BIOGRAPHICAL SKETCH

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NAME: Rosa Bacchetta

eRA COMMONS USER NAME (credential, e.g., agency login): RBACCHETTA

POSITION TITLE: Associate Professor

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, include postdoctoral training and residency training if applicable. Add/delete rows as necessary.)*

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	Completion Date MM/YYYY	FIELD OF STUDY
Medical School, University of Turin, Italy	M.D	07/1987	Medicine
Residency in Pediatric, University of Turin, Italy	Natl. Board	11/1991	General Pediatrics, Immunology

A. Personal Statement

I have long-standing experience in investigating the mechanisms of immunological tolerance, specifically in pediatric patients after hematopoietic stem cell transplantation and in patients with genetic diseases of the immune system. My basic research training in immunology of human diseases, together with my involvement in clinical diagnosis and development of preclinical studies for cellular and gene therapy trials of immune mediated diseases, has strengthened my competence in the field of Translational Medicine. From this work, I have developed extensive expertise in investigating the mechanisms of immunological tolerance and its modulation for clinical applications. I completed the first cellular therapy trial of alloantigen- specific regulatory Type 1 T lymphocytes (Tr1) generated to treat patients transplanted with haploidentical stem cell transplantations to prevent acute graft-versus-host disease. My contribution to the clinical trial extended from the initial observation, that Tr1 lymphocytes are naturally present in tolerant hematopoietic stem cell (HSC)-transplant recipients to the establishment of the in vitro methods to generate the Tr1 lymphocytes, the scale-up and the development of a GMP validated protocol filed and ultimately the clinical trial that was completed in 2009. This work contributed significantly to the development of the current FDA approved (IND# 17292) optimized T-allo10 protocol using purified CD4+ T cells as responder cells and DC-10 as stimulator cells. Over the years, my research has focused on Autoimmune Genetic Diseases and Primary Immunodeficiencies, from identifying causative genes, understanding the pathogenesis, defining novel diagnostic and prognostic markers and investigating new therapeutic approaches. My work has significantly contributed, in the past twenty years, to dissecting the role of FOXP3 and Treg cells in immune responses in humans, as well as to raising the interest of scientists and clinicians in investigating diseases with severe immune dysregulation, such as IPEX Syndrome. At Stanford, I will continue pursuing research on dissecting the immunological mechanisms underlying monogenic autoimmune diseases, especially those with onset in childhood and with progressive course. Importantly, my goal is to establish cell and gene transfer-based therapies for IPEX Syndrome and similar monogenic diseases with immune dysregulation. I have pioneered the application of CRISPR/Cas9 gene editing to restore FOXP3 in IPEX patient HSCs. The goal of this R01 is to refine this gene editing strategy to restore physiological FOXP3 expression from autologous HSC and definitively improve the therapies of IPEX and other monogenic immune regulatory disorders.

B. Positions and Honors

05/15- Present	Associate Professor, Pediatric Division of Stem Cell Transplantation and Regenerative Medicine, Stanford University, School of Medicine
10/14- 10/16	Pediatrician (consultant), Pediatric Immunology Hematology, San Raffaele Hospital, Milan Italy
05/14-04/15	Senior Research Scientist, Pediatric Division of Stem Cell Transplantation and Regenerative Medicine, Stanford University, School of Medicine
01/14	Associate Professor in Pediatrics: Qualification awarded at National Level, based on scientific achievements, Italian Ministry of Health
08/12-04/14	Visiting Scholar and Instructor /Research Associate, Pediatric Division of Immunology and Allergy, Stanford University, School of Medicine
2007- 09/14	Group Leader at San Raffaele Telethon Institute for Gene Therapy (HSR-TIGET) Milan (Dir. Luigi Naldini)
2004-2012	Responsible for the Outpatients Clinic Pediatric Immunology Hematology Unit, San Raffaele Hospital (Dir. Maria Grazia Roncarolo)
2001-2006	Project Leader at San Raffaele Telethon Institute for Gene Therapy (HSR-TIGET) Milan (Dir. Maria Grazia Roncarolo)
1997-2000	Medical Researcher at Cellular Therapy Laboratory Telethon Institute for Gene Therapy (HSR-TIGET), Scientific Institute H.S. Raffaele, Milan (Dir. Claudio Bordignon)
1990-1995	Postdoctoral Fellow at DNAX Research Institute of Molecular and Cellular Biology, Palo Alto CA, USA (Dir. Jan E. de Vries)
1988-1989	Research Fellow at UNICET Laboratory for Immunological Research, Lyon, France (Dir. Jacques Banchereau)
1985-1987	Pre-doctoral Internship. Department of Pediatric Immunology. School of Medicine, University of Turin, Italy (Dir. Pierangelo Tovo)

Professional Memberships/Activities

Member of the European Society of Immunodeficiency (ESID), Federation of Clinical Immunology Societies (FOCIS and CIS) and Italian Society of Pediatric Oncology and Hematology (AIEOP)

Associate Editor for Frontiers in Pediatrics and Frontiers in Primary Immunodeficiencies, Open Access Journal

Reviewer activity for Blood, J Medical Genetics, J Autoimmunity, J Endocrinology, European Journal of Immunology, Transplantation.

Grants revision activities for European Research Council, INSERM, LSBR Foundation, Wellcome Trust, ANR-France

Teaching activities ad hoc lessons at Vita-Salute University Medical School and Biotechnology, Milan; Tor Vergata University, Rome; Undergraduate and Graduate Course at Stanford University

C. Contribution to Science

My first original work was on SCID patients successfully reconstituted with HLA-mismatched fetal liver hematopoietic stem cells. These studies allowed the first isolation in humans of IL-10–producing Type 1 regulatory T cells that represent a fundamental cell subset for the induction and maintenance of peripheral tolerance. I have also completed the **first cellular therapy trial of allo-antigen-specific regulatory Type 1 T lymphocytes (Tr1)** generated to treat patients transplanted with haploidentical stem cell transplantations to prevent acute graft-versus-host disease. My contribution to the clinical trial extended from the initial observation, that Tr1 lymphocytes are naturally present in tolerant hematopoietic stem cell (HSC)-transplant recipients to the establishment of the *in vitro* methods to generate the Tr1 lymphocytes, the scale-up and the development of a GMP validated protocol filed and approved by the Italian Ministry of Health (2000), and ultimately the clinical trial that started in 2001.

1. **Bacchetta R**., Bigler, M., Touraine, J.-L., Parkman, R., Tovo, P.-A., Abrams, J., De Waal Malefyt, R., De Vries, J.E., Roncarolo, M.G. High levels of interleukin 10 production in vivo are associated with tolerance in SCID patients transplanted with HLA mismatched hematopoietic stem cells. (1994) Journal of Experimental Medicine, 179 (2), pp. 493-502. IF 15.7
2. **Bacchetta R**., Parkman, R., McMahon, M., Weinberg, K., Bigler, M., De Vries, J.E., Roncarolo, M.G. Dysfunctional cytokine production by host-reactive T-cell clones isolated from a chimeric severe combined immunodeficiency patient transplanted with haploidentical bone marrow. (1995) Blood, 85 (7), pp. 1944-1953. IF 8.8
3. **Bacchetta R**., Vandekerckhove, B.A.E., Touraine, J.-L., Bigler, M., Martino, S., Gebuhrer, L., De Vries, J.E., Spits, H., Roncarolo, M.G. Chimerism and tolerance to host and donor in severe combined immunodeficiencies transplanted with fetal liver stem cells. (1993) Journal of Clinical Investigation, 91 (3), pp. 1067-1078. IF 10.9
4. **Bacchetta R**, Lucarelli B, Sartirana C, Gregori S, Lupo Stanghellini MT, Miqueu P, Tomiuk S, Hernandez-Fuentes M, Gianolini ME, Greco R, Bernardi M, Zappone E, Rossini S, Janssen U, Ambrosi A, Salomoni M, Peccatori J, Ciceri F, Roncarolo MG. Immunological Outcome in Haploidentical-HSC Transplanted Patients Treated with IL-10-Anergized Donor T Cells. *Front Immunol*. **2014** Jan 31;5:16. IF5.5

2. I extensively **studied the pathogenesis of Immune-dysregulation-Polyendocrinopathy-Enteropathy-X-linked (IPEX) Syndrome**. My work showed for the first time that in IPEX patients FOXP3 gene mutations resulted in the impaired function of the Treg lymphocytes. In addition, I demonstrated that, unlike in the mouse model of the disease, the FOXP3 abnormalities also affected the effector T (Teff) lymphocytes in humans, suggesting that the role of FOXP3 in the human immune response is not confined to the Treg lymphocytes. I further showed that in parallel to their defective function, Treg and Teff lymphocytes from IPEX patients are more prone to secrete IL-17, leading to an increased frequency of peripheral IL-17 producing T lymphocytes, known to mediate inflammation.

5. **Bacchetta R.,** Passerini, L., Gambineri, E., Dai, M., Allan, S.E., Perroni, L., Dagna-Bricarelli, F., Sartirana, C., Matthes-Martin, S., Lawitschka, A., Azzari, C., Ziegler, S.F., Levings, M.K., Roncarolo, M.G. Defective regulatory and effector T cell functions in patients with FOXP3 mutations. (2006) Journal of Clinical Investigation, 116 (6), pp. 1713-1722. IF 15.8
6. Allan, S.E., Alstad, A.N., Merindol, N., Crellin, N.K., Amendola, M., **Bacchetta R.,** Naldini, L., Roncarolo, M.G., Soudeyns, H., Levings, M.K. Generation of potent and stable human CD4+ T regulatory cells by activation-independent expression of FOXP3. (2008) Molecular Therapy, 16 (1), pp. 194-202. IF 6
7. Di Nunzio, S., Cecconi, M., Passerini, L., McMurchy, A.N., Baron, U., Turbachova, I., Vignola, S., Valencic, E., Tommasini, A., Junker, A., Cazzola, G., Olek, S., Levings, M.K., Perroni, L., Roncarolo, M.G., **Bacchetta R.*** Wild-type FOXP3 is selectively active in CD4+CD25hi regulatory T cells of healthy female carriers of different FOXP3 mutations. (2009) Blood, 114 (19), pp. 4138-4141. IF 10.6
8. Passerini L., Olek S., Di Nunzio S., Barzaghi F., Hambleton S., Abinun M., Tommasini A., Vignola S., Cipolli M., Amendola M., Naldini L., Guidi L., Cecconi M., Roncarolo MG., **Bacchetta R*.** *FOXP3* mutations lead to increased Th17 cells and regulatory T cell instability. (2011). J Allergy and Clinical Immunol. Dec;128(6):1376-1379. IF 9.3

3. Using a newly developed quantitative method to detect Treg lymphocytes in small amount of blood, I have identified a subset of IPEX-like patients with low numbers of Treg lymphocytes but preserved function. As well I have, for the first time, successfully converted IPEX T lymphocytes into functional T regulatory lymphocytes by lentiviral-mediated overexpression of wild type FOXP3 and demonstrated that ectopic FOXP3 expression preserves HSCs and impairs T cell differentiation. These results open the possibility of a new therapeutic approach to control autoimmunity not only in IPEX patients but also in other diseases with immune dysregulation.

9. Barzaghi F, Passerini L, Gambineri E, Ciullini Mannurita S, Cornu T, Kang ES, Choe YH, Cancrini C, Corrente S, Ciccocioppo R, Cecconi M, Zuin G, Discepolo V, Sartirana C, Schmidtko J, Ikinciogullari A, Ambrosi A, Roncarolo MG, Olek S, **Bacchetta R*.** Demethylation analysis of the FOXP3 locus shows quantitative defects of regulatory T cells in IPEX-like syndrome. (2012). J Autoimmun. Feb;38(1):49-58.
10. Passerini L, Rossi Mel E, Sartirana C, Fousteri G, Bondanza A, Naldini L, Roncarolo MG, **Bacchetta R *.** $CD4_+$ T cells from IPEX patients convert into functional and stable regulatory T cells by *FOXP3* gene transfer. Science Translational Medicine 2013, Dec 11, 5 (215). IF 10.7
11. Santoni de Sio FR, Passerini L, Valente MM, Russo F, Naldini L, Roncarolo MG, **Bacchetta R*.**

Ectopic FOXP3 Expression Preserves Primitive Features Of Human Hematopoietic Stem Cells While Impairing Functional T Cell Differentiation. Sci Rep. 2017 Nov 17;7(1):15820.

12. Santoni de Sio FR, Passerini L, Restelli S, Valente MM, Pramov A, Maccari ME, Sanvito F, Roncarolo MG, Porteus M, **Bacchetta R***. Role of human forkhead box P3 in early thymic maturation and peripheral T-cell homeostasis. *J Allergy Clin Immunol*. **2018** Apr 27: 1909-1921.e9.

4. I also contributed to a comprehensive **clinical characterization of IPEX patients including the largest long term follow up, and to defining of new diagnostic tools**, complementary to gene sequencing, which have been useful for the follow-up of IPEX patients undergoing different treatments. I achieved the clinical characterization by establishing collaborations with other centers and gathering clinical data from IPEX patients with different mutations and disease severity.

13. Lampasona V, Passerini L, Barzaghi F, Lombardoni C, Bazzigaluppi E, Brigatti C, **Bacchetta R*,** Bosi E*. Autoantibodies to Harmonin and Villin Are Diagnostic Markers in Children with IPEX Syndrome. PlosOne, 2013, Nov 8 (11). IF 3.7
14. Goudy K, Aydin D, Barzaghi F, Gambineri E, Vignoli M, Mannurita SC, Doglioni C, Ponzoni M, Cicalese MP, Assanelli A, Tommasini A, Brigida I, Dellepiane RM, Martino S, Olek S, Aiuti A, Ciceri F, Roncarolo MG, **Bacchetta R*.** Human IL2RA null mutation mediates immunodeficiency with lymphoproliferation and autoimmunity. Clin Immunol. 2013 Jan 24;146(3):248-261. IF 3.7
15. Barzaghi F, Amaya Hernandez LC, Neven B, et al. and **Bacchetta R***; Primary Immune Deficiency Treatment Consortium (PIDTC) and the Inborn Errors Working Party (IEWP) of the European Society for Blood and Marrow Transplantation (EBMT). Long-term follow-up of IPEX syndrome patients after different therapeutic strategies: An international multicenter retrospective study. J Allergy Clin Immunol. 2017 Dec 11. pii: S0091-6749(17)31893-6.
16. Passerini L, Barzaghi F, et al. and Bacchetta R*. Treatment with rapamycin can restore regulatory T-cell function in IPEX patients. J Allergy Clin Immunol. 2019 Dec 23, pub on line.

5. Recently, I contributed to innovative *in vitro* and *in vivo* approaches, instrumental to study human genetic diseases of the immune system and preparatory to **gene therapy via gene editing**.

17. Hendel A, Bak RO, Clark JT, Kennedy AB, Ryan DE, Roy S, Steinfeld I, Lunstad BD, Kaiser RJ, Wilkens AB, **Bacchetta R**, Tsalenko A, Dellinger D, Bruhn L, Porteus MH. Chemically modified guide RNAs enhance CRISPR-Cas genome editing in human primary cells. Nat Biotechnol. 2015 Jun 29. [Epub ahead of print]
18. Goettel JA, Biswas S, Lexmond WS, Yeste A, Passerini L, Patel B, Yang S, Sun J, Ouahed J, Shouval DS, McCann KJ, Horwitz BH, Mathis D, Milford EL, Notarangelo LD, Roncarolo MG, Fiebiger E, Marasco WA, **Bacchetta R**, Quintana FJ, Pai SY, Klein C, Muise AM, Snapper SB. Fatal autoimmunity in mice reconstituted with human hematopoietic stem cells encoding defective FOXP3. Blood. 2015 Jun 18;125(25):3886-95.
19. Goodwin M., Lee E. and **Bacchetta R*.** CRISPR-based gene editing enables FOXP3 gene repair in IPEX patient cells. *Sci Adv, in press*

Complete List of Published Work in MyBibliography:
https://www.ncbi.nlm.nih.gov/pubmed/?term=bacchetta+r+and+immune

D. Additional Information: Research Support and/or Scholastic Performance
Ongoing Research Support (as PI):

MCHRI/Harman Faculty Scholar Award 9/2019-8/2024
Title:" Harnessing FOXP3 genome editing towards innovative treatment of IPEX syndrome"
Genome editing for FOXP3 project, mainly supporting Faculty salary.

California Institute of Regenerative Medicine (CIRM)-CLIN1 12/2019-6/2021
Title: "Phase 1 Study of Autologous $CD4_{LVFOXP3}$ T Cells in Subjects with IPEX Syndrome".
Supporting IND enabling studies for LV-engineered Treg-like cells.

Harrington Discovery Institute-Innovator Award 01/01/2019 – 01/01/2021

Preclinical safety, dose evaluation, pharmacokinetics and efficacy of Treg-like CD4+T cells genetically modified to express *FOXP3* for the treatment of IPEX syndrome patients.

The goal of this study is to complete the preclinical studies and initiate the process development for the clinical use of the converted LV-FOXP3 T cells to test safety and efficacy in IPEX patients in a Phase 1 trial.

Completed Research Support

R21 AI12389601A1 NIH Bacchetta (PI) 09/01/2016 – 08/31/2018
Genome Editing to rescue FOXP3 Deficiency in IPEX
The goal of this study is to obtain efficient *FOXP3* gene-disruption in Treg cells and HSCs to generate a hu-mouse model of *FOXP3* deficiency and establish functional *FOXP3* gene correction using the CRISPR/Cas9 system in IPEX Treg cells and HSCs.

CIRM QUEST Discovery Award 05/01/2017- 04/30/2019
Genome Editing to rescue FOXP3 Deficiency in IPEX
perform the preclinical studies required for the development of a cure for IPEX patients using a highly efficient genome editing approach to repair autologous patients' HSCs.

Spectrum Pilot Grants for SPARK (Therapeutics) 03/01/2017-02/28/2019
Gene Corrected Autologous HSCs for the Treatment of IPEX Syndrome
Provide a foundation for future external funding applications to develop clinical applications of our FOXP3 gene correction strategy in IPEX patient cells.

Spectrum Pilot Grants for SPARK (Diagnostics) 03/01/2017-02/28/2019
Epigenetic screening for primary immune deficiencies and immune dysregulation
Develop a more sensitive diagnostic tool for newborn screening as well as later diagnostic timepoints to detect essentially all primary immune deficiencies (PIDs).