#0082: A VAV1-Directed Molecular Glue Degrader, MRT-6160, Reduces Joint Inflammation in a Collagen-Induced Arthritis Autoimmune Disease Model

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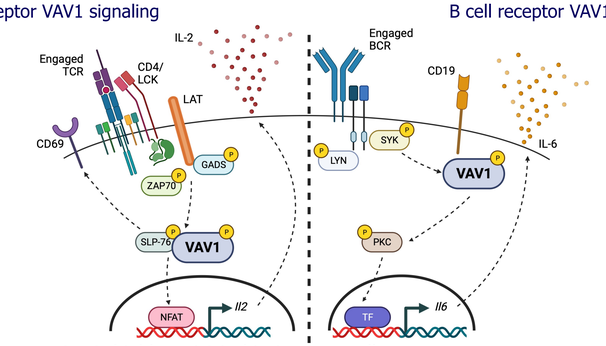
VAV1 is a guanine nucleotide exchange factor with a critical role in T- and B-cell receptor signaling and activity



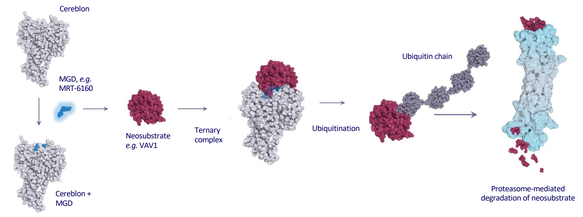
- VAV1 expression is highly restricted to immune cells
- VAV1 is required for antigen receptor-mediated signaling of T- and B-cells
- CRISPR-mediated[1] or genetic loss[2] of VAV1 is associated with decreased effector functions of both T- and B-cells

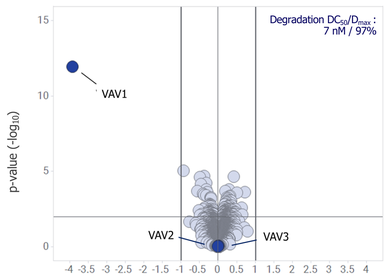
MRT-6160 is a rationally designed molecular glue degrader that selectively degrades VAV1 in human immune cells

Molecular glue degraders (MGD) function to induce structural changes in ubiquitin ligases, such as Cereblon, to drive the formation of ternary structures with a target protein.

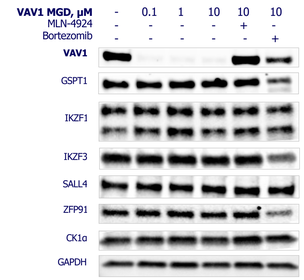
Following binding of Cereblon to the target protein, this target is then ubiquitin tagged and subsequently degraded via the proteasome-mediated degradation machinery of the cell.

MGDs can induce degradation of otherwise 'undruggable' proteins as the mechanism does not require a classical binding pocket contrary to conventional protein inhibitors, significantly increasing the target space and utility across a range of diseases.



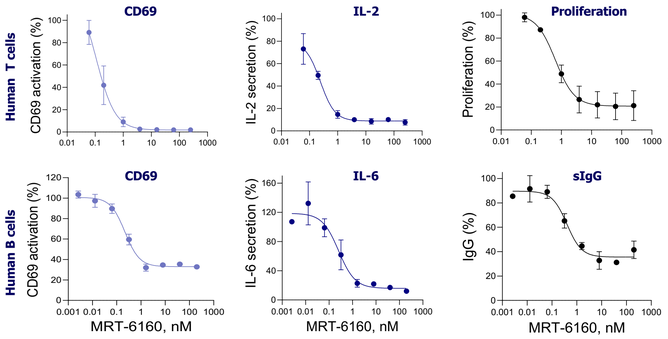


Human PBMCs were treated with 50 nM MRT-6160 or DMSO control for 24 hrs. MRT-6160 selectivity profile was evaluated by TMT-proteomics.



MM1.S and Kelly (SALL4) were treated with MRT-6160 for 6 hrs. To inhibit proteasomal machinery, cells were also treated with 2 μM MLN-4924 or 0.5 μM Bortezomib 30 min prior to treatment with 10uM MRT-6160.

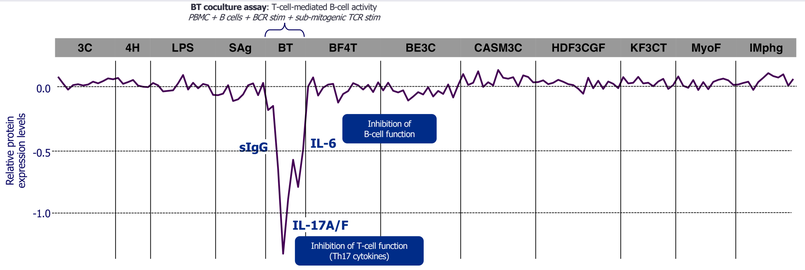
MRT-6160-induced degradation of hVAV1 attenuates T- and B-cell activation and effector functions



Purified primary human pan-T cells were pre-treated with MRT-6160 for 24 hrs followed by αCD3/CD28 TCR stimulation & subsequent analyses after 24 hrs (CD69), 48 hrs (IL-2), or 96 hrs (proliferation). Data are normalized to TCR-stimulated DMSO control. N = 2 donors.

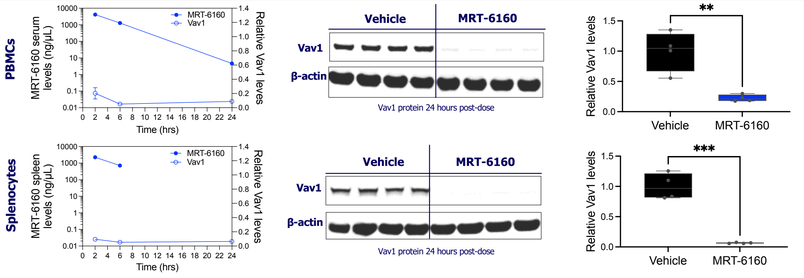
Purified primary human B-cells were pre-treated with MRT-6160 for 24 hrs followed by BCR stimulation and subsequent analyses after 24 hrs (anti-IgM + IL-4: CD69, IL-6) or 144 hrs (anti-IgM, sCD40L, IL-21, IL-2: BAFF-mediated sIgG). Data are normalized to stimulated DMSO controls. N = 3 donors.

Degradation of hVAV1 specifically inhibits key pro-inflammatory protein secretion by T- and B-cells



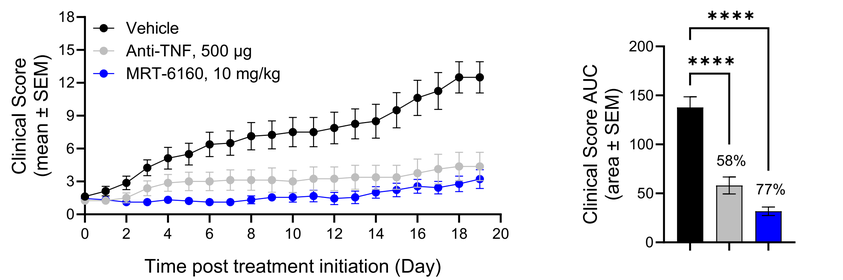
MRT-6160 (1 μM) was characterized using the BioMAP® Diversity Plus Platform (Eurofins). Shark tooth plot shows relative expression levels of indicated proteins in MRT-6160 treated vs. DMSO controls. 3C/4H, venular endothelial cells; LPS/SAg, venular endothelial cells + PBMC; BT, PBMC + B cells; BF4T, Bronchial epithelial cells + dermal fibroblasts; BE3C, bronchial epithelial cells; CASM3C, Coronary artery smooth muscle cells; HDF3CGF, Dermal fibroblasts; KF3CT, keratinocytes + dermal fibroblasts; MyoF, lung fibroblasts; IMphg, macrophages + venular epithelial cells.

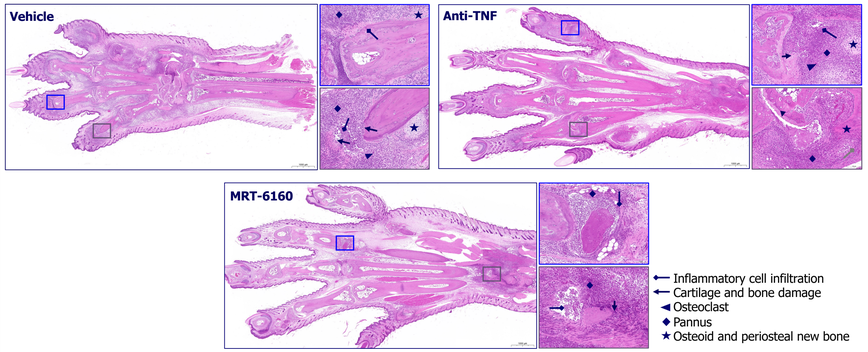
Oral dosing of MRT-6160 leads to rapid degradation of mVAV1 *in vivo*

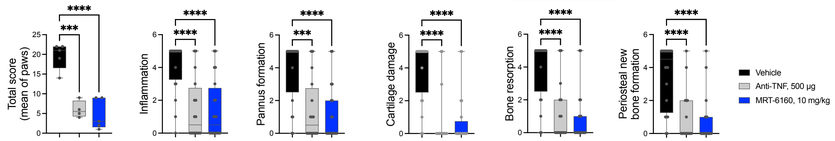


Mice were dosed with a single PO administration of MRT-6160 10 mg/kg. Pharmacokinetics (serum) and pharmacodynamics (PBMC and splenocytes). Quantification relative to vehicle treated mice. Western blot quantification shows 24-hour time point. Bars represent mean ± SD. Statistical analysis performed using an unpaired, two-tailed t-test, ***p<0.001, **p<0.01. N = 4 mice/group.

Oral dosing of MRT-6160 attenuates disease progression in a collagen-induced arthritis (CIA) autoimmune murine disease model







DBA-1 mice were immunized intravenously with chicken collagen-II emulsified in complete Freund's adjuvant on day 0, then boosted with a subcutaneous injection of chicken collagen-II emulsified in incomplete Freund's adjuvant on day 21. Mice were randomly enrolled into treatment groups following disease onset and then treated either PO QD (Vehicle, MRT-6160) or IP TIW (anti-TNF) for 20 days. ON day 22, mice were euthanized and paws were subsequently collected for histopathological analysis. Clinical scores are the sum of individual paw scores from 0–4 based on extent of toe/paw inflammation and ankylosis. Histopathology was scored based on indicated parameters in lower graphs. Graphs show mean ± SEM (*top left*), area ± SEM and disease incidence inhibition (top *left*) or min/max (*lower row*). Statistical analysis was performed using a one-way ANOVA with Dunnett's comparison. ***p<0.001, ****p<0.0001. N = 8 (Vehicle, anti-TNF) or 9 (MRT-6160) mice per group.

Summary and Future Development

- MRT-6160 is a first-in-class selective VAV1 molecular glue degrader that attenuates antigen receptor-mediated activation and effector functions of T- and B-cells.
- Oral dosing of MRT-6160 rapidly degrades Vav1 *in vivo* commensurate with exposure.
- Vav1 degradation attenuates disease progression in the CIA autoimmune disease model.
- Given its *in vitro* and *in vivo* MOA profile, MRT-6160 has strong potential to alleviate disease symptoms in multiple autoimmune and inflammatory diseases including rheumatoid arthritis, multiple sclerosis, inflammatory bowel disease, and psoriasis.
- MRT-6160 is a development candidate with IND submission anticipated in 1H24.

References: 1. Schmidt *et al. Science* (2022); 2. Fujikawa *et al. J. Exp. Med.* (2003)

All authors are employees of Monte Rosa Therapeutics