Proliferating Basidia in *Muribasidiospora indica*

The writer collected a follicular exobasidiaceous fungus in 1964, parasitizing plants of *Rhus mysorensis* at Poona (India). This fungus was placed in a new genus *Muribasidiospora*, M. N. KAMAT and R. B. RAJENDREN[1] on the basis of its ability to produce muriform basidiospores. Critical examination of this material over a period of 2 years revealed an interesting mode of basidial development, not previously reported in the class Basidiomycetes. A brief report on this interesting phenomenon is made in this paper.

Materials and methods. Fresh material of this fungus was examined in scrapings made as by technique described by SAVILE[2]. Microtome sections were stained with iron hematoxylin and light green combinations.

Results. Prior to sporulation the parasitic hyphae concentrate in the sub-stomal chamber and produce the basidia, which push through the stomatal opening displacing the adjacent epidermal cells in the process (Figure 1). At this stage, the entire fructification closely resembles that of *Exobasidium*, in habit. Basidia which are produced in palisade layers are cylindric, clubshaped, strictly bisterigmate and produce spore at each end (Figure 4) capable of producing a chain of arthrospores (Figures 2 and 3). This is the primary crop of basidia and basidiospores in the fructification.

With the discharge of the basidiospores, the basidia shrink and become empty. A new basidium starts growing from the basal septum of the primary basidium in the form of a bud (Figures 5, 6 and 9), with the primary basidia persisting like a sheath over the newly growing basidium within. The secondary basidium soon attains the form of a typical clubshaped basidium closely similar to the primary one differing only in being non-sterigmate. This unique process is repeated over the entire fructification, thus giving rise to a crop of secondary basidia (Figures 6, 7 and 9). Tertiary formation of basidia was rare (Figure 9). On reaching a length of 40 to 50 μ, the secondary basidia become highly granulated over their apical region, which now is separated from the basal portion by a septum. The highly granulated apical region develops septation in various planes and becomes a muriform 'spore-body' in a manner typical of chlamydospore formation (Figures 9 and 10).

The 'spore body' so produced differs from the primary basidiospore in the manner of its formation and in its being persistent and non-deciduous and irregular in form. In rare cases, the entire secondary basidium is converted into a 'spore body' (Figure 8). This phenomenon of proliferation is illustrated in Figures 5–10.

Preliminary observations made on nuclear behaviour of this fungus show typical features of true meiosis and mitosis of uniform pattern in the primary as well as secondary basidia.

Discussion. There is no report of proliferating basidia in the Basidiomycetes except the one by ERIKSSON (1958)[3].

[1] R. B. RAJENDREN, Mycopath. Mycol. appl., in press (1967).
[2] D. B. O. SAVILE, Can. J. Bot. *37*, 641 (1959).
[3] J. ERIKSSON, Symb. Bot. upsal. *16*, (1958).

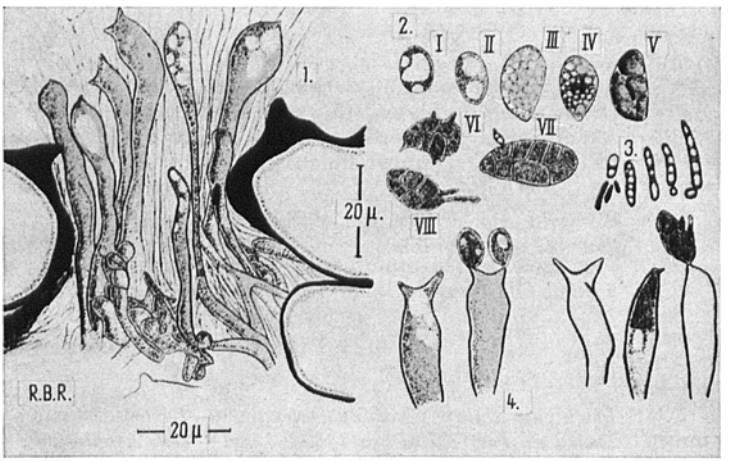
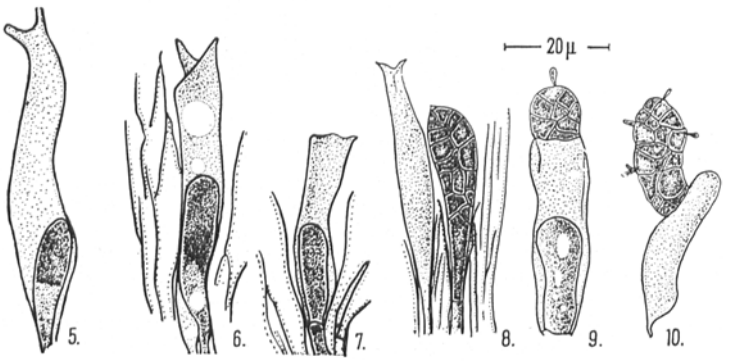


Fig. 1. Super stomal sori.

Fig. 2, I–VIII. Different stages of development of muriform basidiospores.

Fig. 3. Arthrospores.

Fig. 4. Basidium at different stages of development.



Figs. 5–7. Stages in the development of secondary basidia through proliferation.

Fig. 8. Secondary basidium completely converted into a spore body.

Figs. 9 and 10. Apical portion of the secondary basidia producing the spore body.

The phenomenon observed by the author in *M. indica*, M. N. KAMAT and R. B. RAJENDREN, has, however, many features distinct from that obtained by ERIKSSON, in respect of morphology, structure and even manner of formation of basidiospores.

While ERIKSSON noted the occurrence of such a phenomenon under high humidity conditions, the author obtained it under strictly Zerophytic conditions. This probably explains the limited number of crops produced by this fungus. The phenomenon noted by the author in *M. indica* is in the nature of true proliferation, and not repeating basidia[4].

Zusammenfassung. Die Entstehung der sekundären Basidien bei *Muribasidiospora indica*, M. N. KAMAT und R. B. RAJENDREN, und ihre Sporenbildung durch Prolifikation wurde beschrieben.

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[4] Acknowledgment: grateful thanks are offered to Prof. M. N. KAMAT for guidance and interest during the course of this study.

An Autoradiographic Study of the Uptake of ^{3}H-Leucine During Primary Embryonic Induction in vitro

Earlier studies[1,2] imply that the reacting system of primary embryonic induction goes through a phase of determination before the effects of induction becomes histologically visible. To understand the mechanism of primary induction, it is important that one knows what cellular events occur in the reacting system during the determination phase. As a possible approach to this question, a comparison was made during and soon after the determination phase, between protein synthesis in the ectoderm developing under the influence of the dorsal mesoderm and that in the ectoderm not influenced by the same inducing tissue. A comparable study of RNA synthesis in the same induction system has already been published[3].

The ectoderm was isolated from a beginning gastrula of *Triturus pyrrhogaster* and then divided into 2 approximately equal pieces, one of which was fused to a piece of the dorsal mesoderm and cultured in Holtfreter solution (experimental) while the other ectoderm piece was cultured without mesoderm (control). If the culture is extended for more than 4 days, the first piece forms neural tissue, and the other develops into an irregular mass of ciliated cells. Such pairs of explants were cultured for 1, 3, 6, 22 and 48 h at 18 °C, and then incubated in 2 μC/ml of ^{3}H-leucine (10,900 mC/m*M*, Nuclear Chicago Corporation) in culture medium for 3 h after which they were fixed in acetic acid and alcohol. Six μ thick paraplast sections were treated with 5% trichloroacetic acid at 4 °C for 10 min, washed, and then covered with Kodak NTB 3 liquid emulsion.

Silver grains were counted separately per unit nuclear and cytoplasmic areas in each ectoderm explant. The mean value of 75 counts and its 90% confident limits were computed for nucleus and cytoplasm in each ectoderm explant. Comparison of the mean values thus obtained was made between the same cell component of the pair of experimental and control ectoderm. The data are shown in the Table. To ascertain whether a pair of ectoderm explants obtained from the same gastrula gives a valid basis of comparison, a series of control experiments was conducted in which 2 ectodermal explants originating from 1 gastrula were exposed to the labeled precursor. In no pair did we find a significant difference in grain count.

The data summarized in the Table show slight enhancement of uptake in the experimental nuclei for 3- and 22-h groups, and a larger, consistent enhancement in the 48-h group (Figures 1 and 2). In all time groups, regardless of series, the cytoplasm shows values much lower than those of the nucleus. No consistent differences were observed in the cytoplasmic counts between the experimental and control series. Exceptionally low values were obtained for the cytoplasm of the 3-h group. The low cytoplasmic radioactivity throughout all time groups and series is striking. A question may be raised as to whether or not the labeled precursor is first taken up in the cytoplasm,

Comparison of grain counts/unit area of induced and non-induced ectoderm cells at various time intervals from the beginning of culture[a]

h	Nucleus		Cytoplasm	
	Experimental	Control	Experimental	Control
1	13.67 ± 0.83	16.01 ± 1.13	4.03 ± 0.33	[b]5.62 ± 0.38
	9.04 ± 0.52	[b]12.63 ± 0.76	2.25 ± 0.04	[b]4.23 ± 0.31
	13.24 ± 1.20	[b]18.37 ± 1.09	6.80 ± 0.76	8.31 ± 0.50
	[b]13.52 ± 1.09	10.75 ± 0.72	4.60 ± 0.38	3.80 ± 0.31
	(0.85)[c]		(0.80)	
3	11.08 ± 0.84	10.67 ± 0.67	0.25 ± 0.02	0.31 ± 0.02
	[b]13.73 ± 0.86	8.12 ± 0.48	[b]0.37 ± 0.02	0.16 ± 0.01
	[b]12.17 ± 0.78	7.91 ± 0.41	0.23 ± 0.02	0.14 ± 0.02
	[b]15.17 ± 1.10	11.05 ± 0.58	0.19 ± 0.02	0.07 ± 0.02
	(1.38)		(1.53)	
6	[b]10.52 ± 0.68	7.92 ± 0.37	4.40 ± 0.26	3.27 ± 0.22
	8.77 ± 0.47	9.88 ± 0.59	3.73 ± 0.17	3.89 ± 0.24
	14.12 ± 1.15	16.08 ± 1.05	6.16 ± 0.50	[b]8.17 ± 0.73
	18.16 ± 1.28	18.99 ± 1.02	8.77 ± 0.64	9.31 ± 0.53
	(0.98)		(0.94)	
22	[b]18.80 ± 1.10	13.51 ± 0.81	7.55 ± 0.53	6.15 ± 0.46
	[b]14.77 ± 0.78	11.33 ± 0.58	5.16 ± 0.47	4.49 ± 0.37
	[b]20.07 ± 1.22	14.23 ± 0.74	8.07 ± 0.76	6.52 ± 0.51
	13.89 ± 0.88	12.53 ± 0.76	[b]5.68 ± 0.31	4.27 ± 0.83
	(1.31)		(1.23)	
48	[b]23.51 ± 1.09	11.05 ± 0.52	[b]8.36 ± 0.55	3.80 ± 0.83
	[b]27.20 ± 2.54	14.44 ± 0.76	[b]13.69 ± 1.61	5.81 ± 0.44
	[b]19.64 ± 1.09	11.89 ± 0.54	5.63 ± 0.54	4.75 ± 0.44
	[b]19.85 ± 1.42	13.44 ± 0.83	6.09 ± 0.91	5.49 ± 0.53
	(1.77)		(1.70)	

[a] The average of 75 counts is accompanied by 95% confidence limits. Each row indicates a pair of data to be compared, derived from the same ectoderm. [b] The difference is significant with $P < 0.05$. [c] Ratios of experimental to control counts for each time group are in parentheses.

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[3] S. K. BRAHMA, J. Embryol. exp. Morph. *16*, 203 (1966).