ARBS Annual Review of Biomedical Sciences

pdf freely available at http://arbs.biblioteca.unesp.br

2010;13:23-29

NK Cell-Mediated Targeting of Human Ovarian Cancer and Possibilities for Immunotherapy

Rodrigo F Silva[1,2], Paulo CM Alves[1,2], Fernando Guimarães[1*]

[1]Hospital da Mulher Professor Doutor José Aristodemo Pinotti, Centro de Atenção Integral à Saúde da Mulher, Campinas University (UNICAMP), Campinas, SP, BRAZIL
[2]Faculdade de Ciências Médicas, pós-graduação do Departamento de Tocoginecologia, Campinas University (UNICAMP), Campinas, SP, BRAZIL

Received: 13 April 2011; accepted 28 September 2011
Online on 25 January 2012

Abstract

Silva RF, Alves PCM, Guimarães F. NK Cell-Mediated Targeting of Human Ovarian Cancer and Possibilities for Immunotherapy. ARBS Annu Rev Biomed Sci 2011;13:23-29. A better understanding of the NK cell receptor-ligand interaction opened possibilities for new therapeutic strategies. The well known mismatch KIR/ HLA interaction is of great importance for NK cell stimulation. Lately, the interaction of the activating receptor DNAM-1 with its ligand PVR, has been highlighted in many ovarian carcinoma studies. Autologous and allogeneic NK cells, along with monoclonal antibodies, have been tested in current immunotherapies. This review presents information on NK cell receptor-ligand interactions and its possibilities for NK-based immunotherapy against ovarian cancer.

Keywords: Natural killer cells, immunotherapy, ovarian neoplasm, NK cell receptors.

Table of Contents

1. Introduction
2. NK Cells
3. NK Receptors
4. NK Cell in Response Against Ovarian Cancer
5. Conclusion
6. References

1. Introduction

The poor prognosis of ovarian carcinoma has driven studies to develop more efficient therapeutic strategies. The possibility to use natural killer (NK) cells for the treatment of human cancer has increased recently (Ljunggren & Malmberg, 2007; Sutlu & Alici, 2009). This is a consequence of an increased comprehension of the molecular processes and receptor-ligand interactions which influences NK cell

Correspondence
Fernando Guimarães. CAISM, UNICAMP, Rua Alexandre Fleming 101, Campinas, SP, 13083-970, Brazil. Phone: +55 19 35219462, FAX: +55 19 35219433. E-mail: fernando@caism.unicamp.br

Financial support: The "Fundação de Amparo à Pesquisa do Estado de São Paulo" sponsored the project (#2006/04600-2) and conceived a fellowship to RF Silva (2010/17202-7).

recognition and elimination of tumor cells. New cellular isolation and expansion methods ex vivo provide sufficient quantities of human NK cells for clinical trials (Barkholt *et al.*, 2009). Additionally, drugs, antibodies and genetic manipulation can be combined to the therapeutic strategy in order to exploit NK cells antitumor function (Malmberg *et al.*, 2008). The purpose of this review is to present information on NK cell specific molecular receptor-ligand interactions with ovarian cancer cells and its usefulness for the development of an NK-based immunotheraphy.

2. NK Cells

NK cells are lymphocytes initially indentified by its functionality, based on their capacity of eliminating a variety of tumor cells without previous stimulation (Kiessling *et al.*, 1975; Herbermen *et al.*, 1975). In humans, NK cells constitute 5-15% of lymphocytes present in the peripheral blood, identified by the absence of CD3 cell surface molecule and by the expression of CD56 (CD3-CD56+), an isoform of neural cell adhesion molecule (NCAM) also found on a minority of T cells (Robertson *et al.*, 1990; Arnon *et al.*, 2006). NK cells can be further grouped into two functional subtypes considering the expression of the CD56 marker. The majority (90%) of the NK cells that are found in the peripheral blood express low maker density, they are categorized as CD56dim and the minority of them (10%) express high marker density, they are categorized as CD56bright (Lanier *et al.*, 1986; Sutlu & Alici, 2009). CD56dim cells can be found in the bone marrow, peripheral blood and spleen. They express high levels of CD16 (FcγRIIIA) receptors, have great cytolytic potential and produce less variety of cytokines. CD56bright are mainly found in peripheral lymphoid tissues, express low levels of CD16, have low cytolytic capacity and produce the largest variety of immunoregulatory cytokines. It has been hypothesized that CD56bright cells are less mature than CD56dim, since there is an increase in the expression of specific receptors of the subtype "dim" (like activation receptors and CD16+) when the "bright" subtype is stimulated by IL-2 (Lanier *et al.*, 1986; Di Santo, 2006). Another evidence which supports this hypothesis is the existence of large quantities of CD56bright cells associated with lymphocytes CD34+CD45RA+, NK precursor cells in secondary lymphoid tissues. In these tissues there are still plenty of antigen presenting cells expressing interleukin 15 (IL-15) attached to its surface, this cytokine is known to be important in the maturation of NK cells. (Mattei *et al.*, 2001; Fehniger *et al.*, 2003; Freud *et al.*, 2005; Caligiuri, 2008). Therefore, the development of NK cells does not only occur in the bone marrow, but it also occurs in secondary lymphoid tissues.

NK cells are mentioned as lymphocytes of the innate immune response, mediating effector functions against malignant or virus infected cells. However, NK cells production of pro-inflammatory cytokines such as interferon-γ (INF-γ), tumor necrosis factor-α (TNF-α) and granulocyte macrophage colony-stimulation factor (GM-CSF) regulates the innate immune response and contributes to the development of the adaptive immune response (Wiltrout, 2000). The cytotoxic activity of NK cells occurs after contact with target cells, followed by the secretion of perforin and granzyme-B or induction of apoptosis by ligands FasL and TRAIL (TNF-related apoptosis-inducing ligand) (Wiltrout, 2000; Cooper *et al.*, 2001; Wu & Lanier, 2003; French & Yokoyama, 2003; Hayakawa *et al.*, 2004).

3. NK Receptors

NK cells cytolytic function is determined by the balance of inhibitory and activating signals resulted from the interaction between NK cells receptors and target cells ligands (Cerwenka *et al.*, 2001; Lanier, 2005). Several activating and inhibitory receptors have been described in NK cells, some of them have been characterized focusing on the recognition process of ovarian cancer cells. Among the major inhibitory receptors are: KIR (killer immunoglubuline-like receptors) that recognize ligand molecules of the human leukocyte antigen (HLA) class I from the groups A, B and C; CD94-NKG2A receptor that recognizes the HLA class I from the group E; and ILT2 (immunoglobulin-like transcript 2) receptor that recognizes a relatively conserved region in the class I molecule, providing broad class I specificity (Farag *et al.*, 2002; Moretta *et al.*, 2004; Lanier, 2005; Malmberg *et al.*, 2008). HLA class I is expressed by virtually all nucleated cells of a person, playing a key role in the recognition of body cells as "self" by the immune system. Once committed to HLA class I "self", the inhibitory receptors KIR signal with dominance over the signal of activating receptors, therefore hindering the cytolytic function of NK cells (Bryceson *et al.*, 2006; Caligiuri, 2008). Thus, NK cells spare autosomal cells that express normal levels of HLA class I molecules while eliminating abnormal cells, such as tumor cells or cells infected by viruses, which frequently lose their expression of HLA class I (Ljunggren *et al.*, 1990; Garcia-Lora *et al.*, 2003; Schanoski *et al.*, 2004).

Among the activating receptors stand out the natural cytotoxic receptors (NCR) NKp46 and NKp30

that are expressed constitutively in NK cells, and the NKp44 receptor that is expressed after NK cells activation by interleukin 2 (IL-2) (Arnon *et al.*, 2006; Fuchs *et al.*, 2005). These receptors also assist in the recognition of tumor cells, cells infected by viruses and maturation regulation of dendritic cells. However, little is known about the identity of the ligands for NCR (Cooper & Caligiuri, 2004; Arnon *et al.*, 2005). The density of NCR expression varies among individuals and correlates directly with the ability of NK cells to eliminate abnormal cells (Sivori *et al.*, 1999; Pende *et al.*, 2001). Unlike the NCR, the NKG2D is an activating receptor whose expression is not restricted to NK cells, it is also expressed by cytotoxic T lymphocytes (γ/δ T e α/β T-CD8). The NKG2D recognizes as ligands molecules homologous to HLA class I, represented by transmembrane proteins such as MIC/A, MIC/B, ULBP4 (UL16-biding protein) and proteins anchored to the cell surface by glicofosfatidilinositol (GPI) as ULBP 1, 2 and 3 (Bauer *et al.*, 1999; Biassoni *et al.*, 2001; Cosman *et al.*, 2001; Raulet, 2003; Coudert & Held, 2006). In humans, increased expression of MIC and ULBP is related to different forms of cellular stress, such as viral infection and malignant transformation (Bauer *et al.*, 1999; Cosman *et al.*, 2001; Onda *et al.*, 2001). In fact, the expression of MICA/B has been observed in most human epithelial tumors, including breast, ovarian, colon, kidney and lung carcinomas (Biassoni *et al.*, 2001; Diefenbach & Raulet, 2002), contributing to the possible susceptibility of these tumors to the cytotoxic activity of NK cells. DNAX accessory molecule-1 (DNAM-1) is another important NK cells activating receptor. The ligands indentified for DNAM-1 receptor are the Poliovirus receptor (CD155) and Nectin-2 (CD112), with CD155 displaying to have a predominant role in the induction of DNAM-1 responses. Furthermore, CD155 is commonly expressed on normal cells and overexpressed on various tumor types, including ovarian carcinoma (Lanier, 2005; Carlsten *et al.*, 2009). DNAM-1 contributes to tumor immune surveillance and plays a crucial role in NK cell-mediated recognition of several types of human tumors.

4. NK Cell in Response Against Ovarian Cancer

Since NK cells were first described, more than 30 years ago, new cancer therapies based on their capacity to lysis tumor cells have been developed. Although, several studies have demonstrated the ability of NK cells to target tumor cells in vitro and in vivo (Smyth *et al.*, 2002; Wu & Lanier 2003; Malmberg *et al.* 2008), only in the last decade have been obtained direct evidences on how receptor-ligand interactions drive targeting of tumor cells by NK cells in humans. This information has prompted new insights on therapeutic uses for NK cells.

NK cells from ovarian carcinoma patients were initially reported to display none or poor cytolitic activity against ovarian cell lines, fresh ovarian tumors, and even against the prototype NK cell target K562 (Lotzova *et al.*, 1986; Lotzova *et al.*, 1988; Roszkowski *et al.*, 1993; Malberg, 2004). Additionally, NK cell cytolitic activity against tumor cells was significantly lower among patients with ovarian carcinoma than in patients with benign masses (Lutgendorf *et al.*, 2005). However, antitumor function of NK cells from patients with ovarian cancer can be reestablished as demonstrated by in vitro stimulation of effector cells with recombinant IL-2 (Lotzova *et al.*, 1986; Lotzova *et al.*, 1988) or by enriching the preparation of effector cells with large granular lymphocyte, corresponding to the NK cells (Lotzova *et al.*, 1988). These observations support the idea of overcoming the immunosuppression often seen in patients with cancer, by using strategies for ex vivo expansion and stimulation of cytotoxic cells.

Similarly to other malignancies, ovarian cancer exploits an array of immunological ways to create a suppressive environment to prevent being eliminated by the immune response (Yigit *et al.*, 2010). Two immunosupressive mechanisms capable of affecting NK cell functions have already been detected in ovarian carcinoma patients, one involving recruitment of regulatory T CD4+CD25+ (Treg) lymphocytes to the tumor site, and other involving selective down-regulation of NK cell-activating receptor DNAM-1 (Curiel *et al.*, 2004; Yigit *et al.*, 2010; Carlsten *et al.*, 2009). Specific recruitment of Treg lymphocytes to the tumor site and ascites was correlated to a reduced survival of patients with ovarian cancer (Curiel *et al.*, 2004). Additionally, Treg lymphocytes have been reported to affect NK cell proliferation, cytotoxicity and IFN-γ production (Ghiringhelli *et al.*, 2005; Smyth *et al.*, 2006). Ovarian carcinoma cells expressing the DNAM-1 ligand CD155 led to down-regulation of DNAM-1 activating receptor, explaining the hyporesponsiveness found in tumor-associated NK cells compared to the autologous peripheral blood NK cells (Carlsten *et al.*, 2009).

Immunosuppression is not advantageous for immunotherapies, but by knowing the mechanisms of the suppression, new strategies can be developed in an attempt to overcome this therapeutic barrier. Recent studies have demonstrated the possibility of generating CD56+ NK and NKT-like lymphocytes by ex vivo expansion of PBMC (peripheral blood mononuclear cells) from patients with ovarian carcinoma (Alves *et*

al., 2011). Such effector cell preparations displayed antitumor function, showing the feasibility of overcoming the immune impairment often inferred to cancer patients. Additionally, the NK cells present in the ex vivo effector cells expansion were CD16+, indicating their activation status and their cytotoxic potential mediated by antibodies (Borghaei *et al.*, 2009).

Allogeneic human NK cells are also known to recognize and kill freshly isolated ovarian carcinoma cells. The degranulation of NK cells is dependent on signaling through DNAM-1 receptors with an additional contribution of NKG2D receptors by recognizing corresponding ligands expressed on the surface of ovarian carcinoma cells (Carlsten *et al.*, 2007). The relative high expression of CD155 in combination with reduced levels of HLA class 1 molecules on ovarian carcinoma cells, labels them as an ideal target for autologous NK cells as well (Carlsten et al 2009). Allogeneic NK cells from healthy donors can be a promising immunotherapy strategy since they don't exhibit impaired functions consequently derived from induced immune suppression. Besides, allogeneic NK cells can be specifically activated by the mismatch interaction of KIR/HLA and by the positive interaction between the activating receptor DNAM-1 and its ligand PVR (Carlsten *et al.*, 2007). Graft-versus-host disease (GVHD) is of major concern in the usage of allogeneic NK cells for immunotherapy. However, many recent clinical trials have demonstrated no GVHD in patients with malignancies treated with allogeneic NK cells (Passweg *et al.*, 2004; Miller *et al.*, 2005; Barkholt *et al.*, 2009).

Monoclonal antibodies (mAbs) usage in cancer therapy is based on targeting tumor cells that express tumor associated antigens. Many mAbs targeting antigens associated to cancer cells have been developed in the past few years and it is lately, one of the most important drugs approved for the treatment of cancer, including ovarian carcinoma (McCall *et al.*, 2001; Chan *et al.*, 2006; Seimetz *et al.*, 2010; Esser *et al.*, 2011). The efficacy of mAbs on hematological and some solid malignancies has been shown, but in the case of ovarian carcinoma, it has not been yet validated (Mabuchi *et al.*, 2010). Several mAbs have been investigated for a potential treatment against ovarian cancer, such as bevacizumab; a vascular endothelial growth factor-targeted mAb therapy, trastuzumab and cetuximab; an epidermal growth factor-targeted mAb therapy, oregovomab; a CA-125-targeted mAb therapy, the mAb human milk fat globule 1 (HMFG1); a MUC1-targeted therapy and catumaxomab; a trifunctional antibody with two different antigen-binding specificities, epithelial cell adhesion molecule (EpCAM) and CD3 antigen (Seimetz *et al.*, 2010; Mabuchi *et al.*, 2010). Their efficacy and side effects have been demonstrated in clinical trials but still, further research is needed to create more efficient, precise and less toxic immunotherapy strategies for ovarian carcinoma (Mabuchi *et al.*, 2010). Combined therapies using mAbs with NK cells are believed to benefit from antibody-dependent cell-mediated cytotoxicity (ADCC), since NK cells express CD16 (FcγRIIIA) receptor that recognizes the Fc portion of mAbs (McCall *et al.*, 2001; Borghaei *et al.*, 2009).

5. Conclusion

Due to an increased knowledge of the molecular processes and receptor-ligand interactions of NK cells, studies have been conducted using these lymphocytes for the development of cancer treatments. Currently, clinical trials with sufficient amount of NK cells can be conducted, due to improvements on the isolation and expansion methods. In overall, autologous and allogeneic NK cells, the KIR/HLA mismatch, DNAM-1/PVR interaction and monoclonal antibodies, currently are the most promising factors for NK-based immunotherapy against ovarian cancer.

6. References

Alves PC, Andrade LA, Petta CA, Lorand-Metze I, Derchain SF, Guimarães F. Ex vivo expansion of CD56+ NK and NKT-like lymphocytes from PBMC of patients with ovarian neoplasia. Scand J Imm 2011; 74:244-52.

Arnon TI, Achdout H, Levi O, Markel G, Saleh N, Katz G, Gazit R, Gonen-Gross T, Hanna J, Nahari E, Porgador A, Honigman A, Plachter B, Mevorach D, Wolf DG, Mandelboim O. Inhibition of the NKp30 activating receptor by pp65 of human cytomegalovirus. Nat Immunol 2005;6:515-23.

Arnon TI, Markel G, Mandelboim O. Tumor and viral recognition by natural killer cells receptors. Semin Cancer Biol 2006;16:348-58.

Barkholt L, Alici E, Conrad R, Sutlu T, Gilliam M, Stellan B, Christensson B, Guven H, Bjorkstrom NK, Soderdahl G, Cederlund K, Kimby E, Aschan J, Ringden O, Ljunggren HG, Dilber MS. Safety analysis of ex vivo-expanded NK and NK-like T cells administered to cancer patients: a Phase I clinical study. Immunotherapy 2009;1:753-64.

Bauer S, Groh V, Wu J, Steinle A, Phillips JH, Lanier LL, Spies T. Activation of NK cells and T cells by NKG2D, a receptor for stress-inducible MICA. Science 1999;285:727-9.
Biassoni R, Cantoni C, Pende D, Sivori S, Parolini S, Vitale M, Bottino C, Moretta A. Human natural killer cell receptors and co-receptors. Immunol Rev 2001;181:203-14.
Borghaei H, Smith MR, Campbell KS. Immunotherapy of cancer. Eur J Pharmacol 2009;625:41-54.
Bryceson YT, March ME, Ljunggren HG, Long EO. Activation, coactivation, and costimulation of resting human natural killer cells. Immunol Rev 2006;214:73-91.
Caligiuri MA. Human natural killer cells. Blood 2008;112:461-9.
Carlsten M, Björkström NK, Norell H, Bryceson Y, van Hall T, Baumann BC, Hanson M, Schedvins K, Kiessling R, Ljunggren HG, Malmberg KJ. DNAX accessory molecule-1 mediated recognition of freshly isolated ovarian carcinoma by resting natural killer cells. Cancer Res 2007;67:1317-25.
Carlsten M, Norell H, Bryceson YT, Poschke I, Schedvins K, Ljunggren HG, Kiessling R, Malmberg KJ. Primary human tumor cells expressing CD155 impair tumor targeting by down-regulating DNAM-1 on NK cells. J Immunol 2009;183:4921-30.
Chan JK, Hamilton CA, Cheung MK. Enhanced Killing of Primary Ovarian Cancer by Retargeting Autologous Cytokine-Induced Killer Cells with Bispecific Antibodies: A Preclinical Study. Clin Cancer Res 2006;12:1859-67.
Cerwenka A, Lanier LL. Natural killer cells, viruses and cancer. Nat Rev Immunol 2001;1:41-49.
Cooper MA, Caligiuri MA. Isolation and characterization of human natural killer cell subsets. Curr Protoc Immunol 2004;Chapter 7:Unit 7.34.
Cooper MA, Fehniger TA, Caligiuri MA. The biology of human natural killer-cell subsets. Trends Immunol 2001;22:633-40.
Cosman D, Müllberg J, Sutherland CL, Chin W, Armitage R, Fanslow W, Kubin M, Chalupny NJ. ULBPs, novel MHC class I-related molecules, bind to CMV glycoprotein UL16 and stimulate NK cytotoxicity through the NKG2D receptor. Immunity 2001;14:123-33.
Coudert JD, Held W. The role of the NKG2D receptor for tumor immunity. Semin Cancer Biol 2006;16:333-43.
Curiel TJ, Coukos G, Zou L, Alvarez X, Cheng P, Mottram P, Evdemon-Hogan M, Conejo-Garcia JR, Zhang L, Burow M, Zhu Y, Wei S, Kryczek I, Daniel B, Gordon A, Myers L, Lackner A, Disis ML, Knutson KL, Chen L, Zou W. Specific recruitment of regulatory T cells in ovarian carcinoma fosters immune privilege and predicts reduced survival. Nat Med 2004;10:942-9.
Di Santo JP. Natural killer cell developmental pathways: a question of balance. Ann Rev Immunol 2006;24:257-86.
Diefenbach A, Raulet DH. The innate immune response to tumors and its role in the induction of T-cell immunity. Immunol Rev 2002;188:9-21.
Esser R, Müller T, Stefes D, Kloess S, Seidel D, Gillies SD, Aperlo-Iffland C, Huston JS, Uherek C, Schönfeld K, Tonn T, Huebener N, Lode HN, Koehl U, Wels WS. NK cells engineered to express a GD2-specific antiget receptor display built-in ADCC-like activity against tumor cells of neuroectodermal origin. J Cell Mol Med 2011; "Accepted Article"; doi: 10.1111/j.1582-4934.2011.01343.x.
Farag SS, Fehniger T, Ruggeri L, Velardi A, Caligiuri MA. Natural killer cells: biology and application in stem-cell transplantation. Cytotherapy 2002;4:445-6.
Fehniger TA, Cooper MA, Nuovo GJ, Cella M, Facchetti F, Colonna M, Caligiuri MA. CD56bright natural killer cells are present in human lymph nodes and are activated by T cell-derived IL-2: a potential new link between adaptive and innate immunity. Blood 2003;101:3052-7.
French AR, Yokoyama WM. Natural killer cells and viral infections. Curr Opin Immunol 2003;15:45-51.
Freud AG, Becknell B, Roychowdhury S, Mao HC, Ferketich AK, Nuovo GJ, Hughes TL, Marburger TB, Sung J, Baiocchi RA, Guimond M, Caligiuri MA. A human CD34(+) subset resides in lymph nodes and differentiates into CD56 bright natural killer cells. Immunity 2005;22:295-304.
Fuchs A, Cella M, Kondo T, Colonna M. Paradoxic inhibition of human natural interferon-producing cells by the activating receptor NKp44. Blood 2005;106:2076-82.
Garcia-Lora A, Algarra I, Garrido F. MHC class I antigens, immune surveillance, and tumor immune escape. J Cell Physiol 2003;195:346-55.
Ghiringhelli F, Ménard C, Terme M, Flament C, Taieb J, Chaput N, Puig PE, Novault S, Escudier B, Vivier E, Lecesne A, Robert C, Blay JY, Bernard J, Caillat-Zucman S, Freitas A, Tursz T, Wagner-Ballon O,

Capron C, Vainchencker W, Martin F, Zitvogel L. CD4+CD25+ regulatory T cells inhibit natural killer cell functions in a transforming growth factor-beta-dependent manner. J Exp Med 2005;202:1075-85.

Hayakawa Y, Screpanti V, Yagita H, Grandien A, Ljunggren HG, Smyth MJ, Chambers BJ NK cell TRAIL eliminates immature dendritic cells in vivo and limits dendritic cell vaccination efficacy. J Immunol 2004;172:123-9.

Herberman RB, Numm M, Lavrin DH Natural cytotoxic reactivity of mouse lymphoid cells against syngeneic acid allogeneic tumors. I. Distribution of reactivity and specificity. Int J Cancer 1975;16:216-29.

Kiessling R, Klein E, Pross H, Wigzell H. "Natural" killer cells in the mouse. II. Cytotoxic cells with specificity for mouse Moloney leukemia cells. Characteristics of the killer cell. Eur J Immunol 1975;5:117-21.

Lanier LL, Phillips JH, Hackett J Jr, Tutt M, Kumar V. Natural killer cells: definition of a cell type rather than a function. J Immunol 1986;137:2735-9.

Lanier LL. NK cell recognition. Annu Rev Immunol 2005;23:225-74.

Ljunggren HG, Karre K. In search of the missing self. MHC molecules and NK cells recognition. Immunology today 1990;11:237-244.

Ljunggren HG, Malmberg KJ. Prospects for the use of NK cells in immunotherapy of human cancer. Nat Rev Immunol 2007;7:329-39.

Lotzova E, Savary CA, Freedman R, Bowen JM. Natural immunity against ovarian tumors. Comp Immunol Microbiol Infect Dis 1986;9:269-75.

Lotzova E, Savary CA, Freedman RS, Edwards CL, Wharton JT. Recombinant IL-2-activated NK cells mediate LAK activity against ovarian cancer. Int J Cancer 1988;42:225-31.

Lutgendorf SK, Sood AK, Andersson B, McGinn S, Maiseri H, Dao M, Sorosky JI, Geest KD, Ritchie J, Lubaroff DM. Social support, psychological distress, and natural killer cell activity in ovarian cancer. J Clin Oncol 2005;23:7105-13.

Mabuchi S, Morishige K, Kimura T. Use of monoclonal antibodies in the treatment of ovarian cancer. Curr Opin Obstet Gynecol 2010;22:3-8.

MacCall AM, Shahied L, Amoroso AR, Horak EM, Simmons HH, Nielson U, Adams GP, Schier R, Marks JD, Weiner LM. Increasing the Affinity for Tumor Antigen Enhances Bispecific Antibody Cytotoxicity. J Immunol 2001;166:6112-7.

Malmberg KJ, Bryceson YT, Carlsten M, Andersson S, Björklund A, Björkström NK, Baumann BC, Fauriat C, Alici E, Dilber MS, Ljunggren HG. NK cell-mediated targeting of human cancer and possibilities for new means of immunotherapy. Cancer Immunol Immunother 2008;57:1541-52.

Malmberg KJ. Effective immunotherapy against cancer: a question of overcoming immune suppression and immune escape? Cancer Immunol Immunother 2004;53:879-92.

Mattei F, Schiavoni G, Belardelli F, Tough DF. IL-15 is expressed by dendritic cells in response to type I IFN, double-stranded RNA, or lipopolysaccharide and promotes dendritic cell activation. J Immunol 2001;167:1179-87.

Miller JS, Soignier Y, Panoskaltsis-Mortari A, McNearney SA, Yun GH, Fautsch SK *et al.* Successful adoptive transfer and in vivo expansion of human haploidentical NK cells in patients with cancer. Blood 2005;105:3051–7.

Moretta L, Moretta A. Unravelling natural killer cell function: triggering and inhibitory human NK receptors. EMBO J 2004; 23:255-9.

Onda H, Ohkubo S, Shintani Y, Ogi K, Kikuchi K, Tanaka H, Yamamoto K, Tsuji I, Ishibashi Y, Yamada T, Kitada C, Suzuki N, Sawada H, Nishimura O, Fujino M. A novel secreted tumor antigen with a glycosylphosphatidylinositol-anchored structure ubiquitously expressed in human cancers. Biochem Biophys Res Commun 2001;285:235-43.

Passweg JR, Tichelli A, Meyer-Monard S, Heim D, Stern M, Kühne T, Favre G, Gratwohl A. Purified donor NK-lymphocyte infusion to consolidate engraftment after haploidentical stem cell transplantation. Leukemia 2004;18:1835-8.

Pende D, Cantoni C, Rivera P, Vitale M, Castriconi R, Marcenaro S, Nanni M, Biassoni R, Bottino C, Moretta A, Moretta L. Role of NKG2D in tumor cell lysis mediated by human NK cells: cooperation with natural cytotoxicity receptors and capability of recognizing tumors of nonepithelial origin. Eur J Immunol 2001;31:1076-86.

Raulet DH. Roles of the NKG2D immunoreceptor and its ligands. Nat Rev Immunol 2003;3:781-90.

Robertson MJ, Caligiuri MA, Manley TJ, Levine H, Ritz J. Human natural killer cell adhesion molecules. Differential expression after activation and participation in cytolysis. J Immunol 1990;145:3194-201.

Roszkowski PI, Hyc A, Malejczyk J. Natural killer cell activity in patients with ovarian tumors and uterine myomas. Eur J Gynaecol Oncol 1993;14:114-7.
Schanoski AS, Cavalcanti TC, Campos CB, Viera-Matos AN, Rettori O, Guimarães F. Walker 256 tumor MHC class I expression during the shift from A variant to the immunogenic AR variant. Cancer Lett 2004;211:119-27.
Seimetz D, Lindhofer H, Bokemeyer C. Development and approval of the trifunctional antibody catumaxomab (anti-EpCAM x anti-CD3) as a targeted cancer immunotherapy. Cancer Tret Rev 2010;36:458-67.
Sivori S, Pende D, Bottino C, Marcenaro E, Pessino A, Biassoni R, Moretta L, Moretta A. NKp46 is the major triggering receptor involved in the natural cytotoxicity of fresh or cultured human NK cells. Correlation between surface density of NKp46 and natural cytotoxicity against autologous, allogeneic or xenogeneic target cells. Eur J Immunol 1999;29:1656-66.
Smyth MJ, Hayakawa Y, Takeda K, Yagita H. New aspects of natural-killer-cell surveillance and therapy of cancer. Nat Rev Cancer 2002;2:850-61.
Smyth MJ, Teng MW, Swann J, Kyparissoudis K, Godfrey DI, Hayakawa Y. CD4+CD25+ T regulatory cells suppress NK cell-mediated immunotherapy of cancer. J Immunol 2006;176:1582-7.
Sutlu T, Alici E. Natural killer cell-based immunotherapy in cancer: current insights and future prospects. J Intern Med 2009;266:154-81.
Wiltrout RH. Regulation and antimetastatic functions of liver-associated natural killer cells. Immunol Rev 2000;174:63-76.
Wu J, Lanier LL. Natural killer cells and cancer. Adv Cancer Res 2003;90:128-156.
Yigit R, Massuger LFAG, Figdor CG, Torensma R. Ovarian cancer creates a suppressive microenvironment to escape immune elimination. Gynecol Oncol 2010;117:366-72.