



Luciano Piubelli

 UNIVERSITY OF INSUBRIA



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Contact data

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Biography

Luciano Piubelli was born in Milan, Italy, in 1967.

He studied Biological Sciences at the University of Milan where he received his Degree in 1991.

In 1995 he received his Ph.D. in Cellular and Molecular Biology.

From October 1995 to September 1997 he carried out his research activity as a Post-Doc fellow in the laboratory directed by Prof. H.R. Bosshard, at the university of Zurich (Switzerland).

From October 1997 to June 1999 he carried out his research activity as a Post-Doc fellow in the laboratory directed by Prof. G. Zanetti, at the University of Milan.

In 1999 he joined as Research Associate the research group directed by Prof. M.S. Pilone and L. Pollegioni in Varese (Functional Post-Genomics and Protein Engineering Laboratory) at the Dept. of Structural and Functional Biology of the University of Insubria group, where he currently works.

In 2004 he moved, as a co-founder, to the newly established Dept. of Biotechnology and Molecular Sciences still in Varese.

He was a member of the organizing committee of the School on Enzyme Engineering (in 2005) and the National Symposium "Proteine 2000" for the Italian Society of Biochemistry and Molecular Biology.

Qualifications and awards

1995 Ph.D. in Cellular and Molecular Biology, University of Milan, Italy.
Thesis title: "Structural and functional studies on an iron-sulfur flavoproteins by protein engineering" (in Italian).

1991 Degree in Biological Sciences, University of Milan, Italy, Thesis title: "Role of the cysteine 272 in the catalytic mechanism of ferredoxin-NADP+ reductase" (in Italian).

Since 1993 Member of the Italian Society of Biochemistry and Molecular Biology

Research interests

The scientific career of Luciano Piubelli focused, as a general topic, on the structure-function relationships in proteins involved in

oxidoreductive reactions, such as iron-sulfur proteins (ferredoxin) and FAD-dependent enzymatic proteins, both reductases (ferredoxin-NADP⁺ reductase), oxidases (D-amino acid oxidase, cholesterol oxidase), or complex iron-sulfur flavoproteins (glutamate synthase). All these proteins were produced by means of protein engineering techniques (heterologous overexpression in *E. coli* and site-directed mutagenesis).

The functional and structural properties of these proteins have been investigated by a variety of multidisciplinary methodological approaches, including kinetic studies (steady state and pre-steady state kinetics), reaction mechanism studies, structural studies (limited proteolysis), flavin reactivity, protein stability on heating and denaturing agents, analysis of the redox properties.

In the recent years, the scientific activity on Luciano Piubelli focused on two FAD-dependent oxidases, namely D-amino acid oxidase (DAAO) and cholesterol oxidase (CO). He investigated the relationship between the dimeric oligomerisation state of yeast DAAO and its structural (stability and folding) and functional (activity) properties, together with the role for the reactivity of the enzyme of specific amino acidic residues belonging to the active site.

More recently, he started the functional characterisation of CO, by production and characterisation of single point mutant forms of the enzyme, produced by a rational mutagenesis approach, on the basis of the data from 3D structure, involving amino acidic residues putatively important for the functionality of the enzyme.

Teaching experience and appointments

Teaching activities:

Biochemistry

Applied Biochemistry and Biochemical Techniques

Laboratory of Biochemistry (Practical Activities)

Representative publications

1. Pelanda, R., Vanoni, M.A., Perego, M., Piubelli, L., Galizzi, A., Curti, B. and Zanetti, G. (1993) The glutamate synthase genes of the diazotroph *Azospirillum brasilense*: cloning, sequencing, and analysis of functional domains. *J. Biol. Chem.* 268, 3099-3106.
2. Aliverti, A., Piubelli, L., Zanetti, G., Lubberstedt, T., Herrmann, R.G. and Curti, B. (1993) The role of cysteine residues of spinach ferredoxin:NADP⁺ reductase as assessed by site-directed mutagenesis. *Biochemistry* 32, 6374-6380.
3. Piubelli, L., Aliverti, A., Bellintani, F. and Zanetti, G. (1995) Spinach ferredoxin I: overproduction in *Escherichia coli* and purification. *Protein Expression and Purification*, 6, 298-304.
4. Piubelli, L., Aliverti, A., Bellintani, F. and Zanetti, G. (1996) Mutation of Glu92 in ferredoxin I from spinach leaves produce protein fully functional in electron transfer but less efficient in supporting NADP⁺ photoreduction. *Eur. J. Biochem.*, 236, 465-469.
5. Piubelli, L., Zanetti, G. and Bosshard, H.R. (1997) Recombinant wild-type and mutant complexes of ferredoxin and ferredoxin:NADP⁺

reductase studied by isothermal titration calorimetry. *Biological Chemistry*, 378, 715-718.

6. Aliverti, A., Livraghi, A., Piubelli, L., and Zanetti, G. (1997) On the role of the acidic cluster Glu 92-94 of spinach ferredoxin I. *Biochem. Biophys. Acta*, 1342, 45-50.

7. Berger, C., Piubelli, L., Haditsch, U. and Bosshard, H.R. (1998) Diffusion-controlled DNA recognition by an unfolded, monomeric bZIP transcription factor. *FEBS Lett.*, 425, 14-18.

8. Aliverti, A., Deng, Z., Ravasi, D., Piubelli, L., Karplus, P.A. and Zanetti, G. (1998) Probing the function of the invariant glutamyl residue 312 in spinach ferredoxin-NADP⁺ reductase. *J. Biol. Chem.*, 273, 34008-34015.

9. Piubelli, L., Aliverti, A., Arakaki, A.K., Carrillo, N., Ceccarelli, E., Karplus, P.A. and Zanetti, G. (2000) Competition between C-terminal tyrosine and nicotinamide modulates pyridine nucleotide affinity and specificity in plant ferredoxin-NADP⁺ reductase. *J. Biol. Chem.*, 275, 10472-10476.

10. Piubelli, L., Caldinelli, L., Molla, G., Pilone, M.S. and Pollegioni, L. (2002) Conversion of the dimeric D-amino acid oxidase from *Rhodotorula gracilis* to a monomeric form. A rational mutagenesis approach. *FEBS Letts.*, 526, 43-48.

11. Pollegioni, L., Iametti, S., Fessas, D., Caldinelli, L., Piubelli, L., Barbiroli, A., Pilone, M.S. and Bonomi, F. (2003) Contribution of the dimeric state to the thermal stability of the flavoprotein D-amino acid oxidase. *Prot. Sci.*, 12, 1018-1029.

12. Molla, G., Motteran, L., Piubelli, L., Pilone, M.S. and Pollegioni, L. (2003) Regulation of D-amino acid oxidase expression in the yeast *Rhodotorula gracilis*. *Yeast*, 20, 1061-1069.

13. Piubelli, L., Molla, G., Caldinelli, L., Pilone, M.S. and Pollegioni, L. (2003) Dissection of the structural determinants involved in formation of the dimeric form of D-amino acid oxidase from *Rhodotorula gracilis*: role of the size of the F5-F6 loop. *Prot. Eng.*, 16, 1063-1069.

14. Caldinelli, L., Iametti, S., Barbiroli, A., Bonomi, F., Piubelli, L., Ferranti, P., Picariello, G., Pilone, M.S. and Pollegioni, L. (2004) Unfolding intermediate in the peroxisomal flavoprotein D-amino acid oxidase. *J. Biol. Chem.*, 279, 28426-28434.

15. Boselli, A., Piubelli, L., Molla, G., Sacchi, S., Pilone, M.S., Ghisla, S. and Pollegioni, L. (2004) On the mechanism of *Rhodotorula gracilis* D-amino acid oxidase: role of the active site serine 335. *Biochim. Biophys. Acta.*, 1702, 19-32.