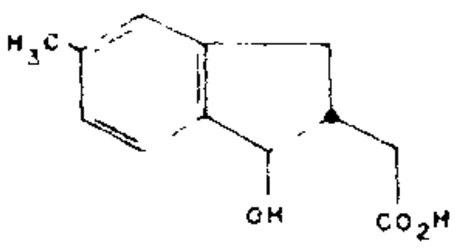
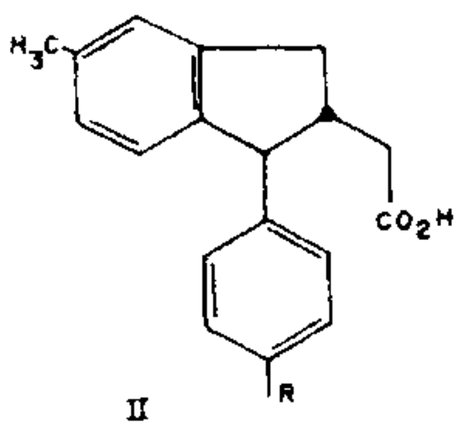
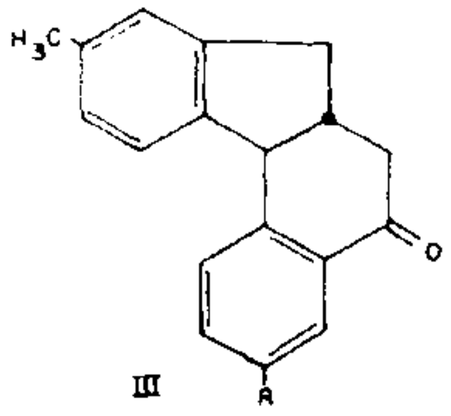


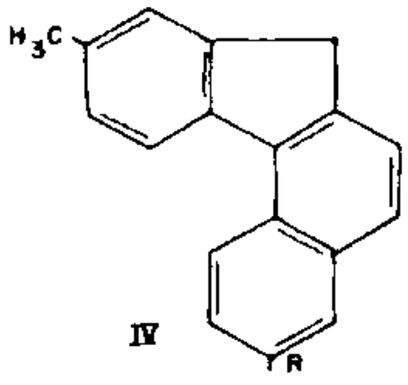
I



II



III



IV

trans) along with the *cis*-lactone (20%), b.p. 160°/2.5 mm., m.p. 53°.

The $AlCl_3$-catalyzed alkylation of benzene with the hydroxyacid (I) in a mixture of sym-TCE and nitrobenzene at low temperature furnished the normal alkylation product, 5-methyl-1-phenyl-trans-2-indanylacetic acid (II, R = H) in 65% yield, m.p. 115°, P.M.R.: dibenzyl proton at δ, 4.2, d, J = 9 Hz (trans), purified and isolated through the methyl ester, b.p. 190–195°/2 mm. Intramolecular Friedel-Crafts acylation of the acid (II, R=H), furnished the benzfluorenone derivative (III, R=H) in 66% yield, b.p. 160–165°/2 mm., which showed the characteristic trans dibenzylic-H at 4.2 δ, d, (J = 8.8 Hz) and the Ar-H, peri to carbonyl group, at 7.8 δ in the PMR spectra. Reduction of the ketone by $LiAlH_4$ followed by dehydration and dehydrogenation furnished 7-methyl-3,4-benzfluorene (IV, R=H), m.p. 70°, picrate, m.p. 136–137°[4], TNB-complex, m.p. 146–147°.

The catalyzed condensation of the hydroxyacid (I) with toluene in a similar manner afforded 5-methyl-1-*p*-tolyl-trans-2-indanylacetic acid (II, R=CH_3), m.p. 116°, methyl ester, m.p. 53°, in 60% yield. The structure of the acid was confirmed by an unambiguous synthesis starting from *p*-tolyl magnesium bromide and methyl 5-methyl-1-keto-2-indanylacetate followed by dehydration and catalytic hydrogenation, as well by the PMR spectra. Intramolecular Friedel-Crafts cyclization of the acid gave the benzfluorenone derivative (III, R=CH_3), m.p. 99–100°, 2,4-DNP derivative, m.p. 222°, which was converted to 7,1′-dimethyl-3,4-benzfluorene (IV, R=CH_3), m.p. 107°, picrate, m.p. 143°, TNB-complex, m.p. 165°. The PMR spectrum of the hydrocarbon shows two ortho coupled meso aromatic protons at δ 8.62 (d, J = 8.8 Hz) and at δ 8.24 (d, J = 8.8 Hz), far downfield from the rest.

The reaction is being fully investigated for the synthesis of highly substituted benzfluorenes and naphthofluorenes, some of which are of interest to us for a study of carcinogenic activity and from the standpoint of stereo-chemistry as they are likely to develop chirality due to molecular overcrowding.

Financial assistance from the Govt. of West Bengal is gratefully acknowledged.

22 July 1985; Revised 15 November 1985

1. Phillips, D. D. and Chatterjee, D. N., *J. Am. Chem. Soc.*, 1958, **80**, 1360; Chatterjee, D. N. and Bhattacharya, S. P., *Tetrahedron*, 1971, **27**, 4153.
2. Roy, J., *J. Indian Chem. Soc.*, 1948, **25**, 69.
3. House, H. O., Babad, H., Toothhill, R. B. and Noltes, A. W., *J. Org. Chem.*, 1962, **27**, 4141.
4. Keene, B. R. T. and Schofield, K., *J. Chem. Soc.*, 1957, 3181.

BIOMASS ENERGETIC YIELD AND MAINTENANCE COEFFICIENTS OF *ASPERGILLUS WENTII* ON DIFFERENT CARBOHYDRATES

V. S. BISARIA and S. NENE
Biochemical Engineering Research Centre, Indian Institute of Technology, New Delhi 110016, India.

ERICKSON *et al*[1] applied mass and energy balance regularities to aerobic microbial growth processes by using equivalents of available electrons in growth substrate, biomass and product. This approach has been used in the present investigation for evaluating the efficiency of conversion of some carbohydrates to fungal biomass using *Aspergillus wentii* Wehmer strain Pt 2804. This fungus was selected as it fulfilled most of the criteria outlined by Imrie and Righelato[2] for selection of suitable strains for production of fungal biomass from carbohydrates.

The fungus was cultivated in a 5-litre bioreactor containing 3.5 litre of modified Mandels and Weber's medium[3]. The cultivation conditions in the bioreactor runs and the determination of growth associated

parameters such as cell mass, substrate consumption, oxygen uptake rate, carbon dioxide evolution rate and product formation have been described by the authors[3]. The method of determination of the calculated variables, viz specific growth rate (μ), specific substrate consumption rate ($Q_s = \mu/Y_s$), biomass carbon yield (y_c), fraction of organic substrate carbon in products (z), fraction of substrate carbon in carbon dioxide (d), fraction of energy in organic substrate evolved as heat (ε), fraction of energy in substrate converted to products (ξ_p), fraction of energy in substrate converted to biomass (η) has also been reported[3].

When determining the values of Y_s^{max} and m_s by the Q_s *vs* μ plot[4], statistical regression was used to give the least square best fit. The correlation factor, χ^2, indicates the closeness of the fit; values of 0.8–1.0 indicate very good fits. The true biomass energetic yield coefficient, η^{max}, was calculated using the correlation,

$$\eta^{max} = \frac{\sigma_b \nu_b}{\sigma_s \nu_s} \cdot Y_s^{max}.$$

The values of true biomass energetic yield coefficient and maintenance coefficient obtained for *A. wentii* on different carbohydrates are summarized in table 1.

The maintenance coefficient for *A. wentii* was comparable to that of *Candida utilis* (the yeast of choice in commercial MBP process) especially when grown at low concentration of glucose, starch and xylan (table 1). Oxygen deficiency can lead to higher maintenance requirements[4]. This seems to be the case with 3.0% starch and 5.0% glucose runs where culture broth viscosities were high and oxygen transfer rates decreased causing higher maintenance requirements. Higher maintenance in the 5% glucose (phosphate deficient) medium was probably due to buffering capacity of phosphate. As maintenance of intracellular pH requires energy, the maintenance coefficient is likely to be higher; besides phosphate deficiency affects the turnover of cell components and therefore increases maintenance requirements.

The values of η^{max} seem to be rather high, especially for 0.5% xylan and 0.5% starch. This was probably due to the poor fits of the straight line plot of Q_s vs μ for these two runs (table 1). The yeast had a high value of η^{max} as 0.67.

The authors thank Dr R. D. Tyagi of this Centre for valuable suggestions.

Nomenclature

m_s—rate of organic substrate consumption for maintenance (g/g dry biomass. h)

Y_s^{max}—true biomass yield based on organic substrate (g dry wt/g substrate)

η^{max}—true biomass energetic yield coefficient (dimensionless)

ν_s, ν_b—reductance degree of organic substrate and biomass respectively (equivalent available electron/g atom carbon)

σ_b, σ_s—weight fraction carbon in biomass and organic substrate respectively (dimensionless)

27 January 1986

1. Erickson, L. E., Minkevich, I. G. and Eroshin, V. K., *Biotechnol. Bioengg.*, 1978, **20,** 1595.
2. Imrie, F. K. E. and Righelato, R. C., In: *Food from wastes* (eds) G. G. Birch, K. J. Parker and J. T. Worgan, Appl. Sci. Pub., London, 1976.
3. Bisaria, V. S. and Nene, S., *J. Ferment. Technol.*, 1984, **62,** 7.
4. Pirt, S. J., *Principles of microbe and cell cultivation*, Blackwell Scientific Publications, London, 1975.

Table 1 *True biomass energetic coefficient η^{max} and maintenance coefficient, m_S, for Aspergillus wentii on different carbohydrates*

Organism	Substrate	η^{max}	m_S	χ^2
Aspergillus wentii	0.5% glucose	0.57	0.020	0.881
	0.5% xylan	0.66	0.023	0.623
	0.5% starch	0.69	0.025	0.775
	3.0% starch	0.57	0.06	0.953
	5.0% glucose	0.48	0.075	0.983
	5.0% glucose (PO_4 deficient)	0.44	0.10	0.972
Candida utilis	1.0% glucose	0.67	0.021	0.907

A RAPID METHOD FOR PLANT ANATOMY USING FLUORESCENCE MICROSCOPY

S. P. TIWARI and T. G. K. MURTHY
Cytogenetics Section, National Research Centre for Groundnut, (I. C. A. R.), Junagadh 362015, India.

THE fluorescence technique is mainly used for observing chloroplasts, porphyrins and pollen grains[1]. Subsequent to the discovery of fluorochromes[2] and fluorescent antibody technique[3], fluorescence microscopy was applied to embryological[4] and biophysical