Electronic Journal of Biotechnology ISSN: 0717-3458 Vol.5 No.3, Issue of December 15, 2002

Teaching of Biotechnology in the Biochemistry Course

Maria Luisa Mourato de Oliveira Marques Serralheiro
Departamento de Química e Bioquímica
Faculdade de Ciências da Universidade de Lisboa
Campo Grande, 1749-016 Lisboa
Portugal
Tel: 351 21 7500925
Fax: 351 21 7500979
E-mail: mls@fc.ul.pt

Ana Maria Ponces Freire *
Departamento de Química e Bioquímica
Faculdade de Ciências da Universidade de Lisboa
Campo Grande, 1749-016 Lisboa
Portugal
Tel: 351 21 7500925
Fax: 351 21 7500979
E-mail: aponces@fc.ul.pt

Biotechnology is taught in the Biochemistry course of the Faculty of Science of the Lisbon University during the last year of the course divide into two semesters. These two disciplines are optional with other disciplines that the students may choose in order to complete their knowledge in Biochemistry. Both disciplines have two credits, one for theory and the other for laboratory: Applied Biochemistry, comes in the first semester and Immobilized Biocatalysts in the second. Each discipline is taught during 13 weeks. The main goal of these two disciplines in the Biochemistry course is to give the students an applied vision, devoted to the industry, of the Biochemistry they have been taught during the previous three years.

Applied Biochemistry is divided into three blocks: I- Applications in Food Industry (four hours); II- Applications in the Environmental Area (three hours); III-Applications in the Health Area (six hours). This makes thirteen hours and completes a semester.

I- Applications in Food Industry

The first block starts with an introduction to the use of genetically modified foods followed by a forum discussion with the participation of the students. It follows the application of enzymes and microorganisms in the dairy (cheese and yoghurt), beverage (beer and fruit juices), bread, meat, sugar and sweeteners industry. Before focusing on the need and benefits of using exogenous biocatalysts, an introduction is given to the natural enzymatic conversion of the raw material into the final product.

In the first session of this block the biochemistry of cheese and yoghurt production is given (Eskin, 1990), focusing mainly on the milk protein precipitation and on the enzymatic transformations of caseins into small peptides (how they can be characterised) is talked, the transformation of lipids into fatty acids and the whole biochemical transformations of the monomers into flavour compounds is also evoked. The symbiosis of two strains for the transformation of milk is highlighted using as an example the production of yoghurt. Next some questions are raised: How the biochemistry can interfere with these processes? How biochemistry can improve them? The production of different starter cultures, as well as the coagulating enzyme rennin and the use of probiotics cultures are some of the subjects that are focused. It is also referred the problem associated with the presence of lactose in the dairy products and how it can be overcome by using free or immobilized enzyme to accomplish its hydrolysis.

The second food industry referred is the beverage industry with the fruit juices and beer included. For the fruit juices it is only mentioned the use of pectinases in the transformation process, for the beer industry it is a process in which the biochemistry takes part and the natural biochemistry transformation is analysed together with the use of exogenous enzymes like amylases and proteases (Eskin, 1990).

The third section is the bread industry together with the

* Corresponding author

starch industry (syrups of glucose versus artificial sweeteners). In the bread formation from wheat flour some biochemical changes of starch and the flour proteins are analysed as well as the fermentation process. The last stage of bread making (the oven process) is mainly analysed on what concerns the Maillard reactions (Eskin, 1990). In this technology the introduction of amylases and lipases to improve the bread making process or even to change it, is developed. The other transformation process of starch into syrups (mainly from maize grains) (Gerhatz, 1990) is also referred together with the use of amylase and amiloglucosidase. As this part of the lesson is about sweeteners, artificial sweeteners are also referred, mainly aspartame with its enzymatic synthesis process (Cheetham, 2000) This block about food industry will end with the biochemical transformation of animal muscle into meat and the use of proteases to change the softness of the meat, finally the production of amino acids used in the food industry, aspartic acid, phenylalanine and glutamic acid, through the fermentation process is also referred. In this topic it is focused the fermentation technology area as well as some aspects of metabolic engineering applied to the phenilalanine production (Eggeling et al. 2001).

II-Applications in the Environmental Area

The second block of Applied Biochemistry is centred on the theme "environment". Here three lessons are also given, the first introduces the pollution problem, the three different types in which the pollutants can be divided (liquid, solid and gaseous) (McKinney and Schoch, 1998) and how the biochemistry can be used to improve the environment quality. Even during the first session it is explained, briefly, on what consist the liquid effluents treatment and the process of activated sludges is described giving some examples with treatment of dairy industry and municipal wastes.

The second session is devoted to the treatment of solid and gaseous effluents, being the emphasis on composting and biofiltration, respectively. The process of composting is analysed on what concerns aeration and temperature (Palmisano and Barlaz, 1996). Some examples are also present to the students: the use of composting to the treatment of solid biodegradable residues from municipal wastes; and to treatment of the olive oil extraction industry. The use of biofiltration as a technique for the treatment of gaseous effluents is also tackled. The mechanisms of biofiltration, the biofilter media and the microbiology of the process, is briefly referred (Devinny et al. 2000). The use of biochemistry through the application of microorganisms in both processes is highlighted.

The last session of this block is concerned with bioremediation and bioconversion aspects of the pollution control. In this session the bioremediation in situ of different compounds is analysed (McKinney and Schoch, 1998) and specific situations in which purified enzymes or cells are used to catalyse the conversion of a pollutant compounds into an inoffensive one is also tackled (Wackett and Hershberger, 2001).

III- Applications in Health Area

The third block is devoted to applications in the health field. In these lectures subjects like the initial production of drugs from biological origin or the more recent production of proteins and hormones with therapeutic interest, antibodies and vaccines and also DNA therapy are analysed. Other subjects included in these lectures are the drug delivering systems, biomaterials and also tissue engineering. The objective is to give a broad view of the actual research in this field and not a deep insight of these different subjects. For this block six seminars are given, as this is the subject more appealing for the biochemistry students. In the first session and introduction to this area is done. In this introduction the evolution in this field is analysed and also the production of antibiotics and steroids through the use of fermentation technology and enzymatic catalysis is referred (Walsh, 1998).

During the second session the importance, production and applications of cytokines (interferons and interleukins) are analysed. The third seminar is devoted to the production and hormones of therapeutic use, insulin, glucagons, human growth factor are some of the examples (Walsh, 1998).

Next the use of DNA vaccines their production and application is explained during the fourth session. In the fifth session the drug delivering systems are discussed focusing mainly the liposomes technology.

The last subject to be analysed is biomaterials (Ratner, 1996) and the tissue engineering (Bell, 2000). These two very complex subjects although briefly described in this session will give a broad view of the application of biochemistry in the medical field. To teach some of these topics different scientists working in this field are invited.
During the term, the students have to prepare and present seminars (20 minute presentation). Usually each group has two students. Several subjects under the theme discussed in the three blocks are proposed and the students can choose whatever they prefer. For the first block the subjects are:

I-1.Genetically Modified Foods: advantages and human health implications;
I-2. Functional and Nutraceutical Foods: What are they, how they can be produced and advantages in their use;
I-3- Probiotic Cheeses and Yoghurts;
I-4- Omega 3, are they useful to the human health?;

I-5- Metabolic engineering in the amino acid production.

The subject to be analysed by the students referring to the environmental block are:

II-1: The bioconversion of whey from the dairy industry into valuable compounds;
II-2. Biodegradation of dyes from the textile industry;
II-3. Biodegradable plastics: production and biodegradation;
II-4. The microbiological process of reduction of VOCs through the use of biofilters.
II-5. Microbial Biofiltration Systems
For the health block the proposed subjects are:
III-1: The influence of growth factors in the wound healing process;
III-2: The use of DNA vaccines to treat diabetes;
III-3. Delivering systems for hormones;
III-4. Bioreaction to the implanting of Biomaterials: Protein Adsorption, Cell Adherence and Tissue Formation
III-5.Biomaterial-Tissue Interaction
III-6: Bioartificial Pancreas
III-7: Tissue Development: Cell-Extracellular Matrix Interactions.

The seminars are included in the credits corresponding to the practical sessions.

The evaluation process consists of a final examination covering all the subjects taught during the semester and the seminar that the students presented. The seminar is divided into a written monograph and the oral presentation during 20 minutes approximately. The examination contributes with 60% to the final mark and the seminar with 40%.

During the second semester Immobilization of Biocatalysts is the discipline taught with the objective of specifying some of the aspects tackled during Applied Biochemistry: I- Production and down-stream processing of enzymes (5 hours); II- Immobilization of biocatalysts (enzymes and cells) (6 hours); III- Biocatalysis in organic systems (2 hours). The first subject is the production of biocatalysts in which the fermentation process is analysed. The kinetics of one substrate fermentation is studied together with the fermentation reactor equations for the batch and chemostat (Eggeling et al. 2001). In the down-stream process the chromatographic methodology is taught briefly since this subject is analysed in others disciplines (Instrumental Methods of Biochemistry Analysis and Enzymology, second and third year, respectively). More time is devoted to the enzyme purification using two-phase systems, either aqueous or organic with reverse micelles (a brief introduction is done about these systems) (Dekker et al. 1989). Next enzyme and cell immobilization process is presented. The advantages of immobilization are focused as well as the methodology to accomplish the immobilization (Pederson and Christensen, 2000). The effect of this process on the enzyme behaviour is analysed on what concerns the enzyme kinetics (Ballesteros and Boross, 2000). The kinetic equations are only developed taking into account the external diffusion effects. Bioreactors are also tackled and the modelling is taught assuming the Michaelis-Menten equation for one substrate without enzymatic loss of activity (Cabral and Tramper, 2002). The medium engineering in the thermal stability of enzymes is also analysed. In this subject the relationship between structure and stability is analysed, as well as the mechanisms of enzyme inactivation and the strategies to stabilise the enzyme through medium engineering or genetic engineering are focused. Another subject is the use of biocatalysts in organic media (Adlercreutz, 2000). The interest in using these systems is focused and the effect of the different systems parameters on the enzymatic activity is analysed. As both disciplines can be chosen independently, during the last session some case-studies of the industrial application of biocatalysts are referred. This discipline has also seminars given by the students and the presentation is done during the practical classes. In these seminars the students must analyse one paper and present it during 10 minutes. This discipline has three hours per week of laboratory classes, half of the time is used for the students to present their seminars and the other half the students have to accomplish four laboratory sessions of 6 hours each. This laboratory work consists of:

L1-Immobilization Methods (occlusion methods for cells, covalent immobilization of enzymes);
L2- Purification of Enzymes by Aqueous two-phase Systems;
L3-Enzymatic Dipeptide Synthesis in Reverse Micellar Medium;
L4- Anaerobic Degradation of Azo Dyes.

After each laboratory session a report is written in which each group presents their results and have to write a small discussion of the problem.

The evaluation process of this discipline consists of a final examination covering all the subjects taught during the semester, the seminar that the students presented and also the reports from the laboratory session. The examination contributes with 60% to the final mark, the seminar and the reports with 20%.

References

ADLERCREUTZ, P. Biocatalysts in non-conventional media. In: STRAATHOF, A.J.J and ADLERCREUTZ, P., eds. *Applied Biocatalysis*. Harwood Academic Publishers,

The Netherlands, 2000, p. 295-316. ISBN 90-5823-024-4.

BALLESTEROS, A. and BOROSS, L. Biocatalyst Performance. In: STRAATHOF, A.J.J and ADLERCREUTZ, P., eds. *Applied Biocatalysis*. Harwood Academic Publishers. The Netherlands, 2000, p. 271-294. ISBN 90-5823-024-4.

BELL, E. Tissue Engineering in Perspective. In: LANZA, R.P.; LANGER, R. and VACANTI, J. eds. *Principles of Tissue Engineering*. Academic Press, N.Y. 2000, p. XXXV-XL. ISBN: 0-12-436630-9.

CABRAL, J.M.S. and TRAMPER, J. Bioreactor Design. In: STRAATHOF, A.J.J and ADLERCREUTZ, P., eds. *Applied Biocatalysis*. Harwood Academic Publishers, The Netherlands, 2000, p. 339-378. ISBN 90-5823-024-4.

CHEETHAM, P.S.J. Case studies in the application of biocatalysts for the production of (bio)chemicals. In: STRAATHOF, A.J.J and ADLERCREUTZ, P. eds. *Applied Biocatalysis*. Harwood Academic Publishers, The Netherlands, 2000, p 93-153. ISBN 90-5823-024-4.

DEKKER, M.; HILHORST, R. and LAANE, C. Isolating enzymes by reversed micelles. *Analytical Biochemistry*, 1989, vol. 178, p. 217-226.

DEVINNY, J.S.; DESHUSSES, M.A. and WEBSTER, T.S. *Biofiltration for air pollution control*. Lewis Publishers, CRC Press, Boca Raton, Florida, USA. 2000, p. 1-23. ISBN 1-56670-289-5.

EGGELING, L.; PFEFFERLE, W. and SAHM, H. Amino acids. In: RATLEDGE, C. and KRISTIANSEN, B., eds. *Basic Biotechnology*. Cambridge University Press, 2001, p. 281-304. ISBN 0-521-77917-0.

ESKIN, N.A.M. *Biochemistry of Foods.* Academic Press, 1990, p. 367-400. ISBN 0-12-242351-8.

GERHARTZ, W. *Enzymes in Industry*. VCH, Verlagsgesellschaft mbH, Weinheim, FRG, 1990, p. 92-102. ISBN 0-89573-937-2.

MCKINNEY, M.L. and SCHOCH, R.M. *Environmental sciences*. systems and solutions. Web Enhanced Edition. Jones and Bartlett Publishers. Barb House, Barb Mews, London, UK. 1998, p. 388-560. ISBN 0-7637-0613-2.

PALMISANO, A.C. and BARLAZ, M.A. *Introduction to solid waste decomposition*. In: PALMISANO, A.C. and BARLAZ, M.A. eds. Microbiology of solid waste. CRC Press, N.Y. 1996, P. 2-31. ISBN 0-8493-8361-7.

PEDERSON, S. and CHRISTENSEN, M.W. Immobilized Biocatalysts In: STRAATHOF, A.J.J and ADLERCREUTZ, P., eds. Applied Biocatalysis. Harwood Academic Publishers. The Netherlands, 2000, p. 213-228. ISBN 90-5823-024-4.

RATNER, B.D. *Biomaterials Science: An interdisciplinary endeavour*. In: RATNER, B.D.; HOFFMAN, A.S.; SCHOEN, F.J. and LEMONS, J.E., eds. Biomaterials Science. An Introduction to Materials in Medicine. Academic Press, N.Y. 1996, p. 1-8. ISBN 0-12-582461-0.

WACKETT, L.P. and HERSHBERGER, C.D. *Biocatalysis and Biodegradation. Microbial Transformation of Organic Compounds*. ASM Press, Washington, D.C. 2001, p. 171-190. ISBN 1-55581-179-5.

WALSH, G. *Biopharmaceuticlas: Biochemistry and Biotechnology*. John Wiley & Sons, NY, USA. 1998, P. 1-36; 235-409. ISBN 0-471-97789-6.