lewis) the specific activity of the nicotinamide-adenine dinucleotide linked glutamate dehydrogenase (GDH_{NAD}) is increased about threefold in both cap and stipe. In sharp contrast, the specific activity of the nicotinamide-adenine dinucleotide phosphate linked enzyme (GDH_{NADP}) increases greatly in cap tissue but remains at a barely detectable level in stipe and basidiospores (Stewart & Moore, 1974). The cap and stipe are intimately connected tissues and are composed of isogenic cells; so this is an extreme example of tissue-specific developmental gene regulation. Further study of this particular situation is being made with a view to determining the developmental significance of the GDH_{NADP} enzyme. In a broader context, however, it is of interest to establish whether the phenomenon is of widespread occurrence in agarics. The results of a survey of common mushrooms are presented here.

Apart from sporophores of *Coprinus cinereus*, which were grown in pure culture in the laboratory, all of the material was collected during the Autumn of 1974 in and around Northenden, a suburban district of S. Manchester, or in the immediate vicinity of the University campus. Whole sporophores were collected, identified and separated into cap and stipe before being frozen for storage at −40°. Most analyses were completed within 14 days of collection. Crude cell-free extracts, enzyme analyses and protein determinations were done as described previously (Al-Gharawi & Moore, 1974), the amination assays being used throughout. No attempt was made to optimize the enzyme assays for each organism tested. Review of the assay mixtures published for a number of different fungi (yeast, *Aspergillus*, *Neurospora* and *Schizophyllum*) revealed only minor differences

Table 1. *Specific activities (nmoles coenzyme oxidized/min/mg protein) of glutamate dehydrogenase enzymes in sporophores at three stages of development produced by four different dikaryons of* Coprinus cinereus

Dikaryon identity	Sporophore stage	Enzyme activity: GDH_{NADP} Cap	GDH_{NADP} Stipe	GDH_{NAD} Cap	GDH_{NAD} Stipe
BC9/6,6 × H1	Primordium*	21		284	
	Immature fruit	292	31	378	327
	Mature fruit	854	52	1136	1200
BC9/6,6 × ZBw601	Primordium	26		775	
	Immature fruit	404	22	1700	757
	Mature fruit	1550	79	2061	1406
PA1 × H1	Primordium	23		680	
	Immature fruit	45	58	697	580
	Mature fruit	526	37	1127	921
PA1 × ZBw601	Primordium	39		1190	
	Immature fruit	103	37	1330	1031
	Mature fruit	740	47	2070	1271

* Primordia are not separated into cap and stipe for this type of gross enzyme analysis. The monokaryotic parents of the dikaryons are of varied origin: H1 was collected in Bayford, England; BC9/6,6 is a 'laboratory' wild type prepared by backcrossing H1 with its sib H9; PA1 originated in Poland, and ZBw601 in Czechoslovakia.

Table 2. *Specific activities of glutamate dehydrogenase enzymes in extracts of mature sporophores of a range of Agaricales*

	Enzyme activity			
	GDH_{NADP}		GDH_{NAD}	
Organism	Cap	Stipe	Cap	Stipe
Agaricus campestris	99	120	257	529
Boletus erythropus	9	0	31	9
B. scaber	3	0	158	84
B. subtomentosus	0	0	20	0
Clitocybe (fragrans ?)	249	450	141	110
Collybia fuscopurpurea	0	0	46	44
C. peronata	0	0	52	57
Coprinus atramentarius	44	10	524	429
C. comatus	94	66	385	308
C. disseminatus	169	54	361	371
C. micaceus	50	15	659	669
C. plicatilis	84	85	924	566
C. silvaticus	328	48	759	776
Hebeloma crustuliniforme	69	47	39	22
Hygrophorus niveus	360	559	19	62
Hypholoma fasciculare	69	4	23	0
Laccaria proxima	49	64	64	19
Lactarius blennius	5	0	51	220
Lepista sordida	616	647	157	398
Lyophyllum decastes	44	116	295	454
Marasmius oreades	273	108	467	327
Mycena flavo-alba	33	50	35	84
Oudemansiella radicata	25	9	41	207
Panaeolus semiovatus	185	53	337	380
Psathyrella lacrymabunda	3	0	142	177
Russula atropurpurea	0	0	21	13
R. virescens	0	0	80	104

from those routinely used in our work with *Coprinus* and encouraged the view that the *Coprinus* assay mixtures even if not optimal for the other fungi would at least be adequate. In any case, although we would hope that the data are sufficiently reliable to allow comparisons to be made between the different organisms, the major emphasis is on comparison between cap and stipe tissues of the same organism.

Two points have been considered. All of our previous work has been done with a single dikaryotic culture, stock number BC9/6,6 × H1; we have tested the sporophores produced by three more dikaryons to confirm that the enzymic behaviour reported earlier is generally true of *Coprinus cinereus* strains (Table 1). Secondly, the specific activities of GDH_{NAD} and GDH_{NADP} in the caps and stipes were determined in mature soprophores of a wide range of Agaricales (Table 2). Table 1 clearly shows that in all *C. cinereus* strains tested the specific activity of GDH_{NADP} is increased greatly in the cap during sporophore development. Despite considerable differences in the maximum specific activity attained, the ratio between GDH_{NADP} activity in cap and stipe shows remarkable consistency, varying only from about 14:1 to about 20:1 in the mature sporophores. Table 2 demonstrates that such behaviour is only rarely observed in other agarics. Among *Coprinus* spp only *C. silvaticus* and *C. disseminatus*, and among other genera only *Panaeolus semiovatus*, *Marasmius oreades* and perhaps *Hypholoma fasciculare* (though here the generally low specific activities lessen the significance of the differences) have a pattern of enzyme activities in any way similar to that observed in *C. cinereus*. In the long term the diversity of habit, morphology and structure exhibited by these fungi may assist in understanding the function of GDH_{NADP} in sporophore development; but at this stage it is not possible to recognize any relationship between these fungi which correlates with the unusual behaviour of GDH_{NADP}. Nevertheless, it is abundantly clear that derepresssion of GDH_{NADP} in the sporophore cap is not a feature which is shared by many agarics; nor, even, is it common among *Coprinus* species.

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REFERENCES

AL-GHARAWI, A. & MOORE, D. (1974). Effects of D-glutamate on mycelial growth and glutamate dehydrogenase enzymes of *Coprinus lagopus*. *Journal of General Microbiology* **85**, 274–282.

STEWART, G. R. & MOORE, D. (1974). The activities of glutamate dehydrogenases during mycelial growth and sporophore development in *Coprinus lagopus* (*sensu* Lewis). *Journal of General Microbiology* **83**, 73–81.