Top 10 Publications (highlighting the important discoveries/contributions described in them briefly)

1. Mehto S, Jena KK, Nath P, Chauhan S, Kolapalli SP, Das SK, Sahoo PK, Jain A, Taylor GA, Chauhan S*. The Crohn's disease risk factor IRGM limits NLRP3 inflammasome activation by impeding its assembly and by mediating its selective autophagy. **Molecular Cell. 2019** Feb 7;73(3):429-445.e7. ***Corresponding author** (Impact Factor- 17.9)

2. Mehto S, Chauhan S, Jena KK, Chauhan NR, Nath P, Sahu R, Dhar K, Das SK, Chauhan S*. IRGM restrains NLRP3 inflammasome activation by mediating its SQSTM1/p62-dependent selective autophagy. **Autophagy**. **2019** Jun 20:1-3. ***Corresponding author**. (Impact Factor- 16)

Highlight of discovery in above two publications:

IRGM is an established genetic risk factor for Crohn disease. However, the mechanisms employed by IRGM to restrain the inflammation in Crohn disease are not known. The studies showed that IRGM negatively regulates NLRP3 inflammasome activation. IRGM employs 2 parallel approaches to constrain NLRP3 inflammasome activation. First, IRGM directly interacts with NLRP3 and ASC, and mediates their autophagic degradation. Second, IRGM impedes inflammasome assembly by blocking the polymerization of NLRP3 and ASC. The studies also found that IRGM suppresses NLRP3-mediated exacerbated outcomes of dextran sodium sulfate (DSS)-induced colitis in a mouse model. Since inflammasome are implicated in cancer, neurodegeneration, diabetes, and other inflammatory disorders, this work provides a framework for designing newer therapeutics.

3. Jena KK, Mehto S, Nath P, Chauhan NR, Sahu R, Dhar K, Das SK, Kolapalli SP, Murmu KC, Jain A, Krishna S, Sahoo BS, Chattopadhyay S, Rusten TE, Prasad P, Chauhan S, Chauhan S*. Autoimmunity gene IRGM suppresses cGAS-STING and RIG-I-MAVS signaling to control interferon response. **EMBO Rep. 2020** Jul 27:e50051. doi: 10.15252/embr.202050051**.* Corresponding author** (Impact Factor- 8.8)

4. Nath P, Jena KK, Mehto S, Chauhan NR, Sahu R, Dhar K, Srinivas K, Chauhan S, Chauhan S*. IRGM Links Autoimmunity to Autophagy. **Autophagy. 2020** Aug 19. doi: 10.1080/15548627.2020.1810920. PMID: 32813580. ***Corresponding author.** (Impact Factor- 16)

Highlight of discovery in above two publications:

IRGM deficiency is genetically and functionally associated with autoimmune diseases. However, the mechanism of IRGM mediated protection in autoimmunity remains undetermined. The abnormal activation of type I interferon (IFN) response is one of the significant factors in the pathogenesis of several autoimmune diseases. The studies showed that IRGM is a master suppressor of the interferon response. The depletion of IRGM results in constitutively activated cGAS-STING, RIG-I-MAVS, and TLR3-TRIF signaling pathways resulting in upregulation of almost all IFN responsive genes. Mechanistically, IRGM utilizes a two-pronged mechanism to suppress interferon response. One, it mediates p62-dependent selective autophagy of nucleic acid sensor proteins, including cGAS, RIG-I, and TLR3. Second, it facilitates the removal of defective mitochondria by mitophagy and avoids a buildup of mito-ROS and mito-DAMPs. Thus, IRGM deficiency results in increased nucleic acid sensors and DAMPs engaging a vicious cycle of aberrant activation of IFN response that is

known to occur in systemic autoimmune-like conditions. These studies suggest that IRGM could an excellent therapeutic target for autoimmune diseases.

5. Nath P, Chauhan NR, Jena KK, Datey A, Kumar ND, Mehto S, De S, Nayak TK, Priyadarsini S, Rout K, Bal R, Murmu KC, Kalia M, Patnaik S, Prasad P, Reggiori F, Chattopadhyay S*, Chauhan S*. *Inhibition of IRGM establishes a robust antiviral immune state to restrict pathogenic viruses*. **EMBO Rep**. 2021 Sep 1:e52948. doi: 10.15252/embr.202152948. *** Corresponding author.** (Impact Factor- 8.8)

Highlight of discovery in above publication:

The type I interferon (IFN) response is the major host arsenal against invading viruses. IRGM is a negative regulator of IFN responses under basal conditions. However, the role of human IRGM during viral infection has remained unclear. In this study, we show that IRGM expression is increased upon viral infection. IFN responses induced by viral PAMPs are negatively regulated by IRGM. Conversely, IRGM depletion results in a robust induction of key viral restriction factors including IFITMs, APOBECs, SAMHD1, tetherin, viperin, and HERC5/6. Additionally, antiviral processes such as MHC-I antigen presentation and stress granule signaling are enhanced in IRGM-deficient cells, indicating a robust cell-intrinsic antiviral immune state. Consistently, IRGM-depleted cells are resistant to the infection with seven viruses from five different families, including Togaviridae, Herpesviridae, Flaviviverdae, Rhabdoviridae, and Coronaviridae. Moreover, we show that Irgm1 knockout mice are highly resistant to chikungunya virus (CHIKV) infection. Altogether, our work highlights IRGM as a broad therapeutic target to promote defense against a large number of human viruses, including SARS-CoV-2, CHIKV, and Zika virus.

6. Kolapalli SP, Sahu R, Chauhan NR, Jena KK, Mehto S, Das SK, Jain A, Rout M, Dash R, Swain R, Lee DY, Rusten TE, Chauhan S*, Chauhan S*. RNA binding RING E3-ligase DZIP3/hRUL138 is a novel driver of cell cycle and cancer progression by employing a unique mechanism to stabilize Cyclin D1. **Cancer Research**, 2021 Jan 15;81(2):315-331. ***Corresponding author**. (Impact Factor- 12.7)

Highlight of discovery in above publication:

This study demonstrates DZIP3 as a novel driver of cancer progression by controlling RNA and protein stability of Cyclin D1 in cell cycle phase-dependent manner. In mice and zebrafish cancer models, DZIP3 was found to promote tumor growth and metastasis. DZIP3 was found to be frequently overexpressed and amplified in several cancer types, and its amplification was found to be associated with poor prognosis in cancer patients. The growth defect observed in DZIP3 depleted cells is because of reduced expression of Cyclin D1, leading to cell cycle arrest in the G1 phase.Mechanistically, DZIP3 utilizes its two different domains to interact and stabilize Cyclin D1 both at mRNA and protein levels. Using an RNA-binding lysine-rich region, DZIP3 interacts with 3'UTR of Cyclin D1 mRNA and stabilizes it. Using a RING E3-ligase domain, DZIP3 interacts and increases the K63-linked ubiquitination of Cyclin D1 protein to stabilize it. Interestingly, DZIP3 interacts, ubiquitinates, and stabilizes Cyclin D1 predominantly in the G1 phase of the cell cycle, where it is needed to be stabilized for cell cycle progression. This study, for the first time, demonstrates that DZIP3 is a crucial driver of cancer progression by increasing the stability of Cyclin D1 by a unique two-pronged mechanism.

7. Jena KK, Kolapalli SP, Mehto S, Nath P, Das B, Sahoo PK, Ahad A, Syed GH, Raghav SK, Senapati S, Chauhan S, Chauhan S*. TRIM16 controls assembly and degradation of protein aggregates by modulating the p62-NRF2 axis and autophagy. **EMBO J.** 2018 Sep 14;37(18). ***Corresponding author**. (Impact Factor- 11.6)

Highlight of discovery in above publication:

The formation of protein aggregates is linked to several diseases collectively called proteinopathies. The mechanisms and the molecular players that control the turnover of protein aggregates are not well defined. This study showed that TRIM16 acts as a key regulatory protein to control the biogenesis and degradation of protein aggregates. TRIM16 interacts with, enhances K63-linked ubiquitination of, and stabilizes NRF2 leading to its activation. The activated NRF2 upregulates the p62 and ubiquitin pathway proteins, which interact with and ubiquitinate the misfolded proteins resulting in protein aggregate formation. TRIM16 is physically present around the protein aggregates and acts as a scaffold protein to recruit SQSTM1 and macroautophagy/autophagy initiation proteins for sequestration of the protein aggregates within autophagosomes, leading to their degradation. This study could provide a framework for understanding the mechanisms of protein aggregate formation in neurodegeneration. The enhancement of TRIM16 activity could be a beneficial therapeutic approach in proteinopathies. On the flip side, cancer cells appear to hijack this machinery for their survival under stress conditions; hence, depleting TRIM16 could be a beneficial therapeutic strategy for treating cancer.

8. Chauhan, S*, Mandell, M. and Deretic, V*. IRGM governs the core autophagy machinery to conduct antimicrobial defense. May 7;58(3):507-21, **Molecular Cell.** 2015 ***Corresponding author**. (Impact Factor- 17.9)

Highlight of discovery in above publication:

IRGM, encoded by a uniquely human gene conferring risk for inflammatory diseases, affects autophagy through an unknown mechanism. This study delineate the mechanism by which IRGM controls autophagy. IRGM interacts with core autophagy proteins governing the formation of autophagy initiation complexes. IRGM interacts with pattern recognition receptors including NOD2. IRGM, NOD2, and ATG16L1, all of which are Crohn's disease risk factors, form a molecular complex to modulate autophagic responses to microbial products. NOD2 enhances K63-linked polyubiquitination of IRGM, which is required for interactions of IRGM with the core autophagy factors and for microbial clearance. Thus, IRGM plays a direct role in organizing the core autophagy machinery to endow it with antimicrobial functions.

9. Chauhan S, Kumar S, Jain A, Ponpuak M, Mudd MH, Kimura T, Choi SW, Peters R, Mandell M, Bruun JA, Johansen T, Deretic V (2016) TRIMs and Galectins Globally Cooperate and TRIM16 and Galectin-3 Co-direct Autophagy in Endomembrane Damage Homeostasis. **Developmental Cell** 39: 13-27. (Impact Factor- 12.27)

Highlight of discovery in above publication:

Macroautophagy/autophagy is a homeostatic process delivering cytoplasmic targets, including damaged organelles, to lysosomes for degradation; however, it is not completely understood how compromised endomembranes are recognized by the autophagic apparatus. This study uncovered the property of TRIMs to directly interact with members of the family of cytosolic lectins termed galectins. Galectins patrol the cytoplasm and recognize compromised membranes. TRIM16 uses LGALS3 (galectin 3) to detect damaged lysosomes and

phagosomes. TRIM16 assembles the core autophagic machinery and is found in protein complexes with MTOR and TFEB, thus regulating their activity to set in motion endomembrane quality control. The TRIM16-LGALS3 system plays a key role in autophagic homeostasis of lysosomes and in the control of Mycobacterium tuberculosis in vivo.

10. Chauhan S, Ahmed Z, Bradfute SB, Arko-Mensah J, Mandell MA, Won Choi S, Kimura T, Blanchet F, Waller A, Mudd MH, Jiang S, Sklar L, Timmins GS, Maphis N, Bhaskar K, Piguet V, Deretic V (2015) Pharmaceutical screen identifies novel target processes for activation of autophagy with a broad translational potential. **Nature Communications** Oct 27 6: 8620. (Impact Factor- 14.9)

Highlight of discovery in above publication:

Autophagy is a conserved homeostatic process active in all human cells and affecting a spectrum of diseases. In this study, we use a pharmaceutical screen (>3000 FDA approved drugs) to discover new mechanisms for activation of autophagy. We identify a subset of pharmaceuticals inducing autophagic flux with effects in diverse cellular systems modelling specific stages of several human diseases such as HIV transmission and hyperphosphorylated tau accumulation in Alzheimer's disease. One drug, flubendazole, is a potent inducer of autophagy initiation and flux by affecting acetylated and dynamic microtubules in a reciprocal way. Disruption of dynamic microtubules by flubendazole results in mTOR deactivation and dissociation from lysosomes leading to Transcription Factor EB nuclear translocation and activation of autophagy. By inducing microtubule acetylation, flubendazole activates JNK1 leading to Bcl-2 phosphorylation, causing release of Beclin-1 from Bcl-2-Beclin-1 complexes for autophagy induction, thus uncovering a new approach to inducing autophagic flux that may be applicable in disease treatment.

11. Chauhan, S., Goodwin, J.G., Chauhan, S., Manyam, G., Wang, J., Kamat, A.M., and Boyd, D.D. (2013). ZKSCAN3 Is a Master Transcriptional Repressor of Autophagy. **Molecular Cell.** Apr 11;50(1):16-28. (Impact Factor- 17.9)

Highlight of discovery in above publication:

Autophagy constitutes a major cell-protective mechanism that eliminates damaged components and maintains energy homeostasis via recycling nutrients under normal/stressed conditions. This study established ZKSCAN3, a zinc finger family DNA-binding protein, as a master transcriptional repressor of autophagy. Silencing of ZKSCAN3 induced autophagy and increased lysosome biogenesis. ZKSCAN3 represses transcription of a large gene set (>60) integral to, or regulatory for, autophagy and lysosome biogenesis/function and that a subset of these genes, including Map1lC3b and Wipi2, represent direct targets. Interestingly, ZKSCAN3 and TFEB are oppositely regulated by starvation and in turn oppositely regulate lysosomal biogenesis and autophagy, suggesting that they act in conjunction. This study uncovers an autophagy master switch regulating the expression of a transcriptional network of genes integral to autophagy and lysosome biogenesis/function.