Published in final edited form as:

Crit Rev Immunol. 2018 ; 38(6): 453–470. doi:10.1615/CritRevImmunol.2018026335.

IL-33 in Tumor Immunity: Nothing to Sneeze At

Donye Dominguez^{a,b}, Yi Zhang^b, Bin Zhang^{a,b,*}

^aRobert H. Lurie Comprehensive Cancer Center, Department of Medicine–Division of Hematology/Oncology, Northwestern University Feinberg School of Medicine, Chicago, Illinois

^bBiotherapy Center, The First Affiliated Hospital of Zhengzhou University, Zhengzhou, Henan, China

Abstract

Although immunotherapy has been at the forefront of cancer therapy for the last several years, better clinical responses are still desired. Interleukin-33 is perhaps one of the most overlooked antitumor cytokines. Its ability to promote type 1 immune responses, which control tumor growth in preclinical animal models is overshadowed by its association with type 2 immunity and poor prognosis in some human cancers. Accumulating evidence shows that IL-33 is a powerful new tool for restoring and enhancing the body's natural antitumor immunity cycle. Furthermore, the antitumor mechanisms of IL-33 are two-fold, as it can directly boost CD8⁺ T cell function and restore dendritic cell dysfunction *in vivo*. Mechanistic studies have identified a novel pathway induced by IL-33 and its receptor ST2 in which dendritic cells avoid dysfunction and retain cross-priming abilities in tumor-bearing conditions. Here, we also comment on IL-33 data in human cancers and explore the idea that endogenous IL-33 may not deserve its reputation for promoting tumor growth. In fact, tumors may hijack the IL-33/ST2 axis to avoid immune surveillance and escape antitumor immunity.

Keywords

IL-33; ST2; CD8⁺ T cells; dendritic cells; antitumor immunity

I. INTRODUCTION

Interleukin-33 is a cytokine that was discovered in 2005 and has been classified as a member of the IL-1 family.¹ Initially, IL-33 was primarily associated with induction of type 2 immunity due to its effects in promoting allergy and asthma via Th2 CD4⁺ T cells.² The IL-33/ST2 axis is now known to play a role in diseases other than allergy which range from obesity to Alzheimer's to cancer. Due to its ability to induce and enhance type 1 antiviral immunity when released from virally infected epithelial barrier cells, IL-33 has been defined as a novel "alarmin" or damage-associated molecular pattern (DAMP).³ IL-33 has thus been gradually recognized as a pleiotropic cytokine because, in addition to promoting

*Address correspondence to: Bin Zhang, Robert H. Lurie Comprehensive Cancer Center Department of Medicine–Division of Hematology/Oncology, Northwestern University Feinberg School of Medicine; 300 E Superior Street-Tarry 4-726, Chicago, IL 60611; Biotherapy Center, The First Affiliated Hospital of Zhengzhou University, Zhengzhou, 450052, Henan, China; Tel: 312-503-2435; Fax: 312-503-0189, bin.zhang@northwestern.edu.

inflammation, it provides protection against atherosclerosis and plays a role in tissue repair.
4-6

The role of IL-33 in cancer remains controversial, with many holding the opinion that IL-33 is a protumor cytokine, especially in humans. A growing amount of evidence in preclinical mouse models supports the idea that exogenous IL-33 can act to induce robust type 1-mediated antitumor immunity. Often glossed over is that a number of human studies also point to the fact that endogenous IL-33 may act as an antitumor cytokine and that loss of IL-33 expression in tumor tissue may result in metastasis.

II. BASICS

IL-33 is primarily expressed in the nucleus of nonimmune cells, including epithelial barrier cells and fibroblasts.³ IL-33 initiates intracellular molecular pathways via its receptor ST2 (IL-1R-like-1), which is expressed on a wide variety of both innate and adaptive immune compartments.^{7,8} Innate immune cells that respond to IL-33 include group 2 innate lymphoid cells (ILC2s), mast cells, eosinophils, and natural killer cells. IL-33 regulates adaptive immunity through direct action on dendritic cells (DCs), CD4⁺ T cells, and CD8⁺ T cells.

A. A Nonclassical Cytokine

IL-33 is located on chromosome 9 at 9p24.1 in humans and at 19qC1 in mice.^{8,9} It is expressed in several splice variants that do have an effect on protein activity.^{10,11} The IL-33 protein contains a nuclear/chromatin-binding region, a cleavage region for generation of active short forms, and a consensus region for caspase cleavage. Normally, IL-33 expression is confined to the nucleus through chromatin-binding motif interaction with nucleosomes.¹² Due to its sequence homology, IL-33 has been classified as the newest IL-1 family member, most closely resembling IL-18 in structure. IL-33 contains the IL-1 signature of beta tree foils, which accounts for its strong covalent interaction with its receptor ST2.¹³

IL-33 can be released from necrotic cells or by fibroblasts and cardiac cells under mechanical stress in its full-length form.¹⁴ When released from necrotic cells, full-length IL-33 acts as an “alarmin” to enhance inflammation. In a second processing, step IL-33 is cleaved by extracellular enzymes to produce “mature” IL-33, which engages in more activity.^{15,16} As a mechanism to prevent IL-33 inflammation under nonpathological conditions, nuclear IL-33 is rendered inert by either caspase-1 or caspase-7 in apoptotic cells.^{17,18}

B. Molecular Signaling

The IL-33 receptor, originally termed suppressor of tumorigenesis 2 (ST2 or IL-1R1), was initially discovered in 1989.⁷ ST2 is found in three forms, including two membrane-bound forms and one soluble form. The crystal structure forms of the IL-33, ST2, and the IL-33/ST2 complex have given insight into the interaction of the two molecules.¹³ IL-33 has two regions that form strong covalent bonds with ST2 to induce conformational change in the receptor. ST2's conformational change is necessary for initiation of intracellular cascade events.

IL-33 recruits the IL-1 receptor accessory protein (IL-1RAcP) as a necessary adaptor protein to form a dimer.¹⁹ ST2, IL-1Rap, and MyD88 then form scaffolding for IRAK1,4 and TRAF6 to activate JAK-STAT, NF- κ B, JNK, ERK, and p38 activity.^{1,9,20–22} Soluble ST2 (sST2) does not have the transmembrane domain of ST2 and can be produced by mast cells and T cells as a decoy receptor to regulate extracellular IL-33.^{23–25}

C. Type 2 Cytokine with Type 1 Abilities

IL-33 is well known for its role in Th2-mediated pathogen expulsion and Th2-driven disease pathology. In particular, IL-33 is crucial for protection against helminth infections through induction of Th2 cytokine production and activation of mast cells and ILC2s.^{26–29} IL-33 is also associated with type 2-mediated airway inflammation in allergy and asthma. IL-33 expression correlates with disease severity in humans, and it has recently been shown that IL-33 can promote antigen-driven asthma by enhancing Th2 derived IL-4, IL-5 and IL-13.^{30–32}

Initially, it was believed that only Th2 CD4⁺ T cells were able to directly respond to IL-33.³³ However, the paradigm of IL-33 has shifted and it is now known that IL-33 plays a much broader role in many T cell subsets.³⁴ In fact, IL-33 has been implicated in many type 1–associated diseases. The literature has described evidence of IL-33 driving type 1 immunity from human NK cells and even driving IFN- γ from allergen-specific Th2 cells.^{35,36} IL-33 can also drive or exacerbate Th1 responses through different mechanisms in chronic obstructive pulmonary disease (COPD), viral infections, and graft versus host disease (GVHD).^{37–43} A major shift in IL-33 biology came with the discovery that IL-33 plays a central role in orchestrating viral clearance.⁴⁴

D. Promotion of Type 1 Immunity *in Vivo*

Type 1 immunity response is characterized by production of cytokines such as IL-2, IL-12, IFN- γ , and TNF- α . IL-12 production by DCs and macrophages induces skewing of naïve CD4⁺ T cells into Th1 cells. Th1 cells produce TNF- α and IFN- γ , which directly suppress tumor growth⁴⁵ while IL-2 production supports the effector activity of tumor-reactive CD8⁺ T cells. Antitumor immunity can also be elicited by NK cell immune surveillance through direct killing of tumor cells and by their ability to produce IFN- γ .^{46,47} The current goal of immunotherapy is to restore or boost type 1 immune responses against established tumors observed in clinical patients. In humans, there is a strong correlation of Th1 type immune responses and better patient prognosis when compared to inflammation driven by Th2 and Th17 cells.^{48,49}

Worthy of mention is that Th2 and Th17 express high levels of ST2 and sST2, which act as negative regulators of IL-33 and hamper its ability to drive type 1 immunity in cancer. Recently, it came to be appreciated that both Tc1 CD8⁺ T cells (a subset associated with IFN- γ production)⁵⁰ and Th1 CD4⁺ T cells can respond directly to IL-33. Furthermore, lack of ST2 expression on type 1 T cells diminishes their effector functions.^{51,52} ST2 expression on T cells is regulated by the Tc1/Th1-associated transcription factor T-bet in Tc1 cells and T-bet and STAT-4 in Th1 cells.^{51,52}

Our own data have confirmed that IL-33 can directly enhance the proliferation of purified tumor antigen-specific CD8⁺ T cells as well as boost their IFN- γ production and cytolytic activity.⁵³ IL-33 also activates human Th1 and Th2 cells.⁵⁴ Interestingly, in response to interferons produced during viral infections, Th2 cells exhibit plasticity that allows them to become a Th1/Th2 hybrid memory cell, expressing the Th1 transcription factor T-bet while producing IFN- γ .⁵⁵ It may be worth investigating whether IL-33-induced IFN- γ production *in vivo* can reprogram Th2 cells into a Th1 memory phenotype in tumor-bearing conditions, thereby enhancing antitumor T cell immunity. In unpublished data, we confirmed that the addition of IL-33 to T cell activation conditions *in vitro* can induce a stem memory phenotype of tumor antigen-specific CD8⁺ T cells.

III. PROMOTION OF ANTITUMOR IMMUNITY *IN VIVO*

The cytokine milieu created in the tumor microenvironment is highly suppressive and drives the adaptive immune response into a tolerant state. One idea to reverse this effect is through the administration of immune-activating recombinant cytokines. IL-2 was the first cytokine used in clinical settings for its ability promote T cell proliferation and effector functions and to control melanoma in mouse models when used in combination with adoptive T cell therapy.^{56,57} IL-12 is another cytokine that has shown promise and is still being manipulated for better delivery to the tumor microenvironment in clinical settings.⁵⁸ Cytokine therapy is accepted as a viable option for the treatment of human cancers, but new options, including IL-33, are desperately needed to save lives.

Two studies, one using transgenic mice engineered to release IL-33 and another using IL-33-expressing cell lines, reported that IL-33 can inhibit tumor growth in a CD8⁺ T cell- and NK cell-dependent fashion.^{59,60} IL-33 can also act as an adjuvant to enhance antitumor immunity.^{61,62} When IL-33 is expressed via DNA vaccine, CD4⁺ T cells increase IFN- γ secretion but do not increase IL-4 production. This means that IL-33 can induce type 1 immunity over type 2 immunity when used as a cancer therapy. In a mechanistic study, replicating viral vectors induced necrotic release of endogenous IL-33 from virally infected cells, which led to a significant increase in tumor-infiltrating CD8⁺ effector T cells and significant tumor growth control.⁶³

A. Recombinant IL-33 in Treatment of Advanced Cancers

Although the results of several IL-33 reports support each other, many of the routes of administration are not feasible for human patients. Therefore, we tried to mimic human clinical settings in our models by waiting to treat established tumors with systemically administered exogenous recombinant IL-33 (rIL-33).⁵³ This is a route that can be used in humans via intravenous infusion. We also used the clinically relevant Braf^{V600E}PTEN mouse model, which harbors the same mutations observed in human melanoma patients.^{64,65} Using “high-dose” IL-33 (1 μ g/day), we found that rIL-33 can significantly inhibit tumor growth through a CD8⁺ T cell dependent mechanism, which is supported by other IL-33 therapeutic strategies.

Specifically, rIL-33 promotes proliferation of tumor antigen-specific CD8⁺ T cells and enhances their IFN- γ production. In agreement with the literature, we also observed an

increase in CD4⁺Foxp3⁺ regulatory T cells (Tregs) and Gr1⁺CD11b⁺ myeloid-derived suppressor cells (MDSCs) both in the spleen and in tumor tissue. However, the ratio of effector cells to both Treg and MDSC populations highly favored CD8⁺ T cells. Similar results were observed in B16F10 melanoma models. However, in B16-SIY models, which are engineered to express the model antigen SIY, we were surprised to find a decrease in tumor-infiltrating Tregs.⁵³ This may have been because SIY expression enhances the immunogenicity of B16F10, which is notorious for its poor immunogenicity.^{66,67} In a leukemia model, IL-33 enhanced adoptive T cell therapy by preventing exhaustion of antigen-specific CD8⁺ T cells.⁶⁸

B. Restoration of DC Function in the Tumor Immunity Cycle

In addition to T cells, IL-33 regulates DCs. IL-33 is generally thought to condition DCs to skew or promote Th2 conversion and function, but it also promotes DC function and IFN- γ response in viral challenges.^{69–72} In the tumor immunity cycle, DCs engulf antigen at the tumor site and then proceed to migrate to tumor-draining lymph nodes. In a process known as cross-priming, which is indispensable for tumor rejection, DCs present exogenous tumor antigen on their MHC class I molecules to prime tumor antigen-specific CD8⁺ T cells.^{73–75} Tumors disrupt this cycle through the generation of dysfunctional DCs, which leads to poor-quality antitumor T cell response characterized by high levels of T cell anergy and an increase in Tregs.^{76,77}

rIL-33 reprograms dysfunctional DCs in tumor-bearing conditions and restores the tumor immunity cycle.⁵³ IL-33 upregulates costimulatory molecules on DCs both in draining lymph nodes and in tumor tissue. Functional studies of DCs from draining lymph nodes of rIL-33-treated mice show a dramatic difference in their cross-priming ability when compared to control mice. These *ex vivo* rIL-33-treated DCs induce significantly more IFN- γ secretion from tumor antigen-specific T cells than their PBS-treated counterparts.

At a molecular level, IL-33 induces activation and phosphorylation of signal transducer and activator of transcription 1 (STAT-1). STAT-1 phosphorylation in DCs is essential for their ability to mature and to prime peptide-specific cytotoxic lymphocytes; its deficiency impairs their Th1 skewing.^{78–80} This novel molecular pathway requires intact signaling of IL-33/ST2/MyD88 and STAT-1. STAT-1 is constitutively activated in DCs of rIL-33-treated mice, indicating that DCs exposed to high-dose rIL-33 are reprogrammed to resist tumor-induced suppression mechanisms. IL-33 also seems to work as an adjuvant to other DC-activating treatments. The combination of rIL-33 with agonistic anti-CD40 significantly improved tumor inhibition and induced regression in some mice.⁵³

As IL-33 can directly expand antigen-specific T cells and restore the antigen cross-priming ability of DCs, we believe that rIL-33 may help alleviate the need for peptide vaccinations. Instead of trying to force the immune system to attack a specific peptide that may or may not be expressed by all tumor cells, it is probably more relevant to readily restore the inherent antitumor mechanisms. IL-33 appears to be powerful as a monotherapy or as an adjuvant to existing immunotherapies; it deserves a serious look in clinical settings as a way to improve patient outcomes.

C. Adjuvant for Adoptive T Cell Therapy

Allogeneic hematopoietic cell transplant represents a curative therapy for treatment of hematological malignancies. The problem with it is that the killing of malignant cells by the donor transplant coincides with donor T cells attacking healthy host tissues. The addition of exogenous rIL-33 to conventional Th9 skewing conditions (IL-4 and TGF- β) creates both Th9_{IL-33} and Tc9_{IL-33} cells for adoptive therapy. These cells exhibit strong antileukemia features with minimal GVHD side effects. Human rIL-33-induced Tc9_{IL-33} cells increased CD8 α expression and enhanced killing of AF9 leukemia cells *in vitro*.⁸¹ Treatment of human TCR V γ 9⁺ $\gamma\delta$ T cells mixed with CD4⁺ T cells supported inductions “self”-production of IL-2 without exogenous IL-2 supplementation.⁸² The key point here is that IL-33 treatment prior to adoptive transfer eliminated the need for administration of supplemental exogenous IL-2 to support TCRV γ 9⁺ $\gamma\delta$ T cells *in vivo*. IL-2 can be counterproductive to immunotherapies because it supports Treg suppressive functions that play a central role in suppression of antitumor immunity.

D. Innate Immune Tumor Control

Using depletion assays, we did not find that NK cells were related to rIL-33 tumor inhibition⁵³ as shown with transgenic expression of IL-33.⁵⁹ We hypothesize that in late-stage tumors, NK cells are unable to respond to IL-33 due to increased immune suppression. New findings show that IL-33 can induce tumor regression through the activation of eosinophils to promote CD8⁺ T cell tumor control.⁸³ These data are supported by a study that reported IL-33 promotion of eosinophil invasion of the tumor tissue while in metastatic disease the eosinophils are only found in the peripheral tissue of metastatic cancer.⁸⁴ Although there are conflicting reports,⁸⁵ we have published evidence that ILC2s, which others have reported as antitumor, actually suppress NK-mediated killing through expression of inhibitory factors.⁸⁶ According to our data, ILC2s play a role in hampering IL-33 antitumor functions in which the immune-suppressive molecule CD73 plays a role.

E. Conflicting Reports of IL-33 in Protumor Immune Response

A number of studies have reported that ST2/IL-33 is a protumor axis that can promote breast cancer metastasis through the recruitment of suppressive immune cells.^{87–89} Two studies from the same group used ST2^{−/−} Balb/c mice and saw enhanced antitumor response in the 4T1 breast cancer model.^{87,90} In this scenario, ST2⁺ Tregs, which outcompete Th1 cells for available IL-33, would be able to expand and suppress antitumor immunity. In these protumor studies, the treatment schedule of rIL-33 is completely different from the antitumor studies using rIL-33 described previously. IL-33 is administered only twice weekly or every other day at 0.4 μ g/mouse.⁹¹ In a wild-type Balb/c mouse that is already prone to Th2 responses, this amount of IL-33 is probably insufficient to oversaturate ST2 and sST2 on Th2 cells and ST2⁺ Tregs, leaving no available IL-33 to activate Tc1/Th1 cells. The threshold of ST2 signaling that must be overcome to activate type 1 immunity probably exists as a mechanism to prevent exaggerated responses to minor barrier tissue injuries.

Finally, it may be the case that in response to high-dose IL-33, MDSCs may lose suppressive function.⁹² This hypothesis is controversial, and we have observed an increase in MDSC populations in tumor tissue following rIL-33 treatment.⁵³ IL-33 seems to promote an

overwhelming increase in the CD8⁺ T cell/MDSC ratio. MDSC function in response to high-dose IL-33 needs to be further evaluated. We infer that low IL-33 concentration promotes tumor growth while high amounts overcome IL-33 suppressive mechanisms to promote type 1 immunity. This theory warrants further investigations, particularly in humans.

IV. IL-33/ST2 AXIS IN CANCER

The typical opinion of IL-33 in human cancer is that it is a protumor cytokine. However, several studies have suggested that endogenous IL-33 may play a role in immune surveillance in human patients (Table 1). Modulation of the IL-33 axis through either treatment with exogenous IL-33 or blockade of the IL-33 decoy receptor sST2 may prove beneficial.

A. Tumor Cell Apoptosis

In at least two human cell lines, IL-33 can directly promote apoptosis through ST2 signaling. In the human pancreatic cell line MIA PaCa-2, which constitutively expresses ST2, IL-33 promotes apoptosis by enhancing the expression of antiproliferative molecules and the upregulation of apoptotic genes p21, cdk4, and TRAIL.⁹³ Lung cancer cells in both mouse and human express ST2. rIL-33 treatment of ST2⁺ lung cancer cells increased phosphorylation of p38 MAPK and JNK, which are validated targets of ST2 and induce cell death. In the same study, human data mining determined that IL-33 correlates with a higher malignancy index, indicating that loss of IL-33 promotes tumor outgrowth.⁹⁴

B. Hepatocellular Carcinoma (HCC)

In a study of resected hepatocellular carcinoma, an effector population of CD8⁺ IL-33⁺ T cells was associated with prolonged patient survival. The tumor-infiltrating T cells had a phenotype of CD8⁺ KLRG⁺ CD107a⁺ effector memory cells and were the main source of IL-33 production in the tumor.⁹⁵ In unpublished work, we, too, confirmed that rIL-33-treated mice express higher levels of KLRG in T cells infiltrating tumor tissue. Our data indicate that T cell-derived IL-33 may provide an unexplored mechanism for tumor control. IL-33 expression in tumor cells in HCC indicates a more differentiated cell type that is associated with a better prognosis.⁹⁶

C. Osteosarcoma and Bone Marrow Cancers

Like many cancers, metastatic osteosarcoma has a very poor prognosis. In human breast cancer metastasis, osteoclasts are responsible for carving out a metastatic niche for breast cancer cells to metastasize to bone.⁹⁷ In a multi-institutional GWAS study to determine survival in patients with osteosarcoma, the GLDC/IL-33 locus was associated with overall survival. It turns out that low IL-33 expression is associated with poor survival in patients with osteosarcoma.⁹⁸ Interestingly, IL-33/ST2 induces osteoclast apoptosis in animal models.⁹⁹ In chronic lymphocytic leukemia and multiple myeloma, cancers that arise in bone marrow, patients have reduced IL-33 plasma expression, suggesting that loss of IL-33 may dampen immune surveillance in bone marrow.^{100,101}

D. Lung Adenocarcinoma

In lung adenocarcinoma, high expression of IL-33 mRNA from tissue is correlated with better a prognosis, while plasma levels of NSCLC patients had lower serum IL-33 compared to healthy controls.^{102,103}

E. Colon Cancer

In human colon cancer, tumor cells expressed lower levels of ST2 compared to adjacent healthy tissue. Similar to lung and pancreatic cells, IL-33 may be controlling early tumor growth by directly inducing apoptosis of ST2⁺ tumor cells.¹⁰⁴ Downregulation of ST2 gene signatures correlated with late-stage colorectal cancer (CRC) when compared to early stage CRC, supporting a role for an IL-33/IFN- γ axis initiated by ST2 to control CRC.¹⁰⁵ A novel chimeric transcript of IL-33 (LTR-IL-33) found in CRC was recently described and compared to natural IL-33. LTR-IL-33 is optimal for growth of CRC 3D colonospheres.¹⁰⁶ An important point is that the primers used in studies that correlate IL-33 with poor prognosis are unable to distinguish LTR-IL-33 from native IL-33. It may be necessary to re-evaluate the expression of LTR-IL-33 in other human cancers.

F. Cervical Cancer

Severe intraepithelial neoplasia exhibited the lowest amount of IL-33 mRNA compared to mild neoplasia and cancer-free patients. IL-33 also positively correlated with production of IFN- γ .¹⁰⁷ IL-33 is well known to promote antiviral immune response, and it may be the case that it is valuable in thwarting virally induced cancers.

G. Head and Neck Cancer

In malignant head and neck cancers, including myoepithelial, acinic cell, and oncolytic carcinomas IL-33 has favorable prognosis due to driving cell differentiation. Specifically, loss of IL-33 leads to loss of myofibroblasts through loss of differentiation status and correlates with a poor prognosis.¹⁰⁸

H. Loss of IL-33 as a Driver of Metastasis

A detailed study of human tumor cell lines, patient tissue samples, patient survival, and *in vivo* mouse models, helps validate the human data presented in this review. Loss of IL-33 expression by tumor cells was identified as a key factor in metastatic transition and also promoted the formation of MHC I loss variants. MHC I loss variants fail to display tumor antigens, allowing them to evade immune surveillance and outgrow or metastasize. In mouse models, reintroduction of IL-33 to metastatic cell lines restored antitumor CD8⁺ T cell infiltrates and reduced the amount of circulating cancer cells. IL-33 was identified as a critical factor in the immune recognition of murine lung and prostate carcinomas. Downregulation of human IL-33 is significantly associated with reduced survival time in human prostate cancer and renal clear-cell cancer patients.⁸⁴

I. Role of ST2 and sST2

The expression of sST2 has generally been associated with a poor prognosis. sST2 plasma levels are increased in patients with gastric cancer and breast cancer and significantly

correlate with more advanced tumors, metastatic disease, and disease duration.^{109–111} sST2 expressed by T cells is upregulated in malignant pleural effusions. In a study of chronic myeloid leukemia, patients in molecular remission were observed to experience a drop in sST2 compared to patients with full-blown disease.^{112,113}

It may be that tumors, including those that form in the bone marrow, recruit sST2-expressing T cells, or other sST2-expressing cells, as a mechanism to suppress IL-33 antitumor responses. It has already been well established that T cells and mast cells can produce sST2 and that sST2 can suppress IL-33–induced inflammation *in vivo*.^{24,114,115} In human Th2 cells, sST2 can block IL-33 from inducing IFN- γ production.³⁵ Given that IL-33 antitumor immunity shares features with IL-33-driven antiviral immunity, it is interesting that similar patterns of high sST2 levels are also observed in HIV.

In two studies of HIV, sST2 levels increased while IL-33 decreased in patient serum when compared to healthy controls.^{116,117} This indicates that expression of sST2 may be a mechanism by which HIV limits the effectiveness of IL-33 antiviral immunity. The lack of available free IL-33, which is necessary for control of many types of pathogens, may be a factor in the ability of HIV patients to ward off secondary infections. In both HIV and cancer, sST2 currently serves as a potential prognostic marker for disease severity. However, the data presented indicate that secretion of sST2, an IL-33 decoy receptor, may be a previously unappreciated checkpoint employed by viruses and cancer to escape IL-33–induced inflammation.

J. ST2 and sST2 as Drivers of Mutated Cell Proliferation

ST2 and sST2 may play a direct role in promoting tumor growth independently of IL-33. In murine fibroblasts, oncogenic Ras mutations can induce the expression of sST2 and ST2 protein. Furthermore, gene expression of sST2 increased the focus formation and colony formation of Ras-transformed cells compared to Ras transformed cells without coexpression of sST2.¹¹⁸ Direct treatment with sST2 induces proliferation of murine fibroblasts, indicating that sST2 may play a larger role in fibroblast and tumor cell transformation than previously appreciated.¹¹⁹ It could be the case that cancer-associated fibroblasts and tumor cells enhance secretion of sST2, which has a dual function: to directly enhance proliferation and to act as a sink and reduce IL-33 to levels that favor tumor growth. If IL-33 is added directly to fibroblasts before they enter the proliferation cycle, IL-33 suppresses proliferation. Alternatively, if fibroblasts are in a proliferative state, IL-33 enhances proliferation.¹²⁰ It remains unclear if this process is dependent on ST2 or sST2 levels in response to IL-33 treatment.

K. Rethinking IL-33 Analysis

Many studies not detailed here have found that IL-33 increases in metastatic disease more so than in non-metastatic disease and correlates with a poor prognosis.¹²¹ In human cancers, it is crucial to analyze how serum IL-33 is defined and detected. Currently, the reagents to detect mature IL-33 from serum are unable to distinguish between free IL-33 and the IL-33/sST2 complex. This is a crucial point, as complexing IL-33 to sST2 renders it inactive. It is well established that IL-33 splice variants, extracellular processing, and binding to sST2 can

all effect downstream molecular signals of IL-33/ST2 binding. It is unknown if mutated cancer cells produce novel IL-33 variants as has been discovered in transformed cells and in human colorectal cancer.¹¹ Furthermore, the stage of cancer must be considered. High IL-33 serum levels often correlate with poor prognosis, yet it is unclear if this IL-33 is active or if it is accompanied by high levels of sST2, which would render it inert. Tumor-infiltrating neutrophils and other cells may also promote cleavage and inactivate IL-33, so even though IL-33 is detected by commercially available products, it may be that those products are unknowingly detecting a suppressive cleavage form. A very relevant study describes a mechanism by which IL-33 loses its ability to perform alarmin functions through inactivation by oxidation.¹²² It is unclear if this mechanism is exploited in tumors.

As in mice, it may be that high-dose amounts of rIL-33 that exceed the endogenous levels observed in cancer patients can act to reverse tolerance and restore antitumor type 1 immunity. The benefit of using recombinant IL-33 cytokine is that the isoform of IL-33 can be designed in the lab and patients can receive optimized, activating forms of it. Also, anti-sST2 antibodies may act to restore and increase the amount of free IL-33 available to activate DCs and Th1/Tc1 cells, which express ST2 at much lower levels and are activated only when excess IL-33 is available.

Another problem with evaluating the IL-33/ST2 axis is that ST2 may be a promiscuous receptor. Evolutionary studies have proposed that ST2 and IL-33 developed independently, as evidence shows that ST2 is conserved across many species but IL-33 is expressed only in mammals.¹²³ Therefore, there is a decent probability that ST2 has other ligands or that it has unappreciated functions independent of IL-33, which may be tumor-promoting.

V. CONCLUDING REMARKS

In the last century, immune surveillance and immunotherapy have gone through waves of promise and rejection. In the last decade, immunotherapy has come a long way from the days of doubt, with many therapies making it to clinical trials and some progressing through FDA approval as first-line treatments for human cancers. One hurdle is that, for many unknown reasons, not all patients respond to all immunotherapies.

In the past, IL-33, has received little attention for its ability to harness the power of the immune system against cancer. Accumulating evidence clearly demonstrates that IL-33 therapy is extremely effective at driving type 1 antitumor immunity by action on both the innate and adaptive arms of the immune system. High-dose therapy strategies favor the proliferation and effector function of tumor antigen-specific CD8⁺ T cells to control aggressive and poorly immunogenic tumors *in vivo*. Additionally, rIL-33 can reprogram DCs to maintain cross-priming ability and resist dysfunction in tumor-bearing conditions. This process happens through a previously undescribed IL-33 → ST2 → MyD88 → STAT-1 pathway that may be taken advantage of for novel conditioning regimens of human DCs.⁵³

One important feature of rIL-33 is that it works as a monotherapy without the conditioning regimens of antigen vaccine and irradiation needed by other cytokine therapies such as IL-2.

Naturally, the possibility of side effects exists with any systemically administered cytokine, including IL-33. It has been well known for over 20 years that exogenous IL-2 and IL-12 therapies induce toxic side effects that limit the tolerable dosage in human patients.^{124,125} In some experimental models, rIL-33 has the ability to exacerbate conditions such as acute GVHD, arthritis, atopic dermatitis, and hepatic fibrosis.^{41,126–128} Only in the case of acute GVHD with complete MHC mismatch does administration of recombinant IL-33 contribute to faster lethality. In this clinically irrelevant model, 100% of mice already succumb to lethal GVHD without rIL-33. The literature also shows a protective role for IL-33 in GVHD.¹²⁹ In the case of fibrosis, studies have generally focused on the role of endogenous IL-33 rather than that of rIL-33. To our knowledge, rIL-33 can enhance allergy and asthma pathology in mouse models but toxicity has not been a limiting factor in any therapeutic model for cancer.

In order to decrease the possibility of systemic toxicity and enhance the efficacy of cytokine therapies, including rIL-33, several strategies have evolved. One that may be applied to rIL-33 is the use of antibody-cytokine fusion delivery systems that help to release active cytokine only in the tumor microenvironment.⁵⁸ Another method, designed by a group of researchers experienced in IL-33 antitumor biology, is to deliver IL-33 to the local tumor environment using CAR T cells harboring an IL-33 expression cassette.¹³⁰ Yet another option is to unleash endogenous IL-33 by blocking sST2 with novel small-molecule inhibitors. As mentioned in earlier sections, human patients can express high levels of serum IL-33 that may be inactivated by high levels of serum sST2. In order to promote endogenous IL-33 activity in GVHD models, researchers recently developed anti-ST2 small-molecule inhibitors that decrease the amount of serum sST2.¹³¹ Importantly, these small-molecule inhibitors ameliorate GVHD while enhancing graft-versus-leukemia effects in *in vivo* MLL-AF9 mouse models.

IL-33 is a flexible cytokine that may work to improve monotherapies that provide only partial benefit when compared to combination therapies. This area needs to be dissected further to optimize combination strategies, but it remains under intense investigation.¹³² When combined with Treg depletion and agonistic anti-CD40 in mouse models of advanced melanoma, some levels of regression can be achieved. These same studies also show that IL-33 alone can perform as well as IL-12 and agonistic anti-CD40 monotherapies, which are both in clinical trials.^{53,60} Furthermore, IL-33 can enhance adoptive T cell therapy when used to condition T cells prior to reinfusion⁸¹ and may provide benefit for DC vaccines because it directly promotes DC priming.⁵³

The data on human IL-33 as a protumor cytokine may also be misleading. IL-33 serum levels may represent a useful biomarker for cancers such as breast and lung, where increased serum levels may predict disease stage, but the data need to be reexamined for therapeutic opportunities. Current methods of detecting and analyzing IL-33 may be found wanting as new studies begin to elucidate more about IL-33 biology. Many factors influence IL-33 isoforms, including cell transformation, intracellular processing, and extracellular processing. Oxidation can now be added to the list of factors that influence IL-33 signaling capability.¹²² Based on the isoform or modification, IL-33 can mature to become functional or to be deactivated. It is unclear if current methods of detection (antibodies for Western blot and imaging, primers for PCR) can distinguish IL-33 variants. It is also unclear if IL-33

quantification from serum is a true indicator of active IL-33, as current assays do not distinguish between free IL-33 and inactive IL-33 complexed with sST2. It may be that the increase in sST2 serves as an immune checkpoint response that is hijacked by tumors in response to high IL-33 exposure, similarly to how the immune-suppressive molecule PD-L1 is upregulated in response to the antitumor cytokine IFN- γ .

When developing therapies, the patient must always be kept in mind. Often, it seems that new breakthroughs in cancer research consist of advanced technologies that are not feasible for broad clinical use. Eventually, personalized medicine and advanced immunotherapies will become more affordable and accessible to more patients. As the situation stands now, a lack of straightforward immunotherapy treatments is handcuffing clinicians while patients continue to die. IL-33 represents a cytokine that may help to fill that void and provide a new option for those who suffer from cancer.

ACKNOWLEDGMENTS

This research was in part supported by National Institutes of Health grants (CA149669, CA208354, and CA222963), the Lynn Sage Cancer Research Foundation, the Melanoma Research Alliance Foundation, a SPOR Pilot Award (P50 CA090386), the Northwestern University RHLCCC Flow Cytometry Facility, a Cancer Center support grant (NCI CA060553), and the National Natural Science Foundation of China (Grant No. 81773046).

REFERENCES

1. Liew FY, Pitman NI, McInnes IB. Disease-associated functions of IL-33: the new kid in the IL-1 family. *Nature Rev Immunol.* 2010;10(2):103–10. [PubMed: 20081870]
2. Oshikawa K, Yanagisawa K, Tominaga S, Sugiyama Y. Expression and function of the ST2 gene in a murine model of allergic airway inflammation. *Clin Exper Allergy.* 2002 10;32(10):1520–6. Epub 2002/10/10. eng. [PubMed: 12372135]
3. Moussion C, Ortega N, Girard JP. The IL-1-like cytokine IL-33 is constitutively expressed in the nucleus of endothelial cells and epithelial cells in vivo: a novel ‘alannin’? *PLoS One.* 2008;3(10):e3331 Epub 2008/10/07. eng. [PubMed: 18836528]
4. Miller AM, Xu D, Asquith DL, Denby L, Li Y, Sattar N, Baker AH, McInnes IB, Liew FY. IL-33 reduces the development of atherosclerosis. *J Exper Med.* 2012;209(13):2515.
5. Humphreys NE, Xu D, Hepworth MR, Liew FY, Grecis RK. IL-33, a potent inducer of adaptive immunity to intestinal nematodes. *J Immunol.* 2008; 180(4):2443–9. [PubMed: 18250453]
6. Jones LA, Roberts F, Nickdel MB, Brombacher F, McKenzie ANJ, Henriquez FL, Alexander J, Roberts CW. IL-33 receptor (T1/ST2) signalling is necessary to prevent the development of encephalitis in mice infected with-Toxoplasma gondii. *Eur J Immunol.* 2010;40(2):426–36. [PubMed: 19950183]
7. Tominaga S-i. A putative protein of a growth specific cDNA from BALB/C-3T3 cells is highly similar to the extracellular portion of mouse interleukin 1 receptor. *FEBS Lett.* 1989;258(2):301–4. [PubMed: 2532153]
8. Schmitz J, Owyang A, Oldham E, Song Y, Murphy E, McClanahan TK, Zurawski G, Moshrefi M, Qin J, Li X, Gorman DM, Bazan JF, Kastelein RA. IL-33, an interleukin-1-like cytokine that signals via the IL-1 receptor-related protein ST2 and induces T helper type 2-associated cytokines. *Immunity.* 2005;23(5):479–90. [PubMed: 16286016]
9. Baekkevoeld ES, Roussigne M, Yamanaka T, Johansen FE, Jahnsen FL, Amalric F, Brandtzaeg P, Erard M, Haraldsen G, Girard JP. Molecular characterization of NF-HEV, a nuclear factor preferentially expressed in human high endothelial venules. *Am J Pathol.* 2003 7;163(1):69–79. Epub 2003/06/24. eng. [PubMed: 12819012]
10. Gordon ED, Simpson LJ, Rios CL, Ringel L, Lachowicz-Scroggins ME, Peters MC, Wesolowska-Andersen A, Gonzalez JR, MacLeod HJ, Christian LS, Yuan S, Barry L, Woodruff PG, Ansel KM,

- Nocka K, Seibold MA, Fahy JV. Alternative splicing of interleukin-33 and type 2 inflammation in asthma. *Proc Natl Acad Sci U S A*. 2016 8 2;113(31):8765–70. Epub 2016/07/20. eng. [PubMed: 27432971]
11. Tsuda H, Komine M, Karakawa M, Etoh T, Tominaga S, Ohtsuki M. Novel splice variants of IL-33: differential expression in normal and transformed cells. *J Investigative Dermatol*. 2012 11; 132(11):2661–4. Epub 2012/07/13. eng.
 12. Roussel L, Erard M, Cayrol C, Girard JP. Molecular mimicry between IL-33 and KSHV for attachment to chromatin through the H2A-H2B acidic pocket. *EMBO Rep*. 2008 10;9(10):1006–12. Epub 2008/08/09. eng. [PubMed: 18688256]
 13. Lingel A, Weiss TM, Niebuhr M, Pan B, Appleton BA, Wiesmann C, Bazan JF, Fairbrother WJ. Structure of IL-33 and its interaction with the ST2 and IL-1RAcP receptors—insight into heterotrimeric IL-1 signaling complexes. *Structure (London: 1993)*. 2009 10 14;17(10):1398–410. Epub 2009/10/20. eng.
 14. Veeraveedu PT, Sanada S, Okuda K, Fu HY, Matsuzaki T, Araki R, Yamato M, Yasuda K, Sakata Y, Yoshimoto T, Minamino T. Ablation of IL-33 gene exacerbates myocardial remodeling in mice with heart failure induced by mechanical stress. *Biochem Pharmacol*. 2017 8 15;138:73–80. Epub 2017/04/30. eng. [PubMed: 28450225]
 15. Clancy DM, Sullivan GP, Moran HBT, Henry CM, Reeves EP, McElvaney NG, Lavelle EC, Martin SJ. Extracellular neutrophil proteases are efficient regulators of IL-1, IL-33, and IL-36 Cytokine activity but poor effectors of microbial killing. *Cell Rep*. 2018 3 13;22(11):2937–50. Epub 2018/03/15. eng. [PubMed: 29539422]
 16. Lefrancais E, Roga S, Gautier V, Gonzalez-de-Peredo A, Monsarrat B, Girard JP, Cayrol C. IL-33 is processed into mature bioactive forms by neutrophil elastase and cathepsin G. *Proc Natl Acad Sci U S A*. 2012 1 31; 109(5):1673–8. Epub 2012/02/07. eng. [PubMed: 22307629]
 17. Cayrol C, Girard JP. The IL-1-like cytokine IL-33 is inactivated after maturation by caspase-1. *Proc Natl Acad Sci U S A*. 2009 6 02;106(22):9021–6. Epub 2009/05/15. eng. [PubMed: 19439663]
 18. Luthi AU, Cullen SP, McNeela EA, Duriez PJ, Afonina IS, Sheridan C, Brumatti G, Taylor RC, Kersse K, Vandenabeele P, Lavelle EC, Martin SJ. Suppression of interleukin-33 bioactivity through proteolysis by apoptotic caspases. *Immunity*. 2009 7 17;31(1):84–98. Epub 2009/06/30. eng. [PubMed: 19559631]
 19. Chackerian AA, Oldham ER, Murphy EE, Schmitz J, Pflanz S, Kastelein RA. IL-1 receptor accessory protein and ST2 comprise the IL-33 receptor complex. *J Immunol*. 2007; 179(4):2551–5. [PubMed: 17675517]
 20. Milovanovic M, Volarevic V, Radosavljevic G, Jovanovic I, Pejnovic N, Arsenijevic N, Lukic ML. IL-33/ST2 axis in inflammation and immunopathology. *Immunol Res*. 2012;52(1-2):89–99. [PubMed: 22392053]
 21. Allakhverdi Z, Smith DE, Comeau MR, Delespesse G. Cutting edge: the ST2 ligand IL-33 potently activates and drives maturation of human mast cells. *J Immunol*. 2007;179(4):2051–4. [PubMed: 17675461]
 22. Ho LH, Ohno T, Oboki K, Kajiwarra N, Suto H, Iikura M, Okayama Y, Akira S, Saito H, Galli SJ, Nakae S. IL-33 induces IL-13 production by mouse mast cells independently of IgE-Fc RI signals. *J Leuk Biol*. 2007;82(6):1481–90.
 23. Gachter T, Werenskiold AK, Klemenz R. Transcription of interleukin-1 receptor-related T1 gene is initiated at different promoters in mast cells and fibroblasts. *J Biol Chem*. 1996 1 5;271(1):124–9. Epub 1996/01/05. eng. [PubMed: 8550546]
 24. Bandara G, Beaven MA, Olivera A, Gilfillan AM, Metcalfe DD. Activated mast cells synthesize and release soluble ST2—a decoy receptor for IL-33. *Eur J Immunol*. 2015 11;45(11):3034–44. Epub 2015/08/11. eng. [PubMed: 26256265]
 25. Zhang J, Ramadan AM, Griesenauer B, Li W, Turner MJ, Liu C, Kapur R, Hanenberg H, Blazar BR, Tawara I, Paczesny S. ST2 blockade reduces sST2-producing T cells while maintaining protective mST2-expressing T cells during graft-versus-host disease. *Sci Trans Med*. 2015 10 7;7(308):308ral60 Epub 2015/10/09. eng.

26. Humphreys NE, Xu D, Hepworth MR, Liew FY, Grecis RK. IL-33, a potent inducer of adaptive immunity to intestinal nematodes. *J Immunol (Baltimore: 1950)*. 2008 2 15;180(4):2443–9. Epub 2008/02/06. eng.
27. Neill DR, Wong SH, Bellosi A, Flynn RJ, Daly M, Langford TK, Bucks C, Kane CM, Fallon PG, Pannell R, Jolin HE, McKenzie AN. Nuocytes represent a new innate effector leukocyte that mediates type-2 immunity. *Nature*. 2010 4 29;464(7293):1367–70. Epub 2010/03/05. eng. [PubMed: 20200518]
28. Hepworth MR, Maurer M, Hartmann S. Regulation of type 2 immunity to helminths by mast cells. *Gut Microbes*. 2012 Sep-Oct; 3(5):476–81. Epub 2012/08/16. eng. [PubMed: 22892692]
29. Allakhverdi Z, Smith DE, Comeau MR, Delespesse G. Cutting edge: the ST2 ligand IL-33 potently activates and drives maturation of human mast cells. *J Immunol (Baltimore, MD: 1950)*. 2007 8 15; 179(4):2051–4. Epub 2007/08/07. eng.
30. Prefontaine D, Lajoie-Kadoch S, Foley S, Audusseau S, Olivenstein R, Halayko AJ, Lemiere C, Martin JG, Hamid Q. Increased expression of IL-33 in severe asthma: evidence of expression by airway smooth muscle cells. *J Immunol (Baltimore, MD: 1950)*. 2009 10 15;183(8):5094–103. Epub 2009/10/06. eng.
31. Sjoberg LC, Nilsson AZ, Lei Y, Gregory JA, Adner M, Nilsson GP. Interleukin 33 exacerbates antigen driven airway hyperresponsiveness, inflammation and remodeling in a mouse model of asthma. *Sci Rep*. 2017 6 26;7(1):4219 Epub 2017/06/28. eng. [PubMed: 28652606]
32. Muehling LM, Lawrence MG, Woodfolk JA. Pathogenic CD4(+) T cells in patients with asthma. *J Allergy Clin Immunol*. 2017 12; 140(6):1523–40. Epub 2017/04/27. eng. [PubMed: 28442213]
33. Xu D, Chan WL, Leung BP, Huang F, Wheeler R, Piedrafita D, Robinson JH, Liew FY. Selective expression of a stable cell surface molecule on type 2 but not type 1 helper T cells. *J Exper Med*. 1998 3 2;187(5):787–94. Epub 1998/03/28. eng. [PubMed: 9480988]
34. Peine M, Marek RM, Lohning M. IL-33 in T cell differentiation, function, and immune homeostasis. *Trends Immunol*. 2016 5;37(5):321–33. Epub 2016/04/09. eng. [PubMed: 27055914]
35. Smithgall MD, Comeau MR, Yoon BR, Kaufman D, Annitage R, Smith DE. IL-33 amplifies both Th1- and Th2-type responses through its activity on human basophils, allergen-reactive Th2 cells, iNKT and NK cells. *Int Immunol*. 2008 8;20(8):1019–30. Epub 2008/06/14. eng. [PubMed: 18550585]
36. Bourgeois E, Van LP, Samson M, Diem S, Barra A, Roga S, Gombert JM, Schneider E, Dy M, Gourdy P, Girard JP, Herbelin A. The pro-Th2 cytokine IL-33 directly interacts with invariant NKT and NK cells to induce IFN-gamma production. *Eur J Immunol*. 2009 4;39(4):1046–55. Epub 2009/03/07. eng. [PubMed: 19266498]
37. Nabekura T, Girard JP, Lanier LL. IL-33 receptor ST2 amplifies file expansion of NK cells and enhances host defense during mouse cytomegalovirus infection. *J Immunol (Baltimore, MD: 1950)*. 2015 6 15;194(12):5948–52. Epub 2015/05/01. eng.
38. Scott IC, Majithiya JB, Sanden C, Thornton P, Sanders PN, Moore T, Guscott M, Corkill DT, Erjefalt JS, Cohen ES. Interleukin-33 is activated by allergen- and necrosis-associated proteolytic activities to regulate its alannin activity during epithelial damage. *Sci Rep*. 2018 2 20;8(1):3363 Epub 2018/02/22. eng. [PubMed: 29463838]
39. Sesti-Costa R, Silva GK, Proenca-Modena JL, Carlos D, Silva ML, Alves-Filho JC, Arruda E, Liew FY, Silva JS. The IL-33/ST2 pathway controls coxsackievirus B5-induced experimental pancreatitis. *J Immunol (Baltimore, MD: 1950)*. 2013 7 1; 191 (1):283–92. Epub 2013/06/05. eng.
40. Kayamuro H, Yoshioka Y, Abe Y, Arita S, Katayama K, Nomura T, Yoshikawa T, Kubota-Koketsu R, Ikuta K, Okamoto S, Mori Y, Kimisawa J, Kiyono H, Itoh N, Nagano K, Kamada H, Tsutsumi Y, Tsunoda S. Interleukin-1 family cytokines as mucosal vaccine adjuvants for induction of protective immunity against influenza virus. *J Virol*. 2010 12;84(24):12703–12. Epub 2010/10/01. eng. [PubMed: 20881038]
41. Reichenbach DK, Schwarze V, Matta BM, Tkachev V, Lieberknecht E, Liu Q, Koehn BH, Pfeifer D, Taylor PA, Prinz G, Dierbach H, Stickel N, Beck Y, Warncke M, Junt T, Schmitt-Graeff A, Nakae S, Follo M, Wertheimer T, Schwab L, Devlin J, Watkins SC, Duyster J, Ferrara JL, Turnquist HR, Zeiser R, Blazar BR. The IL-33/ST2 axis augments effector T-cell responses during acute GVHD. *Blood*. 2015 5 14; 125(20):3183–92. Epub 2015/03/31. eng. [PubMed: 25814531]

42. Villarreal DO, Svoronos N, Wise MC, Shedlock DJ, Morrow MP, Conejo-Garcia JR, Weiner DB. Molecular adjuvant IL-33 enhances the potency of a DNA vaccine in a lethal challenge model. *Vaccine*. 2015 8 20;33(35):4313–20. Epub 2015/04/19. eng. [PubMed: 25887087]
43. Kearley J, Silver JS, Sanden C, Liu Z, Berlin AA, White N, Mori M, Pham TH, Ward CK, Criner GJ, Marchetti N, Mustelin T, Erjefalt JS, Kolbeck R, Humbles AA. Cigarette smoke silences innate lymphoid cell function and facilitates an exacerbated type I interleukin-33-dependent response to infection. *Immunity*. 2015 3 17;42(3):566–79. Epub 2015/03/19. eng. [PubMed: 25786179]
44. Bonilla WV, Frohlich A, Senn K, Kallert S, Fernandez M, Johnson S, Kreutzfeldt M, Hegazy AN, Schrick C, Fallon PG, Klemenz R, Nakae S, Adler H, Merkler D, Lohning M, Pinschewer DD. The alarmin interleukin-33 drives protective antiviral CD8+ T cell responses. *Science*. 2012;335(6071):984–9. [PubMed: 22323740]
45. Braumuller H, Wieder T, Brenner E, Assmann S, Hahn M, Alkhaled M, Schilbach K, Essmann F, Kneilling M, Griessinger C, Ranta F, Ullrich S, Mocikat R, Braungart K, Mehra T, Fehrenbacher B, Berdel J, Niessner H, Meier F, van den Broek M, Haring HU, Handgretinger R, Quintanilla-Martinez L, Fend F, Pesic M, Bauer J, Zender L, Schaller M, Schulze-Osthoff K, Rocken M. T-helper-1-cell cytokines drive cancer into senescence. *Nature*. 2013 2 21;494(7437):361–5. Epub 2013/02/05. eng. [PubMed: 23376950]
46. Smyth MT, Thia KY, Street SE, Cretney E, Trapani JA, Taniguchi M, Kawano T, Pelikan SB, Crowe NY, Godfrey DI. Differential tumor surveillance by natural killer (NK) and NKT cells. *J Exper Med*. 2000 2 21;191(4):661–8. Epub 2000/02/24. eng. [PubMed: 10684858]
47. Banow AD, Edeling MA, Trifonov V, Luo J, Goyal P, Bohl B, Bando JK, Kim AH, Walker J, Andahazy M, Bugatti M, Melocchi L, Vermi W, Fremont DH, Cox S, Cella M, Schmedt C, Colonna M. Natural killer cells control tumor growth by sensing a growth factor. *Cell*. 2018 1 25;172(3):534–48.e19. Epub 2017/12/26. eng. [PubMed: 29275861]
48. Galon J, Costes A, Sanchez-Cabo F, Kirilovsky A, Mlecnik B, Lagorce-Pages C, Tosolini M, Camus M, Berger A, Wind P, Zinzindohoue F, Bruneval P, Cugnenc PH, Trajanoski Z, Fridman WH, Pages F. Type, density, and location of immune cells within human colorectal tumors predict clinical outcome. *Science*. 2006 9 29;313(5795):1960–4. Epub 2006/09/30. eng. [PubMed: 17008531]
49. Fridman WH, Pages F, Sautes-Fridman C, Galon J. The immune contexture in human tumours: impact on clinical outcome. *Nature Rev Cancer*. 2012 3 15;12(4):298–306. Epub 2012/03/16. eng. [PubMed: 22419253]
50. Ngoi SM, St Rose MC, Menoret AM, Smith DE, Tovey MG, Adler AJ, Vella AT. Presensitizing with a Toll-like receptor 3 ligand impairs CD8 T-cell effector differentiation and IL-33 responsiveness. *Proc Natl Acad Sci U S A*. 2012 6 26; 109(26):10486–91. Epub 2012/06/13. eng. [PubMed: 22689946]
51. Yang Q, Li G, Zhu Y, Liu L, Chen E, Turnquist H, Zhang X, Finn OJ, Chen X, Lu B. IL-33 synergizes with TCR and IL-12 signaling to promote the effector function of CD8+ T cells. *Eur J Immunol*. 2011 11;41(11):3351–60. Epub 2011/09/03. eng. [PubMed: 21887788]
52. Baumann C, Bonilla WV, Frohlich A, Helmstetter C, Peine M, Hegazy AN, Pinschewer DD, Lohning M. T-bet- and STAT4-dependent IL-33 receptor expression directly promotes antiviral Th1 cell responses. *Proc Natl Acad Sci U S A*. 2015 3 31; 112(13):4056–61. Epub 2015/04/02. eng. [PubMed: 25829541]
53. Dominguez D, Ye C, Geng Z, Chen S, Fan J, Qin L, Long A, Wang L, Zhang Z, Zhang Y, Fang D, Kuzel TM, Zhang B. Exogenous IL-33 restores dendritic cell activation and maturation in established cancer. *J Immunol (Baltimore, MD: 1950)*. 2017 2 01;198(3):1365–75. Epub 2016/12/25. eng.
54. Blom L, Poulsen LK. IL-1 family members IL-18 and IL-33 upregulate the inflammatory potential of differentiated human Th1 and Th2 cultures. *J Immunol (Baltimore, MD: 1950)*. 2012 11 1; 189(9):4331–7. Epub 2012/10/03. eng.
55. Hegazy AN, Peine M, Helmstetter C, Panse I, Frohlich A, Bergthaler A, Flatz L, Pinschewer DD, Radbruch A, Lohning M. Interferons direct Th2 cell reprogramming to generate a stable GATA-3(+)T-bet(+) cell subset with combined Th2 and Th1 cell functions. *Immunity*. 2010 1 29;32(1):116–28. Epub 2010/01/19. eng. [PubMed: 20079668]

56. Morgan DA, Ruscetti FW, Gallo R. Selective in vitro growth of T lymphocytes from normal human bone marrows. *Science*. 1976 9 10;193(4257):1007–8. Epub 1976/09/10. [PubMed: 181845]
57. Rosenberg SA, Lotze MT, Muul LM, Leitman S, Chang AE, Ettinghausen SE, Matory YL, Skibber JM, Shiloni E, Vetto JT, Seipp CA. Observations on the systemic administration of autologous lymphokine-activated killer cells and recombinant interleukin-2 to patients with metastatic cancer. *N Engl J Med*. 1985 12 05;313(23):1485–92. Epub 1985/12/05. [PubMed: 3903508]
58. Rudman SM, Jameson MB, McKeage MJ, Savage P, Jodrell DI, Harries M, Acton G, Erlandsson F, Spicer JF. A phase I study of AS1409, a novel antibody-cytokine fusion protein, in patients with malignant melanoma or renal cell carcinoma. *Clin Cancer Res*. 2011 4 01; 17(7):1998–2005. Epub 2011/03/31. [PubMed: 21447719]
59. Gao K, Li X, Zhang L, Bai L, Dong W, Gao K, Shi G, Xia X, Wu L, Zhang L. Transgenic expression of IL-33 activates CD8+ T cells and NEC cells and inhibits tumor growth and metastasis in mice. *Cancer Lett*. 2013;335(2):463–71. [PubMed: 23499895]
60. Gao X, Wang X, Yang Q, Zhao X, Wen W, Li G, Lu J, Qin W, Qi Y, Xie F, Jiang J, Wu C, Zhang X, Chen X, Turnquist H, Zhu Y, Lu B. Tumoral expression of IL-33 inhibits tumor growth and modifies the tumor microenvironment through CD8+T and NEC cells. *J Immunol*. 2014; 194(1):438–45. [PubMed: 25429071]
61. Villarreal DO, Wise MC, Walters JN, Reuschel EL, Choi MT, Obeng-Adjei N, Yan J, Morrow MP, Weiner DB. Alarmin IL-33 acts as an immunoadjuvant to enhance antigen-specific tumor immunity. *Cancer Res*. 2014 3 15;74(6):1789–800. Epub 2014/01/23. eng. [PubMed: 24448242]
62. Rose WA 2nd, Okragly AJ, Patel CN, Benschop RJ. IL-33 released by alum is responsible for early cytokine production and has adjuvant properties. *Sci Rep*. 2015 8 14;5:13146 Epub 2015/08/15. eng. [PubMed: 26272855]
63. Kallert SM, Darbre S, Bonilla WV, Kreutzfeldt M, Page N, Muller P, Kreuzaler M, Lu M, Favre S, Kreppel F, Lohning M, Luther SA, Zippelius A, Merkler D, Pinschewer DD. Replicating viral vector platform exploits alarmin signals for potent CD8(+) T cell-mediated tumour immunotherapy. *Nature Commun*. 2017 5 26;8:15327 Epub 2017/05/27. eng. [PubMed: 28548102]
64. Dankort D, Curley DP, Carlidge RA, Nelson B, Karnezis AN, Damsky WE Jr, You MT, DePinho RA, McMahon M, Bosenberg M. BrafV600E cooperates with Pten loss to induce metastatic melanoma. *Nature Genetics*. 2009; 41(5):544–52. [PubMed: 19282848]
65. Hooijkaas AI, Gadiot J, van der Valk M, Mooi WJ, Blank CU. Targeting BRAFV600E in an inducible murine model of melanoma. *Am J Pathol*. 2012;181(3):785–94. [PubMed: 22796458]
66. Dranoff G, Jaffee E, Lazenby A, Golumbek P, Levitsky H, Brose K, Jackson V, Hamada H, Pardoll D, Mulligan RC. Vaccination with irradiated tumor cells engineered to secrete murine granulocyte-macrophage colony-stimulating factor stimulates potent, specific, and long-lasting anti-tumor immunity. *Proc Natl Acad Sci U S A*. 1993 4 15;90(8):3539–43. Epub 1993/04/15. eng. [PubMed: 8097319]
67. Seliger B, Wollscheid U, Momburg F, Blankenstein T, Huber C. Characterization of the major histocompatibility complex class I deficiencies in B16 melanoma cells. *Cancer Res*. 2001 2 1;61(3):1095–9. Epub 2001/02/28. eng. [PubMed: 11221838]
68. Qin L, Dominguez D, Chen S, Fan J, Long A, Zhang M, Fang D, Zhang Y, Kuzel TM, Zhang B. Exogenous IL-33 overcomes T cell tolerance in murine acute myeloid leukemia. *Oncotarget*. 2016 9 20;7(38):61069–80. Epub 2016/08/16. eng. [PubMed: 27517629]
69. Mayuzumi N, Matsushima H, Takashima A. IL-33 promotes DC development in BM culture by triggering GM-CSF production. *Eur J Immunol*. 2009 2009;39(12):3331–42. [PubMed: 19750479]
70. Rank MA, Kobayashi T, Kozaki H, Bartemes KR, Squillace DL, Kita H. IL-33-activated dendritic cells induce an atypical TH2-type response. *J Allergy Clin Immunol*. 2009; 123(5):1047–54. [PubMed: 19361843]
71. Besnard A-G, Togbe D, Guillou N, Erard F, Quesniaux V, Ryffel B. IL-33-activated dendritic cells are critical for allergic airway inflammation. *Eur J Immunol*. 2011 2011/5/25;41(6):1675–86. [PubMed: 21469105]

72. Liang Y, Jie Z, Hou L, Yi P, Wang W, Kwota Z, Salvato M, de Waal Malefyt R, Soong L, Sim J. IL-33 promotes innate IFN-gamma production and modulates dendritic cell response in LCMV-induced hepatitis in mice. *Eur J Immunol*. 2015 11;45(11):3052–63. Epub 2015/08/08. eng. [PubMed: 26249267]
73. vail Mierlo GJD, Boonman ZFHM, Dumortier HMH, den Boer AT, Franssen MF, Nouta J, van der Voort EIH, Offringa R, Toes REM, Melief CJM. Activation of dendritic cells that cross-present tumor-derived antigen licenses CD8+ CTL to cause tumor eradication. *J Immunol*. 2004; 173(11):6753–9. [PubMed: 15557168]
74. Albert ML, Pearce SFA, Francisco LM, Sauter B, Roy P, Silverstein RL, Bhardwaj N. Immature Dendritic cells phagocytose apoptotic cells via $\alpha\beta 5$ and CD36, and cross-present antigens to cytotoxic T lymphocytes. *J Exper Med*. 1998;188(7):1359–68. [PubMed: 9763615]
75. Hildner K, Edelson BT, Purtila WE, Diamond M, Matsushita H, Kohyama M, Calderon B, Schraml BU, Unanue ER, Diamond MS, Schreiber RD, Murphy TL, Murphy KM. Batf3 deficiency reveals a critical role for CD8 α + dendritic cells in cytotoxic T cell immunity. *Science*. 2008 11 14;322(5904):1097–100. Epub 2008/11/15. eng. [PubMed: 19008445]
76. Lin A, Schildknecht A, Nguyen LT, Ohashi PS. Dendritic cells integrate signals from the tumor microenvironment to modulate immunity and tumor growth. *Immunol Lett*. 2010; 127(2):77–84. [PubMed: 19778555]
77. Gabrilovich D. Mechanisms and functional significance of tumour-induced dendritic-cell defects. *Nature Rev Immunol*. 2004;4(12):941–52. [PubMed: 15573129]
78. Jackson SH, Yu CR, Mahdi RM, Ebong S, Egwuagu CE. Dendritic cell maturation requires STAT1 and is under feedback regulation by suppressors of cytokine signaling. *J Immunol*. 2004 ; 172(4):2307–15. [PubMed: 14764699]
79. Johnson LM, Scott P. STAT1 expression in dendritic cells, but not T cells, is required for immunity to *Leishmania major*. *J Immunol*. 2007; 178(11):7259–66. [PubMed: 17513775]
80. Pilz A, Kratky W, Stockinger S, Simma O, Kalinke U, Lingnau K, von Gabain A, Stoiber D, Sexl V, Kolbe T, Rulicke T, Muller M, Decker T. Dendritic cells require STAT-1 Phosphorylated at its transactivating domain for file induction of peptide-specific CTL. *J Immunol*. 2009;183(4):2286–93. [PubMed: 19620292]
81. Ramadan A, Griesenauer B, Adom D, Kapur R, Hanenberg H, Liu C, Kaplan MH, Paczesny S. Specifically differentiated T cell subset promotes tumor immunity over fatal immunity. *J Exper Med*. 2017 12 4;214(12):3577–96. Epub 2017/10/19. eng. [PubMed: 29038366]
82. Duault C, Franchini DM, Famillides J, Cayrol C, Roga S, Girard JP, Fournie JJ, Poupot M. TCRVgamma9 gammadelta T cell response to IL-33: A CD4 T cell-dependent mechanism. *J Immunol (Baltimore, MD: 1950)*. 2016 1 1; 196(1):493–502. Epub 2015/11/27. eng.
83. Lucarini V, Ziccheddu G, Macchia I, La Sorsa V, Peschiaroli F, Buccione C, Sistigu A, Sanchez M, Andreone S, D'Urso MT, Spada M, Macchia D, Afferni C, Mattei F, Schiavoni G. IL-33 restricts tumor growth and inhibits pulmonary metastasis in melanoma-bearing mice through eosinophils. *Oncoimmunology*. 2017;6(6):e1317420 Epub 2017/07/07. eng.
84. Saranchova I, Han J, Huang H, Fenninger F, Choi KB, Munro L, Pfeifer C, Welch I, Wyatt AW, Fazli L, Gleave ME, Jefferies WA. Discovery of a metastatic immune escape mechanism initiated by the loss of expression of file tumour biomarker interleukin-33. *Sci Rep*. 2016 9 13;6:30555 Epub 2016/09/14. eng. [PubMed: 27619158]
85. Kim J, Kim W, Moon UJ, Kim HJ, Choi HJ, Sin JI, Park NH, Cho HR, Kwon B. Intratumorally establishing Type 2 Innate lymphoid cells blocks tumor growth. *J Immunol (Baltimore, MD: 1950)*. 2016 3 1;196(5):2410–23. Epub 2016/02/03.
86. Long A, Dominguez D, Qin L, Chen S, Fan J, Zhang M, Fang D, Zhang Y, Kuzel TM, Zhang B. Type 2 Innate lymphoid cells impede IL-33-mediated tumor suppression. *J Immunol (Baltimore, MD: 1950)*. 2018 10 29; 201(11):3456–64. Epub 2018/10/31. eng.
87. Jovanovic I, Radosavljevic G, Mitrovic M, Lisnic Juranic V, McKenzie ANJ, Arsenijevic N, Jonjic S, Lukic ML. ST2 deletion enhances innate and acquired immunity to murine mammary carcinoma. *Eur J Immunol*. 2011;41(7):1902–12. [PubMed: 21484786]

88. Kim JY, Lim SC, Kim G, Yun HJ, Ahn SG, Choi HS. Interleukin-33/ST2 axis promotes epithelial cell transformation and breast tumorigenesis via upregulation of COT activity. *Oncogene*. 2014;34(38):4928–38. [PubMed: 25531326]
89. Jovanovic IP, Pejnovic NN, Radosavljevic GD, Pantic JM, Milovanovic MZ, Arsenijevic NN, Lukic ML. Interleukin-33/ST2 axis promotes breast cancer growth and metastases by facilitating intratumoral accumulation of immunosuppressive and innate lymphoid cells. *Int J Cancer*. 2014 4 1;134(7):1669–82. Epub 2013/10/10. eng. [PubMed: 24105680]
90. Milosavljevic MZ, Jovanovic IP, Pejnovic NN, Mitrovic SL, Arsenijevic NN, Simovic Markovic BJ, Lukic ML. Deletion of IL-33R attenuates VEGF expression and enhances necrosis in mammary carcinoma. *Oncotarget*. 2016 4 5;7(14):18106–15. Epub 2016/02/27. eng. [PubMed: 26919112]
91. Jovanovic IP, Pejnovic NN, Radosavljevic GD, Pantic JM, Milovanovic MZ, Arsenijevic NN, Lukic ML. Interleukin-33/ST2 axis promotes breast cancer growth and metastases by facilitating intratumoral accumulation of immunosuppressive and innate lymphoid cells. *Int J Cancer*. 2013;134(7):1669–82. [PubMed: 24105680]
92. Lim HX, Choi S, Cho D, Kim TS. IL-33 inhibits the differentiation and immunosuppressive activity of granulocytic myeloid-derived suppressor cells in tumor-bearing mice. *Immunol Cell Biol*. 2017 1;95(1):99–107. Epub 2016/08/11. eng. [PubMed: 27507556]
93. Fang Y, Zhao L, Xiao H, Cook KM, Bai Q, Herrick EJ, Chen X, Qin C, Zhu Z, Wakefield MR, Nicholl MB. IL-33 acts as a foe to MIA PaCa-2 pancreatic cancer. *Med Oncol (Northwood, London)*. 2017 2;34(2):23 Epub 2017/01/07. eng.
94. Akimoto M, Hayashi JI, Nakae S, Saito H, Takenaga K. Interleukin-33 enhances programmed oncosis of ST2L-positive low-metastatic cells in the tumour microenvironment of lung cancer. *Cell Death Disease*. 2016 1 21;7:e2057 Epub 2016/01/19. eng. [PubMed: 26775708]
95. Brunner SM, Rubner C, Kesselring R, Martin M, Griesshammer E, Ruemmele P, Stempf T, Teufel A, Schlitt HJ, Fichtner-Feigl S. Tumor-infiltrating, inter-leukin-33-producing effector-memory CD8(+) T cells in resected hepatocellular carcinoma prolong patient survival. *Hepatology (Baltimore)*. 2015 6;61(6):1957–67. Epub 2015/02/04. eng. [PubMed: 25645298]
96. Yang Y, Wang JB, Li YM, Zhao YU, Wang R, Wu Q, Zheng RS, Ou YR. Role of IL-33 expression in oncogenesis and development of human hepatocellular carcinoma. *Oncol Lett*. 2016 7;12(1):429–36. Epub 2016/06/28. eng. [PubMed: 27347162]
97. Le Pape F, Vargas G, Clezardin P. The role of osteoclasts in breast cancer bone metastasis. *J Bone Oncol*. 2016 9;5(3):93–5. Epub 2016/10/21. eng. [PubMed: 27761364]
98. Koster R, Panagiotou OA, Wheeler WA, Karlins E, Gastier-Foster JM, Caminada de Toledo SR, Petrilli AS, Flanagan AM, Tirabosco R, Andrulis IL, Wunder JS, Gokgoz N, Patino-Garcia A, Lecanda F, Serra M, Hattinger C, Picci P, Scotlandi K, Thomas DM, Ballinger ML, Gorlick R, Barkauskas DA, Spector LG, Tucker M, Belsynda DH, Yeager M, Hoover RN, Wacholder S, Chanock SJ, Savage SA, Mirabello L. Genome-wide association study identifies file GLDC/IL33 locus associated with survival of osteosarcoma patients. *Int J Cancer*. 2018 4 15; 142(8):1594–601. Epub 2017/12/07. eng. [PubMed: 29210060]
99. Lima IL, Macari S, Madeira MF, Rodrigues LF, Colavite PM, Garlet GP, Soriani FM, Teixeira MM, Fukada SY, Silva TA. Osteoprotective effects of IL-33/ST2 link to osteoclast apoptosis. *Am J Pathol*. 2015 12;185(12):3338–48. Epub 2015/11/26. eng. [PubMed: 26598236]
100. Musolino C, Allegra A, Profita M, Alonci A, Saitta S, Bonanno A, Gerace D, Calabro L, Gangemi S. Reduction in IL-33 plasma levels might be involved in T cell dysregulation in chronic lymphocytic leukemia. *Acta Haematol*. 2014;131(3):165–6 Epub 2013/11/13. eng. [PubMed: 24217710]
101. Musolino C, Allegra A, Profita M, Alonci A, Saitta S, Russo S, Bonanno A, Innao V, Gangemi S. Reduced IL-33 plasma levels in multiple myeloma patients are associated with more advanced stage of disease. *British J Haematol*. 2013 3;160(5):709–10. Epub 2012/12/05. eng.
102. Yang M, Feng Y, Yue C, Xu B, Chen L, Jiang J, Lu B, Zhu Y. Lower expression level of IL-33 is associated with poor prognosis of pulmonary adenocarcinoma. *PLoS One*. 2018;13(3):e0193428 Epub 2018/03/03. eng. [PubMed: 29499051]
103. Barrera L, Montes-Servin E, Barrera A, Ramirez-Tirado LA, Salinas-Parra F, Banales-Mendez JL, Sandoval-Rios M, Arrieta O. Cytokine profile determined by data-mining analysis set into

- clusters of non-small-cell lung cancer patients according to prognosis. *Annals Oncol.* 2015 2;26(2):428–35. Epub 2014/12/04. eng.
104. O'Donnell C, Mahmoud A, Keane J, Murphy C, White D, Carey S, O'Riordain M, Bennett MW, Brint E, Houston A. An antitumorigenic role for the IL-33 receptor, ST2L, in colon cancer. *British J Cancer.* 2016 1 12;114(1):37–43. Epub 2015/12/19. eng.
 105. Eissmami MF, Dijkstra C, Wouters MA, Baloyan D, Mouradov D, Nguyen PM, Davalos-Salas M, Putoczki TL, Sieber OM, Mariadason JM, Ernst M, Masson F. Interleukin 33 signaling restrains sporadic colon cancer in an interferon-gamma-dependent manner. *Cancer Immunol Res.* 2018 2 20 Epub 2018/02/22. eng.
 106. Lock FE, Babaian A, Zhang Y, Gagnier L, Kuali S, Weberling A, Karimi MM, Mager DL. A novel isoform of IL-33 revealed by screening for transposable element promoted genes in human colorectal cancer. *PLoS One.* 2017; 12(7):e0180659 Epub 2017/07/18. eng. [PubMed: 28715472]
 107. Wang L, Li H, Liang F, Hong Y, Jiang S, Xiao L. Examining IL-33 expression in the cervix of HPV-infected patients: a preliminary study comparing IL-33 levels in different stages of disease and analyzing its potential association with IFN-gamma. *Med Oncol (Northwood, London).* 2014 9;31(9):143 Epub 2014/08/12. eng.
 108. Rossle M, Cathomas G, Bonapace L, Sachs M, Dehler S, Storz M, Huber G, Moch H, Junt T, Mertz KD. Interleukin-33 expression indicates a favorable prognosis in malignant salivary gland tumors. *Int J Surg Pathol.* 2016 8;24(5):394–400. [PubMed: 26912475]
 109. Bergis D, Kassis V, Radeke HH. High plasma sST2 levels in gastric cancer and their association with metastatic disease. *Disease Markers A.* 2016; 16(1):117–25. Epub 2016/02/03. eng.
 110. Lu DP, Zhou XY, Yao LT, Liu CG, Ma W, Jin F, Wu YF. Serum soluble ST2 is associated with ER-positive breast cancer. *BMC Cancer.* 2014Mar 18;14:198 Epub 2014/03/19. eng. [PubMed: 24636276]
 111. Yang ZP, Ling DY, Xie YH, Wu WX, Li JR, Jiang J, Zheng JL, Fan YH, Zhang Y. The association of serum IL-33 and sST2 with breast cancer. *Disease Markers.* 2015;2015:516895 Epub 2015/10/13. eng. [PubMed: 26456994]
 112. Oshikawa K, Yanagisawa K, Ohno S, Tominaga S, Sugiyama Y. Expression of ST2 in helper T lymphocytes of malignant pleural effusions. *Am J Resp Crit Care Med.* 2002 4 1; 165(7):1005–9. Epub 2002/04/06. eng. [PubMed: 11934729]
 113. Levescot A, Flamant S, Basbous S, Jacomet F, Feraud O, Anne Bourgeois E, Bonnet ML, Giraud C, Roy L, Barra A, Chomel JC, Turhan A, Guilhot F, Girard JP, Gombert JM, Herbelin A. BCR-ABL-induced deregulation of the IL-33/ST2 pathway in CD34+ progenitors from chronic myeloid leukemia patients. *Cancer Res.* 2014May 15;74(10):2669–76. Epub 2014/03/29. eng. [PubMed: 24675360]
 114. Lecart S, Lecoite N, Subramaniam A, Alkan S, Ni D, Chen R, Boulay V, Pene J, Kuroiwa K, Tominaga S, Yssel H. Activated, but not resting human Th2 cells, in contrast to Th1 and T regulatory cells, produce soluble ST2 and express low levels of ST2L at the cell surface. *Eur J Immunol.* 2002 10;32(10):2979–87. Epub 2002/10/02. eng. [PubMed: 12355452]
 115. Hayakawa H, Hayakawa M, Kume A, Tominaga S. Soluble ST2 blocks interleukin-33 signaling in allergic airway inflammation. *J Biol Chem.* 2007 9 7;282(36):26369–80. Epub 2007/07/12. eng. [PubMed: 17623648]
 116. Mehraj V, Jenabian MA, Ponte R, Lebouche B, Costiniuk C, Thomas R, Baril JG, LeBlanc R, Cox J, Tremblay C, Routy JP. The plasma levels of soluble ST2 as a marker of gut mucosal damage in early HIV infection. *AIDS (London).* 2016 6 19;30(10):1617–27. Epub 2016/04/06. eng.
 117. Miyagaki T, Sugaya M, Yokobayashi H, Kato T, Ohmatsu H, Fujita H, Saeki H, Kikuchi Y, Tamaki T, Sato S. High levels of soluble ST2 and low levels of IL-33 in sera of patients with HIV infection. *J Invest Dermatol.* 2011 3;131(3):794–6. Epub 2010/12/15. eng. [PubMed: 21150924]
 118. Tago K, Ohta S, Kashiwada M, Funakoshi-Tago M, Matsugi J, Tominaga SI, Yanagisawa K. ST2 gene products critically contribute to cellular transformation caused by an oncogenic Ras mutant. *Heliyon.* 2017 10;3(10):e00436 Epub 2017/12/12. eng. [PubMed: 29226265]

119. Tominaga SI, Ohta S, Tago K. Soluble form of the ST2 gene product exhibits growth promoting activity in NIH-3T3 cells. *Biochem Biophys Rep.* 2016 3;5:8–15. Epub 2015/11/21. eng. [PubMed: 28955802]
120. Tominaga S, Tago K, Tsuda H, Komine M. Dual function of IL-33 on proliferation of NIH-3T3 cells. *Cytokine.* 2015 3;72(1):105–8. Epub 2015/01/13. eng. [PubMed: 25573803]
121. Zhang P, Liu XK, Chu Z, Ye JC, Li KL, Zhuang WL, Yang DJ, Jiang YF. Detection of interleukin-33 in serum and carcinoma tissue from patients with hepatocellular carcinoma and its clinical implications. *J Int Med Res.* 2012;40(5):1654–61. Epub 2012/12/05. eng. [PubMed: 23206447]
122. Cohen ES, Scott IC, Majithiya JB, Rapley L, Kemp BP, England E, Rees DG, Overed-Sayer CL, Woods J, Bond NJ, Veyssier CS, Embrey KJ, Sims DA, Snaith MR, Vousden KA, Strain MD, Chan DT, Carmen S, Huntington CE, Flavell L, Xu J, Popovic B, Brightling CE, Vaughan TJ, Butler R, Lowe DC, Higazi DR, Corkill DJ, May RD, Sleeman MA, Mustelin T. Oxidation of the alannin IL-33 regulates ST2-dependent inflammation. *Nature Commun.* 2015 9 14;6:8327 Epub 2015/09/15. eng. [PubMed: 26365875]
123. Rivers-Auty J, Daniels MJD, Colliver I, Robertson DL, Brough D. Redefining the ancestral origins of the interleukin-1 superfamily. *Nature Commun.* 2018 3 20;9(1):1156. [PubMed: 29559685]
124. Rosenberg SA, Packard BS, Aebersold PM, Solomon D, Topalian SL, Toy ST, Simon P, Lotze MT, Yang JC, Seipp CA, Simpson C. Use of tumor-infiltrating lymphocytes and interleukin-2 in the immunotherapy of patients with metastatic melanoma. A preliminary report. *N Engl J Med.* 1988 12 22;319(25):1676–80. Epub 1988/12/22. eng. [PubMed: 3264384]
125. Bajetta E, Del Vecchio M, Mortarini R, Nadeau R, Rakhit A, Rimassa L, Fowst C, Borri A, Anichini A, Panniani G. Pilot study of subcutaneous recombinant human interleukin 12 in metastatic melanoma. *Clin Cancer Res.* 1998 1;4(1):75–85. Epub 1998/05/14. eng. [PubMed: 9516955]
126. Xu D, Jiang HR, Li Y, Pushparaj PN, Kurowska-Stolarska M, Leung BP, Mu R, Tay HK, McKenzie AN, McInnes IB, Melendez AJ, Liew FY. IL-33 exacerbates autoantibody-induced arthritis. *J Immunol (Baltimore: 1950).* 2010 3 1;184(5):2620–6. Epub 2010/02/09. eng.
127. Imai Y, Yasuda K, Sakaguchi Y, Haneda T, Mizutani H, Yoshimoto T, Nakanishi K, Yamanishi K. Skin-specific expression of IL-33 activates group 2 innate lymphoid cells and elicits atopic dermatitis-like inflammation in mice. *Proc Natl Acad Sci U S A.* 2013 8 20; 110(34):13921–6. Epub 2013/08/07. eng. [PubMed: 23918359]
128. McHedlidze T, Waldner M, Zopf S, Walker J, Rankin AL, Schuchmann M, Voehringer D, McKenzie AN, Neurath MF, Pflanz S, Wirtz S. Interleukin-33-dependent innate lymphoid cells mediate hepatic fibrosis. *Immunity.* 2013 8 22;39(2):357–71. Epub 2013/08/21. eng. [PubMed: 23954132]
129. Matta BM, Reichenbach DK, Zhang X, Mathews L, Koehn BH, Dwyer GK, Lott JM, Uhl FM, Pfeifer D, Feser CJ, Smith MJ, Liu Q, Zeiser R, Blazar BR, Turnquist HR. Peri-alloHCT IL-33 administration expands recipient T-regulatory cells that protect mice against acute GVHD. *Blood.* 2016 7 21;128(3):427–39. [PubMed: 27222477]
130. Chen Y, Lu B. Guided delivery of the “alarming” cytokine IL-33 to tumor by chimeric antigen receptor T cells. *J Immunol.* 2017;198(Suppl 1):204:23–23.
131. Ramadan AM, Daguindau E, Rech JC, Chinnaswamy K, Zhang J, Hura GL, Griesenauer B, Bolten Z, Robida A, Larsen M, Stuckey JA, Yang CY, Paczesny S. From proteomics to discovery of first-in-class ST2 inhibitors active in vivo. *JCI Insight.* 2018 7 25;3(14).
132. Sharma P, Allison JP. The future of immune checkpoint therapy. *Science.* 2015 4 3;348(6230):56–61. Epub 2015/04/04. eng. [PubMed: 25838373]
133. Schmieder A, Multhoff G, Radons T. Interleukin-33 acts as a pro-inflammatory cytokine and modulates its receptor gene expression in highly metastatic human pancreatic carcinoma cells. *Cytokine.* 2012 11;60(2):514–21 Epub 2012/07/24. eng. [PubMed: 22819319]
134. Chen SF, Nieh S, Jao SW, Wu MZ, Liu CL, Chang YC, Lin YS. The paracrine effect of cancer-associated fibroblast-induced interleukin-33 regulates the invasiveness of head and neck squamous cell carcinoma. *J Pathol.* 2013 10;231(2):180–9. Epub 2013/06/19. eng. [PubMed: 23775566]

135. Kim MS, Kim E, Heo JS, Bae DJ., Lee JU, Lee TH, Lee HJ, Chang HS, Park JS, Jang AS, Koh ES, Hwang HG, Lim G, Kim S, Park CS. Circulating IL-33 level is associated with file progression of lung cancer. *Lung Cancer (Amsterdam)*. 2015 11;90(2):346–51. Epub 2015/09/08. eng.
136. Wang K, Shan S, Yang Z, Gu X, Wang Y, Wang C, Ren T. IL-33 blockade suppresses tumor growth of human lung cancer through direct and indirect pathways in a preclinical model. *Oncotarget*. 2017 9 15;8(40):68571–82. [PubMed: 28978138]
137. Liu X, Zhu L, Lu X, Bian H, Wu X, Yang W, Qin Q. IL-33/ST2 pathway contributes to metastasis of human colorectal cancer. *Biochem Biophys Res Commun*. 2014 10 24;453(3):486–92. Epub 2014/10/05. eng. [PubMed: 25280997]
138. Maywald RL, Doerner SK, Pastorelli L, De Salvo C, Benton SM, Dawson EP, Lanza DG, Berger NA, Markowitz SD, Lenz HJ, Nadeau JH, Pizarro TT, Heaney JD. IL-33 activates tumor stroma to promote intestinal polyposis. *Proc Natl Acad Sci U S A*. 2015 5 12;112(19):e2487–96. Epub 2015/04/29. eng. [PubMed: 25918379]
139. Hu H, Sun J, Wang C, Bu X, Liu X, Mao Y, Wang H. IL-33 facilitates endocrine resistance of breast cancer by inducing cancer stem cell properties. *Biochem Biophys Res Commun*. 2017 4 8;485(3):643–50. Epub 2017/02/22. eng. [PubMed: 28216163]
140. Lu B, Yang M, Wang Q. Interleukin-33 in tmnorigenesis, tumor immune evasion, and cancer immunotherapy. *J Mol Med (Berlin)*. 2016 5;94(5):535–543. Epub 2016/02/29. eng. [PubMed: 26922618]
141. Landis MD, Seachrist DD, Abdul-Karim FW, Keri RA. Sustained trophism of the mammary gland is sufficient to accelerate and synchronize development of ErbB2/Neuinduced tumors. *Oncogene*. 2006 6 1 ;25(23):3325–34. Epub 2006/01/26. eng. [PubMed: 16434967]
142. Saranchova I, Han J, Zaman R, Arora H, Huang H, Fenninger F, Choi KB, Munro L, Pfeifer CG, Welch I, Takei F, Jefferies WA. Type 2 innate lymphocytes actuate immunity against tumours and limit cancer metastasis. *Sci Rep*. 2018 2 13;8(1):2924 Epub 2018/02/15. eng. [PubMed: 29440650]

TABLE 1:

Comparison of IL-33 in human and mouse cancers

Cancer type		Protumor		Antitumor	
		Endogenous	Exogenous	Endogenous	Exogenous
Pancreatic	Human	No data	IL-33-driven inflammation may contribute to inflammation-induced carcinogenesis ¹³³	No data	Human cell line PaCa-2 ST2+ cells undergo apoptosis in response to IL-33 ⁹³
	Human	High IL-33 in CAFs and cancer cells, associated with poor prognosis ¹³⁴	IL-33 treatment enhances migration invasion <i>ex vivo</i> ¹³⁴	High IL-33 decreases metastasis, increases survival ¹⁰⁸	No data
Head and neck	Mouse	No data	No data	No data	IL-33 suppresses fibroblast proliferation ¹²⁰
	Human	ST2 upregulated on CD34+ hematopoietic stem cells in CML ¹¹³	IL-33 increases colony forming of human CD34+ cells that engraft better after IL-33 treatment ¹¹⁵	GWAS associates low IL-33 with decreased survival in OS, CLL, and MM patients have reduced serum IL-33 ^{98,100,101}	No data
Leukemia	Mouse	No data	No data	No data	rIL-33 treatment expands IFN- γ CD8+ antileukemia T cells and prevents T cell exhaustion ⁶⁸
	Human	Plasma IL-33 associated with increased progression and metastasis ¹³⁵	No data	High IL-33 from tissue correlates with better prognosis. Low plasma levels correlate with better prognosis ¹⁰²	No data
Lung	Mouse	No data	Blockade of rIL-33 treatment inhibits NSCLC growth in human xenografts through inhibition of M2 macrophage polarization ¹³⁶	No data	No data
	Human	IL-33 expression correlates with more advanced stage of CRC ¹³⁷	No data	Tumor cells downregulate ST2; ST2 gene signatures decreased in late-stage CRC ¹⁰⁴	Novel IL-33 isoform promotes CRC growth compared to native IL-33 ¹⁰⁶
Colon	Mouse	IL-33 ^{-/-} mice show reduced polyp formation ¹³⁸	No data	No data	No data
	Human	IL-33 promotes metastasis and induces cancer cell stemness ¹³⁹	No data	IL-33 downregulated in ErbB2/Neu-induced tumors ^{140,141}	No data
Breast	Mouse	ST2 ^{-/-} mice have increased antitumor response ⁸⁷	IL-33 promotes metastasis and recruits tumor-infiltrating myeloid-derived suppressor cells ⁸⁹	No data	No data
	Human	No data	No data	Loss of IL-33 promotes immune evasion through loss of MHC I expression and promotes metastasis ⁸⁴	No data
Prostate	Human	No data	No data	No data	IL-33-treated ILCs promote CD8+ T cell killing of metastatic prostate cell line ¹⁴²

Cancer type	Protumor		Antitumor	
	Endogenous	Exogenous	Endogenous	Exogenous
Cervical	No data	No data	Severe neoplasias exhibited lowest amounts of IL-33 correlating with low IFN- γ production ¹⁰⁷	No data
Melanoma	No data	No data	No data	rIL-33 inhibits melanoma growth through type 1 immunity ^{53,59-61,68} and NK immunity ⁸⁷