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Patents and Literature

The objective of this section is to keep readers aware of significant inventions and trends in industrial research as well as to highlight those areas of research that may lead to new biotechnological opportunities. Four major areas of biochemistry will be covered corresponding to enzymes, cells, bioproducts, and nucleic acids. The patent section will briefly cover each area in every issue of the journal. The literature section will focus on one area per issue.

Patents

This section will identify patents and published patent applications from the international patent literature. The title, name(s) of the inventor(s), the patent number, the date of filing, the assignee, and a short description of the invention will be given. Copies of US patents can be obtained for 50¢ each from the Commissioner of Patents and Trademarks, Washington, DC 20231.

Enzymes

Process for the Microbial Production of Alcohol Oxidase

L. Eggling; M. Paschke; H. Sahm
US 4,250,261 (Jul. 9, 1979)
Kernforschungsanlage Julich Gesellschaft m.b.H.

Alcohol oxidase is produced from *Hansenula polymorpha* yeast by cultivating them in a methanol–nutrient broth and then adding to the broth a noncatabolite substrate repressive to alcohol oxidase such as glycerin, sorbite, or xylose.

Novel Neutral Glucoamylase and Method for its Production

M. Tamura; M. Shimizu; M. Tago
US 4,254,225 (Jul. 9, 1979)
CPC International Inc.

0273-2289/81/0900-0247$02.00

A method is described for the production of a glucoamylase with a molecular weight of 50,000. Optimum enzyme activity occurs at pH 6.0–6.5 at an incubation temperature of 60°C.

Thermal Destabilization of Microbial Rennet

S. Branner-Jorgensen
US 4,255,454 (Dec. 28, 1978)
S. Branner-Jorgensen

Mucor miehei or *Mucor pusillus* rennet is acylated, retaining at least 50% of its activity. This results in reduced thermal stability of the enzyme preparation which can then be used to advantage in milk coagulation.

Method for Isolation of Glucose Isomerase

H. Hirohara; S. Nabeshima; S. Mitsuda; T. Nagase
US 4,255,521 (May 21, 1979)
Sumitomo Chemical Company, Limited

A *Streptomyces* cell suspension is subjected to high pressure and then passed through an aperture to instantaneously drop the pressure: this renders the cells fragile and susceptible to fracture when sprayed against a wall at high velocity.

Glycerol Oxidase and Process for the Production Thereof

O. Terada; T. Uwajima; H. Akita
US 4,255,519 (Jul. 19, 1979)
Kyowa Hakko Kogyo Company Ltd.

An oxidizing enzyme is produced with specific activity for glycerol and dihydroxyacetone. The enzyme has a pH optimum of 7.8–8.6, an optimum temperature of 37°C, and a Michaelis constant for glycerol of $8.0 \times 10^{-3} M$.

Method of Purification of Glucose Isomerase

D. M. Jackson; Y. Tsuda; V. Winans
US 4,256,838 (Nov. 13, 1979)
UOP Inc.

A method is described for purifying glucose isomerase which uses a nucleic acid precipitation step followed by adsoption and elution of the enzyme from DEAE-cellulose.

Immobilized Enzyme Catalyst

R. W. Glass; J. Glogowski
US 4,259,445 (Jun. 1, 1979)

A catalyst is prepared of subcellular particulates of vegetable material containing an enzyme system composed of different carbohydrate-transforming enzymes. The particulates are encapsulated in a matrix that is permeable to permit access of carbohydrate substrate, but also acts to immobilize the enzymes and preserve their activity.

Process for the Production of Urokinase in Pure Condition

R. Hafeli
US 4,259,447 (Jun. 29, 1979)

Urokinase of high concentration and purity is obtained by adsorption on an immobilized aprotinin matrix at pH 6 or above and eluted from the matrix at a pH of 4.5 or lower.

Process for the Enzymatic Softening of Furs

A. Asbeck; H. F. Pfeiffer; J. Plapper
US 4,260,686 (Aug. 17, 1979)
Henkel KGAA

Fur is softened by contacting it with an aqueous acid liquor containing an acid protease from a fungus strain of the genus *Rhizopus*.

Alkaline Protease Produced by a Bacillus

B. Tenijenhuis
US Re. 30,602 (Dec. 12, 1977)
Gist-Brocades N V NL

A novel enzyme that has high proteolytic activity in alkaline media and is suitable for inclusion in washing compositions is produced by *Bacillus* strain PB 92.

Cells

Yeast Autolysis Process

K. C. Chao; E. F. McCarthy; G. A. McConaghy
US 4,218,481 (Oct. 6, 1978)
Standard Oil Company (Indiana)

A process for autolysing active yeast cells by incubating them with protease enzymes with continuous mixing to yield an autolyzed yeast slurry.

Process for Producing Acrylamide or Methacrylamide Utilizing Microorganisms

I. Watanabe; Y. Satoh; T. Takaro
US 4,248,968 (Mar. 28, 1979)
Nitto Chemical Industry, Co.

Bacteria from the genus *Corynebacterium* or *Nocardia* are grown in media containing acrylonitrite or methacrylonitrite at pH 6–10.

Preparation of Bacterial Cell Aggregates

A. H. Chen; Y. Jao
US 4,251,632 (Sept. 11, 1978)
Miles Laboratories, Inc.

An extrudable bacterial cell aggregate of *Streptomyces olivaceus* having improved hardness is prepared using a crosslinking agent such as glutaraldehyde and a soluble cationic polymer.

Process for Growing Human Epidermal Cells in Tissue Culture

M. G. Eisenger; J. M. Hefton
US 4,254,226 (Sept. 13, 1979)
Sloan Kettering Institute for Cancer Research

A process is described for growing human epidermal cells in a tissue culture medium having a pH of 5.6–5.8.

Method of Cultivating Cells of Animal and Human Tissues

H. Katinger; W. Scheirer
US 4,259,449 (Aug. 23, 1978)
Chemap AG

Cells of animal and human tissues are cultivated in a compartmentalized container; the cells are kept suspended in a nutrient solution and prevented from settling by a controlled oxygen-containing gas at a pressure somewhat greater than that necessary to counteract the weight of the nutrient solution.

Organism ATCC 31643

S. M. Steenbergen; K. M. Young
US 4,259,451 (Jun. 20, 1980)
Merck & Co., Inc.

A pure culture of a variant of *Agrobacterium radiobacter*, ATCC 31643, produces a heteropolysaccharide when cultivated.

Bioproducts

Interferon Stabilization

S. E. Grossberg; J. J. Sedmak
US 4,252,791 (Oct. 19, 1979)
The Medical College of Wisconsin, Inc.

Interferon is stabilized by mixing it with a soluble lanthanide or calcium salt.

Process for Producing Syrups or Syrup Solids Containing Fructose-Terminated Oligosaccharides

S. Okada; S. Kitahata; S. Yoshikawa; K. Miyake
US 4,254,227 (Mar. 8, 1979)
Kabushiki Kaisha Hayashibara Seibutsu Kagaku Kenkyujo

A process is described for producing syrups with fructose-terminated oligosaccharides that uses an immobilized cyclodextrin glucanotransferase.

Amine Salts of Acidic Microbial Polysaccharides and their Use in Recovery of Such Polysaccharides

C. W. Schroeck
US 4,254,257 (Feb. 21, 1979)
The Lubrizol Corporation

Acidic polysaccharides produced by microbial fermentation are converted to methanol-insoluble amine salts in which the amine has a molecular weight of at least 150 and contains at least three amino nitrogen atoms.

Modified Heterpolysaccharides and Their Preparation

L. A. Naslund; A. I. Laskin
US 4,256,590 (Apr. 11, 1979)
Exxon Research and Engineering Co.

A heteropolysaccharide from the genus *Xanthomonas* is isolated and prepared by a special saline and heat process; the hot solution is then injected underground to displace oil that can be recovered at a production well.

Drilling Fluid Containing Crosslinked Polysaccharide Derivative

C. H. Kucera; D. N. DeMott
US 4,257,903 (Oct. 19, 1978)
The Dow Chemical Company

A drilling fluid is produced that is composed of a hydroxylalkyl galactomannan crosslinked with a water soluble alkali metal aluminate.

Process for Resolving D,L-Leucine

D. P. Bauer
US 4,259,441 (Sept. 17, 1979)
Ethyl Corporation

Process for resolving D,L-leucine that involves the following: acylation of the racemic mixture; preparation of *N*-acyl-D,L-leucine esters; reacting the esters with microbial-derived serine proteinases; separating the unreactive *N*-acyl-D-leucine ester from the resulting *N*-acyl-L-leucine; and, recovering D- and L-leucine.

Synthesis of Ascorbic Acid from Lactose

J. P. Danehy
US 4,259,443 (Jun. 12, 1979)
Bernard Wolnak and Associates

A method to synthesize ascorbic acid from lactose that involves the hydrolysis of lactose and the conversion of D-glucose and D-galactose to the *gamma*-lactones of L-galactonic acid and L-gulonic acid. The lactones are enzymatically oxidized to ascorbic acid.

Process for Making Cellulase-Free Xanthum Gum

K. S. Kang
US 4,259,477 (Jun. 4, 1979)
Merck & Co., Inc.

Cellulase is removed from xanthum gum by addition of an alkali metal hypochlorite at a pH of 6.0–7.0 and heating the mixture at 85–95°C for 2–10 min.

Method and Apparatus for Measurement of Glucose Content

K. Muramatsu; K. Samizo
US 4,260,680 (Oct. 6, 1978)
Mitshubishi Chemical Industries, Ltd.

A method and apparatus is described for measuring the glucose content in a liquid sample by passing the sample consecutively through an ion exchange resin and a column of immobilized glucose oxidase and then determining the amount of hydrogen peroxide produced.

Method and Glycoprotein Composition for Inhibition of Growth of Transformed Cells and Tumors

K. Isselbacher; D. K. Podolsky; M. W. Weiser
US 4,261,976 (Oct. 10, 1978)
Massachusetts General Hospital

A glycoprotein that inhibits malignant growth is obtained from animals and humans having malignant cells or tumors. It has a molecular weight of 3600 and is a substrate for both the GT-I and GT-II isoenzymes of serum galactosyltransferase.

Method for Producing Microbial Cells and Use Thereof to Produce Oxidation Products

H. C. Tsang; A. Laskin; R. N. Patel
US 4,266,034 (Mar. 3, 1979)
Exxon Research and Engineering Company

Newly dicovered strains of microorganisms grow aerobically in the presence of methane as the major carbon and energy source. The cells possess a high protein content and can be used as feedstuffs. They also contain enzymes capable of converting C1–C6 alkanes to alcohols, and C3–C6 secondary alcohols to methyl ketones.

Nucleic Acids

A Novel Plasmid and its Microbiological Preparation

J. J. Manis
UK Patent Application GB 2 046 272 A (Apr. 2, 1980)
The Upjohn Company

The novel plasmid pUC7 can be obtained from the microorganism *Streptomyces espinosus* subsp. *acanthus* NRRL 11081. The plasmid can be used as a cloning vehicle in recombinant DNA work.

Intercalation Inhibition Assay for Compounds that Interact with DNA or RNA

C. L. Richardson; G. E. Schulman
US 4,257,774 (Jul. 16, 1979)
Meloy Laboratories, Inc.

The binding of a compound to a nucleic acid is quantitatively determined by a competitive inhibition fluorescent polarization assay that uses an intercalating dye like ethidium bromide or acridine orange.

Microorganisms Having Multiple Compatible Degradative Energy-Generating Plasmids and Preparation Thereof

A. M. Chakrabarty
US 4,259,444 (Jun. 7, 1972)
General Electric Company

A bacterium from the genus *Pseudomonas* is prepared to contain at least two stable plasmids, each of which provides a separate hydrocarbon degradative pathway.

Process for the Preparation of Deoxyribonucleases

T. Ando; T. Shibata; E. Hayase; S. Ikawa
US 4,259,446 (Jul. 19, 1979)
Rikagaku Kenkyusho

Restriction enzymes are prepared from *Bacillus* of at least two different substrate specificities selected from the group Bsu R, Bsu 1247 I, Bsu 123 I, and Bsu 1231 II.

Interferon Production

C. Colby; D. W. Denney
US 4,262,090 (Jun. 6, 1979)

Hybrid mammalian cell clones are induced to produce interferon messenger RNA in amounts sufficient to produce complementary DNA. The single-stranded complementary DNA is then used to prepare double-stranded DNA, which can then be combined with a replicon capable of transforming microorganisms. The transformants are capable of replicating and expressing the recombinant DNA.

DNA Joining Method

J. Shine
US 4,264,731 (Apr. 4, 1978)
Regents, University of California

DNA molecules having reactant ends that are capable of being joined in a ligase-catalyzed reaction are pretreated to remove the 5′-terminal phosphate groups. This treatment prevents certain ends from joining to each other and enhances the frequency of joining the desired combination

Literature Survey

The objective of the literature ssurvey is to make a thorough, recent review of publications in one specific area. This issue's survey will be on bioproducts. Future surveys will be on nucleic acids, enzymes, and cells. The articles will be chosen for their impact on current biotechnology processes and for their potential to break new ground that may lead to new applications. The entries are listed in alphabetical order by the first author's name.

Bioproducts

1. Polyhydric Alcohol Production and Intracellular Amino Acid Pool in Relation to Halotolerance of the Yeast Debaryomyces Hansenii, L. Adler and L. Gustafsson, *Arch.. Microbiol.* **124,** 123–130 (1980).
2. Production of Useful Nucleotides with Immobilized Microbial Cells, Y. Ado, K. Kimura, and H. Samejima, *Enzyme Eng.* **5,** 295–304 (1980).
3. Production of Biomass from Enzymatic Hydrolysate of Agricultural Waste, A. Araujo and J. D'Souza, *J. Ferment. Technol.* **58,** 399–401 (1980).
4. Production of Stereospecific Alpha-Amino Acids by Biological Hydrolysis of Racemic Alpha-Amino Nitriles, A. Arnaud, P. Galzy, and J. C. Jallageas, *Bull. Soc. Chim. Fr.* **1–2,** 87-90 (1980).

5. Purification of Fecal Waste Waters Combined with Algal Protein Production, K. Benderliev and Kh. Dilov, *Khidrobiologiya* **10,** 12–26 (1979).
6. Production of Ethanol from Plant Biomass, Y. S. Bhatia, *Indian Chem. J.* **15,** 31–36 (1980).
7. Production of High-Fructose Syrup by a Heat-Fixed Lactobacillus Sp, M. Bhatia and K. A. Prabhu, *Biotechnol. Bioeng.* **22,** 1957–1977 (1980).
8. Production of Insulin-Like Growth Factors and Their Carrier By Rat Pituitary Gland and Brain Explants In Culture, M. Binoux, P. Hossenlopp, C. Lassarre, and N. Hardouin, *FEBS Lett.* **124,** 178–184 (1981).
9. Evaluation of the Use of Biomass from *n*-Alkanes in Animal Nutrition by Analyzing Animal Production, L. Boniforti, M. De Vincenzi, and V. Silano, *Ann. Inst. Super. Sanita* **15,** 587–589 (1980).
10. The Potential Use of Immobilized Plant Cells for the Production and Transformation of Natural Products, P. Brodelius, B. Deus, K. Mosbach, and M. H. Zenk, *Enzyme Eng.* **5,** 373–381 (1980).
11. Immobilized Whole Cells of the Yeast Trigonopsis Variabilis Containing D-Amino Acid Oxidase for the Production of Alpha-Keto Acids, P. Brodelius, B. Hagerdal, and K. Mosbach, *Enzyme Eng.* **5,** 383–387 (1980).
12. Hydrolysis of Cellulosic Wastes, K. Buchholz, J. Puls, B. Godelman, and H. H. Dietrichs, *Process Biochem.* **16,** No. 1, 37–43 (1980/81).
13. Enzymic Modification of Vegetable Protein: Mechanism, Kinetics, and Production of Soluble and Partially Soluble Protein in a Batch Reactor, A. Constantinides and B. Adu-Amankwa, *Biotechnol. Bioeng.* **22,** 1543–1565 (1980).
14. Microbial Production of Primary Metabolites, A. L. Demain, *Naturwissenschaften.* **67,** 582–587 (1980).
15. Continuous Ethanol Production by Immobilized Cells of Zymomonas Mobilis, W. Grote, K. J. Lee, and P. L. Rogers, Biotechnol. Lett. **2,** 481–486 (1980).
16. Petroleum and Microorganisms. 8. Production of Amino Acids, Y. Hirose, *Petrotech (Tokyo)* **2,** 814–820 (1979).
17. Use of Cellulase-Depressed Mutant of Cellulomonas in the Production of a Single-Cell Protein Product from Cellulose, E. V. Hitchner and J. M. Leatherwood, *Appl. Environ. Microbiol.* **39,** 382–386 (1980).
18. Silk Protein Production by the Immobilized Silk Gland, Y. Ikariyama, M. Aizawa, and S. Suzuki, *J. Solid-Phase Biochem.* **4,** 69–73 (1979).
19. Methane Production from Wastewaters by Immobilized Methanogenic Bacteria, I. Karube, S. Kuriyama, T. Matsunaga, and S. Suzuki, *Biotechnol. Bioeng.* **22,** 847–858 (1980).
20. Biomass Production of Tobacco Cells. VI. Growth-Substrate Relationship of Tobacco Cells in Suspension Culture, A. Kato and K. Tsuji, *J. Ferment. Technol.* **59,** 33–36 (1981).
21. Production of Sugar Nucleotides by Fermentation, K. Kawaguchi, H. Kawai, and T. Tochikura, *Methods Carbohydr. Chem.* **8,** 261–269 (1980).
22. Biomass Production, S. Kawamura, *Hakko Kogaku Kaishi* **58,** 266–270 (1980).
23. Lysine Production with Brevibacterium sp 22 LD Using Sugar Cane Molasses. I. Study of Optimization, A. M. Khalaf Allah, B. Janzso, and J. Hollo, *Acta Aliment.* **9,** 107–116 (1980).
24. Microbial Protein Production, J. H. Litchfield, *Bioscience* **30,** 387-396 (1980).
25. Production of Extracellular Amino Acids by Some Wood-Attacking Fungi, R. A. Masova, *Mikol. Fitopatol.* **13,** 381-384 (1979).

26. De Novo Protein Synthesis by Human Chondrosarcoma in Cell and Organ Culture: Evidence of Unusually High Collagen Production by a Neoplastic Tissue, D. R. Miller, B. V. Treadwell, and H. J. Mankin, *Connect. Tissue Res.* **8,** 9–20 (1980).
27. Application of Temperature-Sensitive Mutants for Single-cell Protein Production, Y. Miyasaka, C. Rha, and A. J. Sinskey, *Biotechnol. Bioeng.* **22,** 2065–2079 (1980).
28. Extracellular Production of Proteins by Microorganisms. Part V. Stimulatory Effect of Inhibitors of Cell Wall Synthesis on Protein Production by *Bacillus Brevis*, S. Miyashiro, H. Enei, K. Takinami, Y. Hirose, T. Tsuchida, and S. Udaka, *Agric. Biol. Chem.* **44,** 2297–2303 (1980).
29. Induced Production of Acidic Polysaccharide by Benzalkonium Chloride in a Bacteria and Some Properties of the Acidic Polysaccharide Produced, K. Nishikawa, S. Oi, and T. Yamamoto, *Agric. Biol. Chem.* **43,** 2305–2310 (1979).
30. Bacterial Utilization of Pure and Mixed Cl Compounds for Single Cell Protein Production, E. Papoutsakis, W. Hirt, and H. C. Lim, *Biotechnol. Bioeng.* **23,** 235–242 (1981).
31. High Productivity Ethanol Fermentations with *Zymomonas Mobilis*, P. L. Rogers, K. J. Lee, and D. E. Tribe, *Process Biochem.* **15,** No. 6, 7–11 (1980).
32. Microbial Methanogenesis from Acetate, M. R. Smith, S. H. Zinder, and R. A. Mah, *Process Biochem.* **15,** No. 4, 34–39 (1980).
33. Biomass Yields and Maintenance Requirements for Growth on Carbohydrates, B. O. Solomon and L. E. Erickson, *Process Biochem.* **16,** No. 2, 44–49 (1981).
34. Production of Intracellular and Extracellular Protein from *n*-Butane by *Pseudomonas butanovora* sp. nov, J. Takahashi, *Adv. Appl. Microbiol.* **26,** 117–127 (1980).
35. Continuous Production of Salicylic Acid: Effect of Dilution Rate, pH, Substrate Concentration, and Cell Recycle, S. K. Tangnu and T. K. Ghose, *Process Biochem.* **16,** No. 1, 22–24 (1980/81).
36. Vitamin B 12 Production from Methanol by Continuous Culture of *Pseudomonas* Am-1, Y. Tsuchiya, N. Nishio, and S. Nagai, *J. Ferment. Technol.* **58,** 485–487 (1980).
37. High Value Utilization of Molasses for Fermentative Production of L-Lysine, J. S. I. Wang, *Taiwan Sugar* **26,** 155–160 (1979).
38. Production of Plasma Proteins by Human Renal Cell Carcinoma Line (GEP-1) in Culture, T. Watanabe, K. Iijima, T. Tajima, and Y. Ohshika, *Igaku No Ayumi* **110,** 604–606 (1979).
39. Extracellular Polysaccharide Biosynthesis by Pseudomonas NCIB 11264. Studies on Precursor-Forming Enzymes and Factors Affecting Exopolysaccharide Production by Washed Suspensions, A. G. Williams and J. W. T. Wimpenny, *J. Gen. Microbiol.* **116,** 133–141 (1980).
40. Production of D-Phenylglycine-Related Amino Acids by Immobilized Microbial Cells, H. Yamada, S. Shimizu, H. Shimada, Y. Tani, S. Takahashi, and T. Ohashi, *Biochimie* **62,** 395–399 (1980).