

## Thyroid cancer phenotypes in relation to inflammation and autoimmunity

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### 1. ABSTRACT

Thyroid cancer represents the most frequent endocrine neoplasm and is epidemiologically linked to a growing incidence worldwide, which is only in part explained by the increased detection of small cancers in a preclinical stage. Understanding the molecular pathogenesis of well-differentiated thyroid cancers and poorly-differentiated thyroid cancers has prompted interest into the identification of crucial signaling pathways and molecular derangements related to genetic and epigenetic alterations. Increasing attention has been recently focused on inflammation and immunity as major culprit mechanisms involved in thyroid tumorigenesis, through the detection of activated immune cells, pro-inflammatory cytokines, as well as signal integrations between inflammatory and proliferative pathways within the thyroid tumour micro-environment. In addition to playing important roles in tumour surveillance and rejection, the presence of tumour-associated macrophages and the activation of NF- $\kappa$ B signaling pathway are now reckoned as hallmarks and crucial mediator of inflammation-induced growth and progression of thyroid cancer. Thorough understanding of this immunological link and identification of novel molecular targets could

provide unprecedented opportunities for research and development of diagnostic, prognostic and treatment strategies for thyroid cancer.

### 2. INTRODUCTION

Thyroid cancer is the most frequent endocrine neoplasia and its incidence rates are on the rise (1). Thyroid cancers are usually follicular or para-follicular in their origin, and lesions developing from follicular cells include well differentiated thyroid cancers (DTC), poorly differentiated (PDTC) and anaplastic (ATC) thyroid carcinomas (2). DTCs, which encompass papillary cancers (PTC) and follicular cancers (FTC), usually show a good prognosis after surgery and radioiodine therapy, yet 5–10% of cases progress to radioiodine refractory-disease. On the other hand, PDTCs and ATCs are therapy-resistant and prognosis is unfavorable (3). Medullary thyroid carcinoma (MTC), which originates from the para-follicular C-cells, is either sporadic or familial and, by the time of diagnosis, shows high rate of lymph node metastases, which can elude detection pre-operatively or even intra-operatively (4).

The role of inflammation in DTCs has been the focus of several studies published in the last 10 years, which have demonstrated positive associations between chronic inflammation and increased risk of developing DTC, suggesting that the inflammatory microenvironment is an essential component of cellular transformation and tumour progression (5-8). In support of this inference, there has been demonstration that local activities engaged in thyroid tumorigenesis and thyroid cancer progression are positively influenced by two major inflammatory components: inflammatory cells along with their humoral mediators presenting within the cancer site, and activation of oncoprotein-mediated signalling present in epithelial cancer cells. Inflammatory cells and mediators enrich the tumour stroma and partake in several processes such as tissue remodelling, tissue repair and neo-angiogenesis (9). Cancer stroma englobes both inflammatory cells engaged in antitumour effects and activated immune cells capable of pro-tumour immune responses, and the balance of antitumour/protumour immune responses culminates in the regulation of cancer suppression vs cancer progression (7,10). Cancer stroma thus plays a dominant role in the development and progression of DTCs (11-13), as well as it shows a potential role in MTC and PDCs (5,14). The aim of this article is to provide an overview on the relationship between inflammation and thyroid cancer in relation to the immune mechanisms regulating thyroid cancer progression. To this purpose, our at-a-glance description will mainly focus on DTCs, where essential evidences related to different histotypes and relative clinical aspects will be collectively described, whereas information resumed in MTC and ATC will be presented separately.

### 3. INFLAMMATION AND DTCs

In general, two pathways have been proposed to explain the link between inflammation and DTCs (15): the extrinsic (microenvironment-driven) and the intrinsic (oncogene-driven) pathways.

#### 3.1. Extrinsic pathway

##### 3.1.1. Tumour-infiltrating inflammation

The extrinsic pathway is triggered by tumour-infiltrating inflammation that includes leukocyte infiltration, tumour-associated macrophages (TAM), cytokines, chemokines and vascular endothelial growth factor (VEGF) (16). In PTC, TAM and immature dendritic cells accumulate both in tumour stroma and at the invasive front of the tumor, and the strength of their infiltrate positively correlates with capsular invasion, extra-thyroidal extension and aggressive behaviour of the tumour (17).

According to a classical view, tumours are encircled by extracellular matrix (ECM) with its structural and specialized proteins, as well as by stromal cells, which comprises cancer-associated fibroblasts (CAFs), innate and adaptive immune cells, specialized mesenchymal cells, endothelial cells and pericytes. CAFs impact cancer progression by a number of mechanisms, such as remodelling of the ECM, induction of angiogenesis, recruitment of inflammatory cells, and by redirecting cancer cell proliferation via secretion of growth factors, immune suppressive cytokines, and mesenchymal-epithelial cell interactions (18). Cancer stroma contributes to create the so-called tumour microenvironment (TME), which enfolds a significant inflammatory cell infiltrate capable of acting pleiotropically (19). While some cells are engaged in antitumour effects (i.e., dendritic cells, tumour infiltrating lymphocytes, M1 macrophages), others instigate a pro-tumour immune response (i.e., neutrophils, mast cells, NK cells, tumour infiltrating lymphocytes, M2 macrophages), such that the (un)balance of antitumour/protumour immune responses intervenes to regulate cancer suppression vs cancer progression (7,10). Per se, TME acts as a reservoir of protumorigenic and proangiogenic cytokines, which are either produced by tumour cells, CAFs, innate or adaptive immune cells. The resulting inflammatory microenvironment is associated with the production of reactive oxygen species and oxidative stress initiated by the transcription of NF- $\kappa$ B, and perpetuated by activation of the MAPK pathway (19). Several lines of evidence state that TAMs promote cancer cell survival, proliferation, metastases, angiogenesis and immune suppression (20). Likely, the impact of TAMs on tumour progression depends on their specific reprogramming within the tumour. This process is influenced by factors related to local microenvironment such as hypoxia, locally released mediators (i.e. cytokines, growth factors), as well as metabolic products either released by cancer cells or other immune and stroma-related cells (21). Although pathways and mechanisms involved in TAMs reprogramming are not completely understood, activation of the AKT/mTOR pathway has been hypothesized to influence the inflammatory phenotype of DTC-associated macrophages (22).

Furthermore, Melillo et al (6) demonstrated the existence of a complex relationship between mast cells and thyroid cancer cells. In PTC, an increased density of mast cells was observed with respect to normal tissues, and this process is seemingly associated with a worse prognosis (6). *In vitro*, PTC cells recruited mast cells through tumour-derived VEGF-A; in turn, mast cells acted via histamine and chemokines (ex. CXCL-1, CXCL10) to induce cancer cell proliferation, survival and migration, by means of autocrine and a paracrine loops (6). Also TAMs and

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mast cells have been experimentally and clinically shown to promote the progression of PTC (23).

### 3.1.2. Obesity-dependent inflammation

Obesity enhances the risk of at least 13 different cancers, and is a risk factor for tumour recurrence after curative surgery, poor survival, non-cancer-related as well as cancer-related mortality (24,25). Epidemiological linkage between excess body weight and thyroid cancers has been highlighted in cross-sectional studies (26,27) and confirmed in meta-analyses of cross-sectional (28) and prospective studies (26,27,29). In a pooled scrutiny of five U.S prospective studies, BMI was associated with thyroid cancer risk in both men and women, independent of tumour histology (30). Several lines of evidence suggest a potential role for adipose tissue (AT) accumulation in regulating TME pathophysiology, supported by associations found between obesity-dependent inflammation and cancer (31). As such, there is demonstration that hypoxia, chronic inflammation and oxidative stress, which are typical of obese subjects, could favour the development of a subgroup of DTCs characterized by resistance to both <sup>131</sup>I treatment and chemotherapy (31-33). The relevance of AT biology in thyroid tumourigenesis is mainly related to its ability to intervene both as a reservoir and regulator of key elements of the immune system, which involve production of immunomodulatory molecules and expression of their receptors within the AT (34,35).

It is known that AT englobes different cell populations, e.g. preadipocytes, mature adipocytes as well as immune and stromal cells. Pre-adipocytes have functional characteristics and transcriptional patterns of multipotent cells that are similar to immune cells, and can transdifferentiate into macrophages both *in vitro* and *in vivo* (36,37). Mature adipocytes share the ability to secrete cytokines acting as pro- or anti-inflammatory factors related to AT accumulation. Immune cells englobed in AT include pro-inflammatory T lymphocytes (predominantly CD8+), which contribute to local inflammatory cell activation by attracting macrophages, which perpetuate the inflammatory response within the AT (38,39). Both M1 and M2 macrophages can be found in AT (40). While resident M2 macrophages play dominant roles in AT physiology and are able to produce anti-inflammatory cytokines, M1-like macrophages are recruited and clustered within the AT as crown-like structures (CLSs) and, upon stimulation by IFN $\gamma$  or lipopolysaccharide, they are able to produce pro-inflammatory cytokines, thus contributing to inflammatory pathways relating to insulin resistance (41-44). This suggests that M1 and M2 play opposed roles in AT pathophysiology as compared to that seen in DTCs. AT hypoxia may promote the M2 to M1 switching (45), and these macrophages are responsible for AT expression of TNF $\alpha$ , as well as production of discrete amounts of iNOS and IL6 (43). TNF $\alpha$  is a cytokine

capable of anti-proliferative actions in a human PTC line, through a receptor-mediated mechanism (46). Nevertheless, the high TNF $\alpha$  exposure related to obesity seems to induce a state of TNF $\alpha$  resistance, which ultimately facilitates thyroid tumour progression (31,46) and metastatic diffusion (47). IL-6 is a cytokine involved in tumourigenesis (48), but its role in thyroid cancer is still confusing. Although results linking directly thyroid cancer to IL-6 are scant (49,50), IL-6 could be important for the inflammation microenvironment in thyroid carcinogenesis, influencing DTC development and progression (7,48). Recently, Kobawala et al (51) demonstrated that IL-6 mRNA expression is higher in the primary tumour tissues of PTC patients as compared to the corresponding adjacent normal tissues, and that serum IL-6 correlates with larger tumour size, presence of distant metastasis, extra-thyroidal extension and poor overall survival.

The obesity-related adipocytokine network could also play a role in relation to the development of thyroid cancer (7,31,33,52). Mature adipocytes produce leptin, which is involved in activation of monocytes and macrophages, stimulation of VEGF and angiogenesis, and suppression of anti-inflammatory cytokines. Leptin promotes cell migration of PTC, while inhibiting the migration of follicular cells (53). Studies *in vitro* demonstrated that leptin is able to stimulate a more aggressive PTC phenotype by activating the PI3K/AKT pathway (54). Moreover, leptin promotes the de-differentiation of thyroid cancer cells via the JAK2/STAT3 signalling pathway (55). Recently, Fan *et al* (56) demonstrated that leptin had negative prognostic significance in PTC, whereas it may play a protective role in FTC.

On the contrary, adiponectin, an adipocytokine capable of exerting strong anti-inflammatory, proapoptotic and antiproliferative effects, appears to be inversely correlated with occurrence and proliferation of DTCs, hence suggesting its protective effect against thyroid tumourigenesis (57). Accordingly, Cheng *et al* (58) found that, when tissues were negative for adiponectin receptors, tumours were significantly associated with extrathyroidal invasion, multicentricity, and higher TNM stage, demonstrating that the expression of adiponectin receptors could associate with a better prognosis. Further potential links between obesity-related inflammation and DTC are represented by ghrelin and obestatin. In particular, a recent review reported that lower levels of ghrelin would favour thyroid cell proliferation, whereas supra-physiological levels would have an inhibitory effect (33).

Obesity-induced inflammation involves other inflammatory components that could contribute to tumourigenesis. These components include matrix metalloproteinases (MMPs), which are associated with cancer-cell invasion and metastasis (59-62).

Finally, the possible association between obesity and autoimmune thyroid diseases might also play a role because of the link between chronic autoimmune thyroiditis and thyroid cancer (63).

### 4. INTRINSIC PATHWAY

#### 4.1. Genetic alterations

The intrinsic pathway is driven by genetic alterations most frequently found in association with DTC, such as RET/PTC rearrangement and BRAF point mutation. Up to 70% of PTCs express non-overlapping mutations of RET, TRKA, RAS and BRAF genes, which encode the transcription of components of the mitogen-activated protein kinase (MAPK) cascade (5). Both point-mutations and genetic rearrangements can promote the constitutive activation of the tyrosine kinase activity of RET in the absence of ligands. Activation of RET by physiological ligands or by oncogenic conversion results in the phosphorylation of intracellular tyrosine residues, which serve as docking sites for the recruitment of signalling adapters (64-66).

Local activities engaged in thyroid tumorigenesis and thyroid cancer progression are positively influenced by the activation of oncoprotein-mediated signalling present in epithelial cancer cells. Studies investigating the role of the RET/PTC3 oncoprotein in the recruitment of immune cell populations into the tumour site (67-70) showed that the transplantation of RET/PTC3-expressing thyrocytes activates an inflammatory transcriptional program both *in vitro* and *in vivo*, and PTC-like lesions in mice were characterized by a leukocytic infiltrate mainly constituted by macrophages, with parallel increase in cytokine production within the tumour (71). Main humoral components of this program include mediators responsible for different pro-tumour effects, such as growth factors implicated in leucocyte recruitment and survival (G-CSF; GM-CSF, M-CSF), chemokines (CCL2, CXCL12), chemokine receptors (i.e. CXCR4) implicated in monocyte recruitment, angiogenesis and tumour-cell homing to lymph nodes, IL-8, L-selectin and proteases responsible for tumour invasion and dissemination. RET/PTC3-positive thyroid cancers were also found to induce recruitment of CD11b+, Gr1+ cells capable of mediating tumour escape from the immune surveillance (72). Oppositely, the expression of the RET/PTC3 isoform in a rat thyroid cell line (PC Cl3) was demonstrated to increase NF- $\kappa$ B DNA-binding activity with consequent increase in the pro-inflammatory cytokine secretion (73).

NF- $\kappa$ B is a transcription factors laying at the intersection between the intrinsic and extrinsic proinflammatory pathways related to tumorigenesis (15). High constitutive expression of NF- $\kappa$ B is a primary feature of cancer cells but not normal cells, indicating

a crucial role for NF- $\kappa$ B in regulating tumorigenesis (74). NF- $\kappa$ B comprises a family of transcription factors involved in transcription of different genes controlling apoptosis, immune response and inflammation, as well as cancer development and progression. Activation of NF- $\kappa$ B results from different signalling pathways triggered by cytokines, growth factors, and tyrosine kinases (75,76). NF- $\kappa$ B is also recognized to play a major role in the initiation and progression of thyroid carcinoma (77,78). In thyroid cancer cells, oncogenic proteins RET/PTC, RAS and BRAF can induce NF- $\kappa$ B activation in PTC, FTC, and MTC, while constitutively de-regulated NF- $\kappa$ B activity has been found in ATC (75). In a subset of PTCs associated with unfavourable outcome, it has been shown that NF- $\kappa$ B-mediated anti-apoptotic effects are enhanced by over-activation of Ras-related C3 botulinum toxin substrate 1 (RAC-1b), a hyperactive variant of the RAS superfamily of small GTP-binding proteins (79).

#### 4.2. The immune network

Conflicting reports deal with the association between the prognosis of PTC and the degree of lymphocytic infiltration surrounding and/or inside the tumour (80-83). Several studies suggest that the immune response might be important in preventing metastases and recurrence of thyroid cancer, improving disease-free survival (63,84,85). On the contrary, other studies showed that patients with tumour-associated lymphocytes exhibited higher disease stage and increased incidence of invasion and lymph node metastases compared to patients without lymphocytes, or with background thyroiditis (13,86,87).

Moreover, recent studies (88,89) showed important clinical implication of autoimmunity on tumour behaviour; in particular, Stassi et al (89) demonstrated that IL-4 and IL-10 activation induce thyroid cancer cells resistance to chemotherapeutic agents.

Autoimmunity stimulates the production of higher levels of proinflammatory cytokines with growth factor activity (IL-17, IFN- $\gamma$  and TNF- $\alpha$ ) and the angiogenesis enhanced by TNF- $\alpha$  and VEGF (84,90-94). Moreover, high-mobility group box 1 protein (HMGB1), a late inflammatory cytokine that signals danger to the immune system, and nitric oxide can be detected both in thyroiditis and PTC patients, and have been found to promote matrix remodelling, inhibit immune response and suppress cell cycle regulators, thus increasing the risk of PTC proliferation (95). Reactive oxygen species (ROS) also contribute to DNA damage and promote the epithelial-to-mesenchymal transition (62). Two hypotheses may explain the association between autoimmune thyroiditis and differentiated thyroid cancer. In both cases, RET/PTC rearrangement can contribute to modulate the autoimmune response (84,94,95). In fact, RET/

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PTC rearrangement is considered specific for PTC but can also occur in non-neoplastic conditions like Hashimoto's thyroiditis (97). *In vivo* studies showed that RET/PTC rearrangement is more represented in PTC when it is associated with thyroiditis, whereas BRAF<sup>V600E</sup> are more often observed in PTC alone (67,88). According to this hypothesis, free radicals production, cytokine secretion, cellular proliferation as well as other phenomena related to local inflammation could predispose to RET/PTC rearrangement in follicular cells and favour tumorigenesis (98). The second hypothesis is supported by the observation that RET/PTC3 rearrangement expresses high levels of proinflammatory cytokines and proteins involved in the immune response (6,70,73,99). Likewise, there is evidence that RET/PTC1 rearrangement is able to induce the expression of genes involved in inflammation and tumour invasion, including chemokines, chemokine receptors, cytokines, adhesion molecules and matrix-degrading enzymes (69). Other gene alterations have been proposed to explain the association between thyroid cancer and autoimmune thyroiditis. For example, p63 protein is commonly expressed in both PTC and Hashimoto's thyroiditis (100), suggesting p63 expression as a potential link between these conditions (101). Moreover, the increased expression of p-Akt, Akt1, and Akt2 in thyroid cancer and autoimmune thyroiditis suggests PI3K/Akt pathway to be involved in both disorders (102).

Other molecules involved in the immune network relating to DTCs include chemokines, which contribute to the development and progression of cancer (103,104) and tumour metastasis (105,106). Chemokines are a family of about 50 chemotactic proteins (8–10 kDa) classified into four highly conserved groups—CXC, CC, C, and CX3C—based on the position of their first two cysteines adjacent to the amino-terminal region (107,108). These molecules can stimulate cell migration during inflammation, as well as the homeostatic transport of hematopoietic stem cells, lymphocytes, and dendritic cells (109,110). The activity of chemokines is mediated by receptors, which promote the signaling leading to the transcription of genes required for cell motility, invasion, interaction with the extracellular matrix, and cell survival (107,111,112). The main chemokine receptors expressed in thyroid cancer include CXCR4 and CCR7 (113,114). CXCR4 has been studied due to its association with the presence of extranodal extension and thus a more aggressive behavior and negative prognosis (115). CCR7 is also expressed in thyroid carcinoma cell lines (TPC-1) and thyroid cancer tissues (113,114), and is thought to contribute to tissue invasion and cellular proliferation (113). CCR3 is another receptor associated with development, progression, and aggressiveness of several types of cancer, included DTC (116,117).

Finally, recent analysis has focused on immune checkpoints as a prognostic and therapeutic tool for DTCs. In a study assessing immunostaining and mRNA levels of programmed death-ligand 1 (PD-L1), a macrophage-related cell surface glycoprotein regulating local inflammatory responses, more intense expression was observed in samples from DTCs than those from benign tumours, and increasing PD-L1 mRNA expression was demonstrated in more advanced tumour stages (118). Similarly, an increased expression of PD-L1 has been observed in advanced DTCs and ATCs, both at the cellular level and on tumour-associated lymphocytes (119). While these findings await confirmation in Tregs, TAMs, and immature dendritic cells, it appears feasible that studies focusing on immune checkpoint inhibitors in DTCs could lead the way to test new therapeutic strategies (120).

Immunotherapies show promise for providing oncologists with a novel array of therapeutic tools in the near future (121). Cancer immunotherapies have been approved in recent years, including preventive and therapeutic cancer vaccines (122), the first immune checkpoint inhibitors (123,124), a bi-specific T-cell engager, and an oncolytic virus (125). Experience with ipilimumab (CTLA-4 antagonist), nivolumab and pembrolizumab (PD-1 antagonists), and atezolizumab (PD-L1 antagonist) has shown a marked impact on overall survival in cancer patients. Immune checkpoint inhibitors that target the PD-1 pathway generated the greatest interest, with response rates across tumour types that averaged 20-30% (126). Intuitively, the effectiveness of the combination of CTLA-4 and PD-1/PD-L1 blockade on overall survival (OS) will be proved to be higher than single therapy (such as in other cancers) (127). However, the use of immune checkpoint inhibitors in aggressive thyroid cancer has not been extensively investigated yet, and further studies in a large number of patients are warranted.

## 5. INFLAMMATION AND MTC

Data regarding the potential link between MTC and cancer-related inflammation (CRI) originate from few *in vitro* studies. In MTC, the intrinsic pathway, which is driven by genetic alterations associated with thyroid carcinogenesis, seems to play a major role in this link. Germline point mutations of proto-oncogene RET are known to be responsible for almost all familial MTC in multiple endocrine neoplasia (MEN) type 2A and 2B, and familial medullary thyroid carcinoma (FMTc), while somatic point mutations are found in up to 50% of patients with sporadic MTC (128).

RET encodes for the tyrosine kinase receptor of growth factors belonging to the Glial cell-Derived Neurotrophic Factor (GDNF) family, the stimulation

of which activates a variety of signalling pathways, such as the RAS/ERK, the PI3-K/AKT and the MAPK pathways, which are involved in cell survival and differentiation. Gain-of-function mutations of RET cause a constitutive activation of the tyrosine kinase activity of the receptor in the absence of ligands, leading to tumour development and progression (129).

*In vitro* studies (129) demonstrated that GDNF stimulation induced high level of interleukin-8 (IL-8) production in the TT medullary thyroid carcinoma cell lines. IL-8 is a pro-inflammatory, mitogenic and proangiogenic chemokine that is known to be involved directly in tumour growth, cell migration, and angiogenesis in an autocrine or paracrine way, or indirectly by attracting infiltration cells, including neutrophils and macrophages; therefore, its expression in tumour cells may affect their biological properties such as invasion and metastatic ability (16,129). Transcription of IL-8 is known to depend on activation of Nuclear factor interleukin-6 (NF-IL-6) and NF- $\kappa$ B (129,130).

Two reports have associated RET-mediated carcinogenesis with NF- $\kappa$ B activation, so far (76,130). Ludwig *et al.* (131) found that NF- $\kappa$ B was strongly expressed in tissue specimens from parafollicular C-cell carcinomas, and *in vitro* data suggested that NF- $\kappa$ B-dependent transcription plays an essential role in the development of MTC induced by both oncogenic RET isoforms, i.e. those harboring the mutations C634R or M918T, responsible for MEN 2A and MEN 2B, respectively. Gallel *et al.* (76) also demonstrated that the expression of mutated RET induces an increase in NF- $\kappa$ B DNA-binding activity and a consequent increase in pro-inflammatory cytokine secretion.

RET-mediated transformation would be dependent on NF- $\kappa$ B delivered anti-apoptotic and mitogenic signals. Since most part of pro-inflammatory molecules are under NF- $\kappa$ B transcriptional control, it has been hypothesized that NF- $\kappa$ B could be involved in the regulation of pro-inflammatory program of thyroid cells, and this event would contribute to the onset of thyroid cancer (75).

## 6. INFLAMMATION AND ATC

There are studies suggesting that inflammation could also be a key factor involved in the development of ATC, one of the most lethal human malignancies. Many characteristics of the inflammatory status leading to enhanced tumour growth, invasion, angiogenesis, and metastasis, are similar between ATC and PDTC, that is the bridge between DTC and ATC.

Compared to DTC and normal thyroid, ATC and PDTC show an increased amount of TAMs accounting for about more than 50% of immune cells infiltrating

ATC. Infiltrate of TAMs is higher in PDTCs and ATCs than in PTCs and FTCs, and positively correlated with the poor prognosis of PDTCs (17). TAMs usually form a “microglia-like” structure that is in close contact with cancer cells, and their inter-connection and density correlate with invasive features and worse prognosis of the tumour (21,132,133), as confirmed by the evidence that TAMs infiltrate promoted the invasiveness of ATC cell lines *in vitro* through production of CXCL8/IL-8 (21). In contrast to TAMs, infiltration of lymphocytes and dendritic cells, which are involved in antitumour response (7,10), is reduced or absent in ATC (134). In ATC cell lines, CXCL8/IL-8 plays a central role in cell proliferation both during unstimulated conditions and under the effect of pro-inflammatory stimuli, such as IL-1 and TNF- $\alpha$  (21,134). Recruitment of neutrophils within the thyroid gland, a crucial metastasis-promoting factor, is dependent on the amount of CXCL8 produced by the tumour cells when exposed to TNF- $\alpha$  (135,136). Moreover, there is evidence that reduced expression of CXCL8/IL-8 and MCP-1/CCL2 pathways by the oncolytic adenovirus dl922-947 is able to impair angiogenesis and macrophage infiltration, and to promote ATC cell death *in vitro* as well as tumour regression *in vivo* (137). In a recent study on a xenograft mouse model, it has been shown that several cytokines expressed in ATC cell lines and tumour tissues, such as IL-8, TGF- $\alpha$ , and TNF- $\alpha$ , can be down-regulated by suppressing the ubiquitin-like containing PHD and RING finger domains 1 (UHRF1) (138). The protein binds to specific DNA sequences, and recruits a histone deacetylase to regulate gene expression. This UHRF1-mediated effect was found to be associated with inhibited proliferation of ATC, both *in vitro* and *in vivo* (138).

Moreover, inflammatory conditions relating to production of INF- $\gamma$  and TNF- $\alpha$  can promote the autocrine production of IL4, IL10, CXCL1/GRO- $\alpha$ , CXCL10/IP-10 in primary human ATC cells (as well as in DTCs), and potentially contribute to up-regulate anti-apoptotic pathways and chemo-resistance (89,139). Also mast cells are present in ATC, and their infiltrate is directly correlated with tumour invasiveness through production of soluble factors involved in epithelial-to-mesenchymal transition, with CXCL8/IL-8 acting, again, as main effector of this mechanism (133). Furthermore, mast cell-derived CXCL1/GRO- $\alpha$  and CXCL10/IP10 production increased ATC proliferation through the engagement of CXCR2 and CXCR3 expressed on thyroid cells (6). Interestingly, the activation of the CXCL10-CXCR3 axis was also induced by NK cell migration in ATC cell lines. Prostaglandin-E2 was identified as the main responsible for the ATC-mediated NK cell suppression (140).

At odds with results obtained in DTCs, there is no evidence of a connection between the oncogenetic

background of ATCs and inflammation. Likewise, no associations have been found between Hashimoto's thyroiditis or thyroid lymphoma and ATC incidence.

Finally, it is worth mentioning that observational studies in humans have investigated different inflammatory biomarkers as a tool to assess aggressiveness of different thyroid cancers subtypes. The neutrophil-to-lymphocyte ratio, a simple surrogate index of the systemic inflammatory response, was shown to be a prognostic factor in some types of cancers. In a cohort of 3,870 patients affected by benign or malignant thyroid diseases, the neutrophil-to-lymphocyte ratio differed between tumour cancer subtypes and was 3.8.-fold higher in ATC than in PDTC or PTC patients (141). Moreover, also eosinophilia refractory to steroids has been recently reported in an ATC patient (142). Finally, serum levels of IL10 and C-reactive protein in ATC patients were directly correlated with higher peripheral blood myeloid cells (MDSSCs), which are known to be immunosuppressive and cancer promoting (143).

## 7. CONCLUSIONS

The link between inflammation and thyroid cancer involves multiple components of the immune system, ECM, stroma, and AT (5), with pro-tumoural activity of inflammation being opposed to anti-inflammatory effects favoring protection against cancer progression (7,10).

Within the tumour microenvironment, inflammatory cells, belonging both to innate (macrophages) and adaptive (lymphocytes) immune responses, are interconnected with fibroblasts, endothelial cells, adipocytes, and ECM through cytokines, chemokines and adipocytokines (16). Under the influence of transcriptional regulators, such as NF- $\kappa$ B, PI3K-AKT and MAPK, oncogenes connected to the different subtypes of thyroid carcinomas promote their furthestmost proliferative effect on the tumor microenvironment.

As recently reviewed by Antonelly and coworkers, cancer-related inflammation could represent an important target for innovative diagnostic and therapeutic strategies in thyroid cancer (127). The molecular patterns of cytokines and chemokines are key orchestrators and could explain the involvement of the immune system in tumour progression. In fact, anticancer immunotherapy, in particular the immune checkpoint inhibitors, act by promoting lymphocyte activation in order to destroy cancer cells and counteract immune-suppressive signals produced by cancer cells (118). By doing so, they also activate immune memory, leading to a sustained anti-tumour response (118).

Further information on the inflammatory microenvironment may help to explain tumour aggressive behaviour and identify potential new targets of therapy.

## 8. ACKNOWLEDGEMENT

The authors declare that they have no competing interest.

## 9. REFERENCES

1. H. Lim, SS. Devesa, JA. Sosa, D. Check, CM. Kitahara: Trends in thyroid cancer incidence and mortality in the United States, 1974–2013. *JAMA* 317, 1338-1348 (2017) DOI: 10.1001/jama.2017.2719
2. T. Kondo, S. Ezzat, SL. Asa: Pathogenetic mechanisms in thyroid follicular-cell neoplasia. *Nat Rev Cancer* 6, 292–306 (2006) DOI: 10.1038/nrc1836
3. J. Rosai, ML. Carcangiu, RA. De Lellis. Tumors of the thyroid gland. In: Atlas of tumor pathology, Third series, Fascicle 5. Eds: Armed Forces Institute of Pathology, Washington, D.C. (1992)
4. SA. Jr Wells, SL. Asa, H. Dralle, R. Elisei, DB. Evans, RF. Gagel, N. Lee, A. Machens, JF. Moley, F. Pacini, F. Raue, K. Frank-Raue, B. Robinson, MS. Rosenthal, M. Santoro, M. Schlumberger, M. Shah, SG. Waguespack. American Thyroid Association Guidelines Task Force on Medullary Thyroid Carcinoma: Revised American Thyroid Association guidelines for the management of medullary thyroid carcinoma. *Thyroid* 25, 567–610 (2015) DOI: 10.1089/thy.2014.0335
5. V. Guarino, MD. Castellone, E. Avilla, RM. Melillo: Thyroid cancer and inflammation. *Mol Cell Endocrinol* 321, 94–102 (2010) DOI: 10.1016/j.mce.2009.10.003
6. RM. Melillo, V. Guarino, E. Avilla, MR. Galdiero, F. Liotti, N. Prevet, FW. Rossi, F. Basolo, C. Ugolini, A. de Paulis, M. Santoro, G. Marone: Mast cells have a protumorigenic role in human thyroid cancer. *Oncogene* 29, 6203–15 (2010) DOI: 10.1038/onc.2010.348
7. LL. Cunha, MA. Marcello, LS. Ward: The role of the inflammatory microenvironment

- in thyroid carcinogenesis. *Endocr Relat Cancer* 21, R85–R103 (2014)
8. C. Resende de Paiva, C. Grønhøj, U. Feldt-Rasmussen, C. von Buchwald: Association between Hashimoto's Thyroiditis and Thyroid Cancer in 64,628 Patients. *Front Oncol*, 7, 53 (2017)
  9. D. Hanahan, RA. Weinberg: The hallmarks of cancer. *Cell* 100, 57–70 (2000)  
DOI: 10.1016/S0092-8674(00)81683-9
  10. I. Poschke, D. Mougiakakos, R. Kiessling: Camouflage and sabotage: tumor escape from the immune system. *Cancer Immunol Immunother* 60, 1161–1171 (2011)  
DOI: 10.1007/s00262-011-1012-8
  11. J. Modi, A. Patel, R. Terrell, RM. Tuttle, GL. Francis: Papillary thyroid carcinomas from young adults and children contain a mixture of lymphocytes. *J Clin Endocrinol Metab* 88, 4418–25 (2003)  
DOI: 10.1210/jc.2003-030342
  12. Y. Yano, H. Shibuya, W. Kitagawa, M. Nagahama, K. Sugino, K. Ito: Recent outcome of Graves' disease patients with papillary thyroid cancer. *Eur J Endocrinol* 157, 325–9 (2007)  
DOI: 10.1530/EJE-07-0136
  13. JD. French, GR Kotnis, S. Said, CD. Raeburn, RC Jr McIntyre, JP. Kloppner, BR. Haugen: Programmed death-1CT cells and regulatory T cells are enriched in tumor-involved lymph nodes and associated with aggressive features in papillary thyroid cancer. *J Clin Endocrinol Metab* 97, E934–43 (2012)  
DOI: 10.1210/jc.2011-3428
  14. O. Koperek, C. Scheuba, C. Puri, P. Birner, C. Haslinger, W. Rettig, B. Niederle, K. Kaserer, P. Garin Chesa: Molecular characterization of the desmoplastic tumor stroma in medullary thyroid carcinoma. *Int J Oncol* 31, 59–67 (2007)  
DOI: 10.3892/ijo.31.1.59
  15. A. Mantovani, P. Allavena, A. Sica, F. Balkwill: Cancer-related inflammation. *Nature* 454, 436–44 (2008)  
DOI: 10.1038/nature07205
  16. F. Liotti, C. Visciano, RM. Melillo: Inflammation in thyroid oncogenesis. *Am J Cancer Res* 2, 286–97 (2012)
  17. M. Ryder, RA. Ghossein, JC. Ricarte-Filho, JA. Knauf, JA. Fagin: Increased density of tumor-associated macrophages is associated with decreased survival in advanced thyroid cancer. *Endocr Relat Cancer* 15, 1069–74 (2008)  
DOI: 10.1677/ERC-08-0036
  18. R. Kalluri, M. Zeisberg: Fibroblasts in cancer. *Nat Rev Cancer* 6, 392–401 (2006)  
DOI: 10.1038/nrc1877
  19. M. Xing: Molecular pathogenesis and mechanisms of thyroid cancer. *Nat Rev Cancer* 13, 184–99 (2013)  
DOI: 10.1038/nrc3431
  20. R. Noy, JW. Pollard: Tumor-associated macrophages: from mechanisms to therapy. *Immunity* 41, 49–61 (2014)  
DOI: 10.1016/j.immuni.2014.06.010
  21. MR. Galdiero, G. Varricchi, G. Marone: The immune network in thyroid cancer. *Oncoimmunology* 5, e1168556 (2016)
  22. RJ. Arts, TS. Plantinga, S. Tuit, T. Ulas, B. Heinhuis, M. Tesselaar, Y. Sloot, GJ. Adema, LA. Joosten, JW. Smit, MG. Netea, JL. Schultze, RT. Netea-Maier: Transcriptional and metabolic reprogramming induce an inflammatory phenotype in non-medullary thyroid carcinoma-induced macrophages. *Oncoimmunology* 5, e1229725 (2016)
  23. M. Ryder, M. Gild, TM. Hohl, E. Pamer, J. Knauf, R. Ghossein, JA. Joyce, JA. Fagin: Genetic and pharmacological targeting of CSF-1/CSF-1R inhibits tumor-associated macrophages and impairs BRAF-induced thyroid cancer progression. *PLoS One* 8, e54302 (2013)
  24. CB. Steele, CC. Thomas, SJ. Henley, GM. Massetti, DA. Galuska, T. Agurs-Collins, M. Puckett, LC. Richardson: Vital Signs: Trends in Incidence of Cancers Associated with Overweight and Obesity - United States, 2005–2014. *MMWR Morb Mortal Wkly Rep* 66, 1052–1058 (2017)  
DOI: 10.15585/mmwr.mm6639e1
  25. EE. Calle, C. Rodriguez, K. Walker-Thurmond, MJ. Thun: Overweight, obesity, and mortality from cancer in a prospectively studied cohort of U.S. adults. *N Engl J Med* 348, 1625–38 (2003)  
DOI: 10.1056/NEJMoa021423



26. C. Samanic, G. Gridley, WH. Chow, J. Lubin, RN. Hoover, JF Jr Fraumeni: Obesity and cancer risk among white and black United States veterans. *Cancer Causes Control* 15, 35–43 (2004)  
DOI: 10.1023/B:CACO.0000016573.79453.ba
27. A. Engeland, S. Tretli, LA. Akslen, T. Bjorge: Body size and thyroid cancer in two million Norwegian men and women. *Br J Cancer* 95, 366–70 (2006)  
DOI: 10.1038/sj.bjc.6603249
28. L. Dal Maso, C. La Vecchia, S. Franceschi, S. Preston-Martin, E. Ron, F. Levi, W. Mack, SD. Mark, A. McTiernan, L. Kolonel, K. Mabuchi, F. Jin, G. Wingren, MR. Galanti, A. Hallquist, E. Glattre, E. Lund, D. Linos, E. Negri: A pooled analysis of thyroid cancer studies. V. Anthropometric factors. *Cancer Causes Control* 11, 137–44 (2000)  
DOI: 10.1023/A:1008938520101
29. AG. Renehan, M. Tyson, M. Egger, RF. Heller, M. Zwahlen: Body-mass index and incidence of cancer: a systematic review and meta-analysis of prospective observational studies. *Lancet* 371, 569–578 (2008)  
DOI: 10.1016/S0140-6736(08)60269-X
30. CM. Kitahara, EA. Platz, LE. Freeman, AW. Hsing, MS. Linet, Y. Park, C. Schairer, A. Schatzkin, JM. Shikany, A. Berrington de González: Obesity and thyroid cancer risk among U.S. men and women: a pooled analysis of five prospective studies. *Cancer Epidemiol Biomarkers Prev* 20, 464–72 (2011)  
DOI: 10.1158/1055-9965.EPI-10-1220
31. MA. Marcello, P. Malandrino, JF. Almeida, MB. Martins, LL. Cunha, NE. Bufalo, G. Pellegriti, LS. Ward: The influence of the environment on the development of thyroid tumors: a new appraisal. *Endocr Relat Cancer* 21, T235–54 (2014)
32. MI. Ilie, S. Lassalle, E. Long-Mira, V. Hofman, J. Zangari, G. Bénaim, A. Bozec, N. Guevara, J. Haudebourg, I. Birtwisle-Peyrottes, J. Santini, P. Brest, P. Hofman: In papillary thyroid carcinoma, TIMP-1 expression correlates with BRAF (V600E) mutation status and together with hypoxia-related proteins predicts aggressive behavior. *Virchows Arch* 463, 437–44 (2013)  
DOI: 10.1007/s00428-013-1453-x
33. F. Santini, P. Marzullo, M. Rotondi, G. Ceccarini, L. Pagano, S. Ippolito, L. Chiovato, B. Biondi: Mechanisms in endocrinology: the crosstalk between thyroid gland and adipose tissue: signal integration in health and disease. *Eur J Endocrinol* 171, R137–52 (2014)
34. G. Frühbeck: Intracellular signalling pathways activated by leptin. *Biochem J* 393, 7–20 (2006)  
DOI: 10.1042/BJ20051578
35. A. Schäffler, J. Schölmerich: Innate immunity and adipose tissue biology. *Trends Immunol* 31, 228–35 (2010)  
DOI: 10.1016/j.it.2010.03.001
36. B. Cousin, O. Munoz, M. Andre, AM. Fontanilles, C. Dani, JL. Cousin, P. Laharrague, L. Casteilla, L. Penicaud: A role for preadipocytes as macrophage-like cells. *FASEB J* 13, 305–312 (1999)  
DOI: 10.1096/fasebj.13.2.305
37. G. Charriere, B. Cousin, E. Arnaud, M. Andre, F. Bacou, L. Penicaud, L. Casteilla: Preadipocyte conversion to macrophage. Evidence of plasticity. *J Biol Chem* 278, 9850–9855 (2003)  
DOI: 10.1074/jbc.M210811200
38. S. Nishimura, I. Manabe, M. Nagasaki, K. Eto, H. Yamashita, M. Ohsugi, M. Otsu, K. Hara, K. Ueki, S. Sugiura, K. Yoshimura, T. Kadowaki, R. Nagai: CD8+ effector T cells contribute to macrophage recruitment and adipose tissue inflammation in obesity. *Nat Med* 15, 914–20 (2009)  
DOI: 10.1038/nm.1964
39. U. Kintscher, M. Hartge, K. Hess, A. Foryst-Ludwig, M. Clemenz, M. Wabitsch, P. Fischer-Posovszky, TF. Barth, D. Dragun, T. Skurk, H. Hauner, M. Blüher, T. Unger, AM. Wolf, U. Knippschild, V. Hombach, N. Marx: T-lymphocyte infiltration in visceral adipose tissue: a primary event in adipose tissue inflammation and the development of obesity-mediated insulin resistance. *Arterioscler Thromb Vasc Biol* 28, 1304–10 (2008)  
DOI: 10.1161/ATVBAHA.108.165100
40. J. Haase, U. Weyer, K. Immig, N. Klötting, M. Blüher, J. Eilers, I. Bechmann, M. Gericke: Local proliferation of macrophages in adipose tissue during obesity-induced

- inflammation. *Diabetologia* 57, 562–71 (2014)  
DOI: 10.1007/s00125-013-3139-y
41. AA. Hill, W. Reid Bolus, AH. Hasty: A decade of progress in adipose tissue macrophage biology. *Immunol Rev* 262, 134–52 (2014)  
DOI: 10.1111/imr.12216
  42. S. Cinti, G. Mitchell, G. Barbatelli, I. Murano, E. Ceresi, E. Faloia, S. Wang, M. Fortier, AS. Greenberg, MS. Obin: Adipocyte death defines macrophage localization and function in adipose tissue of obese mice and humans. *J Lipid Res* 46, 2347–55 (2005)  
DOI: 10.1194/jlr.M500294-JLR200
  43. SP. Weisberg, D. McCann, M. Desai, M. Rosenbaum, RL. Leibel, AW. Jr Ferrante: Obesity is associated with macrophage accumulation in adipose tissue. *J Clin Invest* 112, 1796–1808 (2003)  
DOI: 10.1172/JCI200319246
  44. H. Xu, GT. Barnes, Q. Yang, G. Tan, D. Yang, CJ. Chou, J. Sole, A. Nichols, JS. Ross, LA. Tartaglia, H. Chen: Chronic inflammation in fat plays a crucial role in the development of obesity related insulin resistance. *J Clin Invest* 112, 1821–1830 (2003)  
DOI: 10.1172/JCI200319451
  45. S. Fujisaka, I. Usui, M. Icutani, A. Aminuddin, A. Takikawa, K. Tsuneyama, A. Mahmood, N. Goda, Y. Nagai, K. Takatsu, K. Tobe: Adipose tissue hypoxia induces inflammatory M1 polarity of macrophages in an HIF-1 $\alpha$ -dependent and HIF-1 $\alpha$ -independent manner in obese mice. *Diabetologia* 56, 1403–12 (2013)  
DOI: 10.1007/s00125-013-2885-1
  46. XP. Pang, NS. Ross, JM. Hershman: Alterations in TNF-alpha signal transduction in resistant human papillary thyroid carcinoma cells. *Thyroid* 6, 313–7 (1996)  
DOI: 10.1089/thy.1996.6.313
  47. F. Coperchini, P. Pignatti, A. Carbone, R. Bongianino, CA. Di Buduo, P. Loporati, L. Croce, F. Magri, A. Balduini, L. Chiovato, M. Rotondi: TNF- $\alpha$  increases the membrane expression of the chemokine receptor CCR6 in thyroid tumor cells, but not in normal thyrocytes: potential role in the metastatic spread of thyroid cancer. *Tumour Biol* 37, 5569–75 (2016)  
DOI: 10.1007/s13277-015-4418-7
  48. F. Lumachi, SM. Basso, R. Orlando: Cytokines, thyroid diseases and thyroid cancer. *Cytokine* 50, 229–33 (2010)  
DOI: 10.1016/j.cyto.2010.03.005
  49. JP. Couto, L. Daly, A. Almeida, JA. Knauf, JA. Fagin, M. Sobrinho-Simões, J. Lima, V. Máximo, P. Soares, D. Lyden, JF. Bromberg: STAT3 negatively regulates thyroid tumorigenesis. *Proc Natl Acad Sci* 109, E2361–70 (2012)  
DOI: 10.1073/pnas.1201232109
  50. JW. Chang, KY. Yeh, YC. Shen, JJ. Hsieh, CK. Chuang, SK. Liao, LH. Tsai, CH. Wang: Production of multiple cytokines and induction of cachexia in athymic nude mice by a new anaplastic thyroid carcinoma cell line. *J Endocrinol* 179, 387–94 (2003)  
DOI: 10.1677/joe.0.1790387
  51. TP. Kobawala, TI. Trivedi, KK. Gajjar, DH. Patel, GH. Patel, NR. Ghosh: Significance of Interleukin-6 in Papillary Thyroid Carcinoma. *J Thyroid Res* 2016, 6178921 (2016)
  52. M. Dalamaga, KN. Diakopoulos, CS. Mantzoros: The role of adiponectin in cancer: a review of current evidence. *Endocr Rev* 33, 547–594 (2012)  
DOI: 10.1210/er.2011-1015
  53. SP. Cheng, PH. Yin, YC. Chang, CH. Lee, SY. Huang, CW. Chi: Differential roles of leptin in regulating cell migration in thyroid cancer cells. *Oncol Rep* 23, 1721–7 (2010)
  54. S. Uddin, P. Bavi, AK. Siraj, M. Ahmed, M. Al-Rasheed, AR. Hussain, M. Ahmed, T. Amin, A. Alzahrani, F. Al-Dayel, J. Abubaker, R. Bu, KS. Al-Kuraya: Leptin-R and its association with PI3K/AKT signaling pathway in papillary thyroid carcinoma. *Endocr Relat Cancer* 17, 191–202 (2010)  
DOI: 10.1677/ERC-09-0153
  55. WG. Kim, JW. Park, MC. Willingham, SY. Cheng: Diet-induced obesity increases tumor growth and promotes anaplastic change in thyroid cancer in a mouse model. *Endocrinology* 154, 2936–47 (2013)  
DOI: 10.1210/en.2013-1128
  56. YL. Fan, XQ. Li: Expression of leptin and its receptor in thyroid carcinoma: distinctive prognostic significance in different subtypes. *Clin Endocrinol (Oxf)* 83, 261–7 (2015)  
DOI: 10.1111/cen.12598

57. N. Mitsiades, K. Pazaitou-Panayiotou, KN. Aronis, HS. Moon, JP. Chamberland, X. Liu, KN. Diakopoulos, V. Kyttaris, V. Panagiotou, G. Mylvaganam, S. Tseleni-Balafouta, CS. Mantzoros: Circulating adiponectin is inversely associated with risk of thyroid cancer: *in vivo* and *in vitro* studies. *J Clin Endocrinol Metab* 96, E2023–8 (2011)  
DOI: 10.1210/jc.2010-1908
58. SP. Cheng, CL. Liu, YC. Hsu, YC. Chang, SY. Huang, JJ. Lee: Expression and biologic significance of adiponectin receptors in papillary thyroid carcinoma. *Cell Biochem Biophys* 65, 203–10 (2013)  
DOI: 10.1007/s12013-012-9419-1
59. M. Egeblad, Z. Werb: New functions for the matrix metalloproteinases in cancer progression. *Nat Rev Cancer* 2, 161–74 (2002)  
DOI: 10.1038/nrc745
60. I. Marecko, D. Cvejic, S. Selemetjev, S. Paskas, S. Tatic, I. Paunovic, S. Savin: Enhanced activation of matrix metalloproteinase-9 correlates with the degree of papillary thyroid carcinoma infiltration. *Croat Med J* 55, 128–37 (2014)  
DOI: 10.3325/cmj.2014.55.128
61. JR. Wang, XH. Li, XJ. Gao, SC. An, H. Liu, J. Liang, K. Zhang, Z. Liu, J. Wang, Z. Chen, W. Sun: Expression of MMP-13 is associated with invasion and metastasis of papillary thyroid carcinoma. *Eur Rev Med Pharmacol Sci* 17, 427–35 (2013)
62. N. Wang, R. Jiang, JY. Yang, C. Tang, L. Yang, M. Xu, QF. Jiang, ZM. Liu: Expression of TGF- $\beta$ 1, SNAI1 and MMP-9 is associated with lymph node metastasis in papillary thyroid carcinoma. *J Mol Histol* 45, 391–9 (2014)  
DOI: 10.1007/s10735-013-9557-9
63. LL. Cunha, RC. Ferreira, MA. Marcello, J. Vassallo, LS. Ward: Clinical and pathological implications of concurrent autoimmune thyroid disorders and papillary thyroid cancer. *J Thyroid Res* 2011, 387062 (2011)
64. M. Takahashi: The GDNF/RET signaling pathway and human diseases. *Cytokine Growth Factor Rev* 12, 361–373 (2001)  
DOI: 10.1016/S1359-6101(01)00012-0
65. ET. Kimura, MN. Nikiforova, Z. Zhu, JA. Knauf, YE. Nikiforov, JA. Fagin: High prevalence of BRAF mutations in thyroid cancer: genetic evidence for constitutive activation of the RET/PTC–RAS–BRAF signaling pathway in papillary thyroid carcinoma. *Cancer Res* 63, 1454–7 (2003)
66. RM. Melillo, MD. Castellone, V. Guarino, V. De Falco, AM. Cirafici, G. Salvatore, F. Caiazzo, F. Basolo, R. Giannini, M. Kruhoffer, T. Orntoft, A. Fusco, M. Santoro: The RET/PTC–RAS–BRAF linear signaling cascade mediates the motile and mitogenic phenotype of thyroid cancer cells. *J Clin Invest* 115, 1068–81 (2005)  
DOI: 10.1172/JCI200522758
67. DJ. Jr. Powell, J. Russell, K. Nibu, G. Li, E. Rhee, M. Liao, M. Goldstein, WM. Keane, M. Santoro, A. Fusco, JL. Rothstein: The RET/PTC3 oncogene: metastatic solid-type papillary carcinomas in murine thyroids. *Cancer Res* 58, 5523–8 (1998)
68. MD. Castellone, V. Guarino, V. De Falco, F. Carlomagno, F. Basolo, P. Faviana, M. Kruhoffer, T. Orntoft, JP. Russell, JL. Rothstein, A. Fusco, M. Santoro, RM. Melillo: Functional expression of the CXCR4 chemokine receptor is induced by RET/PTC oncogenes and is a common event in human papillary thyroid carcinomas. *Oncogene* 23, 5958–67 (2004)  
DOI: 10.1038/sj.onc.1207790
69. MG. Borrello, L. Alberti, A. Fischer, D. Degl'innocenti, C. Ferrario, M. Gariboldi, F. Marchesi, P. Allavena, A. Greco, P. Collini, S. Pilotti, G. Cassinelli, P. Bressan, L. Fugazzola, A. Mantovani, MA. Pierotti: Induction of a proinflammatory program in normal human thyrocytes by the RET/PTC1 oncogene. *Proc Natl Acad Sci USA* 102, 14825–30 (2005)  
DOI: 10.1073/pnas.0503039102
70. E. Puxeddu, JA. Knauf, MA. Sartor, N. Mitsutake, EP. Smith, M. Medvedovic, CR. Tomlinson, S. Moretti, JA. Fagin: RET/PTC-induced gene expression in thyroid PCCL3 cells reveals early activation of genes involved in regulation of the immune response. *Endocr Relat Cancer* 12, 319–334 (2005)  
DOI: 10.1677/erc.1.00947
71. JP. Russell, JB. Engiles, JL. Rothstein: Proinflammatory mediators and genetic background in oncogene mediated tumor progression. *J Immunol* 172, 4059–67 (2004)  
DOI: 10.4049/jimmunol.172.7.4059

72. JS. Pufnock, JL. Rothstein JL: Oncoprotein signaling mediates tumor-specific inflammation and enhances tumor progression. *J Immunol* 182, 5498–506 (2009)  
DOI: 10.4049/jimmunol.0801284
73. JP. Russell, S. Shinohara, RM. Melillo, MD. Castellone, M. Santoro, JL. Rothstein: Tyrosine kinase oncoprotein, RET/PTC3, induces the secretion of myeloid growth and chemotactic factors. *Oncogene* 22, 4569–77 (2003)  
DOI: 10.1038/sj.onc.1206759
74. M. Karin: Nuclear factor-kappaB in cancer development and progression. *Nature* 441, 431–436 (2006)  
DOI: 10.1038/nature04870
75. F. Pacifico, A. Leonardi: Role of NF-kappaB in thyroid cancer. *Mol Cell Endocrinol* 321, 29–35 (2010)  
DOI: 10.1016/j.mce.2009.10.010
76. P. Gallel, J. Pallares, X. Dolcet, D. Llobet, N. Eritja, M. Santacana, A. Yeramian, V. Palomar-Asenjo, H. Lagarda, D. Mauricio, M. Encinas, X. Matias-Guiu: Nuclear factor-kappaB activation is associated with somatic and germ line RET mutations in medullary thyroid carcinoma. *Hum Pathol* 39, 994–1001 (2008)  
DOI: 10.1016/j.humpath.2007.11.015
77. A. Bommarito, P. Richiusa, E. Carissimi, G. Pizzolanti, V. Rodolico, G. Zito, A. Criscimanna, F. Di Blasi, M. Pitrone, M. Zerilli, MC. Amato, G. Spinelli, V. Carina, G. Modica, MA. Latteri, A. Galluzzo, C. Giordano: BRAFV600E mutation, TIMP-1 upregulation, and NF-kappaB activation: closing the loop on the papillary thyroid cancer trilogy. *Endocr Relat Cancer* 18, 669–85 (2011)  
DOI: 10.1530/ERC-11-0076
78. X. Li, AB. Abdel-Mageed, D. Mondal, E. Kandil: The nuclear factor kappa-B signaling pathway as a therapeutic target against thyroid cancers. *Thyroid* 23, 209–18 (2012)  
DOI: 10.1089/thy.2012.0237
79. M. Faria, P. Matos, T. Pereira, R. Cabrera, BA. Cardoso, MJ. Bugalho, AL. Silva: RAC1b overexpression stimulates proliferation and NF-kB-mediated anti-apoptotic signaling in thyroid cancer cells. *PLoS One* 12, e0172689 (2017)
80. RN. Hirabayashi, S. Lindsay: The relation of thyroid carcinoma and chronic thyroiditis. *Surg Gynecol Obstet* 121, 243–52 (1965)
81. OH. Clark, FS. Greenspan, JE. Dunphy: Hashimoto's thyroiditis and thyroid cancer: indications for operation. *Am J Surg* 140, 65–71 (1980)  
DOI: 10.1016/0002-9610(80)90419-5
82. J. Aguayo, Y. Sakatsume, C. Jamieson, VV. Row, R. Volpè: Nontoxic nodular goiter and papillary thyroid carcinoma are not associated with peripheral blood lymphocyte sensitization to thyroid cells. *J Clin Endocrinol Metab* 68, 145–9 (1989)  
DOI: 10.1210/jcem-68-1-145
83. S. Matsubayashi, K. Kawai, Y. Matsumoto, T. Mukuta, T. Morita, K. Hirai, F. Matsuzuka, K. Kakudoh, K. Kuma, H. Tamai: The correlation between papillary thyroid carcinoma and lymphocytic infiltration in the thyroid gland. *J Clin Endocrinol Metab* 80, 3421–4 (1995)
84. M. Ehlers, M. Schott: Hashimoto's thyroiditis and papillary thyroid cancer: are they immunologically linked? *Trends Endocrinol Metab* 25, 656–64 (2014)
85. A. Ieni, R. Vita, E. Magliolo, M. Santarpia, F. Di Bari, S. Benvenga, G. Tuccari: One-third of an Archival Series of Papillary Thyroid Cancer (Years 2007–2015) Has Coexistent Chronic Lymphocytic Thyroiditis, Which Is Associated with a More Favorable Tumor-Node-Metastasis Staging. *Front Endocrinol (Lausanne)* 1, 8:337 (2017)
86. W. Qing, WY. Fang, L. Ye, LY. Shen, XF. Zhang, XC. Fei, X. Chen, WQ. Wang, XY. Li, JC. Xiao, G. Ning: Density of tumor-associated macrophages correlates with lymph node metastasis in papillary thyroid carcinoma. *Thyroid* 22, 905–10 (2012)  
DOI: 10.1089/thy.2011.0452
87. H. Yu, X. Huang, X. Liu, H. Jin, G. Zhang, Q. Zhang, J. Yu: Regulatory T cells and plasmacytoid dendritic cells contribute to the immune escape of papillary thyroid cancer coexisting with multinodular non-toxic goiter. *Endocrine* 44, 172–81 (2013)  
DOI: 10.1007/s12020-012-9853-2
88. M. Muzza, D. Degl'Innocenti, C. Colombo, M. Perrino, E. Ravasi, S. Rossi, V. Cirello, P. Beck-Peccoz, MG. Borrello, L. Fugazzola: The tight relationship between

- papillary thyroid cancer, autoimmunity and inflammation: clinical and molecular studies. *Clin Endocrinol (Oxf)* 72, 702–8 (2010)  
DOI: 10.1111/j.1365-2265.2009.03699.x
89. G. Stassi, M. Todaro, M. Zerilli, L. Ricci-Vitiani, D. Di Liberto, M. Patti, A. Florena, F. Di Gaudio, G. Di Gesù, R. De Maria: Thyroid cancer resistance to chemotherapeutic drugs via autocrine production of interleukin-4 and interleukin-10. *Cancer Res* 63, 6784–90 (2003)
  90. LF. Fajardo, HH. Kwan, J. Kowalski, SD. Prionas, AC. Allison: Dual role of tumor necrosis factor-alpha in angiogenesis. *Am J Pathol* 140, 539–44 (1992)
  91. B. Ji, Y. Liu, P. Zhang, Y. Wang, G. Wang: COX-2 expression and tumor angiogenesis in thyroid carcinoma patients among northeast Chinese population-result of a single-center study. *Int J Med Sci* 9, 237–42 (2012)  
DOI: 10.7150/ijms.4173
  92. S. Mardente, A. Zicari, F. Consorti, E. Mari, M. Di Vito, M. Leopizzi, C. Della Rocca, A. Antonaci: Cross-talk between NO and HMGB1 in lymphocytic thyroiditis and papillary thyroid cancer. *Oncol Rep* 24, 1455–61 (2010)  
DOI: 10.3892/or\_00001005
  93. G. Landskron, M. De la Fuente, P. Thuwajit, C. Thuwajit, MA. Hermoso: Chronic inflammation and cytokines in the tumor microenvironment. *J Immunol Res* 2014, 149185 (2014)
  94. F. Felicetti, MG. Catalano, N. Fortunati: Thyroid Autoimmunity and Cancer. *Front Horm Res* 48, 97–109 (2017)  
DOI: 10.1159/000452909
  95. S. Mardente, E. Mari, F. Consorti, C. Di Gioia, R. Negri, M. Etna, A. Zicari, A. Antonaci: HMGB1 induces the overexpression of miR-222 and miR-221 and increases growth and motility in papillary thyroid cancer cells. *Oncol Rep* 28, 2285–9 (2012)  
DOI: 10.3892/or.2012.2058
  96. MG. Borrello, D. Degl'Innocenti, MA. Pierotti: Inflammation and cancer: the oncogene-driven connection. *Cancer Lett* 267, 262–70 (2008)  
DOI: 10.1016/j.canlet.2008.03.060
  97. A. Wirtschafter, R. Schmidt, D. Rosen, N. Kundu, M. Santoro, A. Fusco, H. Mulhaupt, JP. Atkins, MR. Rosen, WM. Keane, JL. Rothstein: Expression of the RET/PTC fusion gene as a marker for papillary carcinoma in Hashimoto's thyroiditis. *Laryngoscope* 107, 95–100 (1997)  
DOI: 10.1097/00005537-199701000-00019
  98. M. Gandhi, M. Medvedovic, JR. Stringer, YE. Nikiforov: Interphase chromosome folding determines spatial proximity of genes participating in carcinogenic RET/PTC rearrangements. *Oncogene* 25, 2360–6 (2006)  
DOI: 10.1038/sj.onc.1209268
  99. KJ. Rhoden, K. Unger, G. Salvatore, Y. Yilmaz, V. Vovk, G. Chiappetta, MB. Qumsiyeh, JL. Rothstein, A. Fusco, M. Santoro, H. Zitzelsberger, G. Tallini G: RET/papillary thyroid cancer rearrangement in nonneoplastic thyrocytes: follicular cells of Hashimoto's thyroiditis share low-level recombination events with a subset of papillary carcinoma. *J Clin Endocrinol Metab* 91, 2414–23 (2006)  
DOI: 10.1210/jc.2006-0240
  100. P. Unger, M. Ewart, BY. Wang, L. Gan, DS. Kohtz, DE. Burstein: Expression of p63 in papillary thyroid carcinoma and in Hashimoto's thyroiditis: a pathobiologic link? *Hum Pathol* 34, 764–9 (2003)
  101. G. Pellegrini, E. Dellambra, O. Golisano, E. Martinelli, I. Fantozzi, S. Bondanza, D. Ponzin, F. McKeon, M. De Luca: p63 identifies keratinocyte stem cells. *Proc Natl Acad Sci USA* 98, 3156–61 (2001)  
DOI: 10.1073/pnas.061032098
  102. SD. Larson, LN. Jackson, TS. Riall, T. Uchida, RP. Thomas, S. Qiu, BM. Evers: Increased incidence of well-differentiated thyroid cancer associated with Hashimoto thyroiditis and the role of the PI3k/Akt pathway. *J Am Coll Surg* 204, 764–73; discussion 773–5 (2007)
  103. M. Baggiolini: Chemokines in pathology and medicine. *J Intern Med* 250, 91–104 (2001)  
DOI: 10.1046/j.1365-2796.2001.00867.x
  104. MJ. Frederick, GL. Clayman: Chemokines in cancer. *Expert Rev Mol Med* 3, 1–18 (2001)  
DOI: 10.1017/S1462399401003301

105. YX. Sun, J. Wang, CE. Shelburne, DE. Lopatin, AM. Chinnaiyan, MA. Rubin, KJ. Pienta, RS. Taichman: Expression of CXCR4 and CXCL12 (SDF-1) in human prostate cancers (PCa) *in vivo*. *J Cell Biochem* 89, 462–473 (2003)  
DOI: 10.1002/jcb.10522
106. M. Kato, J. Kitayama, S. Kazama, H. Nagawa: Expression pattern of CXC chemokine receptor-4 is correlated with lymph node metastasis in human invasive ductal carcinoma. *Breast Cancer Res* 5, R144–R150 (2003)
107. D. Raman, PJ. Baugher, YM. Thu, A. Richmond: Role of chemokines in tumor growth. *Cancer Lett* 256, 137–165 (2007)  
DOI: 10.1016/j.canlet.2007.05.013
108. F. Balkwill: Cancer and the chemokine network. *Nat Rev Cancer* 4, 540–550 (2004)  
DOI: 10.1038/nrc1388
109. K. Beider, M. Abraham, A. Peled: Chemokines and chemokine receptors in stem cell circulation. *Front Biosci* 13, 6820–6833 (2008)  
DOI: 10.2741/3190
110. D. Rossi, A. Zlotnik: The biology of chemokines and their receptors. *Annu Rev Immunol* 18, 217–242 (2000)  
DOI: 10.1146/annurev.immunol.18.1.217
111. S.J. Allen, SE. Crown, TM. Handel: Chemokine: receptor structure, interactions, and antagonism. *Annu Rev Immunol* 25, 787–820 (2007)  
DOI: 10.1146/annurev.immunol.24.021605.090529
112. NF. Neel, E. Schutyser, J. Sai, GH. Fan, A. Richmond: Chemokine receptor internalization and intracellular trafficking. *Cytokine Growth Factor Rev* 16, 637–658 (2005)  
DOI: 10.1016/j.cytogfr.2005.05.008
113. M. Sancho, J. Vieira, C. Casalou, M. Mesquita, T. Pereira, BM. Cavaco, S. Dias, V. Leite: Expression and function of the chemokine receptor CCR7 in thyroid carcinomas. *J Endocrinol* 191, 229–238 (2006)
114. PL. Wagner, TA. Moo, N. Arora, YF. Liu, R. Zarnegar, T. Scognamiglio, TJ. Fahey: The chemokine receptors CXCR4 and CCR7 are associated with tumor size and pathologic indicators of tumor aggressiveness in papillary thyroid carcinoma. *Ann Surg Oncol* 15, 2833–2841 (2008)  
DOI: 10.1245/s10434-008-0064-2
115. H. Yamashita, S. Noguchi, N. Murakami, M. Toda, S. Uchino, S. Watanabe, H. Kawamoto: Extracapsular invasion of lymph node metastasis. A good indicator of disease recurrence and poor prognosis in patients with thyroid microcarcinoma. *Cancer* 86, 842–849 (1999)  
DOI: 10.1002/(SICI)1097-0142(19990901)86:5<842::AID-CNCR21>3.0.CO;2-X
116. K. Johrer, C. Zelle-Rieser, A. Perathoner, P. Moser, M. Hager, R. Ramoner, H. Gander, L. Hottl, G. Bartsch, R. Greil, M. Thurnher: Up-regulation of functional chemokine receptor CCR3 in human renal cell carcinoma. *Clin Cancer Res* 11:2459–2465 (2005)
117. HE. González, A. Leiva, H. Tobar, K. Böhmwald, G. Tapia, J. Torres, LM. Mosso, SM. Bueno, P. Gonzalez, AM. Kalergis, CA. Riedel: Altered chemokine receptor expression in papillary thyroid cancer. *Thyroid* 19, 957–65.
118. LL. Cunha, MA. Marcello, EC. Morari, S. Nonogaki, FF. Conte, R. Gerhard, FA. Soares, J. Vassallo, LS. Ward: Differentiated thyroid carcinomas may elude the immune system by B7H1 upregulation. *Endocr Relat Cancer* 20, 103–10 (2013)  
DOI: 10.1530/ERC-12-0313
119. JJ. Bastman, HS. Serracino, Y. Zhu, MR. Koenig, V. Mateescu, SB. Sams, KD. Davies, CD. Raeburn, RC. Jr. McIntyre, BR. Haugen, JD. French: Tumor-Infiltrating T Cells and the PD-1 Checkpoint Pathway in Advanced Differentiated and Anaplastic Thyroid Cancer. *J Clin Endocrinol Metab* 101, 2863–73 (2016)  
DOI: 10.1210/jc.2015-4227
120. LL. Cunha, MA. Marcello, V. Rocha-Santos, LS. Ward: Immunotherapy against endocrine malignancies: immune checkpoint inhibitors lead the way. *Endocr Relat Cancer* 24, T261–T281 (2017)
121. J. Couzin-Frankel: Breakthrough of the year 2013. Cancer immunotherapy. *Science* 342, 1432–3 (2013)  
DOI: 10.1126/science.342.6165.1432
122. LA. Emens, PA. Ascierto, PK. Darcy, S. Demaria, AMM. Eggermont, WL. Redmond, B. Seliger, FM. Marincola:

- Cancer immunotherapy: Opportunities and challenges in the rapidly evolving clinical landscape. *Eur J Cancer* 81, 116–29 (2017)  
DOI: 10.1016/j.ejca.2017.01.035
123. C. Robert, A. Ribas, JD. Wolchok, FS. Hodi, O. Hamid, R. Kefford, JS. Weber, AM. Joshua, WJ. Hwu, TC. Gangadhar, A. Patnaik, R. Dronca, H. Zarour, RW. Joseph, P. Boasberg, B. Chmielowski, C. Mateus, MA. Postow, K. Gergich, J. Ellassaiss-Schaap, XN. Li, R. Iannone, SW. Ebbinghaus, SP. Kang, A. Daud: Anti-programmed death-receptor-1 treatment with pembrolizumab in ipilimumab-refractory advanced melanoma: a randomised dose-comparison cohort of a phase 1 trial. *Lancet* 384, 1109–17 (2014)  
DOI: 10.1016/S0140-6736(14)60958-2
124. SL. Topalian, M. Sznol, DF. McDermott, HM. Kluger, RD. Carvajal, WH. Sharfman, JR. Brahmer, DP. Lawrence, MB. Atkins, JD. Powderly, PD. Leming, EJ. Lipson, I. Puzanov, DC. Smith, JM. Taube, JM. Wigginton, GD. Kollia, A. Gupta, DM. Pardoll, JA. Sosman, FS. Hodi: Survival, durable tumor remission, and long-term safety in patients with advanced melanoma receiving nivolumab. *J Clin Oncol* 32, 1020–30 (2014)  
DOI: 10.1200/JCO.2013.53.0105
125. RH. Andtbacka, HL. Kaufman, F. Collichio, T. Amatruda, N. Senzer, J. Chesney, KA. Delman, LE. Spitler, I. Puzanov I, SS. Agarwala, M. Milhem, L. Cranmer, B. Curti, K. Lewis, M. Ross, T. Guthrie, GP. Linette, GA. Daniels, K. Harrington, MR. Middleton, WH. Miller, JS. Zager, Y. Ye, B. Yao, A. Li, S. Doleman, A. VanderWalde, J. Gansert, RS. Coffin: Talimogene Laherparepvec Improves Durable Response Rate in Patients With Advanced Melanoma. *J Clin Oncol* 33, 2780–8 (2015)  
DOI: 10.1200/JCO.2014.58.3377
126. EJ. Lipson, PM. Forde, HJ. Hammers, LA. Emens, JM. Taube, SL. Topalian: Antagonists of PD-1 and PD-L1 in Cancer Treatment. *Semin Oncol* 42, 587–600 (2015)  
DOI: 10.1053/j.seminoncol.2015.05.013
127. A. Antonelli, SM. Ferrari, P. Fallahi: Current and future immunotherapies for thyroid cancer. *Expert Rev Anticancer Ther* 2018, 18, 149–159.
128. BR. Haugen, EK. Alexander, KC. Bible, GM. Doherty, SJ. Mandel, YE. Nikiforov, F. Pacini, GW. Randolph, AM. Sawka, M. Schlumberger, KG. Schuff, SI. Sherman, JA. Sosa, DL. Steward, RM. Tuttle, L. Wartofsky: 2015 American Thyroid Association Management Guidelines for Adult Patients with Thyroid Nodules and Differentiated Thyroid Cancer: The American Thyroid Association Guidelines Task Force on Thyroid Nodules and Differentiated Thyroid Cancer. *Thyroid* 26, 1–133 (2016)
129. N. Iwahashi, H. Murakami, Y. Nimura, M. Takahashi: Activation of RET tyrosine kinase regulates interleukin-8 production by multiple signaling pathways. *Biochem Biophys Res Commun* 294, 642–9 (2002)  
DOI: 10.1016/S0006-291X(02)00528-4
130. N. Mukaida, S. Okamoto, Y. Ishikawa, K. Matsushima: Molecular mechanism of interleukin-8 gene expression. *J Leukoc Biol* 56, 554–8 (1994)  
DOI: 10.1002/jlb.56.5.554
131. L. Ludwig, H. Kessler, M. Wagner, C. Hoang-Vu, H. Dralle, G. Adler, B. Bohm, RM. Schmid: Nuclear Factor -kB is constitutively active in C-Cell carcinoma and required for RET-induced transformation. *Cancer Res* 61, 4526–35 (2001)
132. B. Caillou, M. Talbot, U. Weyemi, C. Pioche-Durieu, A. Al Ghuzlan, JM. Bidart, S. Chouaib, M. Schlumberger, C. Dupuy: Tumor-associated macrophages (TAMs) form an interconnected cellular supportive network in anaplastic thyroid carcinoma. *PLoS One* 6, e22567 (2011)
133. C. Visciano, N. Prevete, F. Liotti, G. Marone: Tumor-Associated Mast Cells in Thyroid Cancer. *Int J Endocrinol* 2015, 705169 (2015)
134. C. Ugolini, F. Basolo, A. Proietti, P. Vitti, R. Elisei, P. Miccoli, A. Toniolo: Lymphocyte and immature dendritic cell infiltrates in differentiated, poorly differentiated, and undifferentiated thyroid carcinoma. *Thyroid* 17, 389–93 (2007)  
DOI: 10.1089/thy.2006.0306
135. M. Rotondi, F. Coperchini, P. Pignatti, F. Magri, L. Chiovato: Metformin reverses the secretion of CXCL8 induced by TNF- $\alpha$  in primary cultures of human thyroid cells: an additional indirect anti-tumor effect of the drug. *J Clin Endocrinol Metab* 100, E427–32 (2015)  
DOI: 10.1210/jc.2014-3045

136. JE. De Larco, BR. Wuertz, LT. Furcht: The potential role of neutrophils in promoting the metastatic phenotype of tumors releasing interleukin-8. *Clin Cancer Res* 10, 4895–900 (2004)  
DOI: 10.1158/1078-0432.CCR-03-0760
137. C. Passaro, F. Borriello, V. Vastolo, S. Di Somma, E. Scamardella, V. Gigantino, R. Franco, G. Marone, G. Portella: The oncolytic virus dl922-947 reduces IL-8/CXCL8 and MCP-1/CCL2 expression and impairs angiogenesis and macrophage infiltration in anaplastic thyroid carcinoma. *Oncotarget* 7, 1500–15 (2016)  
DOI: 10.18632/oncotarget.6430
138. BC. Wang, GH. Lin, B. Wang, M. Yan, B. He, W. Zhang, AK. Yang, ZJ. Long, Q. Liu: UHRF1 suppression promotes cell differentiation and reduces inflammatory reaction in anaplastic thyroid cancer. *Oncotarget* (2016)
139. M. Rotondi, F. Coperchini, P. Pignatti, R. Sideri, G. Gropelli, P. Leporati, L. La Manna, F. Magri, S. Mariotti, L. Chiovato: Interferon- $\gamma$  and tumor necrosis factor- $\alpha$  sustain secretion of specific CXC chemokines in human thyrocytes: a first step toward a differentiation between autoimmune and tumor-related inflammation? *J Clin Endocrinol Metab* 98, 308–13 (2013)
140. E. Wennerberg, A. Pfefferle, L. Ekblad, Y. Yoshimoto, V. Kremer, VO. Kaminsky, CC. Juhlin, A. Höög, I. Bodin, V. Svjatoha, C. Larsson, J. Zedenius, J. Wennerberg, A. Lundqvist: Human anaplastic thyroid carcinoma cells are sensitive to NK cell-mediated lysis via ULBP2/5/6 and chemoattract NK cells. *Clin Cancer Res* 20, 5733–44 (2014)  
DOI: 10.1158/1078-0432.CCR-14-0291
141. JS. Cho, MH. Park, YJ. Ryu, JH. Yoon: The neutrophil to lymphocyte ratio can discriminate anaplastic thyroid cancer against poorly or well differentiated cancer. *Ann Surg Treat Res* 88, 187–92 (2015)  
DOI: 10.4174/astr.2015.88.4.187
142. J. Shiraishi, H. Koyama, M. Seki, M. Hatayama, M. Naka, M. Kurajoh, H. Okazaki, T. Shoji, Y. Moriwaki, T. Yamamoto, Y. Tsuchida, Y. Tsukamoto, S. Hirota, N. Onoda, M. Namba: Anaplastic thyroid carcinoma accompanied by uncontrollable eosinophilia. *Intern Med* 54, 611–6 (2015)  
DOI: 10.2169/internalmedicine.54.3446
143. S. Suzuki, M. Shibata, K. Gonda, Y. Kanke, M. Ashizawa, D. Ujiie, S. Suzushino, K. Nakano, T. Fukushima, K. Sakurai, R. Tomita, K. Kumamoto, S. Takenoshita: Immunosuppression involving increased myeloid-derived suppressor cell levels, systemic inflammation and hypoalbuminemia are present in patients with anaplastic thyroid cancer. *Mol Clin Oncol* 1, 959-964 (2013)  
DOI: 10.3892/mco.2013.170

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