

Properties	<i>C. scotii</i>	BY 2
1. Growth in malt extract .	Sediment	Pellicle
2. Assimilation of potassium nitrate. . . . .	Positive	Negative
3. Utilization of ethanol . .	Occasionally some growth.	Positive
4. Riboflavin excretion . .	Nil	Positive

The question arises as to how far these differences in properties are of taxonomic importance. Pellicle formation has been used as the criterion for differentiating certain species of *Candida* (e.g. *C. mycoderma* from *C. zeylanoides*)<sup>6</sup> capable of assimilating only glucose. Similarly, the ability to utilize potassium nitrate separates *C. pelliculosa* from *C. albicans* and *C. parapsilosis*. Finally, while the mutant yeast, BY 2, excretes into the medium an appreciable quantity of riboflavin, there is no record of a similar behaviour in *C. scotii*.

Therefore the riboflavin producing mutant yeast, BY 2, has to be considered as a new species, and the name<sup>7</sup> *Candida ghoshii* n. sp. is proposed for it.

Its position in the key to the species of the genus *Candida* given by LODDER *et al.*<sup>6</sup> is indicated below: No sugar fermentation. Glucose, galactose, saccharose and maltose assimilated.

a) Early pellicle formation; nitrate not assimilated; riboflavin excreted ... *C. ghoshii* n. sp.

b) No pellicle; nitrate assimilated; no riboflavin excretion ... *C. scotii*.

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### Résumé

Pour une détermination taxonomique, nous avons étudié les caractères morphologiques, physiologiques et culturels d'une levure mutante produisant la lactoflavine. Tout d'abord cette souche mutante est produite en traitant à l'acenaphthène et levure de bière à fermentation basse. Cette souche est asporogène et présente des formes pseudomycéliques. Elle est incapable de fermenter le glucose et n'utilise pas le KNO<sub>3</sub> comme source d'azote. Elle n'assimile pas le lactose. Après une étude approfondie sur les différents caractères de cette souche, nous estimons qu'elle peut être considérée comme une nouvelle espèce de levure du genre *Candida*. Nous l'avons appelée *C. ghoshii* n. sp.

<sup>7</sup> The species is named after Sir J. C. GHOSH, who gave a fillip to these investigations on yeast at the Indian Institute of Science, Bangalore.

## Glycolytic Enzymes in the Human Amniotic Fluid

Very few enzymatic activities have so far been described in the amniotic fluid. Only scanty data exist on cholinesterase<sup>1</sup>, phosphatase<sup>2</sup>, and lysozyme<sup>3</sup>.

As it is well known that glucose, fructose and, probably, other reducing sugars are present in the amniotic fluid<sup>4</sup>, it was a matter of interest to investigate whether glycolytic activities could be detected in this liquid.

The following enzymatic activities have been examined: phosphohexose-isomerase, ribose-5-phosphate-isomerase, aldolase and lactic-dehydrogenase.

Determinations have been made of the total protein content of the amniotic fluids under examination.

**Experimental.**—The samples of amniotic fluid (of about 15 ml each) were collected from pregnant women at term by transabdominal puncture, *intrapartum* by vaginal puncture of amniotic sac or during Caesarean section.

The samples were filtered and stored at 0° for no longer than 24–48 h. Samples contaminated by blood or meconium were discarded.

The protein content was determined by the GOA's micromethod<sup>5</sup>.

**Phosphohexose-isomerase.**—The activity of this enzyme was determined according to BODANSKY<sup>6</sup>: in a centrifuge tube 0.3 ml veronal buffer pH 7.8 0.1 M, 0.3 ml glucose-6-phosphate sodium salt 0.045 M, 0.2 ml amniotic fluid were pipetted. The mixture was incubated at 37° for 1 h and the reaction stopped by addition of 3 ml of 20% trichloroacetic acid (TCA). After centrifugation, fructose was determined on 2 ml of the supernatant according to ROE<sup>7</sup>.

**Aldolase.**—Determinations were carried out in a centrifuge tube mixing 1 ml of TRIS-HCl buffer pH 7.4 0.1 M, 0.25 ml hydrazine 0.56 M pH 7.4, 0.25 ml iodoacetate 0.002 M, 0.25 ml distilled water and 1 ml of amniotic fluid and 0.25 ml hexose 1-6-diphosphate Na salt 0.06 M<sup>8</sup>.

The mixture was incubated for 1 h at 37°, and successively deproteinized by 3 ml of 10% TCA. After centrifugation, triosephosphate was determined on 1 ml of the supernatant according to SIBLEY and LEHNINGER<sup>9</sup>. For standardization of method, in some samples alkali-labile phosphorus was determined<sup>10</sup> in duplicate.

**Ribose-5-phosphate-isomerase.**—This activity was tested according to AXELROD<sup>11</sup>: 0.5 ml of TRIS-HCl buffer pH 7.4 0.1 M, containing 0.5 mg Ribose-5-phosphate (Ba salt) were mixed with 0.5 ml of amniotic fluid; after incubation at 37° for 30 min ribulose-5-phosphate

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Protein content and glycolytic activities of human amniotic fluid

Samples	Protein <sup>1</sup>	Aldolase <sup>2</sup>	PEI <sup>3</sup>	LDI <sup>4</sup>	PRI <sup>5</sup>
1	208	0.278	0.340	0.250	—
2	296	0.094	0.108	0.054	—
3	206	0.237	0.340	0.155	—
4	140	0.121	—	—	—
5	262	0.244	0.150	0.056	—
6	193	0.057	0.052	0.088	0.045
7	164	0.025	0.045	0.079	0.095
8	148	0.048	0.018	0.034	—
9	255	0.040	0.097	0.029	0.149
10	241	0.149	0.059	0.021	—
11	302	0.087	0.108	0.043	0.023
12	182	0.312	0.185	—	—
13	145	0.067	0.140	0.137	0.034
14	200	0.055	0.106	0.067	0.318
15	160	0.056	0.090	—	0.220
16	206	—	0.106	—	0.089
17	205	0.024	—	—	0.263
18	162	0.019	—	—	0.405
19	175	0.103	—	—	0.240
20	217	0.032	—	—	0.350
21	207	0.034	—	—	0.130
Mean values	203 ± 46	0.104 ± 0.091	0.149 ± 0.101	0.181 ± 0.126	0.085 ± 0.066

<sup>1</sup> Protein = mg%ml.<sup>2</sup> Aldolase =  $\mu$ M hexosediphosphate/h/mg of protein.<sup>3</sup> PEI = phosphohexoso-isomerase = mg fructose/h/mg of protein.<sup>4</sup> LDI = lactic-dehydrogenase =  $\mu$ M/min/mg of protein.<sup>5</sup> PRI = phosphoribose-isomerase =  $\mu$ M ribulose/h/mg of protein.

was determined with the sulfuric acid-cistein-carbazole reagent.

**Lactic dehydrogenase.**—This activity was determined spectrophotometrically<sup>12</sup> as follows: in a cuvette of the Beckman spectrophotometer were pipetted: 0.4  $\mu$ M DPNH in 0.1 ml, 3  $\mu$ M piruvate in 0.1 ml, 0.3 ml NaHCO<sub>3</sub> 0.02 M, 2.5 ml phosphate buffer 0.1 M pH 7.8.

The reaction was started by addition of 0.1 ml amniotic fluid and the decrease of O.D. at 340 m $\mu$  was followed for 10 min at room temperature taking readings every minute.

In the Table the data obtained in these experiments are reported with their mean and standard deviation. It is evident that all enzymatic activities tested are present in considerable amounts in the amniotic fluid.

The enzymatic activity referred to the protein content is much higher in amniotic fluid than in blood serum of either mother or newborn<sup>13</sup>.

The values obtained from the different samples are considerably scattered. It has not been possible to relate such differences to the way in which the fluid was obtained nor the time the liquid have been stored.

The protein values obtained are in good agreement with those previously reported by other authors<sup>14</sup>.

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### Riassunto

È stata messa in evidenza nel liquido amniotico di donne a termine di gravidanza la presenza di alcuni enzimi glicolitici. Sono riportati i dati quantitativi ottenuti per l'attività della fosforiboso-isomerasi, aldolasi, fosfoesoso-isomerasi e lattico-deidrogenasi. È stato determinato inoltre il contenuto proteico dei campioni di liquido amniotico presi in esame.

### Post Nephrectomy Increase in Serum Ribonuclease Activity after Total Hepatectomy or Nitrogen Mustard Derivatives Administration\*

It has been previously shown that serum ribonuclease (Rase) activity markedly increases after bilateral nephrectomy in the rat<sup>1</sup>, this phenomenon being observed in animals undergoing a number of experimental procedures, including 'functional evisceration'<sup>2</sup>. Although our results suggested that the liver is not the source of the enzyme increase, the disadvantages of leaving that organ *in situ*<sup>3</sup> led us to study the effects of total hepatectomy and evisceration. These were performed by the two stage technique of INGLE<sup>4</sup> on Wistar rats of both sexes (average body weight 250 g). The second stage was carried out under ether anesthesia, and was immediately followed by bilateral nephrectomy, non-nephrectomised

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