MINISTERIO DE CIENCIA E INNOVACIÓN

Cofinanciado por la Unión Europea

AGENCIA ESTATAL DE INVESTIGACIÓN

CV date	10/01/2022

Part A. PERSONAL INFORMATION

First name	Roberto		
Family name	Fernández Lafuente		
Gender (*)	Male	Birth date	21/11/1964
ID number	50811609D		
e-mail	rfl@icp.csic.es		
Open Researcher and Contributor ID (ORCID)		0000-0003-4976-7096	

A.1. Current position

Position	Research Professor		
Initial date	7/21/2008		
Institution	CSIC		
Department/Center	ICP (Biocatalysis)		
Country	Spain	Teleph. number	+34915854804
Key words	Design of biocatalysts and biosensors, biomacromolecules immobilization, enzyme stabilization, protein purification, bioprocess design		

A.3. Education

PhD, Licensed, Graduate	University/Country	Year
Licensed	UCM	1987
PhD	UAM	1992

Part B. CV SUMMARY

My entire scientific career has been focused in the area of design of biocatalysts, biosensors, and bioprocess. Since 2009, I am the leader of the group “Optimization of biocatalysts and bioprocess” at ICP-CSIC. The main original contributions in the area of biomacromolecules immobilization are summarized below:

1- New protocols to achieve multipoint covalent immobilization of enzymes (e.g., vinyl sulfone activated supports, new strategies to use old supports such as glutaraldehyde, improved supports to immobilize lipases).

2- New heterofunctional supports for the immobilization, purification and stabilization of lipases: acyl-glyoxyl or acyl-vinyl sulfone.

3- Preparation of more porous and active crosslinked magnetic enzyme aggregates.

4- Integration of immobilization and chemical or physical modification of enzymes in the design of biocatalysts (e.g., preventing the release of reversibly immobilized enzymes).

5- New strategies for tuning specificity, selectivity and activity of lipases by its immobilization (e.g., changes in the immobilization conditions).

6- Use of core-shell supports for the immobilization of enzymes

7- Correlation of functional alterations after enzyme immobilization with conformational alterations.

8- Study of the enzyme distribution inside the biocatalyst particle.

9- New biosensors in collaboration with the Company BIOTICA, which commercializes a biosensor versus *Legionella* prepared following our protocols.

10- Determination of the enzyme crowding effects on immobilized enzyme biocatalysts features.

11- Tuning of enzyme features by altering the supports surface: effect of these interactions on the final enzyme performance and in the enzyme inactivation path-way.

12- The enzyme immobilization protocol determines the intensity and sense of the effect of some stabilizing or destabilizing agents.

13- Design of simple protocols to produce multilayers of immobilized enzymes, ensuring the spatial ordering.
14- Our group has detected some usually ignored problems in the design of coimmobilized enzyme biocatalysts, and in the previous project has proposed some solutions for these problems.
In the area of bioprocess design:
1- Production of esters with aroma or flavor interest via esterification. The biocatalysts design has been a key in these reactions, as water accumulation in the biocatalyst particle becomes a problem in the design of these processes. The use of very hydrophobic supports or ultrasounds reduces its negative impact.
2- Production of biodiesel. We have produced a new biocatalyst that have allowed reaching reaction rates in the same order of magnitude than with alkaline catalysis and even better yields from used cooking oils.
3- Definition of the advantages of the use of mixtures of enzymes: combi-enzymes. In many instances, and notably using substrates composed of many different components or multifunctional molecules, the best solution is to mix several enzymes to get the full substrate modified.
4- Production of biolubricants using lipases.
5- We have developed some bioprocesses of juice clarification using immobilized enzymes.
6.- Production of 1,2 diacetin by regioselective hydrolysis of triacetin and its transformation via chemoenzymatic routes.
7.- Development of the concept of combi-enzymes.
I am (co)author of over 500 papers in ISI journals, more than 235 since 2011. The total citations (Source data: **scopus**) are over 33,500 (around 13,800 for papers published since 2011). My global H index is 86, with an H index for the papers published since 2011 of 55. I am (co)inventor of 20 patents. In Google scholar, my H index is 98.
The number of citations received by my papers is growing steadily, from 1371 in 2011 to 4,328 in 2021, at 1-10-2021. Considering only the citations in 2021, I have 9 papers with more than 70 citations, 5 of them with more than 100 citations. I have been recognized as the most influent worldwide author in the area of enzyme immobilization in several recent bibliometric publications (e.g., DOI: 10.1016/j.procbio.2018.09.016). In another publication (DOI: 10.1371/journal.pbio.3000918), I have been placed in the 16th position among the most cited authors in biotechnology worldwide, the first one in Spain. Using total citations, I have 64 papers with more than 100 citations, and 181 publications with 50 or more citations. This growing impact of my publications has positioned me within the lists of **Highly Cited Researchers (HCR) 2019, 2020 and 2021.** These bibliometric data clearly show the international impact of our research. I have co-supervised 8 doctoral theses since 2011, out of a total of 22.

Part C. RELEVANT MERITS *(sorted by typology)*

C.1. Publications (2011-2022)

10 Some Research papers

1.- Morellon-Sterling, R., Carballares, D., Arana-Peña, S., Siar, E.-H., Braham, S.A., Fernandez-Lafuente, R. Advantages of Supports Activated with Divinyl Sulfone in Enzyme Coimmobilization: Possibility of Multipoint Covalent Immobilization of the Most Stable Enzyme and Immobilization via Ion Exchange of the Least Stable Enzyme (2021) ACS Sustainable Chemistry and Engineering, 9, 7508-7518. **Cited 2 time**. DOI: 10.1021/acssuschemeng.1c01065
2.-Arana-Peña, S., Rios, N.S., Carballares, D., Gonçalves, L.R.B., Fernandez-Lafuente, R. Immobilization of lipases via interfacial activation on hydrophobic supports: Production of biocatalysts libraries by altering the immobilization conditions (2021) Catalysis Today, 362, 130-140. **Cited 24 times**. DOI: 10.1016/j.cattod.2020.03.059

3.-Arana-Peña, S., Rios, N.S., Carballares, D., Mendez-Sanchez, C., Lokha, Y., Gonçalves, L.R.B., Fernandez-Lafuente, R. Effects of Enzyme Loading and Immobilization Conditions on the Catalytic Features of Lipase From Pseudomonas fluorescens Immobilized on Octyl-Agarose Beads (2020) Frontiers in Bioengineering and Biotechnology, 8, art. no. 36, . **Cited 31 times**. DOI: 10.3389/fbioe.2020.00036
4.-Arana-Peña, S., Rios, N.S., Mendez-Sanchez, C., Lokha, Y., Carballares, D., Gonçalves, L.R.B., Fernandez-Lafuente, R. Coimmobilization of different lipases: Simple layer by layer enzyme spatial ordering (2020) International Journal of Biological Macromolecules, 145, 856-864. **Cited 20 times.** DOI: 10.1016/j.ijbiomac.2019.10.087
5.- Arana-Peña, S., Mendez-Sanchez, C., Rios, N.S., Ortiz, C., Gonçalves, L.R.B., Fernandez-Lafuente, R. New applications of glyoxyl-octyl agarose in lipases co-immobilization: Strategies to reuse the most stable lipase (2019) International Journal of Biological Macromolecules, 131, 989-997. **Cited 46 times**. DOI: 10.1016/j.ijbiomac.2019.03.163
6.- Fernandez-Lopez, L., Pedrero, S.G., Lopez-Carrobles, N., Gorines, B.C., Virgen-Ortíz, J.J., Fernandez-Lafuente, R. Effect of protein load on stability of immobilized enzymes (2017) Enzyme and Microbial Technology, 98, 18-25. **Cited 137 times**. DOI: 10.1016/j.enzmictec.2016.12.002
7.- Sanchez, A., Cruz, J., Rueda, N., Dos Santos, J.C.S., Torres, R., Ortiz, C., Villalonga, R., Fernandez-Lafuente, R. Inactivation of immobilized trypsin under dissimilar conditions produces trypsin molecules with different structures (2016) RSC Advances, 6, 27329-27334. **Cited 93 times.** DOI: 10.1039/c6ra03627a
8.- Manoel, E.A., dos Santos, J.C.S., Freire, D.M.G., Rueda, N., Fernandez-Lafuente, R. Immobilization of lipases on hydrophobic supports involves the open form of the enzyme (2015) Enzyme and Microbial Technology, 71, 53-57. **Cited 340 times**. DOI: 10.1016/j.enzmictec.2015.02.001
9.- Dos Santos, J.C.S., Rueda, N., Barbosa, O., …. Fernandez-Lafuente, R. Characterization of supports activated with divinyl sulfone as a tool to immobilize and stabilize enzymes via multipoint covalent attachment. Application to chymotrypsin (2015) RSC Advances, 5 (27), 20639-20649. **Cited 80 times.** DOI: 10.1039/c4ra16926c
10.- Rueda, N., Dos Santos, J.C.S., Torres, R., Ortiz, C., Barbosa, O., Fernandez-Lafuente, R. Improved performance of lipases immobilized on heterofunctional octyl-glyoxyl agarose beads (2015) RSC Advances, 5, 11212-11222. **Cited 106 times**. DOI: 10.1039/c4ra13338b

Some review papers

1.- Arana-Peña, S., Carballares, D., Morellon-Sterlling, R., Berenguer-Murcia, Á., Alcántara, A.R., Rodrigues, R.C., Fernandez-Lafuente, R. Enzyme co-immobilization: Always the biocatalyst designers' choice…or not? (2021) Biotechnology Advances, 51, art. no. 107584, . **Cited 48 times.** DOI: 10.1016/j.biotechadv.2020.107584

2.- Rodrigues, R.C., Virgen-Ortíz, J.J., dos Santos, J.C.S., Berenguer-Murcia, Á., Alcantara, A.R., Barbosa, O., Ortiz, C., Fernandez-Lafuente, R. Immobilization of lipases on hydrophobic supports: immobilization mechanism, advantages, problems, and solutions (2019) Biotechnology Advances, 37 (5), pp. 746-770**. Cited 215 times**. DOI: 10.1016/j.biotechadv.2019.04.003
3.-Cipolatti, E.P., Valério, A., Henriques, R.O., Moritz, D.E., Ninow, J.L., Freire, D.M.G., Manoel, E.A., Fernandez-Lafuente, R., De Oliveira, D. Nanomaterials for biocatalyst immobilization-state of the art and future trends (2016) RSC Advances, 6 (106), pp. 104675-104692. **Cited 204 times**. DOI: 10.1039/c6ra22047a
4.- Rodrigues, R.C., Ortiz, C., Berenguer-Murcia, Á., Torres, R., Fernández-Lafuente, R. Modifying enzyme activity and selectivity by immobilization (2013) Chemical Society Reviews, 42 (15), pp. 6290-6307. **Cited 1261 times**. DOI: 10.1039/c2cs35231a
5.- Garcia-Galan, C., Berenguer-Murcia, A., Fernandez-Lafuente, R., Rodrigues, R.C. Potential of different enzyme immobilization strategies to improve enzyme performance (2011) Advanced Synthesis and Catalysis, 353 (16), pp. 2885-2904. **Cited 1142 times**. DOI: 10.1002/adsc.201100534