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Chapter 2

The NKG2A–HLA-E axis as a novel checkpoint in the tumor microenvironment

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ABSTRACT

The success of checkpoint blockade therapy revolutionized cancer treatment. However, we need to increase the fraction of responding patients and overcome acquired resistance to these therapies. Recently, the inhibitory receptor NKG2A received attention as a new kid on the block of immune checkpoints. This receptor is selectively expressed on cytotoxic lymphocytes, including natural killer cells and CD8 T cells, and NKG2A$^+$ T cells are preferentially residing in tissues, like the tumor microenvironment. Its ligand, HLA-E, is a conserved non-classical HLA class I molecule that binds a limited peptide repertoire and its expression is commonly detected in human cancer. NKG2A blockade as a standalone therapy appears poorly effective in mouse tumor models, however in the presence of activated T cells, for example induced by PD-1/PD-L1 blockade or cancer vaccines, displays strongly enhanced efficacy. Clinical trials demonstrated safety of the humanized NKG2A-blocking antibody monalizumab and first results of phase II trials demonstrate encouraging durable response rates. Further development of this axis is clearly warranted.

Keywords: cancer immunotherapy, NKG2A, HLA-E, checkpoint blockade, cancer vaccinations, combination therapy, acquired resistance

INTRODUCTION

The unprecedented clinical impact of immune checkpoint blockade therapy induced a new era of cancer treatment[1]. After introduction in clinical practice of antibodies to CTLA-4 and the PD-1/PD-L1 axis, additional checkpoints like TIM-3, TIGIT and VISTA and stimulation of activating receptors like CD27, CD40 and 4-1BB are evaluated for their anti-tumor inducing immunity. Recently, the inhibitory immune inhibitory receptor NKG2A draw the attention as the new kid on the block of checkpoints[2-5]. NKG2A forms heterodimers with the CD94 chain[6] and recognizes the non-classical HLA class I molecule HLA-E. These recent studies demonstrated high expression of NKG2A on natural killer (NK) cells and cytotoxic CD8 T cells in the tumor microenvironment as a result of PD-1 blockade therapy as well as after immune activation by cancer vaccines[7,8]. First clinical trials in gynecological cancers and head-and-neck carcinoma with the NKG2A-blocking antibody Monalizumab shows safety and promising clinical responses in refractory disease, including regressions of target lesions[8,9]. Interestingly, protein levels of HLA-E, which is the sole ligand of the CD94/NKG2A receptor[10], are frequently upregulated in many cancers, suggesting that this axis functions as an acquired resistance mechanism after immune activation in the tumor microenvironment. Here, we provide an overarching view on the current understanding of NKG2A and HLA-E in cancer and touch upon gaps in our knowledge.

THE SIMILARITIES AND DIFFERENCES WITHIN THE NKG2A FAMILY

Activation of T cells is the result of T cell receptor (TCR) ligation with antigen-HLA complexes in combination with co-stimulatory signals, like CD28 and CD27, and cytokines. To avoid unrestricted immune responses that might result in pathology towards host tissues, T cells are also equipped with multiple inhibitory receptors, which are expressed in a programmed way[11]. As revealed, CTLA-4 and PD-1 inhibitory receptors are also considered as markers of recent T cell activation. The spaciotemporal dynamics are different for each inhibitory receptor, resulting in distinct profiles of immune pathology revealed in knockout mice. The CTLA-4 knockout mouse displays spontaneous and overt lymphoproliferation, whereas PD-1 and NKG2A knockouts show much milder phenotypes[12-15]. The cytoplasmic tail of the NKG2A receptor contains two immunoreceptor tyrosine-based inhibition motifs (ITIM) capable of recruiting both SHP-1[16] and SHP-2 phosphatases, but not the polyinositol phosphatase SHIP[17-19] (Figure 1). Both ITIMs are required to mediate the maximal inhibitory signal, but the membrane-distal ITIM is of primary importance rather than the membrane-proximal ITIM[20]. The partner CD94 has only seven cytoplasmic amino acids, thus lacks ITIMs and has no role in downstream signaling. Interestingly, CD94 can also form a heterodimer with a very close family member of NKG2A (encoded by the Klrc1 gene), which is named NKG2C (encoded by the Klrc2 gene). NKG2C is an activating receptor transducing an stimulating signal via association with DAP12, bearing an immunoreceptor tyrosine-based activation motifs (ITAM)[21]. The NKG2C protein carries some amino acid differences compared to NKG2A, resulting in a 6-fold lower affinity for the shared ligand HLA-E[22,23]. In NK cells, the two family members are usually not expressed together in the same cell and a switch to NKG2C marks

more mature NK cells, also referred to as 'adaptive' NK cells[24,25]. Another activating family member, NKG2D (encoded by the Klrk1 gene) is more distantly related and does not partner with CD94, but rather forms homodimers, and is engaged by stress induced self-proteins, e.g. MICA and ULBP members[26].

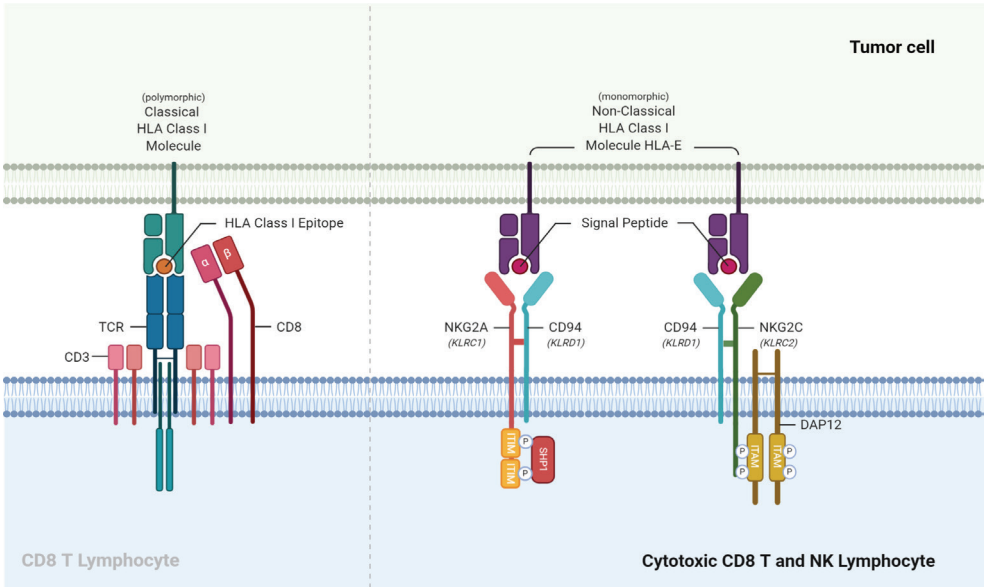

Figure 1|NKG2 family and their ligands.
The polymorphic classical HLA class I molecules present antigenic peptide-epitopes to the T cell receptor (TCR) in complex with CD3 and coreceptor CD8 at the surface of CD8 T cells. In contrast, HLA-E is a mono-morphic non-classical HLA class I molecule that presents a limited set of conserved signal peptides. These peptides are derived from leader sequences of classical HLA class I molecules. NKG2A and NKG2C form heterodimer receptors with CD94 and both target the same p/HLA-E complex, but ligation induces an inhibitory signal for NKG2A and an activation signal for NKG2C. The NKG2 locus of mouse and man also encode the activating family member NKG2E (not shown here), but its function remains elusive. The more distantly related activating homodimer NKG2D receptor binds MICA/B and ULBP1-6, 'empty' molecules that fold like HLA class I heavy chains but do not contain β2-microglobulin (not shown here). The NKG2 family is expressed by both cytotoxic CD8 T cells and NK lymphocytes. Figure is created with BioRender.com.

EXPRESSION AND FUNCTION OF NKG2A IN CYTOTOXIC LYMPHOCYTES

Approximately half of peripheral NK cells display the NKG2A receptor and these cells are mostly present in the CD56^{high} fraction, which contain the more immature cells. Intratumoral NK cells have somewhat higher frequencies of NKG2A[7]. Interestingly, NKG2A expression on CD8 T cells seems to be highly regulated, as peripheral cells hardly express the receptor, but a substantial fraction of intratumoral T cells, especially those in immune reactive milieu, display NKG2A[7,27]. These CD8 T cells often display a late effector memory phenotype and lack expression of typical central memory markers CCR7, CD27 and CD28 or late effector markers KLRG1. Interestingly, we and others found NKG2A expression on CD8 T cells

harboring a tissue-resident signature, marked by specific integrins like CD103[7,28-30]. Although induction of NKG2A is initiated by TCR triggering, the presence of tissue cytokines like IL-15 and TGF-β might enhance its expression[31-33]. TGF-β is often overtly present in the tumor microenvironment[34]. However, the functional relationship between the tissue-residence program, NKG2A and TGF-β is unclear at the moment.

Asides from its expression regulation, the NKG2A downstream effects are also not completely unraveled. The inhibitory signals induced by NKG2A receptor engagement results in decreased capacity of NK cells and CD8 T cells to lyse target cells[33,35-39]. NKG2A triggering inhibits cytotoxic effector functions on NK cells by disrupting the actin network at the immunological synapse of activating receptor NKG2D[40]. For CD8 T cells such mode of action is still to be elucidated, though NKG2D or the immunological synapse of the TCR could be lucid candidates[41]. In this context, the recent findings on the operational mechanism of PD-1, which was suggested to dephosphorylate T cell costimulatory receptor CD28, illustrate our limited understanding at molecular level of these important checkpoints[42,43]. CTLA-4 executes its inhibitory effects by competing with CD28 for the same ligands on APCs, thereby limiting T cell costimulation[44]. Such molecular details are lacking for NKG2A, let alone our insight on direct downstream target genes affected by NKG2A triggering.

BOX 1: THE BASICS OF HLA-E

In contrast to classical MHC class I molecules, HLA-E is virtually non-polymorphic with only 2 functional alleles present in the human population: the HLA-E*01:01 and the HLA-E*01:03 variants. These two alleles only differ in a single amino acid at position 107, being arginine (01:01) or glycine (01:03). Position 107 is located just outside the peptide-binding groove on the loop between the β-strands outside of the α2 domain of the heavy chain[53]. The crystal structure of the peptide binding groove, single alanine substitutions and peptide elution, have demonstrated that HLA-E has an optimal structure to bind peptides with two primary anchor residues at positions 2 and 9, and secondary anchor residues at position 7 and possibly 3[54-59]. Surprisingly, a very limited repertoire of peptides was found under homeostatic conditions and the most prominent are the signal peptides of classical MHC class I molecules. The majority of HLA class I alleles share a consensus sequence in their signal peptides with the amino acid motif of VMAPRTLLL (Table 1). Position 7 and 8 vary between leucine and valine, without affecting binding affinity. Some HLA alleles however contain a substitution at residues p2, 3 or 6, which impair binding[23]. In addition, the immunotolerance molecule HLA-G, which is expressed in immune-privileged sites and frequently in cancers, encodes a unique leader peptide that binds with high affinity to HLA-E[60,61]. Interestingly, the leader peptide motif is extremely conserved among mammalian species, implying an important role in immune defense[62] and is even copied by the human cytomegalovirus gpUL40 protein in order to sustain HLA-E surface display on infected cells while shutting down classical HLA class I antigen presentation of host cells[63-65]. Importantly, the CD94/NKG2A receptor exhibits clear preference for peptide-containing HLA-E, especially for this consensus leader peptide[56]. Under stress, HLA-E can present a peptide of multidrug resistance-associated protein 7 (ALALVRMLI)[66] and under high cell densities the leader peptide QMRPVSRVL of heat shock protein 60 (hSP60) stabilizes HLA-E[67]. In cancers, classical HLA class I molecules are frequently lost to prevent T cell-mediated recognition[68,69], but surprisingly, expression of the HLA-E molecule is often enhanced[70-78]. Finally, HLA-E surface display is determined by expression levels of classical alleles through the availability of their leader peptide and the binding affinity of this peptide to HLA-E, as peptides with a threonine at position 2 poorly bind (see also Table 1)[79,80].

HLA-E AS A MOLECULE OF IMMUNE TOLERANCE

The ligand of the heterodimeric receptor CD94/NKG2A is the human histocompatibility leucocyte antigen E (HLA-E) and its mouse orthologue Qa-1^b. These non-classical MHC class Ib molecules are surprisingly conserved in the population and present signal peptides of classical MHC class I molecules (Box 1)[45]. Interestingly, whereas most tissues express low basal cell surface levels, HLA-E is highly expressed in immune privileged sites of the body, including trophoblast cells of the placenta and ductal epithelial cells in the testis and epididymis, suggesting that HLA-E has a role to counter potential attack by CD8 T cells and NK cells against the partly HLA-mismatched fetus and haploid reproductive cells (Figure 2)[46]. Interestingly, CD94/NKG2A is rather peptide-specific and preferentially interacts with HLA-E when canonical peptides are bound, but not when these are absent[22,47]. These canonical peptides are derived from the leaders of HLA class I molecules, including the immune tolerant molecule HLA-G[48]. Additionally, the expression of HLA-E or its mouse orthologue Qa-1^b provides protection of tissue integrity during viral clearance and limits excessive activation and apoptosis of CD8 T cells[15,49-52].

Table 1. Sequences and origins of HLA-E binding peptides

	HLA derived leader peptides		
	Leader peptide sequence	**HLA-locus**	**allele**
substitution at residues P7 and P8	MAVMAPRTLLLLLSGALALTQTWA[1]	HLA-A	A*01, A*03, A*11, A*29, A*30, A*31, A*32, A*33, A*36, A*74
		HLA-C	C*02, C*15
	VMAPRTL**V**L	HLA-A	A*02, A*23, A*24, A*25, A*26, A*34, A*43, A*66, A*68, A*69
	VMAPRT**V**LL	HLA-B	B*07, B*08, B*14, B*38, B*39, B*42, B*48, B*67, B*73, B*81
	VMAPRTLI**L**	HLA-C	C*01, C*03, C*04, C*05, C*06, C*08, C*12, C*14, C*16
	VMAPRT**VF**L	HLA-G	G*01
substitution at residues P2, P3, P6	VM**PP**RTLLL	HLA-A	A*80
	VT**A**PRT**V**LL	HLA-B	B*15, B*35, B*40, B*41, B*45, B*46, B*49, B*50, B*52, B*53, B*57, B*58
	VT**A**PRTLLL	HLA-B	B*13, B*18, B*27, B*37, B*44, B*47, B*54, B*55, B*56, B*59, B*82, B*83
	VMAPR**A**LLL	HLA-C	C*07, C*18
	VMAP**QA**LLL	HLA-C	C*17
	. M**VDG**TLLL[2]	HLA-E	E*01
	. MAPR**S**LLL[2]	HLA-F	F*01

1. consensus HLA-E-binding peptide sequence
2. leader peptides from HLA-E and HLA-F are shorter and are therefore unable to serve as HLA-E binders

IPD-IMGT/HLA Database, http://www.ebi.ac.uk/ipd/imgt/hla/

HLA-E EXPRESSION IN CANCER

Whereas classical HLA alleles are frequently lost in human cancer to prevent T cell recognition[68,69], we and others reported high levels of HLA-E in several cancer types, including gynecological cancers (up to 90% of tumor samples)[70-72] and up to 50% in breast cancer, non-small cell lung carcinoma (NSCLC), liver, pancreas, kidney, melanoma, prostate, head and neck, stomach, rectal, and colorectal cancer[73-78]. Figure 2 displays some examples of tissue sections of human cancers and their healthy counterparts. The surface expression of HLA-E is correlated with functional antigen processing components and infiltration of CD8 T cells, however, expression can also be observed in tumors with downregulated classical HLA class I expression. In a cohort of patients diagnosed with high grade serous ovarian carcinoma, HLA-E expression correlated with a significantly worse survival[81]. A poor relapse-free survival rate was also observed in breast cancer, but only when classical class I expression was lost[73]. CD8 T cell infiltration and retained expression of classical HLA class I strongly associate with a better prognosis for patients with non-small cell lung (NSCLC), cervical and ovarian carcinomas. However, this predictive value is abrogated when tumor display high HLA-E levels, suggesting that HLA-E mediates resistance against CD8 T cell attack[70,71,74]. In primary colorectal cancers, patients with high HLA-E expression showed a significantly decreased disease-free survival for Duke's C patients[76]. In a another retrospective cohort of 234 colorectal patients the expression of HLA-E, β2m, CD94, CD8, and NKp46 was examined by immunohistochemistry on tissue microarray. HLA-E/β2m was overexpressed in microsatellite instable tumors and to a lesser extend in microsatellite stable ones (45% vs. 19%, respectively) and corresponded with worse survival[78]. Unfortunately, NKG2A expression cannot be quantified in paraffin embedded tissues due to lack of specific antibody. An independent study of colon cancers, the researchers concluded that the total absence of HLA-class I, including HLA-E and HLA-G, related to a better overall and disease-free survival[77]. This was attributed to NK cell-mediated killing in the portal vein of MHC class I negative cancer cells, which give rise to liver metastases. It should be stated that there are also several studies of patient cohorts in which the researchers did not observe a relevant correlation with HLA-E expression[75,82]. However, as previously demonstrated, the role of HLA-E expression seems to be dependent on immune contexture, like lymphocyte infiltration and presence of classical HLA class I molecules. Stratification on these factors might also signify the negative effect of HLA-E in these cohorts as observed in others. Finally, it should be noted that HLA-E directed antibodies fail to distinguish between peptide-filled molecules and empty conformers (see Box 2), complicating the interpretation of these tissue slide results, as especially HLA-E filled with leader peptides constitute ligands for the CD94/NKG2A receptor[56].

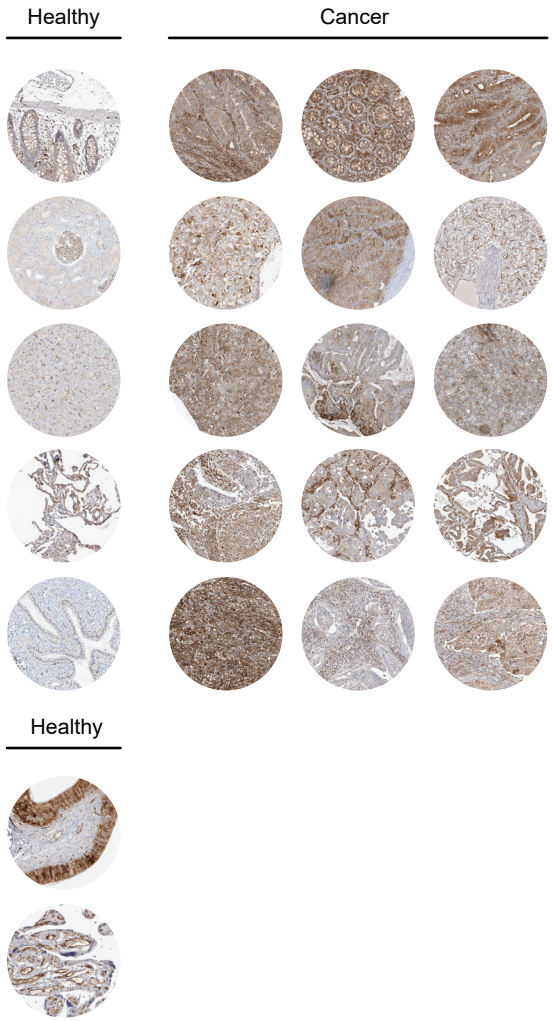

Figure.
HLA-E expression in healthy and cancer tissue. Immunohistochemical staining of healthy and cancer tissues for HLA-E, as deposited in the Human Protein Atlas (www.proteinatlas.org). Tissue slides were stained either with MEM-E/02 antibody or rabbit polyclonal HPA031454 (85, 86). Of note: HLA-E antibodies also binds free heavy chains.

INDUCTION OF HLA-E

The fact that HLA-E is generally upregulated in cancers compared to healthy tissue touches upon responsible gene transcription mechanisms. Interferon (IFN)-γ is proposed as an important cytokine and is locally produced by tumor-infiltrating T cells and NK cells. At the transcription level, this is substantiated through binding of a STAT1-containing complex to distinct IFN-γ-responsive region (IRR) present upstream of the HLA-E gene[92]. In addition, the upstream UIRR region mediates a three- to eight-fold increase in HLA-E transcription in response to IFN-γ upon binding of GATA-1[93]. *In vitro* experiments indeed showed that IFN-γ significantly increases the surface expression of HLA-E and the shedding of soluble HLA-E by melanoma cells, in a metalloproteinase-dependent fashion[94]. This also corresponds to studies showing enhanced HLA-E expression on tumors upon increased cytotoxic lymphocyte infiltration[71] and enhanced Qa-1^b expression in mice in response to immune therapy leading to increased T cell infiltration[7]. Nevertheless, there are also tumors and tumor cell lines that constitutively upregulate HLA-E molecules, soluble and at the cell surface, irrespective of IFN-γ treatment[94]. Importantly, HLA-E surface expression is post-translationally regulated by availability of the conserved leader peptides, the peptide transporter TAP and proteolytic enzymes. Moreover, HLA-E can present alternative peptides aside from the HLA class I leaders and can be recognized by αβ T cell receptors[95,96]. Clearly, novel research tools are required to distinguish HLA-E complexed with the monomorphic leader peptides from those complexed with alternative peptides, which might actually be present in tumors[58].

BOX 2: ANTIBODIES AGAINST HLA-E

Generation of specific antibodies to HLA-E was challenging due to its close homology with classical HLA class I molecules. Three antibodies stand out as most specific: 1. Clone 3D12 is frequently used in flow cytometry and was generated against soluble HLA-E with the HLA-A2 leader peptide[83] and is capable to stain the native conformation of the molecule and also binds to β2m-free HLA-E conformations[84]. 2. Clone MEM-E/02 recognizes a linear epitope on HLA-E and is mostly used in formalin-fixed paraffin embedded tissue slide staining[84,85]. 3. Clone TFL-033 binds HLA-E in a highly specific manner and is reactive to the α1 and α2 helices of HLA-E that are the same sequences recognized by the CD94/NKG2A inhibitory receptors[86]. A debate on the exact specificity and potential cross-reactivity with classical HLA molecules of these antibodies is ongoing[83-85,87,88], and stirred by recent findings that HLA-E and its mouse ortholog Qa-1^b poorly associate with β2m and accumulates as an β2m-free and peptide-empty form at the cell surface[84,89-91]. As currently available antibodies fail to distinguish between peptide-loaded or open HLA-E conformers and the biological relevance of open conformations remains to be determined, novel antibodies to peptide-containing forms of HLA-E and preferably even peptide-specific antibodies, are urgently required to promote the development of this field.

TARGETING THE NKG2A - HLA-E AXIS

Similarities between the PD1 – PD-L1 axis and the NKG2A – HLA-E axis are obvious, as they both involve lymphocytes and represent inhibitory immune receptors and their ligands are expressed on cancer cells and inducible by the pro-inflammatory cytokine IFN-γ. Of note, HLA-E transcripts in the TCGA database exceed those of PD-L1, suggesting a high and general overexpression (figure 3). These two pairs reflect feedback signals to dampen overt T-cell mediated tissue damage at affected lesion sites, which might result in resistance and immune escape of cancer[97,98]. In contrast to PD-1, NKG2A is selectively expressed on lymphocytes with cytolytic function, including NK cells, NKT cells and a subset of CD8 T cells. Thereby the NKG2A – HLA-E axis is suggested to predominantly act at the terminal tumor-attack stage and not to be involved in priming or regulation of immune responses. Interruption of this axis by blocking antibodies can be envisaged at both sides. However, NKG2A is the preferred target instead of HLA-E, since blockade of HLA-E would also prevent interaction with the activating NKG2C receptor. A NKG2C-expressing subset of NK cells, also called "adaptive NK cells", displays an altered receptor profile and is associated with chronic viruses, like CMV[99,100]. Its role in immunity to CMV remains elusive, but anti-HLA-E antibodies would also block activation of such NK cells through NKG2C. Disappointingly, mouse cancer models demonstrated limited success of NKG2A blockade therapy when provided as a standalone blocking antibody, however it greatly improved anti-tumor efficacy of other forms of immunotherapy, like PD-L1 blockade or cancer vaccines[7,8,101]. Cancer vaccines induced strong tumor-directed T cell responses and infiltration of CD8 T cells in the tumors, leading to local IFN-γ release and increase of the mouse HLA-E ortholog[7]. In addition, frequencies of NKG2A$^+$ CD8 T cells were increased in TIL. The association of immune reactivity and NKG2A$^+$ T cell frequency was corroborated in human TIL of oropharyngeal carcinomas[7]. Together, these observations designated a role for NKG2A as an acquired resistance mechanism[102]. This concept is further substantiated by pre-clinical *in vitro* data demonstrating a requirement

of immune activation signals to reveal beneficial effects of NKG2A blockade[8,27,38]. Design of clinical trials should therefore be based on combination therapy leading to inflammatory responses in cancer patients. Induced expression of HLA-E or enhanced frequencies of NKG2A$^+$ immune cells in the tumor might serve as predictive biomarkers.

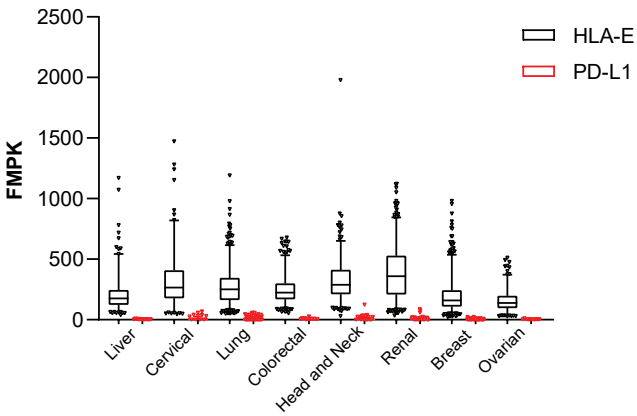

Figure 3.
HLA-E and PD-L1 transcription levels in various cancer types. RNA-seq data for HLA-E and PD-L1 is reported as median FPKM (number Fragments Per Kilobase of exon per Million reads), generated by The Cancer Genome Atlas (TCGA). Normal distribution across the dataset is visualized with box plots, shown as median and 25th and 75th percentiles. Points are displayed as outliers if they are above the 97.5 or below the 2.5 percentile.

NKG2A IN CLINICAL TRIALS

Monalizumab (IPH2201, Innate Pharma/AstraZeneca) is a humanized IgG4 antibody blocking the interaction of human NKG2A with HLA-E and showed a therapeutic effect in immunodeficient mice harboring human leukemia[103]. The NKG2A-blocking antibody monalizumab was furthermore administered in a dose ranging phase II clinical trial in patients with gynecologic malignancies[9]. Administration of 10 mg/kg i.v. every 2 weeks was well tolerated and even short-term disease stabilizations were observed[9]. In another recent clinical trial, monalizumab was combined with the EGFR-blocking antibody cetuximab in previously treated squamous cell carcinoma of the head and neck. This combination showed a promising 31% objective response rate[8]. Cetuximab is able to activate NK cells via Fcγ-receptors, leading to antibody-mediated cellular cytotoxicity. Addition of monalizumab enhanced NK cell effector functions[8]. Currently, several trials in which monalizumab is tested are enrolling patients. Different combinations with anti-EGFR, anti-PD-L1, tyrosine kinase inhibitors and chemotherapy are tested in several cancer indications, including patients with resectable non-small cell lung carcinoma (NCT03794544), PD-1 therapy-resistant NSCLC patients (NCT03833440), and advanced non-resectable stage III NSCLC patients (NCT03822351) as well as patients with advanced squamous cell carcinoma of the head and neck (NCT02643550), refractory chronic lymphocytic leukemia (NCT02557516) or other hematologic malignancies after stem cell transplantation (NCT02921685), and also a basket trial with advanced solid malignancies (NCT02671435). This latter trial reported a manageable safety profile and durable partial responses in patients with microsatellite-stable colorectal carcinoma treated with combinations of FOLFOX chemotherapy and blocking antibodies to VEGF, PD-L1 and NKG2A[104]. Based on these promising results, a larger

phase II trial will start in the near future for high-risk MSS colorectal carcinoma patients who received radical surgery, testing different adjuvant therapies based on standard-of-care chemotherapy FOLFOX in combination with durvalumab (anti-PD-L1) and monalizumab (anti-NKG2A) (Columbia 2) (NCT04145193).

CONCLUSIONS

The current challenge of checkpoint blockade therapy is to increase the fraction of responding patients and, secondly, to overcome acquired resistance. NKG2A blockade displays unique characteristics compared to other immune checkpoints, as this target is selectively expressed within tumor lesions on cytotoxic lymphocytes and is not involved in priming or regulation of immunity against cancer. NKG2A^{+} T cells are mostly residing in the tumor microenvironment where they are effector cells attacking the transformed cells. Moreover, we and others showed increased frequencies of these cytotoxic lymphocytes upon administration of therapeutic cancer vaccines. These observations plea for combination therapies of the NKG2A-blocking antibody monalizumab with such vaccines. In the meanwhile, fundamental investigation on NKG2A and its ligand HLA-E should provide mode-of-action of this novel immune checkpoint.

DECLARATIONS

Conflicts of interest

S.H.v.d.B. and T.v.H. hold a patent on NKG2A and received a research grant from Innate Pharma.

Author Contributions

Conceptualization, S.H.v.d.B. and T.v.H.; Methodology, N.v.M., L.B., M.J.K., M.S., K.A.M., S.J.S., M.J.P.W., S.J.P., and T.v.H.; Formal Analysis, P.C., S.J.S., N.v.M., and L.B.; Investigation, N.v.M., L.B., M.J.K., K.A.M., M.S., S.J.S., V.J.v.H., I.E., and S.J.P.; Resources, P.A. and N.W.; Writing – Original Draft, N.v.M., L.B., and T.v.H.; Writing – Review & Editing, S.J.P., S.H.v.d.B., and T.v.H.; Visualization, L.B. and S.J.P.; Supervision, Y.J.K., S.H.v.d.B., and T.v.H.; Funding Acquisition, S.J.P., S.H.v.d.B., and T.v.H.

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