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ORIGINAL ARTICLE

Expression of interleukin-15 in canine mammary carcinoma: relationships with histologic grades, Bcl-2, recurrence, and overall survival

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Abstract

Spontaneous tumors arising from mammary tissue are considered the most common tumor in female dogs and are of great importance both in veterinary and comparative medicine. Interleukin-15 is a cytokine involved in many physiologic processes such as activation of immune cells, autoimmune diseases, and cancer pathogenesis; however, the role of interleukin-15 in canine mammary cancers has not been well understood. We designed this study to examine the expression of interleukin-15 (IL-15) in canine mammary carcinoma by means of immunohistochemistry and any possible association with histologic malignancy grades, Bcl-2 expression, tumor recurrence, and overall survival. Results revealed that 14 (46.66%) of tumor samples strongly express IL-15, 7 cases (23.33%) moderately, and 9 cases showed weak immunoreactivity (30%). The expression of interleukin-15 is increased in canine mammary tumors in comparison to healthy tissues ($p < 0.05$). Additionally, interleukin-15 higher expression is significantly in relationship with high histologic grade ($p < 0.05$), high Bcl-2 expression ($p < 0.05$), and shorter overall survival ($p < 0.05$), but not with tumor recurrence ($p > 0.05$). Interleukin-15 could have a role mammary carcinogenesis in dogs but more studies are needed to understand its exact roles.

Keywords Canine · Histologic grade · Immunohistochemistry · Interleukin-15 · Mammary carcinoma · Survival

Introduction

Canine mammary carcinoma (CMC) is the most diagnosed tumor in female dogs and shares several characteristics with human breast cancer and is a good model for comparative breast cancer studies (Khanna et al. Khanna et al. 2006; Munson Munson and Moresco 2007; Queiroga et al. 2011). Finding an appropriate model for human breast cancer has been always a challenge for scientist. Pet animal like dogs are exposed to the same insulting agents causing cancer as human beings and their tumors are spontaneous and naturally occurring as well.

Interleukin-15 (IL-15) basically regulates immune responses such as activation of immune cells particularly natural killer (NK) cell (Bancroft 1993; Fehniger and Caligiuri 2001; Meazza et al. 1996; Trinchieri 1997). IL-15 also activates Janus kinase (JAK/STAT) pathway, *src*-related tyrosine kinases, MAPK pathway, and Bcl-2 induction (Ihle et al. 1995; Miyazaki et al. 1995), all of which are involved in mechanisms that contribute to cancer development (Dhillon et al. 2007; Sen and Johnson 2011; Thomas et al. 2015).

Hyper-expression of IL-15-transgenic mice in vivo has led to development of some types of cancer. This chronic increased expression of IL-15 induces JAK1/3 and STAT3/5 signaling pathways which eventually cause neoplastic transformation. Upon activation of STAT3 pathway, the expression of Myc increases. This will result in Aurora kinase A and B overexpression which leads to chromosome instability and mir-29b downregulation which results in silencing of oncosuppressor genes (Mishra et al. 2012).

There are evidences suggesting that IL-15 could be involved in tumorigenesis. In human breast cancer tissues, IL-15 protein has been found to be decreased (Sanders et al. 2011). Likewise, an immunohistochemical study has demonstrated that IL-15 mRNA levels are decreased in lung cancer in man and is correlated with higher tumor stages (Faisal Adhami et al. 2012; Sanders et al. 2011). In opposite,

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increased levels of IL-15 mRNA and protein have been observed in some human solid tumors including osteosarcoma, rhabdomyosarcoma, melanoma, small cell lung cancer, renal cell carcinoma, glioblastoma, neuroblastoma, gastric cancer, melanoma cell lines, and mesothelioma (Barzegar et al. 1998; Kuniyasu et al. 2003; Lollini et al. 1997; Meazza et al. 1996; Trinder et al. 1999).

Targeted drug therapy against IL-15 has been tested in different hematopoietic and solid cancers including breast cancer, leukemia, renal cancer, melanoma, lymphoma, lung cancer, brain cancers, squamous cell carcinoma of the head and neck, and bladder tumors with beneficial results (Rautela and Huntington 2016; Steinway and Loughran 2013). The mentioned data has made IL-15 a promising target for anticancer therapy which is still under many investigations.

Bcl-2 belongs to the family of Bcl-2 proteins (Gross et al. 1999). It is an anti-apoptotic oncoprotein that blocks apoptosis (Cory et al. 2003; Tsujimoto et al. 1984). Inhibition of apoptosis is a strategy which is used by many of tumor cells to survive (O'Brien and Kirby O'Brien and Kirby 2008). There are several studies pointing out dysregulation of apoptosis pathways in canine mammary tumors in which Bcl-2 levels have been found to be increased or unaltered (Kumaraguruparan et al. 2006; Shokoohi et al. 2018; Shoorei et al. 2018; Yang et al. 2006).

It has been shown that IL-15 induces anti-apoptotic proteins such as Bcl-2 family of proteins; one of our goals in the present study is to determine any possible relationships between IL-15 and Bcl-2 protein expression in canine mammary carcinoma (Mishra et al. 2014; Shenoy et al. 2014).

Cytokines could be considered as potential targets for anticancer therapy in canine mammary carcinoma in future; it has been reported that transforming growth factor beta-1 (TGF-β1) and interleukin-17 are overexpressed in canine mammary carcinoma and are of prognostic significance (Rezaee et al. 2017a; Rezaee et al. 2017b). Likewise, there are other articles showing that interleukin-1, interleukin-6, and interleukin-8 are in association with some malignancy indices (Kim et al. 2010; Zuccari et al. 2011). To date, there is no data available regarding IL-15 roles in canine mammary carcinoma and this article is investigating IL-15 expression in this tumor for the first time.

This study examined IL-15 expression in canine mammary carcinoma by immunohistochemistry and its possible associations with histologic grades, Bcl-2 expression, tumor recurrence, and overall survival.

Materials and methods

Ethical approval

The conducted research is not related to animals use. No ethical approval was obtained because this study did not involve laboratory animals and only involved non-invasive procedures.

Thirty female dogs—whose mammary tumors had been removed by surgery—were included in this study. Fixation of the tissues carried on using 10% formalin, after routine processing and staining with H&E stains. "World Health Organization histological classification of the canine and feline mammary tumors" was used for classification of the slides (Misdorp 1999).

Positive controls for IL-15 and Bcl-2 were mouse kidney and tonsil lymph node respectively.

We used a previously developed method for histologic grading of the canine mammary carcinomas for each slide and finally grade them as I, II, and III (Misdorp 2002).

Immunohistochemistry

Tissues were dewaxed in warm xylene and were rehydrated in graded alcohols 100%, 95%, 80%, and 70%. We put the specimens in water bath in 95 °C for 20 min using Tris-EDTA buffer (pH 9) to retrieve possibly masked antigens. 0.3% H_2O_2 diluted in PBS was added to the slides to neutralize endogenous peroxidase enzyme.

To reduce unspecific binding, the slides were incubated with 1.5% normal goat serum in PBS for 1 h. Antibodies against IL-15 (Ab109082, Abcam) and Bcl-2 (SC-492, SantaCruz Biotechnology) were added to the slides at proper dilutions suggested by the manufacturer and were incubated overnight at 4 °C.

The method avidin-biotin complex (ABC) was used for immunostaining; biotinylated secondary antibody was added to the slides at 1:200 dilution for 30 min. Then, avidin and biotinylated horse radish peroxidase (HRP) were added and incubated for 30 min and DAB was used as chromogen. Counterstaining with Harris hematoxylin and dehydration were the final steps.

Interpretation of IHC results

Scoring was performed by two authors (MR, MM) according to a previously described method. The "distribution" of positive cells per slide was obtained by calculating mean percentage of positive-stained cell in 10 HPFs as 0 = 0%, 1 = 1–5%, 2 = 6–20%, 3 = 21–50%, or 4 = > 50% of cells. Additionally, the "intensity" of the observed signals was graded as 0 = negative, 1 = weak, 2 = moderate, 3 = strong, and 4 = intense. Final scores were obtained by multiplying the "distribution score" and the "intensity score", ranging from 0 to 16. The examined slides were finally categorized into 4 groups as follows: negative (0), weak (1–3), moderate (4–7), and strong expression (8–16).

Statistical analysis We performed statistical analysis by SPSS using chi-square test and $p < 0.05$ was considered as significant.

Results

Thirty tumor samples were classified as anaplastic carcinoma (5/30, 16.66%), carcinosarcoma (8/30, 26.66%), solid carcinoma (7/30, 23.33%), mixed-type carcinoma (5/30, 16.66%), and complex-type carcinoma (5/30, 16.66%). Ten samples were grade III (33.33%), 15 were grade II (50%), and 5 were grade I (16.66%).

Bcl-2 cytoplasmic immunolabeling was observed in neoplastic cells in different levels. Twelve cases (40%) showed strong immunoreactivity (Fig. 1), 8 cases (26.66%) moderate immunoreactivity, and 10 cases (33.33%) weak.

In healthy mammary glands, IL-15 cytoplasmic and nuclear staining was absent (Fig. 2). Increased cytoplasmic IL-15 expression was observed in most of CMCs in comparison to normal tissues. IL-15 was expressed in all of the examined cases in different extends.

Fourteen (46.66%) of tumors samples show strong immunoreactivity, 7 cases (23.33.%) showed moderate reactions while 9 cases showed weak immunoreactivity (30%) for IL-15. Those cases with grades II and III had stronger immune-labelling for IL-15 than tumors with grade I. There was a significant relationship between higher IL-15 expression and higher histological grade ($p < 0.05$). Additionally, IL-15 expression was increased significantly in CMCs compared to normal tissues ($p < 0.05$). Statistical analyses showed that there is a significant correlation between high immunohistochemical expression of IL-15 protein with high Bcl-2 expression ($p < 0.05$) and shorter overall survival ($p < 0.05$), but not with tumor recurrence ($p > 0.05$) (Fig. 3).

Fourteen (46.66%) dogs were still alive after 1 year, and 16 (53.33%) dogs had died due to mammary cancer and recurrence of tumors was observed in 12 cases. The results are summarized in detail in Table 1.

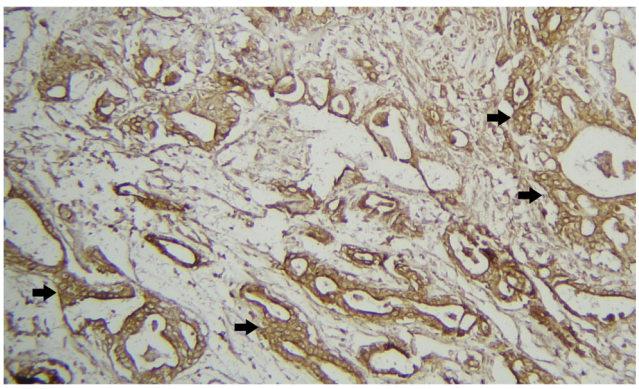

Fig. 1 Anaplastic carcinoma, strong expression of Bcl-2 in hyper-cellular cancerous epithelial parts (arrows), mammary gland tumor, Dog. 200 ×. IHC for Bcl-2

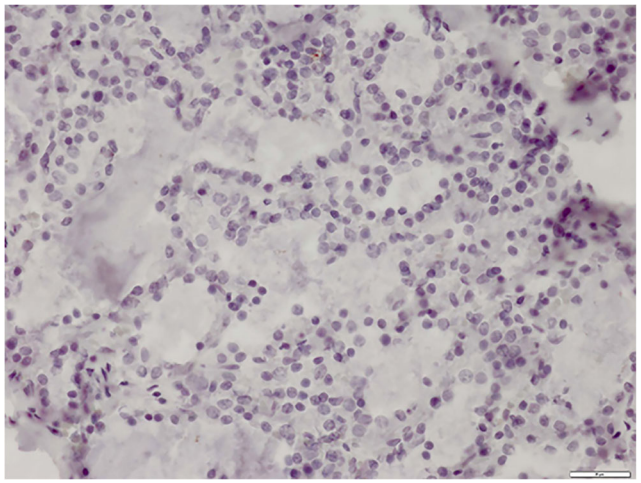

Fig. 2 Healthy mammary gland tumor, absent expression of IL-15, Mammary gland tumor, Dog. 200 ×. IHC for IL-15

Discussion

This study is designed to evaluate IL-15 expression in CMC and its possible relationships with histologic grades, Bcl-2 expression, tumor recurrence, and survival. It is not yet clear whether IL-15 can promote cancer or is an inhibitory factor for tumor formation; there are several studies supporting both ideas which are briefly discussed below.

In the present study, IL-15 higher protein expression is in association with higher histologic grades and shorter overall survival. Finding reliable markers to predict canine mammary tumor prognosis is important and is a common interest among veterinary pathologists and oncologists. Among different potential markers, cytokines are considered interesting subjects and there are increasing interests to evaluate cytokines' roles in oncogenesis.

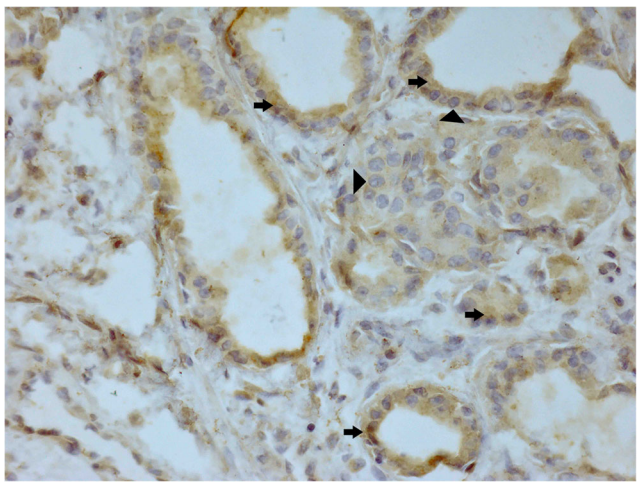

Fig. 3 Anaplastic carcinoma, strong expression of IL-15 in the cytoplasm (arrows), nuclei of the epithelial cells (arrowhead) mammary gland tumor, Dog. 400 ×. IHC for IL-15

Table 1 Details about IL-15 expression, Bcl-2 expression, histologic grades, overall survival, and recurrence in the samples used in this study

No.	Type	Grade	IL-15 expression level	Bcl-2 expression level	Recurrence	Survival
1	Anaplastic carcinoma	3	Strong	Strong	No	No
2	Complex type carcinoma	1	Strong	Moderate	No	No
3	Solid carcinoma	3	Weak	Weak	No	Yes
4	Solid carcinoma	2	Strong	Strong	No	Yes
5	Solid carcinoma	2	Weak	Weak	No	Yes
6	Solid carcinoma	2	Moderate	Weak	No	No
7	Carcinosarcoma	3	Strong	Strong	No	No
8	Mixed-type carcinoma	2	Weak	Weak	No	Yes
9	Carcinosarcoma	2	Weak	Weak	No	Yes
10	Carcinosarcoma	2	Moderate	Moderate	No	No
11	Complex type carcinoma	1	Strong	Strong	No	Yes
12	Solid Carcinoma	3	Strong	Weak	Yes	Yes
13	Complex type carcinoma	1	Weak	Weak	No	No
14	Solid carcinoma	3	Strong	Strong	Yes	Yes
15	Mixed-type carcinoma	2	Weak	Strong	No	Yes
16	Carcinosarcoma	2	Moderate	Moderate	No	No
17	Solid carcinoma	3	Strong	Moderate	No	No
18	Mixed-type carcinoma	2	Moderate	Strong	No	No
19	Complex type carcinoma	2	Strong	Strong	No	No
20	Carcinosarcoma	2	Moderate	Weak	Yes	No
21	Carcnosarcoma	3	Strong	Strong	No	No
22	Complex type carcinoma	1	Weak	Weak	Yes	Yes
23	Mixed-type carcinoma	2	Weak	Weak	No	Yes
24	Anaplastic carcinoma	3	Strong	Strong	No	No
25	Anaplastic carcinoma	2	Strong	Moderate	No	No
26	Anaplastic carcinoma	3	Strong	Strong	Yes	No
27	Anaplastic carcinoma	3	Strong	Strong	No	No
28	Carcinosarcoma	2	Moderate	Moderate	No	Yes
29	Carcinosarcoma	1	Weak	Moderate	No	Yes
30	Mixed-type carcinoma	2	Moderate	Moderate	No	Yes

Furthermore, we found that IL-15 protein expression is significantly increased in CMCs in comparison to normal mammary tissues. In contrast to our findings, there are some studies showing that IL-15 protein expression is decreased in breast cancer tissues; IL-15 mRNA levels are also reduced in human lung cancer which is in correlation with higher tumor stages (Faisal Adhami et al. 2012; Sanders et al. 2011). IL-15 transcripts and protein levels have been observed in some human solid tumors including osteosarcoma, rhabdomyosarcoma, melanoma, small cell lung cancer, renal cell carcinoma, glioblastoma, neuroblastoma, melanoma cell lines, and mesothelioma (Barzegar et al. 1998; Lollini et al. 1997; Meazza et al. 1996; Trinder et al. 1999). Additionally, human colon cancer cells express IL-15 that is responsible for progression of tumor and angiogenesis (Kuniyasu et al. 2003).

IL-15 has been used to treat different cancers including human breast cancer (Rautela and Huntington 2016). Furthermore, IL-15 has inhibitory effects on metastasis rate in breast cancer (Gillgrass et al. 2014).

The importance of Bcl-2 expression in this study is its relationships with IL-15 expression and finding a clue for possible role of IL-15 in apoptosis induction in CMC. In the present study, we found a significant relationship between IL-15 and Bcl-2 expression ($p < 0.05$). One of many biologic roles of IL-15 is induction of anti-apoptotic factors such as Bcl-2. IL-15 is able to support cell expansion and maintenance in different ways: one way is inducing JAK/STAT and Ras/MAPK signaling pathways which lead to proliferation. Besides, IL-15 prevents cell death by inducing anti-apoptotic proteins such as Bcl-2 as well as inhibiting pro-apoptotic proteins like Bim and Puma (Adunyah et al. 1997; Huntington et al. 2007; Johnston et al. 1995; Miyazaki et al. 1994; Miyazaki et al. 1995; Steelman et al. 2004).

IL-15-mediated inhibition of apoptosis has been observed in different neoplasms including multiple myeloma, lymphoma, and leukemia in which presence of an autocrine loop of IL-15 production by malignant cells is suggested (Dobbeling et al. 1998; Hodge et al. 2009; Tinhofer et al. 2000). Our data may suggest that production of IL-15 by CMC cells can activate Bcl-2 and results in evasion of apoptosis, but more studies are needed.

Here, we observed IL-15 overexpression in CMC samples compared to normal mammary tissues. IL-5 higher expression was seen mainly in samples with higher histologic grades of malignancy and this association was statistically significant. Besides, there is a significant relationship between increased expression of Bcl-2 and IL-15. This could shed light to possible roles of IL-15 in CMC progression. As discussed above, IL-15 may activate some subcellular signaling pathways such as JAK/STAT and subsequently help cancer cell continue growing. Possible activation of Bcl-2 by IL-15 autocrine loop is also another mechanism which is also possible in CMC. These hypotheses are needed to be further tested to become facts. For example, we can use CMC cell culture to see whether IL-15 induction leads to Bcl-2 higher expression and higher growth rate or not.

The present article indicates that IL-15 expression is significantly correlated with higher histologic grades, Bcl-2 expression, and shorter overall survival. Repeating similar studies to ours with more samples and evaluating IL-15 mRNA level can lead to better clarification of IL-15 roles in CMC. To better understand the exact role of IL-15 in CMC, more studies are required.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval All applicable international, national, and/or institutional guidelines for the care and use of animals were followed.

References

Faisal Adhami, Jason C Steel, Morris JC 2012 Interleukin-15 expression in lung cancer. In: ASCO Annual Meeting,

Adunyah SE, Wheeler BJ, Cooper RS (1997) Evidence for the involvement of LCK and MAP kinase (ERK-1) in the signal transduction mechanism of interleukin-15. Biochem Biophys Res Commun 232: 754–758. https://doi.org/10.1006/bbrc.1997.6367

Bancroft GJ (1993) The role of natural killer cells in innate resistance to infection. Curr Opin Immunol 5:503–510. https://doi.org/10.1016/0952-7915(93)90030-V

Barzegar C, Meazza R, Pereno R, Pottin-Clemenceau C, Scudeletti M, Brouty-Boyé D, Doucet C, Taoufik Y, Ritz J, Musselli C, Mishal Z, Jasmin C, Indiveri F, Ferrini S, Azzarone B (1998) IL-15 is produced by a subset of human melanomas, and is involved in the regulation of markers of melanoma progression through juxtacrine loops. Oncogene 16:2503–2512. https://doi.org/10.1038/sj.onc.1201775

Cory S, Huang DC, Adams JM (2003) The Bcl-2 family: roles in cell survival and oncogenesis. Oncogene 22:8590–8607. https://doi.org/10.1038/sj.onc.1207102

Dhillon AS, Hagan S, Rath O, Kolch W (2007) MAP kinase signalling pathways in cancer. Oncogene 26:3279–3290

Dobbeling U, Dummer R, Laine E, Potoczna N, Qin JZ, Burg G (1998) Interleukin-15 is an autocrine/paracrine viability factor for cutaneous T-cell lymphoma cells. Blood 92:252–258

Fehniger TA, Caligiuri MA (2001) Interleukin 15: biology and relevance to human disease. Blood 97:14–32

Gillgrass A, Gill N, Babian A, Ashkar AA (2014) The absence or overexpression of IL-15 drastically alters breast cancer metastasis via effects on NK cells, CD4 T cells, and macrophages. J Immunol 193:6184–6191. https://doi.org/10.4049/jimmunol.1303175

Gross A, McDonnell JM, Korsmeyer SJ (1999) BCL-2 family members and the mitochondria in apoptosis. Genes Dev 13:1899–1911

Hodge DL, Yang J, Buschman MD, Schaughency PM, Dang H, Bere W, Yang Y, Savan R, Subleski JJ, Yin XM, Loughran TP, Young HA (2009) Interleukin-15 enhances proteasomal degradation of bid in normal lymphocytes: implications for large granular lymphocyte leukemias. Cancer Res 69:3986–3994. https://doi.org/10.1158/0008-5472.can-08-3735

Huntington ND, Puthalakath H, Gunn P, Naik E, Michalak EM, Smyth MJ, Tabarias H, Degli-Esposti MA, Dewson G, Willis SN, Motoyama N, Huang DCS, Nutt SL, Tarlinton DM, Strasser A (2007) Interleukin 15-mediated survival of natural killer cells is determined by interactions among Bim, Noxa and Mcl-1. Nat Immunol 8:856–863. https://doi.org/10.1038/ni1487

Ihle JN, Witthuhn BA, Quelle FW, Yamamoto K, Silvennoinen O (1995) Signaling through the hematopoietic cytokine receptors. Annu Rev Immunol 13:369–398. https://doi.org/10.1146/annurev.iy.13.040195.002101

Johnston JA, Bacon CM, Finbloom DS, Rees RC, Kaplan D, Shibuya K, Ortaldo JR, Gupta S, Chen YQ, Giri JD (1995) Tyrosine phosphorylation and activation of STAT5, STAT3, and Janus kinases by interleukins 2 and 15. Proc Natl Acad Sci U S A 92:8705–8709

Khanna C, Lindblad-Toh K, Vail D, London C, Bergman P, Barber L, Breen M, Kitchell B, McNeil E, Modiano JF, Niemi S, Comstock KE, Ostrander E, Westmoreland S, Withrow S (2006) The dog as a cancer model. Nat Biotechnol 24:1065–1066

Kim JH, Yu CH, Yhee JY, Im KS, Sur JH (2010) Lymphocyte infiltration, expression of interleukin (IL) -1, IL-6 and expression of mutated breast cancer susceptibility gene-1 correlate with malignancy of canine mammary tumours. Journal of comparative pathology 142:177-186. https://doi.org/10.1016/j.jcpa.2009.10.023

Kumaraguruparan R, Prathiba D, Nagini S (2006) Of humans and canines: immunohistochemical analysis of PCNA, Bcl-2, p53, cytokeratin and ER in mammary tumours. Res Vet Sci 81:218–224. https://doi.org/10.1016/j.rvsc.2005.08.002

Kuniyasu H, Ohmori H, Sasaki T, Sasahira T, Yoshida K, Kitadai Y, Fidler IJ (2003) Production of interleukin 15 by human colon cancer cells is associated with induction of mucosal hyperplasia, angiogenesis, and metastasis. Clin Cancer Res 9:4802–4810

Lollini P-L, Palmieri G, de Giovanni C, Landuzzi L, Nicoletti G, Rossi I, Griffoni C, Frabetti F, Scotlandi K, Benini S, Baldini N, Santoni A, Nanni P (1997) Expression of interleukin 15 (IL-15) in human rhabdomyosarcoma, osteosarcoma and Ewing's sarcoma. Int J Cancer 71:732–736. https://doi.org/10.1002/(SICI)1097-0215(19970529)71:5<732::AID-IJC7>3.0.CO;2-S

Meazza R, Verdiani S, Biassoni R, Coppolecchia M, Gaggero A, Orengo AM, Colombo MP, Azzarone B, Ferrini S (1996) Identification of a novel interleukin-15 (IL-15) transcript isoform generated by alternative splicing in human small cell lung cancer cell lines. Oncogene 12:2187–2192

Misdorp W (1999) Histological classification of mammary tumors of the dog and the cat. Armed Forces Institute of Pathology,

Misdorp W (2002) Tumors of the mammary gland. In: DJ M (ed) tumors in domestic animals. 4th edn. Iowa State Press, Ames, IA, pp 575–606, 764

Mishra A, Liu S, Sams GH, Curphey DP, Santhanam R, Rush LJ, Schaefer D, Falkenberg LG, Sullivan L, Jaroncyk L, Yang X, Fisk H, Wu LC, Hickey C, Chandler JC, Wu YZ, Heerema NA, Chan KK, Perrotti D, Zhang J, Porcu P, Racke FK, Garzon R, Lee RJ, Marcucci G, Caligiuri MA (2012) Aberrant overexpression of IL-15 initiates large granular lymphocyte leukemia through chromosomal instability and DNA hypermethylation. Cancer Cell 22:645–655. https://doi.org/10.1016/j.ccr.2012.09.009

Mishra A, Sullivan L, Caligiuri MA (2014) Molecular pathways: Interleukin-15 signaling in health and in cancer. Clin Cancer Res 20:2044–2050. https://doi.org/10.1158/1078-0432.ccr-12-3603

Miyazaki T et al (1994) Functional activation of Jak1 and Jak3 by selective association with IL-2 receptor subunits. Science (New York, NY) 266:1045–1047

Miyazaki T, Liu ZJ, Kawahara A, Minami Y, Yamada K, Tsujimoto Y, Barsoumian EL, Perlmutter RM, Taniguchi T (1995) Three distinct IL-2 signaling pathways mediated by bcl-2, c-myc, and lck cooperate in hematopoietic cell proliferation. Cell 81:223–231

Munson L, Moresco A (2007) Comparative pathology of mammary gland cancers in domestic and wild animals. Breast Dis 28:7–21

O'Brien MA, Kirby R (2008) Apoptosis: a review of pro-apoptotic and anti-apoptotic pathways and dysregulation in disease. J Vet Emerg Crit Care 18:572–585. https://doi.org/10.1111/j.1476-4431.2008.00363.x

Queiroga FL, Raposo T, Carvalho MI, Prada J, Pires I (2011) Canine mammary tumours as a model to study human breast cancer: most recent findings. In Vivo 25:455–465

Rautela J, Huntington ND (2016) IL-15 signaling in NK cell cancer immunotherapy. Curr Opin Immunol 44:1–6. https://doi.org/10.1016/j.coi.2016.10.004

Rezaee M, Movassaghi AR, Dehghani H (2017a) Strong expression of interleukin-17 is associated with higher histologic grades in canine mammary carcinoma. Comp Clin Pathol 26:477–481. https://doi.org/10.1007/s00580-017-2403-9

Rezaee M, Movassaghi AR, Maleki M (2017b) Immunohistochemical expression of transforming growth factor Beta-1 in canine mammary carcinomas: its relationships with histologic grading, survival rate, and recurrence. Comparative Clinical Pathology:1-6 doi: https://doi.org/10.1007/s00580-017-2410-x

Sanders A, Ye L, Wei X, Mansel R, Jiang W (2011) P1-01-08: expression of Interleukin-15 (IL-15) and the IL-15 receptor in human breast cancer. Cancer Research 71:P1-01-08 https://doi.org/10.1158/0008-5472.sabcs11-p1-01-08

Sen B, Johnson FM (2011) Regulation of Src family kinases in human cancers. Journal of Signal Transduction 2011:14–14. https://doi.org/10.1155/2011/865819

Shenoy AR, Kirschnek S, Häcker G (2014) IL-15 regulates Bcl-2 family members Bim and Mcl-1 through JAK/STAT and PI3K/AKT pathways in T cells. Eur J Immunol 44:2500–2507

Shokoohi M, Shoorei H, Soltani M, Abtahi-Eivari SH, Salimnejad R, Moghimian M (2018) Protective effects of the hydroalcoholic extract of Fumaria parviflora on testicular injury induced by torsion/detorsion in adult rats.e13047. https://doi.org/10.1111/and.13047

Shoorei H, Khaki A, Ainehchi N, Hassanzadeh Taheri MM, Tahmasebi M, Seyedghiasi G, Ghoreishi Z, Shokoohi M, Khaki AA, Abbas Raza SH (2018) Effects of Matricaria chamomilla extract on growth and maturation of isolated mouse ovarian follicles in a three-dimensional culture system. Chin Med J 131:218–225. https://doi.org/10.4103/0366-6999.222324

Steelman LS, Pohnert SC, Shelton JG, Franklin RA, Bertrand FE, McCubrey JA (2004) JAK/STAT, Raf/MEK/ERK, PI3K/Akt and BCR-ABL in cell cycle progression and leukemogenesis. Leukemia 18:189–218. https://doi.org/10.1038/sj.leu.2403241

Steinway SN, Loughran TP (2013) Targeting IL-15 in large granular lymphocyte leukemia. Expert Rev Clin Immunol 9:405–408. https://doi.org/10.1586/eci.13.28

Thomas SJ, Snowden JA, Zeidler MP, Danson SJ (2015) The role of JAK/STAT signalling in the pathogenesis, prognosis and treatment of solid tumours. Br J Cancer 113:365–371. https://doi.org/10.1038/bjc.2015.233

Tinhofer I, Marschitz I, Henn T, Egle A, Greil R (2000) Expression of functional interleukin-15 receptor and autocrine production of interleukin-15 as mechanisms of tumor propagation in multiple myeloma. Blood 95:610–618

Trinchieri G (1997) Cytokines acting on or secreted by macrophages during intracellular infection (IL-10, IL-12, IFN-gamma). Curr Opin Immunol 9:17–23. https://doi.org/10.1016/s0952-7915(97)80154-9

Trinder P, Seitzer U, Gerdes J, Seliger B, Maeurer M (1999) Constitutive and IFN-gamma regulated expression of IL-7 and IL-15 in human renal cell cancer. Int J Oncol 14:23–31

Tsujimoto Y, Finger LR, Yunis J, Nowell PC, Croce CM (1984) Cloning of the chromosome breakpoint of neoplastic B cells with the t(14;18) chromosome translocation. Science (New York, NY) 226:1097-1099

Yang WY, Liu CH, Chang CJ, Lee CC, Chang KJ, Lin CT (2006) Proliferative activity, apoptosis and expression of oestrogen receptor and Bcl-2 oncoprotein in canine mammary gland tumours. J Comp Pathol 134:70–79. https://doi.org/10.1016/j.jcpa.2005.07.002

Zuccari DA, Castro R, Gelaleti GB, Mancini UM (2011) Interleukin-8 expression associated with canine mammary tumors. Genet Mol Res : GMR 10:1522–1532