

STAFF SCIENTIST

Summary

Research Biologist with more than 10 years in laboratory research. Accomplished in generating and applying new protocols and technologies.

Highlights

- Computer skills
- Used FACSDiva, FlowJo and CellQuest to analyze the flow cytometric (FACS) data.
- Used GraphPad Prism for statistical analysis of the experimental data.
- Used MacVector and Vector NTI to analyze DNA, RNA and protein sequencing data.

DNA isolation expert

Microbiological techniques

Knowledgeable about subcloning

Western blotting

Experimental design

Nucleic Acids Isolation

Immunofluorescence assay expertise

Accomplishments

- B.Sc. (Zoology Honors) Delhi University, Delhi, India 1989
- 1. Early Career Faculty Travel Award, AAI (American Association of Immunologists) 2013
- 2. Elected Fellow, AAAAI (American Academy of Allergy, Asthma and Immunology) 2010
- 3. Eleanor and Miles Shore Faculty Career Development Award, Harvard Medical School 2009
- 4. Eleanor and Miles Shore Faculty Career Development Award, Harvard Medical School 2008
- 6. Travel Award, FCE (Federation of Clinical Immunology Societies-Centers of Excellence) 2004
- 7. Travel Award, FOCIS (Federation of Clinical Immunology Societies) 2002
- 8. Elected Life Member, ICAAI (Indian College of Allergy and Applied Immunology) 1996
- 9. Elected Affiliate Member, EAACI (European Academy of Allergology and Clinical Immunology)
- 10. Young Scientist Award, IUBMB (International Congress of Biochemistry and Molecular Biology)
- 11. International Travel Award, IAACI (International Association of Allergology and Clinical Immunology)
- Eleanor and Miles Shore Faculty Career Development Award Principal Investigator (2008-2013)
- 1. Early Career Faculty Travel Award, AAI (American Association of Immunologists) 2013 2. Elected Fellow, AAAAI (American Academy of Allergy, Asthma and Immunology) 2010 3. Eleanor and Miles Shore Faculty Career Development Award, Harvard Medical School 2009 4. Eleanor and Miles Shore Faculty Career Development Award, Harvard Medical School 2008 5. Invited Speaker, IUIS/WHO (International Union of Immunological Societies/World Health Organization) Symposium of Primary Immunodeficiency Diseases. Jackson Hole, Wyoming 2007 6. Travel Award, FCE (Federation of Clinical Immunology Societies-Centers of Excellence) 2004 7. Travel Award, FOCIS (Federation of Clinical Immunology Societies) 2002 8. Elected Life Member, ICAAI (Indian College of Allergy and Applied Immunology) 1996 9. Elected Affiliate Member, EAACI (European Academy of Allergology and Clinical Immunology) 1995 10. Young Scientist Award, IUBMB (International Congress of Biochemistry and Molecular Biology) 1994 11. International Travel Award, IAACI (International Association of Allergology and Clinical Immunology) 12. Senior Research Fellowship, CSIR (Council of Scientific and Industrial Research), Govt. of India, India 1993 13. Junior Research Fellowship, CSIR (Council of Scientific and Industrial Research), Govt. of India, 3 Lalit Kumar India 1991 14. Qualified NET (National Eligibility Test) for Research Fellowship & Lecturership, University Grant Commission, Govt. of India, India 1991 15. Ranked among top three students, M.Sc. Biotechnology Program, Goa University, India 1991 16. Studentship, DBT (Department of Biotechnology), Govt. of India, India 1989

Experience

Staff Scientist 07/2004 to 06/2014

Thermo Fisher Scientific Inc. Stanford, CA

- My research at the Boston Children's Hospital was focused on studying molecular and genetic mechanisms underlying human immunological, inflammatory and autoimmune diseases. It involved scientific as well as management components. Science
- Identified target gene(s) involved in human primary immunodeficiencies (PIDs), inflammatory and autoimmune diseases and the modulation of immune responses.
- Validated target gene(s) by designing, generation and phenotypic evaluation of global and tissue specific transgenic and knockout mouse models. This involved actively designing and successful execution of in vivo, ex vivo and in vitro experiments to assess the role of target gene under investigation.
- Target genes studied and published: Lrrc8a, Slp-76, Wip, 3bp2, Cip-4, C3a, Tslp, Il-21r and Tlr-2. Management
- Led immunology research activities to achieve program and project-level goals.

- Planned and effectively communicated research goals and objective to junior scientific staff for the timely completion of research projects.
- Designed, successfully performed and troubleshoot experiments, interpreted data, and suggested next steps.
- Presented data in-house and at various national and international scientific meetings in a variety of formats (oral, poster and panel discussion).
- Contributed to the writing of SOPs, manuscripts and research grant proposals.
- Managed day-to-day functioning of the laboratory.
- Trained, supervised and mentored junior associates (research assistants, post-doctoral and clinical research fellows).
- Introduced latest concepts/technologies such as PLA (Proximity Ligation Assay) and CRISPR- CAS to research programs in order to achieve research goals and objectives more efficiently.
- Managed interactions with internal and external scientific collaborators in academia as well as biotechnology and contract research organizations. The above work led to three successful grants from the NIH and private agencies, the publication of 15 manuscripts in high impact journals, and training of several post-doctoral and junior technical staff.

Post-doctoral fellow 03/1998 to 06/2004
City Of Hope Torrance , CA

- Led research projects aimed at the investigation of structure-function relationship of different signaling domains of adaptor proteins SLP-76 in the development and function of T cells and bone marrow derived mast cells (BMMCs). Successfully achieved the objectives of these projects by:
- Designing and generating several different SLP-76 DNA expression constructs for the retroviral reconstitution of SLP-76 deficient BMMCs (derived from SLP-76 knockout mice). 1
- Designing and generating a series of tissue specific wild type and different mutant SLP-76 transgenic mouse models and breeding them in to the SLP-76 knockout background.
- In-depth phenotypic characterization of the reconstituted BMMCs and transgenic mouse models. The data generated in these studies resulted in one successful R01 grant from NIAID, NIH (for my mentor) and the publication of 3 manuscripts in high impact journals.
- LABORATORY TECHNIQUES AND SKILLS Generation, evaluation and use of specific antibodies
- Designed target immunogenic peptides and prepared bacterial and mammalian cell expressed purified fusion proteins as immunogens, immunized mice and rabbits with immunogens for specific antibody response.
- Evaluated antibody response by ELISA, Dot blot and Western blot assays.
- Labeled purified specific antibodies with Biotin, FITC and PE to visualize antigen expression by immunofluorescence confocal microscopy and multi-color flow cytometric assays (by using BD LSR/Canto and LSR/Fortessa). Generation and evaluation of in vivo mouse models of human diseases
- Designed, generated and phenotypically characterized transgenic SLP-76 mouse models to study the role of different domains of SLP-76 in human T cell development and function.
- Designed, generated and phenotypically characterized LRRC8A knockout (regular and tissue specific conditional) mouse model to study its contribution in the development and function of hematopoietic and non-hematopoietic systems.
- Established cell intrinsic role of LRRC8A in T cells development and function by generating and phenotypically characterizing *Lrrc8a*^{-/-} hematopoietic stem cell (HSC) transplanted Rag2^{-/-} radiation chimera mice.
- Studied the role of SLP-76, 3BP2, CIP4 and LRRC8A in in vivo T cell function by eliciting and evaluating Th1 and Th2 specific hapten mediated contact hypersensitivity/delayed type hypersensitivity (CHS/DTH) in *Slp-76*^{-/-}, *3bp2*^{-/-}, *Cip4*^{-/-} and *Lrrc8a*^{-/-} mice, respectively.
- Investigated the role of TSLP, IL-21R and TLR-2 in allergic skin disease by developing OVA immunized mouse models of inflammatory allergic skin disease (atopic dermatitis) in *Tslp*^{-/-}, *Il-21r*^{-/-} and *Tlr-2*^{-/-} mice, respectively.
- Investigated the role of C3a in eczema vaccinatum, a rare but severe adverse reaction to smallpox vaccination, by establishing vaccinia virus infection in *C3a*^{-/-} mouse model of atopic dermatitis. Also, acquired extensive experience in working under BSL2+ safety conditions. Mammalian cell culture and cell-based (in vivo, ex vivo and in vitro) functional assays
- Generated embryonic fibroblasts and bone marrow derived mast cells from wild type and *Lrrc8a*^{-/-} mice.
- Purified T and B cells from human and mouse by Miltenyi AutoMACS and FACS based cell sorting for cell signaling experiments.
- Unraveled the role of SLP-76 and LRRC8A in T cell, B cell, Dendritic cell and NK cell function by evaluating Th-Differentiation, Cell Proliferation (by 3H incorporation and CFSE dye dilution methods), Cytokine Production (by FACS, ELISA and RT-PCR), Activation Markers Expression (by FACS), Apoptosis (by FACS, TUNEL and Immunohistochemistry), Cell Cycle Progression (by FACS), and Cell Mediated Cytotoxicity (by 51Cr release assay). 2 Molecular biology, biochemical characterization and signal transduction techniques
- Quantitatively evaluated gene expression profile in skin tissues, derived under regular and inflammatory experimental conditions, by real time quantitative PCR (for cytokines) and microarray (for global gene expression) analysis.
- Established PCR and Southern blot based genotype-screening protocols for SLP-76 transgenic and LRRC8A regular and conditional knockout mice.
- Participated in a collaborated project aimed at studying the role of LRBA in T cells responses. Helped in designing and testing of several candidate siRNA for the study. Additionally, designed CRISPR based DNA constructs for creating *Lrba*^{-/-} mouse model.
- Cloned tagged and untagged LRRC8A cDNA in its full length and truncated mutant forms in appropriate expression vectors for their expression in bacterial and mammalian cells (293T and Jurkat).
- By using a series of pull-down and multi-color flow cytometry based ligand binding and competition functional assays, demonstrated that non-hematopoietic cells express LRRC8A ligand.
- By using a series of immunoprecipitation, SDS-PAGE and Western blot assays, demonstrated that LRRC8A exist as a trimer and oligomer of higher order and act as a cell surface receptor.

Ph.D. : Medical Biochemistry 1998 Delhi University City , State , IN Ph.D. (Medical Biochemistry) Delhi University, Delhi, India 1998

M.Sc. : Biotechnology 1991 Goa University M.Sc. (Biotechnology) Goa University, Goa, India 1991

B.Sc. : Zoology 1989 Delhi University City , State , IN B.Sc. (Zoology Honors) Delhi University, Delhi, India 1989

Certifications

CAS SLP RT AAI

Memberships and Affiliations

1. , AAAS (American Association for the Advancement of Science)

Publications

. . Molecular characterization of major allergen of typhoides pollen Ph.D. Thesis, Delhi University. receptor-mediated activation of basophils. *Int Immunol.* 24(11): 719- 27. (PMID: 22875843) 5. (2010). CI Koduru S, Kumar L, Massaad M, Ramesh N, Le Bras S, Ozcan E, Oyoshi M, Kaku M, Fujiwara Y, Kremer L, King is essential for integrin dependent T cell trafficking. 107(37): . (PMID: 20805498) 6. Oyoshi MK *, Elkhail A*, Kumar L*, HC, Murphy GF, Geha RS. (2009). Vaccinia virus inoculation in sites of allergic skin inflammation elicits a vigorous cutaneous IL-17 response. *Proc Natl Acad Sci USA.* 106(35): 14954-14959. (PMID: 19706451) (*Equal contribution) 7. Le Bras S, Massaad M, Koduru S, Kumar L, Oyoshi MK, Hartwig J, Geha RS. (2009). WIP is critical for T cell responsiveness to IL-2. *Proc Natl Acad Sci USA.* 106(18): 7519-7524. (PMID: 19359486) 8. Jin H*, Kumar L*, Mathias C, Zurakowski D, Oettgen H, Gorelik L, Geha R. (2009). Toll-like receptor 2 is important for the T(H)1 response to cutaneous sensitization. *J Allergy Clin Immunol.* 123(4): 875-882. (PMID: 19348925) (*Equal contribution) 9. Jin H, Oyoshi MK, Le Y, Bianchi T, Koduru S, Mathias CB, Kumar L, Le Bras S, Young D, Collins M, Grusby MJ, Wenzel J, Bieber T, Boes M, Silberstein LE, Oettgen HC, Geha RS. (2009). IL-21R is essential for epicutaneous sensitization and allergic skin inflammation in humans and mice. *J Clin Invest.* 119(1): 47-60. (PMID: 19075398) 10. He R, Oyoshi MK, Garibyan L, Kumar L, Ziegler SF, Geha RS. (2008). TSLP acts on infiltrating effector T cells to drive allergic skin inflammation. *Proc Natl Acad Sci USA.* 105(33): 11875-11880. (PMID: 18711124) 11. Hidano S, Sasanuma H, Ohshima K, Seino K, Kumar L, Hayashi K, Kurosaki T, Taniguchi M, Geha RS, Kitamura D, and Goitsuka R. (2008). Distinct Regulatory Functions of SLP-76 and MIST in NK Cell Cytotoxicity and IFN- γ Production. *Int Immunol.* 20(3): 345-352. (PMID: 18203684) 12. Koduru S, Massaad M, Wilbur C, Kumar L, Geha R, Ramesh N. (2007). A novel anti-WIP monoclonal antibody detects an isoform of WIP that lacks the WASP binding domain. *Biochem Biophys Res Commun.* 353(4): 875-881. (PMID: 17207458) 13. de la Fuente MA, Kumar L, Bao L, Geha RS. (2006). 3BP2 deficiency impairs the response of B cells, but not T cells, to antigen receptor ligation. *Mol Cell Biol.* 26(14): 5214-5225. (PMID: 16809760) 14. Kumar L, Feske S, Rao A, Geha RS. (2005). A ten amino acid long sequence in SLP-76 upstream of the Gads binding site is essential for T cell development and function. *Proc Natl Acad Sci USA.* 102(52): 19063-19068. (PMID: 16354835) 15. Kettner A*, Kumar L*, Anton IM, Sasahara Y, de la Fuente M, Pivniouk VI, Falet H, Hartwig JH, Geha RS. (2004). WIP regulates signaling via the high affinity receptor for immunoglobulin E in mast cells. *J Exp Med.* 199(3): 357-368. (PMID: 14757742) (*Equal contribution) 16. Kumar L, Pivniouk V, de la Fuente MA, Laouini D, Geha RS. (2002). Differential role of SLP-76 domains in T cell development and function. *Proc Natl Acad Sci USA.* 99(2): 884-889. (PMID: 11792851) 17. Kumar L, Sridhara S, Singh BP, Gangal SV. (1998). Characterization of cogon grass (*Imperata cylindrica*) pollen extract and preliminary analysis of grass group 1, 4 and Scott JE, Koduru S, He R, Leung DY, Howell MD, Oettgen homologues using monoclonal antibodies to *Phleum pratense*. 117(3): . (PMID: 9831804) 18. . Immuno-biochemical characterization of *Sorghum vulgare* pollen allergens prevalent in tropical countries. *Ind J Allergy Asthma Immunol.* 16: 33-39. 19. Sridhara S, Kumar L, Verma J, Singh BP, Gangal SV. (2000). Studies on *Cenchrus ciliaris* pollen extract and cross-reactivity among tropical grasses of the family Poaceae. *Allergy and Clin Immunol Int.* 12(3): 110-115. 20. Kumar L, Sridhara S, Singh BP, Gangal SV. (1998). Characterization of cogon grass (*Imperata cylindrica*) pollen extract and preliminary analysis of grass group 1, 4 and 5 homologues using monoclonal antibodies to *Phleum pratense*. 117(3): . (PMID: 9831804) 21. Sridhara S, Singh BP, Kumar L, Verma J, Gaur SN, Gangal SV. (1995). Antigenic and allergenic relationships among airborne grass pollens in India. *Ann Allergy Asthma Immunol.* 75(1): 73-79. (PMID: 7621065) Original research articles (submitted) 1. Yoon J, Wang G, Galand C, Leyva-Castillo JM, Oyoshi MK, Kumar L, Hoff S, He R, Chervonsky A, Oppenheim JJ, Kuchroo VK, van den Brink MRM, Malefyt RDW, Tessier P, Fuhlbrigge R, Terhorst C, Murphy G, and Geha RS. IL-23 induced in keratinocytes by endogenous TLR4 ligands polarizes dendritic cells to drive IL-22 responses to skin immunization. 2. Chou J, Kumar L, Yee C, Fraulino D, Bainter W, Hedayat M, Geha RS. Leucine-Rich Repeat Containing 8a (LRRC8A) is a novel co-stimulatory receptor for T cell activation by non-professional APCs Sridhara S, Kumar L, Verma J, Arora N, Gaur SN and Singh BP.

Skills

Adme, Immunology, Knockout, Laboratory, Mouse, Transgenic, Vitro, Vivo, Biotechnology, Clinical Research, Pids, Proposals, Sops, Training, Antibodies, Antibody, Bacterial, Biochemical, Bone Marrow, Cell Culture, Cytotoxicity, Dna, Dot, Elisa, Flow Cytometry, Gene Expression, Its, Mammalian Cell Culture, Mentor, Molecular Biology, Pcr, Quantitative, Real Time, Sorting, Southern Blot, Testing, Western Blot, Acad, Adv, Assembly, Auto Cad, Award, Career Development, Cellular, Ifn, Information Fuzzy Networks, Journal, Monoclonal, Rdw, Rna, Sbc, Sequencing, Statistical Analysis, Wang, Biochemistry, Zoology