The anti-tumor activity of an anti-CD39 antibody (ES002) in a multiple myeloma xenograft model is dependent on NK cells and myeloid cells

Yangsheng Qiu, Qinglin Du, Meiling Sun, Huimin Kong, Roumei Xing, Yufei Shi, Dan Xu, Jinfeng Zhao, Mengying Xu, Zhihao Wu, Rui Gao, Hongtao Lu

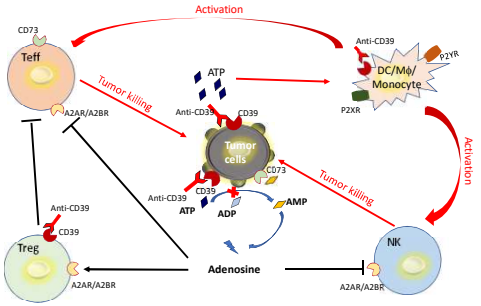
Elpiscience Biopharma, Ltd., 987 Cailun Road, 3rd Floor, Shanghai, China 201203

Poster abstract ID: P790

Elpiscience

BACKGROUND

CD39-CD73-adenosine pathway plays an important immuno-suppressive role within the tumor microenvironment (TME). To overcome the immunosuppression by adenosine in TME, we choose to target CD39 for the following two main reasons: 1). CD39 plays a pivotal role in converting extracellular ATP into final product adenosine. Blocking CD39 enzymatic activity will not only lead to the inhibition of adenosine generation, but also maintain extracellular ATP level which can enhance T cell priming by dendritic cells (DCs). 2). CD39 is expressed highly in Tregs and exhausted T cells, inhibition of CD39 activity will likely suppress Treg inhibition and reinvigorate exhausted T cells.

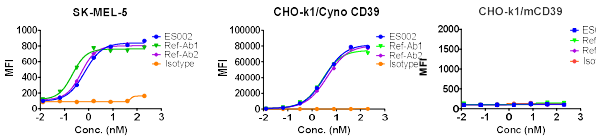


METHODS

We have generated a CD39 antibody, ES002, by hybridoma technology using human CD39 overexpressing HEK293 cells as immunogen, and the antibody was subsequently humanized through complementarity determining region (CDR) grafting. ES002 binding to CD39 and inhibition of ATPase activity were evaluated through protein-based and cell-based assays. The immunological function of ES002 was studied in an in vitro Elpiscience proprietary Immuno-assay platform (ImmunoShine). The in vivo efficacy of ES002 was investigated in a multiple myeloma xenograft cancer model. The effector immune cells were each depleted to analyze their respective roles in tumor growth inhibition.

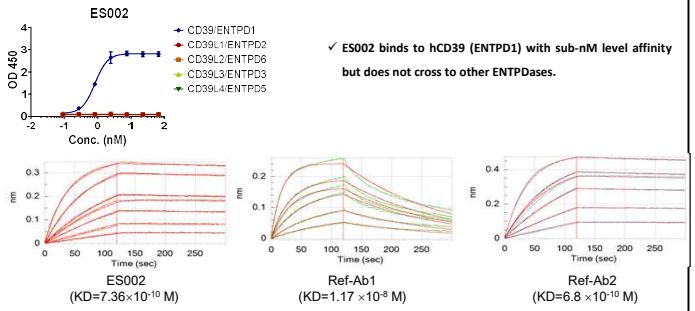
RESULTS

Figure 1. ES002 specifically recognizes human CD39 and monkey CD39, but not mouse CD39



RESULTS

Figure 2. ES002 specifically binds to hCD39 but not to other ENTPDases



✓ **ES002 binds to hCD39 (ENTPD1) with sub-nM level affinity but does not cross to other ENTPDases.**

Figure 3. ES002 is an allosteric blocker of ATP degradation by CD39

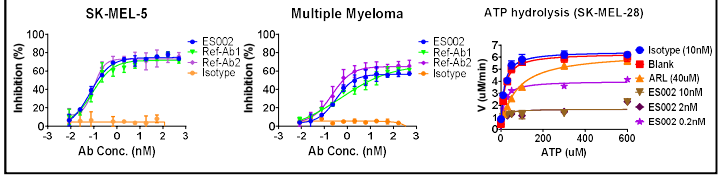
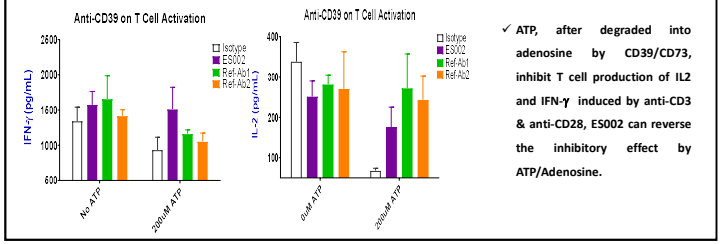
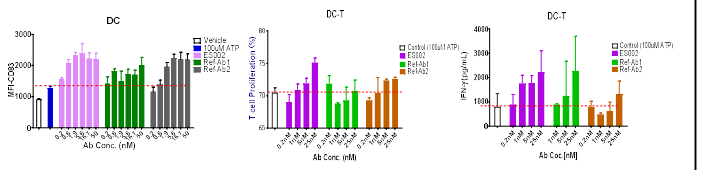


Figure 4. ES002 reverses ATP (Adenosine) mediated inhibition to T cells



✓ **ATP, after degraded into adenosine by CD39/CD73, inhibit T cell production of IL2 and IFN-γ induced by anti-CD3 & anti-CD28, ES002 can reverse the inhibitory effect by ATP/Adenosine.**

Figure 5. ES002-activated DC can functionally stimulate T cells in MLR

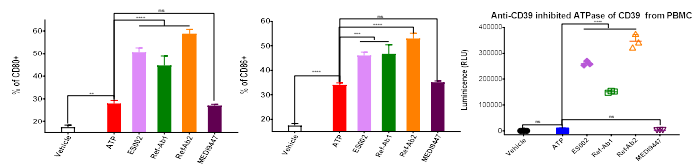


✓ **ES002 enhanced ATP-mediated DC maturation.**

✓ **ES002 activated DC can stimulate T cell proliferation and cytokine production in MLR assay.**

RESULTS

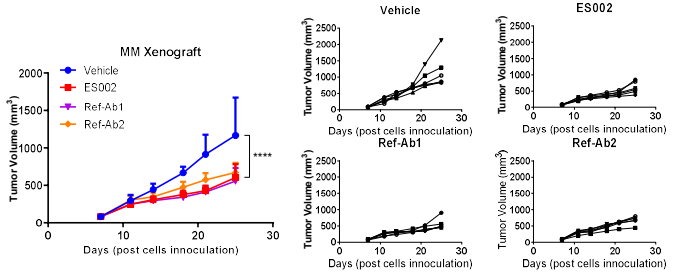
Figure 6. ES002 enhances monocyte activation mediated by ATP



✓ **Anti-CD39 enhanced ATP-mediated CD80 and CD86 expression on monocytes and its activity correlated ATP concentration which prevented to degraded by anti-CD39.**

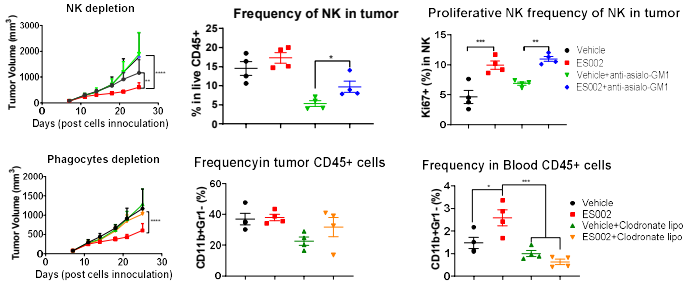
Data are presented as individuals with mean ± SD. ns, not significant; ***, p<0.001; ****, P < 0.0001

Figure 7. ES002 significantly inhibits tumor growth in a MM xenograft model



Data are presented as individuals with mean ± SEM. ****, P < 0.0001

Figure 8. The anti-tumor activity of ES002 in a multiple myeloma model is dependent on NK cells and myeloid cells



✓ **ES002's tumor growth inhibitory effect can be reversed by NK depletion.**

✓ **ES002 treatment slightly increases NK frequency and promotes NK proliferation in tumor, but has no effect on NK in blood (data not shown).**

✓ **Phagocytes depletion also can completely abolish ES002's inhibition function to tumor growth too**

Data are presented as individuals with mean ± SEM. *, P< 0.05; **, P<0.01; ***, p<0.001; ****, p<0.0001